

Molecular Characterization of Resistance and Virulence in Methicillin Resistant *Staphylococcus aureus* (MRSA) from the Private Sector in KwaZulu-Natal (KZN), South Africa

Daniel G. Amoako^a, Linda A. Bester^b, Anou M. Somboro^a, Sooraj Baijnath^c, Chetna N. Govind^d, Sabiha Y. Essack^a

^aAntimicrobial Research Unit, ^bBiomedical Resource Unit, ^cCatalysis and Peptide Research Unit, University of KwaZulu-Natal, Durban, South Africa.

^dLancet Laboratories, Durban, South Africa.

Introduction and Purpose:

MRSA has far reaching consequences in the public health, economic and social sectors. These strains harbor mobile genetic elements (MGEs), including plasmids, pathogenicity islands, transposons, integrons and prophages, which comprise 15-25% of the genome carrying genetic determinants of antibiotic resistance and virulence.¹ This study describes the genetic relatedness, and characterizes the plasmid-encoded antibiotic resistance and virulence profile of 27 clinical MRSA from a private laboratory in KZN, South Africa.

Methods:

Isolates were subjected to antimicrobial susceptibility testing using Clinical Laboratory Standards institute Guidelines.² Molecular characterization of common resistance encoding genes (*blaZ*, *aac (2')-aph (6'')*, *ermC*, *tetK*) and frequently encountered virulence factors (*hla*, *hld*, *eta* and *LukS/F-PV*) was determined by PCR using plasmid DNA as the template. The genetic relatedness was determined by pulsed field gel electrophoresis (PFGE).³

Results:

- All isolates were plasmid positive, and displayed ampicillin, ciprofloxacin, gentamicin, rifampicin, tetracycline, erythromycin & clindamycin resistance.
- All isolates were fully susceptible to daptomycin, linezolid, vancomycin, tigecycline and fusidic acid.
- Multi-drug resistance was evident in 74.1% (20/27) of the isolates.
- The frequency of resistance and virulence genes ranged from 0-100%
- PFGE analysis generated 10 pulsotypes, designated A–J, which correlated well with resistance profiles of isolates.
- 85.2% (23/27) of the isolates clustered into six major PFGE types indicating similar circulating MRSA clones.

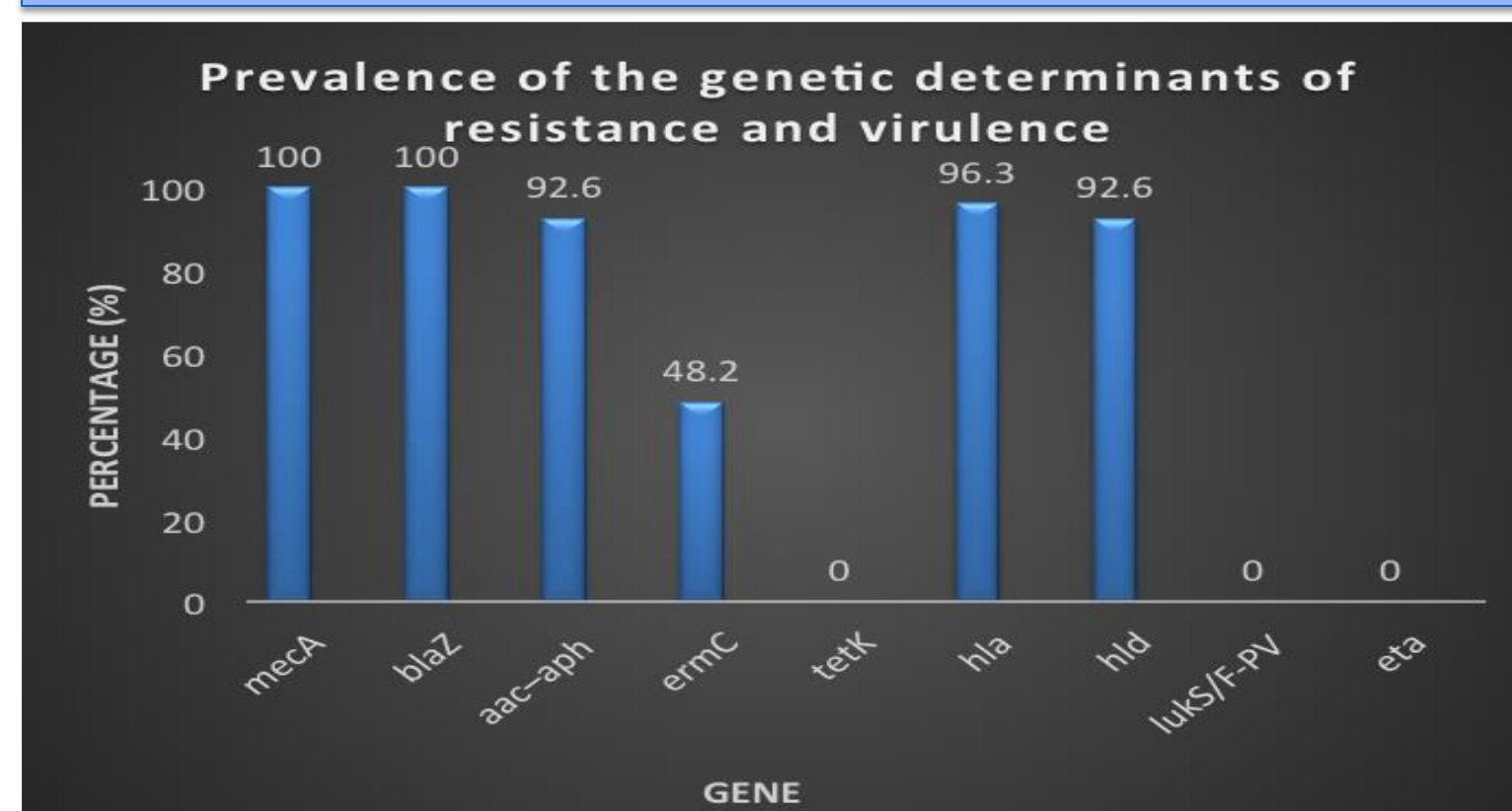


Figure 1. % Prevalence of the genetic determinants of resistance and virulence from MRSA isolates from private sector in KZN.

Table 1. Minimum Inhibitory Concentration (MIC) Distributions

Antibiotic	Resistance pattern, n (%)		Distribution of MIC (mg/ml)												
	R	S	<0.25	0.5	1	2	4	8	16	32	64	128	256	>512	
Ampicillin	27 (100)	0	0	0	0	0	0	0	0	0	0	1	1	25	
Ciprofloxacin	23 (85.2)	4 (14.8)	0	2	2	0	4	1	1	0	2	5	8	2	
Gentamicin	20 (74.1)	7 (25.9)	2	3	1	0	1	0	2	4	7	7	0	0	
Erythromycin	16 (59.3)	11 (40.7)	0	8	2	1	0	2	6	4	4	0	0	0	
Rifampicin	19 (70.4)	8 (29.6)	7	1	0	0	0	0	0	0	1	5	8	5	
Tetracycline	18 (66.7)	9 (33.3)	6	1	1	2	0	0	1	1	8	5	2	0	
Clindamycin	3 (11.1)	24 (88.9)	24	0	1	1	0	0	0	0	0	0	0	1	
Daptomycin	0	27 (100)	7	16	4	0	0	0	0	0	0	0	0	0	
Vancomycin	0	27 (100)	1	10	16	0	0	0	0	0	0	0	0	0	
Linezolid	0	27 (100)	0	0	4	23	0	0	0	0	0	0	0	0	
Fusidic acid	0	27 (100)	26	1	0	0	0	0	0	0	0	0	0	0	
Tigecycline	0	27 (100)	27	0	0	0	0	0	0	0	0	0	0	0	

R, resistant; I, intermediate; S, susceptible, All intermediate MIC values were taken as resistant.

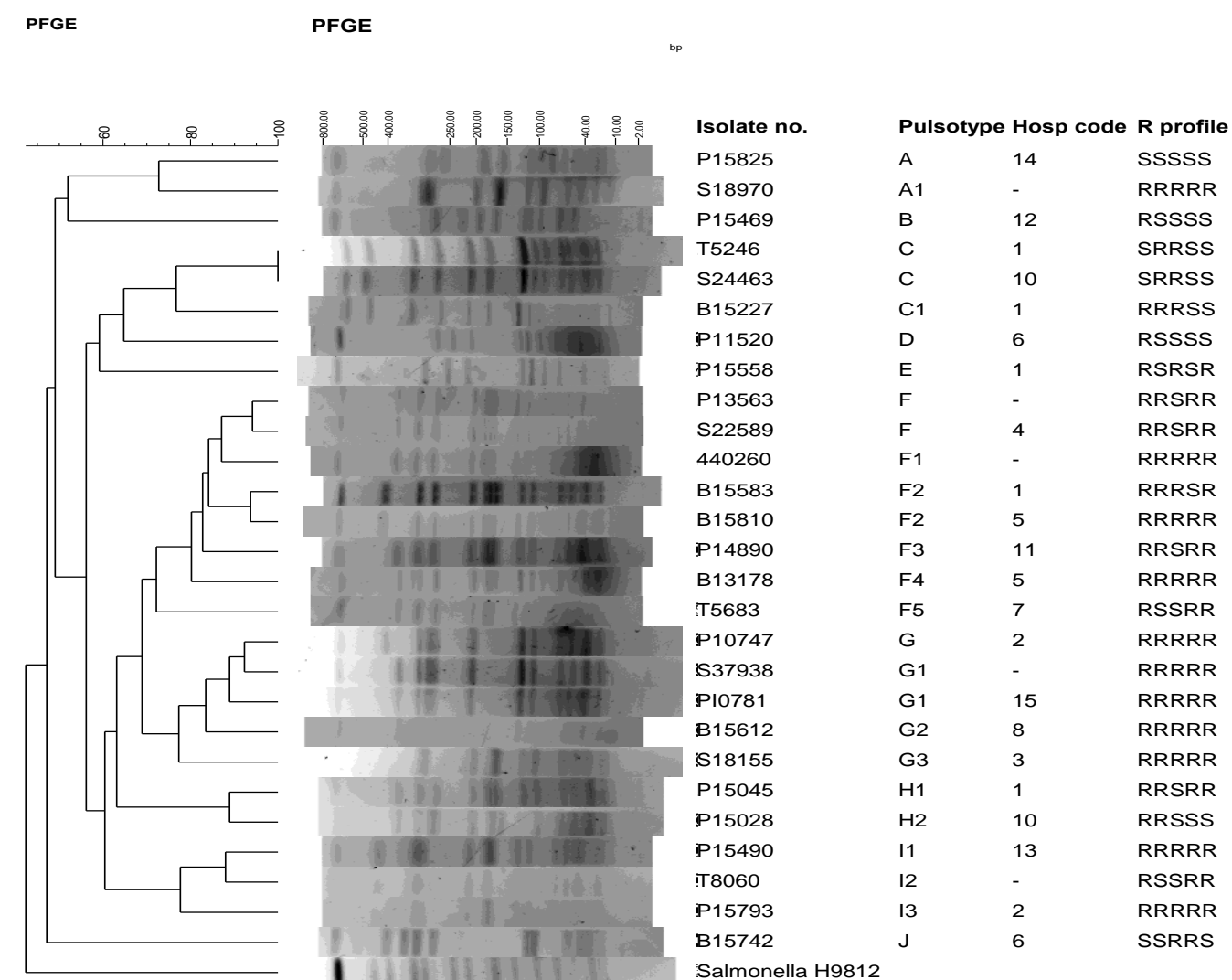


Figure 2: PFGE *Smal* genotypic types generated from 27 clinical MRSA isolates from private sector in KZN.

Pretested *Salmonella* serotype *Braenderup* strain H9812 was used as the quality control strain. The R and S indicate resistance or susceptibility for ciprofloxacin, gentamicin, erythromycin, tetracycline and rifampicin respectively. The alphabets A–J shows the main pulsotype and subtype of each isolate. The numbers 1–15 indicates codes of the hospital centers where the MRSA isolates were collected.

Conclusion:

Inter-health center spread of identical and closely related clones of MRSA is evident in KZN, South Africa, emphasizing the need for the implementation of efficient and effective infection control programs.

References:

- Figueiredo AMS, Ferreira FA. The multifaceted resources and microevolution of the successful human and animal pathogen methicillin-resistant *Staphylococcus aureus*. *Mem Inst Oswaldo Cruz* 2014;**109**:265-78.
- Clinical and Laboratory Standards Institute. Performance standards for antimicrobial susceptibility testing: twenty-fourth informational supplement. CLSI document M100-S24. Wayne, PA: Clinical and Laboratory Standards Institute; 2014.
- Prevost G, Pottecher B, Dahlet M, Bientz M, Mantz J, Piemont Y. Pulsed field gel electrophoresis as a new epidemiological tool for monitoring methicillin-resistant *Staphylococcus aureus* in an intensive care unit. *J Hosp Infect* 1991;**17**:255-69.

Acknowledgements:

The authors are grateful to the University of KwaZulu Natal, the National Research Foundation and the Biomedical Resource Unit for funding the study.

Disclosures:

Professor Essack is a member of the Global Respiratory Infection Partnership sponsored by Reckitt & Benckiser.