LWT - Food Science and Technology 64 (2015) 316–325

Contents lists available at ScienceDirect

LWT - Food Science and Technology

journal homepage: www.elsevier.com/locate/lwt

Effect of thermal treatment on the flavor generation from Maillard reaction of xylose and chicken peptide

Jianbin Liu [a], Mengya Liu [a], Congcong He [a], Huanlu Song [a, *], Feng Chen [b]

[a] Laboratory of Molecular Sensory Science, Beijing Technology and Business University, 100048, China
[b] Department of Food, Nutrition and Packaging Sciences, Clemson University, Clemson, SC 29634, USA

ARTICLE INFO

Article history:
Received 1 December 2014
Received in revised form
24 May 2015
Accepted 28 May 2015
Available online 6 June 2015

Keywords:
Chicken peptides
Maillard reaction
Flavor
Gas chromatography-olfactometry (GC-O-MS)
Sensory perception

ABSTRACT

In this research, effect of the thermal treatment on the flavor generation from Maillard reaction of xylose and chicken peptide was studied. In summary, high temperature (>100 °C) could remarkably increase the formation of meaty aroma generated by xylose and chicken peptide through a Maillard reaction system, while lower temperature and longer heating tended to generate a broth-like taste (i.e., umami and kokumi). Pyrazines were the major contributors to the nutty/roast and basic meaty aroma in the Maillard reaction products (MRP), while the Glu released under the low temperature heating was considered an important contributor to the umami taste of MRPs. The low molecular weight peptide (<500 Da) was considered as the main contributor of the generation of pyrazines and 2-furfurylpyrrole, which could be due to the high reaction activity of the amidogen in these compounds. In addition, cross-linking compounds with molecular weight >1000 Da formed from peptides <500 Da in the low temperature heating might be involved in formation of important kokumi-active compounds, while the high molecular compounds MRPs >3000 Da were very likely responsible for the bitter taste.

1. Introduction

Maillard reaction plays an important role in the generation of aroma and taste-active compounds of processed food (Martins, Jongen, & van Boekel, 2001). The chemistry underlying the Maillard reaction is a complex reaction between carbonyl group of reducing sugars and amine group of free amino acids, peptides and proteins (Hodge, 1953). So far, reaction models between free amino acids and carbonyl compounds have been studied extensively, and suggested that the different reaction mechanisms existed in regards of different reactants, e.g., free amino acids or peptides (van Lancker, Adams & de Kimpe, 2010; Lu, Hao, Payne, & Ho, 2005). Investigations of using pure peptides in Maillard reaction models demonstrated that the peptides could participate in the Maillard reaction in many pathways such as bond cleavage, cyclization, and glycation (Garbe, Würtz, Piechotta, & Tressl, 2008; Horvat & Jakas, 2004; Yang, Wang, & Song, 2012). In addition, the Maillard reaction products (MRPs) of peptides and sugars could exhibit a strong meaty/broth-like (umami and kokumi) flavor in the foods (Liu et al., 2012; Ogasawara, Katsumata, & Egi, 2006).

Thermal treatment is one of the most important factors affecting the reaction rate and flavoring characteristics of the Maillard reaction (Benzing-Purdie, Ripmeester, & Ratcliffe, 1985; Martins et al., 2001). In recent years, several studies have focused on the heating effect on the functional properties of MRPs generated from vegetable protein hydrolyzate (Lertittikul, Benjakul, & Tanaka, 2007; Li et al., 2009). However, the public information of animal proteins involved in the generation of flavor compounds in the Maillard reaction is still limited.

Chicken meat comprises a substantial source of high-quality proteins and is accepted by consumers all over the world (Kurozawa, Park, & Hubinger, 2008). To the best of our knowledge, few researches have clarified the role of chicken peptides on the generation of flavors in the Maillard reaction. In this context, the objective of this research was to investigate the thermal treatment in terms of heating time and temperature on the generation of meaty/broth-like (umami and kokumi) flavors from a Maillard reaction model made of xylose and chicken peptide.

2. Materials and methods

2.1. Chemicals

Commercial enzymes Protamex® and Flavourzyme® 500MG were supplied by Novozymes (Bagsvaerd, Denmark). Xylose (98%, analytical grade), the following chemicals of chromatographic

* Corresponding author. Tel./fax: +86 10 68984025.
E-mail address: songhl@th.btbu.edu.cn (H. Song).

http://dx.doi.org/10.1016/j.lwt.2015.05.061
0023-6438/

reagent grade, including n-alkanes (C_7–C_{30}), 2-methyl-3-heptanone, cytochrome C, bacitracin, aprotinin, glutathione and glycine were purchased from Sigma–Aldrich (St. Louis, MO, U.S.A.). Derivatization reagent o-phthaldialdehyde (OPA) and fluorenylmethyl chloroformate (FMOC) was supplied by Agilent Technologies (Palo Alto, CA, U.S.A.). Standard oligopeptides (TEDVDVK, GASST, CG, 95%) were obtained from Apeptides (Shanghai, China). HPLC grade solvents were purchased from Thermo Fisher Scientific (Waltham, USA). The HPLC-grade H_2O was prepared in our laboratory.

2.2. Chicken breast meat

The frozen chicken breast meat was purchased from a local market in Beijing, China. The main chemical compositions of the meat are as follows: moisture: 72.6 g/100 g; proteins: 20.3 g/100 g; fat: 1.3 g/100 g; ash: 1.1 g/100 g. The meat was cut into paste and stored in the −18 °C until use.

2.3. Preparation of chicken peptides

The endoproteinase Protamex® and exoproteinase Flavourzyme® 500 MG were chosen to prepare the enzymatic hydrolyzate of chicken meat. Detail preparation was as follows: 50 g of meat paste was homogenized with distilled water (1:1, mL/mL) followed by heating at 55 °C for 30 min; the pH of the mixture was adjusted to 6.5 with 2 mol/L NaOH before the Protamex (900 U/g, E/S) and Flavourzyme (200 U/g, E/S) were added to the mixture. The reaction pH was maintained constant by adding 2 mol/L NaOH at regular intervals (30 min). After 4 h of enzymolysis, the hydrolyzate (at a degree of hydrolysis, 11.6 ± 1.2%) was stayed in 90 °C for 5 min to inactive enzyme and then centrifuged at 7500 × *g* for 30 min at 4 °C on a CR 22G high speed refrigerated centrifuge (Hitachi, Tokyo, Japan). The light yellow supernatant was washed away three times with diethyl ether (1:5, mL/mL) and filtered through a 5000 Da cut-off membranes on a Pellicon apparatus (Millipore Ltd., Bedford, MA, U.S.A.) at 4 °C. The permeate liquor was freeze-dried to obtain the chicken peptides. The chemical composition of the chicken peptides was as follows: free amino acids: 7.5 g/100 g; total amino acids: 77.8 g/100 g; total nitrogen: 12.2 g/100 g.

2.4. Preparation of the Maillard reaction products (MRPs)

The mixture of chicken peptides (1.0 g) and xylose (0.2 g) were dissolved in 20 mL deionized water. The mixture was adjusted to pH 6.5 with 6 mol/L NaOH. The solutions were heated with different temperatures (80, 100, 120, 140 °C) and time (30, 60, 90, 120 min) in a high-pressure stainless reactor (Parr Instrument Company, Moline, IL, U.S.A.) for the production of MRPs. After heating, the solutions were rapidly cooled in an ice bath to prevent the further reaction and then stored at 4 °C for 24 h before analysis.

To further investigate the role of thermal degradation of peptide on the flavor generation *via* Maillard reaction, additional experiment was designed: a mixture of thermal degradation reaction products (heating at 80 °C or 140 °C for 90 min) of chicken peptides (1.0 g) and xylose (0.2 g) were dissolved in 20 mL deionized water (pH was adjusted to 6.5 with 6 mol/L NaOH), followed by heating at 120 °C for 90 min to obtain the Maillard reaction products of thermal degradation peptide (80-tMRPs or 140-tMRPs). All the MRPs were prepared in triplicate.

2.5. Thermal degradation reaction

The thermal degradation reaction was done for the comparison of peptide distributions and flavor properties of MRPs and degradation reaction products (TDPs). The TDPs was achieved by heating the solution of chicken peptides (1.0 g/20 mL, pH = 6.5) at 120 °C for 90 min without the addition of xylose in the same way as described above.

2.6. Quantitation analysis of amino acids

The free amino acids in the MRPs were quantified by Zorbax Eclipse-AAA columns and the Agilent 1100 HPLC system (Agilent Technologies, Palo Alto, CA, U.S.A.). For the analysis of free amino acids, MRP samples were mixed with an equivalent volume of trichloroacetic acid (TCA) to precipitate peptides and/or proteins. The suspension obtained was centrifuged at 3000 × *g* for 15 min. The supernatant was freeze-dried and diluted with water to 20 mL. After precolumn derivatization with OPA and FMOC, the amino acids were quantified according to the method of Henderson et al. (Henderson, Ricker, Bidlingmeyer, & Woodward 2000).

2.7. Peptide distribution analysis

The peptide distribution of the samples was analyzed on a TSK G2000 SWXL column (300 × 7.8, 5 μm, Tosoh Co., Tokyo, Japan). The gel chromatography was performed on an Agilent 1100 series preparative HPLC system (Agilent Technologies, Palo Alto, CA, U.S.A.), which consisted of G1311A series prep pump, G1329A autosampler and G1315D ultraviolet detector. Analytical separation was performed on the flow rate of 0.5 mL/min using 20 mL/dL acetonitrile with the adjustment of the pH value to 3.0 with aqueous formic acid. The effluent was monitored at the wave lengths of 220 nm. Aprotinin (6512 Da), bacitracin (1422 Da), heptapeptide (TEDVDVK, 804 Da), pentapeptide (GASST , 421 Da), dipeptide (CG, 175 Da) and Gly (75 Da) were used to construct the calibration curves of standard molecules.

2.8. Extraction of the volatile compounds

The volatile components in the reaction products were extracted by a stratum purge & trap concentrator (Atomx Teklink, Teledyne Tekmar, Ohio, U.S.A.). A total of 2 mL MRPs with 1 μL internal standard (2-methyl-3-heptanone, 1.632 g/L in n-pentane) was put into a purge-and-trap vessel and equilibrated at 50 °C for 30 min. Purge and trap conditions were as follows: the flow rate of high pure nitrogen was controlled at 40 mL/min, purged at 50 °C for 12 min, followed by desorption at 250 °C for 2 min and baked at 280 °C for 5 min. The purge and trap concentrator was connected with a system containing a gas chromatograph coupled with a mass spectrometer and an olfactometer.

2.9. Gas chromatography-olfactometry (GC-O-MS)

The volatiles were analyzed by GC–MS (7890A-7000B; Agilent Technologies Inc., Santa Clara, CA, U.S.A.) which was equipped with an olfactory detector port (sniffer 9000; Brechbühler, Switzerland). After injection, chemical separations by GC were performed on a DB-Wax column (30 m × 0.32 mm × 0.25 m; J & W Scientific, Folsom, CA, U.S.A.). Ultra-high purity helium was used as the carrier gas at 1.2 mL/min. The GC oven program was set at 35 °C, then held for 2 min and ramped at 6 °C min^{-1} to 230 °C, then held at this temperature for 20 min. The effluent from the capillary column was split into 1:1 (ml/ml), between the mass spectrometry detector (MSD) and the sniffing port. The GC–MS transfer line temperature was 240 °C (DB-Wax). Electron-impact mass spectra were generated at 70 eV, with an *m/z* scan range from 40 to 500. The MS source temperature was 230 °C. Three experienced panelists were recruited to perform the sniffing test on GC-O. To prevent drying of