



## Oxidative stability of gelatin coated pork at refrigerated storage

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### ABSTRACT

As consumers are chiefly concerned with odor and appearance, refrigerated meats have a greater possibility of rejection due to their perishability. A treatment was designed to preserve the quality parameters such as color and oxidation rate. The treatment encompassed coating pork loin samples with aqueous porcine gelatin solutions (175 bloom) at three concentrations (0%, 10% and 20%) and studying its effect on lipid oxidation, protein oxidation, oxidation pre-cursors, color and purge of meat. Vacuum packed *Longissimus dorsi* muscle was cut into 10 × 5 × 1 cm samples and randomly dipped in one of three solutions. Samples were stored at 4 °C and analyzed for thio-barbituric acid reactive substances, metmyoglobin content, protein carbonyls, heme iron content and color change. There was a significant difference ( $p < 0.05$ ) for TBARS values, protein carbonyls, heme iron content, total color change, metmyoglobin content with treatments maintaining values more similar to a fresher product when compared to control on days 3, 5 and 7. There was no significant difference ( $p > 0.05$ ) between 10% and 20% gelatin coating on any of the parameters between storage days.

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### 1. Introduction

Meat spoilage occurs due to microbial and chemical deterioration (Sebranek, Sewalt, Robbins, & Houser, 2005). Microbial spoilage of meat arises due to improper handling of meat prior to and after it is purchased by the consumer. This type of spoilage involves simple mechanisms of contamination and spoilage and once identified, the means to counter them are numerous including thermal/non-thermal processing and the use of preservatives. Chemical spoilage of meat generally occurs due to protein and lipid oxidation. Both of these types of oxidation are dependent on the meat composition, meat product processing, handling and storage environment (Saghir, Wagner, & Elmadfa, 2005). Lipid oxidation in meat can vary greatly, ranging from a change in flavor or color, structural damage of proteins to a loss of freshness that would affect the demand by the consumer, thus extensively affecting the meat industry (Mielnik, Aaby, & Skrede, 2003). Despite the utilization of several technologies, oxidation of meats is still a major concern in the meat industry (Jo, Ahn, & Byun, 2002).

Approximately 50% of the worldwide daily protein intake is from pork (Davis & Lin, 2005). Mielche (1995) reported that the rates and amounts of lipid oxidation during storage decreased in meats as turkey > chicken > pork > beef. Pork meat is second to beef in total iron content (Tang, Kerry, Sheehan, Buck-

ley, & Morrisely, 2001). Iron in meat in the form of non-heme iron catalyzes the process of lipid oxidation (Love & Pearson, 1974). Vuorela et al. (2005) reported that the carbonyl content as a result of protein oxidation in pork patties increased by 80% through 7 days of refrigeration. Antioxidants and packaging technologies have been applied to maintain quality, but considering risks due to the consumption of commercial antioxidants, low convenience and cost effectiveness of packaging solutions, there is a need for an alternative technology that minimizes risk, offers convenience and reduces the price of processing. Edible coatings have been applied on meats to preserve the quality, but their application is limited in the meat industry (Gennadios, Hanna, & Kurfh, 1997). Gelatin's potential as an edible coating comes from its film forming capability. Gelatin coatings on meat have been found to decrease moisture loss and control oxidation at frozen conditions and in combination with vacuum packaging (polyamide/polyethylene, 30/70) and oxygen permeable (polyvinyl chloride) packaging (Villegas, O'Connor, Kerry, & Buckley, 1999).

The purpose of this research was to study the effect of gelatin coating in preserving the quality of refrigerated pork for a storage period of 7 days. The objectives were: (a) coating of pork samples with pork gelatin solutions of selected concentrations (0%, 10% and 20%) and subjecting them to refrigerated storage at 4 °C and (b) investigation of change in total color, purge, lipid oxidation, protein oxidation, metmyoglobin content and heme iron content of treated and control samples on days 0, 3, 5 and 7.

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## 2. Materials and methods

### 2.1. Coating

Pork was slaughtered, processed and vacuum packed at a regional slaughter facility. Vacuum packed, never frozen, pork *Longissimus dorsi* was obtained from local market within two days of slaughter. The muscle was cut into samples with approximate dimensions of 10 cm × 5 cm × 1 cm, divided into three groups, weighed and refrigerated (4 °C) until treated. Aqueous solutions of gelatin (Pork skin gelatin, Type A, bloom: 175, Sigma, St. Louis, MO, USA) at 10% and 20% concentrations were made. Treated samples were dipped into gelatin solutions at 60 °C. The control samples were not dipped. After treatment all samples were reweighed. All samples were stored at 4 °C with no packaging (overwrap) as a means to accentuate oxidation in a simulated fresh retail display setting.

### 2.2. Lipid oxidation measurement

Thio-barbituric acid reactive substances (TBARS) for the meat samples were calculated and equated to lipid oxidation of meat. The procedure adopted was similar to that of Ahn et al. (1998). Five grams of meat sample was added to 50 mL distilled deionized water (DDW) and homogenized for 45 s in an Osterizer blender (Osterizer Corporation, Milwaukee, WI). To 1 mL of meat homogenate, 50 µL of 7.2% solution of butylated hydroxytoluene in ethanol and thio-barbituric acid/trichloroacetic acid [20 mM TBA and 15% (w/v) TCA] were added. Samples were vortexed and incubated in a water bath at 90 °C for 15 min. Samples were cooled to room temperature and centrifuged at 3000 × g for 15 min at 5 °C. The absorbance of the supernatant was read at 531 nm against a blank prepared with 1 mL of DDW and 2 mL of TBA/TCA solution. The amount of TBARS was expressed as milligrams of malondialdehyde (MDA) per kilogram of meat.

### 2.3. Preparation of MDA standard curve

The MDA standard stock solution (100 µM) was prepared according to the method adopted by Sim, Salonikas, Naidoo, & Wilcken (2003). To 100 mL of 1% sulfuric acid, 239 µL of 1,1,3,3-tetraethoxypropane (TEP) was added and the solution incubated for 2 h at room temperature. This solution was used to create a standard curve at MDA concentrations of 10, 20, 40, 60, 80 and 100 µM.

### 2.4. Color measurement

The color of meat samples was measured immediately after their separation as individual samples. Surface color measurements were determined using a CR-400 Chromameter (Minolta Co., Osaka, Japan) on days 0, 3, 5 and 7 of storage. The samples were measured in triplicate and averaged. The average *L*, *a*, and *b* values were used in order to calculate  $\Delta E$ , total color change, with the formula as follows:

$$\Delta E = \sqrt{((L - L')^2 + (a - a')^2 + (b - b')^2)}$$

where *L*, *a* and *b* denote values at 0 day of storage time, and *L'*, *a'* and *b'* denote values at 3, 5 and 7 days of storage time (Antoniewski, Barringer, Knipe, & Zerby, 2007). '*L*' measures the lightness of sample; '*a*' measures the variation between red and green; '*b*' measures the variation between blue and yellow.

### 2.5. Purge

Purge of the sample was calculated by measuring the weight loss of the meat samples with respect to storage time, i.e. on days 3, 5 or 7:

$$\% \text{ Weight loss} = \frac{(W_0 - W') \times 100}{W_0}$$

where *W*<sub>0</sub> is the initial weight of sample and *W'* is the weight of sample after 3, 5 or 7 days.

### 2.6. Heme iron content

Total heme pigments were calculated using the protocol of Hornsey (1956). Sample (2 g) was placed into a 50 mL polypropylene tube, and 9 mL of acidified acetone (90% acetone, 8% deionized water, 2% conc. HCl) was added. Using a glass rod, the meat was macerated and the tube was placed in a dark cabinet for 1 h at room temperature. The extract was filtered using Whatman (No. 42) filter paper (Whatman Internations Ltd., Maidstone, England). The filtered extract's absorbance was read at 640 nm against a blank containing acidified acetone.

Total pigments were calculated using the formula:

$$\text{Total pigments } (\mu\text{g/g of meat}) = A_{640} \times 680$$

and heme iron content was calculated as described by Clark, Mahoney, and Carpenter (1997):

$$\text{Heme iron } (\mu\text{g/g of meat}) = \text{total pigment } (\mu\text{g/g}) \times 0.0882$$

### 2.7. Metmyoglobin content

Metmyoglobin content was calculated using the method described by Fernández-López et al. (2003). Minced meat (5 g) was taken from each sample and myoglobin was extracted with 10 volumes of 0.04 M phosphate buffer at 4 °C and pH 6.8. Samples were homogenized for 15 s with a Polytron homogenizer (Brinkman Instruments, New York, NY, USA) at a speed of 10,800 rpm and the homogenates were centrifuged for 30 min at 15,000 rpm at 5 °C. The supernatant was filtered through Whatman (No. 42) filter paper and the absorbance was read at 525, 572 and 730 nm. Percentage of Mmb was determined using the formula (Krzywicki, 1979):

$$\text{Mmb } (\%) = 1.395 - ((A_{572} - A_{730}) / (A_{525} - A_{730})) \times 100$$

### 2.8. Protein oxidation

Protein oxidation was measured using method described by Vuorela et al. (2005). Protein oxidation was calculated by using two measurements: (a) carbonyl quantification and (b) protein quantification. Meat sample (1 g) was taken into a 50 mL polypropylene tube and homogenized with 10 mL of 0.15 M KCl using a Polytron homogenizer (Brinkman Inc., Westbury, NY, USA) for 60 s. Homogenate (100 µL) was transferred into a 2 mL micro-centrifuge tube to which 10% trichloroacetic acid solution was added. The solution was vortexed for 30 s and centrifuged for 5 min at 5000 rpm. The supernatant was removed and the pellet was used for carbonyl content measurement. The pellet was added to 1 mL of 2 M HCl with 0.2% dinitrophenyl hydrazine. After incubation for 1 h (shaken periodically), 1 mL 10% trichloroacetic acid was added. The sample was vortexed and centrifuged for 5 min at 5000 rpm and the supernatant was removed without damaging the pellet. One millilitre 1:1 ethanol/ethyl acetate was used to wash the pellet and then centrifuged for 5 min at 5000 rpm, this step was repeated twice to ensure purity. The pellet was then dis-

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