

PROCEEDINGS OF THE 11th WORLD RABBIT CONGRESS

Qingdao (China) - June 15-18, 2016 ISSN 2308-1910

Session Quality of products

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Full text of the communication

How to cite this paper :

Xie Y.J., He Z.F, Zhang E., Li H.J., 1961 - Characterization of the volatile compounds in rabbit meat using gas chromatography mass spectrometry with simultaneous distillation extraction. *Proceedings* 11th World Rabbit Congress - June 15-18, 2016 - Qingdao - China, 799-802.



CHARACTERIZATION OF THE VOLATILE COMPOUNDS IN RABBIT MEAT USING GAS CHROMATOGRAPHY MASS SPECTROMETRY WITH SIMULTANEOUS DISTILLATION EXTRACTION

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ABSTRACT

The objective of this study was to explore the key volatile compounds in rabbit meat as no information available about them, and to establish foundation for the further research about degradation. Simultaneous distillation extraction with dichloromethane was adopted to extract the volatile compounds from rabbit meat. A total of 35 volatile compounds were identified and quantified by gas chromatograph-mass spectrometry, including volatile aldehydes, alcohols, acids, heterocyclic compounds and esters. Hexanal, heptanal, octanal, nonanal, (E, E)-2, 4-decadienal, 1-octen-3-ol and (Z)-2-decenal were the key odorant compounds as they accounted for higher relative odour activity value (OAV). Furthermore, the concentration of key odorant compounds in male rabbit meat was higher than that in female meat.

Key words: Rabbit meat, Simultaneous distillation extraction, Odour activity value, Volatile aldehydes, GC-MS.

INTRODUCTION

Rabbit meat is a popular food in Europe, Asian, African and South American, especially in China which owns the largest production of 727,000 tons in 2013(FAO, 2015). Actually, the production of rabbit meat maintains steadily increasing in recent few years in China. Meat flavour, which contains variety of tastes and aromas, affects consumers' meat purchasing behavior and preference (Jayasena, et al. 2013). Aldehydes, alcohols, ketones, esters, volatile phenols, acids and terpenes can be obtained through simultaneous distillation extraction. Hence, simultaneous distillation extraction (SDE) is adopted to extract volatile compounds of rabbit meat in this article. Gas chromatography mass spectrometry is used for identifying the volatile compounds in many meat species (Lammers, et al. 2009; Song, et al.2012; Ma, et al. 2013). However, it may be difficult for researchers to find the main volatile chemical compounds from the mass data directly. Odour activity value (OVA), which is apply to evaluate the key odorants in meat (Grosch. 2001; Carrascon, et al. 2014; Sun, et al. 2014), can be regarded as a simple and resultful method for finding the main chemical volatile compounds. No work has yet been published in the literature to evaluate volatile compounds of rabbit meat using simultaneous distillation extraction.

Therefore, the aim of this study was to assess the volatile compounds of rabbit meat first, and to determine the key odorants of overall flavour using OAV method.

MATERIALS AND METHODS

Animals and experimental design

Twenty Hyla rabbits, at 75 days, were purchased from college of animal science and technology, Southwest University. Both ten male and female rabbits were slaughtered and segmented at the same day under the same condition, and the meat were stored at -20 °C until the SDE analysis. The animal experiment was conducted in accordance with the Regulations of Experimental Animal Administration issued by the State Committee of science and Technology of the People's Republic of China.

Chemical Analyses

Different parts of rabbit meat were well minced and homogenized during 1 min in a household blender. Then, 50 g of the rabbit meat and 150 mL saturated NaCl aqueous solution were placed in a 500 mL round bottom flask attached to the appropriate arm of the SDE apparatus. The volatile compounds in rabbit meat analyzed using an Agilent 7890A-5975C gas chromatograph tandem mass spectrometry. Peak areas were calculated using the total ion chromatogram represented for each compound. The content of each compound was reported as percentages, representing the relative amount of each identified peak to the total area of identified peaks in each chromatogram. The identification of volatile compounds in rabbit meat is mainly based on mass spectrum to a data NIST11 and the comparison of GC retention indices (RI). The identified compounds can be calculated by comparing the peak areas with standard substance according to Eq. (1):

 $Concentration(\mu g/kg) = area_{ratio} * Concentration_{IS}$ (1)

where area ratio represents the ratio of analyte peak area and internal standard substance peak area. The standard solution of alkenes (C7-C30) 40mg L^{-1} in hexane was injected into GC-MS and analyzed under similar conditions in previous literature in order to identify the volatile compounds. The RI of volatile compounds in extraction was calculated from the follow Eq. (2):

$$RI = 100Z + 100 \times \frac{tR(X) - tR(Z)}{tR(Z + 1) - tR(Z)}$$
(2)

where x, z and z+1 represent the retain time of analyte, and the time of alkanes of efflux before and after the analyte respectively.

Evaluation of OAV

OAV=C/T, where c and t represents the concentration and threshold of volatile compound respectively. When OAV \ge 1, this means the volatile compound contribute to the overall flavor significantly; while OAV<1, that means no contributions at all.

Compounds	RI	Content[µg kg ⁻¹]	Retention	Method of
			time (min)	identification
Aldehydes				
Hexanal	1081	15	5.997	A,B,C
Heptanal	1182	3.05	8.242	A,B,C
(E)-2-Heptenal	1198	3.25	10.676	A,B,C
Octanal	1207	1.65	10.802	A,B,C
Nonanal	1392	9.15	13.46	A,B,C
(E)-2-Octenal	1345	2.55	11.684	A,B,C
2-Dodecenal	1523	1.25	16.966	A,B
(Z)-2-Decenal	1644	3.05	19.526	A,B
2-Undecenal	1751	2.1	21.965	A,B
(E,E)-2,4-Decadienal	1214	1.7	22.251	A,B
2-Methyl-undecanal	1376	0.35	23.125	A,B
Tetradecanal	1927	1.25	25.57	A,B
Pentadecanal	2042	5.1	27.692	A,B
(Z)-14-Methyl-8-hexadecenal	2219	0.75	32.097	A,B
Octadecanal	2357	1.15	33.506	A,B
(Z)-9-Octadecenal	2693	8.65	33.92	A,B
(Z)-9,17-Octadecadienal	2734	2.65	34.773	A,B
Esters				
Octyl chloroformate	1585	1.8	17.632	А
Dimethyl silanediol	1637	0.7	19.82	А
Hexadecanoic acid methyl ester	2114	0.55	31.252	А
Butyl octyl phthalate	2841	4.55	36.755	А

Table 1: Volatile compounds and their contents detected by SDE-GC-MS in rabbit meat

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Table 1 Continuation	RI	Content[µg kg ⁻¹]	Retention time (min)	Method of identification
2-Chloropropionic acid octadecyl ester	2901	18.1	37.171	А
Dibutyl phthalate	2985	12.5	39.185	А
Octaethylene glycol monododecyl ether	3102	3.95	46.32	А
Alcohols				
1-Pentanol	1256	2.25	9.924	A,B
1-Octen-3-ol	1687	2.25	14.978	A,B
2-Eicosanol	2937	1.65	38.268	A,B
2-Hexyl-1-decanol	2980	11.45	39.69	A,B
Acids				
Nonahexacontanoic acid	2634	0.25	33.822	A,B
trans-13-Octadecenoic acid	2915	17.15	37.112	A,B
Ketone				
3-Hydroxy-2-butanone	1243	18.55	10.682	A,B
3-Pentadecanone	1984	0.45	26.854	A,B
Heterocyclic compounds				
2-Pentyl-furan	1241	1.4	9.362	A,B
Alkanes				
1-Iodo-tridecane	1249	1.35	9.594	А
1-Iodo-hexadecane	2713	0.45	34.368	А

A, mass spectrum; B, liner retention indices are given for a DB-Wax capillary column; C, standard substance.

RESULTS AND DISCUSSION

The volatile compounds in rabbit meat

The rabbit meat flavour is characterized by the presence of a wild range of volatiles belonging to several classes of compounds such as aldehydes, esters, acids, alkanes, alcohols, ketones and furans. A total of 35 volatile compounds were identified and quantified in rabbit meat (Table1).

Key odorant of rabbit meat

Volatile aldehydes may responsible for the undesirable flavour of rabbit meat. Hexanal, heptanal, octanal, nonanal, (E, E)-2, 4-decadienal, 1-octen-3-oland (Z)-2-decenal had higher OAV than 1 (Table 2) and represented that they were the dominant odorants in rabbit meat.

Odour difference between male and female rabbit meat

The volatile compounds of male and female rabbit were identified by gas chromatography mass spectrometry with simultaneous distillation extraction (Figure 1).

Table 2: Odour threshold and description of key volatile compounds in rabbit meat						
Compound	Odour threshold[µg kg ⁻¹]	Odour active value	Odour desciption			
Hexanal	4.5	3.3	Green			
Heptanal	3	1	Fatty			
Octanal	0.7	2.4	Solvent, lemon, bitter			
Nonanal	1	9.1	Green, oil			
(E, E)-2, 4-Decadienal	0.07	24.3	Aldehyde, rancid			
1-Octen-3-ol	1	2.2	Toasted, mushroom, metallic			
(Z)-2-Decenal	0.3	10.2	Poultry, orange			



Figure 1: Overlay of total ion chromatograms of volatile compounds sampled in (blue line) male and (red line) female rabbit meat using simultaneous distillation and extraction

CONCLUSIONS

In this study, the volatile compounds of rabbit meat have been determined using GC-MS in combination with SDE analysis. The dominant flavour compounds of rabbit meat were hexanal, heptanal, octanal, nonanal, (E, E)-2, 4-decadienal, 1-octen-3-ol and (Z)-2-decenal. (E, E)-2, 4-Decadienal contributed to overall flavour for obtaining highest OAV. Furthermore, the concentration of key odorants in male rabbit meat was higher than that in female one. These findings may explain the phenomenon that male rabbit meat had stronger odour than female.

ACKNOWLEDGEMENTS

This work was funded by the basic scientific research program southwest university (XDJK2014D042) and rabbit industry system of education ministry (100030-40305411) in China and Chinese Specialized Fund for the Basic Research Program of College(Grant No. 2120133197) Ministry of Agriculture Public Welfare Industry Science and Technology Research Program (Grant No.201303144).

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