### BOOK OF ABSTRACTS

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#### CREDITS

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## Bacterial resistance to copper and antibiotics: two sides of the same problem in the animal production setting?

E. Silveira<sup>1</sup>, <u>L. Dias</u><sup>1,2</sup>, P. Antunes<sup>1,2</sup>, A. R. Freitas<sup>1</sup>, T. M. Coque<sup>4</sup>, L. Peixe<sup>1</sup>, C. Novais<sup>1,3</sup>
<sup>1</sup>REQUIMTE. Microbiology Department, Pharmacy Faculty, Porto University, Portugal.
<sup>2</sup> Faculty of Nutrition and Food Sciences, Porto University, Portugal.
<sup>3</sup> CEBIMED, Health Sciences Faculty, Fernando Pessoa University, Porto, Portugal.
<sup>4</sup> Microbiology Service. Ramóny Cajal Hospital. Madrid. Spain

**Background**: Copper (Cu) is highly used in the animal production setting as feed supplement or in antiseptic formulations. We hypothesized that the use of Cu might contribute to select antibiotic resistant (AB<sup>R</sup>) pathogenic and non-pathogenic bacteria of animal origin, which might reach human by food chain.

**Methods**: Enterococcus spp (180 E. faecalis, 171 E. faecium, 199 from other species) and Listeria spp (13 L. monocytogenes, 13 L. innocua, 11 Listeria spp) were included in this study. They were collected (1999-2011) from swine feces and piggeries environmental samples, trout aquacultures (Enterococcus spp), poultry carcasses for human consumption (Enterococcus spp, Listeria spp), raw turkey meat and other raw/cooked food types (Listeria spp). Susceptibility to AB was studied by agar dilution/disk diffusion method (CLSI guidelines) and the search of AB/Cu<sup>R</sup> genes by PCR and sequencing. Their ability of transfer was evaluated by conjugation assays and plasmid location by S1-PFGE hybridization. Clonality of representative isolates of different species and sources that carried Cu<sup>R</sup> genes was studied by PFGE/MLST.

**Results**: Only a *L. innocua* (3%) from raw turkey meat and resistant to clindamycin showed the multicopper oxidase gene  $mco_{pLM5578}$  while the incidence of  $Cu^R$  genes among *Enterococcus* spp was 21%. The genes *tcrB* (coding for copper export ATPase) and *cueO* (multicopper oxidase) were detected in 17% and 16% of all *Enterococcus* spp, respectively. A higher incidence of  $Cu^R$  genes was detected in piggeries isolates (31% *vs* 15%- aquacultures, 10%-poultry) and in *E. faecium* (35% *vs* 13% *E. faecalis*, 15% other species). Isolates carrying  $Cu^R$  genes belonged to a polyclonal population: i) 55 *E. faecium*-37 PFGE clones (ST185, ST150/CC5; ST132, ST393/CC17; ST432; ST434; STnew); ii)14 *E. faecalis*-10 PFGE clones (ST224/CC21; ST16; ST445; ST260; ST288; ST49; ST53). Nevertheless, *E. faecium* PFGE type A (ST185/CC5) was detected in different pig farms. *Enterococcus* (n=29) carried plasmids of different sizes (150-320kb) with *tcrB* and/or *cueO* plus genes coding for resistance to tetracycline (*tetM*-18, *tetL*-23), erythromycin (*ermB*-24) and/or vancomycin (*vanA*-4). These plasmids were successfully transferred in all cases studied (n=17), both alone or with other smaller plasmids (25-70kb) also carrying AB<sup>R</sup> genes (n=4; *tetM*, *tetL*, *ermB*, *aac6-aph2*).

**Conclusion**: *Enterococcus* spp seem to be better reservoirs of the  $Cu^R$  genes studied than *Listeria* spp.  $Cu^R$  and  $AB^R$  genes were often co-located within large mobile plasmids or were co-transferred by different plasmids in the same genetic event. The intensive use of Cu might contribute to the maintenance of  $AB^R$  bacteria/genes in the animal setting, which can reach human by food chain.

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