EFFECT OF TENDERNESS CLASSIFICATION AND AGING TIME ON ABUNDANCE OF PEROXIREDOXIN-6 IN PORCINE LONGISSIMUS

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I. OBJECTIVES

Meat tenderness significantly influences meat palatability, consumer satisfaction, and industry profitability. Protein oxidation does influence the development of meat tenderness. Peroxiredoxin-6 (PRDX6), an antioxidant protein, contributes to hydrogen peroxide degradation. Previous studies have suggested that PRDX6 might be a protein marker for tenderness in bovine biopsies and samples collected shortly after slaughter. However, the relationship between PRDX6 and ultimate meat tenderness in porcine skeletal muscle is not defined. This study aimed to examine PRDX6 during postmortem aging to further understand the relationship between PRDX6 and meat tenderness.

II. MATERIALS AND METHODS

Fresh pork loins were collected at 1 d postmortem. Pork chops (2.5 cm) containing the *longissimus dorsi* muscle were fabricated and aged for 1, 8, 14, or 21 d postmortem. After aging, chops were cooked to 68°C, and instrumental tenderness was measured using of the star probe attachment on an Instron (Instron, Norwood, MA). Star probe values were used to classify chops into high (star probe > 7.0 kg, $n = 6$) and low (star probe < 5.8 kg, $n = 6$) star probe groups (21 d values). Sarcoplasmic proteins from *longissimus dorsi* at each aging time were solubilized in ice-cold, low-ionic strength buffer (50 mM Tris-HCl and 1 mM EDTA; pH 8.5), and samples with a reducing agent were prepared. A pooled reference representing every sample in the experiment was prepared. Western blot analysis was employed to identify PRDX6. Each sample was fractionated on a 15% acrylamide separating gel and transferred to PVDF membrane. After 1-h blocking within 5% nonfat dried milk, membranes were incubated overnight with primary antibody (1:10,000 anti-PRDX6; AB1333, AbCam) at 4°C. Membranes were incubated with secondary antibody (1:20,000 Goat anti-rabbit IgG-HRP) for 1 h at room temperature. The results were visualized using the chemiluminescent detection kit (ECL Prime; GE Healthcare, Piscataway, NJ), and images were obtained and analyzed using a ChemiImager 5500 (Alpha Innotech Corp., San Leandro, CA) and Alpha Ease FC software (version 3.03; Alpha Innotech Corp.). Samples were run in duplicate and reported as a ratio to the reference that was run on each gel. PRDX6 abundance (as a ratio to a within gel reference) was analyzed using PROC MIXED of SAS version 9.4 (SAS Institute Inc., Cary, NC) with fixed effects of days aging and star probe category and a random effect of gel. Significance was denoted with $P < 0.05$.

III. RESULTS

Star probe classification (at 21 d postmortem) did not affect PRDX6 abundance (high star probe, 1.16; low star probe, 1.11). PRDX6 abundance was not affected by aging time (Day 1, 1.11; Day 8, 1.19; Day 14, 1.04; Day 21, 1.20).

IV. CONCLUSION

These results indicate that PRDX6 abundance is not changed during postmortem aging. Star probe classification did not result in a difference in PRDX6 abundance. PRDX6 may not be a substrate for degradation by proteinases in postmortem muscle. Alternatively, changes in PRDX6 that do occur in postmortem muscle may be complete by 1 d postmortem. PRDX6 undergoes various post-translational modifications, which, when analyzed under non-reducing conditions, may provide further insight into the relationship between PRDX6 and meat tenderness.

Keywords: peroxiredoxin-6, pork, postmortem aging, tenderness