

Monitoring of microbial ecology and *Salmonella* resistance in a pig slaughterhouse

Alain Le Roux¹, Carole Feurer¹, Paméla Houé², Patricia Le Grandois², Edouard Hirchaud³, Christophe Soumet², Arnaud Bridier²

¹ Department of Fresh and Processed Meat, IFIP, Maisons-Alfort, Le Rheu, France

² Antibiotics, Biocides, Residues and Resistance Unit, ANSES Fougères laboratory, Fougères, France

³ Viral Genetic and Biosecurity Unit, BP53, ANSES Ploufragan-Plouzané-Niort Laboratory, Ploufragan, France.

Introduction

Salmonella was responsible for 22,5% of reported outbreaks in EU in 2020. To guarantee food safety, a better understanding of microbial ecology and adaptation strategies on the food production chain constitutes a prerequisite. In a *One Health* perspective, data on *Salmonella* antibiotic and biocide resistance in food environments are also crucial to decipher transmission routes of resistant foodborne pathogens as well as resistance genetic determinants involved, and the role of process and selection pressures underwent in food industries (as cleaning and disinfection) in bacterial adaptation and antimicrobial resistance emergence.

Methods

Occurrence of *Salmonella* was investigated at seven different areas along a slaughter chain and through three sampling campaign in 2019. *Salmonella* strains were characterized using serotyping and pulsotyping to trace persistent strains and compare *Salmonella* ecology to data collected in 2017 at the same sampling areas (BRAVO project, EcoAntibio1). Minimal inhibiting concentrations (MIC) were also determined for various relevant antibiotics and for biocides used in the slaughterhouse. Associated indigenous bacterial communities were also characterized using 16S rRNA amplicon sequencing to assess microbial ecology evolution over time. Shotgun metagenomics was also performed to compare abundance of genes involved in antimicrobial resistance between 2017 and 2019.

Results

Salmonella was detected after enrichment for 5 among the 7 sampling areas investigated. Similarly to BRAVO projet, five serotypes were identified: S.4,5,12:i:-, Rissen, Typhimurium, Infantis and Derby with a large dominance of the monophasic variant of *S. Typhimurium*. Levels of antimicrobial resistance were also similar to those observed in 2017 during BRAVO project showing no evolution of *Salmonella* resistance during this period. Bacterial diversity analyses showed that populations in the slaughterhouse were recurrently highly dominated by γ -proteobacteria and especially by the Moraxellaceae family (genus *Psychrobacter*, *Moraxella* and *Acinetobacter*) and genus *Pseudomonas* with slight variation depending on areas. Population compositions were stable in time at a given sampling area as revealed by the comparison between samples collected in 2017 and 2019. Shotgun metagenomics analyses are still in progress to analyze functional potential evolution (especially antibiotic and biocide resistance genes and their association) between 2017 and 2019 for two sampling areas.

Conclusions

This study participates to the construction of a comprehensive view of *Salmonella* ecology in food environments integrating associated resident microbial flora and the distribution of antimicrobial resistance in relation to processing conditions. Data about functional genetic potential and its modulation should give original information about the impact of industrial process (including cleaning and disinfection) on microbial communities in the slaughterhouse over time.