Molecular Epidemiology Of Macrolide And/Or Tetracycline Resistant Streptococcus Agalactiae And Streptococcus Uberis From Bovine Mastitis

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Publication date:
2011

Citation (APA):
MOLECULAR EPIDEMIOLOGY OF MACROLIDE AND/OR TETRACYCLINE RESISTANT *STREPTOCOCCUS AGALACTIAE* AND *STREPTOCOCCUS UBERIS* FROM BOVINE MASTITIS

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**Objectives:** To identify macrolide/tetracycline phenotypes and genotypes among *Streptococcus agalactiae* (Group B Streptococcus, GBS) and *S. uberis* from bovine subclinical mastitis, relevant for therapeutic policies, and evaluate further molecular features.

**Methods:** GBS (n=60) and *S. uberis* (n=30) field isolates from 9 herds in Portugal were characterized by pulsed field gel electrophoresis (PFGE). Resistance to macrolides (erythromycin-E), lincosamides (pirlimycin-PRL), tetracycline-TET, and the constitutive macrolide-lincosamide resistance phenotype (cMLS) was evaluated by disk diffusion. Resistance genes (*mef*A; *erm*A; *erm*B; *lin*B; *tet*O; *tet*T; *tet*S; *tet*Q; *tet*K; *tet*W; *tet*L) were screened by PCR among all isolates. The C5a peptidase and *lmb* genes, important virulence factors in human GBS localized in a composite transposon, were screened by PCR among the bovine GBS. Capsular serotyping of GBS was performed by agglutination and by PCR-sequencing the capsule gene cluster. Capsular serotyping of *S. uberis* was performed by agglutination and by PCR-sequencing the capsule gene cluster.

**Results:** A total of four PFGE clusters comprised 50% of the GBS and four PFGE clusters comprised 53% of the *S. uberis*. Co-resistance to E and PRL (18%-27%) and to TET (57%-60%) was observed in both species. Resistance to PRL and susceptibility to E (LSA phenotype) was found in 27% of *S. uberis* isolates. Diverse resistance genotypes were found: *erm*B/*tet*O/*tet*K in GBS and *erm*B/*tet*O or *lin*B/*tet*S in *S. uberis*. Both C5a peptidase and *lmb* genes were present in 20% of GBS. Two known molecular serotypes were detected: III-3 (n=1) and V (n=14), and four types (detected among 75% of GBS) showed *cps*DEF sequences distinct from those described so far, and most were found to be herd-specific.

**Conclusion:** A contagious source for both GBS and *S. uberis* was assessed. Dissemination of antimicrobial-resistance was clonal and by lateral gene transfer. Serotype-specific capsular polysaccharides appear to have evolved in a herd-associated manner. GBS with C5a peptidase and *lmb* were of capsular serotype V – common in human GBS infection.

**Max:** 300 words

**Max space (including names and titles):** 13,5 cm wide x 16,5 cm high

**Topic:** Animal streptococci (24. Mastitis-associated streptococci: *Streptococcus agalactiae*, *Streptococcus uberis*)

**Presentation:** Poster