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Metabolites correlated with dark, firm and dry occurrence in beef (#43)

Alessandra Rosa[1], Nara R. B. Consolo[1], Linda Samuelsson[4], Luiz Carlos G. S. Barbosa[1], Angel Higuera-Padilla[1], Luis Alberto Colnago[3], Evaldo A. L. Titto[1]

[1] University of São Paulo, College of Animal science and Food Engineering, Pirassununga, Brazil; [2] University of São Paulo, College of Veterinary Medicine, Pirassununga, Brazil; [3] EMBRAPA Instrumentation, São Carlos, Brazil; [4] AgResearch, Grasslands Research Centre, Grasslands, New Zealand

Introduction

Dark, firm and dry (DFD) meat is a worldwide problem relating to the quality of fresh meat. DFD meat is characterized by a dark red or purple color and high pH (above 6.0) (Newton & Gill, 1981), and it can influence the shelf-life, tenderness and consumer acceptance, whereas beef with abnormal appearance tends to be rejected (Tarrant, 1989). The occurrence of DFD meat is driven by the glycogen content in muscle immediately after slaughter and also by the muscle glycolytic capacity (Apaoblaza et al., 2015; Gerrard et al., 2015; Immonen & Puolanne, 2000). However, the nature and origin of DFD meat is not yet fully understood; therefore, the objective of this study was to compare the metabolites in DFD and normal meat to shed more light on the biochemical mechanisms involved in DFD.

Methods

A total of 75 crossbreed ½ Angus ½ Nellore young bulls were slaughtered and after 24 hours the carcass pH (pH 24h) was recorded. Carcasses were classified as DFD (pH > 6.0) or normal (pH < 5.8) (Newton & Gill, 1981). The longissimus lumborum (LL) muscle was sampled from five randomly selected carcasses classified as DFD and five classified as normal 24 h post-mortem and the meat was vacuum packed and aged for 14 days at 1.5 . A total of 0.5 g of meat was used for metabolomics analysis and the samples were prepared as previously described [if supportFields]><spanlang=EN-US style='font-size:12.0pt;line-height:107%;font-family:"Times New Roman",serif;mso-fareast-font-family:"Times New Roman";color:black;mso-ansi-language:EN-US;mso-fareast-language:PT-BR;mso-bidi-language:AR-SA'>ADDIN CSL_CITATION {"citationItems":[{"id":"ITEM-1","itemData":{"DOI":"10.1038/nprot.2007.376","author":[{"dropping-particle":"","family":"Beckonert","given":"Olaf","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Keun","given":"HectorC","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Ebbels","given":"TimothyMD","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Bundy","given":"Jacob","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Holmes","given":"Elaine","non-dropping-particle":"","parse-names":false,"suffix":""},{"dropping-particle":"","family":"Lindon","given":"JohnC","non-dropping-particle":"","parse-names":false,"suffix":""}],"container-title":"Nature Protocols","id":"ITEM-1","issued":{"date-parts":[["2007"]]},"page":"2692-2703","title":"Metabolicprofiling , metabolomic and metabonomic procedures for NMR spectroscopy ofurine , plasma , serum and tissueextracts","type":"article-journal","volume":"2"},"uris":["http://www.mendeley.com/documents/?uuid=47935e35-e7f3-47a4-a341-319e1abbe5e3"]}],"mendeley":{"formattedCitation":"(Beckonertet al., 2007)","plainTextFormattedCitation":"(Beckonert etal., 2007)","previouslyFormattedCitation":"(Beckonert etal.,2007)"},"properties":{"noteIndex":0},"schema":"https://github.com/citation-style-language/schema/raw/master/csl-citation.json"}<spanstyle='mso-element:field-separator'><![endif](Beckonert et al., 2007) [if supportFields]><spanlang=EN-US style='font-size:12.0pt;line-height:107%;font-family:"Times New Roman",serif;mso-fareast-font-family:"Times New Roman";color:black;mso-ansi-language:EN-US;mso-fareast-language:PT-BR;mso-bidi-language:AR-SA'><![endif]. One dimensional proton nuclear magnetic resonance (1D ^{1}H-NMR) was used for metabolite profiling. ^{1}H NMR spectra were acquired at 300 K on a Bruker Avance 14.1 T spectrometer (Bruker Corporation, Karlsruhe, Baden-Württemberg, Germany) at 600.13 MHz for ^{1}H, using a BBO 5 mm probe. D_2O was used as a lock solvent and DSS as internal reference for ^{1}H and an internal standard for metabolite quantitation. 1D ^{1}H NMR spectra were processed and metabolites quantified using the Chenomx NMR Suite Professional 7.7 software (Chenomx Inc., Edmonton, Canada). The quantified data of metabolites were imported to MetaboAnalyst 3.0 for principal component analysis (PCA), and volcano plot analysis which combined fold-change analysis and t-tests.

Results

Forty-five metabolites were quantified in meat by ^{1}H NMR including amino acids, small peptides, and metabolites correlated with postmortem glycolysis, Krebs cycle, purine metabolism, fatty acid metabolism, muscle metabolism, and nitrogen compounds. The majority of metabolites did not differ significantly (P> 0.05) in abundance between DFD and normal meat. The PCA scores plot (Fig. 1) showed that the PC1 and PC2 accounted for 45.6% and 18% of the variance, respectively. This result shows clear separation of DFD and normal meat, suggesting differences in metabolites abundances between the two groups. According to the Volcano plot (Fig. 2), glucose-6-phosphate (G6P), creatine phosphate, glucose, threonine and glucose-1-phosphate (G1P) were 2-fold more abundant in the normal meat

Notes

whereas ATP was 2-fold more abundant in DFD meat. As previously described by others, the over-representation of carbohydrates and creatine phosphate (CP) in normal meat could be an indicator of increased energy metabolism, and greater glycolytic potential for normal meat compared to DFD meat (Apaoblaza et al., 2015; Gerrard et al., 2015). In conclusion, energy metabolic pathways including glycolysis and amino acid degradation were affected in DFD meat.

Conclusion

In conclusion, energy metabolic pathways including glycolysis and amino acid degradation were affected in DFD meat.

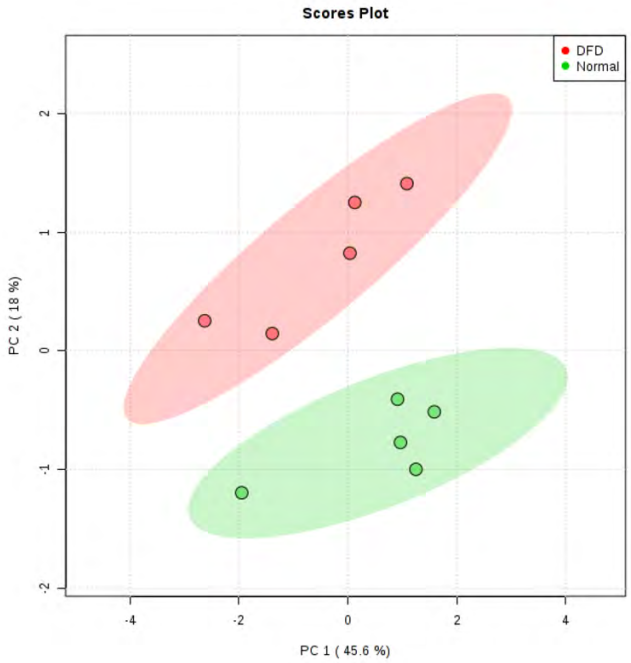

Figure 1. Principal Component Analysis (PCA)
Principal Component Analysis (PCA) of concentrations of 45 metabolites in meat classified as either DFD (pH > 6.0) or Normal (pH < 5.8).

Notes

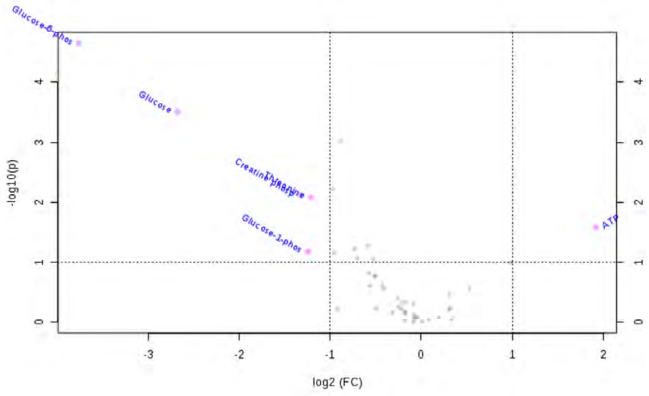

Figure 2. Volcano plot
Volcano plot comparing the metabolite differences between DFD and Normal meat. The coloured points represent metabolites which were 2-fold more concentrated in one group compared to the other (log 2 (FC) -1, P < 0.05). Metabolites in the left hand part of the plot were overabundant in normal meat, while metabolites on the right-hand side were overabundant in DFD meat.

Notes