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Evaluation of volatile compounds in chicken breast meat using simultaneous distillation and extraction with odour activity value

MEHMET TURAN AYSELI – GOKHAN FILIK – SERKAN SELLI

Summary
Limited information is available about the volatile components of raw chicken meat; therefore the current study was aimed at chicken meat volatiles. In addition, odour activity values (*OAV*) were used to evaluate potent volatile components of the chicken meat. Simultaneous distillation and extraction with dichloromethane was used to obtain volatile components. The volatile compounds were analysed by gas chromatography-mass spectrometry. A total of 33 compounds were identified and quantified in the sample. Volatile acids, esters and alcohols were the dominant volatiles in the chicken breast meat. On the basis of *OAV*, the most important aroma compounds identified in the extract were hexanal and *(E)*-2-heptenal, which were described as the green-fresh odour and green-cheesy-fatty odour, respectively.

Keywords
chicken breast meat; aroma; simultaneous distillation-extraction; odour activity value

Chicken meat is certainly one of the most popular items in the human diet, being consumed all over the world as a nutritious food. Turkey is an important chicken meat-producing country with the total production quantity of chicken meat risen from 662 096 tons in 2000 to 1 550 578 tons in 2011. The United States of America is the world's largest chicken meat producer, and Turkey is ranked at the 7th place in the world according to United States Department of Agriculture [1, 2].

Aroma compounds play an important role in the organoleptic characteristics of foodstuffs [3–5]. Over 350 volatile compounds have been found in different chicken meats, with a wide content range varying from hundreds of micrograms or nanograms per kilogram level. Major classes of compounds identified in chicken volatiles include hydrocarbons, aldehydes, ketones, sulphur-containing compounds and heterocycles (furans, pyrroles, pyrazines, thiazoles, thiophenes, oxazoles and pyridines). Many of the compounds have relatively high odour thresholds and present little contribution to the overall aroma and no single compound has been identified as being primarily responsible for the aroma of cooked muscle foods [6–10].

Volatile compounds of chicken meat are dependent on key precursors and various factors including genetic factors, sex, age, diet and as well as different processing factors such as freezing, chilling, prepackaging, cooking, dehydration, irradiation and storage procedures [11]. The chemical reactions by which volatile compounds responsible for chicken aroma and flavour are formed include the Maillard reaction, Strecker degradation, lipid oxidation and degradation of thiamine [12–14]. Carbonyl compounds are a major class of flavour components identified in the cooked chicken meat. They are formed by peroxidation of unsaturated acyl lipids. Carbonyl compounds are important contributors to the "chicken-like" aroma, since their removal from the volatile fraction resulted in a loss of the "chicken odour" and in intensification of the meaty odour [15]. Lipid

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oxidation is well known as the cause of rancidity development but it can also contribute to desirable food flavours. The oxidation of lipids produces a wide range of aliphatic compounds, including both saturated and unsaturated hydrocarbons, alcohols, aldehydes, ketones, acids and esters, as well as some cyclic compounds, such as furans, lactones and cyclic ketones [11, 16, 17]. Saturated and unsaturated aldehydes bearing 6–10 carbons constitute the major part of the volatile compounds found in cooked meat and hence they probably play important role in meat aroma. The odour threshold values of aldehydes are generally lower than those of volatile compounds, thus they have an important potential effect on total flavour of chicken meat [8, 13, 18].

Most studies in the literature deal with the volatile profile of cooked [19, 20] and grilled [21, 22] meat products, and only few research studies have been focused on volatile compounds present in raw meat [23]. No work has yet been published to determine the volatile composition of chicken breast meat from Turkey. Therefore, the aims of the present study were to determine volatile composition of this meat, using simultaneous distillation and extraction, and to evaluate the potent aroma compounds of this sample using odour activity values (*OAV*).

MATERIALS AND METHODS

Chicken breast meat (5 kg) was obtained from a standard processing plant, Banvit chicken company (Balikesir, Turkey). Chicken were manually slaughtered the same day by immersing in ice-cold water and transported under ice in insulated polystyrene boxes. Upon arrival, the samples were transferred to the laboratory and sample portions of 100 g were prepared from distinct parts of chicken breast meat, put in odour-proof bags and stored at +4 °C until analysis.

Volatile compounds of the chicken breast meat were extracted by simultaneous distillation and extraction (SDE) in a Likens-Nickerson apparatus. This method was shown to be reliable for the extraction of volatile components of different meat species, namely, beef [3, 24], chicken [3, 7, 20] and fish species, such as rainbow trout (*Oncorhynchus mykiss*) [4, 5] or sea bream (*Sparus aurata*) [25].

Chicken breast meat was finely minced and homogenized during 1 min in a household blender (Arcelik, Istanbul, Turkey). Then, 100 g of the chicken breast meat and 60 ml of 30% NaCl aqueous solution containing 40 μg of 4-nonanol as internal standard were placed in a 500 ml round-bottom flask attached to the appropriate arm of the SDE apparatus. A 50 ml round-bottom flask containing 25 ml of dichloromethane was linked to the other arm of the SDE apparatus. The steams were cooled by circulation of polyethylene glycol at –5 °C. Contents in the sample and solvent flasks were heated to a boil using heating mantle with magnetic stirrer (Electrothermal Engineering, Stone, United Kingdom). The temperature of dichloromethane flask was maintained by a water bath at 50 °C. The distillation-extraction was continued for 3 h. The volume of the extract was reduced to 5 ml by evaporating the solvent by a Vigreux apparatus and then to 200 μl under a gentle stream of nitrogen. Sample was extracted in triplicate and the content of volatiles, as 4-nonanol equivalents, was obtained as a mean of three repetitions. The extracts were then stored at –20 °C in a glass vial equipped with a Teflon-lined cap until the analysis.

The gas chromatography (GC) system consisted of an Agilent 6890 chromatograph equipped with a flame ionization detector (FID) and Agilent 5973-Network-MSD mass selective detector (all from Agilent Technologies, Santa Clara, California, USA). Aroma compounds were separated on DB-Wax (30 m length × 0.25 mm i.d. × 0.5 μm thickness; J&W Scientific, Folsom, California, USA) column. A volume of 3 μl of the extract was injected in pulsed splitless mode (275760 Pa; 0.5 min). This mode was chosen to minimize artifact formation by thermal degradation of analytes in the injection port. Injector and FID detector were set at 270 °C and 280 °C, respectively. The flow rate of carrier gas (helium) was 1.5 ml·min^{-1}. The oven temperature of the DB-Wax column was first increased from 50 °C to 200 °C at a rate of 5 °C·min^{-1}, and then to 260 °C at 8 °C·min^{-1}, with a final hold at 260 °C for 5 min.

The mass spectrometry (electron impact ionization) conditions were as follows: ionization energy of 70 eV, mass range m/z of 30–300 Da, scan rate of 2.0 s^{-1}, interface temperature of 250 °C, and source temperature of 180 °C. The aroma compounds were identified by comparing their retention index and their mass spectrum on the DB-Wax column with those of a commercial spectral databases Wiley 6 (Wiley, New York, USA), NBS 75k (Gaithersburg, Madison, USA) and of the instrument's internal library created from the previous laboratory studies [25]. Some of the identifications were confirmed by the injection of the chemical standards into the GC-MS system. Retention indices of the compounds were calculated by using an *n*-alkane series.

RESULTS AND DISCUSSION

The volatile compounds identified in the chicken breast meat and linear retention index values on the DB-Wax column for these compounds are listed in Tab. 1. Mean contents of volatile compounds of triplicate extractions and standard deviations are reported. A total of 33 compounds were identified and quantified in the chicken breast meat. It contained 536.1 $\mu g \cdot kg^{-1}$ volatile compounds, which included volatile acids (8), esters (4), alcohols (8), ketones (4), aldehydes (4), volatile phenols (4) and terpene (1). Volatile acids were the dominant volatiles in the chicken breast meat as they accounted for the largest proportion (32.4%) of the total volatile compounds. Esters were the second largest (29%) volatile group in the chicken breast meat. Tab. 2 presents the potent volatile compounds identified in the chicken breast meat extract, their odour descriptions and *OAV* calculated from their contents and odour threshold values obtained from the literature. *OAV* defined as ratio of content to odour threshold, give an idea of the odour potency of a single odourant in a food itself, based on its odour threshold in the respective food matrix [9, 25]. Of all the volatile compounds determined, only those displaying *OAV* greater than 1 were deemed to contribute to overall chicken breast meat aroma.

It is well known that aldehydes play an important role in many meat species. Four aldehydes (hexanal, *(E)*-2-heptenal, 2-furaldehyde and 5-methyl furfural) were identified and quantified in the chicken breast meat extract. The total content of aldehydes was 23.4 $\mu g \cdot kg^{-1}$. Among the aldehydes, hexanal showed the highest content of 12.9 $\mu g \cdot kg^{-1}$ in the studied sample. This aldehyde was found in many previous studies and mainly derives from the oxidation of linoleic acid. Hexanal provides green and fatty character to different meat species [9, 15, 26–31]. Another aldehyde present at high content in the studied sample is *(E)*-2-heptenal (3.1 $\mu g \cdot kg^{-1}$), which could be produced from typical oxidation products of *n*-3 and *n*-6 polyunsaturated fatty acids (PUFA). *(E)*-2-heptenal was found in different meat species such as fish, duck, goat or chicken [9, 12, 27]. Two aldehydes were found to have *OAV* higher than 1, namely, hexanal and *(E)*-2-heptenal (Tab. 2). Because of their low odour threshold values, aldehydes have an important potential effect

Tab 1. Volatile compounds of raw chicken breast meat.

Compounds	*LRI*	Content [$\mu g \cdot kg^{-1}$]	Method of identification
Aldehydes			
Hexanal	1 091	12.9	LRI, MS, Std
(E)-2-Heptenal	1 339	3.1	LRI, MS, Std
2-Furaldehyde	1 440	2.0	LRI, MS, Std
5-Methyl furfural	1 566	5.4	LRI, MS, Std
Total		23.4	
Alcohols			
3-Penten-2-ol	1 177	3.8	LRI, MS, Std
Isoamyl alcohol	1 192	6.2	LRI, MS, Std
3-Pentanol	1 292	13.9	LRI, MS, Std
2-Hexanol	1 307	32.3	LRI, MS, Std
(E)-3-Hexen-1-ol	1 391	4.5	LRI, MS, Std
2-Furan methanol	1 640	3.8	LRI, MS, Std
Benzyl alcohol	1 861	2.7	LRI, MS, Std
Phenylethyl alcohol	1 870	66.9	LRI, MS, Std
Total		134.0	
Ketones			
3-Hydroxy-2-butanone	1 277	6.2	LRI, MS, Std
4-Nonanone	1 334	14.7	LRI, MS tent.
1-Hydroxy-2-pentanone	1 460	1.3	LRI, MS tent.
Cyclopenthyl methyl ketone	1 796	4.7	LRI, MS tent.
Total		26.9	
Esters			
Ethyl lactate	1 326	52.3	LRI, MS, Std
Diethyl succinate	1 682	31.2	LRI, MS, Std
Ethyl-4-hydroxybutanoate	1 783	2.5	LRI, MS, Std
Monoethyl succinate	2 377	69.6	LRI, MS, Std
Total		155.7	
Volatile phenols			
Benzophenone	1 628	0.8	LRI, MS, Std
Phenol	1 995	2.6	LRI, MS, Std
2,3-Dimethylphenol	2 104	3.1	LRI, MS tent.
4-Vinyl-2-methoxyphenol	2 168	8.4	LRI, MS, Std
Total		14.9	
Acids			
Acetic acid	1 473	6.9	LRI, MS, Std
Butanoic acid	1 636	2.7	LRI, MS, Std
3-Methyl pentanoic acid	1 670	3.6	LRI, MS, Std
Pentanoic acid	1 753	2.1	LRI, MS, Std
Octanoic acid	2 091	8.9	LRI, MS, Std
Nonanoic acid	2 157	8.2	LRI, MS, Std
Decanoic acid	2 293	11.9	LRI, MS, Std
Oleic acid	3 184	114.5	LRI, MS, Std
Total		173.9	
Terpenes			
DL-Limonene	1 203	7.3	LRI, MS, Std
Total		7.3	
General total		536.1	

LRI – linear retention index calculated on DB-Wax capillary column. Contents are expressed as the means of three repetitions.
Methods of identification: LRI – linear retention index, MS tent. – tentatively identified by mass spectrometry, Std – chemical standard. When only MS or LRI is available for the identification of a compound, it must be considered as an attempt of identification. Standard deviations for all aroma compounds were below 10%.

Tab 2. Odour activity values of potent volatile compounds in chicken breast meat.

Compound	Odour threshold [$\mu g \cdot kg^{-1}$]	Odour activity value	Odour description
Hexanal	4.5	2.9	Green, fresh
(E)-2-Heptenal	3.0	1.0	Green, cheesy, fatty
4-Vinyl-2-methoxyphenol	3.0	2.8	Spicy

Odour threshold values for hexanal and (*E*)-2-heptenal were taken from the literature SELLI and CAYHAN [25], for 4-vinyl-2-methoxyphenol from PINO [38].
Odour activity values were calculated by dividing the contents by the odour thresholds.

on total flavour of meat species [25]. The first has a green and fresh aroma and the second one has a fatty, green and cheesy aroma.

Chicken breast meat contained a larger number of acids, the total amount of volatile acids being 173.9 $\mu g \cdot kg^{-1}$. Within acids, oleic acid was found in the highest content (114.5 $\mu g \cdot kg^{-1}$). Several acids were previously detected in different meat species, such as acetic acid in cooked and raw chicken meat [11, 15, 32], butanoic acid in duck meat [9, 27], nonanoic acid in goat and fried chicken meat [12, 33], pentanoic acid in roasted chicken [33] and octanoic acid in roasted chicken [15, 27].

Esters ethyl lactate, diethyl succinate, ethyl-4-hydroxybutanoate and monoethyl succinate were detected in the chicken breast meat. The total content of esters was 155.7 $\mu g \cdot kg^{-1}$ (Tab. 1). These compounds might be products of esterification of the alcohols with carboxylic acids that are formed by microorganisms and by degradation of lipids [25]. Ethyl lactate was found naturally in small quantities in a wide variety of foods including wine, chicken, beer, fruits and soya products [34, 35].

Eight alcohols were identified and quantified in the chicken breast meat. It was noted that a larger number of alcohols such as benzyl alcohol, were identified in raw beef, cooked beef and chicken [11, 32], or phenyl ethyl alcohol in cooked chicken meat [11]. Among the higher alcohols, the highest contents of phenyl ethyl alcohol (66.9 $\mu g \cdot kg^{-1}$), 2-hexanol (32.3 $\mu g \cdot kg^{-1}$) and 3-pentanol (13.9 $\mu g \cdot kg^{-1}$) were found in the chicken breast meat extract, while 3-penten-2-ol, 3-pentanol, *(E)*-3-hexen-1-ol, 2-furan methanol, isoamylalcohol and benzyl alcohol were present at lower contents (Tab. 1). Alcohols are known to be formed by a lipoxygenase-initiated peroxidation of *n*-3 and *n*-6 PUFA, which are present in fish tissue. They could have insignificant contribution to odour due to their relatively high odour threshold values [36].

Ketones 3-hydroxy-2-butanone, 4-nonanone, 1-hydroxy-2-pentanone and cyclopenthyl methyl ketone were also detected in the chicken breast meat. The most abundant ketone in the meat extract, 4-nonanone, was found at 14.7 $\mu g \cdot kg^{-1}$. Thermal degradation, oxidation of lipids, degradation of amino acids and Maillard reaction are possible mechanisms for the formation of ketones in cooked meat products [13, 28].

Phenolic acids are secondary metabolites that are commonly found in plant-derived foods. They are degraded thermally or decomposed by microorganisms into phenols, which are then detected in several foods. Volatile phenols benzophenone, phenol, 2,3-dimethylphenol and 4-vinyl-2-methoxyphenol were detected in the chicken breast meat. Dimethylphenols, of which 2,3-dimethylphenol is an isomer, are present in essential oils of various conifers, in tea, in roasted coffee, chicory and in various smoked foods [37]. Based on *OAV*, the main contributor to chicken breast meat aroma from this group would be expected to be 4-vinyl-2-methoxyphenol, providing a spicy odour (*OAV* = 2.8). The odour threshold value for this volatile phenol was determined to be 3.0 $\mu g \cdot kg^{-1}$ [38]. 4-Vinyl-2-methoxyphenol was shown to be formed as the main product of a thermally induced decarboxylation of ferulic acid [39].

Another volatile identified in the chicken breast meat was a terpene, *DL*-limonene. The content of this terpene was found to be 7.3 $\mu g \cdot kg^{-1}$ in the meat extract. *DL*-limonene is one of the most common terpenes in nature. In previous studies, it was found in grey mullet [25], goat meat [12], chicken breast and minced beef meat [11, 34]. Terpenoids can be added intentionally to feed due to their aromatic properties [40].

CONCLUSION

In the present work, the volatile profile of chicken breast meat from Turkey was first de-

termined. A total of 33 volatile compounds were identified in the SDE extracts of chicken breast meat sample. Acids and esters were found as the major compound classes. In terms of odour contribution to raw chicken breast meat, two compounds were more prominent based on *OAV*. Within these, hexanal (*OAV* = 2.9) was the most powerful contributor to the odour of the chicken breast meat, followed by 4-vinyl-2-methyoxyphenol (*OAV* = 2.8).

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