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*Title*

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

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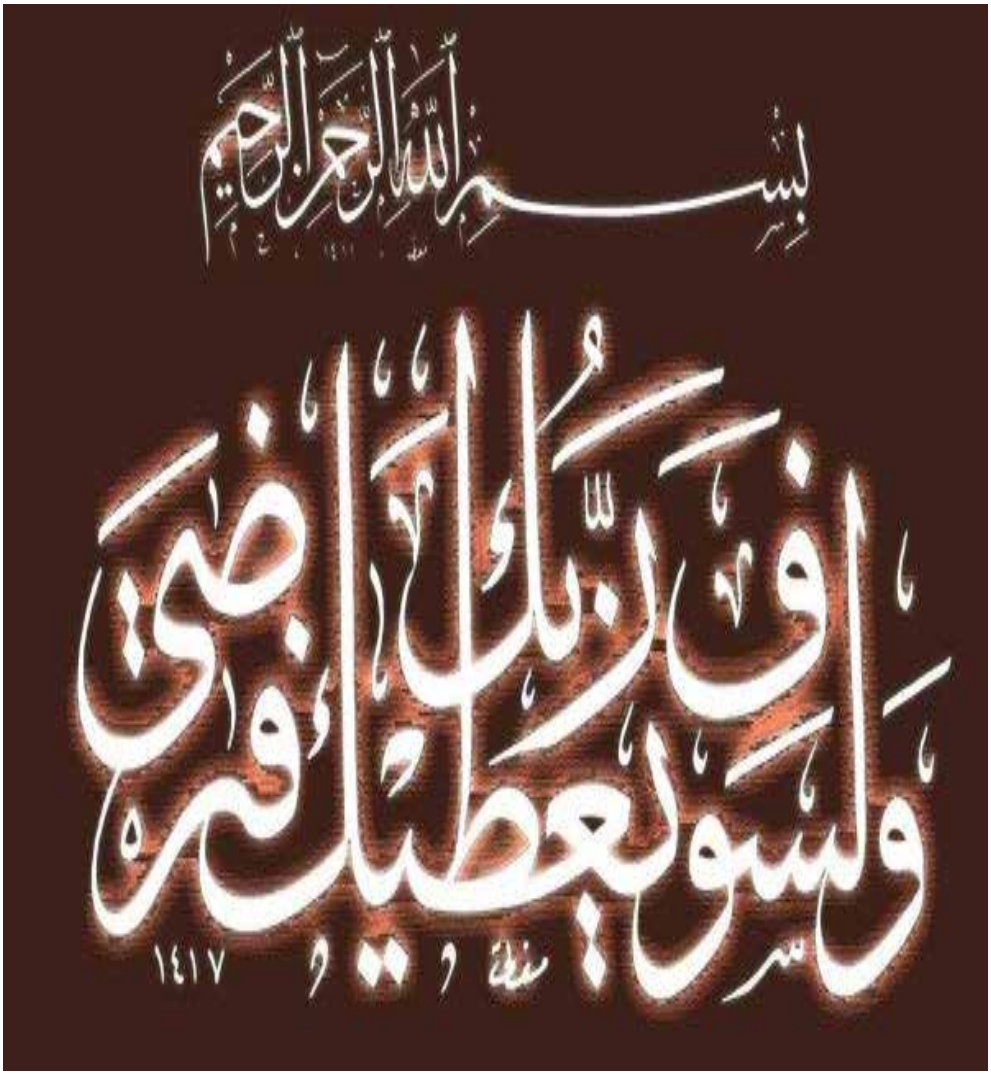
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# ABSTRACT

**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 bacteria. Randomly amplified polymorphic DNA was 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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