

M.Sc thesis (Microbiology)

Submitted by

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<u>Title</u>

Multiplex PCR for direct detection of vancomycin resistant *Staphylococcus aureus* isolated from clinical samples

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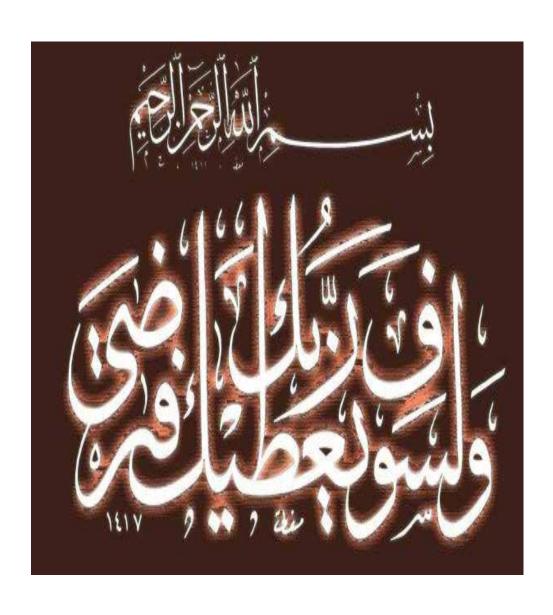
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ABSTRACT

Ahmed El Sayed Mohamed El Aysh. Multiplex PCR for direct detection of vancomycin resistant *Staphylococcus aureus* isolated from clinical samples (M.Sc). Faculty of Science, Ain Shams University.

Vancomycin resistant Staphylococcus aureus (VRSA) has emerged over the last ten years. The most resistant strains (fortunately rare) bear the vanA gene cluster. In this study 439 isolates were collected from hospitalized patients in Zagazig university hospital, ten VRSA strains were isolated from 220 patients infected with S. aureus according to conventional methods then investigated by multiplex PCR for vanA and nuc genes. A high percentage of VRSA was observed in the present study which may be explained by administration of multiple prophylactic and post-operative antibiotics with prolonged hospitalization. All VRSA isolates were sensitive to (cefoperazone, trimethoprime, amikin and rifampin) and resistant to (clavulinic acid, cefoxtin, cefazolin, oxacillin, tetracyclin, vancomycin and Cefexime). Only five of them were positive for vanA gene by multiplex PCR, although all were confirmed as S. aureus by multiplex PCR (nuc gene positive). The resistance of Staphylococci to vancomycin has been found to be reversible under laboratory conditions. Moreover, thickening of the bacterial cell wall may be the underlying mechanism for vancomycin resistance in these Randomly amplified polymorphic bacteria. DNA previously used for typing of MRSA. The present study provided a molecular typing (by RAPD) for the reported VRSA strains using four different pairs of primers. However, more studies are still needed to explore the definite mechanisms by which these strains acquire resistance to vancomycin which may open the door to overcome this problem.

LIST OF ABBREVIATION

AM	Amikin
AMCC	Clavulinc Acid
AMPI	Ampicillin
ATP	Adenosin Tri Phosphates
CDC	Centers For Disease Control And Prevention
CEC	Cefoxtin
CES	Cefoperazon
CTX	Cefexim
CZ	Cefazoline
DNA	Deoxyribonucleic Acid
EDTA	Ethylene Diamine Tetracetic Acid
FW	Fresh Weight
HLR	High-Level Representation
ICU	Intensive Care Unit
MIC	Minimum Inhibition Concentration
MI-VRSA	Michegene Isolates
MRSA	Methicillin-Resistant Staphylococcus aureus
MSSA	Methicillin-Sensitive Staphylococcus aureus
NCCLS	The National Commmittee For Clinical Laboratory
	Standards
NNISS	National Institute of Statistical Sciences
OX	Oxacilin
PA-VRSA	Pennsylvania VRSA
PBP2A	Penicillin-Binding Protein 2A
PBPS	Penicillin-Binding Proteins
PCR	Polymerase Chain Reaction
PFGE	Pulsed Field Gel Electrophoresis
PMNS	Polymorphonuclear Leukocytes
R RNA	Ribosomal Ribonucleic Acid
RAPD	Random Amplification Of Polymorphic
RFLP	Restriction Fragment Length Polymorphism
RIF	Rifampine
RNA	Ribonucleic Acid
SXT	Trimethoprime
TBE	Tris Base, Boric Acid And EDTA.

TE	Tetracycline
VA	Vancomycin
VISA	Vancomycin- Intermediate S. aureus
VRE	Vancomycin-Resistant Enterococci
VRSA	Vancomycin-Resistant Staphylococcus aureus
VSSA	Vancomycin-Susceptible S. aureus

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