

TITLE: PHENOGENOTYPICAL CHARACTERIZATION OF BETA-LACTAMIC RESISTANCE IN *Staphylococcus* spp. FROM A DAIRY FARM

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ABSTRACT: *Staphylococcus* spp. is the most important contagious pathogen of the bovine mastitis, presenting a high level of antimicrobial resistance, especially to beta-lactams, which favors its persistence in the herd. In order to characterize *Staphylococcus* spp. strains from a dairy farm in the South-Fluminense region of Rio de Janeiro, samples of milk, swabs from milking professionals hands and nasal conduits, and nasal swabs from dogs and cats were collected in the period of 2014 and 2015. A total of 321 samples were obtained, resulting in 248 *Staphylococcus* spp. isolates. All isolates were submitted to an interpretative reading antibiogram with penicillin G, oxacillin, amoxicilin + clavulanic acid and cefoxitin to search beta-lactams resistance. The phenotypic test revealed high percentages of resistance to penicillin G, 54,4% (135/248), also positive for *edge* zone test, and confirmed the expected low level of resistance to amoxicilin + clavulanic acid, 2% (5/248). The resistance to oxacilin and cefoxitin observed were 7,3% (18/248) and 1,6% (6/248) respectively. The *mec* gene analyses comprised the detection of classic *mecA*, *mecA* variant described by Melo et al. (2014) and *mecC*. The classic *mecA* gene was detected in three strains, the variant of *mecA* gene in two strains and one isolate presented an amplification product suggestive of *mecC* gene, but the sequencing analysis showed no similarity with this homologue. Analyzing the correlation between cefoxitin and oxacillin markers with the detection of the *mec* gene, was observed that cefoxitin demonstrated greater discriminatory power than oxacillin. The primers to detect the *mec* regulatory system, designed in this study, classified only the isolates that amplified the classic *mecA* gene, which carried the SCC*mec* type V, and was not possible to identify the *mec* regulatory system in isolates *mecA* variant positives. The phenotypical results indicated the production of beta-lactamases as the predominant mechanism of resistance in this farm. Considering *Staphylococcus* species distribution, *S. aureus* was the most prevalent specie, 64,1% (150/234), however, only *Staphylococcus* coagulase negative carried *mec* genes, confirming the important role of this specie as reservoir of resistance genes to *S. aureus*. New approaches will be necessary to better understand how the *mec* regulatory system plays in isolates *mecA* variant positives.

Keywords: beta-lactamic resistance, *mec* genes, *Staphylococcus* spp..

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