P-11-06

Meat metabolites profile changed by ageing time (#84)

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Introduction

Metabolomics can be an important tool to predict meat quality, since metabolites contribute to various metabolic pathways including glycolysis, protein degradation and gustatory sensations (Kodani et al., 2017). The conversion of muscle to meat throughout the ageing process can affect meat quality due to structural breakdown of muscle by endogenous proteases. This can substantially improve meat palatability attributes such as tenderness, flavour, and/or juiciness (Ma et al., 2017). The aim of this study was to evaluate the metabolites of muscle and meat at different aging times using 1D ^{1}H NMR.

Methods

A total of 50 Nellore bulls were slaughtered and *Longissimus lumborum* (LL) muscle was immediately sampled from 5 randomly selected animals. After 24 h, the LL from the same five animals were vacuum packed and aged for 21 days. Meat samples were collected after 1, 7, 14 and 21 days of ageing for metabolites investigation by ^{1}HNMR spectroscopy. A total of 0.5 g of meat or muscle was used and prepared according to Beckonert et al. (2007). 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. 1D ^{1}H NMR spectra were processed and metabolites quantified using the Chenomx NMR Suite Professional 7.7 software (Chenomx Inc., Edmonton, Canada). Principal component analysis was performed using the web-based tool MetaboAnalyst 2.0. The R software (REDENCE) was used to analyse data with a Bayesian Clustered Linear Regression model.

Results

Thirty-one metabolites were identified in muscle and 45 metabolites in the aged LL. The Principal Component scores plot (PC1 73% and PC2 9.9%) of the metabolite concentrations (Figure 1) shows clear clustering of timepoints, suggesting differences occurring over time. A Bayesian Clustered Linear Regression model was applied for this data to explain the variations in metabolite concentrations over time (Figure 2). The vertical bars (Figure 2) represent the time-course behaviour of each metabolite: Time-course curve type A: linear; Time-course curve type B: quadratic with plateau at 7 days; Time-course curve type C: quadratic with plateau at 14 days. Approximately a third of the metabolites (including the majority of amino acids, acetate, hypoxanthine, creatinine, glutathione and choline) increased linearly from day 0 to day 21 (time-course curve type A). It can be explained by the myofibrillar break down during the ageing process, releasing peptides and free amino acids into the meat (Koohmaraie & Gessink, 2006). Several of the metabolites with time-course curve type A found in greater abundance in the aged beef are associated with gustatory sensations: alanine, glutamine and glycine are correlated with sweetness; isoleucine, leucine, phenylalanine, tyrosine and valine with bitterness; and glutamate with umami flavour of meat (Nishimura & Kato, 1988). Glucose, glucose-6-phosphate, ATP, IMP and *O*-acetylcarnitine also increased from day 0, but reached a plateau at day 7 (time-course curve type B). These metabolites are involved in glycolysis and purine metabolism, both of which are highly active immediately following slaughter but decrease in activity as substrates are used up and pH decreases (Muroya et al., 2014). Additionally, IMP and hypoxanthine are important in meat flavour perception, as they hold umami taste characteristics (Muroya et al., 2014). A third of the metabolites (including glutamine, taurine, glucose-1-phosphate, inosine, uridine, glycerate, glycerol, fumarate, malonate, carnitine, lactate, betaine, creatine, pyruvate, niacinamide, carnosine, succinate and urea) increased in concentration from day 0 reaching a plateau at day 14 (time-course curve type C). These metabolites are part of distinctive pathways in meat, such as glycolysis, purine metabolism, Krebs cycle, fatty acids metabolism, protein degradation, and nitrogen metabolism. Some of them are correlated to meat taste and flavour, either directly or indirectly by being substrates in chemical reactions forming flavourful compounds upon cooking: carnosine and inosine are associated with umami flavour; lactate and succinate with sweet flavour (Kim, Kemp & Samuelsson, 2016).

Conclusion

In conclusion, the concentrations of all metabolites increased in the conversion of muscle to meat as well as during ageing. Many of the released metabolites are related to flavour and thus contribute to improving meat flavour with ageing.

References

Beckonert, O., Keun, H. C., Ebbels, T. M. D., Bundy, J., Holmes, E., & Lindon, J. C. (2007). Metabolic profiling , metabolomic and metabonomic procedures

Notes

for NMR spectroscopy of urine , plasma , serum and tissue extracts. *Nature Protocols*, *2*, 2692–2703. https://doi.org/10.1038/nprot.2007.376
Kim, Y. H. B., Kemp, R., & Samuelsson, L. M. (2016). Effects of dry-aging on meat quality attributes and metabolite profiles of beef loins. *Meat Science*, *111*, 168–176. https://doi.org/10.1016/j.meatsci.2015.09.008
Kodani, Y., Miyakawa, T., Komatsu, T., & Tanokura, M. (2017). NMR-based metabolomics for simultaneously evaluating multiple determinants of primary beef quality in Japanese Black cattle. *Scientific Reports*, *7*(1), 1–13. https://doi.org/10.1038/s41598-017-01272-8
Koohmaraie, M., & Geesink, G. H. (2006). Contribution of postmortem muscle biochemistry to the delivery of consistent meat quality with particular focus on the calpain system. *Meat Science*, *74*(1), 34–43. https://doi.org/10.1016/j.meatsci.2006.04.025
Ma, D., Kim, Y. H. B., Cooper, B., Oh, J. H., Chun, H., Choe, J. H., ... Min, B. (2017). Metabolomics Profiling to Determine the Effect of Postmortem Aging on Color and Lipid Oxidative Stabilities of Different Bovine Muscles. *Journal of Agricultural and Food Chemistry*, *65*(31), 6708–6716. https://doi.org/10.1021/acs.jafc.7b02175
Muroya, S., Oe, M., Nakajima, I., Ojima, K., & Chikuni, K. (2014). CE-TOF MS-based metabolomic profiling revealed characteristic metabolic pathways in postmortem porcine fast and slow type muscles. *Meat Science*, *98*(4), 726–735. https://doi.org/10.1016/j.meatsci.2014.07.018
Nishimura, T., & Kato, H. (1988). Taste of free amino acids and peptides. *Food Reviews International*, *4*(2), 175–194. https://doi.org/10.1080/87559128809540828

Notes

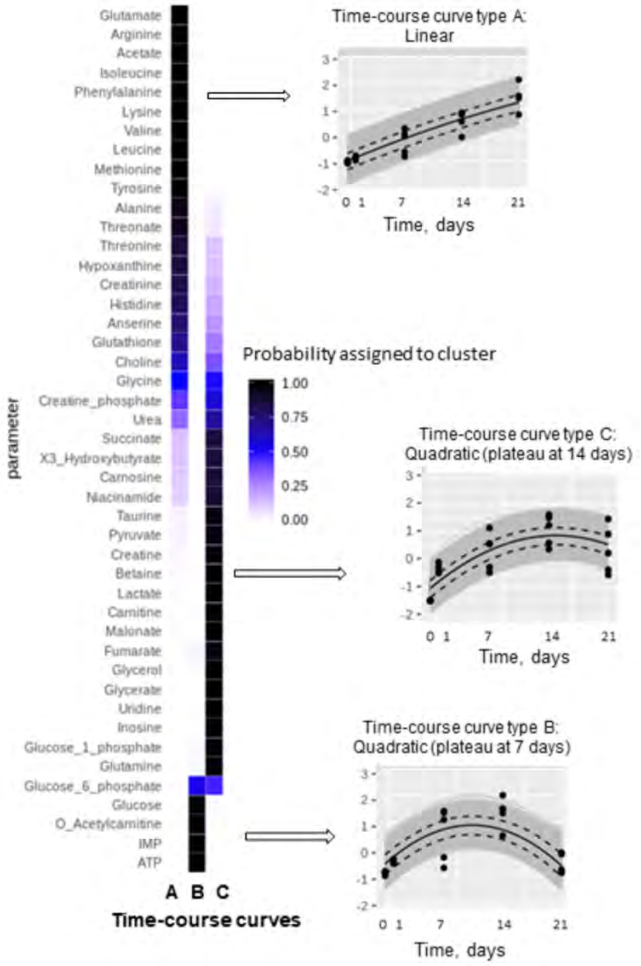

Figure 2
The Bayesian Clustered Linear Regression model applied to the metabolites in muscle and meat samples. Black colour means that a particular metabolite is strongly associated with a particular time-course behaviour. Purple colour means that the time-course behaviour of a certain metabolite can be represented by two time-course behaviours. White colour means that a particular metabolite does not display a certain time-course behaviour. The graphs show the three types of predicted time-course curves for metabolites involved in meat ageing.

Notes

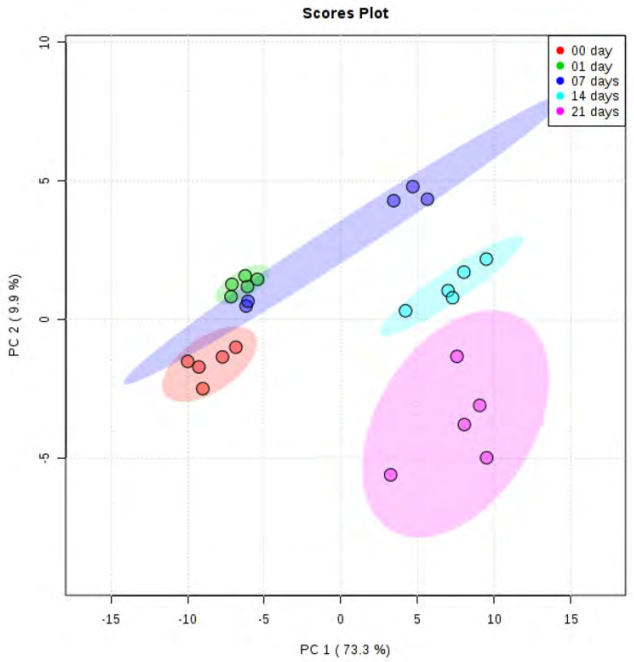

Figure 1
PCA scores plot of metabolite profiles of muscle and meat at different time points.

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