

Session: OS199 One health perspective on MDR Gram-positives: VRE & MRSA

Category: 3a. Resistance surveillance & epidemiology: MRSA, VRE & other Gram-positives

25 April 2017, 14:42 - 14:52
OS1003

Diversity of methicillin-resistant *Staphylococcus aureus* CC9/CC398 genomes and the phylogenetic relationship between food and human clinical isolates

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Background: An expansive spread of antimicrobial resistant bacteria with livestock origin such as Methicillin-resistant *Staphylococcus aureus* (MRSA) of clonal complex (CC)398, also named Livestock-associated (LA)-MRSA has been shown in the past. Direct animal exposure is considered to be the most effective way of MRSA CC398 transmission from livestock to humans. Recently, a novel hybrid LA-MRSA CC9/CC398 genotype has been described in human clinical cases showing both, poultry and human genetic adaptation. Poultry meat was considered to serve as a vehicle of a possible livestock-to-human transmission of CC9/CC398 MRSA. Here, we wish to describe CC9/CC398 genomes from poultry origin and their phylogenetic relationship to human clinical strains.

Material/methods: A collection of >14,000 *S. aureus*, comprising strains mainly of food and livestock origin was screened for CC9/CC398 MRSA strains applying *spa*- and MLST-typing and a multiplex real-time PCR assay targeting *nuc* and *mec*. CC9/CC398 MRSA were further characterized by DNA-microarray- and multiplex-PCR-analysis aiming to identify poultry-associated genomic features, i.e. ϕ Sa3 and ϕ Av β prophages. Whole-genome sequencing by a PacBio RSII platform and subsequent de novo assembling of the reads by HGAP.3 was done with four CC9/CC398 MRSA. Phylogenetic relationship of 33 CC9/CC398 isolates from food and human clinical cases was determined by SNP tree analysis.

Results: Thirty-seven CC9/CC398 MRSA were identified. Among these, 31 strains (all of poultry origin) carried the ϕ Sa3 prophage while 8/31 (all from turkey meat, except one from broiler meat) carried also the ϕ Av β prophage. Raw read de novo assemblies of four selected CC9/CC398 MRSA resulted in a closed draft genome for each isolate, which was further analysed in detail on virulence factors (virulome), antimicrobial resistances (resistome) and mobile genetic elements (mobilome). In general, all four isolates exhibited similar genome features, chromosomal resistance genes and virulence factors. However, distinct differences were observed in the content of mobile genetic elements, e.g. 1/4 isolates carried six different prophages while the others harboured four prophages, only. Isolates also differed in their number of plasmids including those plasmids harbouring genes encoding for antimicrobial resistances such as against phenicols and/or aminoglycosides. Here we provide detailed data on the diversity of the resistome, mobilome and virulome of CC9/CC398 MRSA from food and human origin. Phylogenetic SNP tree analysis confirmed a high identity of CC9/CC398 strains from human cases and of poultry, mainly turkey meat origin. Strains carrying the ϕ Av β prophage formed a separate group within the poultry-associated strains.

Conclusions: There is clear evidence that specific LA-MRSA lineages with unique genetic characteristics are adapting to humans. Poultry may serve as one main reservoir of these LA-MRSA lineages. The dissemination of highly pathogenic MRSA strains via the poultry food chain needs to be prevented by all means.