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Comparative Analysis of *Enterococcus* spp. from Conventional and Organic Farms: Impact of Antibiotic Use on AMR Profiles

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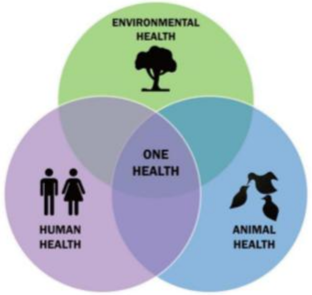
Background

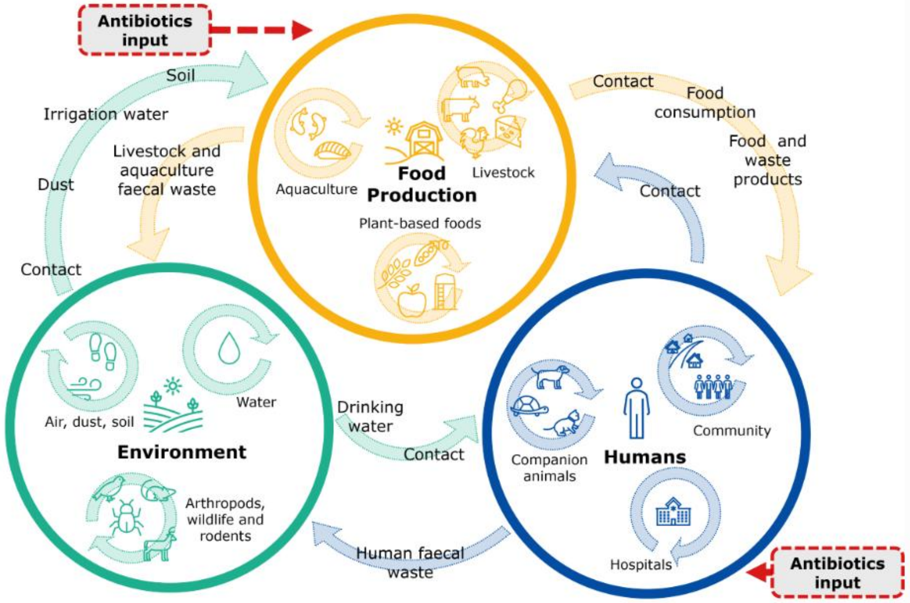
Antimicrobial resistance (AMR) emergence

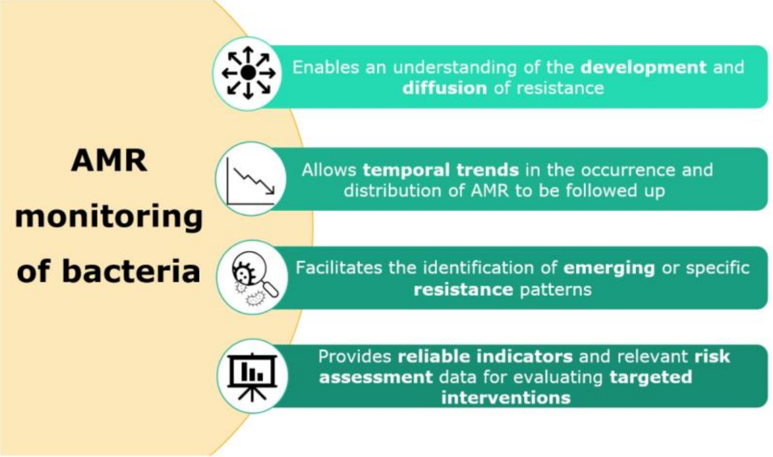
- Significant concern for human and animal health
- Common infection harder to treat
- Spread of AMR bacteria and determinants across settings
- European One Health Action Plan (2017)



Enterococcus spp.

- Hospital-acquired infection (*E. faecium*, *E. faecalis)*
- Gastrointestinal tract commensals
- Adaptability and ability to acquire and transfer resistance genes
- Indicators bacteria in AMR surveillance





AMR monitoring of bacteria

Enables an understanding of the **development** and **diffusion** of resistance

Allows **temporal trends** in the occurrence and distribution of AMR to be followed up

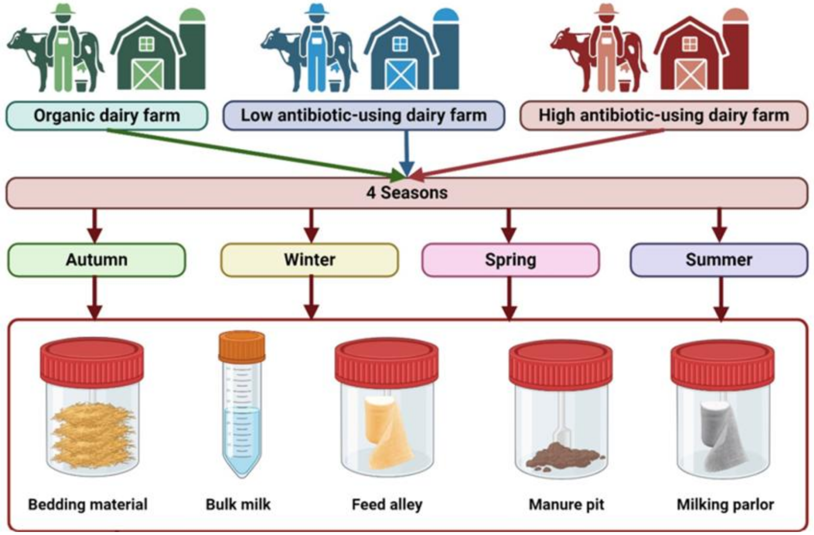
Facilitates the identification of **emerging** or specific **resistance** patterns

Provides **reliable indicators** and relevant **risk assessment** data for evaluating **targeted interventions**

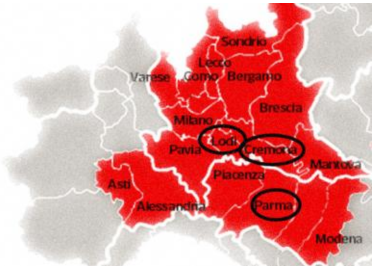
García-Solache M, Rice LB. Clin Microbiol Rev. 2019;32. https://doi.org/10.1128/CMR.00058-18

Objective and Experimental procedure

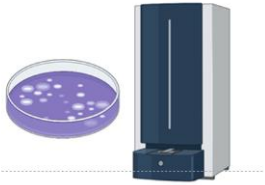
Investigate and compare the diffusion and the AMR profiles of *Enterococcus* spp. from dairy farms with different Defined Daily Dose usage: LOW, HIGH and ORGANIC (no use)



1. Sampling (2023-2024) from:
    - environmental sources
    - milk

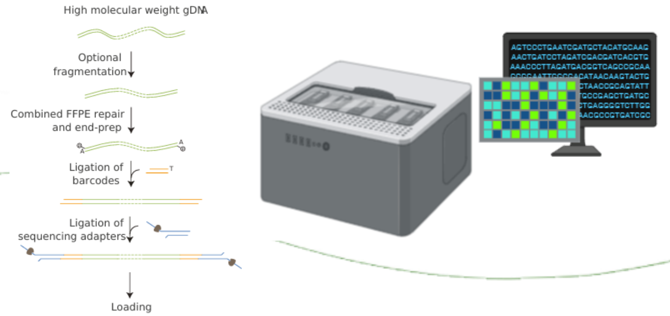


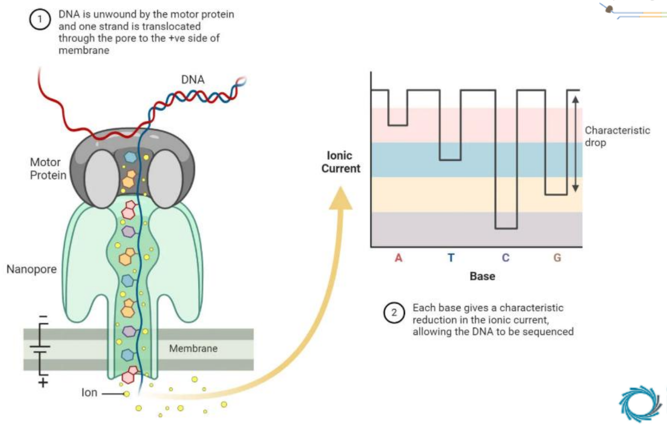
2. Isolation and identification with MALDI-TOF



3. DNA extraction

4. Library preparation and sequencing



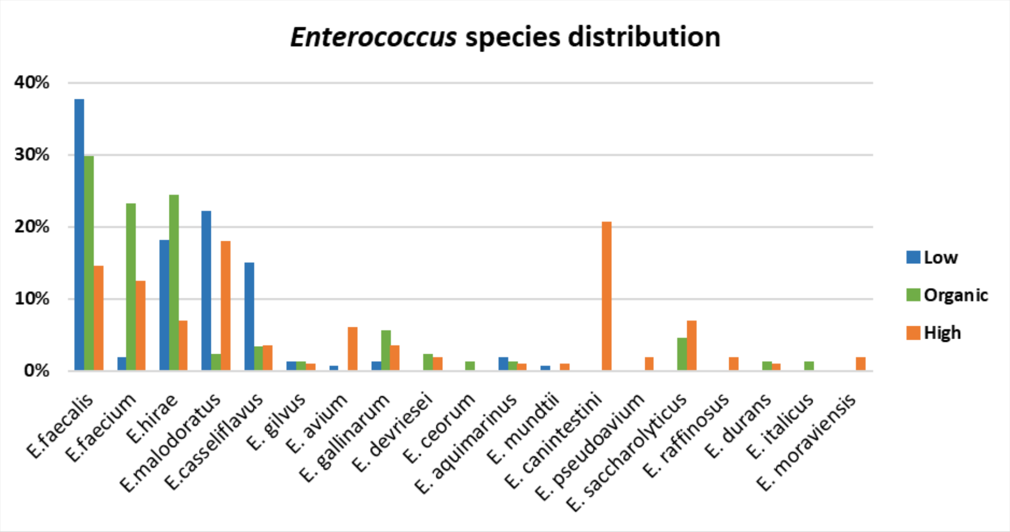


ITINERIS platform

Oxford NANOPORE Technologies

Preliminary results and work in progress

Species identified:
E. coli, *Klebsiella spp* and ***Enterococcus spp***



- ***E. faecalis***, ***E. faecium*** and ***E. hirae*** as potential indicators of farms with low or organic antibiotic use
- ***E. malodoratus*** and ***E. canintestini*** were associated with farms using antibiotics

- ***E. hirae*** the most widespread in environmental samples in all farms
- 28 sample of *E. hirae* were sequenced

Genome assembly results:
AAC(6')-Ia and related *AACs* genes in **90%** of isolates

Resistance to aminoglycosides (streptomycin, gentamicin, tobramycin)

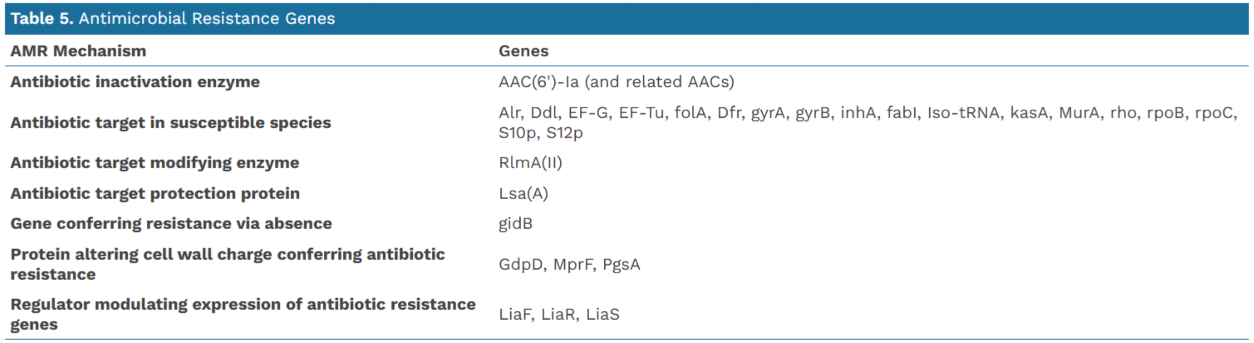


Table 5. Antimicrobial Resistance Genes

AMR Mechanism	**Genes**
Antibiotic inactivation enzyme	AAC(6')-Ia (and related AACs)
Antibiotic target in susceptible species	Alr, Ddl, EF-G, EF-Tu, folA, Dfr, gyrA, gyrB, inhA, fabI, Iso-tRNA, kasA, MurA, rho, rpoB, rpoC, S10p, S12p
Antibiotic target modifying enzyme	RlmA(II)
Antibiotic target protection protein	Lsa(A)
Gene conferring resistance via absence	gidB
Protein altering cell wall charge conferring antibiotic resistance	GdpD, MprF, PgsA
Regulator modulating expression of antibiotic resistance genes	LiaF, LiaR, LiaS

Sequencing and data processing in progress:

E. faecium/ E.faecalis/ E. hirae
E. malodoratus/ E. canintestini
from different environmental and milk sources

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THANKS!

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