Metabolomic fingerprinting of *in vitro* human and *in vivo* pig colonic digests unravels red meat and white meat associated biomarkers

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1. Introduction

Recently, the International Agency for Cancer (IARC) has assigned red and processed meat consumption as probably carcinogenic and carcinogenic for humans (1). However, mechanisms fully explaining the adverse effects of red meat intake are still lacking. Moreover, most epidemiological studies rely on food frequency questionnaires that are prone to participant bias and hence there is an increasing need for more objective and reliable nutritional biomarkers, especially those discriminating between red and white meat intake.

2. Approach

In this study, an untargeted Ultra High Performance Liquid Chromatography coupled to hybrid High Resolution Mass Spectrometry (UHPLC-HRMS/MS) based metabolomics approach (2) was implemented to retrieve metabolic fingerprints discriminating between red and white meat consumption.

To this extent, the gastro-intestinal colonic digestion of meat was simulated with the use of the fecal inocula of 10 human volunteers and next to this, 8 pigs received red and white meat diets for three weeks. Finally, *in vitro* colonic digests and colonic content were analysed, respectively, and multivariate statistics was applied to obtain red meat and white meat associated biomarkers.

3. Results and discussion

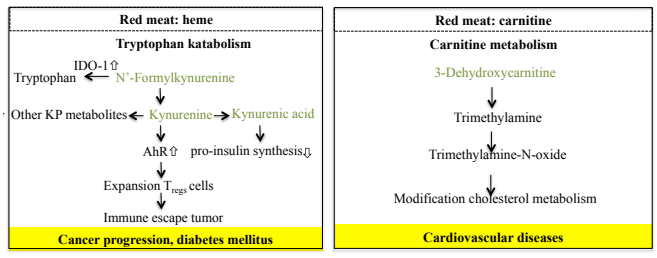

Figure 1. Red meat associated disease pathways

In total, 11 chicken and 6 beef meat associated biomarkers were retained with VIP value > 1.0 and P-value < 0.05 in both pig and human colonic digests. Both biomarker panels reached 100% specificity, and sensitivity was 77% and 87%, respectively.

Also, some promising red meat associated metabolites possibly involved in diseases were identified, including the tryptophan catabolites N-formylkynurenine, L-kynurenine and kynurenic acid and 3-dehydroxycarnitine (Figure 1). In addition, *in vitro* digestions with the red meat constituents heme and carnitine showed their involvement in the formation of these metabolites. The tryptophan catabolites are linked with immune escape of tumors and diabetes mellitus and trimethylamine-N-oxide (metabolite of 3-dehydroxycarnitine) has been linked with the development of colorectal cancer and cardiovascular diseases (Figure 1) (3,4).

4. Conclusion

The used approach in this work enabled to obtain promising biomarker panels for red and white meat intake. Nevertheless, further validation by performing *in vivo* human feeding trials and assessing their potential role as objective measures for meat intake in epidemiological studies is still required.

The discovered red meat associated disease pathways could be further explored and new prevention strategies regarding meat processing could be established to minimize the risk for the development of red meat associated diseases.

References

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