Food Chemistry 269 (2018) 466–472

Contents lists available at ScienceDirect

Food Chemistry

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

Effect of lipid oxidation on the formation of N^{ε}-carboxymethyl-lysine and N^{ε}-carboxyethyl-lysine in Chinese-style sausage during storage

Ligang Yu[a], Meng Chai[b], Maomao Zeng[b], Zhiyong He[b], Jie Chen[b,c,*]

[a] School of Life Science, Shanxi University, Taiyuan 030006, China
[b] State Key Laboratory of Food Science and Technology, Jiangnan University, Wuxi 214122, China
[c] Synergetic Innovation Center of Food Safety and Nutrition, Jiangnan University, Wuxi 214122, China

ARTICLE INFO

Keywords:
N^{ε}-carboxymethyllysine
N^{ε}-carboxyethyllysine
Chinese-style sausage
Lipid oxidation
Model system

ABSTRACT

This work aimed to investigate the effects of lipid oxidation on N^{ε}-carboxymethyllysine (CML) and N^{ε}-carboxyethyllysine (CEL) formation in three types of Chinese-style sausages during storage. Medium-high temperature dried sausage, naturally dehydrated sausage and smoke-dried sausage were prepared, and CML/CEL contents were measured. The result showed that CML and CEL levels increased with the increasing storage time, and an exponential correlation was found between thiobarbituric acid reactive substances (TBARs) and either CML ($r^2 = 0.922$) or CEL ($r^2 = 0.921$) in medium-high temperature dried sausages and smoke-dried sausage, which suggested that lipid oxidation could facilitate CML/CEL formation during storage. Furthermore, lipid oxidation and the Maillard reaction might have a synergistic effect on CML formation in Chinese-style sausages, as evidence by the results of model system during incubation. These findings indicated that lipid oxidation played a key role in CML/CEL formation during the storage of Chinese-style sausages.

1. Introduction

Advanced glycation end products (AGEs) are generated through nonenzymatic reaction between reducing sugars and amino compounds (Singh, Barden, Mori, & Beilin, 2001). These compounds are related to many chronic diseases. AGEs could exist in foods, and daily diet contributed to the level of AGEs in body (Li et al., 2015). It has been reported that AGEs accumulation in human bodies are implicated in a variety of chronic diseases such as atherosclerosis (Basta, Schmidt, & De Caterina, 2004), Alzheimer's disease (Takeuchi and Yamagishi, 2008), and diabetes (Vlassara and Palace, 2002). Thus, AGEs have received much attention not only in medicine but also in food science.

N^{ε}-carboxymethyl-lysine (CML) and N^{ε}-carboxyethyl-lysine (CEL) are stable AGEs in food system, and usually recognized as typical AGEs markers (Poulsen et al., 2013). In food systems, CML and CEL are generated through different pathways, such as Maillard reaction pathway and lipid oxidation pathway (Peng, Ma, Chen, & Wang, 2011). At present, the influencing factors and generation pathway of CML have been widely reported in the Maillard reaction (Nguyen, Van der Fels-Klerx, & Van Boekel, 2014). Briefly, CML is formed via oxidation of N^{ε}-fructosyllysine (FL) and the reaction of glyoxal (GO) with lysine or arginine residues, and CEL is produced by reaction of methylglyoxal (MGO) with lysine residues in the Maillard reaction (Peng et al., 2011). In lipid oxidation pathway, the reactions between the lysine residues and dicarbonyls (such as GO and MGO) formed in lipid oxidation would lead to the formation of CML and CEL (Fu et al., 1996).

Meat products are the most popular foods in the world. Meanwhile, meat products are a rich source of CML and CEL due to high protein and/or high fat content. During the storage and processing of meat product, lipid and protein oxidation will be inevitable, which may affect the formation of CML and CEL (Ahmed, Thorpe, & Baynes, 1986). At present, there have been many studies on the amounts of CML/CEL or influencing factors of CML/CEL generation in meat products. For example, Hull, Woodside, Ames, and Cuskelly (2012) determined CML content in 257 foods, and found that CML level in meat dishes varied from 10.09 to 2354.87 mg/kg protein. Chen and Smith (2015) have been conducted to elucidate the effects of processing conditions on the formation of CML, and indicated that CML content was dependent on types of meat, the sample centre temperature and cooking conditions. However, studies on the effect of lipid oxidation on CML and CEL formation during the storage of meat products are still limited.

Chinese-style sausage is a favorite traditional processed meat product due to its unique flavor characteristics developed during the natural fermentation process (Wang, Jiang, & Lin, 1995). It is made solely from pork with a fat content of approximately 20–30%. Chinese-style sausage is normally vacuumed packed for storage, and is not cooked

* Corresponding author at: State Key Laboratory of Food Science and Technology, Jiangnan University, Wuxi 214122, China.
E-mail address: chenjie@jiangnan.edu.cn (J. Chen).

https://doi.org/10.1016/j.foodchem.2018.07.051
Received 9 October 2017; Received in revised form 23 June 2018; Accepted 8 July 2018
Available online 09 July 2018
0308-8146/

until just before consumption. Traditionally, the sausage is hung outdoors under the sun to naturally dehydrate after stuffing. In modern meat-processing factories, this drying procedure has been replaced by smoking methods or oven drying. This will inevitably lead to lipid oxidation, which may affect the formation of CML and CEL in Chinese-style sausage during storage. Therefore, the objective of this work was to investigate the effects of lipid oxidation on the formation of CML and CEL in three types of Chinese-style sausages, including medium-high temperature dried sausage, naturally dehydrated sausage, and smoke-dried sausage, thereby providing a valuable reference on the formation mechanism of CML and CEL during storage of Chinese-style sausages.

2. Materials and methods

2.1. Materials

CML (purity over 99%), N^{ε}-carboxy[$^2H_2$]methyl-lysine (d_4-CML, purity over 95.7%), CEL (purity over 99%), N^{ε}-carboxy[$^2H_2$]ethyl-lysine (d_4-CEL, purity over 95.7%) and L-leucine were provided from Santa Cruz Biotechnology (Dallas, Texas, USA). Sodium borohydride, linoleic acid, nonafluoropentanoic acid (NFPA, purity over 97%), acetonitrile, and methanol of HPLC grade were obtained from J&K Chemical Co., Ltd. (Shanghai, China). Raw pork and food grade ingredients were obtained from local supermarkets (Wuxi, Jiangsu Province, China).

2.2. Sausages preparation

The preparation of the three types of Chinese-style sausages was based on the method describe by Lin and Huang (2008) with some modifications. As shown in Fig. 1, lean tissue was trimmed off and ground through a 10 mm plate. The pork fat was sheared manually into cubes. The meat mixture was composed of 70% ground lean pork and 30% fat by weight. The lean pork and fat were blended thoroughly in a mixer, and cured with sodium chloride (25 g/kg) and sodium nitrite (0.15 g/kg) (sodium nitrite was not used in naturally dehydrated sausage) at 4 °C for 2 h before being stuffed into collagen casing and linked into 15 cm units (Fig. 1). Following the different drying methods, the sausages were dried, cooled, vacuum-packaged and stored at 25 °C for 0, 15, 30, 60, 90, 120 and 180 days.

2.3. Analysis of moisture content

The sausage samples were ground with a grinder, freeze-dried, smashed into powder, and stored at −80 °C for further analysis. The moisture content in the samples was estimated according to the AOAC methods 950.46 (AOAC, 2002).

2.4. Lipid oxidation

The thiobarbituric acid reactive substance (TBARs) values of sausage samples were measured using the method of Wang and Xiong (2005). Briefly, 1.5 mL of TBA solution (1% TBA solution with 0.075 mol/L NaOH) and 8.5 mL of TCA solution (2.5% TCA solution with 0.036 N HCl) were added to the chopped sample (ca. 0.2 g) and mixed for 30 s. The mixture was placed in 100 °C water bath for 1 h. Absorbance was recorded at λ = 532 nm against reagent blank prepared in the same manner. The result was calculated by using the equation:

$$\text{TBARs (mg/kg sample)} = (A_{532}/Ws) \times 9.48$$

where A_{532} represent the absorbance of the assay solution

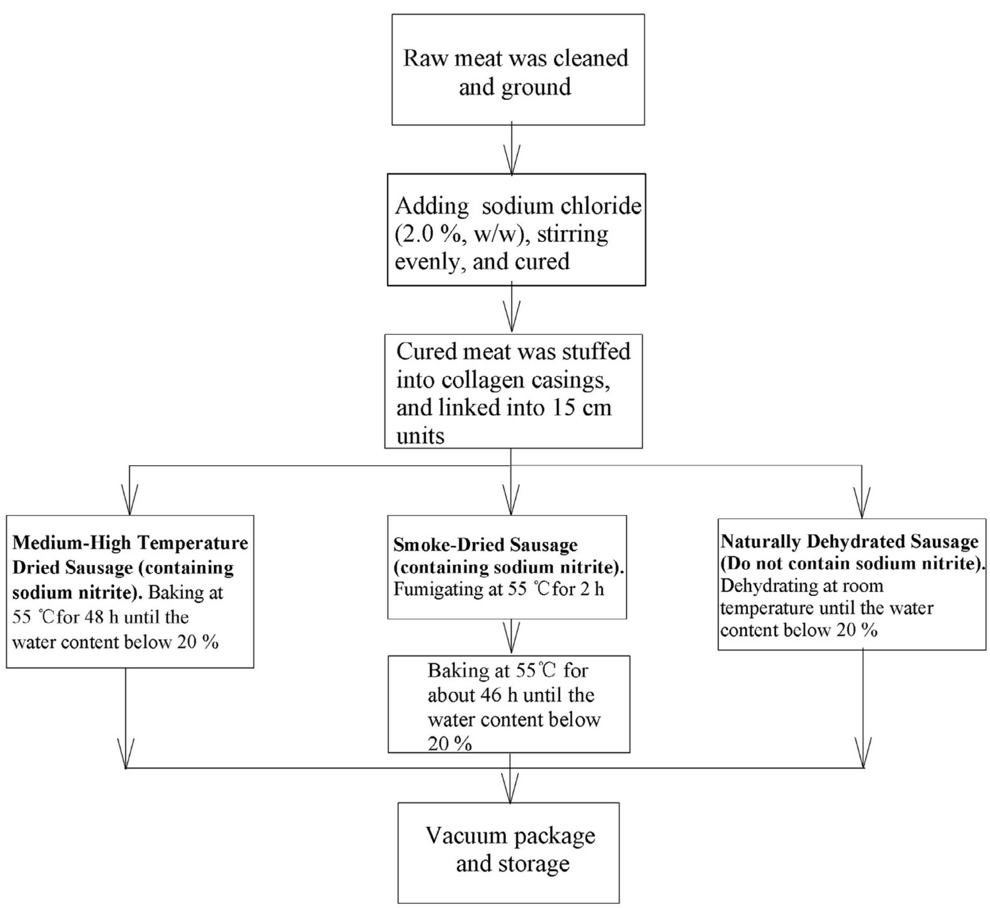

Fig. 1. The processing procedure of Chinese-style sausages.