29th ECCMID Amsterdam, Netherlands 13 – 16 April 2019

The congress of ESCMID

O1108 Genomic characterization of six Panton-Valentine leukocidine-encoding bacteriophage types detected in *Staphylococcus aureus* isolates from South Africa and Nigeria

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Background: The Panton-Valentine leukocidine (PVL) gene in *Staphylococcus aureus* strains was previously associated with community-associated methicillin-resistant *Staphylococcus aureus* (CA-MRSA) strains. However, PVL-positive healthcare-associated MRSA (HA-MRSA) and PVL-positive methicillin-susceptible *S. aureus* (MSSA), have since been reported. Thirteen PVL-encoding bacteriophages have been reported, however, only 10 have been included in a PCR-based typing scheme. Due to limited information on the genomic characterization of PVL-encoding bacteriophage types associated with *S. aureus* strains from South Africa and Nigeria, this study characterized *S. aureus* bacteriophage types using whole-genome sequencing.

Materials/methods: Seventy clinical *S. aureus* isolates were screened for the presence of the PVL gene using multiplex-PCR assays, followed by the bacteriophage PCR-based typing scheme. Six representative isolates were selected and included: two typeable and four non-typeable isolates. Total Genomic DNA was performed using a commercial kit. Isolates were submitted to the National Institute of Communicable Disease/National Health Laboratory Service (NICD/NHLS) for WGS which was carried out on an Illumina MiSeq instrument (Illumina, US). The resultant paired-end sequenced reads (2X300 bp) were quality trimmed and assembled *de novo* using the CLC Genomics Workbench version 11 (Qiagen, Denmark). The PHASTER (PHAge Search Tool Enhanced Release, phaster.ca) tool was used to characterize and identify bacteriophages present in the sequenced isolates.

Results: Seven novel bacteriophage types were identified including four PVL-encoding bacteriophage and two novel non-PVL encoding bacteriophage types belonging to the *Siphoviridae* family of the genera *Biseptimusvirus* and *Triavirus*. Non-PVL encoding bacteriophages carried the *luk*E and *luk*D genes that belonged to the same family as the *luk*S/F-*PV* genes. One non-PVL-encoding bacteriophage did not harbour any leukotoxin genes. Sequence homology of 91% and 94% was reported between the *luk*E and *luk*D genes and the *luk*S-*PV* and *luk*F-*PV* genes explaining the detection of the *luk*E/D genes with the PVL screening PCR assay. Whole-genome sequencing identification of novel bacteriophage types explains the non-typeability of some strains using the PCR-based typing scheme.

Conclusions: Whole-genome sequencing showed that the *luk*S/F-*PV* primer pairs detected partial sequences of the PVL genes and identified novel PVL-encoding bacteriophages. This confirmed that the current PCR-based typing is insufficient to identify all PVL-encoding bacteriophage types in this study setting.