Evaluating sampling methods for the detection of multidrug-resistant Enterobacterales in Nursing Home Wastewater

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Introduction

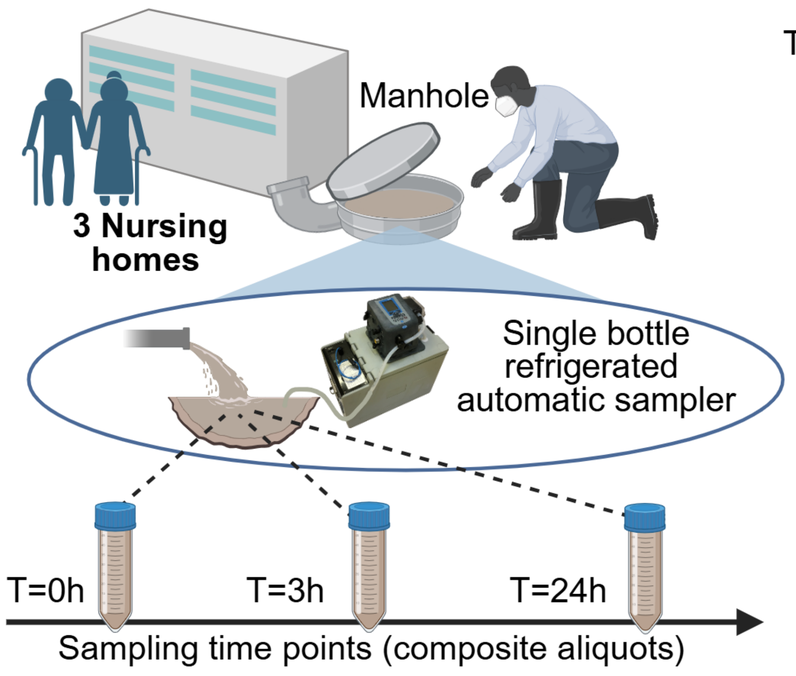
Multidrug-resistant Enterobacterales (MRE), including extended-spectrum beta-lactamase-producing Enterobacterales (ESBL-E) and carbapenemase-producing Enterobacterales (CPE), are major public health threats. Nursing homes (NH) represent important reservoirs of antimicrobial resistance and wastewater-based surveillance could provide a non-invasive population-level monitoring tool, but optimal sampling strategies duration remain unclear.

Objectives

- To develop a practical wastewater-based surveillance protocol to detect MRE in nursing homes.
- To compare sampling duration effect on detection and quantification of MRE.

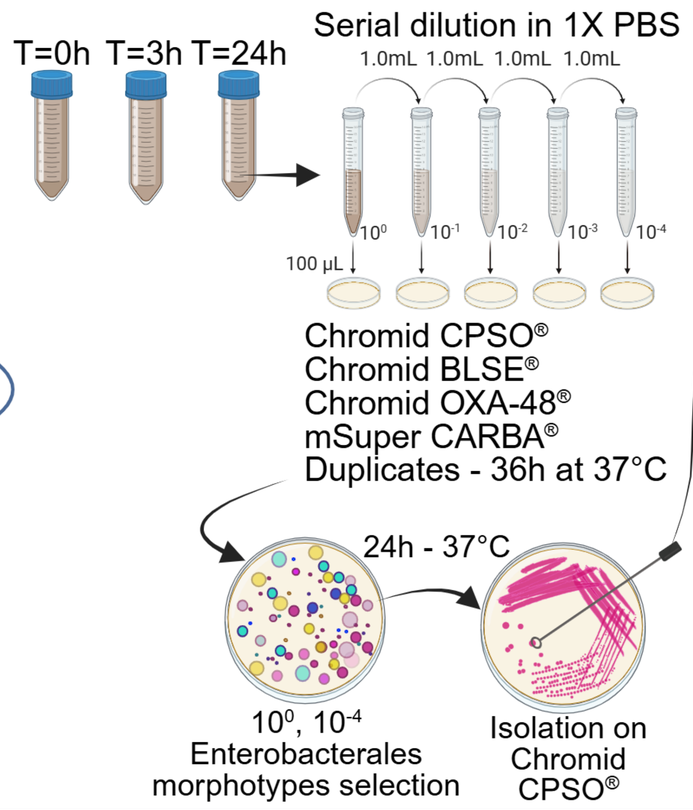
Materials and Methods

Wastewater sampling

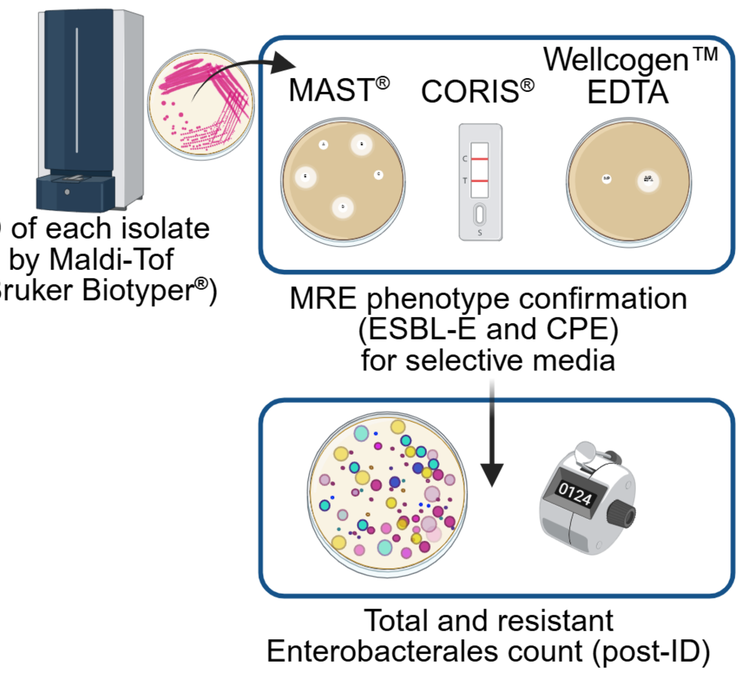


2 sampling campaigns one week apart, per NH
Frequency: 20mL / 10 min
Begining at 9:00 a.m

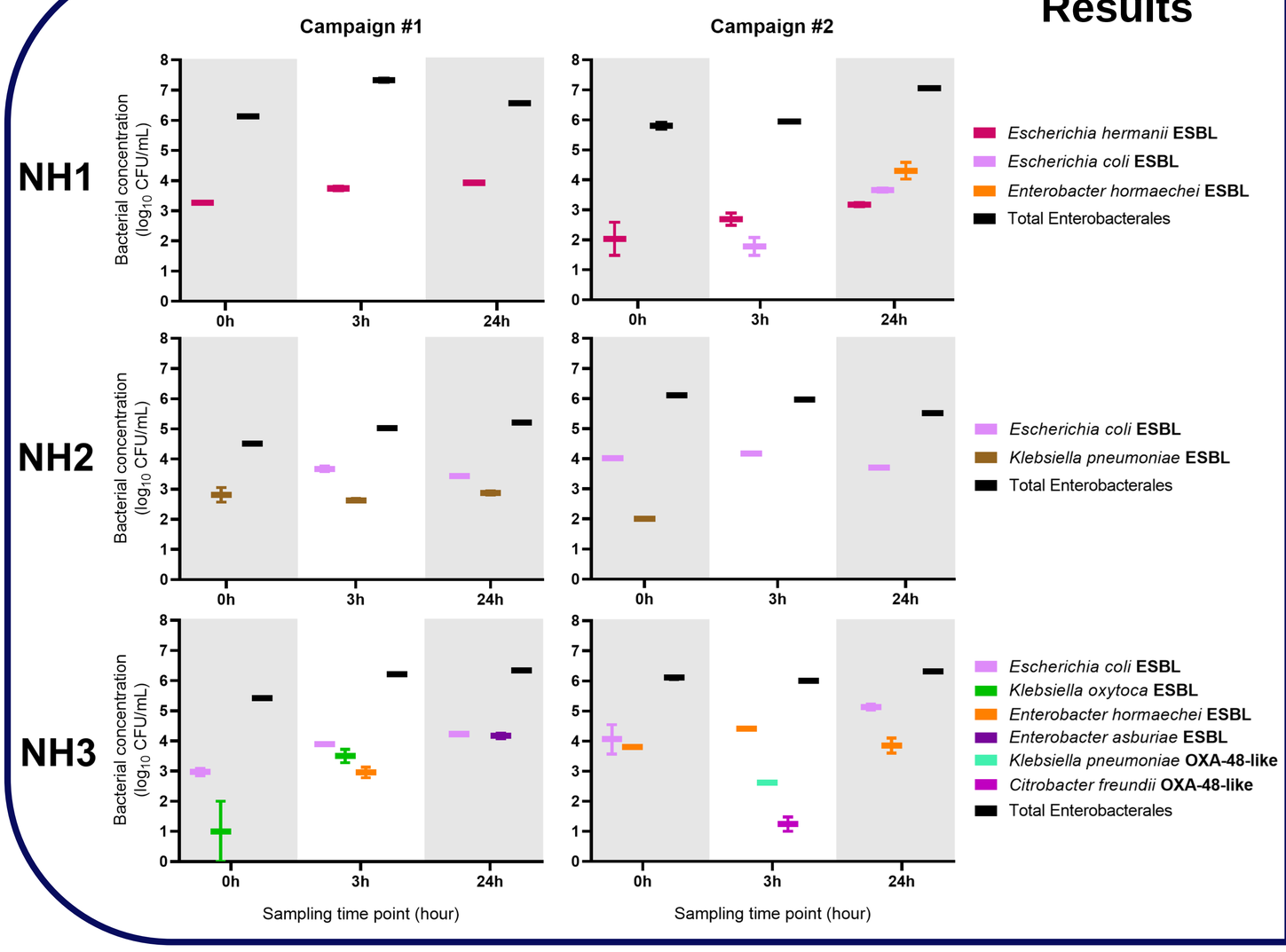
Wastewater dilution and isolation

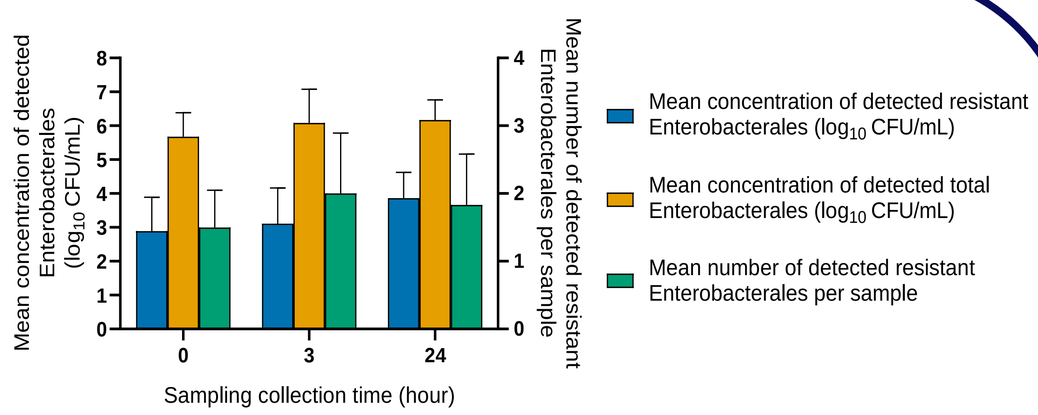


Identification and resistant phenotypes confirmation



Results





All wastewater samples were MRE-positive (18/18)

- ESBL-E were ubiquitous across samples
- *E. coli* predominated (67%), followed by *Enterobacter* spp. (28%) and *K. pneumoniae* (22%)
- CPE were rarely detected (2 OXA-48-like in a single sample)

3-hour sampling frequently detected equal or greater isolates diversity

- Higher isolates yield at T=3h (2.0 isolates/sample) vs T=0h (1.5) and T=24h (1.83)

→ 3-hour composite sampling provides the best balance between performance and feasibility

Discussion

This study suggests that ESBL-E are endemic in French NH wastewater. Three-hour sampling is a compromise between operational feasibility and performance for detecting one-off events, although interpretations remain limited without strain-level molecular typing. CPE may be underdetected in NH, as two strains were identified without known colonised residents in the facility. However, the correlation between wastewater findings and resident colonisation remains to be established.

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