

Oral Communication 3

Bacteriocin dynamics in *Enterococcus faecium* and *Enterococcus lactis*: implications for clinical and commensal strain interactions

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Abstract

Background: *Enterococcus faecium*-Efm and *E. lactis*-Elts (former Efm-cladeB) colonize the human gut, with Efm also being a leading hospital-pathogen. Dynamics influencing strain dominance in competitive environments (e.g., infection/colonization) are not fully understood, but bacteriocins may provide competitive advantage to clinical Efm or commensal Elts strains. **Objective:** We explored bacteriocin content of contemporary Efm and Elts, isolated from healthy/diseased humans, and correlated it with their inhibition profiles against strains across these species. **Methods:** A collection of 129 strains [91 clinical-Efm (77 vancomycin-resistant-VRE); 35 healthy-volunteers (21-Efm;14-Elts)] from 1996-2022 were challenged against each other by a qualitative bacteriocin production/sensitivity-assay (soft-agar-overlay-technique). Eighty-eight representatives were sequenced (Illumina-NovaSeq) to establish clonality, antibiotic profiles (CGE-tool), and bacteriocins (homemade-database) [1]. **Results:** Elts (93%) and Efm (87%) carried ≥ 1 bacteriocin. Twenty-one bacteriocins were found, including 8 newly identified. Efm exhibited greater diversity (1-9; $\bar{x}$ =3.6 vs 1-5; $\bar{x}$ =2.6) and both species presented exclusive bacteriocin genes (Efm:*bac43/AS5/AS9/entA/B/entB*; Elts:*entL50A/B/GM-1*). Bacteriocins 43/AS5/AS11/AS9/entA were significantly associated with clinical-Efm-strains ($p<0.05$), whereas AS8/bac32/entQ/AS4/entL50A/B/GM-1 were exclusive to commensal ones. All were susceptible to inhibition, while 53% of Elts and 65% of Efm (clinical-50%; commensal-39%) inhibited ≥ 1 strain. Those unable to inhibit others were mostly recovered < 2007 or lacked *bac43*. More bacteriocin genes correlated with less inhibition, and similar profiles resulted in comparable inhibition patterns. Among clinical isolates, ST117, ST78 and ST80 showed a higher inhibitory spectrum. ST78-related strains, particularly ST117, demonstrated activity against ST18-related strains previously dominant in Portuguese hospitals, but not vice-versa. VRE were inhibited by 26% of commensal-strains (Efm/Elts with diverse profiles/STs), while inhibiting up to 85% of them. **Conclusions:** Distinct bacteriocin profiles in clinical/commensal isolates, coupled with strain-specific and/or mutual strain inhibition dynamics, suggest a competitive landscape for Efm. Commensal strains inhibited VRE, showcasing their potential to counteract resistant strains. This delicate balance, influenced by unknown factors, underscores the valuable insights bacteriocins could provide for future eco-evo strategies combating human infections caused by Efm.

Keywords: bacteriocins; competitive interactions; vancomycin-resistant *Enterococcus faecium*; *Enterococcus lactis*

Acknowledgments

This work received financial support from national funds (FCT/MCTES, Fundação para a Ciência e Tecnologia and Ministério da Ciência, Tecnologia e Ensino Superior) through the exploratory project EXPL/SAU-INF/0261/2021 and UIDP/04378/2020, UIDB/04378/2020 and i4HB research project LA/P/0140/2020. Ana C. Almeida-Santos acknowledges her UI/BD/151317/2021 fellowship from FCT/MCTES through national funds.

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