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Paper Poster Session I

Recent advances in molecular bacterial typing

Genomic comparison of epidemic and non-epidemic hospital-associated strains of *Enterococcus faecium*

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Objective. The main objective of the work was to perform a comparative genome analysis of clinical isolates of vancomycin-resistant *Enterococcus faecium* (VREF) belonging to the clonal complex 17 (CC-17) in order to identify determinants potentially involved in epidemicity.

Method. We sequenced (using the 454 pyrosequencing technology) and analyzed the genomes of three VREF strains exhibiting different epidemic potentials: UCBN1 was the cause of a large hospital outbreak in France, the UW1107 was isolated from a sporadic infection in Germany and AK-EM40 from a healthy carrier German patient. Using the sequence of the *E. faecium* strain Aus0004 as reference genome (fully sequenced and annotated), we searched for gene clusters present or absent in the different strains. These clusters were then screened among 54 epidemiologically-unrelated strains (epidemic or not epidemic) from the collection of clinical VREF isolates of the French National Reference Center for enterococci as well as in seven environmental isolates.

Results. In term of genes found, no significant differences were observed between the sequences of the two virulent strains UCBN1 and UW1107. By contrast, 219 loci were apparently not present in the non-epidemic strain AK-EM40 compared to the two former isolates. Some of these genes were grouped into six "blocks" (A to F) and could be "markers" of the epidemic trait of *E. faecium* strains belonging to CC-17. The sizes varied from 1.5 to 72.7 kb and the number of open reading frames present into the blocks A, B, C, D, E and F were 6, 13, 80, 58, 2 and 20, respectively. The presence or the absence of these blocks was therefore determined on a panel of 61 strains by multiplex PCR. First, the presence of the IS16 (criterion of belonging to the CC-17) was tested for all strains. Thus, 52 strains (including 2 of environmental origin) could be categorized as a member of the CC-17 and 9 as non-CC-17. The "epidemic potential" of the strains was estimated by the number of patients infected with the same clone during a hospital epidemic. The results indicate that 77% of isolates classified as "epidemic" (involved more than 3 reported infections) had the *msrA* gene encoding the methionine sulfoxide reductase that is representative of the block (A). Furthermore, the total number of blocks present in CC-17 strains appeared linked to the potential of dissemination: 71% and 73% of epidemic strains have more than two and three blocks, respectively.

Conclusion. This work is a preliminary study aimed to develop a fast tool, reliable and applicable in routine, to assess the epidemic potential of *E. faecium* clinical isolates. Further investigations using extensive epidemiological data are in progress to validate these markers.