71st ICoMST
GIRONA - CATALONIA - SPAIN
3rd-8th August - 2025

POST-MORTEM MUSCLE PROTEOMICS REVEALS GENETICS AND ENVIRONMENTAL ENRICHMENT EFFECTS – IMPLICATIONS FOR MEAT AUTHENTICATION

Seren Yigitturk[1,*], Ingrid C. de Jong[2], Sjef Boeren[3], Shai Barbut[4,5],Vincenzo Fogliano[1], Sara W. Erasmus[1,*]

[1]Food Quality and Design, Wageningen University & Research, P.O. Box 17, 6700 AA Wageningen, The Netherlands

[2]Wageningen Livestock Research, Wageningen University & Research, 6700 AH Wageningen, The Netherlands

[3]Laboratory of Biochemistry, Wageningen University & Research, Wageningen, The Netherlands

[4]Adaptation Physiology Group, Department of Animal Sciences, Wageningen University & Research, Wageningen, The Netherlands

[5]Department of Food Science, University of Guelph, Guelph N1G 2W1, Ontario, Canada

*Corresponding authors. E-mails: S. Yigitturk (seren.yigitturk@wur.nl); S.W. Erasmus (sara.erasmus@wur.nl)

I. INTRODUCTION

Poultry meat is a major contributor to global protein intake, and projected to account for 43% of meat-derived protein by 2033[1]. Its popularity is driven by its nutritional profile, affordability, efficiency, and lower environmental impact compared to other livestock [1,2]. To meet this demand at an efficient pace, intensive selection has produced faster-growing (FG) broiler hybrids that reach market weight within 28–42 days [3]. However, this accelerated growth raises concerns about meat quality [4], alongside increasing pressure on the meat industry to adopt more sustainable, welfare-friendly husbandry practices. In response, production systems are adapting by incorporating “extensification factors” [5] such as slower-growing (SG) genetics, reduced stocking densities, and environmental enrichments. These extensification factors add value to the final product but also increase production costs, making such meat more vulnerable to food fraud. To maintain consumer trust in welfare and quality claims, it is crucial to deepen our understanding of intrinsic meat quality at a molecular level and how husbandry practices influence it. Such insights enable the identification of biomarkers for product authentication and the development of fit-for-purpose meat authentication tools [6].

Proteomics offers a valuable approach for characterising molecular adaptations [7]. Although still evolving, proteomic research is more advanced in red meat species than in poultry [8]. Only a limited number of studies have investigated the chicken proteome under different farming systems (organic vs. antibiotic-free) to discover authenticity biomarkers [9-11]. However, there is a paucity of research that considers the individual animal, together with its production data within a large controlled experimental setup, with the aim to study the effect of extensification factors.

This study, conducted within the mEATquality EU project, investigates the effects and interactions of genetics (breed type) and environmental enrichment under controlled, higher-welfare, non-organic conditions. It explores their impact on broiler performance, meat quality, and associated proteomic profiles to support meat authentication strategies.

II. MATERIALS AND METHODS

A total of 3,680 of two different broiler breeds (Hubbard S757N, SG vs. Hubbard JA787, FG) were raised in a higher welfare system enriched with and without lucerne in a net, perch and dust bath with peat in medium-space availability (30 kg/m^2). A total of 160 broilers (2 breeds* 2 enrichment, n=5 pens/treatment, n=8 broilers/pen) were randomly selected for meat quality analysis. Yield characteristics, meat colour (IRIS-Alphasoft electronic eye) and texture (blunt Meullenet-Owens razor shear/ BMORS method) [12] of broilers were measured as related to intrinsic meat quality. Twenty right *pectoralis major* muscles (1 representative broiler/pen, n=5 broilers/treatment) were selected for comparative proteome analysis of which the whole muscle lysate was processed with Protein Aggregation Capture on Microparticles [13] using trypsin. Peptides were measured using nano LC-MS/MS and quantified by label-free quantification method.

III. RESULTS AND DISCUSSION

Proteomics analyses revealed that SG broilers exhibited higher abundance of proteins related to detoxification and oxidative stress management (GPX3, LOC100859645, GSTA3, AKR1A1), actin filament binding and actin filament-based processes (FLNA, MYLK2, MYH11, TPM1, TAGLN, SH3KBP1, ANXA6). This molecular profile corresponded with a 28.7% increase in the shear force ($p < 0.001$) of the cooked chicken meat, indicating a denser, structurally stronger muscle. Additionally, FG broilers adapted to environmental enrichment by upregulating proteasome pathway proteins, such as PSMA4 and PSMD4, suggesting enhanced protein degradation. Phenotypically, on-farm enriched FG birds showed a significant reduction in breast muscle yield ($p = 0.03$), likely reflecting increased

proteolysis, alongside higher b* values in chicken meat colour ($p < 0.01$), while texture parameters remained unchanged. The results indicate that both genetics (breed) as well as environmental enrichment affected the differentially expressed proteins. This highlights the importance of investigating the impact of extensification factors when identifying biomarkers for chicken authentication and developing robust meat authentication strategies.

CONCLUSION

This study provides a comprehensive overview of how environmental enrichments influence protein regulation in SG and FG broilers. FG broilers exhibited a proteome optimised for rapid muscle accretion and softer texture. However, when exposed to enriched conditions, this growth-oriented proteome shifted toward increased proteasome activity, which may explain the observed reduction in breast muscle yield and changes in meat colour. Importantly, both genetics and husbandry practices influence proteomic profiles and authenticity biomarkers, highlighting the need to account for these factors when developing meat authentication tools. This molecular-level understanding supports the development of authentication strategies to ensure the integrity of the meat supply chain. Further research is needed to validate these molecular indicators and explore how other extensification factors influence broader dimensions of meat quality and authenticity.

ACKNOWLEDGEMENTS

This research was part of the mEATquality project (https://meatquality.eu/)- “Linking extensive husbandry practices to the intrinsic quality of pork and broiler meat”, funded by the European Union's Horizon 2020 Research and Innovation Programme (Grant Agreement No. 101000344). We thank Hubbard Breeders for supplying the chickens.

REFERENCES

1 OECD/FAO (2024), OECD-FAO Agricultural Outlook 2024-2033, OECD Publishing, Paris/FAO, Rome, https://doi.org/10.1787/4c5d2cfb-en.

2 Marangoni, F. *et al.* Role of poultry meat in a balanced diet aimed at maintaining health and wellbeing: an Italian consensus document. *Food Nutr Res* **59**, 27606 (2015). https://doi.org:10.3402/fnr.v59.27606

3 Santos, M. N. *et al.* In pursuit of a better broiler: carcass traits and muscle myopathies in conventional and slower-growing strains of broiler chickens. *Poult Sci* **100**, 101309 (2021). https://doi.org:10.1016/j.psj.2021.101309

4 Barbut, S. *et al.* Review: Myopathies in broilers: supply chain approach to provide solutions to challenges related to raising fast growing birds. *Poultry Science* **103**, 103801 (2024). https://doi.org:https://doi.org/10.1016/j.psj.2024.103801

5 Marchewka, J. *et al.* Linking key husbandry factors to the intrinsic quality of broiler meat. *Poultry Science* **102**, 102384 (2023). https://doi.org:https://doi.org/10.1016/j.psj.2022.102384

6 Erasmus, S. W., Sohaib, M., Revilla, I., Vivar-Quintana, A. M. & Giancoli, S. J. Markers for meat provenance and authenticity with an account of its defining factors and quality characteristics - a review. *J Sci Food Agric* **104**, 7027-7084 (2024). https://doi.org:10.1002/jsfa.13492

7 Kiyimba, F., Gagaoua, M., Suman, S. P., Mafi, G. G. & Ramanathan, R. in *Encyclopedia of Meat Sciences (Third Edition)* (ed Michael Dikeman) 528-539 (Elsevier, 2024).

8 Huang, C. *et al.* Proteomics discovery of protein biomarkers linked to meat quality traits in post-mortem muscles: Current trends and future prospects: A review. *Trends in Food Science & Technology* **105**, 416-432 (2020). https://doi.org:https://doi.org/10.1016/j.tifs.2020.09.030

9 Alessandroni, L., Sagratini, G. & Gagaoua, M. Proteomics and bioinformatics analyses based on two-dimensional electrophoresis and LC-MS/MS for the primary characterization of protein changes in chicken breast meat from divergent farming systems: Organic versus antibiotic-free. *Food Chemistry: Molecular Sciences* **8**, 100194 (2024). https://doi.org:https://doi.org/10.1016/j.fochms.2024.100194

10 Alessandroni, L., Sagratini, G., Bravo, S. B. & Gagaoua, M. Data-independent acquisition-based SWATH-MS proteomics profiling to decipher the impact of farming system and chicken strain and discovery of biomarkers of authenticity in organic versus antibiotic-free chicken meat. *Current Research in Food Science* **8**, 100757 (2024). https://doi.org:https://doi.org/10.1016/j.crfs.2024.100757

11 Alessandroni, L., Sagratini, G. & Gagaoua, M. Integrated Chemometrics and Data-Independent Acquisition Proteomics for the Discovery of Meat Authenticity Biomarkers: A Study on Early Post-Mortem Pectoralis major Muscle Proteomes of Ross 308 and Ranger Classic Chicken Produced by Organic versus Antibiotic-Free Farming Systems. *Journal of Agricultural and Food Chemistry* **72**, 20153-20170 (2024). https://doi.org:10.1021/acs.jafc.4c03651

12 LEE, Y. S., OWENS, C. M. & MEULLENET, J. F. THE MEULLENET-OWENS RAZOR SHEAR (MORS) FOR PREDICTING POULTRY MEAT TENDERNESS: ITS APPLICATIONS AND OPTIMIZATION. *Journal of Texture Studies* **39**, 655-672 (2008). https://doi.org:https://doi.org/10.1111/j.1745-4603.2008.00165.x

13 Batth, T. S. *et al.* Protein Aggregation Capture on Microparticles Enables Multipurpose Proteomics Sample Preparation. *Mol Cell Proteomics* **18**, 1027-1035 (2019). https://doi.org:10.1074/mcp.TIR118.001270