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Parallel rather than intermixed populations of Enterococcus faecium from livestock and humans

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Background: *Enterococcus faecium* is an important cause of nosocomial infection, most of which is caused by a hospital-adapted lineage. This may have evolved from an animal-adapted lineage, and is genetically distinct from the human commensal lineage. We performed a large genome sequence-based 'One Health' study to determine whether this major hospital lineage is carried by livestock and define the genetic relatedness between isolates from different reservoirs.

Material/methods: A cross-sectional surveillance study was conducted to isolate *E. faecium* from 29 livestock farms and 20 municipal wastewater treatment plants across East Anglia, United Kingdom between 2014 and 2015. We sequenced 636 *E. faecium* from livestock (256 isolates from cattle, pigs, chickens and turkey) and wastewater (n=383) on an Illumina HiSeq2000 instrument. We obtained additional whole genome data for 782 *E. faecium* associated with bloodstream infection in the British

Isles (47% from East Anglia, 2001-2012), 11 historical strains, and 10 reference strains. Multilocus sequence types (ST) were derived from genome data. Phylogenetic and bioinformatics analyses were performed using open-access tools.

Results: The 1442 *E. faecium* isolates were assigned to 218 STs. Bayesian clustering divided the collection into 10 phylogenetic clusters (BAPS groups). *Enterococcus faecium* from humans and livestock were largely genetically segregated, with 97% human invasive isolates residing in the hospital-adapted lineage and 85% of animal isolates residing basal to this lineage. The four exceptions to this were: (i) BAPS group 5, containing 94 isolates from humans, pigs, cattle and chicken that was ancestral to the hospital-adapted lineage; (ii) 22 isolates from pigs that were nested within the hospital-adapted lineage; (iii) six human isolates in the animal-associated basal lineage; and (iv) human commensal lineage, containing 17 isolates from human infection and all four livestock species. Contrasting with this segregation between human and animal isolates, we observed that highly related *E. faecium* were shared by different animal species (turkeys and chickens), and by different farms. Vancomycin resistant *E. faecium* was not found in any of the farms. Genes encoding resistance to other antibiotics, including aminoglycosides, trimethoprim and tetracyclines were found in both humans and livestock. We identified copper and streptomycin-resistance genes in isolates from pigs that were rare in human-related isolates.

Conclusions: Genetic segregation of *E. faecium* from humans and livestock indicates parallel populations and low rates of bacterial flow between the two reservoirs. Further analysis is ongoing to examine the relatedness of antibiotic resistance genes in humans and livestock.

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