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The active role of aquatic invertebrates in the spread of antimicrobial resistance in *Enterococcus faecalis* in freshwater environments

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The rapid rate of emergence of clinically relevant multi-resistant pathogenic bacteria has triggered investigations into L their potential environmental sources and feedback pathways into hospitals and homes. As reservoirs for antimicrobial residues and antimicrobial resistant bacteria, aquatic environments are likely hotspots for horizontal transfer of antimicrobial resistance. Aquatic invertebrates may facilitate bacterial aggregation and horizontal gene transfer through biofiltration but the extent to which this contributes to the ecological evolution of antimicrobial resistance remains unknown. Therefore, this research investigated the propensity of the zooplankton Daphnia to facilitate the transfer of vancomycin resistance between vancomycin-resistant donor and rifampicin-resistant recipient strains of Enterococcus faecalis. Microcosm experiments exposed filter-feeders to donor and recipient E. faecalis strains, and transconjugants were detected on double-selection agar plates. Donor and recipient E. faecalis strains were first fed simultaneously to Daphnia, followed by a phase of gut evacuation by feeding Daphnia with the recipient strain only. Transconjugant were recovered from all treatments in which Daphnia was simultaneously fed with one particular donor strain and different recipient strains. There was no significant difference between transconjugant numbers obtained from the feeding phase and the gut evacuation phase. Vancomycin resistance transfer efficiencies for the two Daphnia species were not significantly different. Experimental results showed that aquatic invertebrates can facilitate the emergence of multi resistant Enterococcus faecalis. The recovery of transconjugants in Daphnia trails after gut evacuation appears to be the first direct evidence in support of the hypothesis that filter-feeding can facilitate horizontal vancomycin resistance transfer in aquatic environments.

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