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Effects of Number of Washes and Salt Treatment on the Quality Characteristics of Protein Recovered from Alaska Pollock and Pork Leg

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Abstract The aim of this study was to compare the effects of number of washes and salt addition on the meat quality in protein recovered from Alaska Pollock compared with pork leg. Various properties of protein recovered from Alaska Pollock (C, washed twice, no salt) and pork leg (T1, washed twice, no salt; T2, two washes, salt added; T3, washed four times, no salt; and T4, washed four times, salt added) were assessed in this study. Pork leg samples exhibited better color (more whiteness, less yellowness) than Alaska Pollock samples. In pork leg samples, four washes (T3, T4) during processing yielded whiter, less yellow protein than two washes (T1, T2). Overall, the textural property measures were higher in pork leg samples (T2, T3, and T4) than in other samples. Breaking force, jelly strength, and folding resistance were significantly higher in salt-treated pork leg samples (T2, T4) than in the other samples. Our findings demonstrate that protein recovered from pork leg has better color parameters, and physical strength compared with Alaska Pollock-derived protein. A higher number of wash steps and treatment with salt during processing were furthermore found to yield better color, and physical strength in the protein samples.

Keywords Alaska Pollock, pork leg, protein recovery, texture

Introduction

Recently, emerging technology in the production of meat products has allowed developed countries to develop products such as non-meat proteins and to substitute fat or nitrite in meat products with alternative additives (Utama et al., 2018; Yum et al., 2018). Moreover, consumers and meat producers can take advantages from the development of beneficial, palatable and inexpensive functional meat products manufactured by undervalued spent hen meat (Jin et al., 2007). For these reasons, various alternative meat products including protein recovered from meats, crab meat

analogue, and fish cake from undervalued meats especially spent laying hens have been developed. Indeed, the most famous surimi-like product is imitation crab meat or crab stick, which is defined as a refined fish protein product prepared by washing to remove blood, lipids, enzymes, and sarcoplasmic proteins or as minced and washed fish flesh that has been stabilized by cryoprotectants (Vilhelmsson, 1997). It was previously found that water-washed beef or pork also has high salt-soluble protein content, and thus that beef or pork may also be suitable for achieving textures similar to lobster in surimi-like products compared with crab or shrimp flesh (Kang et al., 2007). We previously showed that surimi-like products and sausage can be made using spent laying hen breast meat hydrolysate and that the number of washes during processing and the muscle type affect the physico-chemical properties of chicken-derived surimi (Jin et al., 2007). Little is known, however, about the quality characteristics of surimi-like products made of proteins recovered from Alaska Pollock compared with those made from pork. The purpose of our study was therefore to compare the impacts of number of washes and salt addition on the color, textural traits, and gel characteristics in protein recovered from Alaska Pollock compared with pork leg.

Materials and Methods

Sample preparation

Pig leg meat was obtained from a local slaughterhouse and three individual batches were collected on different days for replicate experiment. The semimembranosus muscle was obtained (above about 1 kg) from hot boning of each pork carcass. After removing external unnecessary adipose tissue, the muscle was divided into three chunks. The experimentally recovered Alaska Pollock- and pork-derived protein samples were divided into five groups: C (Alaska Pollock protein recovered by two washes and no salt treatment), T1 (pork leg protein recovered by two washes and no salt treatment), T2 (pork leg protein recovered by two washes and 2% salt treatment), T3 (pork leg protein recovered by four washes and no salt treatment), and T4 (pork leg protein recovered by four washes and 2% salt treatment). The external fat and skin was removed from the meat and the lean meat was diced into approximately two cubes, which were then ground through a mincer with a 3 mm diameter orifice. Ground samples (500 g) were homogenized in 3 L distilled water using a bowl cutter (AS-30, Ramon Co., Barcelona, Spain) at 7,168×g for 1 min. The slurry was filtered through a 1-mm mesh metal filter to remove connective tissues, after which the filtrate was centrifuged (twice or four times) at 10,000×g (Union 5KR, Hanil, Seoul, Korea) for 25 min at 4°C. The top and bottom layers of the centrifuged slurry were discarded, and the middle layer was recovered and mixed with 0% or 2% salt (99% purified salt) and stuffed into polyvinylidene chloride casings (18 mm diameter). Recovered protein was cooked in a cooking chamber at 78°C for 40 min.

Color assessment

The color (lightness, L^* ; redness, a^* ; yellowness, b^*) of the recovered and processed samples was measured using a Minolta colorimeter (Minolta Chroma Meter CR-300, Minolta Co., Ltd., Tokyo, Japan) that was calibrated with a white standard plate ($Y=92.8$, $x=0.3134$, and $y=0.3193$). Sample whiteness (W) was calculated using the following formula: L^*-3b^* (Park et al., 1996).

Textural properties

Textural properties of the recovered protein samples were determined using a modified version of the method described by Kealy (2006). The brittleness, hardness, cohesiveness, springiness, gumminess, and chewiness were measured using a texture

analyzer (EZ-test, Shimadzu, Tokyo, Japan) equipped with a cylindrical plunger (5 mm diameter) and a 500 N load cell. The crosshead speed was 80 mm/min and the adaptor area was 30 mm².

Gel characteristics and shear force

The gel characteristics of the recovered protein samples were determined according to the method described by Phatcharat et al. (2006). The breaking force, deformation, jelly strength, and gel strength were measured using a texture analyzer (EZ-test, Shimadzu, Tokyo, Japan) equipped with a cylindrical plunger (5 mm in diameter, depression speed of 80 mm/min). For the shear force, samples were sheared once through the center using an Instron 3343 instrument (US/MX50, A&D Co., MA, USA) equipped with a Warner Bratzler shearing device (100 mm/min of crosshead speed).

Statistical analysis

Statistical analysis was carried out for three batches of samples. Data describing the color, textural properties, and gel characteristics of each batch of samples collected were analyzed using the generalized linear model procedure of the SAS software package (SAS Inst. Inc., Cary, NC). The Student-Newman-Keuls multiple-range test was used to compare differences among means. Significant differences ($p < 0.05$) between the mean values of 45 samples were determined for the color, textural properties and gel characteristics ($n=45$).

Results and Discussion

The results of the color assessments of the recovered protein samples are shown in Table 1. The lightness (L^*) and redness (a^*) parameters were significantly higher in pork leg samples (T1–T4) than in the Alaska Pollock sample (C). Among the pork leg samples, yellowness (b^*) was lower in the samples processed with four washes (T3 and T4) and whiteness (W) was higher in the samples washed four times (T3 and T4) compared with the other samples. The results of textural properties in the protein recovery are shown in Table 2. Brittleness, hardness, cohesiveness, gumminess, and chewiness were found to be significantly lower in the T1 samples (protein recovered from pork leg by two washes and no salt treatment) than in the other samples; and the textural property measures were highest in the T2, T3, and T4 protein samples recovered from pork leg compared with the control (C) and T1 samples. The gel characteristics results for the recovered protein samples are shown in Table 3. Breaking force, jelly strength, and folding resistance were significantly higher in salt-treated pork leg samples (T2

Table 1. Effects of number of washes and salt treatment on meat color of protein recovered from pork leg compared with Alaska Pollock

| Treatments | L^* | a^* | b^* | W |
|------------|--------------------------|--------------------------|--------------------------|--------------------------|
| C | 70.71 ^D ±1.03 | 0.56 ^E ±0.25 | 4.80 ^C ±0.09 | 56.30 ^D ±1.10 |
| T1 | 80.82 ^A ±0.97 | 6.81 ^C ±0.22 | 7.29 ^A ±0.12 | 58.94 ^C ±0.98 |
| T2 | 75.55 ^B ±1.01 | 5.73 ^D ±0.19 | 6.52 ^B ±0.30 | 55.99 ^D ±0.67 |
| T3 | 73.61 ^C ±1.10 | 10.87 ^A ±0.37 | 1.98 ^D ±0.24 | 67.68 ^B ±0.93 |
| T4 | 69.33 ^E ±0.88 | 9.90 ^B ±0.35 | -2.13 ^E ±0.31 | 75.73 ^A ±0.35 |

^{A-E} Means with different superscripts in the same column differ significantly ($p < 0.05$).

W, whiteness ; C, protein recovered from Alaska Pollock by two washes and no salt treatment; T1, protein recovered from pork leg by two washes and no salt treatment; T2, protein recovered from pork leg by two washes and salt treatment; T3, protein recovered from pork leg by four washes and no salt treatment; T4, protein recovered from pork leg by four washes and salt treatment.

Table 2. Effects of number of washes and salt treatment on the textural properties of protein recovered from pork leg

| Treatments | Brittleness (kg) | Hardness (kg) | Cohesiveness (%) | Springiness (mm) | Gumminess (kg) | Chewiness (kg/mm) |
|------------|-------------------------|--------------------------|--------------------------|---------------------------|---------------------------|-----------------------------|
| C | 0.41 ^A ±0.01 | 0.53 ^{AB} ±0.02 | 51.58 ^A ±3.31 | 13.87 ^A ±0.25 | 27.55 ^{AB} ±1.60 | 382.22 ^{AB} ±21.54 |
| T1 | 0.18 ^B ±0.02 | 0.25 ^D ±0.01 | 31.61 ^B ±4.98 | 13.80 ^A ±0.21 | 7.92 ^D ±1.56 | 109.18 ^D ±13.70 |
| T2 | 0.41 ^A ±0.02 | 0.57 ^A ±0.01 | 55.67 ^A ±2.54 | 13.35 ^{AB} ±0.16 | 31.84 ^A ±1.20 | 425.88 ^A ±23.65 |
| T3 | 0.36 ^A ±0.03 | 0.49 ^B ±0.02 | 53.02 ^A ±3.01 | 13.03 ^B ±0.18 | 26.17 ^B ±0.98 | 341.49 ^B ±26.93 |
| T4 | 0.36 ^A ±0.01 | 0.36 ^C ±0.03 | 48.42 ^A ±2.87 | 13.66 ^{AB} ±0.22 | 17.55 ^C ±2.97 | 239.62 ^C ±17.68 |

^{A-D} Means with different superscripts in the same column differ significantly ($p < 0.05$).

C, protein recovered from Alaska Pollock by two washes and no salt treatment; T1, protein recovered from pork leg by two washes and no salt treatment; T2, protein recovered from pork leg by two washes and salt treatment; T3, protein recovered from pork leg by four washes and no salt treatment; T4, protein recovered from pork leg by four washes and salt treatment.

Table 3. Effects of number of washes and salt addition on the gel characteristics and shear force of protein recovered from pork leg

| Treatments | Breaking force (g) | Deformation (mm) | Jelly strength (g/cm ²) | Gel strength (g/mm) | Folding test | Shear force (kg/cm ²) |
|------------|-----------------------------|--------------------------|-------------------------------------|--------------------------------|--------------------------|-----------------------------------|
| C | 577.00 ^{AB} ±17.56 | 10.10 ^A ±1.87 | 2,938.64 ^{AB} ±48.75 | 5,828.20 ^A ±531.21 | 3.00 ^C ±0.10 | 1.79 ^B ±0.01 |
| T1 | 436.33 ^D ±20.36 | 7.10 ^{AB} ±0.56 | 2,222.23 ^D ±42.19 | 3,112.12 ^B ±120.65 | 2.33 ^D ±0.23 | 2.49 ^A ±0.04 |
| T2 | 597.33 ^A ±15.68 | 6.92 ^{AB} ±1.25 | 3,075.53 ^A ±67.36 | 4,176.56 ^{AB} ±263.70 | 4.00 ^{AB} ±0.11 | 1.68 ^B ±0.02 |
| T3 | 499.67 ^C ±21.56 | 5.74 ^B ±0.87 | 2,544.78 ^C ±57.12 | 2,867.98 ^B ±487.09 | 3.67 ^B ±0.17 | 2.31 ^A ±0.03 |
| T4 | 525.67 ^B ±16.14 | 7.62 ^{AB} ±1.12 | 2,677.20 ^B ±49.34 | 3,999.02 ^{AB} ±510.87 | 4.67 ^A ±0.20 | 1.17 ^B ±0.01 |

^{A-D} Means with different superscripts in the same column differ significantly ($p < 0.05$).

C, protein recovered from Alaska Pollock by two washes and no salt treatment; T1, protein recovered from pork leg by two washes and no salt treatment; T2, protein recovered from pork leg by two washes and salt treatment; T3, protein recovered from pork leg by four washes and no salt treatment; T4, protein recovered from pork leg by four washes and salt treatment.

and T4) than in pork leg samples processed in the absence of salt (T1 and T3). The correlation coefficient data for the recovered proteins are shown in Table 4 and reveal that breaking force and gumminess of the samples were closely related with the jelly strength and chewiness, respectively. Hardness was found to correlate closely with gumminess and chewiness and deformation correlated closely with gel strength. Lightness in color was found to be negatively related with aroma. Overall, chewiness correlated closely with textural properties and gel characteristics e.g., breaking force, jelly strength, brittleness, hardness, cohesiveness, and gumminess.

From the results of this study, it was found that washing times and salt treatment do affect the physical properties of recovered protein. Surface color of meat products in particular is an important factor because it affects consumer purchase decisions: consumers specifically like a bright white and less yellow color in surimi-like materials such as crab meat analogue or protein recovered from meat. In general, myoglobin (meat pigment) plays an essential role in the whiteness in such recovered proteins, and high-quality surimi-like materials with a whiter appearance can be obtained when dark muscle including myoglobin is removed as much as possible (Ochiai et al., 2001). Jafarpour and Gorczyca (2008) also reported that the effective removal of myoglobin in surimi is washing process, convention method. In this study, washing samples four times (T3 and T4) instead of only twice was found to result in significantly more whiteness and less yellowness, indicating that four washes during processing is better than two washes in terms of color. This difference may be a result of more washes removing more myoglobin, since myoglobin is water-soluble. We furthermore found in this study (data not shown) that the myoglobin content in the samples subjected to four washes (T3 and T4) was lower than that in samples subjected to

Table 4. Correlation coefficients of various parameters used to assess protein recovered from pork leg

| Items | BF | DE | JS | Fo | Br | Ha | Coh | Gu | L* | a* | b* | Fl | Coh | Ju |
|-------|------|------|------|------|------|------|------|------|----|----|------|------|------|------|
| JS | 1.00 | - | - | - | - | - | - | - | - | - | - | - | - | - |
| GS | - | 0.94 | - | - | - | - | - | - | - | - | - | - | - | - |
| Ha | - | - | 0.80 | - | - | - | - | - | - | - | - | - | - | - |
| Coh | - | - | - | - | - | 0.80 | - | - | - | - | - | - | - | - |
| Gu | - | - | - | - | 0.80 | 0.97 | 0.90 | - | - | - | - | - | - | - |
| Che | 0.80 | - | 0.80 | - | 0.81 | 0.97 | 0.89 | 1.00 | - | - | - | - | - | - |
| W | - | - | - | - | - | - | - | - | - | - | 0.94 | - | - | - |
| Ar | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| Fl | - | - | - | 0.85 | - | - | - | - | - | - | - | - | - | - |
| Ju | - | - | - | - | - | - | - | - | - | - | - | - | - | - |
| Te | - | - | - | 0.82 | - | - | - | - | - | - | - | 0.83 | 0.83 | 0.85 |
| OA | - | - | - | 0.88 | - | - | - | - | - | - | - | - | 0.82 | - |

* Level of significance of correlation coefficients: $p < 0.05$.

BF, breaking force; DE, deformation; GS, gel strength; JS, jelly strength; Fo, folding resistance; Br, brittleness; Ha, hardness; Coh, cohesiveness; Gu, gumminess; Che, chewiness; W, $L^* - 3b^*$; Ar, aroma; Fl, flavor; MC, meat color; Ju, juiciness; Te, tenderness; OA, overall acceptability.

only two washes. Generally, the color of processed meat products are heavily influenced by ingredients and additives such as sodium nitrite, salt, or colorants (Eskandari et al., 2013; Hammad HHM et al., 2017). In addition, these samples with salt (T2 and T4) and control exhibited significantly lower shear force than groups with the absence of salt (T1 and T2) ($p < 0.05$). This result was considered that the protein recovered from Alaska Pollock or addition with salt might be influence tenderness of product. In this study, it was found that wash steps and salt treatment positively influenced the color of the pork-derived protein by decreasing the yellowness and increasing the whiteness of the samples.

The number of washes and treatment with salt were furthermore found to influence the textural properties and gel characteristics of the samples. Overall, samples processed by only two washes and in the absence of salt (T1) exhibited significantly less brittleness, hardness, cohesiveness, gumminess, and chewiness than the other samples. These samples (T1) were also shown to have lower breaking force, deformation, jelly strength, gel strength, and folding resistance, while samples processed by four washes and those treated with salt (T2, T3, and T4) exhibited higher textural properties and gel characteristics. In addition, these samples with salt (T2 and T4) and control exhibited significantly lower shear force than groups with the absence of salt (T1 and T2) ($p < 0.05$). This result was considered that the protein recovered from Alaska Pollock or addition with salt might be influence tenderness of product. The number of wash steps and the addition of salt during processing do indeed play an important role in determining the quality characteristics of recovered protein, since the main ingredient of such proteins is wet concentrate of water-insoluble myofibrillar protein (salt-soluble protein), and its structural integrity is influenced by the strength of the interacting forces within the protein network and the binding of free water within this network (Morin et al., 2004). Gaudette and Pietrasik (2017) revealed that reducing salt content restrict myofibrillar protein extraction due to lower both water binding and strength of meat, which means to influence texture properties. In this study, the addition of salt may have improved the binding of protein networks by increasing the solubility of myofibrillar protein since soluble myofibrillar proteins exhibit high cohesive attraction after heating. This is consistent with the better physical strength observed for the T2 and T4 (salt-treated) samples compared with the C, T1, and T3 samples

processed in the absence of salt in this study. The preliminary experiments carried out in this study showed that the myofibrillar protein content was higher in samples subjected to four washes (T3 and T4 by 5.25 and 5.32 mg/g, respectively) than those subjected to only two washes (C, T1, and T2 by 5.09, 5.00, and 5.03, respectively), which is likely to account for the fact that the pork leg recovered protein processed with four washes had higher physical strength than the Alaska Pollock samples and the pork protein samples processed with only two washes.

Conclusions

This study was carried out to assess the effects of number of washes and of salt treatment on the color, textural properties, and gel characteristics outcomes of protein recovered from pork leg compared with that recovered from Alaska Pollock. The findings of this study demonstrate that the protein recovered from pork leg has better color parameters and physical strength compared with that recovered from Alaska Pollock. In terms of the effects of the number of washes and of salt treatment during protein recovery, four washes and the inclusion of salt resulted in better color, and physical strength compared with protein recovered with only two washes in the absence of salt. Sensorial traits are crucial factors for consumer choice and purchase of meat products, and four washes in addition to salt treatment should therefore be carried out in the recovery of protein from pork leg for optimal quality characteristics of the recovered protein. Our findings could be useful and economical for the industry processors to get refined pork protein after 4 washing times rather than refined fish protein.

Conflicts of Interest

The authors declare no potential conflict of interest.

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Author Contributions

Investigation: Jin SK. Formal analysis and Methodology: Jin SK. Writing-original draft: Jin SK, Yim DG, Hur SJ. Writing-review & editing: Jin SK, Lee SY, Yim DG, Hur SJ.

Ethics Approval

This article does not require IRB/IACUC approval because there are no human and animal participants.

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