The Effect of Antibiotics on Toxin Gene Expression in PVL-positive Staphylococcus aureus Strains

BALAKY, SALAH,TOFIK

Declaration

I declare that the work within this thesis, submitted for the degree of Doctor of Philosophy, is my own original work and has not been submitted for a degree at this or any other University.

Signed ……..

Date ……..

Abstract

Staphylococcus aureus is an extra-ordinarily versatile pathogen causing a wide spectrum of infections. The aims of this study are to analyze 10 clinical isolates of *S. aureus* from the UK by Multi Locus Sequence Typing (MLST) and determining their PVL-type variants. In addition to that, to study the effect of several antibiotics at sub inhibitory concentrations on a number of virulence factors at mRNA using quantitative PCR and protein levels using proteomic methods. Western blotting was used to study differential expression of Spa at protein levels.

Data showed that the 10 clinical isolates belong to seven clonal complexes (CCs), which are CC1, CC5, CC8, CC22, CC30, CC88, and CC121. Genetic variation within *lukSF-PV* gene showed that three of these isolates were belong to the same PVL type variant of CA-MRSA USA300 strain, R variant. From which, two isolates were found to belong to the same CC of USA300, CC8. The remaining 7 isolates were found to belong to H variant. Data presented here showed that the sub-MIC levels of both cell wall inhibitors reduced *lukSF-PV* and *spa* steady-state mRNA levels when cells were grown in the presence of these antibiotics for one hour. However, after 5 hrs post antibiotic addition of these two antibiotics, vancomycin remained depressed *lukSF-PV* and *spa* steady-state mRNA levels as well as at protein levels, but oxacillin increased *spa* and *lukSF-PV* mRNA levels, as well as Spa at protein levels. Protein synthesis inhibitors clindamycin and linezolid were both caused an increase of *lukSF-PV* mRNA levels, but they both decreased *spa* mRNA levels, when cultures grown in the presence of these antibiotics for one hour. However, when cultures grown with these antibiotics for 5 hrs, clindamycin remained to increase *lukSF-PV* and decrease *spa* mRNA levels and protein levels, but linezolid decreased both virulence factors at mRNA and protein levels.

The data showed in this study confirmed that growing *S. aureus* in the presence of oxacillin induce toxin expression and might enhance the virulence of this bacterium, therefore using these antibiotics to treat *S. aureus* infections may contribute to worse outcomes. These data also confirmed that linezolid and vancomycin, are both important selections of antimicrobial agents to treat serious infections caused by the bacterium.

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Last, but certainly not least, I would like to thank my Mum, Dad and all loved ones for their continued support, particularly my wife Ahang because without her constant support this would not have been possible.

DEDICATION

This Thesis is Dedicated to My Wife Ahang, My Mum and Dad. It is Also Dedicated Proudly to My Uncle Abdul-Wahid and All Loved Ones.

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