THESIS

IDENTIFICATION OF AROMA ACTIVE COMPOUNDS IN FLASH FRIED RICE

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THESIS

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A Thesis Submitted in Partial Fulfillment of
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This study aimed to identify aroma active compounds in the flash fried rice (FFR) and to characterize the aroma attributes in FFR and stir fried rice (SFR). Gas chromatography-mass selective detector (GC-MS) was used to determine the contents of volatile compounds. A total of 72 compounds were identified in FFR and 69 compounds were identified in SFR. Aroma active compounds in both fried rice samples were identified and tentatively identified using a combine system incorporating both gas chromatography-olfactometry (GC-O) and two dimensional gas chromatography-time of flight mass spectrometry (GCxGC-TOFMS). The prominent odorants that had the $\log_3$FD factor of 5 in the typical SFR were (*E*,*Z*)-2,4-nonadienal (raw white rice) and acetophenone (sweet). Moreover, in the typical SFR also found (*E*,*E*)-2,4-heptadienal (stir-fried oil), (*E*,*E*)-2,4-octadienal (cucumber peel) and octanoic acid (oily, sweet) with $\log_3$FD factor of 4. In FFR, there were 2,3-dimethyl-5-ethylpyrazine and 2-pentylfuran that had wok aroma characteristic with $\log_3$FD factors of 4. The other prominent odorants in flash fried rice that had the $\log_3$FD factor of 5 were octanoic acid (oily, sweet) and acetophenone (sweet). 2,3-dimethylpyrazine (roast), (*E*,*E*)-2,4-heptadienal (stir-fried oil), nonanal (scented candle), 2-octanone (oily, sweet), (*E*,*E*)-2,4-hexadienal (fruity), 1-octen-3-one (mushroom) and hexanoic acid (fatty, oily) with $\log_3$FD factor of 4. The descriptive sensory analysis was performed to define the aroma attributes in fried rice samples. FFR had significant difference in heated oil, smoky, acrid, wok, animal fat, dark brown, burnt, briny and dried aroma from SFR ($p \leq 0.05$).

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IDENTIFICATION OF AROMA ACTIVE COMPOUNDS IN FLASH FRIED RICE

INTRODUCTION

Stir fried technique is a cooking method that used in Asia. This cooking method can be categorized into stir frying and flash frying. The two techniques differ in the level of heat used and their speed of cooking in the wok (Chao, 1945). Flash frying is the special technique of cooking that significantly enhances food flavor during heating. This technique gives a unique smoke/burnt aroma which generated when the edible oil and the main ingredients attract fire around several seconds.

Wu and Chen (1992) reported more than 40 volatile compounds that were found in stir fried soy bean oil at 200 °C. They were groups of aldehydes, ketones and furans. Furthermore, hexanal, (*E*)-2-heptenal, (*E*)-2-octenal and 2,4-decadienal were principal contributors to the prominent aroma of stir fried soy bean oil that gave the rancid and heated oil aromas. Flash frying involves heating of foods at the flash point of the cooking oil. Volatile compounds from flash frying can be generated from pyrolysis. Major products from pyrolysis of vegetable oils are *n*-alkanes and 1-alkenes and minor products are groups of alkylcycolalkanes (Alencar *et al.*, 1983).

Pyrolysis is defined as a chemical degradation reaction that is caused by thermal energy and usually takes place at temperatures higher than 300 °C (Moldoveanu, 1998). Pyrolysis gives special flavor to the flash fried food when ingredients are exposed to high temperature. The main components in the flash-fried rice (FFR) that undergo pyrolysis are carbohydrates in rice and edible oil.

Patwardhan *et al.* (2009) revealed that the main pyrolysis products of carbohydrates were the low molecular weight compounds, furan ring derivatives and anhydrous sugars. Furan derivatives such as 5-methyl furfural, 2-furan methanol, 3-furan methanol were described as burnt sugar and caramel-like (Acree and Arn, 2009). Volatile compounds generated from pyrolized oil were carboxylic acids, aldehydes, alcohols, alkenes and alkadienes (Fortes and Baugh, 2004).

In previous study, Piyachaiseth (2010) reported that the most prominent volatile compounds of flash fried rice were 2,4-heptadienal (stir-fried oil, burnt), nonanal (scented candle), heptanone (metallic, rust) and unknowns that generated fishy, salty and sweet aroma. There were 2 unknowns with FD factors of 9 and 0 that had “wok” or the flash fried aroma characteristics. These compounds were only detected by gas chromatography-olfactometry (GC-O) method but not by the mass selective detector. These unknown compounds could present in low concentrations but had low threshold values.

In this research, we continued our study to better extract and concentrate the flash fried rice aroma in order to identify the aroma compounds for future uses in food manufactures. The identification and extraction of the flash-frying aroma active compounds can benefit to future synthesis of the wok flavor components and eliminate the health problem of associated with carcinogen generated during the high temperature cooking. Piyachaiseth (2010) used different from control test and reported that the untrained panelists could differentiate the aroma of flash fried rice from the stir fried rice. However, her study did not specify the differences on each attribute. In this study the descriptive sensory analysis was performed to defined aroma attributes in fried rice samples.

OBJECTIVES

The objectives of this work were:

1. to characterize the aroma attributes in the flash fried rice
2. to identify aroma active compounds in the flash fried rice

LITERATURE REVIEW

1. Stir frying technique

Stir fried technique is a cooking method that used in Asia. This cooking method can be categorized into stir frying and flash frying. The two techniques differ in the level of heat used and their speed of cooking in the wok (Chao, 1945). Stir frying is a simple cooking technique for preparing food in a wok involving high rates of heat and mass transfer (Nissen, 2007). A wok is heated to a high temperature. Then the small amount of cooking oil is poured down the side of the wok follows by other ingredients. All ingredients are stirred and tossed out very quickly using a cooking utensil. Many dishes can be cooked extremely quickly within minutes.

Flash frying is also a cooking with the same procedure as stir frying but it has the special cooking technique that significantly enhances food flavor during heating. This technique gives a unique smoke/burnt aroma that generated when the edible oil and the main ingredients attract fire for several seconds. Flash frying could preserve flavors that are lost in the slow cooking. For example, a tasty, fat-free substitute might compete with the conventional French-fried potatoes (Squires, 2007).

2. Wok flavor (wok hay)

Wok flavor or wok hay is the description of the unique flavor developed by the flash frying at high temperatures. The “wok” characteristic refers to the pan and the “hay” character refers to the energy or breath (Young, 2004). The key to get wok flavor is the heat. The wok must be heated to the point that the cooking oil generates a lot of smoke before the first ingredient hits into the wok. Moreover, the important technique to get wok flavor is the Chinese cooking technique call "bao". The ingredients are tossed over in the wok, and then the edible oil and the main ingredients attract fire for several seconds. The surfaces of foods will be mild burnt and release the inner essence and the unique smoke/ burnt aroma.

3. Aroma active compounds in flash fried rice and stir fried rice

In previous study, Piyachaiseth (2010) revealed that, there were the unique smoky and burnt aroma perceived in flash fried rice. While stir fried rice showed the heated oil as the prominent odor. The results from the gas chromatography-olfactometry (GC-O) using the aroma extraction dilution analysis (AEDA) technique indicated that, there were aldehydes, ketones, esters, hydrocarbons, furans and pyrans as the aroma active compounds in both fried rice sample (Table 1).

Table 1 Selected aroma active compounds in flash-fried rice and stir-fried rice.

Compound	**Description**	**RI**		**FD factor**	
		FFAP	**HP-5**	**FFR**	**SFR**
butanal	green	<1000	n.a.	27	3
unknown A	fishy, salty	1061	n.a.	81	3
unknown B	sweet, stale	1135	n.a.	81	3
heptanone	metallic (rust)	1204	n.a.	81	9
2-acetyl-1-pyrroline	sweet, pandan like	1220	939	27	3
unknown C	wok	1266	n.a.	9	-
unknown D	acetic acid	1319	n.a.	27	3
unknown E	wok	1321	n.a.	0	-
nonanal	sweet, smoky	1370	n.a.	81	3
(*E*,*E*)-2,4-heptadienal	stir-fried oil, sour	1461	n.a.	27	0
2,4-heptadienal	stir-fried oil, burnt	1475	n.a.	81	-
(*E*)-2-nonenal	stir-fried oil	1531	n.a.	27	3
(Z)-2-decenal	stir-fried oil, burnt	1586	1256	27	-
heptanone	metallic, solvent	n.a.	885	27	3
unknown F	sweet	n.a.	907	27	3
unknown G	fishy	n.a.	917	-	27
acetylfuran	sweet	n.a.	923	27	3
octadienone	metallic (rust)	n.a.	997	27	81
unknown H	sweet	n.a.	1024	-	81

Table 1 (Continued)

Compounds	**Description**	**RI**		**FD factor**	
		FFAP	**HP-5**	**FFR**	**SFR**
limonene	citrus	n.a.	1035	27	-
decanal	sweet, smoky	n.a.	1205	27	9
dodecanal	stir-fried oil, burnt	n.a.	1407	27	9
2-dodecenal	stir-fried oil	n.a.	1461	27	-
methyl dodecanoate	stir-fried oil	n.a.	1511	-	27
hexyl octanoate	stir-fried oil, burnt	n.a.	1568	27	27

Note n.a.= not available

Source: Piyachaiseth (2010)

Table 1 reveals that the most prominent volatile compounds of stir fried rice that have the highest flavor dilution (FD) factor of 81 were octadienone (metallic, rust) and an unknown H (sweet). Flash fried rice had 2,4-heptadienal (stir-fried oil, burnt), nonanal (scented candle), heptanone (metallic, rust), unknown A (fishy, salty) and unknown B (sweet, stale) as its prominent aroma compounds. There were 2 unknowns with FD factors of 9 and 0 that had the wok or the flash fried aroma characteristics. These compounds were only detected by GC-O but not by GC-MS. Therefore, these compounds could present in low concentrations but had low threshold values.

4. Volatile compounds in deep fried food

There are many researches study about volatile compounds from deep fried potato. Several hundred compounds were found in deep fried potato at 150-190°C for 1-3 min. There were 24 acids, 37 alcohols, 30 aldehydes, 10 esters, 20 furans, 105 hydrocarbons, 36 ketones, 5 lactones, 25 oxazoles, 33 pyrazines, 13 pyridines, 1 pyrrole, 50 thiazoles, 19 thiophenes, 16 sulfur compounds and 16 nitrogen compounds (Carlin *et al*., 1986).

Aldehydes from lipid oxidation in deep fried potato can produce thiazole and oxazole. Whitefield (1992) found that 4-methyl-5-ethyl-2-octylthiazole and 4-methyl-5-

ethyl-2-pentylthiazole were aroma active compounds in deep fried potato. Moreover, 3-(methylthio)-butanol and 3-(methylthio)-heptanol were important volatile compound to pronounced unique aroma in deep fried potato (Carlin *et al*., 1990).

Volatile compounds in deep fried potato using palm oil were studied by Wagner and Grosch (1997). There were 48 volatile compounds and 2,3-diethyl-5-methypyrazine, (*E*,*E*)-2,4-decadienal, 4-hydroxy-2,5-dimethyl-3(2*H*)-furanone, methanethiol, dimethyl trisulfide, 3-methylbutanal and 2,3-butanedione were aroma active compounds in this sample.

Eighty five percent of volatile compounds in the deep fried potato formed from sugar degradation and Maillard reaction. Fifteen percent of volatile compounds formed from lipid degradation. Forty one aroma compounds in the deep fried potato were perceived by the panelists using a GC-O. 3-Methylbutanal, 2-methylbutanal, hexanal, 2,3-dimethylpyrazine, 2-methylpropanal, 2,3-butanedione, pyridine, heptanal, 2,5-dimethylpyrazine, 2,6-diemethylpyrazine, ethylpyrazine, dimethyl trisulfide, octanal, phenylacetaldehyde, 2,5-diethylpyrazine, (*E*)-2-nonenal, 3-methylbutanoic acid, 2-methylbutanoic acid, (*E*,*Z*)-2,4-heptadienal, (*E*)-2-octenal, 5-ethyl-2,3-dimethylpyrazine, 2-ethyl- 3,5-dimethylpyrazine, nonanal and 2-methylpyrrole were of the highest frequency being perceived by the panelists. Most of these compounds had the malt, deep fried potato, caramel, butter and heated oil characteristics (van Loon *et al*. 2005).

5. Thermal process flavor

Thermal process flavor is the product obtained after heating the ingredients. The formation of the thermal process flavor involves complex compounds that generated from carbohydrates, proteins and lipids. There are many compounds that give several thousand flavors, depending on the food products. Time, temperature and water content are the main influencing factors. Volatile compounds in FFR are derived from pyrolysis process, Strecker degradation, Maillard reaction, caramelization and lipid oxidation.

5.1 Pyrolysis

Pyrolysis is a chemical degradation of organic materials caused by thermal energy. This process usually takes place at temperatures higher than 250 °C. Mild pyrolysis is considered to take place between 300-500 °C and vigorous pyrolysis is more than 800 °C without the presence of oxygen and does not occur as long as free water is present (Moldoveanu, 1998).

Pyrolysis is the first chemical reaction that occurs in the burning of many materials. In food, pyrolysis occurs when food exposed to high enough temperature in a dry environment such as roasting, frying and grilling. The main food components that undergo pyrolysis are carbohydrates, proteins and lipids. There are many reactions occur after the pyrolysis process. The thermal energy is the only cause of this chemical degradation with the formation of molecules smaller than the starters. Many small volatile compounds, colorant and others compounds can be generated form these small molecules.

5.1.1 Pyrolysis of carbohydrate

Pyrolysis products identified from sugars are common to all saccharides. They give similar compounds in the final products if those saccharides have the same monosaccharide units. There are several classes of compounds formed from pyrolysis of carbohydrates. For examples, anhydrosugars, furan derivatives, lactones, pyran derivatives, phenols, acids and acid esters, and other compounds.

A variety of small molecules are formed during pyrolysis of polysaccharides. Hydroxyacetone, 2-furaldehyde, 2-hydroxycyclopent-2-en-l-one, 5-hydroxymethyl-2- furaldehyde and 1,5-anhydro-4-deoxy-glycero-hex-l-en-3-ulose are known as common pyrolysis products form carbohydrates (Moldoveanu ,1998). Several mechanisms show the formation of smaller molecules from monosaccharides. For example, the formation of hydroxyacetaldehyde is one of the products that may take place by a retro-aldolization (Figure 1).

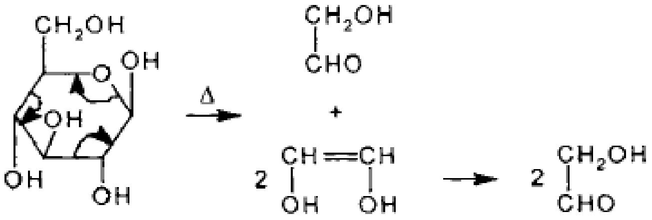


Figure 1 Retro-Aldolization of monosaccharides.

Source: Moldoveanu (1998)

The formation of 5-hydroxy-2-furancarboxaldehyde (hydroxyl methylfurfural) and furancarboxaldehyde are the products that take place by the elimination of water and elimination of water with other small molecules respectively (Figure 2 and 3).

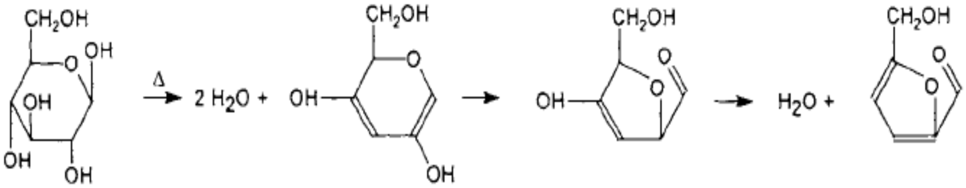


Figure 2 Water elimination of glucose.

Source: Moldoveanu (1998)

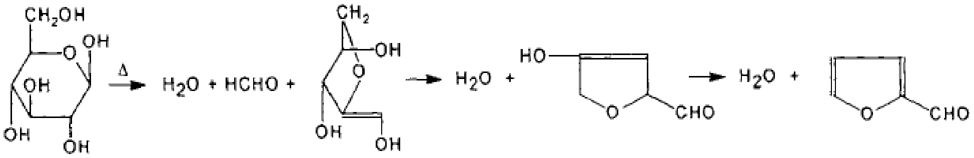


Figure 3 Elimination of water with other small molecules of glucose.

Source: Moldoveanu (1998)

Patwardhan *et al.* (2009) studied the pyrolysis process of glucose, cellobiose, maltose, maltohexose, cellulose in the inert gas condition at 500 °C by using the micro furnance single short pyrolyzer. The main products were levoglucosan (pyranose), formic acid, furaldehyde, furan, 5-hydroxymethyl furfural (Table 2). Table 2 divides the main products into three groups. There were formic acid, acetone, acetic acid, furan derivatives and anhydro sugar.

Table 2 Pyrolysis products from glucose cellobiose maltose, maltohexose, cellulose and maize starch (% by weight).

Compound	**Glucose**	**Cellobiose**	**Maltose**	**Malto hexose**	**cellulose**	**Maize starch**
formic acid	7.25	10.05	7.91	12.41	6.59	5.31
furan	1.25	0.84	0.86	0.93	0.73	0.71
glycolaldehyde	6.62	7.12	8.41	6.09	6.69	6.95
acetic acid	0.07	0.04	0.05	0.08	0.04	0.02
2-methyl furan	1.03	0.62	0.74	0.97	0.37	0.35
acetol	1.14	0.52	0.62	0.96	0.30	0.46
2-furanaldehyde	8.35	6.67	5.64	4.20	1.26	1.06
2-furan methanol	0.60	0.60	0.71	0.82	0.54	0.45
3-furan methanol	0.15	0.21	0.15	0.10	0.25	0.15
5-methy furfural	0.92	0.54	0.55	0.45	0.24	0.52
2-hydroxy-3-methyl cyclopenten-1-one	0.33	0.06	0.06	0.45	0.21	0.45
levoglucosenone	0.16	0.41	0.12	0.36	0.35	0.33
5-hydroxymethy furfural	7.70	8.74	8.87	6.79	2.76	3.70
anhydroxylopyranose	0.05	1.13	1.15	1.11	2.95	5.18
levoglucosan-pyronose	7.00	24.36	20.51	33.11	58.78	48.48
levoglucosan-furonose	3.90	3.89	3.69	3.90	4.08	4.20
other anhydrosugars	2.54	0.90	1.19	1.85	1.43	3.07
unidentified fraction	40.82	23.59	32.44	20.54	7.07	13.20

Source: Modified from Moldoveanu (1998)

Furan and furan derivatives were defined as the main important products from the pyrolysis of glucose. There were furfural [1], 5-hydroxymethyl- furfural [2], furan [3], 5-methylfurfural [4], 2-methylfuran [5], 2,5-dimethylfuran [6], 3-[2*H*]-furanone [7], 2-furyl hydroxymethyl ketone [8], 2-(1,2-di-hydroxyethyl) furan [9], furfuryl alcohol [10], 3-furfural [11], 4-hydroxytetrahydrofuran-3-one [12] and 3-methyl-furan [13] in Figure 4 (Paine *et al.*, 2008) that had the caramel and the burnt sugar characteristics (Acree and Arn, 2009).

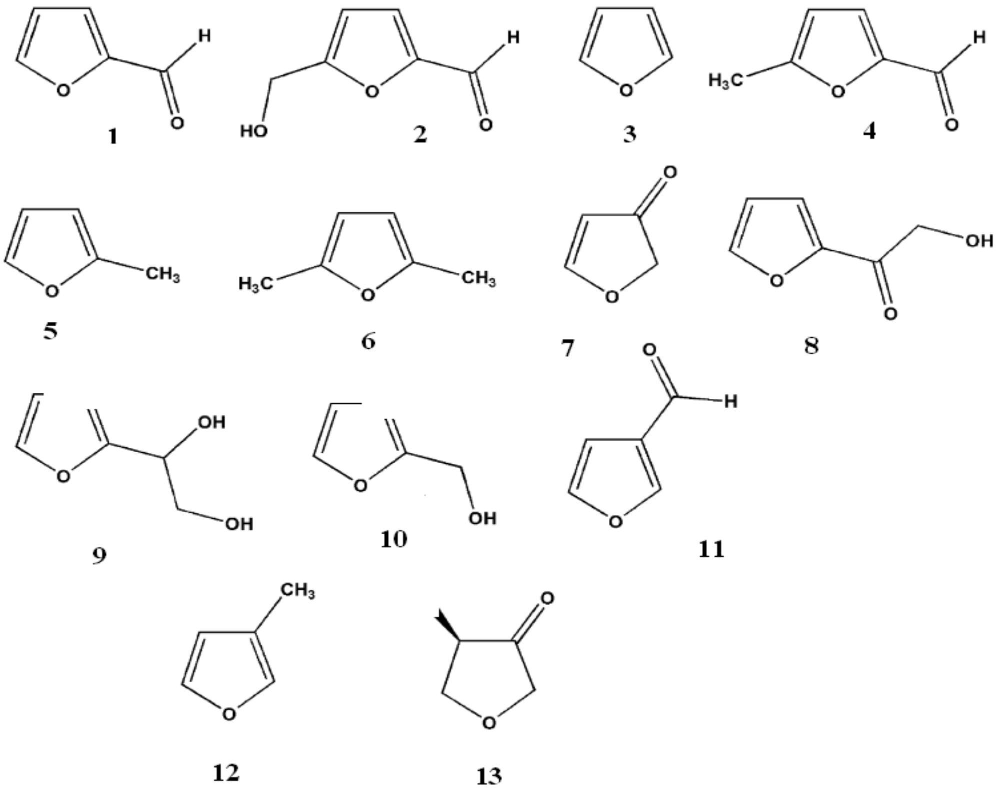


Figure 4 Furan and furan derivative products from pyrolysis of glucose.

Source: Paine *et al.* (2008)

5.1.2 Pyrolysis of lipids

Lipid is the organic compounds refer to fatty acid esters consisted of a variety of compounds such as glycerol esters, waxes, phosphoglycerides, sphingolipids, hydrocarbons and vitamins. There are several classes of compounds that formed from the pyrolysis of lipids such as carboxylic acids, aldehydes, alcohols, alkenes, alkadienes and other compounds.

Several mechanisms show the formation of smaller molecules from the pyrolysis of lipid structure. For example, the formation of polynuclear aromatic hydrocarcon is the product that may take place by the dehydrogenation of sterol (Figure 5).

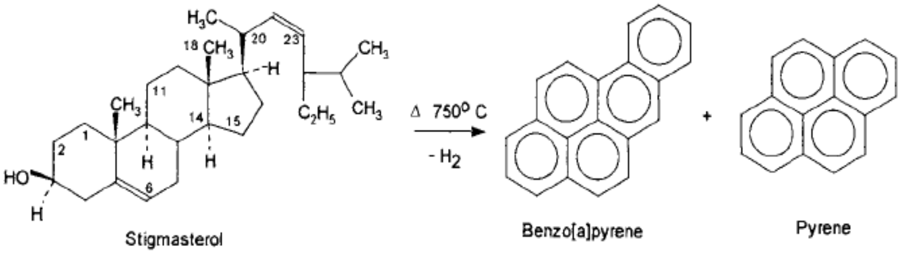


Figure 5 Dehydrogination of stigmasterol.

Source: Moldoveanu (1998)

The formation of alcohols and carboxylic acids are the products that take place by the hydrolysis of triacylglycerols and aromatic hydrocarbons are generated from the diels-alder process (Figure 6 and 7).

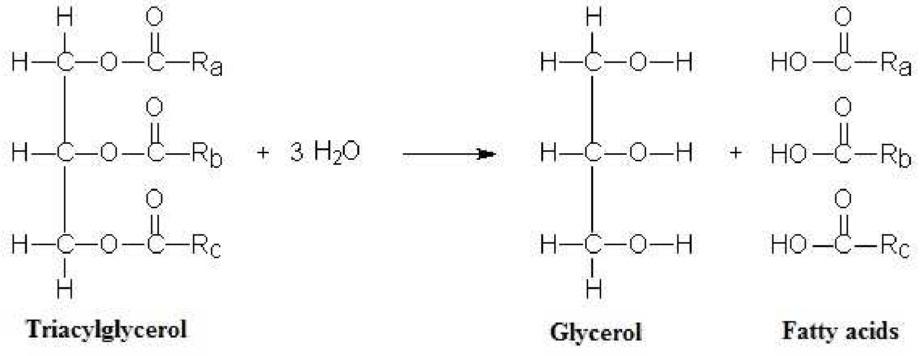


Figure 6 Hydrolysis of triacylglycerol.

Source: Moldoveanu (1998)

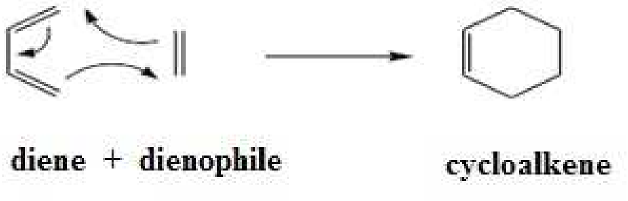


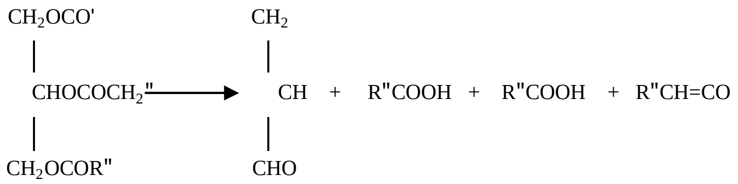
Figure 7 Diels-Alder process.

Source: Moldoveanu (1998)

Several researches have studied on pyrolysis or thermal cracking of triacylglycerols. These studies focused on pyrolysis of triacylglycerols as food materials and also as fuels (Maher and Bressler (2007). Fortes and Baugh (1999, 2004) studied the pyrolysis of macauba fruit oil under conditions of fast pyrolysis. The main products were aldehydes, alkenes and carboxylic acids, the secondary products consisted of alkanes, cycloalkanes and unknowns.

The machanism for the pyrolysis of saturated triacylglycerols was presented by Chang and Wan (1947). There were 16 types of reactions (Figure 8).

(1) Decomposition of the glyceride



$$\begin{array}{l} CH_2OCO' \\ | \\ CHOCOCH_2'' \\ | \\ CH_2OCOR'' \end{array} \longrightarrow \begin{array}{l} CH_2 \\ | \\ CH \\ | \\ CHO \end{array} + R''COOH + R''COOH + R''CH{=}CO$$

(2) Decomposition of fatty acids

$RCOOH \longrightarrow CO_2 + RH$

$2RCOOH \longrightarrow CO_2 + H_20 + RCOR$

(3) Decomposition of ketenes and acrolein

$2R''CH{=}CO \longrightarrow 2CO + RHC{=}CHR$

$CH_2{=}CHCHO \longrightarrow CO + C_2H_2$

$RCOCH_2R \longrightarrow R - R + CH_2CO$

$2RCOCH_2R \longrightarrow 2R_2 + CO + C_2H_4$

(4) Decomposition in elements

$C_nH_{2n+2} \longrightarrow nC + (n+1)\,H_2$

(5) Decomposition of paraffins

$C_nH_{2n+2} \longrightarrow C_nH_{2n} + H_2$

(6) Splitting Decomposition of paraffins

$C_nH_{2n+2} \longrightarrow C_nH_{2n-2m+2} + C_mH_{2m}$

(7) Alkylation of paraffins,the reverse of (6)

(8) Isomerization of paraffins

$N - C_nH_{2n+2} \longrightarrow iso - C_2H_{2n+2}$

(9) Aromatic cyclization of paraffins

$C_{(2n+6)}H_{(2n+14)} \longrightarrow C_nH_{2n+1} + 4H_2$

(10) Polymerization of olefins

$2C_nH_{2n} \longrightarrow C_{2n}H_{4n}$

$C_nH_{2n} + C_mH_{2m} \longrightarrow C_{(n-m)}H_{2(n-m)}$

(11) Depolymerization of olefins,reverse of (10)

(12) Decomposition of olefins to diolefins

(13) Decomposition of olefins to acetylenic hydrocarbon

(14) Aromatization or cyclization of olefins

(15) Hydrogentation of olefins

$C_nH_{2n} + H_2 \longrightarrow C_nH_{2n+2}$

(16) Isomerization of olefins

$n - C_nH_{2n} \longrightarrow iso - C_nH_{2n}$

Figure 8 Decomposition of triacylglycerols.

Source: Chang and Wan (1947)

Vegetable oil also contains unsaturated components. Schwab *et al*., (1988) studied the impact of heat on unsaturated fatty acids and presented a mechanism to generate alkanes, alkenes, alkadienes, aromatics and carboxylic acids from the pyrolysis of unsaturated triacylglecerols (Figure 9).

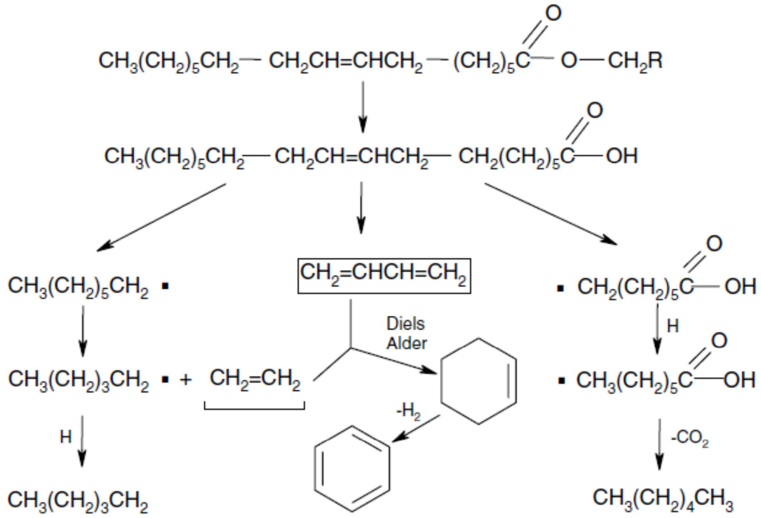


Figure 9 Reaction mechanism for the pyrolysis of triacylglycerols.

Source: (Schwab *et al*., 1988)

5.1.3 Pyrolysis of proteins

Proteins are natural macromolecular compounds that are the major components in foods. The main constituents of proteins are amino acids. The amino acid pyrolysis is relevant to protein pyrolysis. The compounds in the pyrolysates obtained from the pyrolysis of amino acids or peptides containing the same amino acids are the same (Moldoveanu, 1998).

The main mechanisms in Figure 10 are the decarboxylation by CO_2 elimination or the water elimination with the formation of a dipeptide and further of the diketopiperazines. Amino acids may be converted to aldehydes by Strecker degradation. Furthermore, R-H type compounds may be generated. This is the characteristic for the aromatic amino acids. Smaller molecules, some unsaturated, are also formed by further decompositions. The compounds of the type R-CH=N-CH_2-R and R'=C=N-CH_2-R are formed from aliphatic amino acids (Moldoveanu, 1998).

The main pyrolysis products of several amino acids are classified in many groups (Table 3). There were pyrroline, pyridine, pyrrole, aldehyde, indole, phenol and thiazole. Those compounds always show nutty, roast, burnt and popcorn-like characteristic.

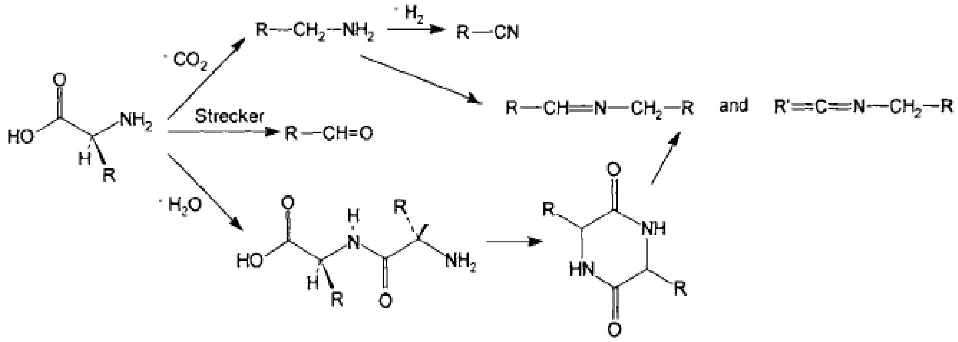


Figure 10 The main mechanism of protein pyrolysis.

Source: Moldoveanu (1998)

Table 3 The main pyrolysis products of several amino acids.

Amino acids	**Compounds**
Alanine	methylamine
	3,4-dimethylpiperazin-2,5-done
Arginine	pyrroline
	3-amino-2-piperidone
Asparagine	maleimide
	succinimide
Cysteine	methylpyridine
	methylthiazolidine
	methylethylthiazole
Glutamine	pyrrole
	2,3-dehydro-2-piperidone
	methyl-2,3-dehydro-2-piperidone
Hydroxyproline	pyrroline
	pyrrole
	2-ethylpyrrole
	1*H*-2,3-dihydropyrrole-2-carboxylic acid
	1,2-dipyrrolylethane
	1,3-dipyrrolylpropane
	diketodipyrrole
Isoleucine	butanenitrile
	2-methylbutene
	2-methylbutanal
	2-methylbutyronitrile
	2-methylbutylamine
	1-butanamine-2-methyI-N-(2-methylbutylidene)
	3,6-(2-methylpropyl)-2,5-diketopiperazine
Leucine	butanenitrile

Table 3 (Continued)

Amino acids	Compounds
	3-methylbutene
	3-methylbutanal
	3-methylbutyronitrile
	3-methylbutylamine
	1 -butana mine-3-methyI-N-(3-methylbutylidene)
	3,6-diisobutyl-2,5-diketopiperazine
Lysine	pyrroline
	3-aminohexahydroazepinone
	methionine
	methanethiol
	vinyl-l-methylthioether
	propane-l-methylthioether
	3-methylthio-1-propylamine
	1-propanamine-*N*-(3-methylthiopropylidene)
Phenylalanine	toluene
	ethylbenzene
	ethenylbenzene
	benzene-ethaneamine
	phenylacetonitrile
	diphenylethane
Proline	pyrroline
	pyrrolidine
	2,5-diketopiperazine
Tryptophan	indole
	3-methylindole
	3-ethenylindole
	3-ethylindole
	2,3-dimethylindole
	indole-3-ethanamine
	indolylacetonitrile

Table 3 (Continued)

Amino acids	**Compounds**
Tyrosine	phenol
	4-methylphenol
	4-ethylphenol
	4-ethenylphenol
	phenyl-acetaldehyde
	4-hydroxyphenetylamine
Valine	isobutene
	2-methylpropanal
	2-methylpropionitrile
	2-methyl-l-propanamine
	methylamine-*N*-(2-methylpropylidene)
	ethylamine-*N*-(2-methylpropylidene)
	2-methylpropylamine-N-(2-methylpropylidene)
	3-isopropyliden-2,5-diketopiperazine
	3,6-diisopropyl-2,5-diketopiperazine

Source: Moldoveanu (1998)

5.2 Maillard Reaction

Maillard reaction is a chemical reaction between an amino acid and a reducing sugar. This reaction is one of the most important reaction which results from food processing. The reactive carbonyl group of the sugar interacts with the nucleophilic amino group of the amino acid. Maillard reaction products affect the food quality attributes such as flavor, aroma, color and texture. Several hundreds of flavor compounds are created from the Maillard reaction. Each type of food has the unique flavor that is formed during Maillard reaction.

Flavors and aromas are developed during Maillard reaction depends on the temperature, time, pH, water content and the types of sugars and amino acids (Yu and Zhang, 2010; Van Boekel, 2006). The intermediate and the final stages of Maillard reaction are the most important to flavor development, especially in Strecker degradation.

Amino acids are degraded by dicarbonyls that previously formed in the reaction, then lead to the amino acid deamination and the decarboxylation in Strecker degradation (Rizzi, 2008).

The volatile products of Maillard reaction can be classified from the sugar dehydration products and the amino acid degradation products. There were furans, pyrones, cyclopentenes, carbonyls and acids, aldehydes and sulfur compounds. After that pyrroles, pyridines, imidazoles, pyrazines,oxazoles, thiazoles, and others were produced by further interactions as shown in Figure 11. Pyrazines and alkylpyrazines are associated with the the cooked and nutty aromas. Alkylpyridines refer to the odors of green, bitter, astringent and burnt. Furans, furanones and pyranones are associated with the sweet, burnt, pungent and caramel-like odor (Bastos *et al*., 2012). The compounds that are essential to generate flavor and aroma of food products normally present in trace levels.

The nitrogen-containing aroma compounds 2-ethyl-3,5dimethylpyrazine, 2,3-diethyl-5-methylpyrazine and 2-acetyl-1-pyrroline are found in foods in the range of 0.001–10 mg/kg. While, sulfur containing compounds from Maillard reaction are the most powerful aroma compounds at trace levels and play an important role in the flavor of cooked meats. These volatile compounds are responsible for the flavor and aroma of stewed beef juice, boiled trout, french fries, bread crust, cooked chicken, roasted chicken, boiled beef, cocoa powder, peanuts, pilsner beer, roasted beef, popcorn and coffee (Bastos *et al*., 2012).

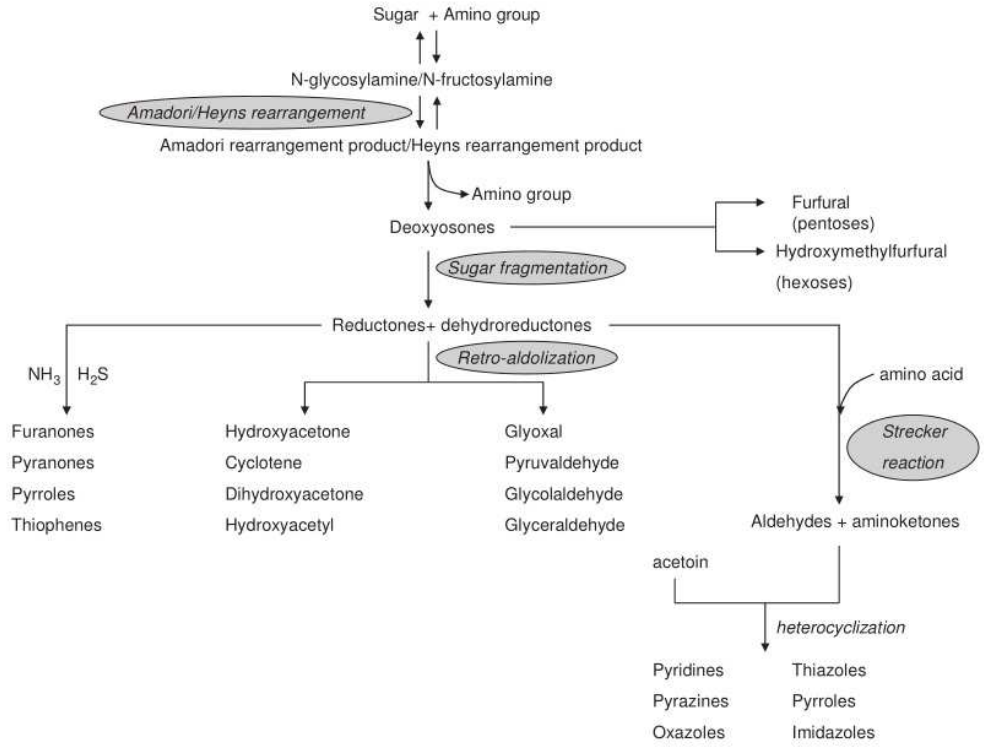


Figure 11 General overview of the Maillard reaction showing flavour compounds as end products.

Source: van Boekel (2006)

5.3 Strecker degradation

Strecker degradation has been known as a sub reaction in Maillard reaction. This reaction is the oxidative decarboxylation reactions of amino acids that can be affected by varieties of reagents and reaction conditions. Strecker degradation of amino acids often occurs in conjunction with Maillard reaction. In Strecker degradation, amino acids are decarboxylated and deaminated in the presence of dicarbonyl compounds forming Strecker aldehydes and aminoketones (Figure 12).

Aldehydes may often be important reactive carbonyls that can take place in Maillard reaction, while aminoketones may take part in the cyclisation reactions to produce heterocyclic compounds such as pyrazines. Dicarbonyl compounds that participate in

Strecker degradation may be derived from lipid oxidation or from Maillard reaction (Ó Hare and Grigor, 2005).

5.4 Caramelization

Caramelization is one of the most important types of browning processes in foods. This reaction is a type of non-enzymatic browning reaction and causes important changes in foods, especially in color and flavor. Caramelization occurs during dry heating and roasting of foods with high concentrations of carbohydrates (sugars). Figure 13 shows the caramelization reaction. The reaction involves the removal of water and the breakdown of the sucrose into glucose and fructose by heating. After the hydrolysis of sucrose and sugar breakdown, volatile compounds are released producing the characteristic caramel flavor (Ó Hare and Grigor, 2005).

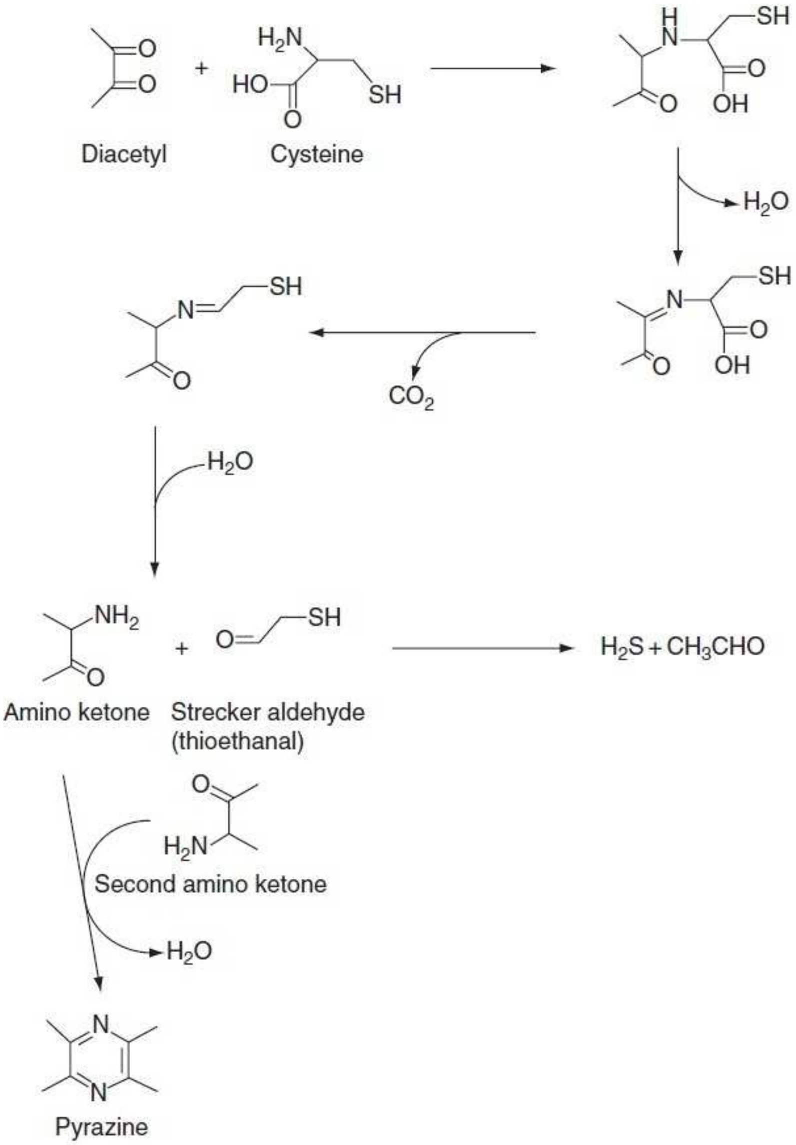


Figure 12 The Strecker degradation (illustrated by the reaction of diacetyl with cysteine).

Source: Ó Hare and Grigor (2005)

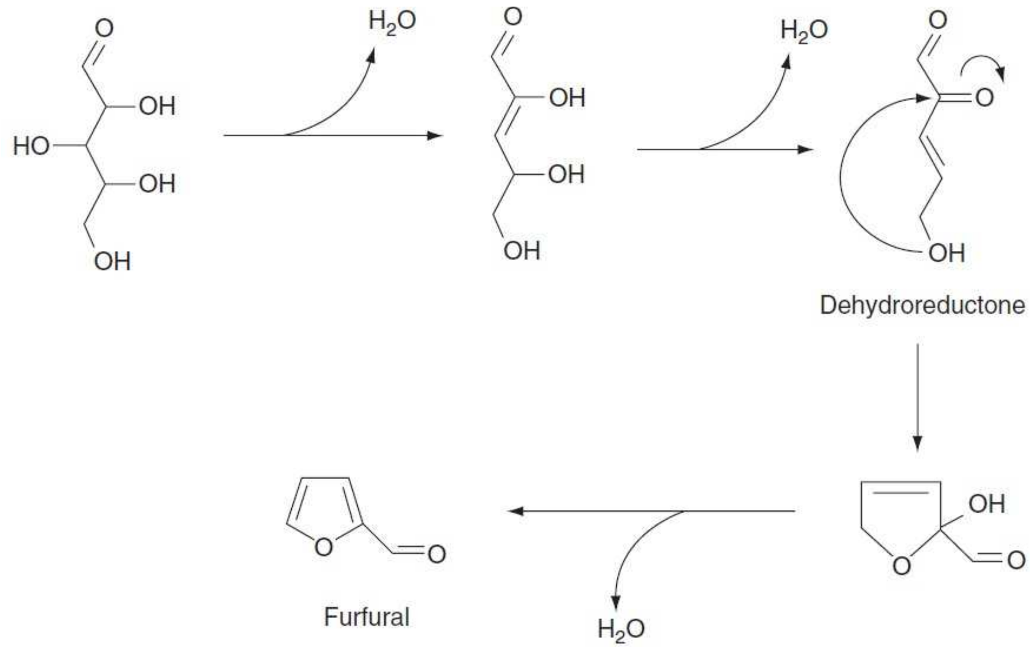


Figure 13 Caramelization.

Source: Ó Hare and Grigor (2005)

5.5 Lipid oxidation

Lipid oxidation generated several hundreds of the volatile compounds. These compounds include aliphatic hydrocarbons, aromatic hydrocarbons, aldehydes, ketones, alcohols, fatty and other carboxylic acids, esters, lactones and alkylfurans.

Lipid oxidation is the significant contributor to cooked meat aroma. Some lipid oxidation products have strong and characteristic aromas, and even at very low levels, compounds such as *trans*,*cis*-2-4-decadienal, *cis*-hex-3-enal and penta-2,4-dienal contribute the prominent and unpleasant rancid aroma of the oxidized fats. The rancid aroma can be generated from autoxidation of unsaturated fatty acids. The reaction occurs in three steps as follows;

1. Initiation

– A radical is formed

RH + (reactive oxygen species) ·OH -->R· + H2O

2. Propagation

– Radicals react and transfer their unpaired electron to other compounds

R· + O2 ------> ROO·
ROO· + RH ------> R· + ROOH
ROOH -------> RO· + HO-

3. Termination

– Two radicals combine to stop the reaction

R· + R· ------> RR
R· + ROO· ------> ROOR
ROO· + ROO· ------> ROOR + O2

Lipid degradation can involve hydrolysis to release free fatty acids. While short-chain fatty acids have distinctive odours, the long-chain fatty acids that are more common in meat fat are not significant aroma chemicals. Hydrolysis is favoured by high water activity and acid/base catalysis (Ó Hare and Grigor, 2005).

Fatty aldehydes with small structures (C6-C10) are perceived as green, fatty or tallow aromas, for examples hexanal, heptanal and octanal. Unsaturated aldehydes may have more interesting aromas, for example 2,4-decadienal has an aroma of fat-fried food. Alkyl derivatives of heterocyclics also have fried or fatty flavors (Ó Hare and Grigor, 2005).

6. Isolation of volatile compounds

6.1 Direct solvent extraction

Direct solvent extraction is the technique that wildly used. The extract is obtained by mixing the sample with the organic solvent. Volatile compounds will transfer from the sample to the solvent. Finally, solvent phase will be separated and collected. The extraction can be done in batches or a continuous flow extraction. When the extraction process is done, the analyst should end up with the extract that its sensory property is closely resembled to those of the original sample. The solvents most commonly used are diethyl ether, diethyl ether/pentane mixtures, hydrocarbons and methylene chloride.

Solvent must have a low boiling point because it can be easily removed from the extract without a loss of volatiles in the sample. Moreover, it must be able to extract polar and non-polar components. Below are the advantages and disadvantages of the solvent extraction method by Costa and Eri (2005).

Advantages

- Good recovery of aroma compounds; the most quantitative of all isolation methods.
- Easy to perform.

Disadvantages

- Solvent removal may cause loss of some of the volatile compounds.
- Use of solvents may be toxic and/or flammable and may introduce contaminants.
- Need for large amounts of sample in order to produce a strong extract.
- Obtained extracts may contain high-boiling and non-volatile materials and color components, which may lead to further problems in analysis and chromatography.
- Emulsion formation may occur during the extraction.
- Solvent peak may cover early eluting volatiles in the chromatogram.

6.2 High vacuum distillation

High vacuum distillation apparatus was developed to isolate volatile compounds from lipid food matrices. A high vacuum distillation technique suitable for the distillation of foods or solvent extracts of the food material. The idea of high vacuum distillation was to transfer the volatiles in an evacuated system. High vacuum distillation system was performed in the extreme temperature differential between two vessels connected via a glass tube, thereby removing the non-volatile material (Engel *et al*., 1999).

The primary principles used in isolation of aroma components are volatility, partition, or both in combination. If the foods have the high lipid content, the release of volatile compounds into the gaseous phase is low. Engel *et al*. (1999) presented a vacuum distillation method to transfer the flavor molecules at lower temperatures. A condenser was used to condense highly volatile flavor compounds under continuous vacuum pumping.

The apparatus currently used for performing the aroma isolation by high vacuum distillation is shown in Figure 14.

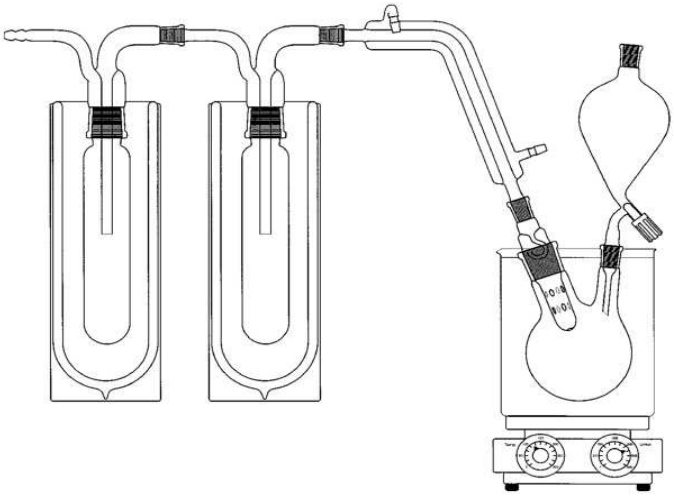


Figure 14 Equipment for high vacuum distillation.

Source: Engel *et al*. (1999)

7. Analysis of volatile compounds by using the comprehensive two dimensional gas chromatography

Aroma extracts from foods, is a complex components. Peak overlapping in many samples occur when the conventional gas chromatography (one dimensional gas chromatography) is performed. Two dimensional gas chromatography was designed to solve this problem by increasing the peak capacity in the two independent separation processes.

In the two independent separation processes, the samples pass through the first column to the second column. Target and non-target of volatile compounds are analyzed in a system as show in Figure 15. This technique, called two dimensional gas chromatography (GC×GC). The important section in GC×GC is modulator that connects the first dimension with the second dimension (Figure 16). Modulator function (Hajslova and Cajka (2009):

1. trapping of small adjacent fraction from the first dimension (1st column)
2. refocusing on the small adjacent fraction
3. injection of the refocused fraction into the second dimension (2nd column)

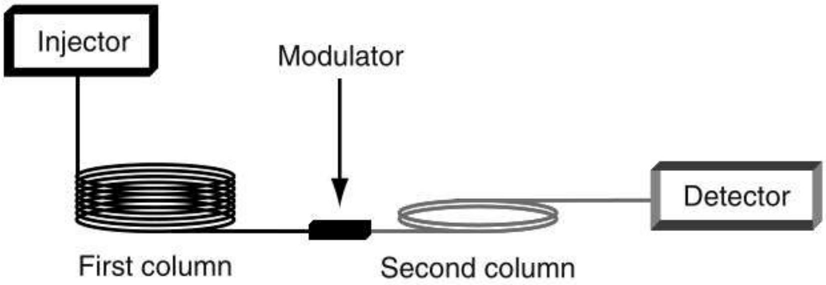


Figure 15 GC×GC configuration.

Source: Hajslova and Cajka (2009)

The convenient method to analyze GC×GC data is a contour plots. A contour plot representing the bird's eye view, where peaks are displayed as spots on a plane using colors and shading to indicate the signal intensity (Hajslova and Cajka, 2009).

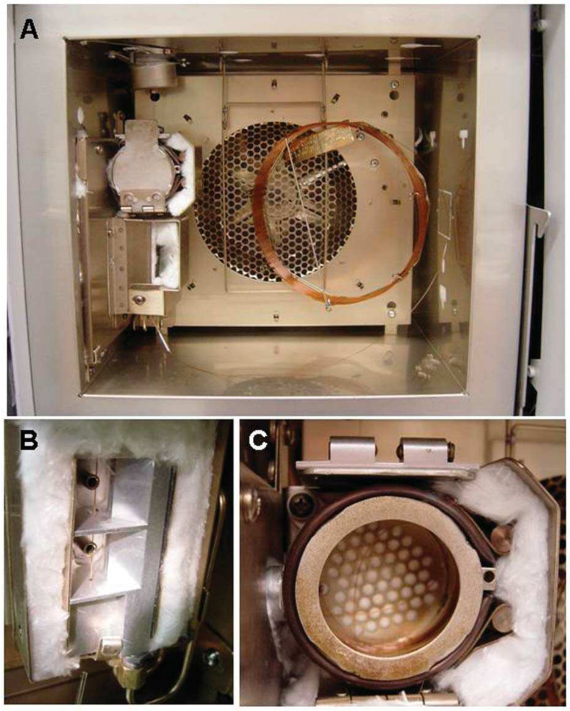


Figure 16 A typical GC×GC setup. (A) Inside of the1stD oven of a GC×GC system equipped with a quad-jet dual-stage cryogenic modulator. (B) Detail of the quad-jet dual-stage cryogenic modulator. (C) Detail of the 2ndD oven with the coiled 2D column.

Advantages of GC×GC (Hajslova and Cajka, 2009)

1. High peak capacity

The peak capacity is a maximal number of chromatographic peaks that can be placed into the chromatogram. The total peak capacity in GC×GC is significantly enhanced in the sample transfer between the two columns. Therefore, GC×GC can overcome the limitation of the conventional GC to gain higher peak capacity.

2. Enhanced sensitivity

Compared to 1D-GC separation, GC×GC system is obtained the improvement of detection limits. GC×GC collected the peak in the modulation before send to the part of the second column (following fast chromatography avoids band broadening of focused peaks). Morover, in the GC×GC system, the signal to noise ratio is also improved.

3. Structured chromatograms

Separation mechanisms in GC×GC occur in both columns. Characteristic patterns are obtained from the reconstructed 2D contour plots. Overall volatile compounds differ in their volatility are ordered along the first-dimension axis, whereas the compounds differing by polarity are spread along the second-dimension axis.

4. Improved identification of unknowns

Non-target screening allows obtaining of overview of the sample components from peak finding and deconvolution, library searching and further post-processing by automated data processing.

MATERIALS AND METHODS

Materials

1. Samples

1.1 Thai white rice (Sao Hai), Tesco Brand (Asia Interrice Co., Ltd., Chacherngsao, Thailand)

1.2 Soybean oil, Morakot brand (Morakot Industry Co. Ltd., Samutprakarn Thailand)

2. Chemical Reagents

2.1 Diethyl ether (anhydrous, 99.8%) (Aldrich Chemical Co. Ltd., USA)

2.2 *n*-Alkane standards (C5-C30) (Aldrich Chemical Co. Ltd., USA)

2.3 Internal standard; 2-Methyl-3-heptanone (Aldrich Chemical Co. Ltd., USA)

2.4 Sodium sulfate anhydrous (Fisher Scientific, UK)

2.5 Deodorized distilled water

2.6 Authentic standard compounds

2.6.1 2,3-Dimethyl-5-ethylpyrazine (Tokyo Chemical Industry Co. Ltd., Japan)

2.6.2 2-Pentyl-furan (Aldrich Chemical Co. Ltd., USA)

2.6.3 2,3-Dimethylpyrazine (Aldrich Chemical Co. Ltd., USA)

2.6.4 Nonanal (Aldrich Chemical Co. Ltd., USA)

2.7 Nitrogen gas (ultra high purity, Linde (Thailand) Co. Ltd., Thailand)

2.8 Helium gas (ultra high purity, Linde (Thailand) Co. Ltd., Thailand)

2.9 Hydrogen (high purity, Linde (Thailand) Co. Ltd., Thailand)

2.10 Air zero (high purity, Linde (Thailand) Co. Ltd., Thailand)

2.11 Liquid nitrogen (Bangkok Industrial Gas Co. Ltd., Thailand)

3. Equipments and Instruments

3.1 Rice cooker (CR-100T, Otto King Glass Co. Ltd., Bangkok, Thailand)

3.2 Iron wok (round bottom)

3.3 Shaker (MS-OR, Major science, Taipei, Taiwan)

3.5 Hot air oven (Memmert, Schwabach, Germany)

3.6 Hot plate stirrer (SS10, Stuart Scientific, Essex, UK)

3.7 Thermocouple Sheath T/C Type K Single Element 1100 °C, 2 m probe length (Engineering system analyzer Inc.,Bangkok, Thailand)

3.8 Balance 2 decimal (ED3202S, Satorius, Goettingen, Germany)

3.9 Balance 4 decimal (ED224S, Satorius, Goettingen, Germany)

3.10 Vigreux column size30 cm x 2.5 cm i.d. (SP Glass, Bangkok, Thailand)

3.10 Freezer -40°C (MDF-435. Sanyo, Osaka, Japan)

3.11 High vacuum equipment (Edwards, West Sussex, UK) consisted of Diffstak® MK2 Diffusion Pump, AV10K Air Admittance Valve)

3.12 Refrigerated circulator (CoolAce Ca-110, Dynamic Corporation, USA)

3.13 Gas chromatography 6890 (Agilent Technologies Inc., Califonia, USA) - mass selective detector (MSD 5973, Agilent Technologies Inc., Califonia, USA)

3.14 Gas chromatography 7890 (Agilent Technologies Inc., Califonia, USA) – olfactory detection port (DATU technology transfer, New York, USA)

3.15 Gas chromatography 6890N (Agilent Technologies Inc., Shanghai, China) equipped with a FID, and thermal desorber (QUI-0002 version 5.1, Unity Markes International Limited, Rhondda Cynon Taf, UK)

3.16 Gas chromatography 7890 (Agilent Technologies, Califonia, USA) equipped with a Pegasus® 4D GC×GC-TOFMS (LECO, Michigan, USA) and cryo-genic modulator (quadjet) cooled with liquid nitrogen. The data were processed using LECO Corp. ChromaTOF-GC software v 4.50.8.0

3.17 Glasswares; prior to use, all glasswares were baked at 120 °C for at least 2 h.

Methods

Experiment 1: Comparison of volatile compounds and aroma attributes between the stir fried rice and the flash fried rice

In order to investigate the unique aroma in the flash fried rice, descriptive analysis and gas chromatography-mass selective detector were performed to compare the aroma attributes and the volatile compounds between the two samples and to reveal that which compounds and aroma attribute made flash fried rice differ from stir fried rice.

1. Sample preparation

Steamed rice – six hundred grams of polished rice, white rice (Sao Hai) (Tesco Brand, Tesco, Thailand) was steamed with 650 ml odor-free water (boiled distilled water until volume were 1/2) using a rice cooker.

Stir fried rice (SFR) – An iron wok was heated until generated a little smoke. Thirty milliliters of soybean oil were added into an iron wok. The soybean oil temperature was measured by using the thermocouple sheath T/C type K until 200 ^{o}C. After that, two hundred grams of steamed rice was added and stir-fried in an iron wok for 1 min.

Flash fried rice (FFR) −An iron wok was heated until generated a little smoke. Thirty milliliters of soybean oil were added into an iron wok. The soybean oil temperature was measured by using the thermocouple sheath T/C type K until 320 ^{o}C (a little fire were in an iron wok). After that, two hundred grams of steamed rice was added and flash-fried in an iron wok for 15 s.

SFR and FFR were separately collected in a glass container before evaluated their aroma attributes and analyzed their volatile compounds.

2. Sensory evaluation

The descriptive analysis of the aroma was performed using eight trained panelists from Kasetsart University Sensory and Consumer Research (KUSCR) center. In this study, a total of four 2-h sessions were arranged to 1. develop aroma attributes, 2. define aroma attributes, 3. select reference standard and their intensities, and 4. individually train the panelist on aroma attributes. After the final session, panelists had agreed on the list of twelve defined terms, the appropriate reference standards and their intensities on the 15-cm intensity line scale (Table 4).

Twenty grams of the SFR and FFR were served to each panelist in the ceramic cups with the ceramic covers at 70^{o}C. During the evaluation, the covers were 1/3 opened, and the samples were only sniffed by the panelists. Aroma intensities were scored on a 15 cm intensity line scale with 0 meaning “none” and 15 meaning “extremely”.

Table 4 Aroma attributes: definitions and corresponding reference standards used in the trained panel evaluation of FFR and SFR samples.

Aroma attribute	Definition	Reference	Reference preparation	Reference intensity
Heated oil	Perception of heated oil associated with brown, cooked, nutty, stale and light acrid.	Heated oil	Heated rice bran oil (King brand) by microwave oven for 4 min.	7
Smoky	Perception of any type of smoke aroma.	Smoke H	¼ tea spoon of liquid smoke with hickory barbecue (Tone's brand) in 500 ml of water.	7
		Smoke L	100 ml of smoke high in 100 ml water.	5
Acrid	The aroma associated with sharp, bitter, stinging, burnt such as burnt rubber or oil.	Acrid	1 drop of liquid smoke with hickory barbecue (Tone's brand) on cotton ball.	7
Wok	The aroma associated with smoke, dried, acrid, mild burnt, briny and heated oil such as flash fried rice or noodle in iron wok.	-	-	-
Animal fat	The aroma associated with animal fat, soup, brown, meaty, sweet aromatic and briny.	Animal fat	Heated 100 g of pork bacon (CP brand) by 1 g of bacon oil. microwave for 1.5 minutes and served	4
Brothy	The aroma associated with meaty, metallic, sweet aromatic and briny in boiled meat soup.	Brothy H	10 ml of condensed soup chicken broth (Campbell brand).	8
		Brothy L	10 ml of condensed soup chicken broth (Campbell brand) in 100 ml of water.	4
Dark brown	The aroma associated with mild burnt perceived by smell.	Dark brown H	1 g of chocolate syrup (Hershey brand).	7

Table 4 (Continued)

Aroma attribute	Definition	Reference	Reference preparation	Reference intensity
		Dark brown L	5 g of chocolate syrup (Hershey brand) in 200 ml of water.	3
Burnt	Perception of any type of burnt aroma.	Burnt H	1 g of roasted coffee (Nescafe’ red cup espresso).	8
		Burnt L	0.5 g of roasted coffee (Nescafe’ red cup espresso) in 300 ml of water.	5.5
Rancid	The aroma associated with extremely oxidized fats or oils perceived by smell.	Rancid	100 ml of Rice bran oil (King brand) by microwave oven for 3 min.	2
Briny	The aroma associated with salt perceived by smell.	Briny H	1 g of Maggi seasoning naturally fermented in 200 ml of water.	6
		Briny L	0.4 g of Maggi seasoning naturally fermented in 200 ml of water.	3.5
Sweet aromatics	The aroma associated with sugar perceived by smell.	Sweet aromatics	20 g of brown sugar (Mitr phol brand) in 200 ml of water.	3
Dried	The aroma associated with dried product perceived by smell such as soy bean powder.	Dried	1 g of soy bean powder (Doi kham brand).	6

3. Isolation of volatile compounds

Volatile compounds were extracted from 480 g of samples using 500 ml diethyl ether. Twelve microliters of 2-methyl-3-heptanone (12.02 mg/ml of diethyl ether) was added as an internal standard. The mixtures were shaken at 150 rpm for 30 min at ambient temperature. The extraction was repeated twice. The three portions of the extracts were combined and concentrated to 50 ml using a vigreux column (30 cm x 1 cm i.d.) in a 40 °C water bath. Volatile components were separated from the oil matrix by using high vacuum distillation at 10^{-5} Torr.

The concentrated extracts were further concentrated to 10 ml under mild nitrogen stream. The extracts were dried and filtered by passing through anhydrous Na_2SO_4 and glass wool. The sample was further concentrated to 0.5 ml under mild nitrogen stream. The extracts were kept in amber bottle at −40 °C before analyzed their volatile compounds.

4. Gas chromatography-mass selective detector (GC-MS)

Volatile compounds from SFR and FFR were analyzed using an HP 6890 gas chromatograph equipped with an HP 5973 mass selective detector (Agilent Technologies, USA). Samples were injected by the cool on-column method (+3 °C temperature tracking mode) into the capillary columns, DB-5 (30m x 0.25m x 0.25µm film thickness) and FFAP (30m x 0.25m x 0.25µm film thickness). The oven temperature program used with DB-5 column started at 35 °C, held for 1 min, increased to 220 °C at the rate of 10°C/min and then held for 15 min.

The oven temperature program for the FFAP column started at 45 °C, held for 1 min, increased to 220 °C at the rate of 5 °C/min and then held for 10 min. Helium was used as a carrier gas at the constant flow rate of 2.2 ml/min. The ionization energy level was 70 eV. Volatile compounds were identified by comparison of mass spectrum data and retention indices (RI) with NIST mass spectrum search 2.0. RIs were calculated using *n*-alkanes series (C_5-C_{30}). Relative concentrations of volatile compounds were calculated from total ion areas relative to those of the internal standard (2-methyl-3-heptanone).

Experiment 2: Identification of aroma active compounds in the flash fried rice

In this experiment, we focused on the identification of aroma active compounds in FFR sample. Some aroma active compounds that generated wok flavor in the FFR sample could not identified by GC-MS (Piyachaiseth, 2010), therefore in this study, gas chromatography-olfactometry and two dimensional gas chromatography-time of flight mass spectrometry were performed to identify those aroma active compounds in FFR.

Sample preparation and isolation of volatile compounds in the FFR and SFR sample followed the experiment 1 issue 1 and 3. Aroma active compounds in both samples were analyzed by using gas chromatography-olfactometry. Aroma active compounds in the SFR were identified by using the retention indices data from GC-MS in experiment 1. While only aroma active compounds in the FFR sample were also analyzed by two dimensional gas chromatography-time of flight mass spectrometry to increase the efficiency of identification of the wok flavor compounds.

1. Gas chromatography-olfactometry

The flavor dilution (FD) factors of the aroma active compounds in FFR and SFR sample were determined by Aroma Extraction Dilution Analysis (AEDA) using a gas chromatography-olfactometry (GC-O). The FFR and SFR extracts from experiment 1 were diluted sequentially with diethyl ether at the ratio of 1:3, 1:9, 1:27, 1:81, and 1:243.

The determination of FD factors was done by two trained panelists in duplicate. The GC-O system consisted of an Agilent 7890 GC (Agilent Technologies, Palo Alto, CA, USA) equipped with flame ionization detector at 250 °C and a sniffing port connected with steam generator at 70 °C. The extract sample was cool on-column injected (+3 °C temperature tracking mode) into a FFAP column (15 m x 0.32 m x 0.25 mm film thickness) and DB-5 column (15 m x 0.32 m x 0.25 mm film thickness). The oven temperature program was used as previously described in experiment 1 issue 4.

2. Two dimensional gas chromatography-time of flight mass spectrometry (GC×GC-TOFMS)

The volatile compounds were separated in a GC×GC/TOFMS system consists of an Agilent© 7890 Gas chromatograph (Agilent Technologies, Palo Alto, CA, USA) equipped with a Pegasus® 4D GC×GC-TOFMS (LECO, St. Joseph, MI, USA) and cryogenic modulator (quadjet) cooled with liquid nitrogen. The data were processed using LECO Corp. ChromaTOF-GC software v 4.50.8.0 optimized for GC×GC. The column set for GC×GC analysis consisted of two columns. The primary column had dimensions of 30m×0.25mm×0.25 µm film thickness Stabilwax® (Restek, Bellefonts, PA, USA), and the second column had dimensions of 1m×0.18mm×0.18µm film thickness Rxi®-5SilMS (Restek, Bellefonts, PA, USA). The injector temperature was 250 °C using splitless mode.

The primary oven temperature program was started at 45 °C, held for 1 min, increased to 220 °C at the rate of 5 °C/min and then held for 10 min. The secondary oven temperature was increased 10 °C over the primary oven temperature; the modulator temperature offset was 45°C; the second dimension separation time was 5 s, the hot pulse time was 0.6 s and the cool time between stages was 1.9 s. Helium was used as the carrier gas with a constant flow rate of 1 ml/min and an injection volume of 1 µl. The mass spectrometer was operated at an acquisition rate of 100 spectra/s. No mass spectra were collected during the solvent delay for the first 4 min of each run. The transfer line temperature was 250 °C, and the ion source temperature was 200 °C. The detector voltage was 1450V, and the electron energy was 70V. Mass spectra were collected from 35–300 m/z. Mass spectra were probability matched to the NIST Mass Spectral Search Program version 2.0.

3. Identification of aroma active compounds

For positive identification, retention indices, mass spectra, and odor properties of unknowns were compared with those of authentic standard compounds analyzed under identical conditions. Tentative identifications were based on comparing mass spectra of unknown compounds with those in the National Institute of Standards and Technology mass spectral database or by matching the retention indices values and odor properties of unknowns against those of authentic standards. For retention indices calculation, a series of *n*-alkanes (C10-C30 and C6-C25 for polar and non-polar column, respectively) were analyzed under the same identical conditions. The retention index (RI) was calculated as shown in the Appendix.

Statistical Analysis

Each experiment was carried out in triplicate and measured sample in duplicates. Paired *t*-test was performed to determine the differences in aroma attributes between FFR and SFR samples at 5% significance level. Principal component analysis (PCA) of mean rating for each sensory attribute was used to show the relationships among variables and samples. PCA was performed using the XLSTAT® software version 2006.

Place and duration

1. Places

Department of Food Science and Technology, Faculty of Agro-Industry, Kasetsart University, Bangkok, Thailand.

2. Duration

March 2010 - November 2012

RESULTS AND DISCUSSIONS

Experiment 1: Comparison of volatile compounds and aroma attributes between the stir fried rice and the flash fried rice

1. Sensory profiles of SFR and FFR

The descriptive analysis of the fried rice samples resulted in significant differences observed between the stir-frying and flash-frying. Aroma attributes from the FFR and the SFR were described by the trained panelists using twelve aroma attributes (Table 5). The aroma profiles of FFR and SFR based on the descriptive sensory analysis are shown in Figure 17.

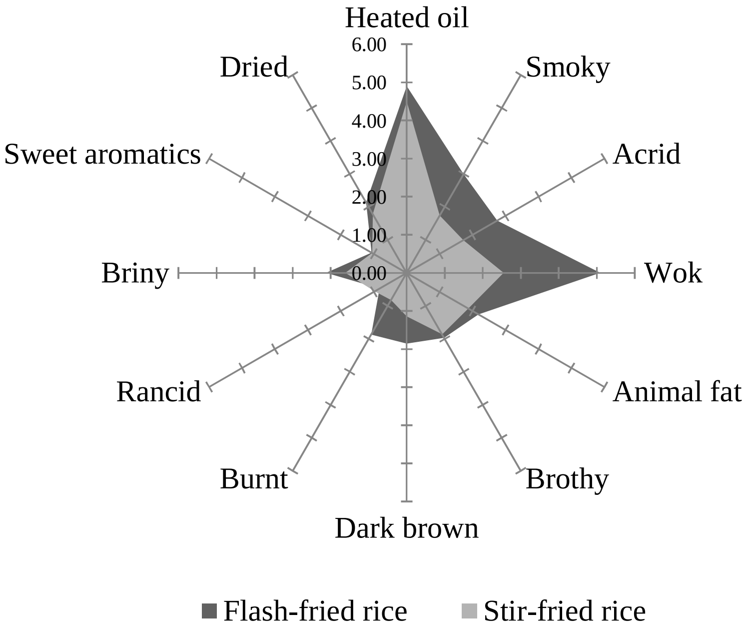


Figure 17 The aroma profiles of flash fried rice and stir fried rice based on the descriptive sensory analysis.

Mean rating and *p*-value for aroma attributes in FFR and SFR are shown in Table 5. The results show that the FFR had higher intensity on aroma attributes namely, heated oil, smoky, acrid, wok, animal fat, dark brown, burnt, briny and dried aroma than those of the SFR sample ($p \leq 0.05$). The brothy, rancid and sweet aromatic aromas had no significantly different between both fried rice samples ($p > 0.05$).

Table 5 Mean rating ± standard deviation (*n*=24) and *p*-value for aroma attribute in FFR and SFR.

Aroma attributes	FFR	SFR	*p*-value
1. Heated oil	4.90 ± 0.64	4.50 ± 0.51	0.00*
2. Smoky	2.99 ± 0.54	1.74 ± 0.48	0.00*
3. Acrid	2.75 ± 0.49	1.72 ± 0.61	0.00*
4. Wok	5.09 ±0.68	2.55 ± 0.65	0.00*
5. Animal fat	2.18 ± 0.37	1.87 ± 0.43	0.00*
6. Brothy	1.97 ± 0.36	1.86 ± 0.43	0.20
7. Dark brown	1.85 ± 0.38	1.15 ± 0.45	0.00*
8. Burnt	1.87 ± 0.76	0.82 ± 0.57	0.00*
9. Rancid	0.83 ± 0.32	0.97 ± 0.32	0.09
10. Briny	2.12 ± 0.40	1.61 ± 0.46	0.00*
11. Sweet aromatics	1.06 ± 0.18	1.05 ± 0.15	0.77
12. Dried	2.14 ± 0.22	1.79 ± 0.37	0.00*

The *p*-value followed by * are significantly different at $p \leq 0.05$.

2. Principal component analysis (PCA) of fried rice samples

The principal component analysis (PCA) was applied to the twelve sensory attributes to investigate and interpret the data structure. The results showed that the dimensionality of the data was reduced from twelve variables to two uncorrelated components, which explained a total of 88.98% of the variation. The score plot (the map of fried rice sample scores) and the loading plot (the map of sensory attribute loadings) on the first two principal components are shown in Figures 18A and 18B, respectively. The score plot could be used to detect the sample patterns, grouping, similarities or differences, while the loading plots could be used to show how much each variable contributed to the meaningful variations in the data and interpret variable relationships (Rosenfeld and Nes, 2000).

PCA was used to show the relative locations of the samples with respect to each other and their characterizing attributes (Meilgaard *et al*., 1999). The explained variance on each attribute in FFR and SFR is shown in Table 6. PC1 accounted for 72.51% of the variation and highly correlated to the aroma namely, heated oil, smoky, acrid, wok, animal fat, dark brown, burnt, briny and dried. PC2 explained the additional 16.47% of the variation that highly correlated to the aroma namely, rancid, brothy and sweet aromatic.

Figure 18A and 18B also shows the same variations on PC1 and PC2 that respectively explained 72.51% and 16.47% of the total variance. Figure 18A shows that SFR samples and FFR samples differ in this plot. There was a clear distinction of the scores from the two types of fried rice on PC1. The FFR samples were more shifted towards the higher values of PC1 than the SFR samples.

From Figure 18B, the aroma attributes was reduced from twelve variables to two components. The aroma attributes namely, heated oil, smoky, acrid, wok, animal fat, dark brown, burnt, briny and dried were mainly represented by the PC1. The aroma attributes namely, rancid, brothy and sweet aromatic were represented by the PC2. From Figures 18A and 18B, the results indicated that FFR could be separated from SFR by PC1. FFR was best described by heated oil, smoky, acrid, wok, animal fat, dark brown, burnt, briny and dried notes.

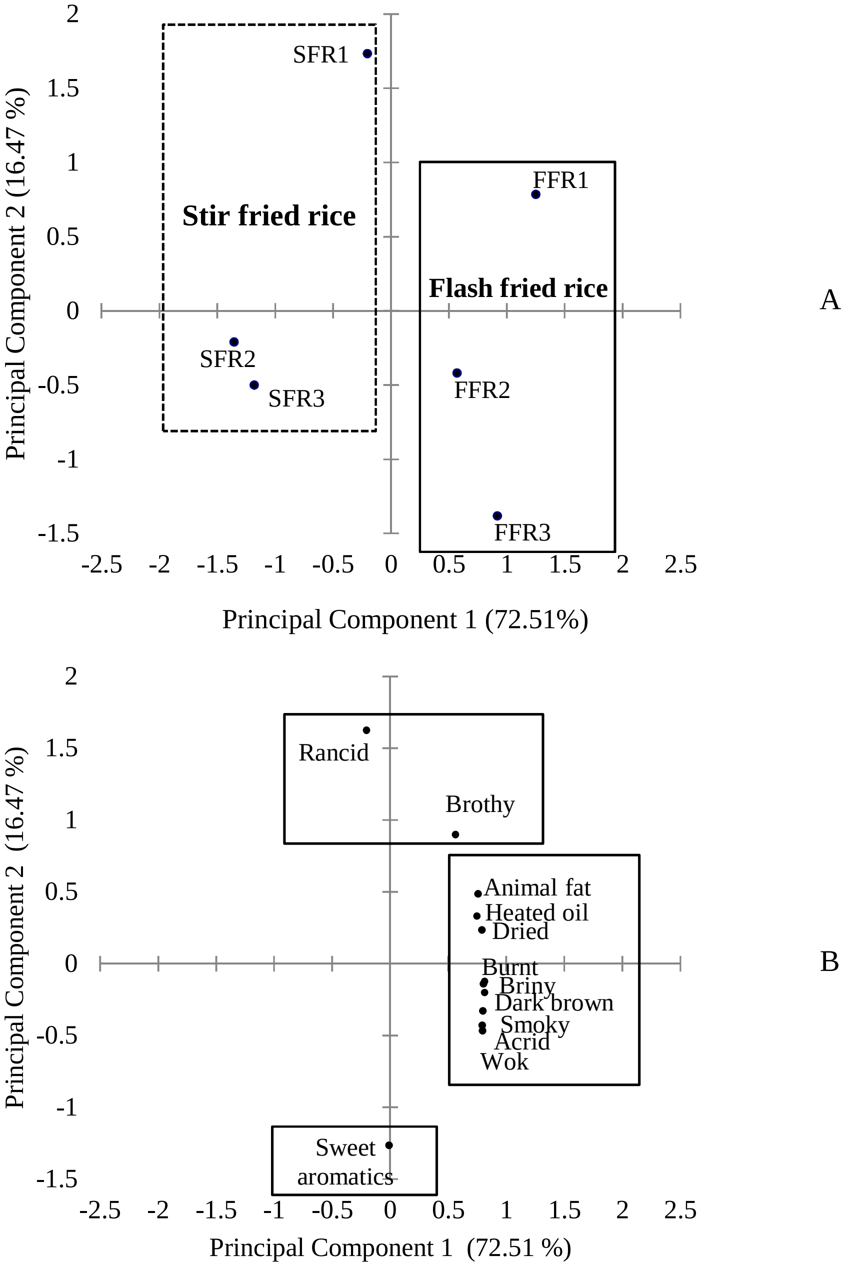


Figure 18 (A) PCA score plot of the 6 fried rice samples performed by sensory data. FFR1-FFR3 were flash fried rice samples and SFR1-SFR3 were stir fried rice samples. (B) Sensory map of the first two principal components for describing twelve sensory attributes of fried rice.

Table 6 Explained variance for the first two principal components (PCs) from the principal component analysis (PCA).

Attributes	Explained variance (%)	
	Principal component 1	Principal component 2
1.Heated oil	9.313	1.802
2.Smoky	10.670	1.815
3.Acrid	10.491	3.074
4.Wok	10.564	3.653
5.Animal fat	9.589	3.892
6.Brothy	5.290	13.306
7.Dark brown	10.782	0.333
8.Burnt	11.066	0.260
9.Rancid	0.702	43.658
10.Briny	11.083	0.687
11.Sweet aromatics	0.002	26.613
12.Dried	10.447	0.907

3. Volatile compounds in stir-fried rice and flash-fried rice by using gas chromatography-mass selective detector (GC/MS)

The difference in cooking temperatures caused the difference in volatile compounds of the fried rice samples. FFR allowed fire to contact the surface of steamed rice and caused the soybean oil to flame for 15 s at the flash point of soybean oil, 328°C (Pryde, 1980). In SFR, the steamed rice was stir fried with soybean oil at the temperature approximately 200°C for 1 min.

Ranking of the chemical classes in the FFR following their relative concentration (%) was as follows: acid 47.51%, aldehydes 19.37%, alcohols 10.54%, furans 9.21%, esters 6.30%, hydrocarbons 4.06%, ketones 1.65%, pyrazines 0.42%, lactones 0.4%, pyrans 0.39% and phenols 0.16%. Meanwhile, the stir fried rice samples had acids 47.35%, aldehydes 31.35%, alcohols 6.56%, esters 6.49%, hydrocarbons 4.62%, furans 1.52%, ketones 1.02%, lactones 0.89% and pyrans 0.20% (Table 7).

A total of 91compounds were identified in FFR and SFR as shown in Table 8. The identified compounds consisted of 17 acids, 14 alcohols, 17 aldehydes, 6 esters, 10 hydrocarbons, 5 ketones, 1 phenol, 13 furans, 2 lactones, 2 pyrans and 2 pyrazines.

There were pyrazines and phenols that were detected only in the FFR samples. Pyrazines are important contributors to the toasted, roasted, nutty and burnt notes (Adams *et al.*, 2002). There are several precursors and pathways for pyrazine formations. The α-amino acid carbonyls, that formed from the reactions between dicarbonyl compounds and amino acids during Strecker degradation, are generally considered to be the precursors of pyrazines (Fisher and Scott, 1997; Lasekan, 2012).

Phenols can be formed from the degradation of chlorogenic acid and decarboxylation of phenolic carboxylic acid (Tressl *et al*., 1978). They had smoky, burnt and spicy characteristics (Sanz *et al*., 2001).

Table 7 Number of volatile compounds and total relative concentrations (%) in the flash-fried rice (FFR) and the stir-fried rice (SFR).

Compound group	**FFR**		**SFR**	
	Number of compound	**Relative concentration (%)**	**Number of compound**	**Relative concentration (%)**
Acids	13	47.51	15	47.35
Alcohols	9	10.54	11	6.56
Aldehydes	14	19.37	17	31.35
Esters	4	6.30	6	6.49
Hydrocarbons	10	4.06	7	4.62
Ketones	3	1.65	5	1.02
Phenols	1	0.16	0	0.00
Furans	13	9.21	5	1.52
Lactones	1	0.40	2	0.89
Pyrans	2	0.39	1	0.20
Pyrazines	2	0.42	0	0.00
Total	**72**	**100.00**	**69**	**100.00**

Table 8 Relative concentration of volatile compounds in the flash-fried rice (FFR) and the stir-fried rice (SFR).

Volatile compound	Odor Description[a]	RI		Relative concentration (ng/g)[b]	
		DB5-MS	FFAP	FFR	SFR
Acids					
acetic acid	strong, pungent, sour, vinegar odor	654	1436	3653.52	3328.68
propanoic acid	pungent, sour milk odor; sour milk	718	1538	343.37	170.63
2-methylpropanoic acid	sour cheesy odor; In dilution a fruity odor and taste	n.a.	1564	89.90	101.14
butanoic acid	strong, cheese, butter-like, sour, rancid	n.a.	1618	212.57	181.70
3-methyl-butanoic acid	−	n.a.	1660	167.89	172.45
isovaleric acid	sour chessy odor; in dilution a fruity odor	843	n.a.		117.38
2-methylbutanoic acid	−	855	n.a.	n.a.	29.94
pentanoic acid	powerful, "sweaty" cheese-like odor; fruity in dilution	897	1723	278.02	299.85
4-pentanoic acid	−	n.a.	1786	28.09	0
hexanoic acid	heavy, fatty, cheesey odor	990	1816	1144.85	1437.46
hexanoic acid, anhydride	−	n.a.	1645	49.47	34.34
heptanoic acid	fatty, sour-sweat-like, rancid odor	1073	1908	55.81	106.90
octanoic acid	fatty, rancid, sweaty odor	1167	2038	43.04	118.27
nonanoic acid	mild, nut-like, fatty odor	n.a.	2143	0	45.69
(*E*)-2-octenoic acid	−	n.a.	2160	0	10.38
phthalic acid, isobutyl 2-pentyl ester	−	n.a.	2497	67.54	0
hexadecanoic acid	almost odorless; bland waxy taste	1958	2834	235.82	191.41

Table 8 (Continued)

Volatile compound	**Odor Description**[a]	**RI**		**Relative concentration (ng/g)**[b]	
		DB5-MS	**FFAP**	**FFR**	**SFR**
Alcohols					
1-butanol	breathtaking, winey, fuel oil-like	668	1131	30.79	26.85
1-pentanol	alcoholic-breathtaking, fusel-like	n.a.	1240	220.49	259.41
1-penten-3-ol	pungent, grassy, alliaceous-like odor	685	1147	0	38.13
(*Z*)-2-penten-1-ol	−	n.a.	1305	0	19.87
1,2-propanediol	odorless when pure with a weak mild	744	n.a.	15.73	0
1,3-butanediol	solvent of flavor	791	n.a.	120.72	71.24
2,3-butanediol	weak	803	1574	135.73	41.31
1-hexanol	chemical, winey, slight fatty-fruity odor	869	1343	n.a.	34.59
(*Z*)-3-nonen-2-ol	−	n.a.	1016	49.00	0
benzenemethanol	faint, sweet, fruity aroma	1033	1841	0	55.33
1-(2-butoxyethoxy)-ethanol	−	n.a.	1765	330.32	265.93
2-(2-butoxyethoxy)-ethalnol	−	1186	n.a.	486.4	0
(*E*)-2-methyl-cyclopentanol	−	n.a.	1328	23.46	14.76
1-octanol	orange-rose, waxy, sweet odor	n.a.	1551	0	31.21
Aldehydes					
N-pentanal	strong, acrid, pungent odor; chocolate and nut-like below 10 ppm	n.a.	<1000	78.77	94.39

Table 8 (Continued)

Volatile compound	**Odor Description**[a]	**RI**		**Relative concentration (ng/g)**[b]	
		DB5-MS	**FFAP**	**FFR**	**SFR**
N-hexanal	strong, penetrating, fatty-green, grassy unripe fruit odor	798	1062	169.02	243.52
N-heptanal	fatty, in dilution sweet, fruity, nutty, fatty-cognac like	902	1173	191.03	115.29
(*E*)-2-hexenal	green, fruity, fresh, apple and woody with leafy and grassy notes	n.a.	1212	27.83	24.00
(*Z*)-2-heptanal	−	957	n.a.	445.3	502.38
benzaldehyde	odor of bitter almond oil	962	1514	126.56	58.50
octanal	fatty-fruity odor; sweet	1003	1277	60.75	43.72
(*Z*)-2-heptenal	−	n.a.	1312	502.38	445.53
(*E*,*E*)-2,4-heptadienal	fatty, green, with and oily, greasy	1011	1457	270.59	381.39
(*E*)-2-octenal	peculiar fatty, green-grassyodor	1059	1417	162.96	122.21
nonanal	fatty-floral-rose, waxy odor	1105	1381	175.95	171.61
decanal	strong, sweet, orange peel odor	1206	1490	0	14.70
(*E*)-2-decanal	waxy, fatty, earthy, coriander, mushroom, green, pork fat note	1261	1628	77.83	151.38
2,4-decadienal	strong, deep fat, fatty-citrus note odor in dilution	1294	1736	0	76.53

Table 8 (Continued)

Volatile compound	Odor Description[a]	RI		Relative concentration (ng/g)[b]	
		DB5-MS	**FFAP**	**FFR**	**SFR**
(*E*,*E*)-2,4-decadienal	strong, deep fat, fatty-citrus note	1318	1740, 1778	277.80	1445.75
undecanal	sweet, fatty, waxy-floral-citrus odor	1368	n.a.	0	99.71
2-undecenal	Fresh, sweet-citrus, orange-lemon peel	n.a.	1727	29.62	113.38
Ester					
ethyl acetate	ethereal, sharp, wine-brandy like odor	614	<1000	146.20	216.59
2-ethylhexyl acetate	−	n.a.	<1000	0	27.87
butoxyethoxyethyl acetate	−	1358	n.a.	521.68	480.55
1,2-benzenedicarboxylic acid, *bis*-(2-methylpropyl)-ester	−	1860	n.a.	166.84	47.94
2,4-ditert-butylphenyl 5-hydroxypentanoate	−	n.a.	2276	10.34	19.64
isobutyl 2-pentyl ester	−	n.a.	2499	0	56.59
Hydrocarbons					
2-methyl-2-butene	−	<600	n.a.	86.96	180.78
2,3-dimethlbutane	−	<600	n.a.	237.95	289.03
1,3-octadiene	−	825	<1000	41.04	0
1-octene	hydrocarbon, lifting	n.a.	<1000	24.45	0

Table 8 (Continued)

Volatile compound	Odor Description[a]	RI		Relative concentration (ng/g)[b]	
		DB5-MS	FFAP	FFR	SFR
2-octene	−	n.a.	<1000	0	25.48
(*Z*)-2-octene	−	n.a.	<1000	39.26	n.a.
butyl-cyclopentane	−	n.a.	<1000	21.32	32.19
1-undecene	−	n.a.	1126	28.19	0
5-undecene	−	n.a.	1135	16.83	0
limonene	fresh, sweet, hydrocarbon and orange citrus odor	n.a.	1178	31.46	11.16
vanillin	powerful, creamy, vanilla-like odor	1395	2524	16.37	65.96
Ketones					
3-hydroxy-2-butanone	creamy-buttery, yogurt-like odor	709	1275	129.44	72.60
3-ethylcyclopentanone	−	958	n.a.	0	18.21
2-hydroxy-2-cyclopenten-1-one	−	n.a.	1180	0	8.45
2-heptanone	fruity, cheese, cinnamon odor; in dilution-blue cheese-fruity	n.a.	1170	36.19	23.05
1,2-cyclopentanedione	strong maple, burnt-sugar, caramel odor	n.a.	1750	55.92	0
1-(2-hydroxy-5-methylphenyl)-ethanone	−	n.a.	2170	0	11.60
Phenol					
Phenol	phenolic medicinal odor	n.a.	1981	21.99	0

Table 8 (Continued)

Volatile compound	Odor Description[a]	RI		Relative concentration (ng/g)[b]	
		DB5-MS	FFAP	FFR	SFR
Furans					
2-furancarboxaldehyde	−	831	n.a.	246.98	9.02
2-furanmethanol	weak, fermented, burnt-sugar, creamy, caramellic notes	852	1656	141.56	28.92
2-(5H)-furanone	−	906	1735	104.01	0
5-methyl-2-furancarboxaldehyde	sweet spicy, bready, caramellic odor	960	1523	130.26	0
2-butyltetrahydro-furan	−	n.a.	1143	20.86	0
2-pentyl-furan	earthy, green, metallic odor	n.a.	1225	63.43	67.70
furfural	sweet, cereal, bread-like, yeasty, caramellic, spicy notes	n.a.	1462	295.65	0
5-ethyldihydro-2(3H)-furanone	−	n.a.	1683	58.94	49.85
2-furancarboxylic acid, methyl ester	−	n.a.	1975	42.72	0
dihydro-3-hydroxy-4,4-dimethyl-2(3*H*)-furanone	−	n.a.	2010	45.19	43.57
3-furancarboxylic acid	−	n.a.	2397	42.06	0
5-hydroxymethylfurfural	−	n.a.	2472	24.17	0
4-hydroxydihydro-2(3*H*)-furanone	−	n.a.	2566	18.68	0

Table 8 (Continued)

Volatile compound	Odor Description[a]	RI		Relative concentration (ng/g)[b]	
		DB5-MS	FFAP	FFR	SFR
Lactones					
γ-butyrolactone	coconut-butter aroma; creamy	907	1618	53.43	25.60
γ-hexalactone	coumarin-like, sweet odor and taste; creamy note	1050	n.a.	0	91.39
Pyrans					
2,3-dihydro-3,5-dihydroxy-6-methyl-4*H*-pyran-4-one	–	1144	2236	33.79	26.76
2*H*-pyran-2,6(3*H*)-dione	–	n.a.	1968	18.61	0
Pyrazines					
pyrazines	pungent, sweet, corn-like, roasted hazelnuts	733	n.a.	22.36	0
methyl pyrazines	green, nutty, cocoa, musty, potato, fishy-amoniacal notes	828	1255	33.48	0.00

Note [a] Odor description from Flavor-base 2004 (Leffingwell, 2004)

[b] Average relative concentration (n=2) n.a. = not available

Several volatiles in FFR and SFR came from soybean oil. Hydrocarbons, aldehydes and ketones were the result of lipid oxidation and degradation. Products from lipid degradation were mostly formed at temperature ≥ 150°. (van Loon *et al.*, 2005; Kuo *et al*., 1989). Most of alcohols identified were oxidative decomposition of lipids. For example, 1-propanol and 1-butanol could be formed from miristoleic acid, 1-pentanol from linoleic acid, 1-hexanol from palmitoleic acid and oleic acid and 1-octanol from oleic oxidation (Flores *et al.*, 1997). 1-penten-3-ol came from linolenic oxidation and had a penetrating, grassy and ethereal odor (Shahidi *et al.*, 1986).

Aldehydes can be formed by oxidation of unsaturated fatty acids. Hexanal came from linoleic acid oxidative decomposition. Nonanal was from the degradation of oleic acid (van Loon *et al.*, 2005). Aldehydes contribute to the loss of desirable flavor in food because of their high rate of formation during lipid oxidation and their low flavor thresholds. (Flores *et al*., 1997). Drumm and Spanier (1991) reported that unsaturated aldehydes undergo further oxidation to form short chain aldehydes as was occurred with 2,4-decadienal and 2-undecenal.

4. Relationship between aroma attributes and volatiles compounds in FFR

The results from the descriptive sensory analysis revealed that, the flash fried rice showed higher degree of aroma attributes namely, heated oil, smoky, acrid, wok, animal fat, dark brown, burnt, briny and dried aroma which differed from stir fried rice samples ($p<0.05$). The increasing of heptanal, benzaldehyde, octanal and (*E*)-octenal that show fatty, oily characteristic was the main factor that led to the increasing of aroma attributes namely, animal fat and heated oil. Meanwhile, the presence of pyrazines, phenol, furfural, 5-hydroxymethylfurfural, 2-(5*H*)-furanone, 2-methyl-2-furancarboxaldehyde and1,2-cyclopentanedione may contribute to the smoky, acrid, wok, dark brown and burnt aroma.

Experiment 2: Identification of aroma active compounds in the flash fried rice

1. Two dimensional gas chromatography-time of flight mass spectrometry (GC×GC-TOFMS)

In this study, GC×GC-TOFMS also used for the analysis of volatile compounds focused on the identification of aroma active compounds in the flash fried rice sample. GC×GC has been developed for an increasing need of complex sample analysis and to undergo the limitations such as peak capacity, dynamic range, and restricted specificity of conventional GC systems (one dimensional gas chromatography). Mass spectra of volatiles that had similarity more than 80% were probability matched to the NIST Mass Spectral Search Program version 2.0.

The totals of 187 compounds of FFR were identified by GC×GC-TOFMS. The identified compounds consisted of 37 ketones, 32 furans, 30 alcohols, 24aldehydes, 24 hydrocarbons, 18 acids, 4 pyrazines, 3 phenols, 3 pyrans, 2 pyrroles and 10 others as shown in Table 9. Pyrrole, pyridine, thiazole and few others were not detected by GC-MS but were detected by GC×GC-TOFMS. There were 102 compounds identified by GC×GC-TOFMS, that were more than the number of compounds previously identified by GC-MS .

These results indicated that GC×GC-TOFMS had more efficiency of the separation than conventional GC. FFR was a complex sample. The complicated position of each compounds in FFR are shown in Figure 19. Previous study by Piyachaiseth (2010) revealed that many aroma active compounds in FFR were unknown. These unknown compounds were detected by GC-O but not by the conventional GC-MS. Therefore, GC×GC-TOFMS had an important role to help us completely identify the aroma active compounds in this sample.

Table 9 GC×GC-TOFMS identification of volatile compounds in flash fried rice

Volatile compound	**t_{R1}[a], t_{R2}[b] (min)**	**Similarity (%)[c]**	**RI[d]**	**Relative concentration (ng/g)**
Acids				
butanoic acid	7.667 , 1.270	86.10	1165	12.13
acetic acid	14.583 , 0.730	91.20	1447	1226.91
propanoic acid, anhydride	16.000 , 0.970	85.00	1503	8.66
formic acid	16.750 , 0.660	89.60	1533	5.40
propanoic acid	16.750 , 0.750	91.70	1533	220.05
2-methyl-propanoic acid	17.500 , 0.760	89.30	1563	65.92
butanoic acid	18.833 , 0.770	93.50	1618	518.39
4-hydroxy-butanoic acid	18.833 , 0.830	93.10	1618	95.27
2-propenoic acid	19.250 , 0.700	80.00	1636	16.45
3-methyl-butanoic acid	19.833 , 0.760	92.90	1661	69.77
(*Z*)-2-butenoic acid	20.750 , 0.720	87.50	1700	3.31
pentanoic acid	21.333 , 0.800	91.30	1729	528.04
4-pentenoic acid	22.917 , 0.730	93.00	1807	7.73
hexanoic acid	23.667 , 0.830	92.60	1841	464.01
5-hexenoic acid	24.917 , 0.750	85.20	1896	9.41
heptanoic acid	25.833 , 0.810	93.00	1943	50.47

Table 9 (Continued)

Volatile compound	$t_{R1}{}^{a}$(min), $t_{R2}{}^{b}$(s)	Similarity (%)^{c}	RI^{d}	Relative concentration (ng/g)
octanoic acid	27.917 , 0.820	92.50	2052	19.96
hexadecanoic acid	45.333 , 1.400	90.90	>2800	11.51
<u>Alcohols</u>				
Ethanol	3.833 , 0.720	80.20	909	93.79
2-propen-1-ol	6.167 , 0.760	95.30	1095	74.18
2-pentanol	6.250 , 0.890	90.00	1100	23.56
1-butanol	6.750 , 0.840	93.00	1123	73.53
1-penten-3-ol	7.167 , 0.850	91.90	1142	98.18
2-hexanol	8.583 , 0.970	93.10	1206	13.59
1-pentanol	9.333 , 0.920	95.50	1234	225.26
3-heptanol	10.500 , 1.080	90.00	1278	5.15
cyclopentanol	10.583 , 0.880	84.00	1281	12.52
4-penten-1-ol	10.667 , 0.840	84.50	1284	12.06
(*E*)-2-penten-1-ol	10.917 , 0.850	94.20	1293	29.42
2-methyl-cyclopentanol	10.917 , 0.960	92.30	1293	1.42
2-methyl-cyclopentanol	11.583 , 0.930	86.90	1322	14.83
1-hexanol	11.917 , 0.950	91.30	1336	78.63

Table 9 (Continued)

Volatile compound	**t_{R1}[a](min), t_{R2}[b](s)**	**Similarity (%)[c]**	**RI[d]**	**Relative concentration (ng/g)**
3-octanol	13.000 , 1.160	92.60	1382	8.75
cyclohexanol	13.167 , 0.950	91.20	1389	15.73
2-octanol	13.667 , 1.120	93.80	1410	9.50
1-octen-3-ol	14.417 , 1.040	90.90	1440	436.62
3,3-dimethylbutane-2-ol	14.583 , 0.840	80.20	1447	31.69
cyclobut-1-enylmethanol	14.750, 0.790	86.90	1453	5.48
2,4-dimethyl-cyclohexanol	15.167 , 1.050	87.00	1470	23.46
2-ethyl-1-hexanol	15.417 , 1.070	90.40	1480	9.04
4-ethylcyclohexanol	16.667 , 1.060	84.20	1530	32.40
1-octanol	17.000 , 1.060	90.90	1543	16.03
2,3-butanediol	17.583 , 0.750	88.50	1567	309.00
(*E*)-2-octen-1-ol	18.333 , 0.990	85.90	1597	14.23
isopropyl alcohol	19.250 , 0.710	81.90	1636	11.76
2-(2-butoxyethoxy)-ethanol	23.583 , 1.230	94.70	1837	891.22
benzyl alcohol	24.250 , 0.800	93.30	1867	63.18
phenylethyl alcohol	24.917 , 0.850	94.80	1896	12.65

Table 9 (Continued)

Volatile compound	**t_{R1}^{a}(min), t_{R2}^{b}(s)**	**Similarity (%)^{c}**	**RI^{d}**	**Relative concentration (ng/g)**
Aldehydes				
pentanal	3.917 , 0.960	89.50	918	108.40
hexanal	5.667 , 1.150	90.40	1063	606.11
heptanal	7.833 , 1.330	93.50	1173	335.90
(*E*)-2-hexenal	8.583 , 1.100	93.90	1206	30.44
cyclohexanecarboxaldehyde	9.167 , 1.290	85.00	1228	8.21
(*Z*)-4-heptenal	9.250 , 1.170	88.60	1231	4.52
cyclohexanecarboxaldehyde	9.917 , 1.290	85.70	1256	23.84
octanal	10.417 , 1.460	94.40	1275	86.63
(*Z*)-2-heptenal	10.667 , 1.200	86.30	1284	324.39
2-ethenyl-2-butenal	10.917 , 1.060	89.50	1293	23.69
nonanal	13.083 , 1.580	95.10	1386	281.04
(*E*,*E*)-2,4-hexadienal	13.333 , 0.970	89.10	1396	7.62
(*E*)-2-octenal	14.000 , 1.280	91.10	1423	328.02
3-cyclohexene-1-carboxaldehyde	14.583 , 1.140	84.20	1447	8.98
(*E*)-2-nonenal	16.583 , 1.350	94.50	1527	35.36
(*E*,*E*)-2,4-heptadienal	15.583 , 1.060	91.10	1487	246.36

Table 9 (Continued)

Volatile compound	**t_{R1}[a](min), t_{R2}[b](s)**	**Similarity (%)[c]**	**RI[d]**	**Relative concentration (ng/g)**
(*E*,*E*)-2,4-octadienal	17.833 , 1.110	88.80	1577	8.93
(*Z*)-2-decenal	19.083 , 1.410	93.40	1628	53.21
benzeneacetaldehyde	19.167 , 0.950	93.00	1632	232.24
2,4-nonadienal	20.417 , 1.160	90.90	1686	11.17
2-undecenal	21.583 , 1.460	91.70	1742	18.13
(*E*,*E*)-2,4-decadienal	21.833 , 1.240	91.60	1754	343.70
Hydrocarbons				
1,3-octadiene	3.833 , 1.400	95.80	909	28.52
decane	4.167 , 3.170	94.30	945	47.67
1-butyl-cyclopentene	4.750 , 2.190	96.00	1005	67.05
(*E*)-1,3-nonadiene	5.000 , 1.890	89.00	1021	75.82
1,3-(*E*),5-(*Z*)-octatriene	6.000 , 1.440	89.10	1084	34.90
1-nonen-4-yne	6.250 , 1.760	83.60	1100	51.53
ethylbenzene	6.500 , 1.310	94.80	1112	83.15
limonene	8.083 , 2.110	93.80	1184	24.05
propyl-benzene	8.417 , 1.510	95.50	1200	66.67

Table 9 (Continued)

Volatile compound	**t_{R1}^{a}(min), t_{R2}^{b}(s)**	**Similarity (%)^{c}**	**RI^{d}**	**Relative concentration (ng/g)**
Ketones				
2,3-butanedione	3.833 , 0.940	89.80	909	16.95
3-methyl-2-butanone	4.333 , 0.940	86.30	963	29.45
2,3-pentanedione	5.333 , 0.900	91.40	1042	29.51
2-hexanone	5.500 , 1.130	81.50	1053	3.57
4-methyl-3-penten-2-one	6.667 , 1.070	89.60	1119	37.76
3-heptanone	7.083 , 1.330	91.90	1138	18.85
cyclopentanone	7.750 , 1.010	93.30	1169	77.32
2-heptanone	7.750 , 1.280	92.30	1169	27.65
2-methyl-cyclopentanone	8.000 , 1.120	93.70	1181	21.38
3-octanone	9.500 , 1.490	90.00	1241	18.14
acetoin	10.250 , 0.810	91.50	1269	105.34
2-octanone	10.250 , 1.410	94.50	1269	68.95
cyclohexanone	10.417 , 1.110	93.00	1275	48.49
1-hydroxy-2-propanone	10.667 , 0.780	93.10	1284	560.36
1-octen-3-one	10.750 , 1.300	85.80	1288	31.82
2-cyclopenten-1-one	12.083 , 0.900	95.20	1343	99.34

Table 9 (Continued)

Volatile compound	**t_{R1}^{a}(min), t_{R2}^{b}(s)**	**Similarity (%)^{c}**	**RI^{d}**	**Relative concentration (ng/g)**
4-hydroxy-4-methyl-2-pentanone	12.250 , 0.900	87.80	1350	19.46
2-methyl-2-cyclopenten-1-one	12.417 , 1.000	92.60	1357	26.08
1-hydroxy-2-butanone	12.500 , 0.810	91.70	1361	29.53
2-nonanone	12.917 , 1.500	92.40	1378	17.15
2-cyclohexen-1-one	14.083 , 0.990	83.10	1427	34.82
2-decanone	15.500 , 1.600	89.70	1483	13.48
2,5-hexanedione	15.750 , 0.890	81.20	1493	23.76
1-(2-furanyl)-ethanone	15.833 , 0.890	89.30	1497	135.13
benzocyclobuten-1(2*H*)-one	15.833 , 1.020	89.70	1497	7.65
3-nonen-2-one	16.000 , 1.320	86.70	1503	13.14
3-methyl-2-cyclopenten-1-one	16.083 , 0.950	88.60	1507	22.67
2,3-dimethyl-2-cyclopenten-1-one	16.667 , 1.050	82.20	1530	6.54
3,6-heptanedione	17.583 , 0.960	83.70	1567	8.15
4-cyclopentene-1,3-dione	17.833 , 0.810	84.30	1577	4.41
acetophenone	19.250 , 0.980	96.10	1636	13.00
1,4-cyclohex-2-enedione	21.250 , 0.850	85.20	1725	6.06
1,2-cyclopentanedione	21.917 , 0.780	88.70	1758	107. 48

Table 9 (Continued)

Volatile compound	t_{R1}^{a}(min), t_{R2}^{b}(s)	Similarity (%)^{c}	RI^{d}	Relative concentration (ng/g)
2*H*-pyran-2-one	21.917 , 0.810	89.20	1758	17.87
3-methyl-1,2-cyclopentanedione	23.167 , 0.820	93.30	1819	18.23
levoglucosenone	26.500 , 0.810	90.00	1978	6.29
2*H*-Pyran-2,6(3*H*)-dione	26.667 , 0.740	84.30	1987	8.31
Phenols				
phenol	26.917 , 0.730	95.60	2000	99.91
3-methyl-phenol	28.417 , 0.750	92.60	2078	27.78
2,4-*bis*-(1,1-dimethylethyl)-phenol	32.500 , 0.960	91.60	2296	4.23
Furans				
2-propyl-tetrahydrofuran	5.083 , 1.470	86.20	1026	45.85
2-butyl furan	6.667 , 1.410	91.80	1119	15.09
2-butyltetrahydro-furan	7.250 , 1.790	92.70	1146	86.45
2-pentyl-furan	9.000 , 1.600	93.40	1222	183.38
dihydro-2-methyl-3(2*H*)-furanone	9.750 , 0.940	88.90	1250	20.18
2-butyltetrahydro-furan	9.750 , 2.020	88.70	1250	14.37
3-furaldehyde	14.000 , 0.830	92.40	1423	143.71
5-methyl-2(3*H*)-furanone	14.083 , 0.880	93.30	1427	30.53

Table 9 (Continued)

Volatile compound	**t_{R1}[a](min), t_{R2}[b](s)**	**Similarity (%)[c]**	**RI[d]**	**Relative concentration (ng/g)**
2-propyl-furan	14.417 , 0.880	80.40	1440	20.68
furfural	14.833 , 0.860	95.50	1457	343.32
5-methyl-2-furancarboxaldehyde	15.000 , 0.920	87.30	1463	10.44
2-(1-hydroxy-1-methyl-2-oxopropyl)-2,5-dimethyl-3(2*H*)-furanone	15.667 , 0.930	82.40	1490	16.96
5-methyl-2-furancarboxaldehyde	17.500 , 0.920	94.60	1563	332.25
2-methyl-benzofuran	18.000 , 1.120	87.00	1583	2.76
dihydro-5-methyl-2(3*H*)-furanone	18.333 , 0.860	83.30	1597	40.13
2-acetyl-5-methylfuran	18.417 , 0.950	88.20	1600	7.05
dihydro-4-methyl-2(3*H*)-furanone	18.500 , 0.860	86.20	1603	7.97
2,5-dihydro-3,5-dimethyl-2-furanone	19.250 , 0.880	89.40	1636	5.62
2-furanmethanol	19.500 , 0.780	94.30	1646	195.87
5-methyl-2(5*H*)-furanone	19.917 , 0.810	90.70	1664	41.31
5-ethyldihydro-2(3*H*)-furanone	20.500 , 0.910	92.70	1689	197.42
2(*5H*)-furanone	20.833 , 0.780	84.70	1704	226.34
3-methyl-2(5*H*)-furanone	20.833 , 0.830	90.00	1704	22.70
5-methyl-2-furanmethanol	20.917 , 0.790	85.40	1708	4.49

Table 9 (Continued)

Volatile compound	**t_{R1}^{a}(min), t_{R2}^{b}(s)**	**Similarity (%)^{c}**	**RI^{d}**	**Relative concentration (ng/g)**
5-ethyl-2(5*H*)-furanone	21.750 , 0.850	86.10	1750	16.84
3,5,5-trimethyl-2(5*H*)-furanone	22.500 , 0.890	86.00	1788	6.31
dihydro-5-propyl-2(3*H*)-furanone	22.667 , 0.950	88.00	1796	21.66
4-methyl-5*H*-furan-2-one	24.500 , 0.800	92.00	1878	19.73
5-butyldihydro-2(3*H*)-furanone	25.083 , 0.980	91.90	1904	48.37
dihydro-5-pentyl-2(3*H*)-furanone	27.333 , 1.020	89.30	2022	6.98
5-hydroxymethylfurfural	35.667 , 0.750	90.10	2484	23.64
dihydro-4-hydroxy-2(3*H*)-furanone	37.333 , 0.730	91.70	2574	41.52
Pyrans				
tetrahydro-2*H*-pyran-2-one	22.750 , 0.850	92.00	1800	38.35
5,6-dihydro-2*H*-pyran-2-one	23.833 , 0.810	92.60	1848	7.27
maltol	26.000 , 0.820	92.20	1952	5.89
Pyrazines				
pyrazines	8.417 , 0.880	95.10	1200	197.67
methyl-pyrazine	9.750 , 0.980	95.50	1250	112.36
ethyl-pyrazine	11.417 , 1.080	92.30	1314	15.93
2,3-dimethyl-pyrazine	11.750 , 1.070	84.70	1329	5.62
2-methyl-1*H*-Pyrrole	17.083 , 0.780	91.60	1547	12.70

Table 9 (Continued)

Volatile compound	t_{R1}[a](min), t_{R2}[b](s)	Similarity (%)[c]	RI[d]	Relative concentration (ng/g)
Pyrroles				
pyrrole	16.167 , 0.740	94.50	1510	339.22
3,5,5-trimethyl-2(5*H*)-furanone	22.500 , 0.890	86.00	1788	6.31
Others				
pyridine	7.667 , 0.920	93.50	1165	20.05
thiazole	9.333 , 0.860	92.80	1234	14.22
methyl formate	14.500 , 0.740	84.20	1443	1298.79
ethyl acetate	16.333 , 0.680	93.60	1517	946.32
1,2-diacetylhydrazine	18.000 , 0.930	81.30	1583	0.82
benzonitrile	18.250 , 0.900	94.10	1593	14.77
2-methyl-furoate	26.833 , 0.790	88.50	1995	27.11
indole	34.750 , 0.780	94.10	2426	5.56
2,3-dideoxyribonolactone	35.333 , 0.750	93.90	2463	37.10
1,2-benzenedicarboxylic acid, *bis*-(2-methylpropyl)-ester	36.333 , 1.250	91.30	2522	21.70

Note [a] 1st dimension retention time on Stabilwax® column. [b] 2nd dimension retention time on Rxi®-5SilMS column.

[c] Similarity of mass spectral data obtained by GC×GC-TOFMS matching to NIST library.

[d] Retention index calculated from t_{R1} for identified compounds in flash fried rice.

2. Aroma active compounds in stir-fried rice and flash-fried rice

Aroma active compounds of FFR and SFR were studied using the GC-O and the AEDA technique. Aroma active compounds listed in Table 10 were identified and tentatively identified using the GC×GC-TOFMS and the reference data. The identification was performed by using their odor description and the retention index on the Stabilwax® column, comparing its mass spectrum and the odor descriptions with those of the authentic standards. Aroma active compounds in FFR were identified by GC×GC-TOFMS as shown in Figure 19 that used the contour plot.

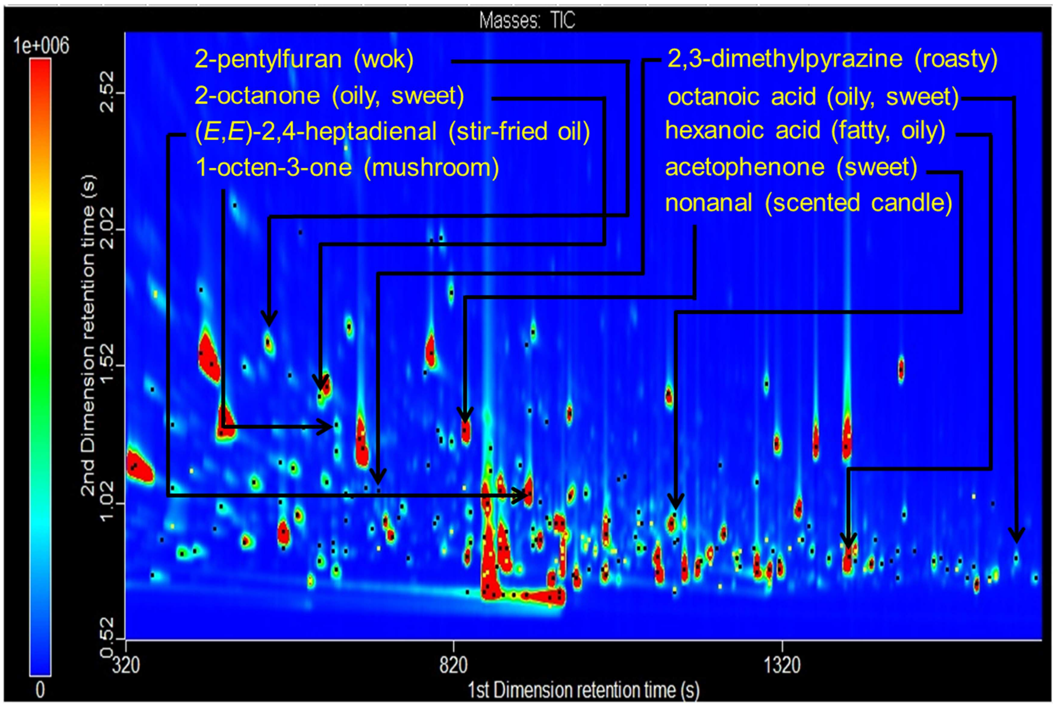


Figure 19 Aroma active compounds in flash-fried rice (FFR) identified by GCxGC contour.

Aroma compounds could be grouped by their odor descriptions to the stir-fried oil, sweet, oily/sweet, fatty, roast, wok (smoke/burnt), mushroom and others descriptions. Most aroma active compounds in FFR and SFR were from degradations of soybean oil that had the stir-fried oil/oily characteristics. Aroma active compounds in FFR and SFR are shown in Table 10. In the typical SFR, the prominent odorants that had the $\log_3$FD factors of 5 were (*E*,*Z*)-2,4-nonadienal (raw white rice) and acetophenone (sweet). Moreover, the SFR also had the prominent compounds that were responsible for the deep-fried aroma as reported by Wagner and Grosch (1997). There were (*E*,*E*)-2,4-heptadienal (stir-fried oil), (*E*,*E*)-2,4-octadienal (cucumber peel) and octanoic acid (oily, sweet) with the $\log_3$FD factor of 4.

On the other hand, wok aroma characteristics in FFR were from the heat-induced condensation between amino acids and sugars through Strecker degradation and sugar thermal breakdown through caramelization that generated pyrazines and furans. There were 2,3-dimethyl-5-ethylpyrazine and 2-pentylfuran that had wok aroma characteristic with $\log_3$FD factors of 4. The other prominent odorants in FFR that had the $\log_3$FD factor of 5 were octanoic acid (oily, sweet) and acetophenone (sweet). 2,3-dimethylpyrazine (roast), (*E*,*E*)-2,4-heptadienal (stir-fried oil), nonanal (scented candle), 2-octanone (oily, sweet), (*E*,*E*)-2,4-hexadienal (fruity), 1-octen-3-one (mushroom) and hexanoic acid (fatty, oily) with the $\log_3$FD factors of 4.

Acetophenone had the highest $\log_3$FD factor in both FFR and SFR. Acetophenone can be formed by the oxidation of ethylbenzene (Arshadia, 2012) and is the precursor of compounds in the aroma resembling almond, cherry, jasmine and strawberry (Dong *et al.*, 2012). In this case, ethylbenzene was also found in FFR and SFR by using GC×GC-TOFMS. The chemical structures of aroma active compounds in FFR are shown in Figure 20.

Table 10 Aroma active compounds in flash fried rice (FFR) and stir fried rice (SFR) samples.

Compound	Odor description[a]	Retention Index		log$_3$FD factor[b]	
		DB-5	FFAP	FFR	SFR
unknown A	salty	n.a.	1018	0	−
n-hexanal[c]	grass	799	1057	3	3
butyl acetate[e]	sweet	n.a.	1077	1	−
2-pentanol[c]	green	n.a.	1110	0	−
1-butanol[c]	rancid	n.a.	1127	0	−
unknown B	oily, rancid	n.a.	1139	0	0
n-heptanal[c]	stir-fried oil	903	1162	0	0
pyrazine[c]	nutty	n.a.	1180	0	0
2-hexenal[c]	oily, rancid	n.a.	1209	2	−
2-pentylfuran[c, std]	wok	n.a.	1222	4	0
(*E*,*E*)-2,4-hexadienal[e]	fruity	n.a.	1263	4	3
2-octanone[c]	oily, waxy	n.a.	1268	4	−
octanal[c]	earthy	977	1275	1	0
1-octen-3-one[c]	mushroom	n.a.	1280	4	3
(*Z*)-2-heptenal[d]	rancid	n.a.	1302	0	0
2,3-dimethyl-pyrazine[c.f,std]	roasted coffee	n.a.	1314	4	0
2-acetyl-1-pyrroline[e]	sweet	922	1334	1	0
2-methyl-2-cyclopentan-1-one[c]	rust, burnt rubber	n.a.	1352	3	3
hexanol[e]	fruity	n.a.	1367	1	−
nonanal[c,std]	scented candle	1104	1372	4	2
methylethylpyrazine[e]	nutty, oily	n.a.	1383	2	−
octanol[e]	mushroom, earthy	n.a.	1384	0	3
3-octen-2-one[e]	nutty	n.a.	1396	0	2
2,4-heptadienal[e]	stir-fried oil,	1004	1407	0	1
5-methyl-2-(3*H*)-furanone[c]	roasted coffee	n.a.	1413	3	2
(*E*)-2-octenal[c]	insect, green, fat	1057	1421	0	1
acetic acid[c]	vinegar	649	1426	3	3
2,3-dimethyl-5-ethyl pyrazine[e,f,std]	wok	n.a.	1436	4	−

Table 10 (Continued)

Compound	Odor description[a]	Retention Index		log$_3$FD factor[b]	
		DB-5	FFAP	FFR	SFR
furfuralc	caramel	n.a.	1453	0	−
unknown C	rice	n.a.	1471	0	3
(*E*,*E*)-2,4-heptadienalc	stir fried oil	1012	1478	4	4
nonanole	stale	n.a.	1505	3	3
(*E*)-2-nonenalc	cucumber peel	1148	1513	0	2
unknown D	brown	n.a.	1518	0	−
unknown E	floral, green	n.a.	1533	0	1
(*E*,*Z*)-2,4-nonadienale	rice	1196	1547	2	5
unknown F	fresh, green	n.a.	1554	1	2
(*E*,*E*)-2,4-octadienalc	cucumber peel	n.a.	1579	2	4
γ-butyrolactone[d]	sweet	n.a.	1593	2	0
butanoic acid[c]	cheesy	n.a.	1606	3	0
2-decenal[e]	musty	n.a.	1609	0	2
benzene acetaldehyde[c]	grass	n.a.	1619	2	0
acetophenone[c]	flower, sweet	n.a.	1634	5	5
3-methyl-butanoic acid[d]	sweaty	n.a.	1649	3	1
pentanoic acid[c]	sweaty	n.a.	1721	3	1
2-(5*H*)-furanone[d]	sweet, waxy	n.a.	1725	1	3
unknown G	coriander, green	n.a.	1730	0	3
2-undecenal[c]	fruity	n.a.	1737	3	1
(*E*,*E*)-2,4-decadienal[c]	fatty, green	n.a.	1761	2	1
unknown H	dried, stale	n.a.	1783	0	−
2-dodecenal[e]	oily, cucumber peel	n.a.	1801	2	2
hexanoic acid[c,d]	fatty, oily	n.a.	1829	4	1
ethyl laurate[e]	fresh	n.a.	1844	0	3
unknown I	fresh, cucumber	n.a.	1878	0	2
2-methyl-fuoate	dried, stale, mushroom	n.a.	1987	2	−
octanoic acid[c,d]	oily, sweet	n.a.	2024	5	4
unknown J	salty	n.a.	2067	3	3
unknown K	burnt rubber	n.a.	2168	4	1
unknown L	burnt	n.a.	2191	3	−

Table 10 (Continued)

Compound	Odor description[a]	Retention Index		$\log_3$FD factor[b]	
		DB-5	FFAP	FFR	SFR
unknown M	sweet, sugar	n.a.	2387	0	−
indolec	zoo, dung	n.a.	2434	3	0
unknown N	sweet, vanilla	1395	2534	3	0

Note [a] Odor description at the sniffing port from GC-O.

[b] Log_3FD Factor on FFAP or DB-5 columns..

[c] Identified by comparing with RI on Stabilwax® column, mass spectra obtained by TOFMS and odor at the sniffing port on the same phase column.

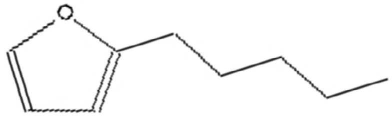
[d] Identified by comparing with RI on FFAP columns, mass spectra obtained by MS and odor at the sniffing port on the same column.

[e] Identified by comparing with RI from Acree and Arn (2009)

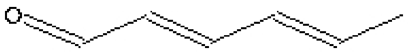
[f] Identified by comparing with RI from Chin et al. (2011)

[std] Identified based on standard compound.

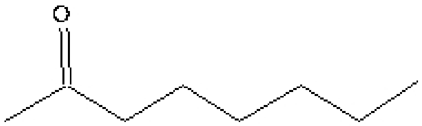
n.a. : not available



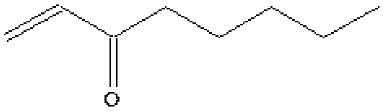
2-pentyl-furan



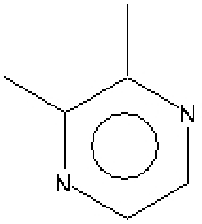
(*E*,*E*)-2,4-hexadienal



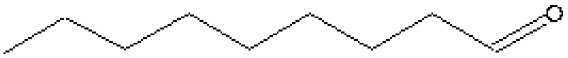
2-octanone



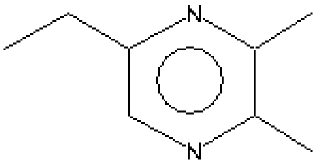
1-octen-3-one



2,3-dimethylpyrazine



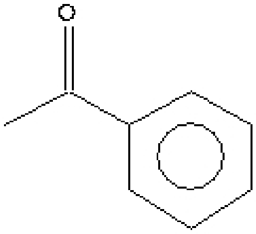
nonanal



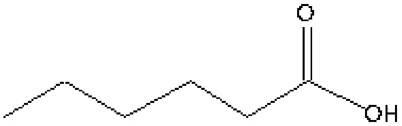
2,3-dimethyl-5-ethylpyrazine

(E,*E*)-2,4-heptadienal

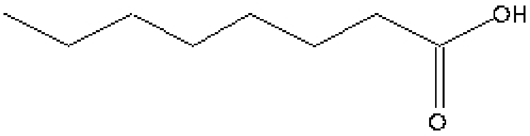
Figure 20 Chemical structures of aroma active compounds in flash fried rice (FFR)



acetophenone



hexanoic acid



octanoic acid

Figure 20 (Continued)

CONCLUSIONS

Flash-frying gave the unique burnt aroma characteristic that was different from the aroma of the typical stir-fried rice. This unique aroma comes from more species and high amount of aroma compounds especially pyrazines and furans from Strecker degradation and caramelization, aldehydes, ketones and acids from degradation and oxidation of lipids.

The results from the descriptive sensory analysis revealed that, the flash fried rice showed higher degree of aroma attributes namely, heated oil, smoky, acrid, wok, animal fat, dark brown, burnt, briny and dried aroma which differed from stir fried rice samples ($p<0.05$). The increasing of heptanal, benzaldehyde, octanal and (*E*)-octenal that show fatty, oily characteristic was the main factor that led to the increasing of aroma attributes namely, animal fat and heated oil. Meanwhile, the presence of pyrazines, phenol, furfural, 5-hydroxymethylfurfural, 2-(5*H*)-furanone, 2-methyl-2-furancarboxaldehyde and1,2-cyclopentanedione may contribute to the smoky, acrid, wok, dark brown and burnt aroma.

Compounds that have high impact on aroma of flash-fried rice are 2,3-dimethyl-5-ethylpyrazine and 2-pentylfuran that had wok aroma characteristic, octanoic acid (oily, sweet), acetophenone (sweet), 2,3-dimethylpyrazine (roast), (*E*,*E*)-2,4-heptadienal (stir-fried oil), nonanal (scented candle), 2-octanone (oily, sweet), (*E*,*E*)-2,4-hexadienal (fruity), 1-octen-3-one (mushroom) and hexanoic acid (fatty, oily).

The identification and extraction of aroma active compounds in flash fried food can benefit to future synthesis of the wok flavor components and eliminate the health problem of associated with carcinogen generated during the high temperature cooking.

LITERATURE CITED

Acree, T. and H. Arn. 2009. **Flavornet and Human Odor Space**. Available source: www.flavornet.org/flavornet.html, September 14, 2012.

Adams, T.B., J. Doull, V.J. Feron, J.I. Goodman, L.J. Marnett, I.C. Munro, P.M. Newberne, P.S. Portoghese, R.L. Smith, W.J. Waddell, and B.M. Wagner. 2002. The FEMA GRAS assessment of pyrazine derivatives used as flavor ingredients. **Food and Chemical Toxicology** 40: 429–45.

Alencar, J.W., P.B. Alves and A.A. Craveiro. 1983. Pyrolysis of tropical vegetable oils. **Journal of Agricultural and Food Chemistry** 31: 1268-1270.

Arshadia, M., M. Ghiaci, A. Rahmaniana, H. Ghaziaskara and A. Gil. 2012. Oxidation of ethylbenzene to acetophenone by a Mn catalyst supported on a modified nanosized SiO_2/Al_2O_3 mixed-oxide in supercritical carbon dioxide. **Applied Catalysis B: Environmental** 119-120: 81-90.

Bastos, D.M., É. Monaro, É. Siguemoto and M. Séfora. 2012. **Maillard Reaction Products in Processed Food: Pros and Cons.** Available source: http://www.intechopen.com/books/food-industrial-processes-methods-and-equipment/maillard-reactionproducts-in-processed-food-pros-and-cons.

Carlin, J.T., C.T. Ho, S.S. Chang, Q.Z. Jin, and T.C. Huang. 1986. Identification of alkyloxazoles in the volatile compounds from French-fried potatoes. **Journal of Agricultural and Food Chemistry** 34: 621.

________, ________, _________, A. Vellus and W. Pickenhagan. 1990. Analysis of French fried potato flavor: identification of 3-(methylthio) alkanals. **LWT-Food Science and Technology** 23: 276

Chang, C.C., S.W. Wan. 1947. Chinas motor fuels from tung oil. **Industrial Engineering Chemistry Research** 39: 1543–1548.

Chao, B.Y. 1945. **How to Cook and Eat in Chinese**. New York: John Day. New York.

Chin, S.T., G.T. Eyres and P.J. Marriott. 2011. Identification of potent odourants in wine and brewed coffee using gas chromatography-olfactometry and comprehensive two-dimensional gas chromatography. **Journal of Chromatography A** 1218: 7487-7498.

Costa, N.C.D., S. Eri. 2005. Flavor generation in food, pp13-15, 25-26. *In* D.J, Rowe, ed. **Chemistry and Technology of Flavors and Fragrances**. CRC Press LLC., New York.

Dong, F., Z. Yang, S. Baldermann, Y. Kajitani, S. Ota, H. Kasuga, Y. Imazeki, T. Ohnishi and N. Watanabea. 2012. Characterization of *L*-phenylalanine metabolism to acetophenone and 1-phenylethanol in the flowers of Camellia sinensis using stable isotope labeling. **Journal of Plant Physiology** 169: 217-225.

Drumm, T.D. and A.M. Spanier. 1991. Changes in the content of lipid autoxidation and sulfur-containing compounds in cooked beef during storage. **Journal of Agricultural and Food Chemistry** 39(2): 336-343.

Engel, W., W. Bahr and P. Schieberle. 1999. Solvent assisted flavour evaporation – a new and versatile technique for the careful and direct isolation of aroma compounds from complex food matrices. **European Food Research and Technology** 209: 237–241.

Fisher, C. and T.R. Scott. 1997. **Food Flavours: Biology and Chemistry.** Royal Society of Chemistry, Cambridge.

Flores, M., C.C. Grimm, F. Toldra and A.M. Spanier. 1997. Correlations of sensory and volatile compounds of spanish “serrano” dry-cured ham as a function of two processing times. **Journal of Agricultural and Food Chemistry** 45: 2178−2186.

Fortes, I.C.P., P.J. Baugh. 1999. Study of analytical on-line pyrolysis of oils from Macauba fruit (*Acrocomia sclerocarpa* M) via GC/MS. **Journal of the Brazilian Chemical Society** 10: 469-477.

___________,_________. 2004. Pyrolysis–GC/MS studies of vegetable oils from Fortes, I.C.P. and P.J. Baugh. 2004. Pyrolysis–GC/MS studies of vegetable oils from Macauba fruit. **Journal of Analytical and Applied Pyrolysis** 72: 103–111.

Hajslova, J. and T. Cajka. 2009. Gas Chromatography in Food Analysis, pp119-142. *In* S. Ötles, ed. Handbook of Food Analysis Instruments. CRC Press LLC., New York.

Kuo, M.C., Y. Zhang, T.G. Hartman, R.T. Rosen and C.T. Ho. 1989. Selective purge and trap method for the analysis of volatile pyrazines. **Journal of Agricultural and Food Chemistry** 37: 1020-1022.

Lasekan, O. 2012. Headspace solid-phase microextraction gas chromatography-mass spectrometry (HS-SPME-GCeMS) determination of volatile compounds in roasted plantains (French sombre and Dwarf Kalapua). **LWT - Food Science and Technology** 46: 536-541.

Leffingwell, J.C. 2004. **Flavor-Base version 2004**. Flavor-Base Database, Version Date July 01, 2004.

Maher K.D., D.C. Bressler. 2007. Pyrolysis of triglyceride materials for the production of renewable fuels and chemicals. **Bioresource Technology** 98: 2351–2368.

Meilgaard, M., G.V. Civille and B.T. Carr. 1999. **Sensory Evaluation Techniques**. 3rd ed., CRC Press, New York.

Moldoveanu, S. C. 1998. **Techniques and Instrumentation in Analytical Chemistry Volume 20; Analytical Pyrolysis of Natural Organic Polymers.** Elsevier Science B.V., Amsterdam.

Nissen, J.A. 2007. Continuous wok-frying of vegetables: Process parameters influencing scale up and product quality. **Journal of Food Engineering** 83: 54-60.

Ó Hare, L. and J. Grigor. 2005. Flavor generation in food, pp. 35-55. *In* D.J, Rowe, ed. **Chemistry and Technology of Flavors and Fragrances**. CRC Press LLC., New York.

Paine III, J.B., Y.B. Pithawalla,, J.D. Naworal. 2008. Carbohydrate pyrolysis mechanisms from isotopic labeling Part 4. The pyrolysis of D-glucose: The formation of furans. **Journal of Analytical and Applied Pyrolysis** 83: 37–63.

Patwardhan, P.R., J.A. Satrio, R.C. Brown and B.H. Shanks. 2009. Product distribution from fast pyrolysis of glucose-based carbohydrates. **Journal of Analytical and Applied Pyrolysis** 86: 323-330.

Piyachaiseth, T. 2010. **Aroma Compounds in Fried Rice.** M.S. Thesis, Kasetsart University.

Pryde, E.H. 1980. **Physical properties of soybean oil.** pp. 33-48. *In* D. R. Erickson, , E.H. Pryde, O.L. Brekke, T.L. Mounts, and R.A. Falb, eds. Handbook of Soy Oil Processing and Utilization. American Soybean Association, St. Louis.

Rizzi, G. 2008. The strecker degradation of amino acids: newer avenues for flavor formation. **Food Reviews International** 24: 416–435.

Rosenfeld, H. J. and A. Nes. 2000. Prediction of sensory quality of strawberry jam by means of sensory quality attributes of fresh fruits. **Journal of the Science of Food and Agriculture** 80: 1895-1902.

Sanz, C., D. Ansorena, J. Bello and C. Cid . 2001. Optimizing headspace temperature and time sampling for identification of volatile compounds in ground roasted Arabica coffee. **Journal of Agricultural and Food Chemistry** 49: 1436-1369.

Schwab, A.W., G.J. Dykstra, E. Selke, S.C. Sorenson, E.H. Pryde. 1988. Diesel fuel from thermal-decomposition of soybean oil. **Journal of the American Oil Chemists' Society** 65(11): 1781–1786.

Shahidi, F., L.J. Rubin and L.A. D'Souza. 1986. Meat flavor volatiles: A review of the composition, techniques of analysis, and sensory evaluations. **Critical Reviews in Food Science and Nutrition** 24: 141-243.

Squires, A.M. 2007. Chemical process opportunities for vibrated powders
3. Provisional design for an experimental side wards flying bed. **Powder Technology** 175: 55–62.

Tressl, R., D. Bahri, H. Kopper and A. Jensen. 1978. **Diphenole und caramelkomponenten in rostkaffees verchiedener sorten.** II. Z. Lebensm. Unters. Forsch. 167: 111-114. Cited. Flament, I. 2002. Coffee Flavor Chemistry. John Wiley & Sons, Ltd. Chichester.

van Boekel M.A.J.S. 2006. Formation of flavour compounds in the Maillard reaction. **Biotechnology Advances** 24: 230–233.

van den Dool, H. and P.D.A. Kratz. 1963. A generalization of the retention index system including linear temperature programmed gas liquid partition chromatography. **Journal of Chromatography** 11: 463-471.

van Loon, W.A.M.V., J.P. Linssen, H.A. Legger, M.A. Posthumus and A.G.J. Voragen. 2005. Identification and olfactometry of french fried flavour extracted at mouth conditions**. Food Chemistry** 90: 417-425.

Wagner, R. and W. Grosch. 1997. Evaluation of potent odorants of French fries. **LWT - Food Science and Technology** 30: 164-169.

Whitefield, F.B. 1992. Volatiles from Interactions of maillard reactions and lipids. **Food Science and Nutrition** 31: 1-58.

Wu, C.M. and S.Y. Chen. 1992. Volatile compounds in oils after deep frying or stir frying and subsequent storage. **Journal of the American Oil Chemists' Society** 69: 858-865.

Young, G. 2004. Wok hay: The breath of a wok. **Gastronomica: The Journal of Food and Culture** 4: 26-30.

Yu, A.N. and A.D. Zhang. 2010. The effect of pH on the formation of aroma compounds produced by heating a model system containing L-ascorbic acid with L-threonine/L-serine. **Food Chemistry** 119: 214–219.

APPENDICES

Appendix A

Analysis of volatile compounds

Calculation of Retention Index

Retention index is a measure of relative retention using normal alkanes as a standard of reference. Retention index can be calculated by using the formula derived by Van den Dool and Kratz (1963).

$$RI = 100N + 100n \frac{t_{Ra}-t_{RN}}{t_{R(N+n)} - t_{RN}}$$

Where

N = carbon number of the lower alkane

n = the difference in carbon number of the two n-alkanes that bracket the compound

t_{Ra} = the retention time of unknown (or test) compound

t_{RN} = the retention time of the lower alkane

$t_{R(N+n)}$ = the retention time of the upper alkane

Calculation of Relative Concentration of Volatile Compounds

$$C_s = \frac{C_I \times A_S \times V_I \times r}{A_I \times W_S}$$

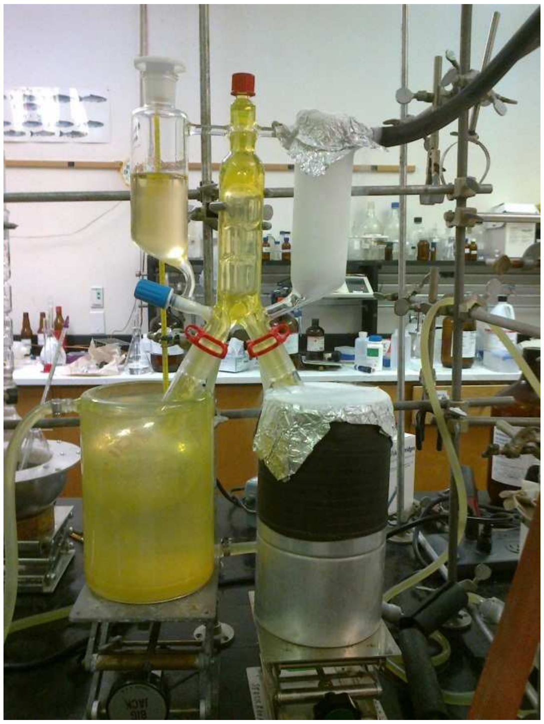
Where

C_S = relative concentration of sample (µg / g of sample)
C_I = concentration of internal standard (mg / ml in diethylether)
A_S = peak area of sample
A_I = peak area of internal standard
V_I = volume of internal standard (µl)
W_S = weight of sample (g)
r = response factor = 1

Isolation of volatile compounds

1 Solvent Assisted Flavor Evaporation (SAFE)

Volatile compounds were extracted from 480 g of samples using 500 ml of diethyl ether for 2 times. The mixtures were shaken at 200 rpm for 30 min at room temperature. The two portions of the extracts were combined and concentrated to 50 ml using a vigreux column (20 cm x 1 cm i.d.) at 40°C. The extract was distilled at 50°C by SAFE apparatus under vacuum to separated volatile compounds from the oil matrix. Liquid nitrogen was used for condensation of volatile compounds. The distillate was further concentrated to 50 ml by using a vigreux column (20cm x 1 cm i.d.) at 50°C. The extract was dried by anhydrous Na_2SO_4 overnight and further concentrated to 0.5 ml under mild nitrogen stream.



Appendix Figure A1 Solvent assisted flavor evaporation (SAFE) apparatus

2 Solid Phase Micro Extraction (SPME)

SPME fiber coated with DVB/CAR/PDMS (Supelco Co., USA) was used. 10 g. of flash-fried rice were weight into a 20 ml headspace vial. The vial was heated at 80°C. SPME fiber was exposed to the headspace of the sample to adsorb the volatile compounds for 2 hr. Then, the fiber was introduced into the GC injector port for desorption at 250°C for 5 min.

Analysis of Volatile Compounds from SAFE and SPME method

1. Gas Chromatography-Mass Spectrometry (GC-MS)

Volatile compounds were analyzed on 7890A gas chromatography equipped with a 5975C MSD mass spectrometer (Agilent Technologies, Palo Alto, CA, USA). Samples were injected by the splitless method into the capillary column, ZB-WAX (60m x 0.25m x 0.25μm film thickness). The oven temperature program for the ZB-WAX column started at 40 °C, held for 1 min, increased to 230 °C at the rate of 5 °C/min and then held for 10 min. Helium was used as a carrier gas at the constant flow rate of 2.2 ml/min. Volatile compounds was identified by comparison of mass spectrum data and retention indexes (RI) with Wiley 275 mass spectrum library, the literatures and from those of authentic standards. RIs were calculated using *n*-alkanes series (C_5-C_{22}).

Appendix Table A1 Identification of volatile compounds from flash fried rice obtained from SAFE and SPME method.

No.	Compound	Odor description[a]	RI $_{cal}$[b]	GC-MS[c]	
				SAFE	SPME
	Acids				
1	acetic acid	strong, pungent, sour, vinegar odor	1345	+	-
2	hexanoic acid	heavy, fatty, cheesey odor	1649	+	-
3	heptanoic acid	fatty, sour-sweat-like, rancid odor	1729	+	-
4	decanoic acid	rancid, fat	1963	+	+
	Alcohol				
5	1-octen-3-ol	−	1448	-	+
6	benzyl alcohol	faint, sweet, fruity aroma	1671	+	+
	Aldehydes				
7	hexanal	strong, fatty-green, grassy unripe fruit odor	1061	-	+
8	heptanal	fatty, in dilution sweet, fruity, nutty, cognac like	1163	-	+
9	(*E*)-hexanal	green, fruity, fresh, apple and woody with leafy and grassy notes	1204	-	+
10	octanal	fatty-fruity odor; sweet	1265	-	+
11	(*Z*)-2-heptenal	−	1309	-	+
12	nonanal	fatty-floral-rose, waxy odor	1374	-	+
13	(*E*)-2-octenal	peculiar fatty, green-grassy-leafy odor	1413	-	+
14	(*E*,*E*)-2,4-heptadienal	fatty, green, with and oily, greasy	1479	-	+

Appendix Table A1 (Continued)

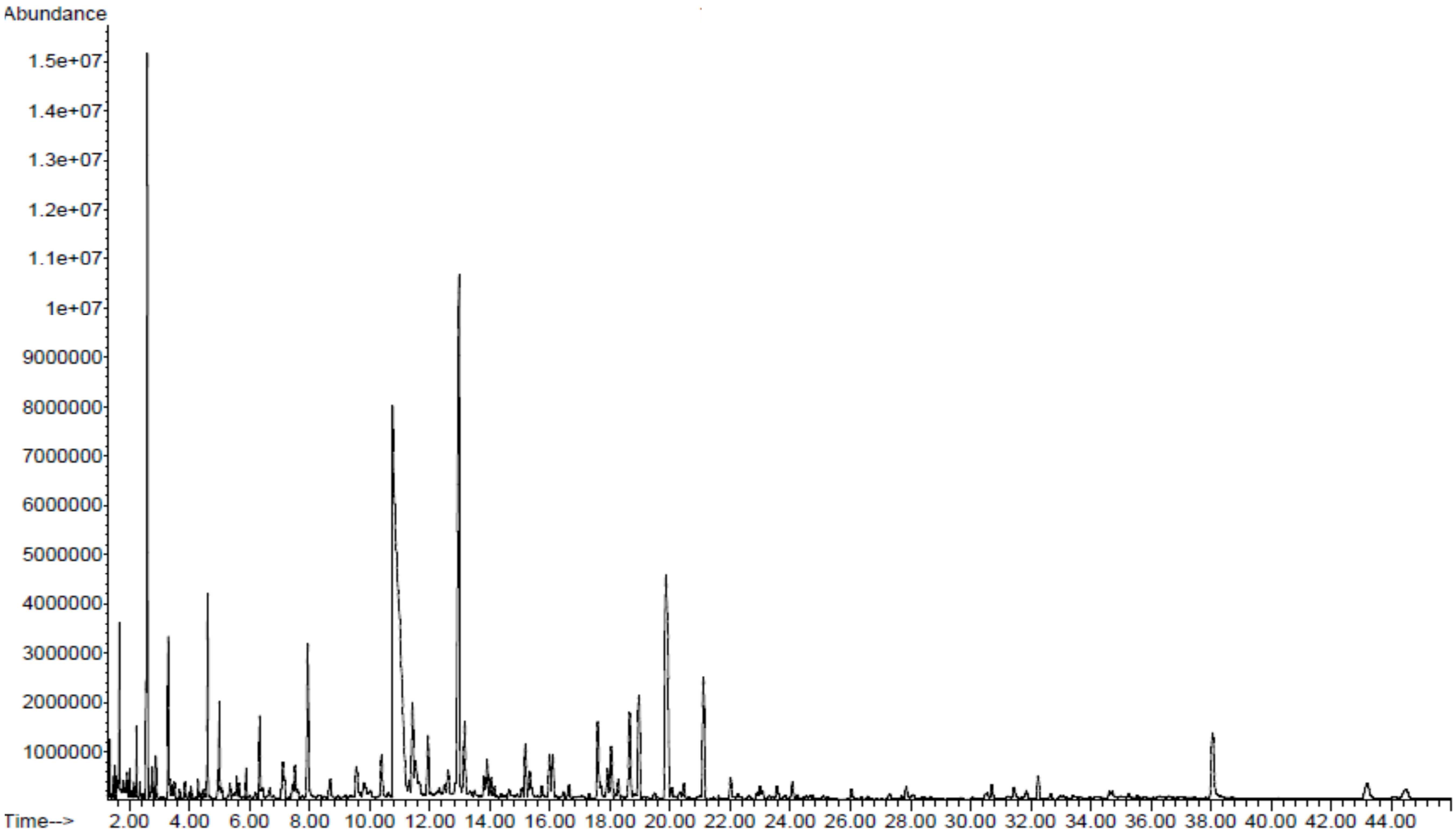
No.	Compound	Odor description[a]	RI $_{cal}$[b]	GC-MS[c]	
				SAFE	SPME
15	benzaldehyde	odor of bitter almond oil	1502	-	+
16	(*E*)-2-decenal	waxy, fatty, earthy, coriander, mushroom, green, pork fat note	1508	+	+
17	(*E*)-2-nonenal	−	1520	-	+
18	2-undecenal	fresh, sweet-citrus, orange peel in dilution	1583	+	+
19	(*E*,*E*)-2,4-decadienal	strong, deep fat flavor; fatty-citrus notes	1754	+	+
20	2-(phenylmethylene)-heptanal	−	1947	+	-
	<u>Furan derivatives</u>				
21	2-pentyl-furan	earthy, green and metallic odor	1194	-	+
22	furfural	sweet, cereal, bread-like, caramellic note	1451	-	+
23	2-furanmethanol	fermented, burnt-sugar, creamy, caramel-like	1523	+	-
24	2-(2-propenyl)-furan	−	1600	-	+
	<u>Hydrocarbons</u>				
25	undecene	−	1074	-	+
26	5-undecene	−	1124	-	+
27	dodecane	Alkane	1156	-	+
28	(*E*)-3-dodecene	−	1164	+	-
29	(*E*)-6-dodecene	−	1180	-	+

Appendix Table A1 (Continued)

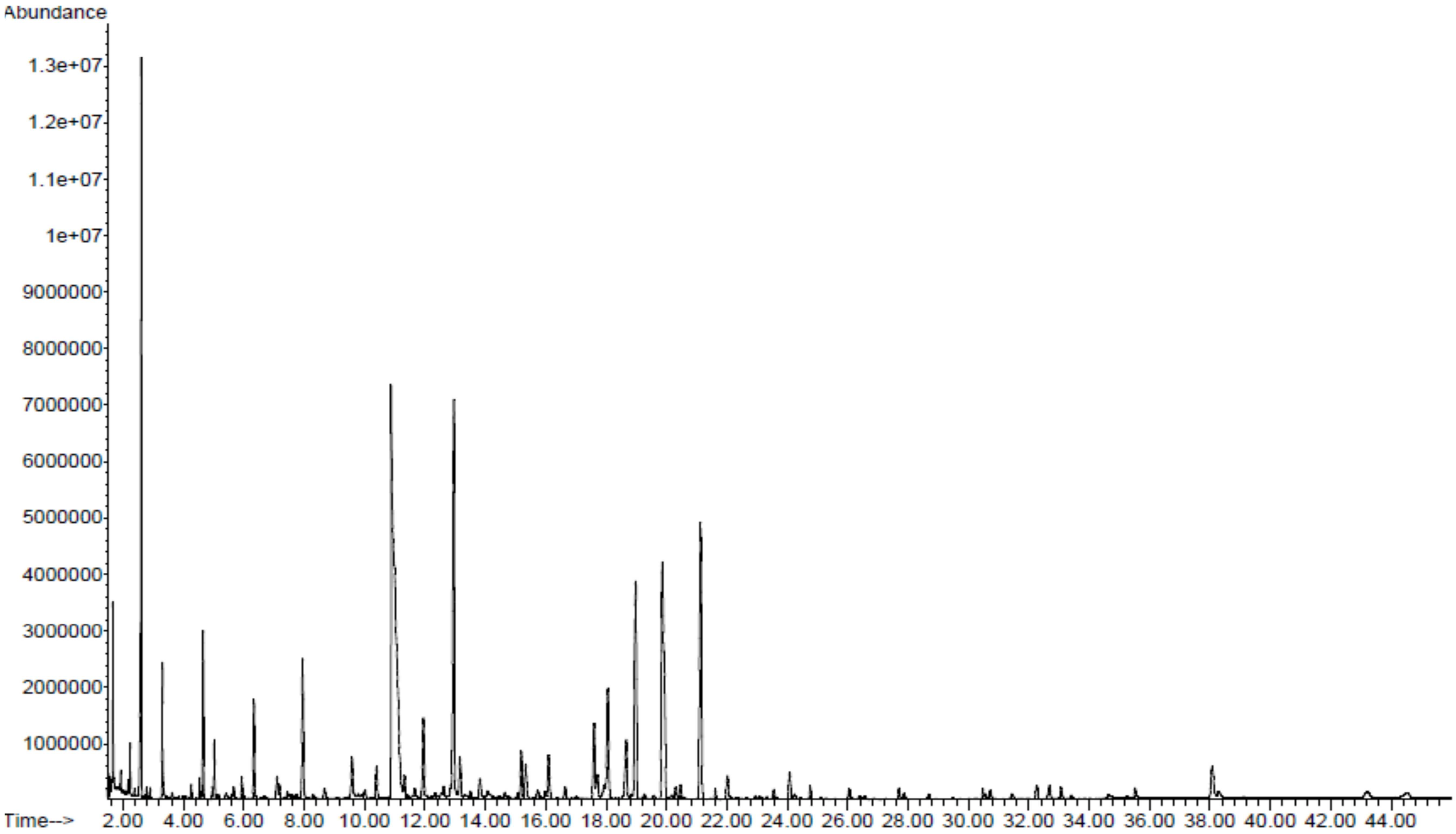
No.	Compound	Odor description[a]	RI $_{cal}$[b]	GC-MS[c]	
				SAFE	SPME
30	1,3,5,7-cyclooctatetraene	−	1227	-	+
31	tridecane	Alkane	1253	-	+
32	(*Z*)-6-tridecane	−	1284	-	+
33	3-methyl-2-methyl-1,3-hexadiene	−	1397	-	+
34	nonyl-cycolpentane	−	1467	-	+
35	2-methyl-bicycol[2.2.2]-octane	−	1546	-	+
36	cyclodecene	−	1555	-	+
37	γ-butyrolactone	coconut-butter aroma; creamy	1699	+	+
38	phenol	phenolic medicinal odor	1765	+	-
39	2-methyl-4-vinylphenol	almond shell	2182	-	+
	Other				
40	butyl-benzene	−	1277	-	+
41	pentyl-benzene	−	1385	-	+
42	azulene	−	1712	-	+
43	methyl-phenyl-oxime	−	1772	-	+
44	indole	mothball, burnt	>2200	-	+

Note [a] Odor description from Flavor-base 2004 (Leffingwell 2004). [b] Retention Index on ZB-WAX column.

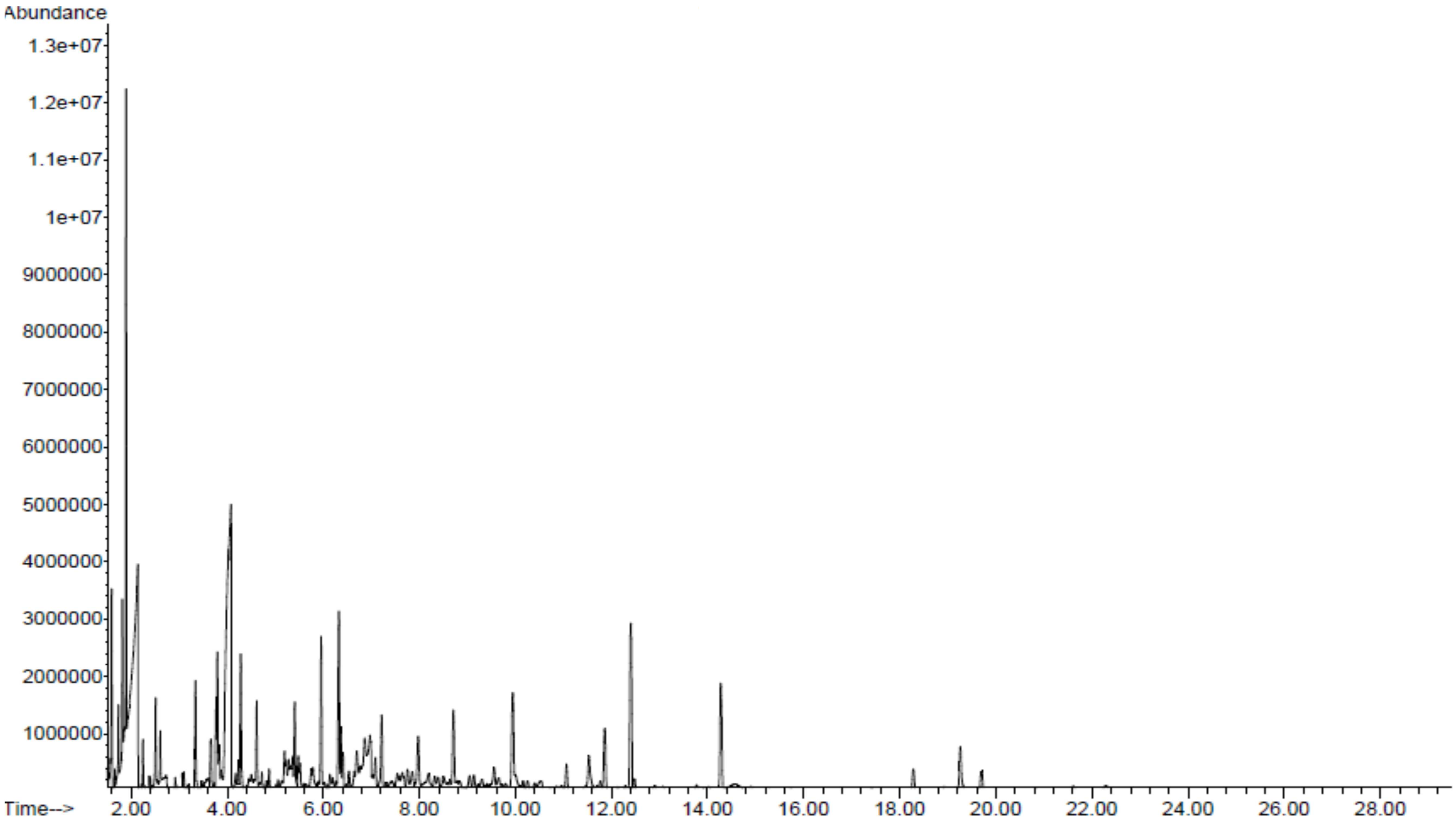
[c] "+" Identified by GC-MS.



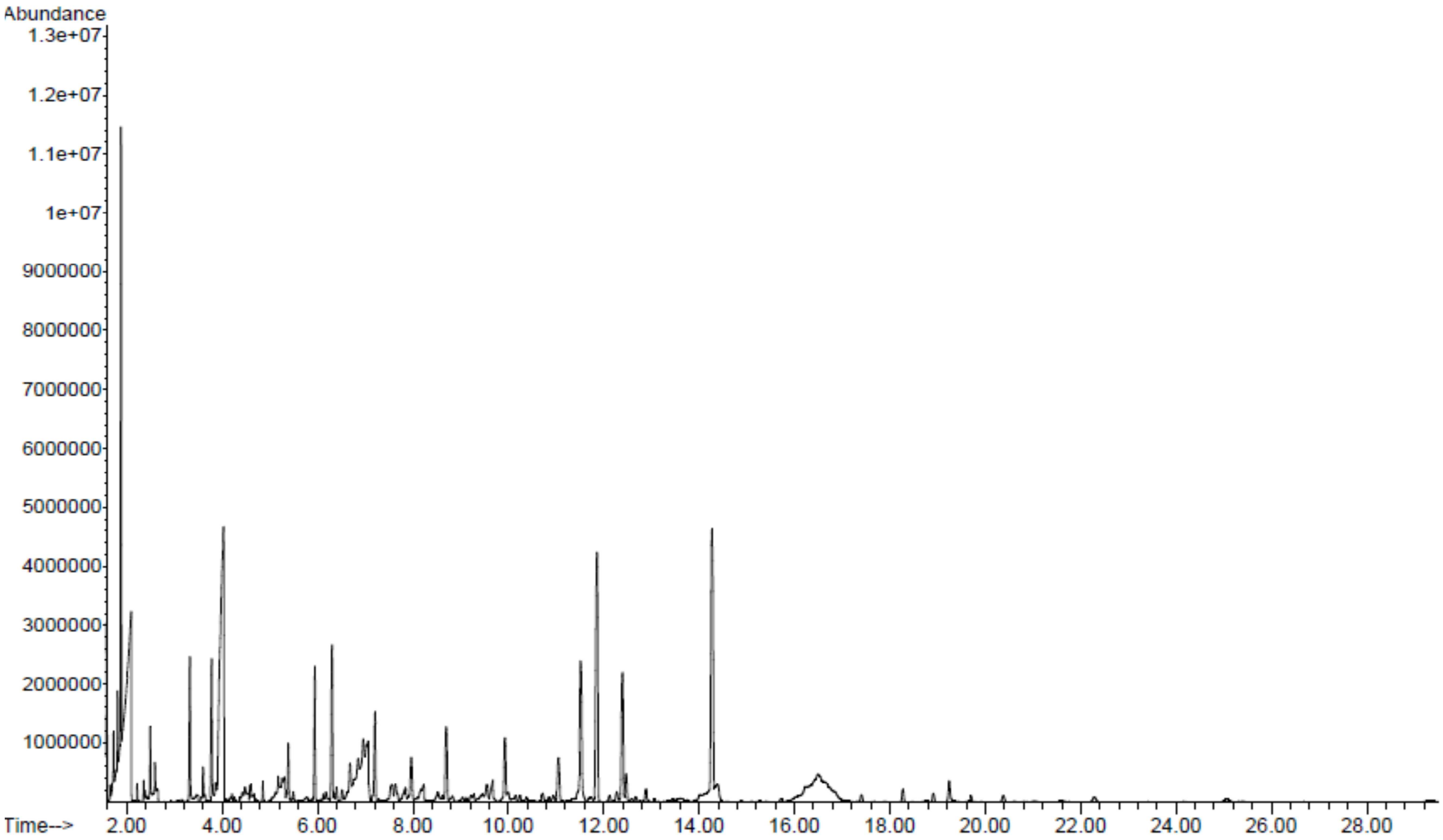
Appendix Figure A2 Total ion current gas chromatogram of flash-fried rice (FFR) injected by using cool on column/GC-MS on FFAP column.



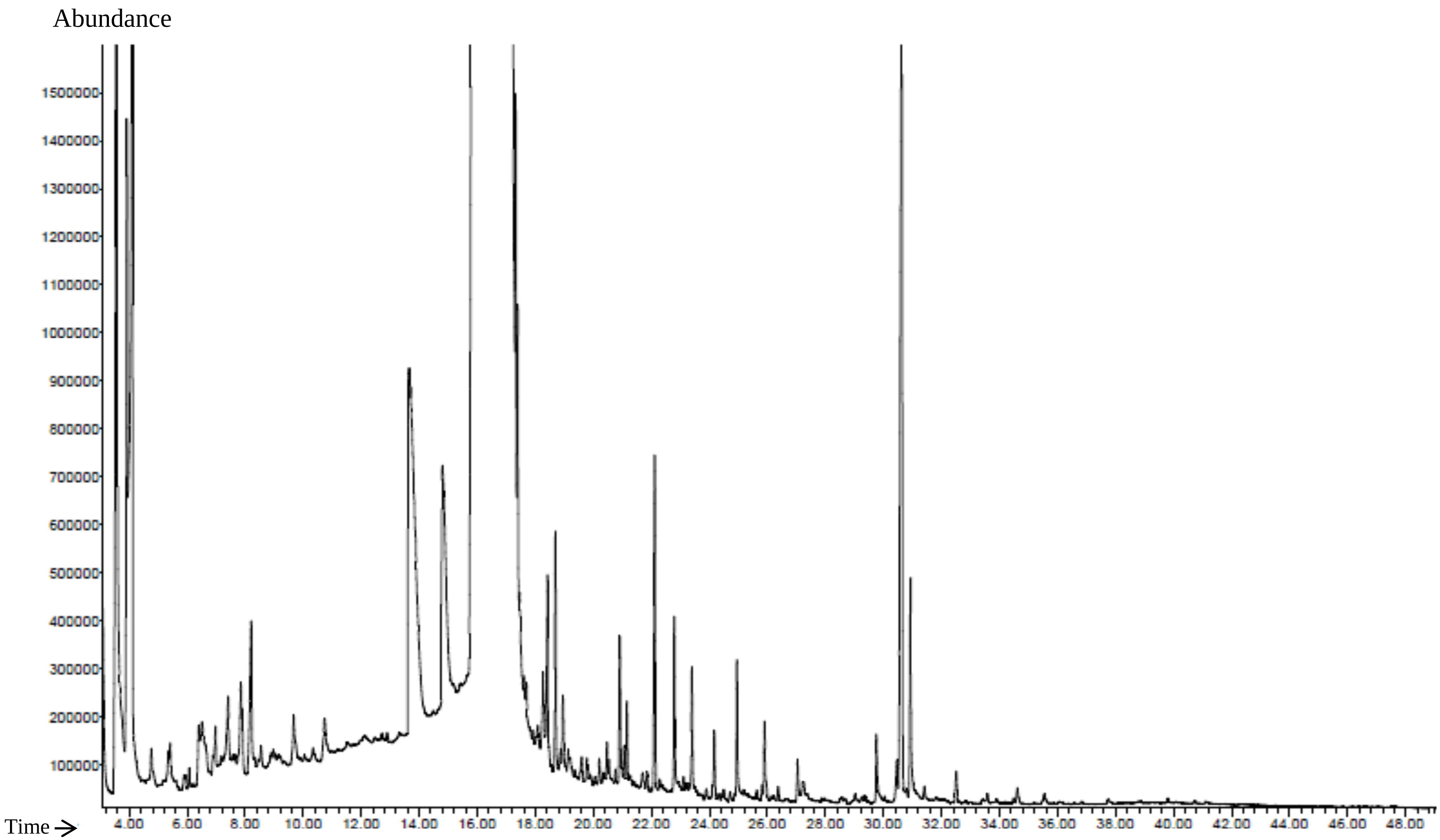
Appendix Figure A3 Total ion current gas chromatogram of stir-fried rice (SFR) injected by using cool on column/GC-MS on FFAP column.



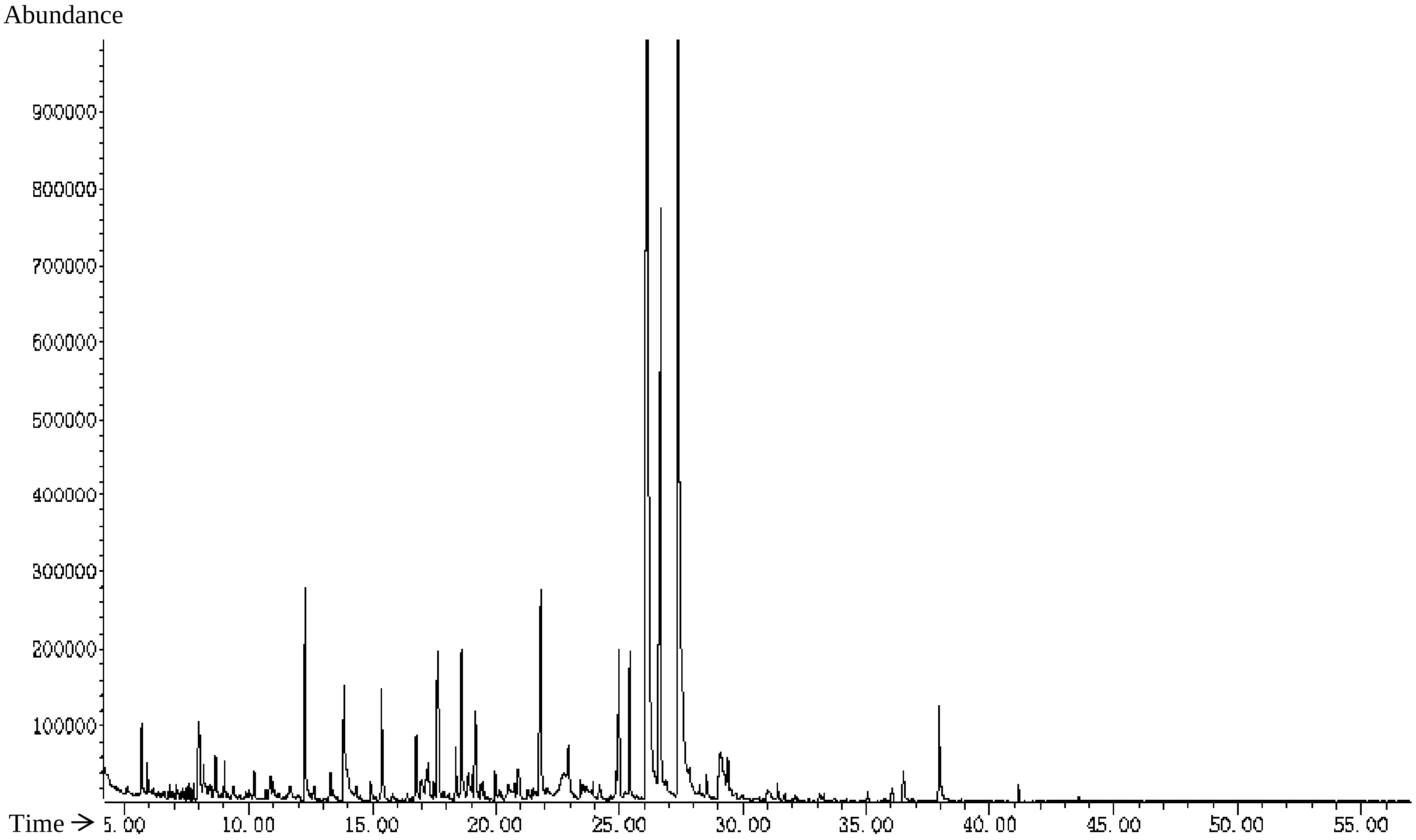
Appendix Figure A4 Total ion current gas chromatogram of flash-fried rice (FFR) injected by using cool on column/GC-MS on DB5-MS column.



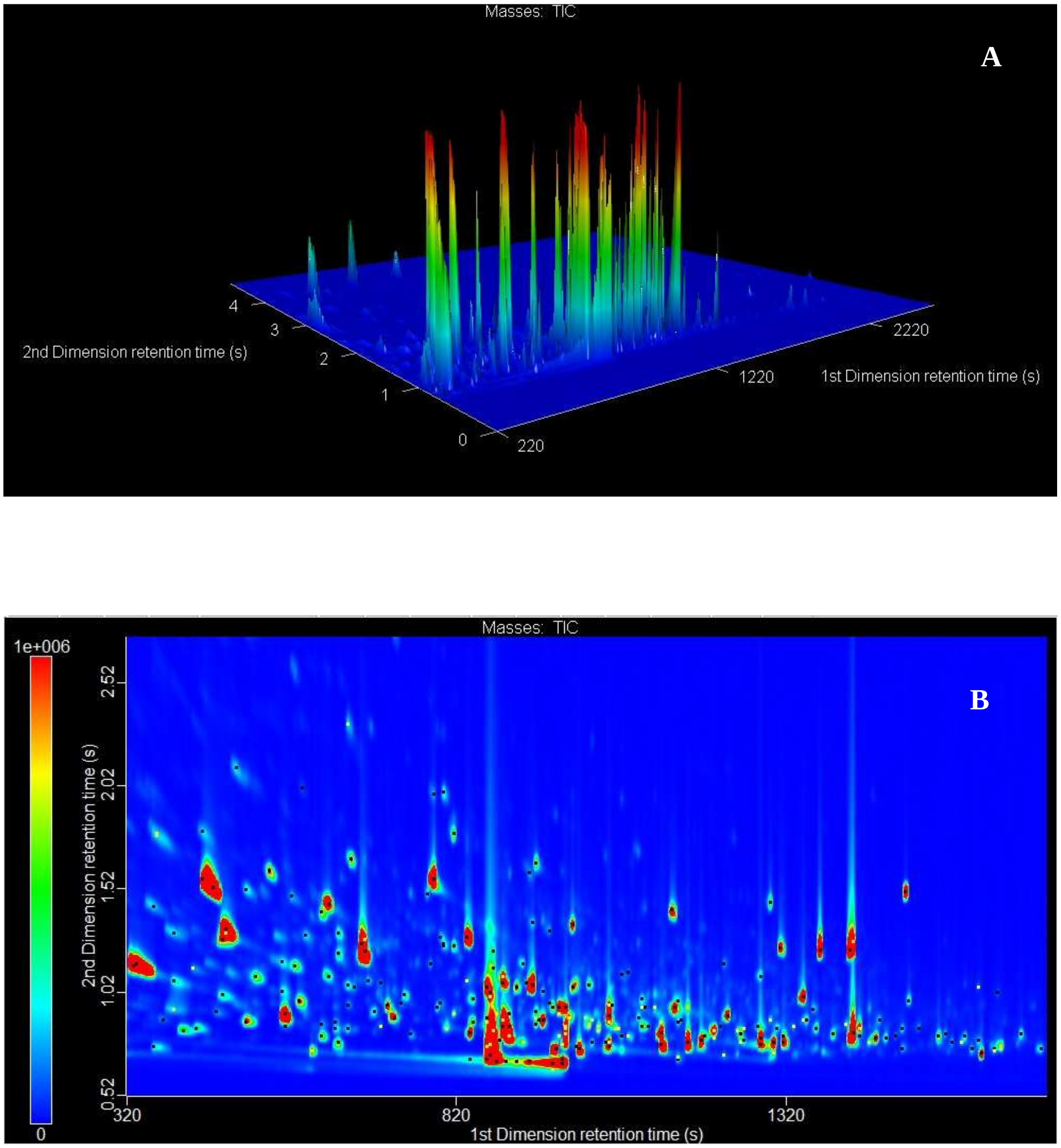
Appendix Figure A5 Total ion current gas chromatogram of stir-fried rice (SFR) injected by using cool on column/GC-MS on DB5-MS column.



Appendix Figure A6 Total ion current gas chromatogram of flash-fried rice (FFR) extracted by using solvent assisted flavor evaporation.



Appendix Figure A7 Total ion current gas chromatogram of flash-fried rice (FFR) extracted by using SPME.



Appendix Figure A8 GC×GC chromatogram of flash fried rice A: surface plot and B: contour plot.

Appendix B

Sensory evaluation

Paired *t*-test of aroma attribute in fried rice samples

1. Heated oil

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	4.904166667	4.5
Variance	0.413460145	0.260869565
Observations	24	24
Pearson Correlation	0.56131867	
Hypothesized Mean Difference	0	
Df	23	
t Stat	3.581502343	
P(T<=t) one-tail	0.00079001	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	0.001580019	
t Critical two-tail	2.068657599	

$P < 0.05$

2. Smoky

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	2.991666667	1.741666667
Variance	0.29384058	0.226884058
Observations	24	24
Pearson Correlation	0.316291086	
Hypothesized Mean Difference	0	
Df	23	
t Stat	10.24338847	
P(T<=t) one-tail	2.42345E-10	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	4.8469E-10	
t Critical two-tail	2.068657599	

$P < 0.05$

3. Acrid

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	2.75	1.720833333
Variance	0.24173913	0.374764493
Observations	24	24
Pearson Correlation	0.10617122	
Hypothesized Mean Difference	0	
df	23	
t Stat	6.782496743	
P(T<=t) one-tail	3.221E-07	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	6.44201E-07	
t Critical two-tail	2.068657599	

$P < 0.05$

4. Wok

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	5.091666667	2.55
Variance	0.457318841	0.417391304
Observations	24	24
Pearson Correlation	0.372188132	
Hypothesized Mean Difference	0	
df	23	
t Stat	16.79744506	
P(T<=t) one-tail	1.04433E-14	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	2.08865E-14	
t Critical two-tail	2.068657599	

$P < 0.05$

5. Animal fat

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	2.179166667	1.870833333
Variance	0.133894928	0.182155797
Observations	24	24
Pearson Correlation	0.466435326	
Hypothesized Mean Difference	0	
df	23	
t Stat	3.659648104	
P(T<=t) one-tail	0.000652248	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	0.001304496	
t Critical two-tail	2.068657599	

$P < 0.05$

6. Brothy

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	1.970833333	1.8625
Variance	0.13259058	0.183315217
Observations	24	24
Pearson Correlation	0.4890849	
Hypothesized Mean Difference	0	
df	23	
t Stat	1.312907101	
P(T<=t) one-tail	0.101081914	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	0.202163827	
t Critical two-tail	2.068657599	

$P < 0.05$

7. Dark brown

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	1.854166667	1.145833333
Variance	0.140851449	0.205199275
Observations	24	24
Pearson Correlation	0.012893705	
Hypothesized Mean Difference	0	
df	23	
t Stat	5.936657514	
P(T<=t) one-tail	2.36142E-06	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	4.72284E-06	
t Critical two-tail	2.068657599	

$P < 0.05$

8. Burnt

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	1.870833333	0.820833333
Variance	0.57259058	0.321721014
Observations	24	24
Pearson Correlation	0.2922089	
Hypothesized Mean Difference	0	
df	23	
t Stat	6.41250889	
P(T<=t) one-tail	7.62005E-07	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	1.52401E-06	
t Critical two-tail	2.068657599	

$P < 0.05$

9. Rancid

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	0.829166667	0.970833333
Variance	0.103025362	0.102155797
Observations	24	24
Pearson Correlation	0.27141335	
Hypothesized Mean Difference	0	
df	23	
t Stat	-1.794994927	
P(T<=t) one-tail	0.042904233	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	0.085808467	
t Critical two-tail	2.068657599	

$P < 0.05$

10. Briny

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	2.116666667	1.6125
Variance	0.16057971	0.212445652
Observations	24	24
Pearson Correlation	0.448433331	
Hypothesized Mean Difference	0	
df	23	
t Stat	5.423797254	
P(T<=t) one-tail	8.2021E-06	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	1.64042E-05	
t Critical two-tail	2.068657599	

$P < 0.05$

11. Overall sweet aromatics

t-test: Paired Two Sample for Means

	Flash-Fried Rice	*Stir-Fried Rice*
Mean	1.0625	1.05
Variance	0.03201087	0.022608696
Observations	24	24
Pearson Correlation	0.218182263	
Hypothesized Mean Difference	0	
df	23	
t Stat	0.29572364	
P(T<=t) one-tail	0.385047288	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	0.770094577	
t Critical two-tail	2.068657599	

$P < 0.05$

12. Dried

t-test: Paired Two Sample for Means

	Variable 1	*Variable 2*
Mean	2.141666667	1.7875
Variance	0.050362319	0.134184783
Observations	24	24
Pearson Correlation	0.27105757	
Hypothesized Mean Difference	0	
df	23	
t Stat	4.637430218	
P(T<=t) one-tail	5.74527E-05	
t Critical one-tail	1.713871517	
P(T<=t) two-tail	0.000114905	
t Critical two-tail	2.068657599	

$P < 0.05$

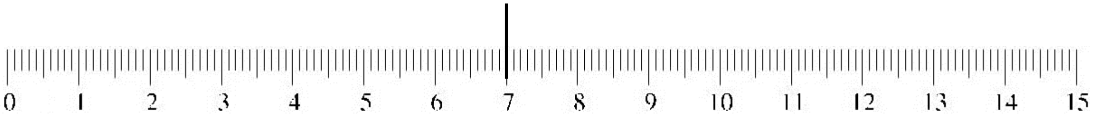
Ballot for descriptive sensory analysis of fried rice sample

Name…………………………………………….....
Date ……………………………………………….
Sample Code……………………………………

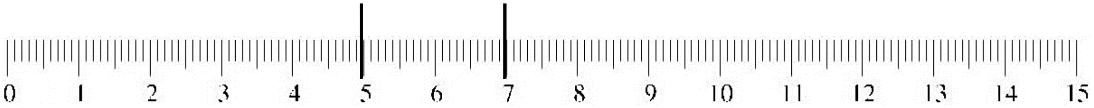
Instruction: Evaluate a sample on each attribute by open the covers of 1/3, sniffing and scoring the sample by placing “ | “ onto the scale

Aroma

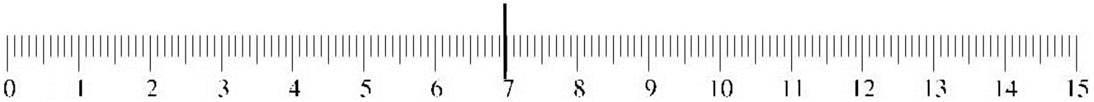
1. Heated oil



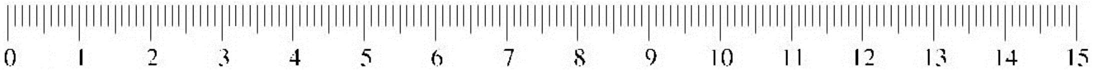
2. Smoky



3. Acrid

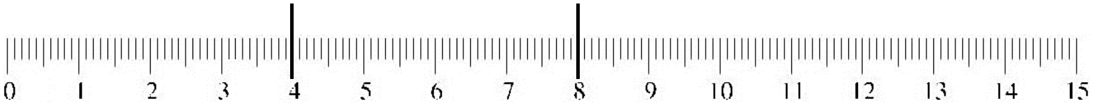


4. Wok



5. Animal fat

6. Brothy

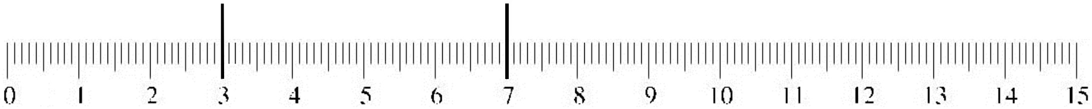


Ballot for descriptive sensory analysis of fried rice sample (continued)

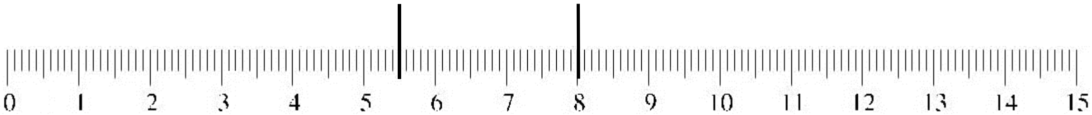
Name……………………………………………………
Date ……………………..…………………………
Sample Code...……..……………………………………

Aroma (Continued)

7. Dark brown

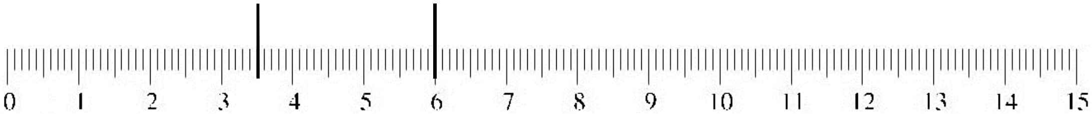


8. Burnt

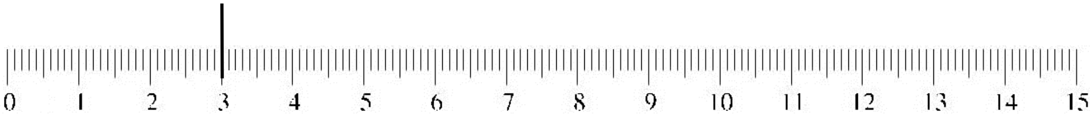


9. Rancid

10. Briny



11. Sweet aromatics



12. Dried

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2011	Exchange student at Oregon State University (March15-September 15, 2012) under the project "The Capacity Building for Kasetsart University Students on Internationalization Scholarship for 2012"