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Contribution of beef base to aroma characteristics of beeflike process flavour assessed by descriptive sensory analysis and gas chromatography olfactometry and partial least squares regression

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ABSTRACT

Descriptive sensory analysis and gas chromatography–mass spectrometry (GC–MS) analysis were conducted to investigate changes in aroma characteristics of beeflike process flavours (BPFs) prepared from enzymatically hydrolyzed beef (beef base) of different DH (degree of hydrolysis) with other ingredients. Five attributes (beefy, meaty, simulate, mouthful and roasted) were selected to assess BPFs. The results of descriptive sensory analysis confirmed that BPF2 from beef base of moderate DH 29.13% was strongest in beefy, meaty and simulate characteristics; BPF4 and BPF5 from beef base of higher DH (40.43% and 44.22%, respectively) were superior in mouthful and roasted attributes respectively; while BPF0 without beef base gave weaker odour for all attributes. Twenty six compounds from GC–MS were selected as specific compounds to represent beef odour based on their odour-active properties assessed by a detection frequency method of GC–O and correlation of their contents with sensory attributes intensity. Correlation analysis of molecular weight (MW) of peptides, odour-active compounds and sensory attributes through partial least squares regression (PLSR) further explained that beef base with DH of 29.13% was a desirable precursor for imparting aroma characteristics of beeflike process flavour.

1. Introduction

Beef flavours have been increasingly found application in meat analogues and processed instant foods. Recently, there have been various types of simulated meat flavours such as simple blended spices [1], recombined flavour components isolated and identified from cooked or fried meat [2], prepared from hydrolyzed vegetable protein (HVP) or hydrolyzed yeast [3], however, the most common type is "thermal process flavour", which is a comparatively recent term given to a food flavour produced by heating a combination of two or more precursor materials under carefully controlled conditions [4]. The primary reaction occurring in this process is the Maillard reaction.

It is well-known that meat flavour is thermally derived and consists of "meaty flavour" and "species-specific flavour", which are imparted through coordination of Maillard reaction and lipid oxidation [5]. Therefore, precursors play an important role in the generation of process flavour. Generally, beef flavours are derived from the complex interactions of flavour precursors such as amino acids, peptides, sugars, thiamine, metabolites of nucleotides, and products of lipid oxidation. Considerable researches and patents have been done to develop beeflike flavour by Maillard reaction with various amino acids and sugars [6]. In contrast to pure amino acids, protein hydrolysates which contain free amino acids, peptides, nucleotides, reducing sugars, carbonyl compounds, and sulfur compounds, are inexpensive and have been used to produce beef flavours.

For many years, HVP like soybean protein has been selected as potential precursor for beeflike process flavour [3,7]. However, meat flavours based on HVP can only partially simulate natural meat aroma, therefore, the thermal reaction model system has been evaluated for other flavour precursors (e.g. enzymatically hydrolyzed animal proteins) [8]. Some early researches related to meat hydrolysates have been involved in the preparation of meat flavour [9]. The first attempt of heating enzymatically degraded meat to produce meat flavour was made by Chhuy and Day [10]. Similar processes starting with the proteolysis of meat and meat by-products have been described by others [11].

The degree of hydrolysis (DH) of meat protein is a very important index for preparing meat flavours. Barbel Lieske and Gerd Konrad [12] confirmed that strong meatlike flavour notes would be intensified by heating the partial hydrolysates of meat protein in the

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Table 1
Changes of molecular weight (MW) distribution in different HBPs.

MW (Da)	Samples				
	HBP1[a]	HBP2	HBP3	HBP4	HBP5
>5000[b]	0.25 ± 0.01[c]	0.00 ± 0.00	0.14 ± 0.01	0.07 ± 0.00	0.06 ± 0.01
1000–5000	6.75 ± 0.12	4.45 ± 0.18	5.00 ± 0.12	3.56 ± 0.01	3.13 ± 0.30
500–1000	19.05 ± 0.06	21.04 ± 0.10	18.11 ± 0.03	13.33 ± 0.09	12.69 ± 0.06
200–500	54.49 ± 0.05	59.31 ± 0.03	62.55 ± 0.12	69.44 ± 0.06	70.77 ± 0.01
>200	2.47 ± 0.02	1.21 ± 0.03	2.20 ± 0.06	4.59 ± 0.06	4.35 ± 0.01

[a] Five samples were denoted by the HBP followed by 1-digit Arabic numbers. Where “HBP” represents for beef enzymatic hydrolysate (beef base), the followed Arabic numbers 1–5 denote DH 25.35%, 29.13%, 35.40%, 40.43% and 44.22%, respectively.
[b] Peptides in HBP as mg/mL of beef base.
[c] Mean ± standard deviation (average of triplicate).

Table 2
Analyses of variance for the main effects and their interactions for each of the five attributes in descriptive analysis.

	F-values						Adjusted *F*-value	
	Sample (S) (df = 5)	Panelist (P) (df = 7)	Replication (R) (df = 2)	S × P (df = 35)	P × R (df = 14)	S × R (df = 10)	Sample[a] (S) (df = 35)	Sample[b] (S) (df = 10)
Beefy	213.41***	2.71*	3.18*	2.61***	0.33	2.32*	81.64***	66.76***
Meaty	416.65***	16.98***	2.96	11.48***	0.75	1.18	36.29***	33.60***
Simulate	401.53***	0.96	0.88	0.90	1.59	1.04	445.91***	441.35***
Roasted	67.36***	6.43***	2.58	2.24***	0.73	1.22	29.38***	22.58***
Mouthful	133.66***	1.11	3.17	2.03**	1.05	0.47	65.84***	71.21***

[a] Adjusted *F*-values of sample effect calculated using $MS_{sample \times panelist}$ instead of MSerror as described in the text.
[b] Adjusted F-values of sample effect calculated using $MS_{sample \times replication}$ instead of MSerror as described in the text.
* Significant at $p \leq 0.05$.
** Significant at $p \leq 0.01$.
*** Significant at $p \leq 0.001$.

presence of appropriate sulfur and carbohydrate sources compared with the total hydrolysates. However, there is still a lack of more systematic study for the impact of beef hydrolysate with different DH on the aroma characteristics of beeflike process flavour.

Even though a great number of volatile compounds (more than 1000) have been reported in cooked beef meat, only some of them are important in terms of the characteristic beef flavour. In recent researches, more great efforts have been made to find and identify key aroma compounds in beef via gas chromatography in combination with olfactometry (GC–O) [13,14]. However, a little was known about the aroma active components of beeflike process flavour prepared from enzymatically hydrolyzed beef, so called beef base.

The objectives of the present study are to (a) apply descriptive sensory analysis to describe and monitor the aroma attributes of beeflike process flavours (BPFs) derived from beef base with different DH, (b) analyze the volatile compounds released from BPFs by GC–MS and investigate the impact of beef base with different DH on their corresponding aroma-active compounds determined by GC–O, and (c) identify which aroma-active compounds and peptides of what MW have significant influence on individual sensory attributes through correlation analysis among molecular weight (MW) of peptides, aroma-active compounds and sensory attributes. Through the above analysis, the desirable beef base with suitable DH is then recommended for controlled proteolysis to prepare characteristic beef flavour precursors.

2. Materials and methods

2.1. Materials

Lean beef was purchased from Wal-Mart supermarket in Wuxi, China. Hydrolyzed vegetable protein (HVP) was provided by Tianning Flavour & Fragrance Co., Ltd. (Shanghai, China). Refined tallow was purchased from Anhui Muyang Oil and Fats Co., Ltd. (Anhui, China). DL-methionine, D-xylose, glucose, L-cysteine, L-glutamic acid, L-proline, thiamine and taurine were purchased from Sinopharm Chemical Reagent Co., Ltd. (Shanghai, China). Alkaline protease, activity 2.4 AU/mL, and flavourzyme, activity 500 LAPU/g, were purchased from Novozymes (Bagsvaerd, Denmark). 1,2-Dichlorobenzene and methanol were of chromatography grade from TCI Development Co., Ltd. (Shanghai, China). Other authentic reference compounds were obtained from commercial sources and Sigma–Aldrich Co. Ltd. (Shanghai, China).

2.2. Sample preparation

2.2.1. Preparation of beef base

Lean beef (water content, 75.98%; protein content, 20.58%) was minced with a tissue-tearor and mixed with deionized water at a meat–water ratio of 7:3. The mixture dispersion was then heated at 95 °C for 10 min in order to make the endogenous enzyme

Table 3
The mean intensity values of the 5 attributes for the 6 BPF samples in descriptive sensory evaluation.[X]

Beefy		Meaty		Simulate		Roasted		Mouthful	
Sample[Y]	Mean score	Sample	Mean score	Sample	Mean score	Sample	Mean score	Sample	Mean score
BPF0	3.25[a]	BPF0	1.21[a]	BPF0	2.39[a]	BPF4	5.71[a]	BPF0	2.88[a]
BPF1	4.04[b]	BPF1	4.33[b]	BPF5	2.79[b]	BPF0	6.25[b]	BPF5	5.25[b]
BPF3	5.54[c]	BPF5	6.13[c]	BPF1	3.54[c]	BPF3	6.63[b]	BPF3	6.50[c]
BPF4	6.00[d]	BPF3	6.13[c]	BPF3	6.00[d]	BPF2	7.50[c]	BPF2	6.58[c]
BPF5	6.00[d]	BPF4	6.88[d]	BPF4	6.46[e]	BPF1	7.92[d]	BPF1	7.21[d]
BPF2	8.50[e]	BPF2	8.58[e]	BPF2	8.67[f]	BPF5	8.75[e]	BPF4	7.25[d]

[X] Mean scores (listed in ascending order) for each attribute within a column with different letters are significantly different ($p \leq 0.05$) using Duncan's multiple comparison test ($n = 24$; 8 panelists with 3 replications).
[Y] Six beaflike process flavours were denoted by the BPF0-5, which were prepared from without HBP and with HBP1, HBP2, HBP3, HBP4 and HBP5, respectively.