

## DETECTION OF NOVEL GENETIC DETERMINANTS OF ANTIMICROBIAL RESISTANCE IN *Staphylococcus aureus* THROUGH PAN-GENOME AND RESISTOME ANALYSIS

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Antimicrobial resistance (AMR) has become a significant global health concern, particularly with methicillin-resistant *Staphylococcus aureus* (MRSA), which has a complex genomic basis and evolving resistance mechanisms. Although genes such as *mecA* and *mecRI* are well known contributors, previous research has not fully identified the genes associated with AMR, particularly those linked to mobile genetic elements. This study aimed to uncover such novel genes to improve AMR management strategies. We retrieved and assembled 91 publicly available *S. aureus* genomes representing Asian human and livestock hosts, which were originally sequenced using Oxford Nanopore technology, and supplemented these with 56 laboratory-validated MRSA and methicillin-susceptible *S. aureus* genomes from the BV-BRC database. Genomes were annotated and sequence typed with Prokka and MLST (v. 2.22.0) tools, respectively, and antibiotic-resistant genes and virulence genes were detected using ABRicate (v. 1.0.1) against CARD, ResFinder, and VFDB databases. Pan-genome analysis (Roary v. 3.13.0), mobile genetic element identification (plasmidFinder, ISEScan v. 1.7.2.3), Fisher's exact test, and the statistical association test, were performed to evaluate the correlations between genotype and phenotype. All analyses were executed on Galaxy and Google Colab platforms. Among the 334 MRSA-specific genes identified, *mecRI* (resistance) and *essC*, *esxB*, *sdrE* and *fnbB* (virulence) exhibited significant association with resistant phenotype ( $p < 0.001$ ). Two novel MGE-associated transposases (LEHPCDPB\_00452, OKANCEOL\_00333) were detected, suggesting a potential for horizontal gene transfer. From the several sequence types detected through MLST, only ST\_8 showed a statistically significant association with methicillin resistance ( $p < 0.01$ ). In conclusion, this research revealed critical genetic determinants for methicillin resistance and virulence in *S. aureus*, providing a promising framework for AMR surveillance and the discovery of novel therapeutic targets. This study highlights the value of integrating next-generation sequencing with laboratory validation in exploring AMR mechanisms. The future work will focus on enhancing the sample size to further refine and strengthen the results.

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