

Detection of Staphylococcus Species from Subclinical Mastitis Cattle Milk Using MALDI-TOF MS, PCR and Sequencing

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Abstract

Mastitis is one of the economically important diseases in dairy industry. Staphylococcus spp. are amongst bacteria that bovine mastitis (BM) worldwide and they produce and contain a spectrum of extracellular protein toxins, virulence factors and antimicrobial resistant properties which are thought to contribute to the pathogenicity of the organism. This study aimed at detecting Staphylococcus spp. from cattle milk with subclinical mastitis using MALDI-TOF MS and 16S rRNA gene Sanger sequencing, virulence genes and screening of antimicrobial resistance (AMR). Out of 166 cows that were screened for subclinical mastitis only 33.13% were considered to have subclinical mastitis at a cow level, while the quarter-level prevalence was 54%. Thereafter from 50 bacterial isolates cultured, 76% were identified as *S. aureus* followed by *S. chromogenes* (12%) *S. epidermidis* (4%), and *S. haemolyticus* (4%) by MALDI-TOF MS. The 16S rRNA gene sequences indicated that *S. aureus* was the dominant species at 76%, followed by *S. chromogenes*, *S. agnetis*, *S. argenteus* and *S. devriesei* at 10%, 4%, 4% and 1% respectively. The AMR test showed that 86% of the isolates were resistant to penicillin followed by ciprofloxacin 80%, vancomycin 76% and ceftiofur 52%. Moreover, our genomic detection of antimicrobial resistant and virulence genes showed that *mecA* gene was detected in 16 % of all isolates, on the other hand 52% of all the isolates carried *Lg* gene, followed by *coa* and *spa* genes with 42% and 40% respectively. While *hla* and *hly* genes were detected in 38% of the isolates whereas *Sea*, and *bap* genes were detected in 10% and 2% respectively. According to our findings, MALDI-TOF MS and 16S rRNA sequence analysis can be used to detect Staphylococcus spp. in milk samples on a routine basis. The study indicated the importance of using various diagnostic tools for bacterial infection surveillance involved in subclinical mastitis to ensure accurate species identification.