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Metals and microbes

SEVEN *MER*A ALLELES ARE SPREAD AMONG *ENTEROCOCCUS* FROM DIFFERENT SPECIES, ENVIRONMENTS AND CLONAL BACKGROUNDS (1926-2012)

A. Sanchez-Valenzuela¹, E. Silveira², A.R. Freitas², P. Antunes³, F. Baquero⁴, T.M. Coque⁴, L. Peixe², C. Novais²

¹Microbiologia, Ramón y Cajal Hospital, Madrid, Portugal

²Faculdade de Farmácia., UCIBIO/REQUIMTE. Universidade do Porto, Porto, Portugal

³Faculdade de Farmácia/Faculdade de Ciências da Nutrição e Alimentação., UCIBIO/REQUIMTE. Universidade do Porto, Porto, Portugal

⁴Microbiologia, Ramón y Cajal Hospital, Madrid, Spain

Background

Successful bacteria accumulated different genetic features shaping their evolution and fitness to diverse environments/hosts. Mercury (Hg) is widespread in Nature and possibly contributed for selection of particular strains.

Objectives

To evaluate the dispersion of diverse *merA* alleles among enterococci from several origins and clonal backgrounds.

Methods

merA alleles was searched in *Enterococcus* available genomes, used to construct a maximum-likelihood phylogenetic tree. A PCR scheme+RFLP+sequencing was developed to detect the six *merA* alleles identified, namely I-(GenBank-AECE01000068), IIA-(AIUL01000023.1), IIB-(ASDU01000008.1), III-(NZ_KB947199.1), IV-(AECE01000068.1) and VI-(NZ_KB030055.1), among 918 *Enterococcus* (Portugal; human/animal/environment/food; 1996-2012). Clonality was evaluated by PFGE/MLST, *merA*-transfer by conjugation and Hg-phenotype in m_Enterococcus+128mg/L HgCl₂.

Conclusions

merA were found in 4% (n=37/918) of *Enterococcus* studied. PCR+RFLP+sequencing distinguished the six alleles previously found at Genbank and a new one (type V). They were distributed among diverse sources and clones, in our and GenBank isolates: types I and IV-human *E. faecalis* (*Efls*; n=5; USA; 1987-88; ST9/ST206); IIA-human/pig/trout/feed/hospital-sewage *E. faecium*-

Efm/E.hirae/Enterococcus_sp (n=17/1/1; Portugal/France/Denmark/Germany; 1989-2012; 12ST-*Efm* including from ST78-ST17-ST18-lineages-CC17); IIB-human *Efls/E.dispar* (n=2/1; Portugal/USA; 1926-2001; ST206/ST105/ST107); III-human/animal/hospital-sewage *Efls/E.casseliflavus* (n=15/1; Portugal/France/USA/Canada/Japan; 1961-2012; ST64/ST9/ST30/ST159/ST245); V-human/trout/pig-manure *Efm* (n=4; Portugal; 2001-2006; ST94/ST890); VI-human/hospital-sewage/piggery-soil/piggery-manure/trout *Efm* (n=23; Portugal, Italy France, Germany, Denmark, Hungary, Switzerland, Ireland, Norway, Brazil, USA; 1961-2012; 14ST including ST78-ST17-ST18-lineages-CC17). The presence of *merA* among major *Efm/ST9-Efls* lineages associated with human infections, suggests that mercurial-compounds potentially contributed for their selection/maintenance. *merA*⁺-*Enterococcus* grown in >128mg/L-HgCl₂. Transfer occurred for IIA and VI alleles. All but VI-*merA* alleles, were also identified in genomes of different species of Firmicutes (data not shown), suggesting genetic exchange of enterococci with bacteria sharing the same communities/environmental challenges.