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Molecular characterization of antimicrobial resistance genes against *Staphylococcus aureus* isolates from Trinidad and Tobago

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Summary *Staphylococcus aureus* continues to pose major public health challenges in many areas because of antibiotic resistance problems. In the Caribbean, especially Trinidad and Tobago, the challenge is not different. This study was performed to evaluate the antimicrobial resistance gene prevalence among *S. aureus* isolates in Trinidad and Tobago.

Standard and molecular microbiological methods, including the Microscan automated system, DNA microarray and multi locus sequence typing (MLST) analysis, were performed on 309 clinical *S. aureus* isolates recovered from patients who were treated at three of the country's main health institutions.

S. aureus exhibited susceptibilities ≥80% to eleven of the 19 antimicrobials tested against it, and these belong to the most commonly used and available antibiotics in the country. While the antibiotic to which it was most susceptible of the commonly used antibiotics was trimethoprim/sulfamethoxazole, the antibiotics to which it was least susceptible or most resistant to were ampicillin and penicillin. *S. aureus* isolates from the pediatric ward produced the greatest rate of susceptibility among the isolates recovered from patients admitted into hospitals, while isolates from Accident and Emergency rooms displayed the greatest susceptibilities among patients from the community.

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S. aureus isolates from the country did not harbor acquired resistant genes targeting clindamycin/macrolides (*erm*B), linezolid (*cfr*) or vancomycin (*van*A). The *bla*Z gene, which is the most common beta lactam (Penicillinase) resistance mechanism for *S. aureus*, was observed in 88.7% of the methicillin susceptible *S. aureus*, while methicillin resistance mediated by the *mec* gene was present in 13.6%. Most of the resistance markers found in MRSA isolates were significantly associated with the ST239-MRSA-III strain in this study, and all isolates that belonged to the USA300 strain, which additionally encoded both the *PVL* gene and ACME cluster, belonged to CC8.

Several resistant genes, such as *van*A, *cfr* and *erm*B, mediating resistance in *S. aureus*, are currently non-existent in Trinidad and Tobago. However, the majority of SCC*mec* genes were observed, suggesting that there is ongoing nosocomial transmission with minimal community transmission. This calls for stringent antibiotic stewardship and policies in the country.

Introduction

Severe *S. aureus* infections require combative treatment, including incision and drainage for abscesses and systematic antibiotics [1]. Systematic antibiotics are necessary for deep-seated and systematic infections [2]. In the 1940s, penicillin, a widely used antibiotic derived from *Penicillium* fungi, was introduced for use [3]. Penicillin proved to be an effective antibiotic as it decreased the incidence and spread of infections as well as deaths caused by *S. aureus*. Penicillin acts by preventing cell wall formation as it competes for protein binding sites on the bacterium. However, resistance to penicillin was observed in the late 1940s with the emergence of the enzyme penicillinase, which inactivates penicillin [3]. Treating penicillin-resistant *S. aureus* led to the synthesis and further introduction of methicillin in 1959 [4]. However, in 1961, there were reports from the United Kingdom of methicillin-resistant *S. aureus* (MRSA). This trend was similarly seen in other European countries, Japan, the United States of America and Australia [4]. In the subsequent years, resistance developed to a range of antibiotics, including macrolides, fluoroquinolones, aminoglycosides, glycopeptides and tetracyclines.

Phenotypic methods have been used to identify and susceptibility test *S. aureus* isolates in many studies, and data on the characterization of *S. aureus* using molecular methods are limited in most undeveloped countries [5,6]. Knowledge of *S. aureus* susceptibility patterns and molecular characterization of genes mediating resistance are very important for developing effective infection control measures and treating or combating staphylococcal infections [5,6].

Many staphylococcal strains are known to resist multiple antibiotics and exhibit reduced susceptibility to glycopeptides, such as vancomycin. However, with the rise of glycopeptide intermediate susceptible *S. aureus* (GISA) and resistant genes, such as *van*A, there is a great concern in administering treatment [7—9]. Alternatively, other drugs have been used, such as linezolid, whose resistance is uncommon and encoded for by the *cfr* gene [8,9]. Several other genes are responsible for antibiotic resistance in *S. aureus*. For example, macrolide/clindamycin resistance is encoded by the *erm* gene, while the *aphA*3 and *sat* genes confer resistance to neo-/kanamycin and streptomycin, respectively. Gentamycin and tobramycin resistance are encoded for by the *accA-aph*D genes and tetracycline resistance is carried on the *tet* genes [8,9]. In an era of rapid antimicrobial resistance gene spread on multi resistance plasmids, our main objective was to evaluate and document the genes encoding antibiotic resistance in *S. aureus* isolates recovered from several major hospitals in Trinidad and Tobago.

Materials and methods

This was an observational cross-sectional study to investigate the susceptibility profiles of several antimicrobial agents on *S. aureus* isolates encountered in several infection types for patients admitted into the hospitals and those coming directly from communities in Trinidad and Tobago over a 10-month period in 2011—2012. The study was conducted at three of the five major regional hospitals across the country. Ethical approval for

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