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► To cite this version:

Alisson Godino Sanchez, Michèle Gourmelon, Melody Lebrun, Alexandre Mrozinski, Joelle Serghine, et al.. Bacterial antimicrobial resistance in coastal waters of two contrasting temperate environments Authors. *WaterMicro* 2025, Jun 2025, Amersfoort, Netherlands. <hal-05588051>

HAL Id: hal-05588051

<https://hal.science/hal-05588051v1>

Submitted on 10 Apr 2026

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Bacterial antimicrobial resistance in coastal waters of two contrasting temperate environments

Authors

Alisson Godino Sanchez, Michèle Gourmelon, Melody Lebrun, Alexandre Mrozinski, Joelle Serghine, Julien Quéré, Emilie Rabillier, Claire Labry, Sophie Schmidt, Marie Latimier, Florian Caradec, Alexandre Cormier, David Goudenege, Sabine Delannoy, Patrick Fach, Dominique Hervio-Heath, Cyril Noël, Stéphanie Madec, Gwenaëlle Le Blay

Abstract

Introduction

Antimicrobial resistance (AMR) poses critical risks to human and animal health and the environment. However, AMR in coastal environments remains underexplored, even though anthropogenic activities and climate change are increasing the risks of its propagation.

This study aims to assess the presence and dynamics of antibiotic-resistant bacteria (ARB) and antibiotic resistance genes (ARGs) in two sites of Western France and to identify key factors contributing to their occurrence.

Methods

Between January 2023 and June 2024, coastal water samples were collected at two sites: Verveur (upstream Le Faou estuary, Bay of Brest), an anthropised area where shellfish harvesting is prohibited, and Barnenez (downstream Bay of Morlaix), a less anthropised site where harvesting is permitted. At Verveur, water from two main rivers was also sampled at their outlets.

In these samples, bacterial communities were described using V3-V4 16S rRNA gene metabarcoding and the presence of ARGs was investigated by screening 45 ARGs from nine antibiotic families using high-throughput microfluidic qPCR. DNA was extracted (PCI extraction) from water samples filtered at 3 µm (particle-attached bacteria) and then sequentially at 0.2 µm (free-living bacteria). ARGs were also searched for in ARBs, isolated on non-selective media (R2A) and phenotypically characterised using EUCAST protocols, by whole-genome Oxford nanopore long-read sequencing (WGS).

Finally, hydrological and biological parameters (e.g., salinity, temperature, nutrients, suspended particulate matter) and concentrations of *E. coli*, biocides and antibiotics were measured.

Results

The coastal waters of Verveur, influenced by strong hydrodynamics (river discharges, tides), had variable salinity values (0.2–32.2), high nutrient concentrations (nitrate > 100 µM) and *E. coli* concentrations of 2.49 ± 0.51 Log/100 ml. In contrast, those from Barnenez showed less variable salinity values (33.2–34.9), lower nutrient concentrations (nitrate < 21 µM) and lower *E. coli* concentrations (1.39 ± 0.52 Log/100 ml).

The bacterial communities differed significantly between the two sites ($p < 0.001$), with the coastal waters of Verveur dominated by *Flavobacteriaceae*, *Rhodobacteraceae*, and *Nitrincolaceae*, while Barnenez had more *Rubritaleaceae*. The coastal communities of Verveur also differed from those of the main river upstream, with less *Comamonadaceae*, due to higher salinity. However, *Flavobacteriaceae* bacteria were present in both coastal and river waters.

ARBs were isolated more frequently at Verveur than at Barnenez. For example, at Verveur, antibiotic-resistant *Pseudomonas* were isolated, including seven multidrug-resistant (MDR) bacteria. Preliminary results obtained from WGS showed the presence of the *adeF*, *abaQ*, and *soxR* genes in these strains.

Discussion

These results are consistent with the few studies that have investigated the impact of anthropogenic activities on the presence of ARBs and ARGs in the coastal environment.

Conclusion

This study highlights the potential role of coastal environments as reservoirs for AMR, with the isolation of MDRs. One perspective will be to study the coastal environment more generally by comparing the results obtained in waters with those obtained in surface sediments and clams, both sampled and analysed at the same time.

Impact

This study provides relevant data on AMR in the coastal environment, which has been little studied. It remains to be determined whether it can be a source of risks to health.