THE USE OF ^{1}H NMR IN THE QUANTIFICATION OF FREE AMINO ACIDS IN DRY-AGED AND WET-AGED BEEF

Cristiano S. Prado[1*], Andressa K. Silva[1,4], Cintia S. Minafra e Rezende[1], Jonatã H. R. Souza[2], Sérgio B. Pflanzer Junior[2], Maria F. C. Santos[3], Claudio F. Tormena[3], Luciano M. Lião[4]

[1]School of Veterinary Medicine and Animal Science, Federal University of Goiás, Goiânia-GO, Brazil

[2] Department of Food Engineering and Technology, State University of Campinas, Campinas-SP, Brazil

[3]Chemistry Institute, State University of Campinas, Campinas-SP, Brazil

[4]Chemistry Institute, NMR Laboratory, Federal University of Goiás, Goiânia-GO, Brazil

*Corresponding author: cristiano_prado@ufg.br

I. INTRODUCTION

Meat from *Bos indicus* cattle has been associated with lower tenderness than *Bos taurus* cattle [1]. Wet and dry aging processes have been used to improve sensory attributes such as flavor and tenderness. During aging, enzymatic proteolysis promotes the release of free amino acids, which influence the flavor of meats, such as sweet taste (glycine, alanine, serine, threonine, proline, and hydroxyproline), sour taste (phenylalanine, tyrosine, and alanine), bitter taste (histidine, arginine, isoleucine, leucine, lysine, phenylalanine, tyrosine, valine), and umami (glutamic acid and aspartic acid). Differences in the amount and type of free amino acids, between breeds and the aging process, may be responsible for differences in sensory attributes. This research aims to evaluate beef from crossbred *Bos taurus* x *Bos indicus* animals and Nellore breed, aged via dry-aging and wet-aging, comparing changes in the profile of free amino acids.

II. MATERIALS AND METHODS

Ten samples were obtained from crossbred castrated male cattle, with at least 50% *Bos taurus* content, kept for at least 120 days in feedlots during finishing, and at 24 months of age. Additionally, eight samples were taken from intact male Nelore cattle raised in a semi-confined system, also aged 24 months. After slaughter and chilling (2°C, 48 hours), samples were collected from the m. *Longissimus thoracis et lumborum* between the 9th thoracic vertebra and the 3rd lumbar vertebra. Samples were separated for evaluation without aging, and the rest were aged for 28 days, divided into two treatments, wet-aging and dry-aging, in an aging chamber (10°C ± 1°C / ± 75% RH). ^{1}H NMR spectra were acquired on a Bruker Avance III 500 spectrometer at 11.75 T, using the BBI probe at 25°C, after the extraction process in 200 mg of meat with water and chloroform. An aliquot of the aqueous phase was transferred to the NMR tube and homogenized with 200 µL of D2O/DSS/Sodium Azide solution. The spectra were processed using Topspin 3.1 software. Chenomx NMR Suite 8.3 software and literature data (Human Metabolome Database (HMDB)) were used to assist in compound identification. Statistical analyses (ANOVA and Tukey HSD test) were performed using Statistica 7 software.

III. RESULTS AND DISCUSSION

The aging processes increased the content of most free amino acids (Figure 1). Dry-aged samples showed a more pronounced increase in amino acid content than wet-aged samples, regardless of animal genetics. When comparing crossbred with Nellore beef, it is noticeable that all amino acids presented more expressive increases in the crossbred beef, regardless of the aging type (dry-aged and wet-aged). This more expressive increase in free amino acid content may influence the more pronounced flavor of dry-aged beef. The most pronounced changes in concentrations of free amino acids during dry aging may be due to two factors: the action of microorganisms on the crust, which can intensify proteolysis, and concentration of compounds by moisture evaporation [2]. The loadings plot (Figure 2) shows that all amino acids are grouped in the same PC of crossbred dry-aged beef (positive PC2 and negative PC1), showing a higher correlation of these compounds with this group, which reinforces the possibility of these free amino acids influencing sensory attributes. Unaged crossbred and Nellore samples (positive PC1 and PC2) were also grouped, indicating similar composition characteristics. Nellore dry-aged and wet-aged samples were also grouped (positive PC1 and negative PC2). Finally, crossbred dry-aged and wet-aged beef are grouped only in negative PC1.

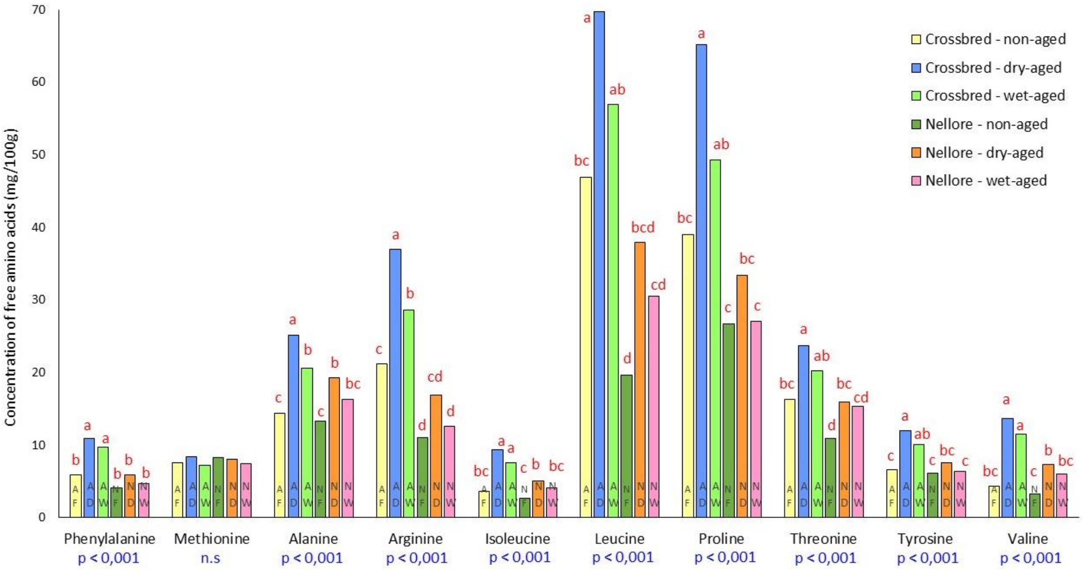

Figure 1. Concentration of free amino acids (mg/100g) in Crossbred and Nellore beef during dry- and wet-aging (28 days).

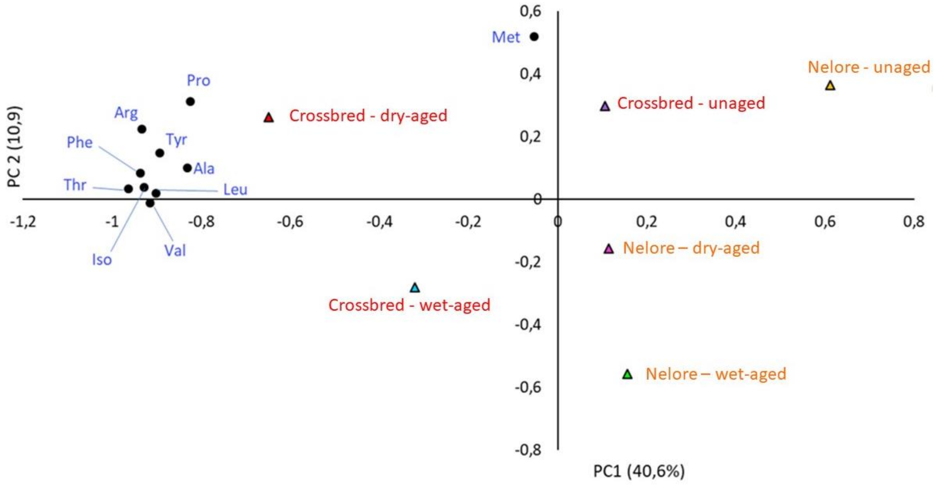

Figure 2. Plot of the variables having the highest discrimination power with significantly differences (P<0.05) among the groups, according to the principal components loading vectors.

IV. CONCLUSION

Through ^{1}H NMR analyses, it was possible to identify that beef from crossbred animals had more free amino acids than Nellore. Additionally, dry-aging resulted in a greater quantity of free amino acids compared to wet-aging.

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