Characteristics of Fermented Pork Jerky Using Cultures of Dairy Lactic Acid Bacteria

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Abstract – This study was performed to evaluate microbiological and physico-chemical properties of unheated minced pork jerky fermented with lactic acid bacteria (LAB) starter cultures. After adding 10-20% LAB starter culture concentrates (LAB-C), the growth of coliform bacteria in the jerky was inhibited by fermentation incubating for >4 h at 43°C. In the pork jerky made after addition of 10% LAB-C, the shear force value was 1.54±0.52 kg at pH 4.73±0.04 with a 0.766±0.004 water activity (a_w). Exopolysaccharides from the bacteria were found in the LAB starter culture. The shear force of the jerky fermented with the LAB-C was lower than the control without the LAB-C or in commercial minced jerky. SDS-PAGE showed the myofibrillar proteins from the minced pork were partly degraded to polypeptides during LAB fermentation. The surviving viable LAB cells were 10^7 to 10^8 cfu/g in the jerky. The developed products demonstrate fermented pork jerky may provide a technology to produce a probiotic dry meat product.

Key Words – *Lactobacillus bulgaricus, Streptococcus thermophilus*, proteolysis, exopolysaccharide

I. INTRODUCTION

The human life span is rapidly increasing where many older adults are at risk of developing malnutrition. It is important for older adults to have intake of a nutrient-dense diet especially high-protein foods such as meat or milk products. Intake of probiotics containing useful live microorganisms such as LAB is also beneficial. LAB are widely used throughout the world in fermented products such as yogurt or cheese, however, their application in meat products is rare except for dry-fermented sausage. We recently studied the application of LAB as yogurt starter cultures in dry meat products such as jerky [1]. Jerky is convenient, ready-to-eat and usually requires no refrigeration. It has easy preparation, is light in weight, rich in nutrients, and is stable without refrigeration making jerky popular for sports enthusiasts, travelers, and mountaineers.

We evaluated unheated fermented minced pork jerky after the addition of LAB starter cultures for its microbiological and physico-chemical properties. The LAB starter cultures were *Lactobacillus delbrueckii* subsp. *bulgaricus* (*Lb. bulgaricus*) and *Streptococcus thermophilus* (*Stc. thermophilus*) that are often used as starter cultures in processed yogurts. The EPS content in the processing of the fermented pork jerky was determined and the effect of the EPS on the physical properties is discussed.

II. MATERIALS AND METHODS

Materials – Frozen pork raw ham was obtained from a commercial processor (Aichi, Japan) and thawed at 3°C, trimmed and minced through a plate with a hole diameter of 3.2 mm. The minced pork was vacuum-packed and stored at -35°C until jerky production. The LAB (YC-380, CHR Hansen, Denmark) starters were added at 1.5 μ/L to a medium composed of 10% whey, 0.5% D-glucose and 0.5% yeast extract; incubated at 43°C until the pH reduced to 4.6. The LAB-C cells were collected centrifuging the culture for 20 min at 10,000×g at 4°C.

Preparation of fermented pork jerky – The LAB-C was added at 5, 10, and 20% of the total minced pork weight and mixed. The preparations were stuffed into a fibrous casing (0.50 mm) (Futamura Chemical Co., Ltd., Aichi, Japan) and extended into a sheet 2 mm in thickness using a pasta machine. Then, the sheets were fermented for 6 h at 43°C in an incubator. The fermented meats were transferred into polyethylene bags, and a seasoning mix, 50% soy sauce and 50% Mirin (a traditional Japanese seasoning), was added to the meats. The bags were heat-sealed and stored for 12 h at 5°C. The seasoned meats were spread on

stainless steel net and dried at 20°C in a smoke house (Maurer TITAN, Germany). The low-temperature drying was performed to lower the water activity to less than 0.87 a_w agreeing with the hygiene standards for dry meat products in Japan.

Analysis – LAB and coliform counts were analyzed using Plate Count Agar with BCP and DOA agar, respectively. EPS in LAB starter cultures and fermented-minced pork were prepared and determined according to the method described by Nishimura et al., [2]. The EPS quantity was determined as glucose using the phenol-H_2SO_4 method. The pH of LAB starter cultures was directly measured with a pH meter; the pH of minced pork or jerky was measured using a 10% (w/w) suspension homogenized after addition of 45 ml D.W.. The water activity values of jerky samples were determined using an a_w meter (AquaLab LITE, USA). The shear force value (SFV) was determined using a Warner-Bratzler Meat Shear (G-R Manufacturing Co., USA). Tensile strength (TS) was determined using a Rheo Meter Compac-100 (Sun Scientific Co., Ltd., Japan). Measurements were taken using two parameters characterizing texture, i.e. maximum tensile force (N) and elongation capacity (expressed in mm). SDS-PAGE was performed according to the method of Laemmli using a 5.0% stacking gel and 12.5% separation gel. Sensory evaluation of fermented pork jerky was performed by 100 randomly selected untrained panelists at Chubu University. They assessed the samples for color, taste, texture, tenderness, and overall-acceptability using a 7-point scoring scale.

III. RESULTS AND DISCUSSION

As shown in Table 1, when the pH of the minced pork during fermentation approached pH 4.6, the viable cell count of the LAB increased to ~10^8 cfu/g. The coliform counts in the minced pork made with LAB-C gradually decrease throughout fermentation, and finally were not detected when the pH and the LAB counts in the minced pork reached pH 4.42 to 5.15 and >10^8 cfu/g, respectively. The minced pork without LAB-C (control) remained near the initial pH 5.85 and the coliform bacteria counts increased from $10^{3.45}$ to $10^{5.43}$ cfu/g during incubation for 6 h at 43°C. Most dry-fermented sausages are usually manufactured using fermentation for 24 - 48 h at 18 - 24°C [3]. However, it was almost impossible to suppress the growth of harmful bacteria during this short fermentation time (6 h or less) at these low temperatures (18 - 24°C) [4]. We found the growth of contaminant pathogenic microorganisms like coliform bacteria may be completely inhibited using 10 - 20% LAB-C in the minced pork and fermentation for 4 - 6 h or more at the optimum temperature (43°C). We considered the metabolic products of the LAB-C of *Lb. bulgaricus* and *Stc. thermophilus*, i.e. organic acids, mainly lactic acid, or bacteriocins [5], may usefully act as antimicrobial agents. Our data described above shows the minced pork with LAB-C was appropriately fermented for 6 h at 43°C to inhibit growth of harmful bacteria.

Table 1 pH, LAB, and coliform bacteria counts in the minced pork during fermentation after addition of LAB starter cultures concentrates (LAB-C)

	Fermentation time (h)			
	0	2	4	6
pH				
Control	5.85±0.07	5.84±0.06	5.83±0.07	5.82±0.07
5% LAB-C	5.70±0.06	5.51±0.09	5.24±0.09	5.15±0.05
10% LAB-C	5.61±0.08	5.13±0.11	4.76±0.07	4.58±0.06
20% LAB-C	5.49±0.04	4.63±0.05	4.46±0.03	4.42±0.04
LAB count (log cfu/g)				
Control	4.68±0.14	4.96±0.21	5.24±0.20	5.22±0.27
5% LAB-C	6.68±0.12	6.87±0.09	6.97±0.10	8.38±0.12
10% LAB-C	7.83±0.08	7.99±0.05	8.26±0.13	8.73±0.12
20% LAB-C	7.89±0.03	8.38±0.21	8.57±0.18	9.05±0.06
Coliform count (log cfu/g)				
Control	3.45±0.57	3.88±0.36	4.64±0.28	5.43±0.35
5% LAB-C	3.32±0.49	3.11±0.42	2.72±0.25	ND
10% LAB-C	3.29±0.48	3.04±0.28	ND	ND
20% LAB-C	2.99±0.18	2.78±0.28	ND	ND

All values represent means of 3 measurements. ND, not detected.

From the results of pH, moisture, and a_w in the samples collected at 2, 4, 6, and 8 h during the drying process at 20°C (data not shown), the processing conditions for unheated minced pork jerky fermented using LAB-C were concluded as follows: LAB-C added to 10% (w/w) of total minced pork, fermentation for 6 h at 43°C, and drying to less than 0.87 of a_w at 20°C (usually 6 - 8 h). The pH values of the experimental pork jerky

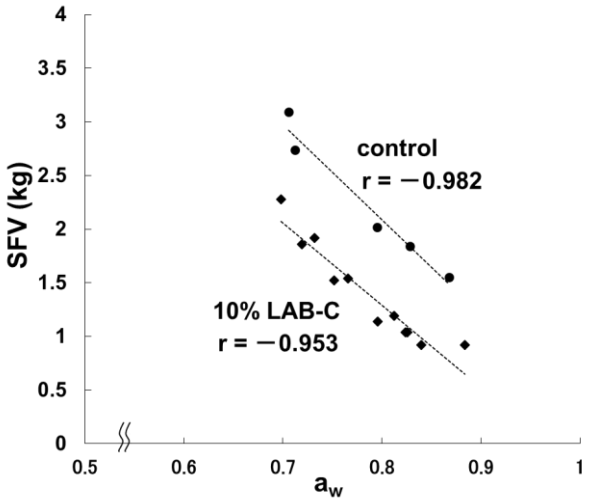

Figure 1. The correlation between a_w and SFV in the experimental minced pork jerky made by addition of 10% LAB-C (◆) or in control (without addition) (●)

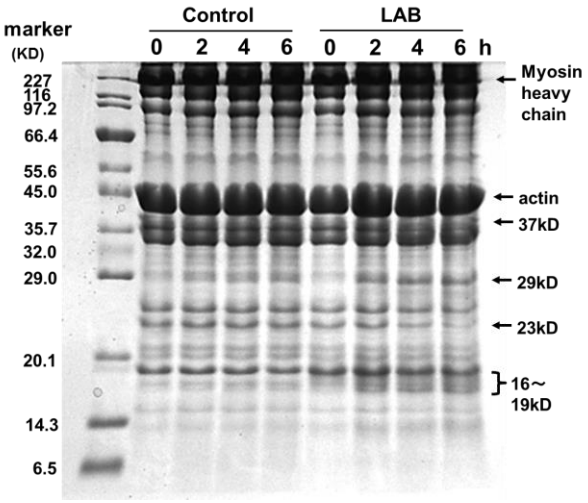

Figure 2. SDS-12.5% polyacrylamide gel electrophoretic pattern of myofibrillar proteins prepared from the minced pork during fermentation for 0, 2, 4, and 6 h at 43℃.

were 4.73±0.04, more acidic than commercial jerky samples. The a_w was 0.766±0.004 in the experimental pork jerky and 0.651 to 0.761 in the commercial jerky. There were no a_w significant differences between experimental and commercial jerky samples. There were significant differences in SFV among the jerky samples. The SFV in the commercial jerky samples were 3.38 to 11.93 kg, while that of the experimental pork jerky was 1.54±0.52 kg and showed the lowest value of all the jerky samples. In addition, the use of starter cultures such as the LAB-C in the processing of fermented-minced pork jerky caused an increase in the tenderness of the jerky.

As shown in Fig. 1, the correlation coefficient between a_w and SFV in the experimental pork jerky made by addition of 10% LAB-C or in the pork jerky without the LAB-C addition (control) was -0.953 or -0.982, respectively. The SFV of jerky was significantly influenced by a_w values. The SFV of the jerky linearly increased as a_w values decreased. Comparing SFV among the jerkies having the same a_w, the shear force of the fermented pork jerky using addition of the LAB-C always showed lower values than the control without addition of the LAB-C. The fermented pork jerky showed a tendency to contain more moisture than the control, regardless of having the same a_w. The lowering of a_w or/and the increase of tenderness in the experimental pork jerky may be caused by metabolites produced during LAB fermentation.

The results of SDS-PAGE shown in Fig. 2 show myofibrillar proteins from the fermented-minced pork with LAB-C was significantly degraded into polypeptides; in contrast to the control sample without the LAB-C. The concentration of the bands with approximate molecular weight of 23 kDa or 37 kDa decreased, while the concentration of the bands with a molecular weight of 16-19 kDa or 29 kDa increased. This myofibrillar protein degradation may be similar to the proteolysis mechanism for myofibrillar proteins reported in dry fermented sausages [6]. From the observed degradation of myofibrillar proteins, the activation of acidic muscle proteases i.e. cathepsins and/or proteinases from the LAB contributed to the hydrolysis of the myofibrillar proteins [7]. And this proteolysis occurred during fermentation of the minced pork with the LAB-C that may affect the decrease in the SFV in the fermented pork jerky described above.

Many LAB species produce EPS and the biopolymers are able to improve the viscosity, texture, and mouth-feel of dairy products. Frengova et al. [8] reported the components of starter cultures for Bulgarian yogurt, *Stc. thermophilus* and *Lb. bulgaricus* revealed extensive EPS production activity when cultivated in whole cow's milk. When the culture pH reduced to near 4.60 after incubation, the amount of EPS increased to 133.0 μg/mg compared to 28.3 μg/mg before incubation. In addition, the

EPS contents in the minced pork jerky with 10% LAB-C during fermentation for 6 h at 43°C significantly increased from 1.33 (0 h) to 69.05 µg/g (after 6 h) (Table 2). The SFV and the tensile strength of the pork jerky decreased with the increase of EPS. When EPS contents in the pork jerky increased from 1.33 to 69.05 g/mg, the SFV decreased from 2.58 kg to 1.25 kg and the values of tensile force and elongation also declined from 14.77 to 9.92 N and 8.0 to 1.6 mm, respectively. Although the moisture contents in the pork jerky increased from 22.3 to 25.6% as the EPS contents increased, the variation in the a_w was relatively small at 0.759 - 0.764. The EPS produced by LAB may cause a decrease in free water in the jerky due to an increase in water-binding capacity. Therefore, the LAB-producing EPS may play an important role in improvement of the texture i.e. tenderness in the fermented-minced pork jerky.

Table 2 EPS content, a_w, moisture, SFV and TS in the pork jerky throughout the process of fermentation for 6h at 43°C

	Fermentation time (h)			
	0	2	4	6
EPS content (µg/g)	1.33±0.09	4.34±2.02	43.0±9.61	69.1±9.94
a_w	0.76±0.01	0.76±0.00	0.76±0.01	0.76±0.01
Moisture (%)	22.3±0.5	23.1±0.4	24.5±0.5	25.6±0.6
SFV (kg)	2.58±0.36	2.22±0.32	1.64±0.29	1.25±0.34
TS				
Tensile strength (N)	14.8±1.3	13.0±0.9	10.0±0.8	9.9±0.7
Elongation (mm)	8.0±1.9	6.8±0.8	2.1±0.2	1.6±0.1

The pork jerky was made by addition of 10% LAB-C, fermentation, and drying for 6-8 h at 20℃. All values represent means of 3 measurements.

From the sensory evaluations of fermented pork jerky experimentally produced using 10% LAB-C, the scores for color, flavor, texture, and tenderness in the experimental jerky were 5.04 - 5.61; especially, the tenderness score was higher. As the overall-acceptability score was 5.27±1.08 (~75% approval), the experimental fermented jerky was evaluated as better on the whole compared to jerky without fermentation.

IV. CONCLUSION

Fermented minced pork jerky processed using the LAB starter cultures showed a more tender texture than the commercial jerky. The increase in the tenderness may be caused by a rise in the hydration of water in the jerky due to the LAB-producing EPS and/or breakdown of the myofibrillar proteins by acidic muscle and/or LAB-producing proteases.

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