Assessment of patient, drain, and hospital wastewater connections in the surveillance of multidrug-resistant Enterobacterales epidemiology

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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 hospital threats associated with infections and limited treatment options. While aquatic environments and wastewater are suspected to contribute to dissemination, their potential as a routine surveillance tool to reflect patient epidemiology remains uncertain.

Objective

To assess the relationships between patient carriage, contamination of sink and shower drains, and hospital wastewater microbiology in the context of ESBL-E and CPE surveillance.

Materials and Methods

University Hospital of Angers

Site 1: Medical ICU, general surgery

Site 2: Surgical ICU, Orthopaedics, Urology

Patients

	Site 1	Site 2
Patients	**34**	**94**
ESBL-E Screening	12	25
CPE Screening	12	30

Patients in ICU were weekly sampled for CPE and ESBL-E carriage

Shower and sink drains

	Site 1	Site 2
Rooms	**23**	**62**
Showers CPE swabbing	15	40
Sinks CPE swabbing	23	62

ESBL swabbing was not performed for sink and shower drains

Wastewater

	Site 1	Site 2
Wastewater site	**Manhole 1**	**Manhole 2**
Sampling per site (one week-appart)	2	2

eSwab® and clinical samples

Internal drains eSwab®

100µL of serial dilution in 9mL of 1X PBS

10^{0} to 10^{-4}

20-hour refrigerated composite sampling
Frequency: 20mL / 10min

Samples	Chromid CarbaSmart®	Chromid ESBL®	Chromid OXA-48®	mSuper Carba™	Chromid CPSO®
Patients	×	×			
Sinks and Showers	×				
Wastewater		×	×	×	×

Agar culture media

Enterobacterales count (wastewater)

Enterobacterales isolation on Chromid CPSO®

Identification by MALDI-TOF-MS Bruker Biotyper®

Disk diffusion test

CORIS®

MRE phenotype confirmation (ESBL-E and CPE) for selective media

Results

Patients and showers/sinks

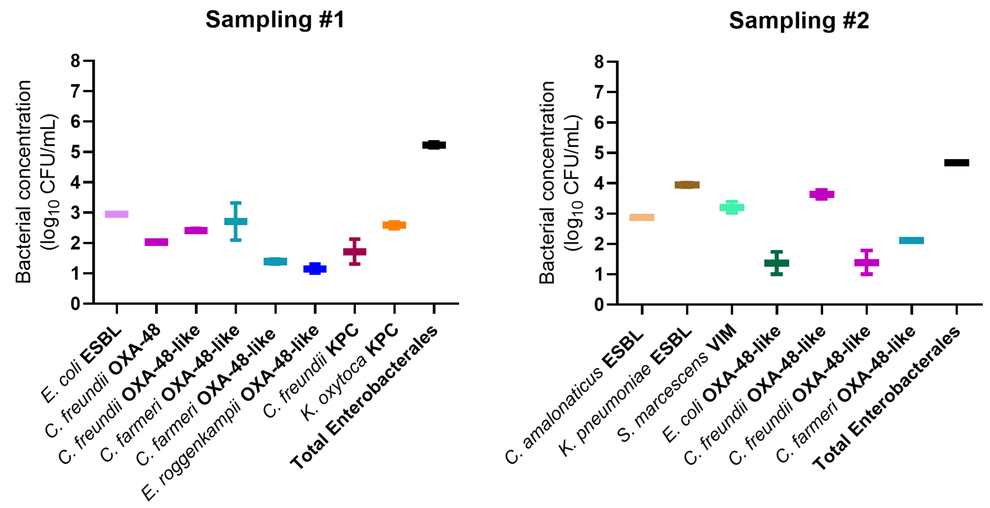
	Site 1	Site 2
ESBLE-E positive patients	**2 / 12 (17%)** 1 *E. coli* ESBL *1 K. oxytoca* ESBL	**7 / 25 (28%)** 3 *E. coli* ESBL 3 *Enterobacter* spp. ESBL 1 *K. pneumoniae* ESBL
CPE positive patients	**0 / 12 (0%)**	**1 / 30 (3.3%)** 1 *Enterobacter* spp. VIM
CPE-positive showers	**0 / 15 (0%)**	**12 / 40 (30%)** *Enterobacter* spp. (6 NDM, 2 VIM) *K. pneumoniae* (3 VIM, 1 OXA-48-like)
CPE-positive sinks	**0 / 23 (0%)**	**0 / 62 (0%)**

Environmental CPE contamination was restricted to showers

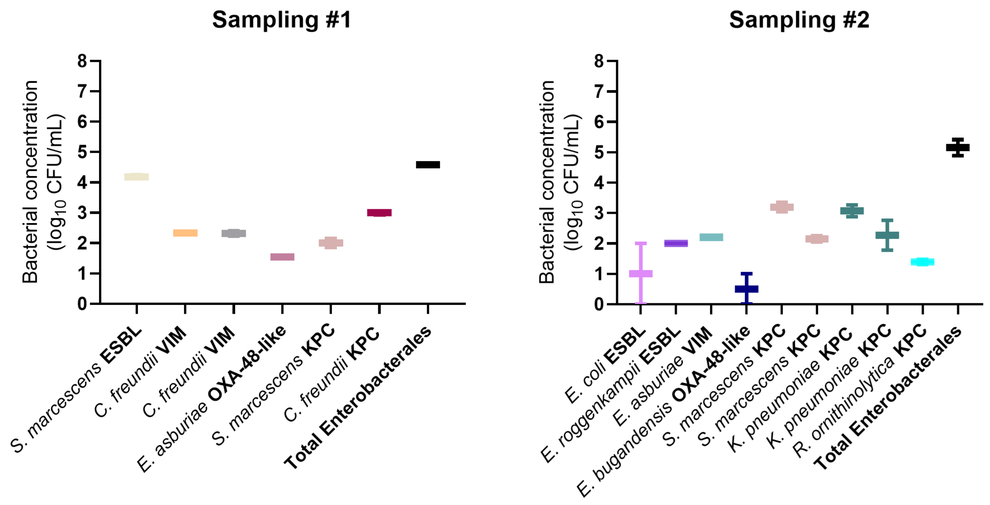
Same species-phenotype isolates found according to patients

	Site 1	Site 2
Patients and wastewater	*E. coli* ESBL	*E. coli* ESBL *Enterobacter* spp. VIM
Showers and wastewater	No same isolates	*Enterobacter* spp. VIM
Sinks and wastewater	No same isolates	No same isolates

Wastewater from site 1



Wastewater from site 2



All wastewater samples were MRE-positive (4/4 samplings)

CPE dominated wastewater isolates (80%, 24/30)

20% of ESBL-E isolates (6/30)

OXA-48-like were the most common (37%), followed by KPC (30%) and VIM (13%)

Limited concordance between wastewater and clinical/environmental isolates

Discussion

The presence of ESBL-producing *E.coli* and VIM-producing *Enterobacter* spp. suggests a possible connection between patient carriage and wastewater detection, although overall concordance remained limited. Interpretation is limited by incomplete patient screening and the absence of strain-level molecular typing. This study suggests that wastewater reflects more the manholes' microbiome than patient epidemiology, limiting its usefulness for routine surveillance.

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