Research Article

Comparison of Dynamic Headspace Trapping on Tenax TA and Headspace Stir Bar Sorptive Extraction for Analysis of Grilled Chicken (Yakitori) Volatiles

Panita Ngamchuachit, Yurika Kitai and Suwimon Keeratipibul*
Department of Food Technology, Faculty of Science, Chulalongkorn University, Bangkok, Thailand

Preecha Phuwapraisirisan
Department of Chemistry, Faculty of Science, Chulalongkorn University, Bangkok, Thailand

* Corresponding author. E-mail: suwimon.k@chula.ac.th

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Abstract

Two headspace extractions, dynamic headspace trapping on Tenax TA (DHS-Tenax TA) and headspace stir bar sorptive extraction (SBSE), were performed to compare the volatile profile in Japanese commercially processed grilled chicken produced in two processing steps: after dipping in thin sauce and then grilling (S1) and S1 after dipping in thick sauce (S2). Eighty-two volatile compounds were identified. Although DHS-Tenax TA extracted a larger number and higher amounts of volatile compounds for MS than SBSE, SBSE detected more aromas at the olfactometer than DHS-Tenax. Lipid oxidation products (aldehydes, alcohols, ketone, and phenols) and Maillard reaction products (Strecker aldehydes, nitrogen- and sulfur-containing compounds, furan(one)s, and pyrrole) are major compounds. DHS-Tenax TA was more suitable for the detection of volatile compounds from the Maillard and Strecker degradation products, such as pyrazines and furan(one)s, whereas SBSE was adequate for hydrocarbons and terpenes, and aroma compounds that were not detected by MS. Furan(one)s were only identified in S2 samples due to the presence of sugars in the thick sauce.

Keywords: Grilled chicken, Yakitori, Volatile compounds, Dynamic headspace, Headspace stir bar sorptive extraction

1 Introduction

Chicken meat consumption has grown significantly over the past fifty years. Chicken meat, the cheapest commercial livestock meat, is more superior to red meat due to several other reasons including its health benefits, as it contains less fat and high protein, easy to handle portions and less religious barriers [1]. It can be processed and prepared into various types of ready-to-eat products. Thailand is one of the world major producers of frozen ready-to-eat chicken products, which are exported to several countries, particularly Japan. One of the main frozen products exported to Japan is a Japanese-style grilled chicken skewer called *yakitori*, which is one of the most popular Japanese grilled food. Yakitori is charcoal-grilled bite-sized chicken meat skewed with a bamboo stick. The chicken meat is marinated with various seasoning ingredients, such as soy sauce, sugar, Japanese rice wine (Mirin), cooking sake, garlic, and ginger. The charcoal grilling imparts a desirable smoky flavor to the chicken meat, which influences consumer preference. The aroma characteristics of yakitori play the most important role in consumer acceptance.

Although sensory evaluation by trained panelists is an important tool for assessing the flavor quality of food

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products [2], frequent disagreements concerning product flavor consistency between the food manufacturers and customers occur. Therefore, the analysis of aroma-active volatile compounds as quality indicators could promote better quality control. Several conventional extraction methods, such as steam distillation, solvent extraction, and Soxhlet extraction, have been utilized to obtain volatile extracts. However, these extraction methods are extensively time-consuming which might not be suitable for a large number of samples. In addition, loss of highly volatile compounds, decomposition of volatile compounds, and heating-induced artifact formation can occur during extraction. Therefore, headspace extraction, such as dynamic headspace (DHS) and solid-phase microextraction (SPME), has been widely used for extracting volatiles in food applications owing to its simplicity, speed, solvent-free procedures, lack of contamination by non-volatile compounds, and lack of artifact formation [3]. In DHS, during sample extraction, a carrier gas is purged through the sample and causes volatile compounds to be released from the sample into the gas stream in the headspace. Subsequently, the volatile compounds exiting the sample container are trapped in an adsorptive trap for subsequent gas chromatography analysis. Tenax sorbent is the most common due to its wide volatility range, high temperature stability, low water affinity and long shelf life [4]. For SPME, the principle of headspace SPME is the equilibrium partitioning between the analytes and the coating fiber [5]. However, there is a limit to extraction because of the small quantity of the extraction phase. Based on the SPME theory, a headspace stir bar sorptive extraction (SBSE) has been developed [6]. In this extraction technique, a stir bar coated with polydimethyl siloxane (PDMS) is used as the volatile absorbent. In general, SBSE improves the extraction capability because of its 50- to 250-times larger extraction phases than that of SPME. Recently, the SBSE technique has been applied to meat products, such as grilled pork, grilled beef, and cooked ham [7]–[9].

Therefore, the objectives of this study are to compare DHS-Tenax TA and SBSE techniques and propose a simple and reliable method for industrial application of aroma compound extraction from Japanese-style grilled chicken. In addition, the volatile profiles of a Japanese commercially processed grilled chicken meat obtained from gas chromatography–olfactometry–mass spectrometry (GC-O-MS) are reported for applications in process control.


(S1) (S2)

Figure 1: Japanese commercially processed grilled chicken samples (Yakitori) used in the present study. (S1) Grilled with thin sauce, (S2) Grilled with thick sauce.

2 Materials and Methods

2.1 *Sample preparation*

Two types of frozen Japanese-style grilled chicken meat (500 g per each) with different dipping sauce (S1 and S2) were obtained from a Thai chicken meat factory in 2018. In the factory, steamed meat was skewered with a bamboo stick, dipped in thin seasoning soy sauce, and manually grilled over charcoal. The skewered meat was turned every 30 s for 2 min and dipped in thick seasoning sauce. The meat was frozen at –25°C and sent to our laboratory. Sample 1 (S1) was a chicken meat dipped in thin seasoning soy sauce without the final dip in thick seasoning sauce. Sample 2 (S2) was a chicken meat which was dipped in thin sauce and in thick sauce after grilling. The appearance of the products is shown in Figure 1. The samples were kept in a freezer (–20°C) until analysis. Fifty grams of each sample was ground using a grinder (LAB MILL; OSAKA CHEMICAL Co., Ltd. Japan). Five grams of ground sample was spiked with 10 µL of 2-methyl-3-heptanone (10 mg/L in methanol) as an internal standard and placed in a 20 mL vial with a polytetrafluoroethylene (PTFE)/silver screw cap. The sample in the vial was heated in a water bath (100°C). The heating time was varied between 1, 3, 5, 7, and 10 min.

2.2 *Dymamic headspace trapping on Tenax TA (DHS)*

The volatile compounds in the headspace were trapped in a Tenax TA adsorbent tube (60/80 mesh; Gerstel, Mülheim an der Ruhr, Germany). The trapping volume was 1,800 mL with a nitrogen purge stream at a flow rate of 30 mL/min. The sampling temperature was at 55°C, and the adsorbent tube temperature was set at 65°C to prevent moisture condensation. After sample purging, the tube was desorbed in a Gerstel thermal desorption unit (TDU) on a GC-MS.

2.3 *Headspace stir bar sorptive extraction (SBSE)*

Stir bars (Twister bar; PDMS, 0.5 mm film thickness × 10 mm length) were obtained from Gerstel. For each sample, two stir bars were installed at the headspace using a headspace insert vial before heating the sample in a water bath as described in Section Sample preparation. The stir bars were further exposed to the sample at 55°C for 60 min [8]. After sampling had finished, the water droplets on the stir bars were removed with a lint-free tissue and then the stir bars were placed in a TDU tube for the analysis of the volatile compounds. The volatile compounds from the stir bars were desorbed in the TDU attached to a GC-MS.

2.4 *Gas chromatography–olfactometry–mass spectrometry analysis (GC-O-MS)*

The volatile compounds were analyzed using a GC-MS (7890B GC and 5977B MSD; Agilent Technologies Inc., Santa Clara, CA, USA) equipped with an olfactory detector port (ODP3; Gerstel). The volatile separation was conducted using an HP-5MS column (30 m × 0.25 mm internal diameter, 0.25 µm film thickness; Agilent). Analytes were introduced to the TDU, which was run in splitless mode at an initial temperature of 25°C, followed by ramping at 400°C/min to 300°C and then holding at 300°C for 5 min. The analytes were trapped at 10°C in the Gerstel cooled injection system (CIS) inlet with a Tenax TA liner, which was run in splitless mode. The initial temperature of the CIS was 10°C, and it was then raised to 300°C at a rate of 10°C/s and held at this temperature for 5 min. The transfer temperature between the TDU and CIS was maintained at 300°C. The GC oven temperature was 40–170°C at a ramp rate of 4°C/min, which was then increased to 250°C at a ramp rate 10°C/min and held for 5 min. The flow rate of the helium carrier gas was 2 mL/min. The effluent of the capillary column was split 1:2 by volume and transferred to a single quadrupole mass detector (MSD) and ODP. The ion source temperature was 230°C. The MS transfer line temperature was 280°C. The electron impact ionization energy was 70 eV. The mass scan range was between 35 and 350 amu. The ODP was maintained at 200°C (ODP transfer line) and at 200°C (ODP mixing chamber). Humidified air was supplied (50 mL/min) to the ODP to prevent drying of the nasal mucosa. The odor-active compounds were perceived by three trained panelists in separate time. The panelists were also required to note the perceived odor characteristic and the retention time of odor-active compound individually. Each sample was performed in duplicates by each panelist.

2.5 *Volatile compound identification and semi-quantification*

The tentative identification of volatile compounds was carried out by matching mass spectra with those of references obtained from the NIST MS 14.0 library (National Institute of Standards and Technology, Gaithersburg, MD, USA), and comparing linear retention index (LRI) and odor descriptions. The LRIs of the compounds were calculated by the Kovats method using a homologous series of n-alkanes (C_8–C_{20}) under the same chromatographic conditions, which were calculated using the following Equation (1):

$$LRI(x) = 100 \times z + \left(\frac{RT(x) - RT(z)}{RT(z+1) - RT(z)} \right) \tag{1}$$

where $LRI(x)$ is the retention index of unknown compound (x), $RT(z)$ is the number of carbon atoms of standard n-alkane eluted before unknown compound (x), $RT(x)$ is the retention time of unknown compound (x), and $RT(z)$ and $RT(z+1)$ are the retention times of the standard n-alkanes eluted before and after unknown compound (x), respectively. The internal standard (IS) method was used to semi-quantify the volatile compounds. The concentration calculated as 2-methyl-3-heptanone equivalents using the following Equation (2):

$$C_I = \frac{X_{IS} \times PA_I}{PA_{IS}} \quad (2)$$

where C_I is the concentration of interest compound, C_{IS} is the concentration of internal standard, PA_I is the area of interest compound, and PA_{IS} is the peak area of internal standard, respectively.

3 Results and Discussion

3.1 *Sample preparation*

In general, the reheating process used by consumers for frozen Japanese-style grilled chicken product is microwave or boil-in-bag heating. In this study, each ground frozen sample was reheated in a closed 20 mL glass-vial with a metal cap in a 100°C water bath because the glass vial has a metal cap that could not be reheated in the microwave. The aroma characteristics and intensities of the samples reheated in a water bath for five different reheating times (1, 3, 5, 7, and 10 min) were compared to those obtained by microwave and boil-in-the bag heating by three trained panelists to ensure that the samples reheated in the water bath had the same aroma characteristics and aroma intensity as the product reheated by microwave heating. As a result, an optimal reheating time of 7 min was chosen because of the similarity of the aroma characteristics and intensity of this sample to those of the microwave reheated product. For reheating times of less than 7 min, the aroma intensity was quite low. However, the 10 min reheating time yielded burnt and rancid aromas.

3.2 *Comparison of DHS-Tenax TA and SBSE techniques for the analysis of volatile compounds*

The results from two different headspace extraction techniques, DHS-Tenax TA and SBSE, for the analysis of the Japanese grilled chicken samples were shown in Table 1. DHS-Tenax TA shows a greater number and higher amounts of volatile compounds extracted from both grilled chicken S1 and S2 than the SBSE technique. Most of the volatile compounds were compounds formed by lipid oxidation (48), including aldehydes, alcohols, ketones, hydrocarbons, esters, and phenols (Table 2). In addition, terpenes (9) and volatile compounds (20) formed from the Maillard reaction and the Strecker degradation, including Strecker aldehydes, N- and S-containing compounds, furan(one)s, and pyrrole, were also detected.

Table 1: Volatile compounds of grilled chicken samples extracted by DHS-Tenax TA and SBSE techniques

No	RI [1]	RI-ref [2]	Name	Concentration (μg/kg)				LogK_{ow} [3]	Odor Note	Identification Methods [4]	Olfactometry Detected by Panelists			
				Thin Sauce (S1)		Thick Sauce (S2)					Thin Sauce (S1)		Thick Sauce (S2)	
				Tenax TA	SBSE	Tenax TA	SBSE				Tenax TA	SBSE	Tenax TA	SBSE
Aldehydes														
1	<800	640	3-Methyl-butanal	11.79±2.91	4.46±2.45	6.49±1.86	30.20±6.68	1.27	Sour	MS, O	✓	✓	✓	✓
2	<800	651	2-Methyl-butanal	14.70±1.41	4.53±1.49	8.78±2.79	20.98±3.22	1.27	Sour, ferment	MS, O	✓	✓	✓	✓
3	800	800	Hexanal	48.40±2.56	28.02±3.00	6.76±2.21	8.00±2.19	1.78	Green	MS, RI, O	✓	✓	✓	✓
4	903	903	Heptanal	2.69±1.87	0.39±0.25	0.43±0.13	2.03±0.93	2.44	Fresh	MS, RI, O	✓	✓	✓	✓
5	962	960	Benzaldehyde	4.06±2.98	5.30±2.39	3.40±0.05	5.31±3.25	1.48	Ferment	MS, RI, O	✓	✓	✓	✓
6	1004	1001	Octanal	5.76±1.92	1.87±1.27	2.27±0.83	4.56±0.41	2.95	Fresh, soap	MS, RI, O	✓	✓	✓	✓
7	1104	1102	Nonanal	9.15±2.90	14.16±7.71	4.46±2.08	12.10±2.84	3.46	Fresh	MS, RI, O	✓	✓	✓	✓
8	1143	1147	2-Nonenal	–5	-	-	-		Old, wax	RI, O	✓	✓	✓	✓
9	1208	1207	Decanal	0.82±0.25	2.44±1.28	0.28±0.10	1.47±0.11	3.97	Vegetable	MS, RI, O		✓		
10	1224	1224	Cumin aldehyde	-	-	-	-		Dry chili	RI, O		✓		✓
11	1252	1262	2-Decenal	-	-	-	-		Fresh	RI, O	✓	✓	✓	✓
12	1360	1350	2-Undecenal	-	-	-	-		Waxy	RI, O	✓	✓	✓	✓
			total aldehydes	**97.38**	**61.17**	**32.87**	**84.66**							
Alcohols														
13	<800	730	3-Methyl-1-butanol	18.15±8.05	18.52±10.55	18.86±8.00	17.43±1.41	1.16	-	MS				
14	981	980	1-Octen-3-ol	2.54±2.32	-	-	-	2.52	Mushroom, green	MS, RI, O	✓	✓	✓	✓
15	1040	1041	Acetophenone	-	-	-	-		Rose, bitter	RI, O	✓	✓	✓	✓

Table 1: Volatile compounds of grilled chicken samples extracted by DHS-Tenax TA and SBSE techniques (Continued)

No	RI[1]	RI-$_{ref}$[2]	Name	Concentration (µg/kg)				LogK_{ow}[3]	Odor Note	Identification Methods[4]	Olfactometry Detected by Panelists			
				Thin Sauce (S1)		Thick Sauce (S2)					Thin Sauce (S1)		Thick Sauce (S2)	
				Tenax TA	SBSE	Tenax TA	SBSE				Tenax TA	SBSE	Tenax TA	SBSE
16	1093	1182	Isopinocarveol	0.52±0.07	0.85±0.22	-	-	-	-	MS, RI				
17	1101	1100	Linalool	1.63±0.73	3.16±0.96	1.04±0.15	1.25±0.28	2.97	Wood	MS, RI, O	✓	✓	✓	✓
18	1112	1118	2-Phenylethyl alcohol	-	-	-	-		Bitter, woody	RI, O	✓	✓	✓	✓
19	1177	1173	Menthol	0.54±0.29	0.33±0.19	0.70±0.01	0.91±0.42	3.22	-	MS, RI				
20	1291	-	(S)-(+)-5-Methyl-1-heptanol	0.25±0.12	0.23±0.07	0.39±0.18	1.02±0.20	2.82	-	MS				
21	1211	1214	Isodihydrocarveol	-	-	-	-		Ferment, wood	RI, O		✓		
			total alcohols	**23.63**	**23.07**	**20.99**	**20.61**							
Ketones														
22	<800	680	Acetoin	-	-	25.07±4.62	7.25±1.19	-0.36	-	MS				
23	989	988	5-Hepten-2-one, 6-methyl-	1.11±0.45	0.32±0.06	-	-	1.947	-	MS, RI				
24	1036	1035	Cyclohexanone, 2,2,6-trimethyl-	0.78±0.22	3.87±1.83	-	-	2.41	-	MS, RI				
25	1064	1075	2-Hydroxy-3,4-dimethyl-2-cyclopenten-1-one	-	-	-	-		Sweet, caramel	RI, O	✓	✓	✓	✓
26	1094	1091	2-Nonanone	0.52±0.07	0.85±0.22	-	-	3.14	-	MS, RI				
			total ketones	**2.41**	**5.03**	**25.07**	**7.25**							
Hydrocarbons														
27	<800	769	Toluene	7.17±1.19	5.21±3.11	0.45±0.19	1.81±0.77	2.73	-	MS, RI				
28	800	800	Octane	1.44±0.84	1.34±0.15	0.36±0.17	1.01±0.46	5.18	-	MS, RI				
29	860	864	Ethylbenzene	0.61±0.30	0.65±0.17	-	0.42±0.14	3.15	-	MS, RI				
30	869	866	p-Xylene	0.52±0.24	1.63±0.22	1.24±0.38	2.57±0.79	3.15	-	MS, RI				
31	902	896	Nonane	0.07±0.01	0.28±0.12	-	-	5.29	-	MS, RI				
32	1025	1030	β-Cymene	0.18±0.11	0.69±0.51	-	-	4.50	-	MS, RI				
33	1099	1100	Undecane	0.41±0.01	0.60±0.26	0.55±0.11	3.57±1.10	6.31	-	MS, RI				
34	1170	1170	3-Methyl-undecane	1.07±0.44	1.16±0.90	-	-	6.67	-	MS, RI				
35	1191	1205	(E)-2-Dodecene	-	-	0.39±0.08	0.94±0.30	6.41	-	MS, RI				
36	1199	1202	Dodecane	4.40±2.23	1.38±0.72	1.88±0.26	5.40±0.84	6.10	-	MS, RI				
37	1262	1252	Hexyl-benzene	-	-	0.36±0.17	0.78±0.38	-	-	MS, RI				
38	1370	1369	3-Methyl-tridecane	0.28±0.16	0.08±0.01	-	-	7.68	-	MS, RI				
39	1399	1401	Tetradecane	1.53±0.36	0.30±0.01	0.79±0.18	1.78±0.32	7.20	-	MS, RI				
40	1465	1501	Pentadecane	0.30±0.01	4.15±1.35	0.08±0.02	0.15±0.05	8.35	-	MS, RI				
			total hydrocarbons	**17.98**	**17.47**	**6.10**	**18.43**							
Esters														
41	<800	600	Ethyl acetate	-	-	1.66±0.30	13.24±3.19	0.73	-	MS				
42	815	815	Ethyl lactate	-	-	33.61±13.54	-	-0.04	-	MS, RI				
43	909	910	Butyl propanoate	1.19±0.65	0.53±0.06		-	2.31	-	MS, RI				
44	1377	1375	2-Ethyl-3-hydroxyhexyl 2-methylpropanoate	0.53±0.29	0.70±0.17	0.23±0.01	1.02±0.25	2.772	-	MS, RI				
45	1383	1380	(Z)-3-Hexenyl hexanoate	0.34±0.14	0.07±0.02	0.15±0.21	0.29±0.09	4.44	-	MS, RI				
46	1599	-	Pentan-2-yl undecyl sulfite	1.10±0.58	5.40±0.57	1.01±0.58	1.87±0.32	-	-	MS				
47	1921	1927	Methyl hexadecanoate	2.83±0.31	1.58±0.49	0.98±0.40	4.19±2.22	7.38	-	MS, RI				
48	>1995	2023	Isopropyl palmitate	0.33±0.02	4.19±0.23	-	-	8.27	-	MS				
			total esters	**6.32**	**12.47**	**37.64**	**20.61**							

Table 1: Volatile compounds of grilled chicken samples extracted by DHS-Tenax TA and SBSE techniques (Continued)

No	RI [1]	RI-ref [2]	Name	Concentration (µg/kg)				LogK_{ow} [3]	Odor Note	Identification Methods [4]	Olfactometry Detected by Panelists			
				Thin Sauce (S1)		Thick Sauce (S2)					Thin Sauce (S1)		Thick Sauce (S2)	
				Tenax TA	SBSE	Tenax TA	SBSE				Tenax TA	SBSE	Tenax TA	SBSE
N-S-containing														
49	824	828	Methyl pyrazine	4.20±1.71	-	0.48±0.16	-	0.21	Potato	MS, RI, O	✓	✓	✓	✓
50	908	903	Methional	1.10±0.47	-	-	-	0.44	-	MS, RI				
51	912	911	2,5-Dimethyl-pyrazine	2.68±0.49	0.30±0.02	3.99±0.70	2.01±0.56	0.63	Roasty	MS, RI, O	✓	✓	✓	✓
52	970	971	Dimethyl trisulfide	-	1.48±0.22	-	-	1.93	-	MS, RI				
53	1043	1047	2-Methyl-3-ethyl-pyrazine	-	-	-	-		Cooked rice	RI, O	✓	✓		
54	1080	1081	3-Ethyl-2,5-dimethyl-pyrazine	0.60±0.21	0.10±0.04	0.47±0.10	0.63±0.08	2.07	Roasty	MS, RI, O	✓	✓	✓	✓
55	1100	1105	2-Acetyl-2-thiazoline	-	-	-	-		Cooked rice	RI, O	✓	✓	✓	✓
56	1243	1240	Benzothiazole	-	-	-	-		Rubber	RI, O			✓	✓
			total N-S containing compounds	**8.58**	**1.88**	**4.94**	**2.64**							
Furan(one)s														
57	808	804	Dihydro-2-methyl-3(2H)-furanone	-	-	13.35±7.46	-	-		MS, RI				
58	835	835	Furfural	-	-	23.75±11.02	-	0.41		MS, RI				
59	859	868	2-methyl-3-furan-thiol	-	-	-	-		Cooked chicken	RI, O	✓	✓	✓	✓
60	862	857	Furfuryl alcohol	-	-	6.23±3.20	-	0.28		MS, RI				
61	1044	1044	4-Hydroxy-5-methyl-3-(2H)-furanone	-	-	-	-		Sweet	RI, O	✓			
62	1138	1139	Ethyl-4-hydroxymethyl- 3(2H)-furanone	-	-	-	-		Caramel	RI, O			✓	✓
63	1150	1147	3-(Acetylthio)-2-methylfuran	-	-	-	-		Roast sesame	RI, O	✓	✓	✓	✓
64	1170	1170	2-Methyl-3-(methyldithio)-furan	-	-	-	-		Cooked chicken	RI, O	✓	✓		
65	1179	1180	S-(2-Furfuryl)-ethanethioate	-	-	-	-		Malty, roasty	RI, O	✓	✓		
			total furans	-	-	**43.33**	-							
Pyrroles														
66	915	923	2-Acetyl-1-pyrroline	-	-	-	-		Cooked jasmine rice	RI, O	✓	✓	✓	✓
Terpenes														
67	933	935	α-Pinene	0.63±0.03	0.48±0.13	-	-	4.83		MS, RI				
68	948	951	Camphene	1.22±0.53	16.81±4.01	0.41±0.13	2.93±0.51	4.35		MS, RI				
69	991	992	β-Myrcene	1.55±0.53	1.26±0.86	-	-	4.17		MS, RI				
70	1029	1028	D-Limonene	6.27±2.62	4.65±1.55	1.52±0.73	2.23±0.73	4.57		MS, RI				
71	1031	1038	Eucalyptol	4.20±1.12	4.16±1.79	3.75±0.67	13.52±5.60	2.74	Menthol like	MS, RI, O	✓	✓	✓	✓
72	1146	1146	Camphor	0.49±0.08	0.21±0.11	0.53±0.11	0.71±0.18	2.38		MS, RI				
73	1485	1483	α-Curcumene	0.90±0.52	0.34±0.15	0.19±0.03	1.05±0.45	6.02		MS, RI				
74	1499	1490	α-Zingiberene	1.00±0.40	4.90±0.30	1.68±0.52	1.93±0.65	6.38		MS, RI				
75	1527	1525	β-Sesquiphellandrene	0.59±0.29	1.57±0.57	-	-	6.52		MS, RI				
			total terpenes	**16.85**	**34.38**	**8.08**	**22.37**							
76	1085	1089	2-Methoxyphenol	-	-	-	-		Old, medicine	RI, O	✓	✓	✓	✓
77	1189	1192	o-Cresol	-	-	-	-		Woody, sharp	RI, O	✓	✓		

Table 1: Volatile compounds of grilled chicken samples extracted by DHS-Tenax TA and SBSE techniques (Continued)

No	RI [1]	RI-$_{\text{ref}}$ [2]	Name	Concentration (µg/kg)				LogK_{ow} [3]	Odor Note	Identification Methods [4]	Olfactometry Detected by Panelists			
				Thin Sauce (S1)		Thick Sauce (S2)					Thin Sauce (S1)		Thick Sauce (S2)	
				Tenax TA	SBSE	Tenax TA	SBSE				Tenax TA	SBSE	Tenax TA	SBSE
Unknowns														
78	<800		Unknown	-	-	-	-		Sour	O	✓	✓	✓	✓
79	<800		Unknown	-	-	-	-		Alkaline	O		✓		✓
80	1132		Unknown	-	-	-	-		Vegetable, green, bitter	O		✓		✓
81	1168		Unknown	-	-	-	-		Floral	O				✓
82	1185		Unknown	-	-	-	-		Medicine	O	✓	✓		
			total compounds	**173.15**	**155.47**	**179.02**	**176.57**		**39**		**31**	**35**	**27**	**31**

Semi-quantification values are expressed in µg/kg as the means ± standard deviation in triplicates. [1] Retention index on HP-5 MS column. [2] Retention indices from NIST14.0 mass spectral database. [3] LogK_{ow} value obtained from the good scents company (http://www.thegoodscentscompany.com). [4] MS, mass spectrum identified by NIST database; RI, retention index agreed with literature value; O, odor description of odor compounds identified by database (pherobase, thegoodscentscompany, flavornet). 5 - means not detected.

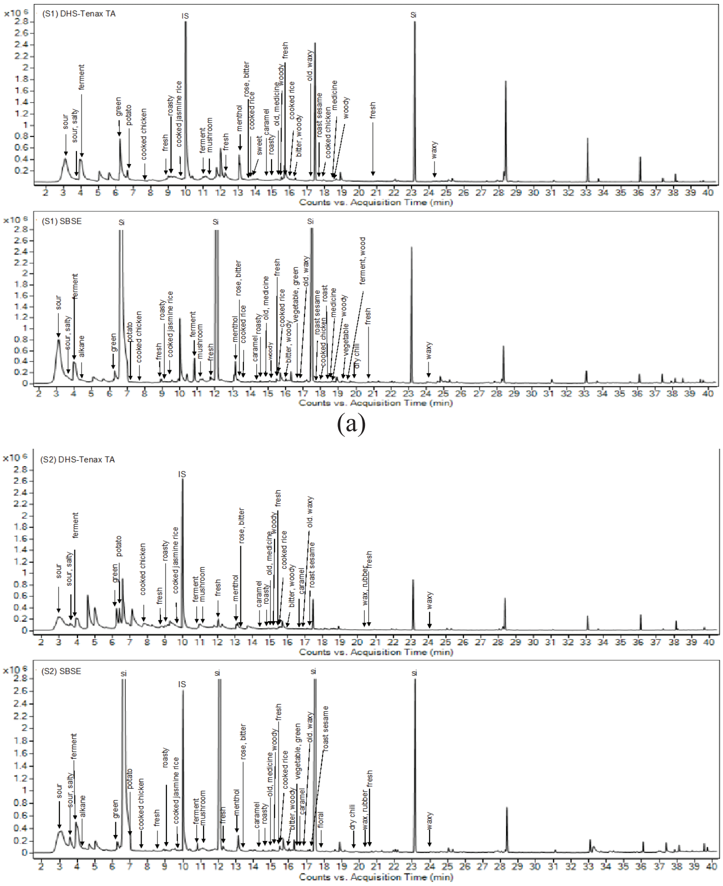
Table 2: The number of detected volatile compounds in Japanese grilled chicken extracted by dynamic headspace trapping on glass liner containing DHS-Tenax TA and SBSE

Volatile Compounds	Total by Chemical Classes	Thin Sauce (S1)		Thick Sauce (S2)	
		Tenax TA	SBSE	Tenax TA	SBSE
Lipid oxidation	48	41	43	34	35
Aldehydes	10	9	10	9	10
Alcohols	9	8	9	7	7
Ketones	5	4	4	2	2
Hydrocarbons	14	12	12	9	10
Esters	8	6	6	6	5
Phenols	2	2	2	1	1
Maillard reaction	20	14	13	14	11
Strecker aldehydes	2	2	2	2	2
N-S-containing compounds	8	6	6	5	5
Furan(one)s	9	5	4	6	3
Pyrrole	1	1	1	1	1
Terpenes	9	9	9	6	6
Unknown	5	2	4	1	4
Total compounds	82	64	65	54	52

In agreement with previous studies, the lipid oxidation and the Maillard reaction products mainly contribute to the aroma of grilled chicken. Wettasinghe *et al*. [10] detected lipid oxidation products, such as hexanal and nonanal, in roasted chicken . Maillard reaction products, such as pyrazines, thiazoles, and furans, were identified in cooked chicken and soy sauce [11]–[13].

In this study, not all identified volatile compounds contributed to the aroma notes. Figure 2 shows the extraction and separation patterns of volatile compounds and odor compounds based on the retention times obtained by GC-O-MS. Figure 2(a) and (b) show the chromatograms of the DHS-Tenax TA and SBSE extracts of grilled chicken samples S1 and S2, respectively. The results show that SBSE could extract more compounds contributing to aroma notes than DHS-Tenax TA. However, the quantity of these compounds was not high enough for MS detection. Therefore, to increase the quantity of compounds extracted, an additional aroma compound concentrating step, such as solvent assisted flavor extraction (SAFE), could be used to improve MS detection.

Interestingly, furan(one)s in S2 were detected by both DHS-Tenax TA and SBSE. From our results, although DHS-Tenax TA allowed the extraction of a higher concentration of furan(one)s, as shown by the peak intensity in the MS spectrum, SBSE allowed more detection of aroma notes by panelists at the olfactometer. The different extraction characteristics of DHS-Tenax TA and SBSE could be explained by the polarity of the compounds. SBSE extraction is based on the octanol–water partitioning coefficient (K_{ow}), which is proportional to the partitioning coefficient between the coated stir bar sorbent (polydimethylsiloxane (PDMS)) and the sample matrix [6]. A compound with a higher logK_{ow} value is more hydrophobic and has a higher affinity for PDMS. Furan(one)s have lower logK_{ow}



(a)

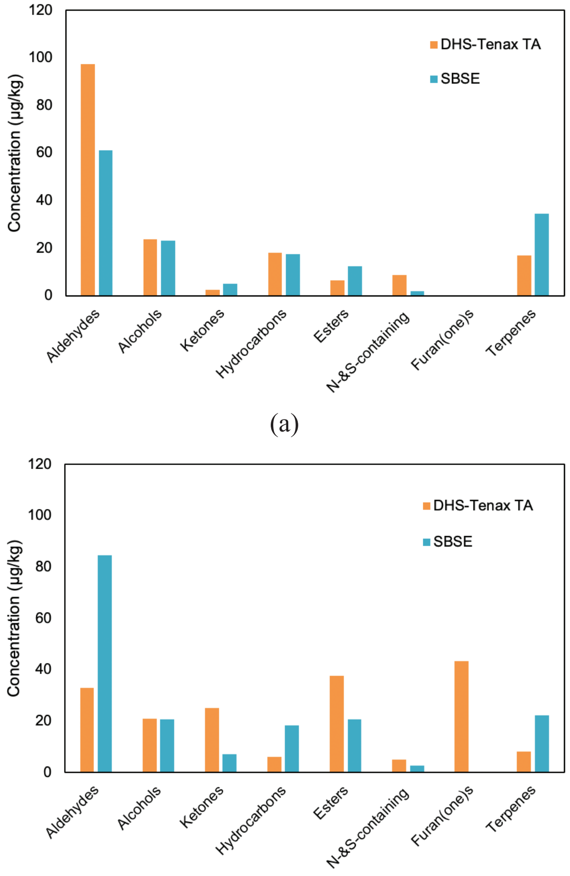
(b)

Figure 2: Total ion chromatograms of the grilled chicken samples extracted by DHS-Tenax TA and SBSE. Grilled chicken S1(a) and S2 (b) (IS = internal standard, Si=Siloxane).

value ($\log K_{ow}$ = 0.41 and 0.28 in furfural and furfuryl alcohol) than other compounds. Therefore, furans were less absorbed in the PDMS in SBSE. From our results, compounds with a $\log K_{ow}$ of less than 0.4 are more difficult to extract by SBSE. However, there is a slight anomaly, particularly for acetoin. Acetoin can be extracted by SBSE, even though it has very low $\log K_{ow}$ (–0.36). This may because of the large amount of acetoin in the samples. However, acetoin is extracted in three-times greater quantity by DHS-Tenax TA than SBSE.

The chemical profiles varied with the extraction technique (Figure 3). In the SBSE extract of both grilled chicken samples, aldehydes are the major volatile compounds, whereas DHS-Tenax TA extract showed aldehydes as the major compounds only in S1. The lower aldehyde extraction in S2 might be potentially from the interference of water during extraction because the sample had a higher water content (S2, 65.83%) than that of S1 (62.74%). Interestingly, only a 3% difference in the water content in the samples interfered with the extraction efficiency. Thus, preventing the introduction of water into the GC-MS is important.

Consequently, additional dry purging with an inert gas to remove water retained on the adsorbent tube is necessary for DHS-Tenax TA extraction [14]. This result implies that, for the application of DHS-Tenax TA, the composition of the food matrix, such as water and sugar contents, should be taken into consideration because the small compositional differences during each processing step could reduce the efficiency of the DHS-Tenax TA extraction.



(a)

(b)

Figure 3: Proportion of the chemical classes of volatile compounds in the grilled chicken samples extracted by DHS-Tenax TA and SBSE. Grilled chicken S1 (a) and S2 (b). Concentration are expressed in µg/kg as the means in triplicates.

3.3 *Aroma-active compounds in Japanese grilled chicken*

Aldehydes were the most abundant volatile compounds in the grilled chicken samples. Because of the effects of water interference on the efficiency of DHS-Tenax TA for the detection of aldehydes, the aldehyde results were investigated using SBSE extraction. As shown in Figure 4, the major aldehydes in S2, including 2-methyl-butanal and 3-methyl-butanal, were obtained in greater quantity than those in S1. These compounds are Strecker degradation products, which might be formed during the heating of the thick sauce in S2.

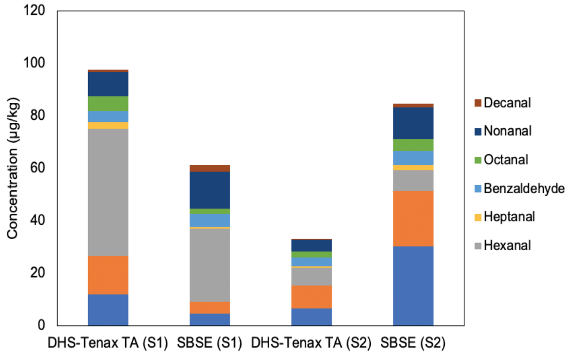


Figure 4: Proportion of the aldehyde compounds in grilled chicken samples extracted by DHS-Tenax TA and SBSE. Grilled chicken S1 and S2. Concentration are expressed in µg/kg as the means in triplicates.

The Maillard reaction products, nitrogen- and sulfur-containing compounds, and furan(one)s have been reported to be important contributors to the meaty flavor because of their low odor detection threshold [15]. Eight N- and S- containing compounds were identified. 2,5-Dimethyl pyrazine was one of the major compounds, and this compound contributes to nutty and roasted aroma notes in the grilled chicken. Liu [10] reported that among the Maillard reaction products, 2,5-dimethyl pyrazine was more abundant than the other alkyl pyrazines.

Methional and dimethyl trisulfide are potentially formed via the Strecker degradation of sulfur-containing amino acids, such as cysteine, cystine, and methionine [16] and contributes sulfurous, cooked potato, and onion aroma notes.

Nine furan(one)s, such as dihydro-2-methyl-3(2H) furanone, furfural, and furfuryl alcohol, were identified only in S2 sample, accounted for 24% of total volatile compounds in S2 sample, due to the presence of glucose and fructose sugars in the thick sauce. Furan(one)s contribute to the sweet, caramel, and burnt aroma notes of the samples. Furfural is formed by the thermal degradation of pentose sugars, such as xylose, ribonucleotides, and inosine-5'-monophosphate, in meat by reacting with leucine and isoleucine in the Maillard reaction [17], [18]. Furfuryl alcohol can be generated by the reduction of furfural [19].

One alcohol (3-methyl-1-butanol), one ketone (acetoin), and two esters (ethyl acetate and ethyl lactate) were detected at moderately high concentrations

(1.6–33.6 μg/kg). These compounds might be derived from the sauce ingredients because 3-methyl-1-butanol, ethyl acetate, and ethyl lactate have been detected in soy sauce [20], [21] and contribute to fruity aromas. Moreover, acetoin has been found in many fermented products, being generated by bacteria and yeasts [22] and contributing to buttery aromas.

Fourteen hydrocarbons were identified in the grilled chicken samples. It has been reported that toluene and ethyl benzene are generated from the pyrolysis of phenylalanine [23]. Even though several hydrocarbons in cooked chicken has been previously reported [10], [24], they are not the main contributors to the meaty aroma [12], [25].

Nine terpenes were identified in the grilled chicken samples. D-Limonene and eucalyptol have been previously identified in cooked chicken [24]. However, other terpenes are not generally found in cooked chicken. In general, terpenes are produced by plants. Therefore, they might arise from the Bamboo skewers used in this study. It has been reported that *α*-pinene, D-limonene, and other terpenes are present in bamboo [26]. Therefore, the terpenes might derive from the skewer material.

4 Conclusions

Two headspace extraction methods, DHS-Tenax TA and SBSE, were tested to identify the appropriate extraction method for the analysis of Japanese commercially processed grilled chicken meat at two different processing steps. The volatile and aroma profiles obtained from the grilled chicken samples varied with the extraction methods. DHS-Tenax TA was suitable for the detection of the Maillard and Strecker degradation products, such as pyrazines and furan(one)s. SBSE could extract more compounds that contributed to the product aroma than DHS-Tenax TA. Although the SBSE technique is easy to optimize and could be used with various products obtained during different food processing steps, the quantity of aroma compounds obtained was not sufficiently high for MS detection. Therefore, further studies on the concentration of volatile aroma compounds are needed. In addition, the optimization of the extraction conditions should be carried out to maximize the volatile aroma compound profile for better quality control and product consistency.

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