# CALPAIN AUTOLYSIS AND PROTEOLYSIS OF BOS TAURUS AND BOS INDICUS LONGISSIMUS LUMBORUM DURING 14-DAY AGING

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## I. OBJECTIVES

The objective of this study was to determine calpain autolysis and protein degradation in aged *Bos taurus* (Angus) and *Bos indicus* (Brahman) steaks and their relationship with tenderness.

### II. MATERIALS AND METHODS

Angus and Brahman steers (n = 14 per breed) were reared in the same conditions. Samples from the longissimus lumborum (LL) were taken at 1 h and 24 h postmortem, immediately frozen in liquid nitrogen, and stored at -80°C until analysis. At 48 h, an approximately 8-cm section was removed posterior to the 12th and 13th rib, which was subsequently cut into three 2.54-cm-thick steaks. Steaks were vacuum packaged and aged at 3°C for 14 d postmortem. After aging, 2 steaks were frozen at  $-40^{\circ}$ C until further analysis of tenderness, and the remaining steak was used for protein biochemistry. For protein extraction, pulverized LL was diluted in extraction buffer, homogenized, and centrifuged. Protein concentration of supernatants was determined using a bicinchoninic acid protein assay, and samples were diluted to obtain equal protein concentrations. Proteins were separated using sodium dodecyl sulfate-polyacrylamide gel electrophoresis. Proteins were then transferred to nitrocellulose membranes. Membranes were stained for total protein, which was used as an index to normalize protein content. Membranes were blocked and incubated with primary antibodies for desmin, troponin-T, and µ-calpain, followed by secondary antibody conjugated with fluorescent dye. Bands were detected using the LI-COR Odyssey and quantified using ImageStudio software. For tenderness evaluation, steaks were thawed at 4°C and cooked to an internal temperature of 71°C. Tenderness was determined objectively by Warner-Bratzler shear force (WBSF) and a trained sensory panel (1 = extremely tough; 8 = extremely tender). Data were analyzed using SAS mixed procedure (SAS Institute Inc., Cary, NC) with fixed effects of time, breed, and the interaction; time was considered a repeated measure for proteolysis. Tenderness and WBSF were analyzed using the fixed effect of breed.

#### III. RESULTS

Breed affected  $\mu$ -calpain autolysis (P = 0.0085). Brahman showed less  $\mu$ -calpain autolysis at 24 h (P = 0.0040) than Angus. Degradation of troponin-T (P = 0.0028) and desmin (P = 0.0210) were lower for Brahman compared to Angus. Troponin-T degradation was lower at 24 h (P = 0.0288) and 14 d (P = 0.0106) in Brahman compared to Angus. Desmin degradation was also lower at 24 h (P = 0.0421) in Brahman. Brahman were less tender than Angus, evidenced by higher WBSF (P = 0.0029) and lower sensory panel scores (P = 0.0007). Brahman also showed greater variability in tenderness than Angus (sensory:  $4.51 \pm 0.91$  vs.  $5.60 \pm 0.55$ , respectively). Within Brahman samples, protein degradation (14 d) of desmin (P = 0.0161) and troponin-T (P = 0.0090), and  $\mu$ -calpain autolysis at 24 h (P = 0.0211), were associated with tenderness.

## IV. CONCLUSION

As expected, there are differences between proteolysis of steaks from Angus and Brahman breeds. There is a large amount of tenderness variability within Brahman. Tender Brahman do exist, and greater  $\mu$ -calpain autolysis is positively associated with proteolysis and tenderness. In order to enhance value of Brahman and reduce variability, it is important to identify ante- and postmortem predictors of tenderness specifically aimed at Brahman.

Keywords: Brahman, calpain, desmin, tenderness, troponin-T