

Genetic landscape of *Streptococcus uberis* isolated from bovine mastitis; typing and antibiotic resistance in-depth analysis

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Bovine mastitis remains the leading cause of economic loss at the dairy herd level. *Streptococcus uberis* is one of the most important mastitis pathogens and has shown increased resistance to antibiotics. Though clinically relevant, *Strep. uberis* is still poorly characterized in terms of subtypes and their genotypic and phenotypic antimicrobial resistance (AMR) profiles.

The aim of our study was to characterized genotypically and phenotypically 109 *Strep. uberis* mastitis strains.

All strains were subjected to whole genome sequencing and tested for phenotypic antimicrobial resistance using the minimum inhibitory concentration (MIC) assay. The WGS data were used for subtyping the strains by MLST, evaluating their genotypic AMR profile and presence of mobile genetic elements using bioinformatics tools. RAPD-PCR (Random Amplified Polymorphic DNA-PCR) was used as an additional method for genotyping 43 strains for which supplementary clinical data had been collected.

Our results showed high level of variability in the genotypes identified by both MLST and RAPD-PCR methods. Regarding antibiotic resistance, seventy strains (64%) were susceptible to all the 26 antibiotics tested; thirty-nine strains were resistant to cephalosporins (2%), lincosamides (12%), macrolides (12%), and tetracycline (33%). Eleven strains (10%) were resistant to three different classes of antibiotics (multi-drug resistant strains). Fifteen ARGs were detected with a majority related to tetracycline resistance (*tetL*, *tetM*, and *tetO*) carried by mobile genetic elements integrated in the genome.

The present study explains the observed phenotypic tetracycline resistance of *Strep. uberis* at the genetic and genomic level and demonstrates a non-understood phenomenon of a high Sequence Type diversity.