Journal of Food Engineering 194 (2017) 58-66

Contents lists available at ScienceDirect

Journal of Food Engineering

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

Effect of mechanical curing treatments on particle distribution to simulate non-motile bacteria migration in cured raw ham

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A R T I C L E I N F O

Article history: Received 14 March 2016 Received in revised form 26 June 2016 Accepted 3 September 2016 Available online 6 September 2016

Keywords: Tumble Freeze-thaw Vacuum Meat starter cultures Blue Lake penetration Image analysis

1. Introduction

The traditional dry-curing prevents meat from spoilage and the growth of pathogenic microorganisms through applying different hurdles (Hayes et al., 2007; Pegg, 2004; Shahidi and Samaranayaka, 2004). Dry-curing is a time intensive processing step limited by the diffusion of salt and curing agents from the outside into the core of the ham (Marriott et al., 1992). The use of accelerated curing techniques such as injection, tumbling and vacuum can increase the salt distribution, but also the risk of introducing microorganisms from the surface into the core of the meat piece (Elmossalami and Wassef, 1971; Warsow et al., 2008). As had been demonstrated by previous studies, microorganisms appeared to be able to penetrate muscle tissue and the authors attributed this to a wide range of different mechanisms (Anderson et al., 1991; Bosse et al., 2015; Elmossalami and Wassef, 1971; Gill et al., 1984; Gill and Penney, 1977, 1982; Gill et al., 2008; Gupta and Nagamohini, 1992; Sikes and Maxcy, 1980; Thomas et al., 1987; Warsow et al., 2008). To minimize the risk of the undesired microbiota, the application of

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ABSTRACT

In this study, the migration and distribution of Blue Lake particles with various mechanical curing treatments (freeze-thawing, tumble, vacuum and vacuum-tumble) was studied after injection and drycuring of raw hams to simulate non-motile bacteria. The influence of injection pressure and various mechanical treatments on particle penetration was examined by using image analysis at the beginning and end of curing. The results showed that particles were able to penetrate the muscle after injection. An increase of the injection pressure from 0.5 bar to 0.9 bar resulted in a broader distribution of particles around the injection zone. Moreover, all additional mechanical treatments significantly increased the penetrated area of the particles by up to 64% due to an increase in damaged tissue and an introduction of new penetration pathways. The results provide insights that may be transferable from particles to starter cultures, which can further be applied in cured raw ham manufacture.

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bacterial starter cultures is a standard method in fermented sausages to maintain constant quality and a high degree of food safety (Toldrá, 2002). But not much is known about the application of starter cultures in cured whole muscle products such as cured ham. Some authors have shown that the application of meat tenderizing (Boyd et al., 1978; Marsden et al., 2001; Raccach and Henrickson, 1979) or vacuum (Warsow et al., 2008) allow the penetration of microorganisms by leading to small amounts of damage in the dense meat structure. Moreover, treatments like immersion in brine (Arnau et al., 2007; Shahidi and Samaranayaka, 2004), tumbling (Krause et al., 1978; Marriott et al., 1992; Ockerman and Organisciak, 1978; Xiong and Kupski, 1999) or freeze-thaw processes (Barat et al., 2005, 2006, 2004; Grau et al., 2011; Uttaro and Aalhus, 2007) can lead to tissue damage, too. Stained colloidal particles were used as model system for non-motile bacteria to visualize and study their distribution, location and pathways in the muscle tissue (Anderson et al., 1991; De Zuniga et al., 1991; Gill et al., 2008; Uttaro et al., 2011) and into eggs (Favier et al., 2000, 2009; Kim and Slavik, 1996; Xie et al., 2002). In addition, the application of image analysis was useful in conducting not only qualitative, but also quantitative analysis of bacterial penetration during meat processing (Bosse et al., 2015; Jackman and Sun, 2013; Jackman et al., 2011; Nunan et al., 2001; Schönholzer et al., 2002).







The aim of this study was to investigate the influence of various mechanical treatments on the distribution of stained colloidal particles injected into pork loin as a model system for non-motile bacteria in cured raw ham. To do this, the injection of particles in fresh and freeze-thawed pork loin was used to introduce the particles in the interior of the pork loin to reduce the penetration pathways in the whole muscles. For a more widespread particle distribution, further mechanical curing techniques such as tumbling, vacuum and vacuum-tumbling were applied to avoid latent curing defects in respect to future production of cured raw hams with starter cultures. Image analysis was used to determine the particle area in the injected cured raw hams at the beginning and the end of curing. We hypothesized that mechanical treatments would increase the penetration of particles in the meat structure due to a combined effect of damage of the meat structure and degassing of pores. Hence, this study was conducted as a first step towards an industrial use of starter cultures to manufacture cured raw hams with an injection process and further mechanical treatments

2. Materials and methods

2.1. Experimental setup and cured raw ham production

Experimental setup: The study of the influence of mechanical treatments on the particle distribution in cured raw ham included eight settings during curing and is illustrated in Fig. 1a to d. Two kinds of raw materials were used for this purpose: fresh and freeze-thawed pork loin (Fig. 1a). Freezing was done for 60 h at $-20 \,^{\circ}$ C and thawing was conducted for 60 h at between $4 \,^{\circ}$ C and $5 \,^{\circ}$ C to prevent microbial spoilage. The mechanical treatments were applied after injection and dry-curing (Fig. 1b and c); the hams were transferred to plastic bags (polyethylene/polyamide bags; Type C400, Multivac,

Wolfertschwenden, Germany) and the following treatments were applied:

- (1) Control (injection (105 MC2 R, Günther, Dieburg, Germany) and dry-curing)
- (2) Tumble (without vacuum, 12 h tumbling with 5 min rotation (12 rpm) and 25 min rest (total rotation time 60 min) (Tumbler Type 140, Vakona, Lienen, Germany))
- (3) Vacuum (vacuum packed with 100 mbar residual pressure, no tumble)
- (4) Vacuum-Tumble (vacuum packed with 100 mbar residual pressure, 12 h tumbling with 5 min rotation (12 rpm) and 25 min rest (total rotation time 60 min))

To study the influence of injection pressure and mechanical treatments, two experiments were carried out. In the first trial, the injection pressure varied between 0.5 bar and 0.9 bar to reach injection weights of approximately 5%–15% (w/w). The second trial was focused on the influence of the mechanical treatments with at an injection pressure of 0.7 bar to reach an injection weight of approximately 10%. In both studies, 5 mm thick samples were taken with a slicer (Type VS8A, Bizera, Balingen, Germany) after one day, to prevent brine loss by slicing directly after injection, and at the end of curing (day 7) (Fig. 1d). The second trial was conducted in triplicate.

Cured raw ham production: Fresh pork loin (*Musculus longissimus dorsi*; pH: 5.48 \pm 0.12) was obtained from a local central market (MEGA, Stuttgart, Germany) and cured raw ham production was carried out in the pilot plant at the University of Hohenheim (Germany). Determination of the pH was performed on three locations of the raw hams with a pH meter (pH537, WTW, Weilheim i. OB, Germany). For all the batches, the brine (30 L) was prepared with 10% (w/w) nitrite curing salt (Südsalz, Heilbronn, Germany;



Fig. 1. Schematic of experimental setup and image analysis for the particle distribution. The experimental setup showed the (a) raw materials used, fresh and freeze-thawed pork loin, that were (b) injected with a brine containing stained blue particles (Blue Lake) using a multi needle injector. (c) After dry-curing, the hams were transferred to the mechanical treatments as (1) control without further treatment, (2) tumble for 12 h, (3) vacuum with 100 mbar residual pressure and (4) vacuum-tumble as combination of tumble and vacuum treatment. (d) On sampling day, the hams were sliced and scanned. (e) The images were used for image analysis. Therefore, the images were split into (f) red and (g) blue channels and (h) subtracted from each other. (i) Inverted images were created, and a mask algorithm was applied to separate the ham slices. After (j) thresholding, the area of the blue particles was measured and set into respect to the whole slice area.

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