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Evaluation of antimicrobial resistance and virulence genes in commensal *Escherichia coli* isolated from different sources.

INTRODUCTION

Antimicrobial resistance has been recognised as one of the world's most pressing and ever-growing public health problems, affecting both human and veterinary medicine. In the last decades we have witnessed a dramatic spread and diffusion of resistant bacterial pathogens, showing in many cases insensibility to different antimicrobial classes and high virulence. Horizontal gene transfer, mediated by mobile genetic elements, plays an essential role in the evolution of bacteria. Indeed, bacteria may acquire a mobile genetic element carrying multiple antimicrobial-resistance genes and/or virulence-associated genes in a single horizontal gene transfer event. This phenomenon enables the diffusion and the acquisition of antimicrobial resistance and virulence genes between different microbial population and different environments, representing a risk for public health.

AIM OF THE STUDY

The aim of the study is the evaluation of virulence and antimicrobial resistance genes carried by different mobile elements in commensal *Escherichia coli*, typically chosen as a representative indicator in Gram-negative bacteria. The data, obtained from different «environments» (human, food, food producing, companion and wild animals), are essential for a better understanding of the epidemiology traits involved in virulence and antimicrobial resistance diffusion.

MATHERIALS & METHODS

A total of 300 commensal *Escherichia coli* were collected from 12 different sources (beef, dairy, swine, boar, poultry, fish, companion animal, rabbit, vegetable, mollusc, human, wild animal) during the period November 2016 - January 2018. Genomic DNA was extracted using a commercial kit (DNeasy Blood and Tissue Kit, Qiagen) following manufacturer instruction. *Escherichia coli* identification was performed through a multiplex PCR targeting 4 different genes (cytochrome bd complex, lactose permease, b-d-glucuronidase, and b-d-galactosidase) following Horakova *et al.* (2008) suggestions. Library preparation was performed using the Nextera Flex library preparation kit (Illumina, USA) and sequencing was executed with Illumina NextSeq 500. All gene screening was performed using ARIBA (Hunt *et al*., 2017) and publicly available reference.

RESULTS & DISCUSSION

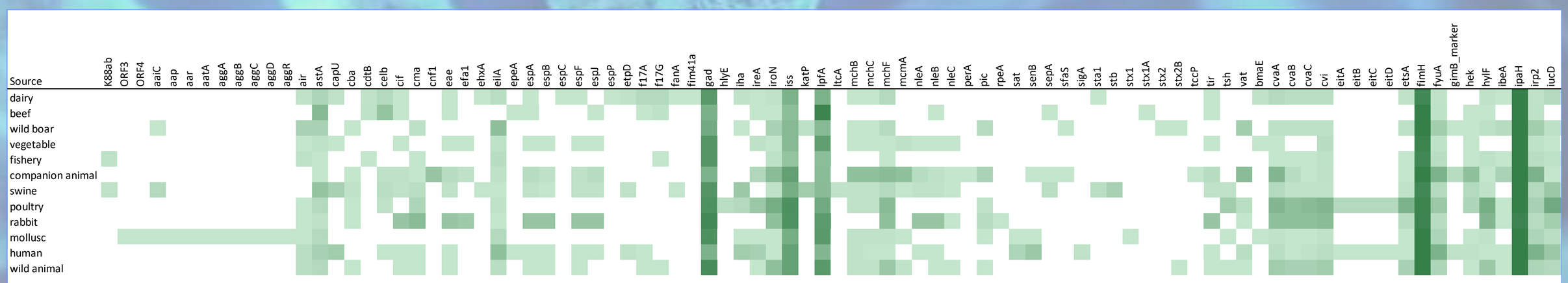

Fig.1a : Virulence genes associated with the 12 investigated sources.

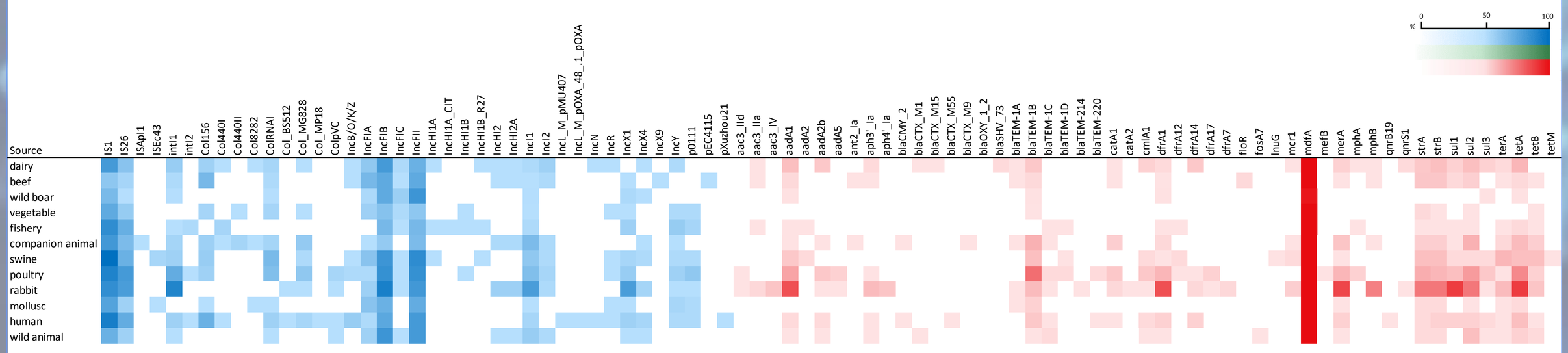

Fig.1b : Mobile elements (in blue) and resistance genes (in red) associated with the 12 investigated sources.

CONCLUSION & FUTURE PROSPECTS

Concerning, commensal *Escherichia coli* showed many antimicrobial resistance and virulence genes, some of them usually associated with important pathogens, carried by different mobile elements. The data suggest that the investigated environments could represent a reservoir of antimicrobial resistance and virulence genes, and also a possible threat for human health, and underline the importance of a constant monitoring, representing the key for a better understanding of virulence and antimicrobial resistance spatial and temporal trends. In the future we will focus on integron structures and related gene cassettes and we will perform long read sequencing of the strains showing the most interesting resistance pattern.

References:
Horakova K, Mlejnkova H, Mlejnek P, 2008. Specific detection of Escherichia coli isolated from water samples using polymerase chain reaction targeting four genes: cytochrome bd complex, lactose permease, β-d-glucuronidase, and β-d-galactosidase.
Hunt M, Mather AE, Sánchez-Busó L, Page A.J, Parkhill J, Keane JA, Harris SR (2017). ARIBA: rapid antimicrobial resistance genotyping directly from sequencing reads. Microbial genomics, 3(10).