

Influence of sex class of feedlot crossbred Angus x Nellore cattle on post-mortem proteolysis and beef tenderness

Pollyana Garbossa¹, Mirele Poleti², Taiane Martins¹, Hellencris Rocha¹, Tamyres de Amorim³, Ana Giulia do Barco⁴, Hugo dos Reis³, Letícia Augusta Stella¹, Lenise Mueller³, Adrielle Ferrinho³, Julian Andrés Muñoz³, Júlio Cesar Balieiro¹, Angélica Simone Pereira¹

¹ School of Veterinary Medicine and Animal Science, University of São Paulo (FMVZ/USP), Department of Animal Nutrition and Production, Pirassununga, Brazil

² Faculty of Animal Science and Food Engineering, University of São Paulo (FZEA/USP), Department of Veterinary Medicine, Pirassununga, Brazil

³ Faculty of Animal Science and Food Engineering, University of São Paulo (FZEA/USP), Department of Animal Science, Pirassununga, Brazil

⁴ Hermínio Ometto Foundation (Uniararas), Araras, Brazil

Introduction: Meat consumers have been increasingly demanding high-quality beef (Warner et al., 2010). In this sense, crossbred bovine breeds have been widely used to produce this product in tropical regions (Favero et al., 2019). Furthermore, it is possible to adopt other strategies to complement the advantages of the crossbred breeds, as the use of different sex classes (Bonny et al., 2016). However, the molecular mechanisms that contribute to the variations in beef tenderness resulting from the sex class and also from the aging process are not well known. Therefore, we aimed to evaluate the influence of sex class on beef tenderness, as well as to deepen the knowledge by the evaluation of Longissimus muscle protein profile during post-mortem proteolysis.

Materials and Methods: A total of 150 F1 Angus x Nellore cattle was used from the same contemporary group, with an average slaughter age of 16 months, divided into heifers, steers, and bulls. The animals were confined and fed with the same diet and management for 150 days. Subsequently, the animals were harvested and during boning samples of the Longissimus muscle were collected and aged at 0 to 2 °C for the Warner-Bratzler shear force (two, seven, 14, and 21 days) and sensory analysis (14 days). For the protein profile analysis, 36 samples of Longissimus muscle were randomly collected during slaughter and at the end of the aging periods of two and seven days. The steps followed were protein extraction, unidimensional electrophoresis (SDS/PAGE), analysis of the gel images, statistical analysis to identify the differentially abundant bands, digestion of the "in-gel" bands, identification of the proteins by liquid chromatography-mass spectrometry (LC-MS/MS; $P < 0.0001$), and bioinformatics analysis of functional enrichment and protein-protein interaction network.

Results: Sex classes, as well as aging periods, changed the Longissimus muscle protein profile, influencing post-mortem proteolysis, consequently affecting the beef tenderness. Steers and heifers were the cattle sex classes that allow the production of tender beef, with a shorter aging period. Consumer panelists preferred steers' meat, which they considered to be more tender and juicier than their counterparts. Heifers and bulls had a similar bands profile, while steers had the most differential bands profile, both only in terms of sex classes and in terms of sex classes throughout the aging periods. The main proteome changes occurred between the slaughter time and the second aging day, demonstrating the importance of the first aging days for obtaining beef tenderness. In summary, the unidimensional electrophoresis bands, which showed particularities in each sex class, throughout the aging periods, were those in which proteins are mainly found in energy metabolism pathway.

Conclusions: Therefore, it is believed that the key to the particularity between the cattle sex classes, as well as post-mortem proteolysis, lies in this pathway. Moreover, the importance of the biological processes of muscle structuring and contraction, ion transport, and response to cellular stress is highlighted for the development of beef tenderness. Finally, the present study confirmed previously identified proteins (AGL, ANXA6, EEF1A2, LDB3, PGAM2, AK1, and TNNC2) as potential biomarkers for beef tenderness.

Acknowledgements and Financial support statement: We are grateful to São Paulo Research Foundation (FAPESP) for the scholarship and financial support (Grants #2018/24624-7 and #2012/50788-0) and to Brazilian National Council for Scientific and Technological Development (CNPq) for the financial support (process n. 409186/2018-0).

Literature:

Bonny, S. P. F., Hocquette, J.-F., Pethick, D. W., Farmer, L. J., Legrand, I., Wierzbicki, J., Allen, P., Polkinghorne, R. J., & Gardner, G. E. (2016). The variation in the eating quality of beef from different sexes and breed classes cannot be completely explained by carcass measurements. *Animal*, 10, 987–995. doi:10.1017/S175173111500292X

Favero, R., Menezes, G. R. O., Torres Jr., R. A. A., Silva, L. O. C., Bonin, M. N., Feijó, G. L. D., Altrak, G., Niwa, M. V. G., Kazama, R., Mizubuti, I. Y., & Gomes, R. C. (2019). Crossbreeding applied to systems of beef cattle production to improve performance traits and carcass quality. *Animal*, 13, 2679–2686, 2019. doi:10.1017/S1751731119000855

Warner, R. D., Greenwood, P. L., Pethick, D. W., & Ferguson, D. M (2010). Genetic and environmental effects on meat quality. *Meat Science*, 86, 171–183. doi:10.1016/j.meatsci.2010.04.042