

TITLE: CRISPR, RESISTOME AND MOBILOME OF *Staphylococcus epidermidis* RECOVERED FROM RECREATIONAL COASTAL WATERS

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ABSTRACT:

Coagulase-negative *Staphylococcus* (CoNS) such as *S. epidermidis* can play an important role as reservoir of antimicrobial resistance and virulence genes. Some authors suggest that this almost unidirectional horizontal gene transfer (HGT) from CoNS to *S. aureus* may be explained by differences in abundances of CRISPR systems, so that *S. epidermidis* would have more CRISPR than *S. aureus*. However, the few studies conducted with *S. epidermidis* genomes described a small number of CRISPR. These were performed mainly in clinical samples, which may be under a positive pressure for antibiotic resistance genes (ARG) and the opposite for CRISPR. On the other hand, the abundance of phages in seawater (5-10 phages/bacteria) may work as a positive pressure for maintenance of CRISPR. In this context, the present study aims to analyze the features of 52 *S. epidermidis* genomes isolated from coastal waters, focusing on the presence of CRISPR loci, ARG and mobile genetic elements. We isolated *Staphylococcus* strains from four beaches of the southern area of Rio de Janeiro, Brazil. A total of 1008 isolates were recovered among which 477 were identified as *Staphylococcus* or were not classified in any genus by MALDI-TOF. These isolates were analyzed by species-specific PCR and *rrs* sequencing, when necessary. Then, all *S. epidermidis* (n = 52) had their antimicrobial susceptibility profile assessed by disk diffusion, and their whole genome sequenced (Illumina MiSeq). Most isolates were resistant to Penicillin (40,38%), followed by Erythromycin (28,85%), Oxacillin/Cefoxitin (13,46%), Tetracycline (3,84%), Trimethoprim-sulfamethoxazole (1,92%). The scaffolds and contigs were assembled (CLC) and annotated (RAST) and in silico MLST identified 22 different STs, including 14 new ones. ARG found included *aadD*(n=3), *mecA*(n=3), *blaZ*(n=29), *fosB*(n=52), *mphC*(n=6), *msrA*(n=13), *tetK*(n=1), *dfrG*(n=1). By PlasmidFinder, we detected four different plasmids in eight strains, in which two (pUB110 and pWBG744) showed strong evidence that they are intact using Blastn and Mauve. The plasmid pUB110 is known as an aminoglycoside resistance determinant and may be associated with type II SCCmec and pWBG744 is native to *S. aureus* and encodes multidrug efflux protein. Complete or incomplete phage sequences were found in all strains, Staphy_StB20_like (n=29) and Staphy_StB12 (n=22) were the most recurrent. Although all strains had preliminarily presented CRISPR array, it is necessary to confirm this finding and *cas* genes were detected only in five strains. Our results may help elucidate this relationship between CRISPR and ARG in *Staphylococcus*. Studies to date have not allowed us to state that there is more CRISPR loci in *S. epidermidis* than in *S. aureus*. We can analyze the influence of the environment of isolation on these results or corroborate a small number of CRISPR loci in *S. epidermidis*, suggesting that this is not the major determinant of HGT among *Staphylococcus*.

Keywords: *Staphylococcus epidermidis*, CRISPR, antibiotic resistance genes, HGT

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