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Tandem Mass Tag Labeling to Identify Proteome Changes in Beef *Longissimus Lumborum* and *Psoas Major* Muscles During Early Postmortem Period

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Objectives

Longissimus lumborum (LL) and *psoas major* (PM) are important muscles in beef hindquarters that exhibit variation in meat quality attributes. Postmortem metabolism (muscle-to-meat conversion) affects biochemical properties of muscles and in turn influence the meat quality. Although previous research has indicated that variation in the proteome profile of LL and PM post-rigor influences meat quality attributes such as tenderness and color stability during retail display, limited research has examined the influence of early postmortem metabolism on meat quality. Tandem mass tag (TMT) labeling is a chemical labeling approach used for accurate mass spectrometry-based quantification and identification of biological macromolecules. Therefore, the objective of this study was to use TMT labeling to examine proteome profile variation between beef LL and PM during the early postmortem period.

Materials and Methods

Muscle biopsy samples were collected from carcasses (n = 4) at 45 min, 12 h, and 36 h postmortem from a commercial beef processing facility. Samples were frozen immediately in liquid nitrogen and stored at –80°C until proteomic analysis. Proteome was analyzed using TMT label containing ten different isobaric compounds with the same mass and chemical structure composed of an amine-reactive NHS-ester group, a spacer arm, and a mass reporter. After labeling and peptide fractionation, all the samples were multiplexed and ran through the Orbitrap Velos mass spectrometer equipped with a Nanospray Flex ion source to identify differentially abundant proteins. The proteins exhibiting 1.5-fold or more intensity difference and a statistical difference ($P < 0.05$) between LL and PM or within the muscles during the postmortem were reported as differentially abundant.

Results

Seventy differentially abundant proteins ($P < 0.05$) were identified from three comparisons between the muscles (31 proteins in PM 45 min vs. LL 45 min, 41 proteins in PM 12 h vs. LL 12 h, 49 proteins in PM 36 h vs. LL 36 h). However, no difference ($P > 0.05$) in protein expression within a muscle was observed during these time points. The differentially abundant proteins were mainly involved in oxidative phosphorylation and ATP-related transport, tricarboxylic acid cycle, NADPH regeneration, fatty acid degradation, muscle contraction, calcium signaling, chaperone activity, oxygen transport, as well as degradation of the extracellular matrix. At early postmortem, overabundant anti-apoptotic proteins in LL could cause high metabolic stability, enhanced autophagy, and delayed apoptosis, while overabundant metabolic enzymes and pro-apoptotic proteins in PM could accelerate the reactive oxygen species generation and programmed cell death.

Conclusion

Differentially abundant proteins between LL and PM during the early postmortem were primarily associated with cellular metabolism and programmed cell death. The greater oxidative and color stability in LL compared to PM could be related to the increased expression of anti-apoptotic proteins and the decreased expression of metabolic enzymes and proapoptotic proteins in LL.