UPLC-MS/MS-based metabolomics reveals spoilage metabolic dynamics in modified atmosphere/airpackaged chilled and super- chilled fresh pork loins

Anthony Bassey [1], Zongshuai Zhu [1], Abdulmumini Amin [2], Cong Zhou [1], Okon Johnson Esua [3], Keping Ye [1], Changbo Tang [1], Chunbao Li [1], Guanghong Zhou [1]

[1] National Center of Meat Quality and Safety Control, Synergistic Innovation Center of Food Safety and, Nutrition, College of Food Science and Technology, Nanjing Agricultural University, 210095, Nanjing, Jiangsu, PR China, [2] Centre for Ruminant Nutrition and Feed Engineering Research, College of Animal Science and, Technology, Nanjing Agricultural University, Nanjing, 210095, China, [3] School of Food Science and Engineering, South China University of Technology, Guangzhou 510641, China

OBjective: Bacterial mechanism is complex, and undetermined adaptation strategies are difficult to unravel, especially with conventional targeted methods. Nonetheless, novel omics tools consisting of full genome, proteome, transcriptome, or metabolome analysis enable the presentation of a non-targeted overall overview of bacterial metabolism (Bassey et al., 2021c). Although transcriptomic (Wang et al., 2018) and proteomic (Kolbeck et al., 2020) techniques have shown remarkable outcomes in uncovering the metabolism, adaptation, and interactive mechanism of predominant meat spoilers, there is limited information in the application of metabolomics to assess the metabolic profiles associated with spoilage. Hence, the objective of this study.

Materials and Methods: To elucidate spoilage-related metabolic characteristics in air packaged (AP) and modified atmosphere packaged (MAP) chilled (4 °C) and super-chilled (-2 °C) fresh pork loins, the samples were subjected to UPLC-MS/MS metabolomics analysis on 0, 7, 14 d and 0, 14, 28 d, respectively, synonymous to the period of significant bacterial growth reported previously (Bassey et al., 2021a,b).

Results and Discussion: From the overall metabolites (2,061) identified in the samples, 599 (AP) and 524 (MAP) were identified at chilled storage, while 506 (AP) and 472 (MAP) were detected at super-chilling. The metabolites were mostly related to lipids and proteins, suggesting the significance of both nutrients in meat quality. Notwithstanding the effect of temperature and gas composition in inhibiting lipid- protein oxidation during storage, hypoxanthine, guanine, xanthine, urocanate, histidine, and glutamate dominated the AP groups, while guanosine, xanthosine, inosine, and glutamate were prevalent in MAP samples, indicating their roles in spoilage processes. These metabolites were mainly linked to purine, histidine, arginine biosynthesis, ABC transporters, glycerophospholipids, and alanine, aspartate, and glutamate metabolic pathways. Satisfactory results with precision (RSD < 8.1%), linearity ($R2 > 0.96$), and recovery (87 - 104%) demonstrated the tool's efficacy in elucidating spoilage-related differential metabolites in samples. These pathways have been demonstrated to induce spoilage in refrigerated pork (Li et al., 2019), vacuum-packed chilled beef (Frank et al., 2020), MAP meat (Kolbeck et al., 2020), chilled chicken meat (Wen et al., 2020; Zhang et al., 2021) , goat meat (Jia et al., 2021), and dry-cured ham (Liao et al., 2022).

Conclusion: These findings provide a comprehensive insight into the differential metabolites and metabolic pathways underlying the mechanisms of chilled and super-chilled AP and MAP pork spoilage.

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