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Analysis of flavor volatiles of some Iranian rice cultivars by SPME-GC-MS

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Abstract:

A combined gas chromatography–mass spectrometry (GC–MS) with headspace solid-phase microextraction (HS-SPME) method has been employed for the analysis of the flavor volatiles of some Iranian rice cultivars (Domsiah, Deylamani, Hashemi, Shirodi and Tarom)**.** The samples were grown in different cities of Mazandaran province. The sampling procedure and chromatographic conditions were same for all samples. The separated chemicals were tentatively identified from their corresponding mass spectra (NIST mass libraries). A total of 49, 31, 29, 16 and 24 peaks were identified and assigned to Domsiah, Deylamani, Hashemi, Shirodi and Tarom rice samples, respectively. The volatile components identified in the five rice cultivars belong to the chemical classes of aldehydes, ketones, alcohols and heterocyclic compounds, as well as fatty acids and esters, phenolic compounds, hydrocarbons, etc. The main identified chemicals in these samples were not identical while some minor chemicals were same, which indicate the differences among the rice samples.

Key words: Gas chromatography-mass spectrometry, Rice flavor volatiles, Solid-phase microextraction, Headspace sampling

1. Introduction

Rice is a popular cereal crop commonly used as human food. It is actually a type of grass and belongs to a family of plants that includes other cereals such as; wheat and corn. Rice is rich in nutrients and contains a number of vitamins and minerals. It is an excellent source of complex carbohydrates and energy; which is the staple food of more than half of the world's population. Small variations in sensory properties, especially aroma, can make rice highly desired or unacceptable to consumers [1]. A recent review on extraction and pre-concentration techniques for the analysis of volatiles in rice samples can be found in the literature [2]. Standard methods for analysis of the flavor volatiles of rice employ schemes to capture the volatile compounds, enrich, separate and quantify them. There are many sample preparation techniques such as; purge and trap [3], steam distillation-solvent extraction [4], and direct solvent extraction [5] for analysis of the flavor volatiles of rice. These traditional methods have various drawbacks such as: complicated and time consuming procedures, requirement of a large amount of samples or organic solvents and loss of volatile compounds during solvent removal. Furthermore, due to the low concentrations of rice volatiles the development and optimization of sensitive analytical methods with low detection limits are necessary. The bibliography frequently refers to the use of the gas chromatography with mass spectrometry (GC/MS) for the analysis of these compounds together with a previous step of pre-concentration of the sample such as; solid-phase microextraction (SPME) [6]. Solid-phase microextraction is an interesting and promising technique for the extraction and concentration or enrichment of volatile compounds from different sample matrices [7]. It uses a fine rod with a polymeric coating to extract organic compounds from their matrix and directly transfer them into the injector of a gas chromatograph for thermal desorption and analysis [8].

This method is a growing sample preparation technique, and an attractive alternative to conventional extraction methods, that reduces solvent usage and exposure, disposal costs and extraction time for sample separation and concentration purposes. It provides enhanced sensitivity because the extracted fraction on the fiber can be introduced quantitatively into the gas chromatography (GC) by thermal desorption. Herrero Latorre et al. [9] reviewed the different analytical methods based on diverse solid phase extraction (SPE) sorbents for the speciation and preconcentration of inorganic selenium in water samples. Using several speciation strategies, and measuring the element by various analytical techniques have proven useful for this task. The use of SPE has led to enhancements in the sensitivity and selectivity of the primary methods for water samples and also avoided interferences due to other compounds in the sample matrix. Moreover, Ulrich [10] developed Applications of solid-phase microextraction techniques in biomedical analysis In this review, the optimization of SPME as well as advantages and disadvantages are discussed. It is concluded that, because of some unique characteristics, SPME can be introduced with benefit into several areas of biomedical analysis. SPME was introduced in food analysis [11, 12] and has been widely used in the analysis of the volatile compounds in recent years [13]. In order to analyzing of volatiles chemicals headspace solid phase microextraction (HS-SPME) technique was developed [14-17]. The advantage of making the extraction of analytes by means of HS-SPME is that the deterioration of the fiber is avoided, especially when complex samples are analyzed. Altaki et al. [18] achieve LOD from 8 to 70 pg g−1 when a headspace-solid-phase microextraction (HS-SPME) coupled to gas chromatography–ion trap mass spectrometry (GC–IT-MS) method is proposed for the analysis of furan in different heat-treated carbohydrate-rich food samples.

Moreover, this technique was introduced in studies on the flavor indicators of drug [19, 20]. Bonadio et al. [21] reported the Determination of venlafaxine in post-mortem whole blood by HS-SPME and GC-NPD. In the present work, the HS-SPME technique coupled with GC-MS was used for the simultaneous extraction, pre-concentration and determination of volatiles of Iranian rice's samples.

2. Materials and methods

2.1. Sample preparation

Five native rice cultivars from the northern provinces of Iran in the bordering of the Caspian Sea were used in this study. The samples were grown in different city of Mazandaran province. Among the various samples, Domsiah and Deylamani from Tonekabon, Hashemi from Noor, Shirodi from Amol and Tarom from Sari were selected for analyzing. The samples were chilled at 4 °C and then powdered to an aperture size of 150 µm. The exact weight of rice powder (1.0 gr) was placed into a 10 ml headspace vial and 5 ml water was added then the vial was sealed with a Teflon lined magnetic cap. The samples were placed in an oil bath and were preheated for 5 min at 80 °C prior to evaluation and were maintained at this temperature until analyzed.

The extraction and enrichment of the flavor volatiles of rice were performed using an SPME fiber (Supelco, Bellefonte, Pennsylvania, United States) 1 cm long, coated with polydimethylsiloxane (PDMS) preconditioned in a SPME fiber conditioner at 270°C for 1 hr before the first measurement. The fiber, mounted in the manual SPME holder, was in its protective sheath when it was inserted through the Teflon-coated silicone septum into the sampling port of the Pyrex glass sampling apparatus. Inside the sampling vial, the fiber was lowered to expose it to the effluent of the flavor volatiles of the rice. It was left there for adsorption and then withdrawn at the end of 30 min. Then the SPME fiber thermally desorbed the flavor volatiles in the injection port of the GC-MS instrument for 4 min at 250 °C. Then it was left in the SPME fiber conditioner for exposing to the flavor volatiles of the next sample.

2.2. Instrumentation

The chromatographic system used to be a Varian CP-3800 gas chromatograph (Markham (Greater Toronto Area) Ontario L3R 3V6 Canada) equipped with a mass spectrometric detector. A PC interfaced to the GC using MS data review software was used for data acquisition and processing. The chromatographic separations were carried out on a DB-5 capillary column (Agilent Technologies, USA), 60¬ m×0.25¬ mm i.d., 0.25¬ µm d.f., with 0.5 µm film thickness. The carrier gas was helium and adjusted at a constant flow-rate of 1.0 ml. min−1 and the split ratio was 5:1. Oven program temperature was 40 ˚C (hold 7 min), rate 8 ◦C/min up to 240 ˚C (hold 10 min). The ionization energy was 70 eV; the ion source and transfer line temperatures were 250 ˚C and 170 ˚C, respectively. The mass spectra of the sample constituents were compared with a spectral library (NIST) compatible with chemical classes of the components under study.

3. Results and discussion

The flavor volatiles of the five rice cultivars were directly extracted using HS-SPME method and analyzed by GC–MS. The sampling procedure and chromatographic conditions were same for all samples. Fig. 1 indicated the total ion chromatograms for the volatiles of these samples on the DB-5 capillary column. The separated chemicals were tentatively identified from their corresponding mass spectra (NIST mass libraries).

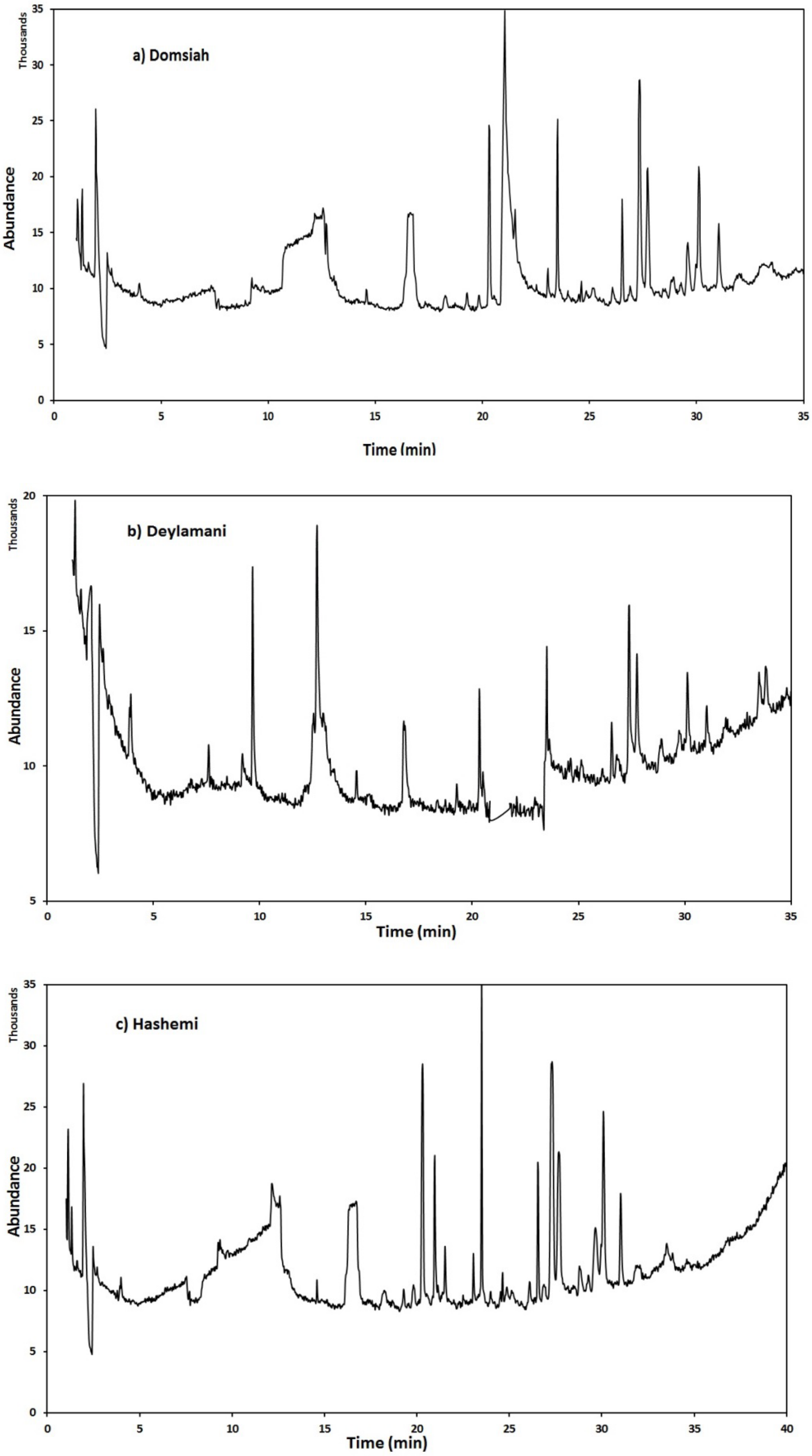
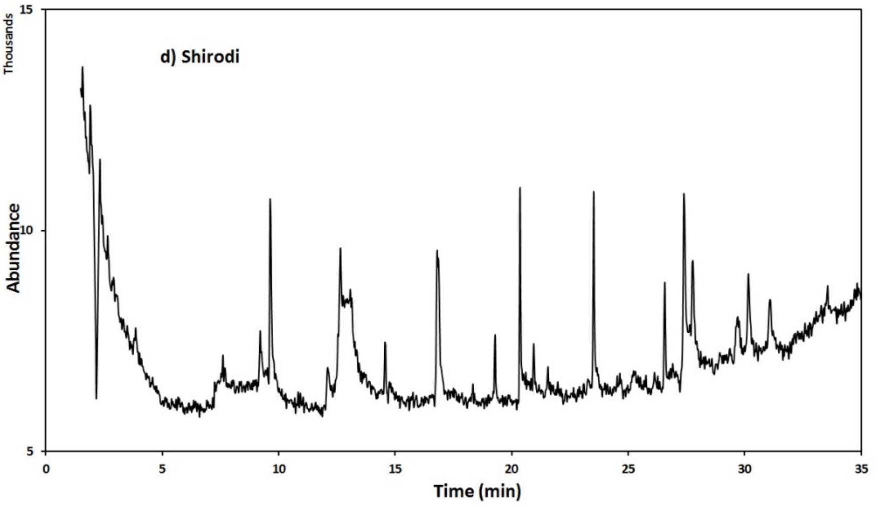


Fig. 1. Total ion current chromatograms for the flavor volatiles of the five types of rice cultivars ((a) Domsiah, (b)Deylamani, (c) Hashemi, (d) Shirodi and (e) Tarom) (The flow-rate of helium was 1.0 ml. min−1 with split ratio of 5:1. Oven program temperature was 40 ◦C (hold 7 min), rate 8 ◦C/min up to 240 ◦C (hold 10 min).



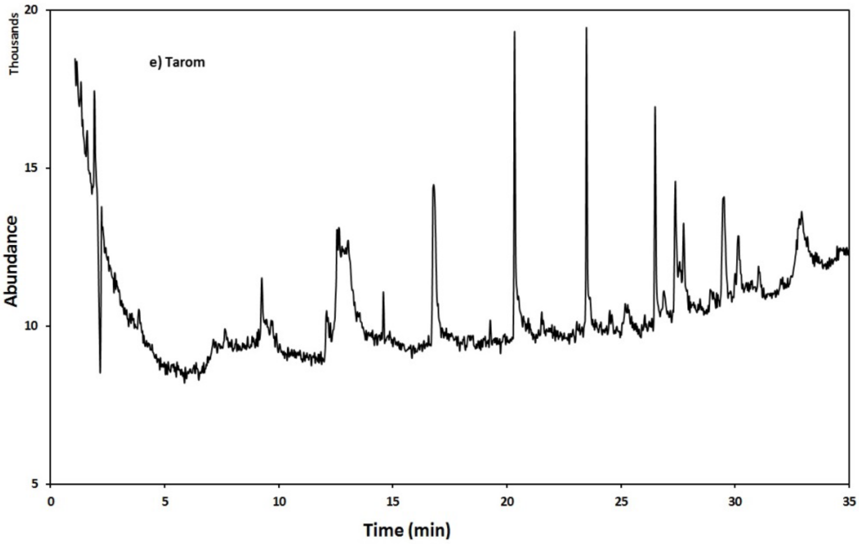


Fig. 1: continued

A total of 49, 31, 29, 16 and 24 peaks were assigned to Domsiah, Deylamani, Hashemi, Shirodi and Tarom, respectively (table 1). Table 1 lists the names of these compounds and their retention times. The main identified volatiles in these samples were belonged to the aldehydes, ketones, alcohols and heterocyclic class of chemicals, as well as fatty acids and esters, phenolic compounds, hydrocarbons, etc.

Among identified chemicals, the relative abundance of main detected species are;pentadecylamine(4.26%) and dibromodifluoro-methane (4.19%) in Domsiah sample , (Z)-9-octadecenoic acid methyl ester (20.25%) and 3-methyl-benzenamine (10.14%) in Deylamani sample, tert-butyl-benzene (38.37%) and 1,4,5,8-tetraamino-9,10-anthracenedione (13.22%) in Hashemi sample, p-aminotoluene (28.96%) and N-(2-methoxyphenyl)-3-oxo-butanamide (15.19%) in Shirodi sample and carbromal (29.84%) and isothiocyanato-methane (18.80%) in Tarom sample.

Table 1 Major volatiles identified in the rice samples determined by HS-SPME-GC-MS

No.	Sample	R_t	m/z	Abundance % Domsiah	Deylamani	Hashemi	Shirodi	Tarom
1	Carbromal	1.0000	208.0	-	-	2.14	-	29.84
2	Digitoxin	1.0359	355.0	2.50	7.19	-	-	-
3	(Z)-9-Octadecenoic acid methyl ester	1.0372	55.0	-	20.25	-	-	-
4	1-Chloro-2-nitro-Propane	1.1243	77.0	-	5.65	3.35	-	-
5	Ethanethioamide	1.1245	77.0	-	-	-	1.19	-
6	1,2-Dichloro-1,1,2,2-tetrafluoro-ethane	1.1618	151.0	-	-	0.52	-	-
7	N-(2-Methoxyphenyl)-3-oxo-butanamide	1.3027	207.0	-	-	-	15.19	-
8	2,3-Dimethyl-2,3-Butanediol	1.3123	59.0	-	2.09	-	-	-
9	2,3,6-Trichlorobenzaldehyde	1.3139	207.0	-	5.10	5.98	-	2.83
10	Dibromodifluoro-Methane	1.5759	75.2	2.03	-	-	-	-
11	(2-Amino-5-chlorophenyl)(2-chlorophenyl)-methanone,	1.5941	265.0	-	0.94	-	-	-
12	Eicosane	1.8640	75.2	1.90	-	0.81	-	0.37
13	N,N-Dimethyl-methanamine N-oxide	1.9226	75.0	-	-	-	-	18.80
14	Isothiocyanato-methane	1.9242	75.0	-	-	-	13.20	-
15	Oxybenzone	2.0579	151.0	-	3.10	-	-	-
16	2-Methoxyphenothiazine	2.1167	229.0	-	-	-	-	7.83
17	Spiro[2.4]hepta-4,6-diene	2.1689	91.0	-	-	0.49	-	-
18	2,4,6-Trinitro-phenol	2.1996	75.2	1.40	-	-	3.18	-
19	Hexanal	2.5436	75.2	2.06	0.05	0.07	0.21	0.17
20	12,13-Dihydro-7H-dibenzo(a,g)carbazole	2.6309	269.0	-	1.49	-	-	-
21	2,4,6-Trinitro-phenol	3.1870	75.2	1.73	-	-	-	-
22	(4-Nitrophenyl)phenyl-diazene	3.5173	75.2	1.70	-	-	-	-
23	Pyridine	3.9094	79.0	1.66	3.85	1.94	2.79	3.02
24	2-Hydroxy-3-(3-methyl-2-butenyl)-1,4-naphthalenedione	3.9374	75.2	1.67	-	-	-	-
25	Trichloro-methanesulfenyl chloride	3.9689	151.0	-	0.56	-	-	-
26	Acetyl bromide	5.0548	75.2	1.47	-	-	-	-
27	Benzoic acid 2-bromoethyl ester	5.7271	75.2	1.55	-	6.93	-	-
28	Phenyl-phosphoramidic acid diphenyl ester	6.2313	75.2	1.60	-	-	-	-
29	Pentadecylamine	8.0129	75.2	1.42	-	-	-	-
30	2,2-Dimethyl-3-hexanone	9.1739	57.0	-	0.36	-	-	-
31	2-hydroxy-propanenitrile	9.1878	56.0	-	0.07	-	-	-
32	(Butoxymethyl)-oxirane	9.2038	55.0	-	-	-	3.70	-
33	p-Aminotoluene	9.6376	79.0	-	-	-	28.96	-
34	3-Methyl-benzenamine	9.6654	107.0	-	10.14	-	-	-
35	o-Toluidine	9.7349	107.0	-	-	0.30	-	1.21
36	(E)-9-Tetradecen-1-ol	12.0995	55.0	-	-	-	-	10.12
37	(Z)-11-Hexadecen-1-ol	12.1616	55.0	-	-	5.09	-	-
38	9,10-Dihydro-9,9-dimethyl-acridine	12.4077	281.7	2.83	-	-	-	-
39	Vincamine	12.4260	207.0	-	-	-	-	4.42
40	5-Nitro-1H-Isoindole-1,3(2H)-dione	12.5385	193.0	-	0.65	-	-	-
41	2-Methyl-pentadecane	12.5420	193.0	-	-	-	3.90	-
42	Tributyl phosphate	12.5678	99.0	-	1.55	0.65	-	-
43	1-(Bromomethyl)-2-chloro-benzene	12.6023	208.0	-	-	0.32	-	3.52
44	7-Benzoyloxybicyclo[2.2.1]hepta-2,5-diene	12.7328	105.0	-	0.13	1.42	-	
45	2,4-Dinitro-phenol acetate	13.0032	226.0	-	-	-	-	0.54
46	1-Octene	13.6650	75.2	1.54	-	-	-	-
47	Sulfisoxazole acetyl	14.1912	267.0	1.57	0.07	-	-	-
48	Dienochlor	14.4740	404.0	1.52	0.15	-	-	-
49	2,2-Dimethyl-3-hexanone	14.5540	57.0	-	0.63	-	-	-
50	4-Amino-3-nitro-benzoic acid	14.5552	75.2	1.52	-	-	-	-
51	Nonane	15.2005	75.2	1.44	-	-	-	-
52	m-(Chlorosulfonyl)benzoic acid	16.3718	220.0	-	-	0.40	-	0.98
53	Methoserpidine	16.4620	355.6	2.38	-	-	-	-
54	Triphenyl phosphate	16.5459	327.0	-	-	-	0.85	0.57

55	Diisopropyl methanephosphonate	16.8629	97.0	-	0.30	-	2.54	-
56	2-Propenoic acid oxiranylmethyl ester	16.8806	75.2	1.80	-	-	-	-
57	Undecane	17.0801	75.2	1.44	-	-	-	-
58	3-(4-Methoxyphenoxy)-benzaldehyde	17.4602	75.2	1.44	-	-	-	-
59	Benzylcarbamate	17.7987	75.2	1.40	-	-	-	-
60	Azulene	17.8797	75.2	1.43	-	-	-	-
61	1,4,5,8-Tetraamino-9,10-anthracenedione	18.2123	75.2	1.57	-	13.22	-	3.59
62	Tridecane	19.2673	57.0	1.64	0.45	2.23	3.63	3.19
63	Pentadecylamine	20.3022	429.8	4.26	-	-	-	-
64	Dibromodifluoro-methane	20.3303	429.7	4.19	-	-	-	-
65	3-(4-Methoxyphenoxy)-benzaldehyde	21.3933	79.3	2.93	-	-	-	-
66	Pentadecylamine	21.6337	79.3	2.15	-	-	-	-
67	Leucinocaine	22.3378	75.3	1.73	-	-	-	-
68	1-(Bromomethyl)-4-fluoro-benzene	22.5332	75.3	1.71	-	-	-	-
69	6-Methyl-6-hepten-4-yn-2-ol	23.2295	75.3	1.63	-	-	-	-
70	3-Phenyl-pyridine,	23.2658	75.2	1.65	-	-	-	-
71	4-(Butylnitrosoamino)-1-butanol	23.4221	75.3	1.72	-	-	-	-
72	Phenol	23.6360	94.0	1.71	0.78	2.81	1.62	0.73
73	2-Methoxy-2-methyl-propane	23.9892	73.0	-	-	0.39	-	-
74	Kepone	24.0165	272	1.64	1.52	-	-	-
75	1,3-Dihydro-2H-Inden-2-one	24.3895	75.2	1.52	-	-	-	-
76	N,N-diethyl-4-methyl-benzamide	24.9909	75.3	1.60	-	-	-	-
77	Cyclopentadecanol	26.4772	208.0	-	-	-	-	1.88
78	Isobutane	26.6432	58.0	-	-	-	-	0.29
79	2-Ethyl-butanal	26.7879	100.0	-	0.85	-	-	-
80	3,5-Diamino-1,2,4-triazole	26.7973	100.0	-	0.21	0.52	1.39	0.74
81	3-Methyl-3-pentanol	26.8922	73.0	-	-	1.53	-	-
82	N-(1,1,3,3-tetramethylbutyl)-acetamide	26.9678	75.3	1.68	-	-	-	-
83	1-(Phenylazo)-2-naphthalenol	27.3735	221.0	-	9.85	-	-	-
84	2-Chloro-dibenzo[b,e][1,4]dioxin	27.6431	221.0	-	-	-	-	0.17
85	Drometrizole	27.7452	207.5	3.24	-	-		-
86	N-butyl-1-butanamine	27.7493	129.0	2.77	2.75	0.06	6.63	0.15
87	Dibutyl methanephosphonate	29.2769	209.0	-	-	0.79	-	-
88	2-[[4-(dimethylamino)phenyl]azo]-benzoic acid	29.3393	75.3	1.73	-	-	-	-
89	2,4-Bis(1,1-dimethylethyl)-phenol	29.5364	191.0	-	-	1.64	-	-
90	Ephedrine	29.8872	105.0	-	-	0.53	-	-
91	9,10-Dihydro-9,9-dimethyl acridine	29.9619	194.0	-	-	5.79	-	-
92	Tert-butyl-benzene	30.0898	119.0	-	-	38.37	-	2.56
93	Bis(p-tolyl)-ethandione	30.1227	119.0	-	3.60	-	10.90	-
94	2-Toluic hydrazide	30.1509	119.0	-	-	-	-	3.10
95	N-methyl-1-octanamine	31.0343	143.0	-	1.23	-	-	-
96	Pentadecylamine	31.4091	75.3	1.76	-	-	-	-
97	Benzenecarboxylic acid	31.8434	105.0	-	1.45	-	-	-
98	Benzophenone	32.0900	105.0	-	-	0.38	-	-
99	Benzofuran	32.5730	75.3	1.82	-	-	-	-
100	Phenyl 3-pyridyl ketone	33.1282	75.3	2.09	-	-	-	-
101	Triethanolamine	33.4813	149.0	-	-	1.18	-	-
102	3-Bromo-1-propene	33.8827	75.3	1.93	-	-	-	-
103	1,1,1-Trinitro-ethane	34.3688	75.3	1.95	-	-	-	-

Of particular significance, flavor volatiles are the especially essential factors in evaluation of the rice quality because aroma is rated as the highest desired trait [22]. The flavor volatiles were representative of their chemical classes based on their importance as potential impact odorants for these rice cultivars, which were selected as the flavor molecular markers. An understanding of the similarities and differences of these rice cultivars has been achieved through a comparison of the volatile components. As illustrated in Fig. 1, some differences were found in the profiles of flavor volatiles. The major volatile components in all cases were acids, aldehydes and ketones. Five chemical volatiles are same in all samples, which are; hexanal, pyridine, tridecane, phenol and N-butyl-1-butanamine. By concerning the flavor volatile profiles of the five rice cultivars, it was concluded that they were quite different and only minor similarity in their composition. Because of different indicator components in five rice samples, aroma of various rice samples is different from each other.

Apart from these observations, there were some differences in the composition of the minor volatiles among the five different rice cultivars, as presented in Table 1. For example, some minor volatiles such as 2,4,6-trinitro-phenol, acetyl bromide, benzylcarbamate, azulene, undecane, pentadecylamine and 1-octadecanol were detected only in Domsiah, prednisolone acetate, 1-chloro-2-nitro-propane, 2,3-dimethyl-2,3-butanediol, 2,2-dimethyl-3-hexanone and phenol were detected only in Deylamani and o-toluidine, (Z)-11-hexadecen-1-ol, 1-(bromomethyl)-2-chloro-benzene were identified just in Hashemi and ethanethioamide, p-aminotoluene and 2-methyl-pentadecane are only in shirodi sample and (E)-9-tetradecen-1-ol, 2,4-dinitro-phenol acetate, cyclopentadecanol and isobutane are detected only in Tarom sample, which reflected some differences in the profiles of flavor volatiles among the five rice cultivars.

4. Conclusion

This study has demonstrated the application of a headspace SPME method in conjunction with GC–MS to analyze the flavor volatiles in five rice cultivars. A total of 49, 31, 29, 16 and 24 peaks were assigned to Domsiah, Deylamani, Hashemi, Shirodi and Tarom, respectively. The major volatile components in all cases were acids, aldehydes and ketones. Five chemical volatiles are same in all samples, which are; hexanal, pyridine, tridecane, phenol and N-butyl-1-butanamine. Concerning the flavor volatile profiles of the five rice cultivars, they were quite different and only minor similarity in their composition. Because of different indicator components in five rice samples, aroma of various rice samples is different from each other.

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