

MEMBER OF BASQUE RESEARCH & TECHNOLOGY ALLIANCE



## Targeting mercury bioremediation of marine sediments by using *omics* and culture-driven approaches

Laura Alonso-Sáez (1), Blanca Rincón-Tomás (1), Carla Pereira-García (2), Haiyan Hu (3), Pablo Sánchez (4), Carla Pérez-Cruz (1), Mónica Estupiñan (1), Anders Lanzén (1), Elena Hernández-Del Amo (2), Isabel Sanz-Sáez (4), Andrea Garcia Bravo (4), Olga Sánchez (2), Stefan Bertilsson (3), Silvia G. Acinas (4).

<sup>1</sup> AZTI, BRTA, 48395 Sukarrieta, Spain

<sup>2</sup> Universitat Autonoma de Barcelona, 08193 Bellaterra, Spain

<sup>3</sup> Swedish University of Agricultural Sciences, 75007 Uppsala, Sweden

<sup>4</sup> Institut de Ciències del Mar, ICM-CSIC, 08003 Barcelona, Spain



Mercury clean-up system based on bioremediation by marine bacteria



MEMBER OF BASQUE RESEARCH & TECHNOLOGY ALLIANCE

Anders Lanzén



Silvia G. Acinas

Universitat Autònoma de Barcelona Olga Sánchez



E David Amouroux



Ulrich Soltmann



Stefan Bertilsson





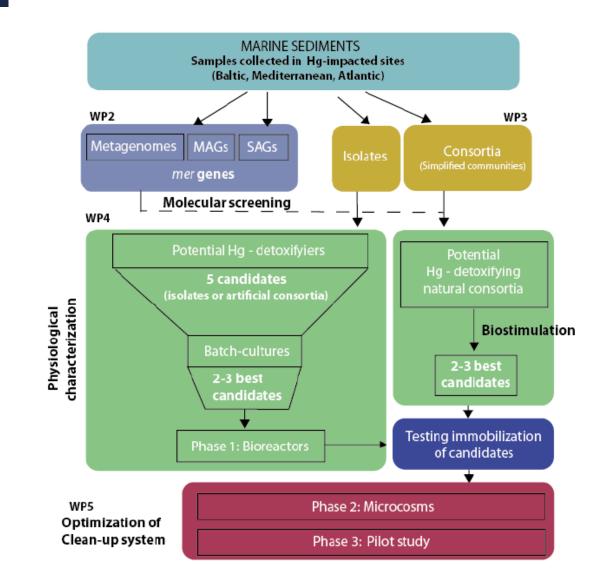
Mercury clean-up system based on bioremediation by marine bacteria The objective of MER-CLUB is to advance our understanding of mercury biogeochemistry in marine sediments and design a novel bioremediation solution for this complex environmental matrix.





Mercury clean-up system based on bioremediation by marine bacteria The objective of MER-CLUB is to advance our understanding of mercury biogeochemistry in marine sediments and design a novel bioremediation solution for this complex environmental matrix.





WHICH BACTERIA ARE INVOLVED IN MERCURY TRANSFORMATION IN MARINE POLLUTED SEDIMENTS?

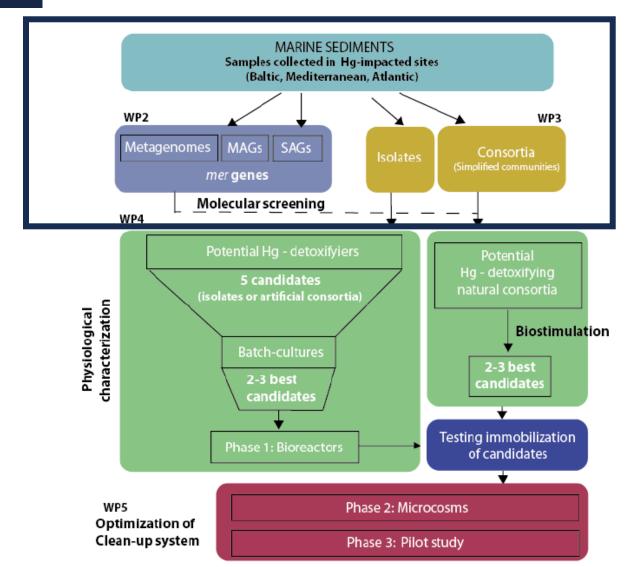
ISOLATION AND PHYSIOLOGICAL CHARACTERIZATION IN LABORATORY CULTURES

**UP-SCALING** 



Mercury clean-up system based on bioremediation by marine bacteria The objective of MER-CLUB is to advance our understanding of mercury biogeochemistry in marine sediments and design a novel bioremediation solution for this complex environmental matrix.



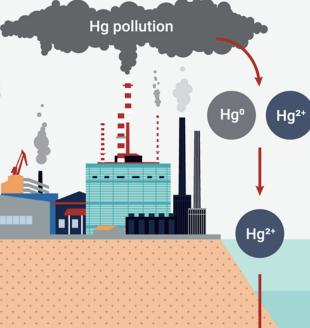


WHICH BACTERIA ARE INVOLVED IN MERCURY TRANSFORMATION IN MARINE POLLUTED SEDIMENTS?

#### Anthropogenic Hg Impact

Hg⁰

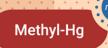
#### Microbial Methyl-Hg Transformation



hgcAB Hg2+ hgcAB

MER-CLUB

Mercury clean-up system based on bioremediation by marine bacteria



Biomagnification

Bioaccumulation

merA Hg<sup>2+</sup> merA

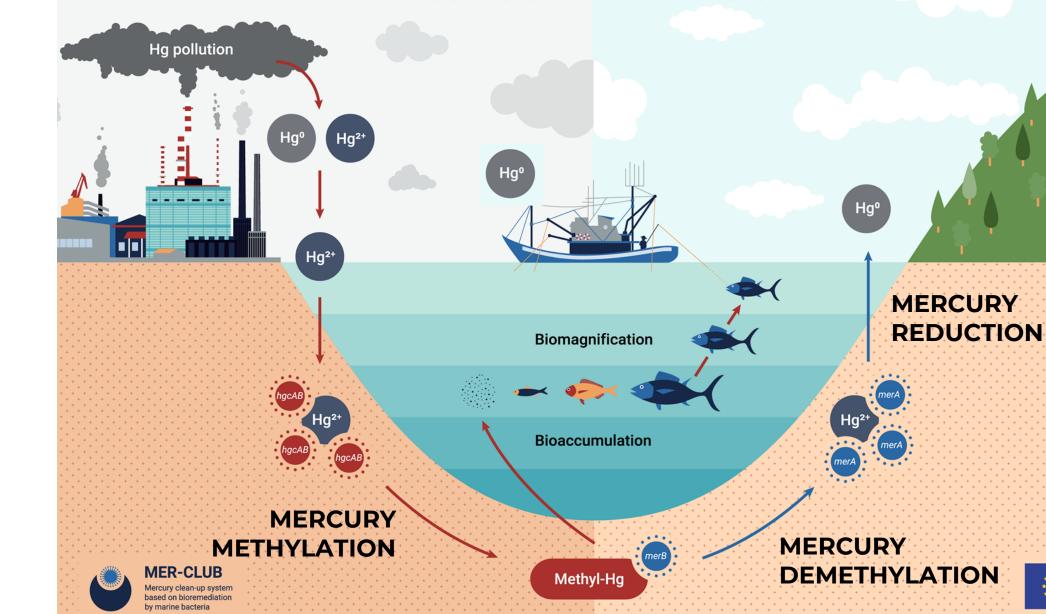
www.mer-club.eu

Hg⁰



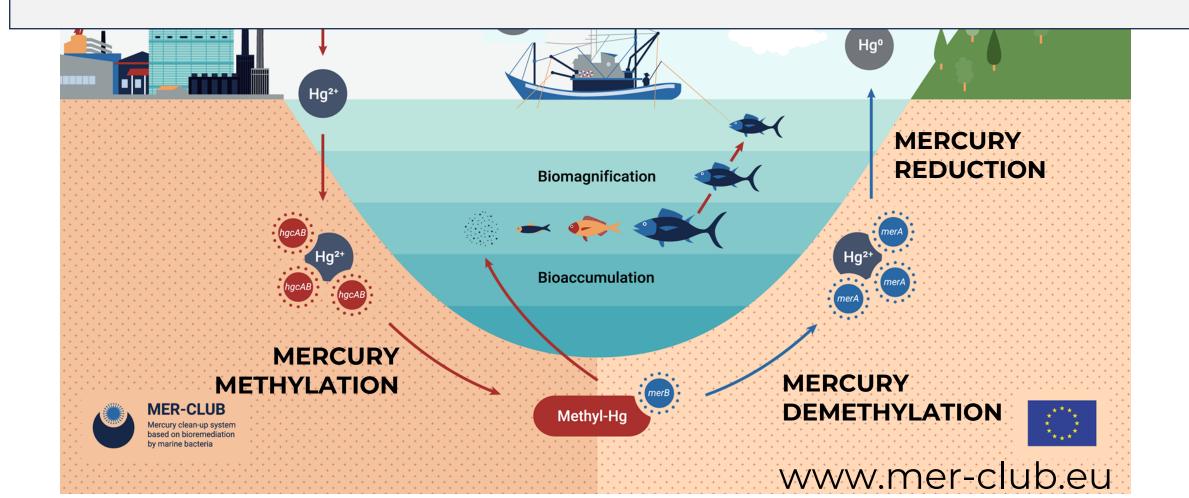
#### Anthropogenic Hg Impact

#### **Microbial Methyl-Hg Transformation**



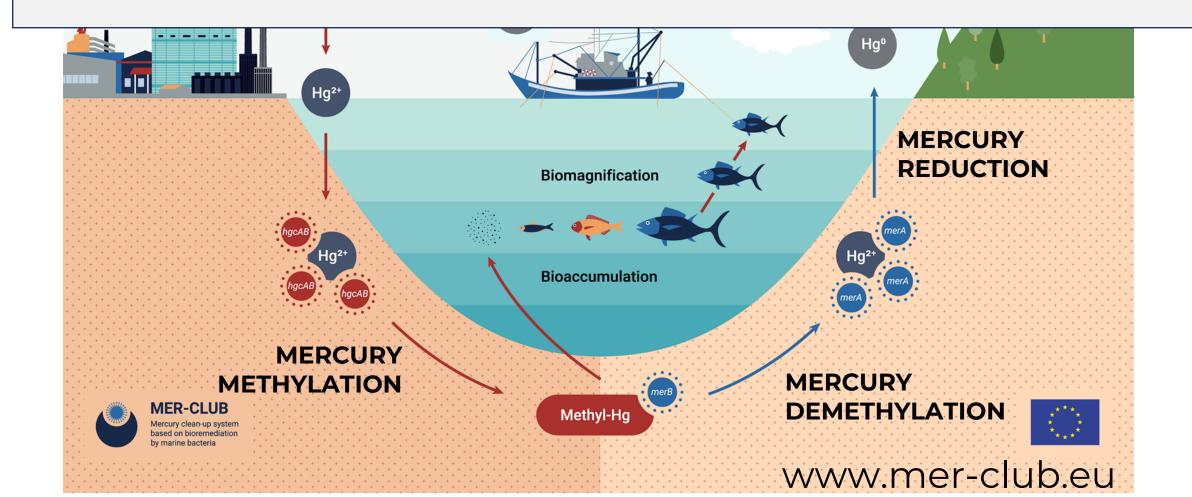
WWW.mer-club.eu

### WHICH BACTERIA ARE INVOLVED IN MERCURY TRANSFORMATIONS IN MARINE POLLUTED SEDIMENTS?



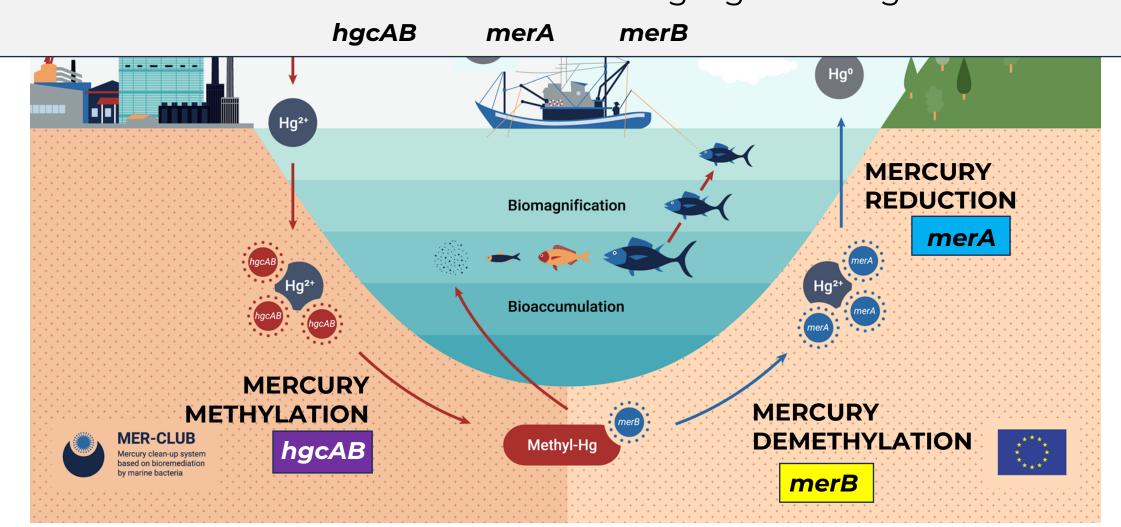
### WHICH BACTERIA ARE INVOLVED IN MERCURY TRANSFORMATIONS IN MARINE POLLUTED SEDIMENTS?

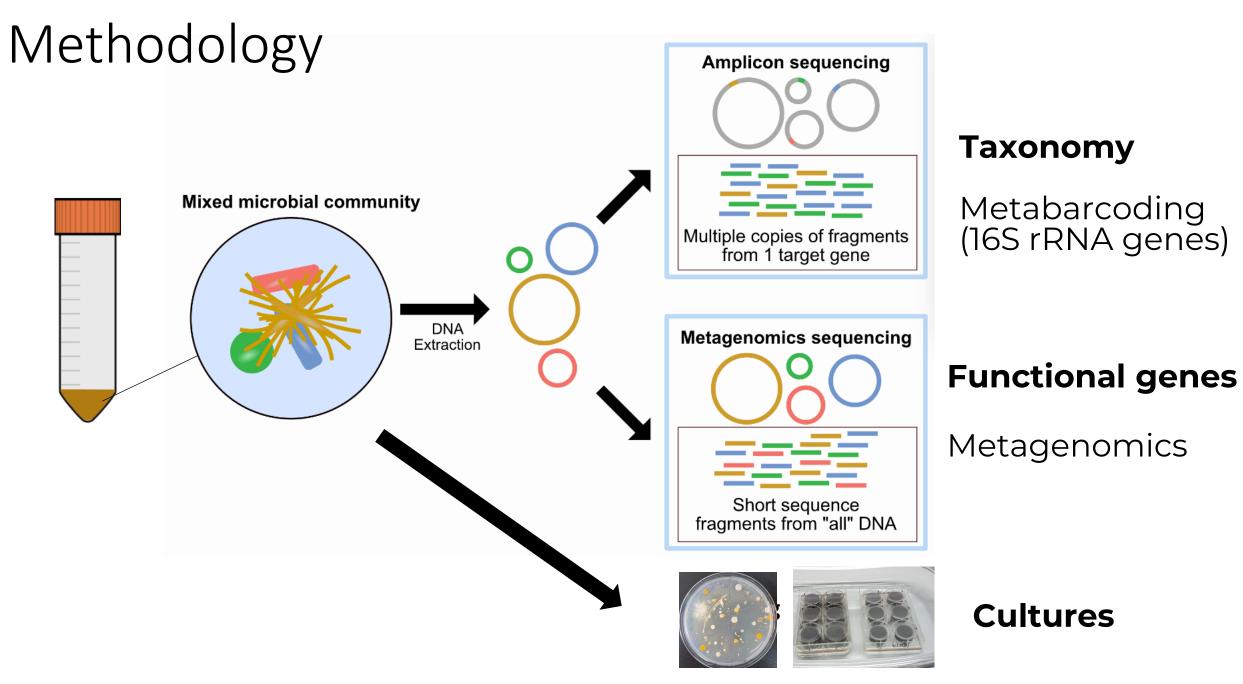
Identification of bioindicators of Hg pollution



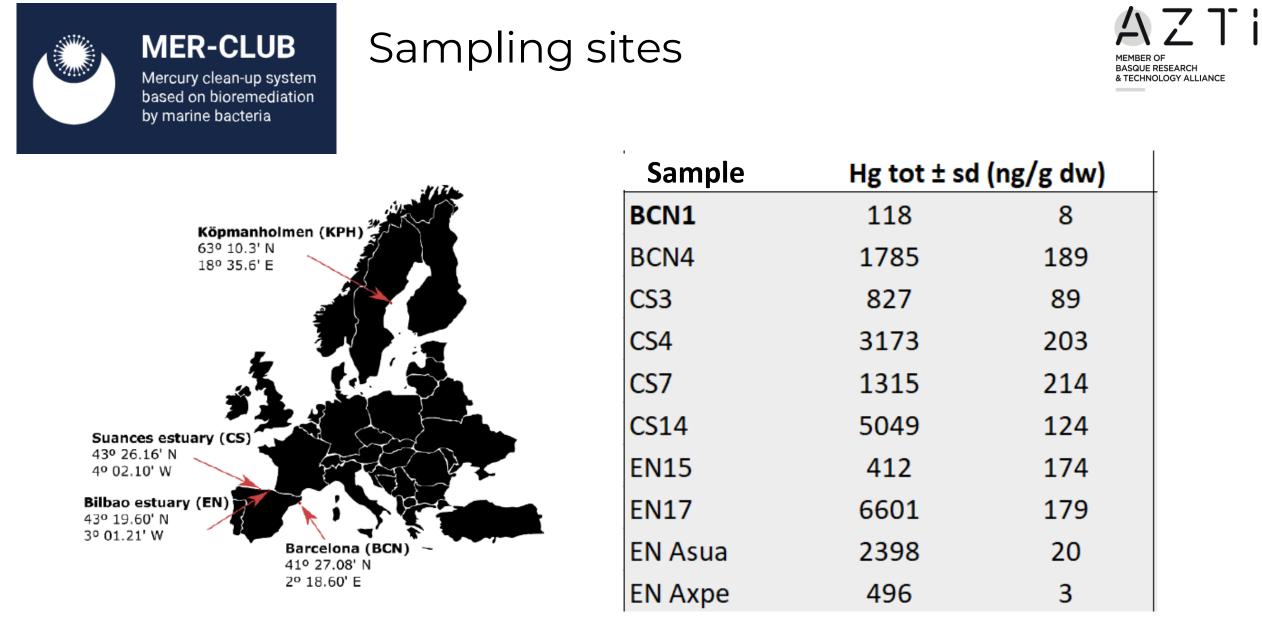
### WHICH BACTERIA ARE INVOLVED IN MERCURY TRANSFORMATIONS IN MARINE POLLUTED SEDIMENTS?

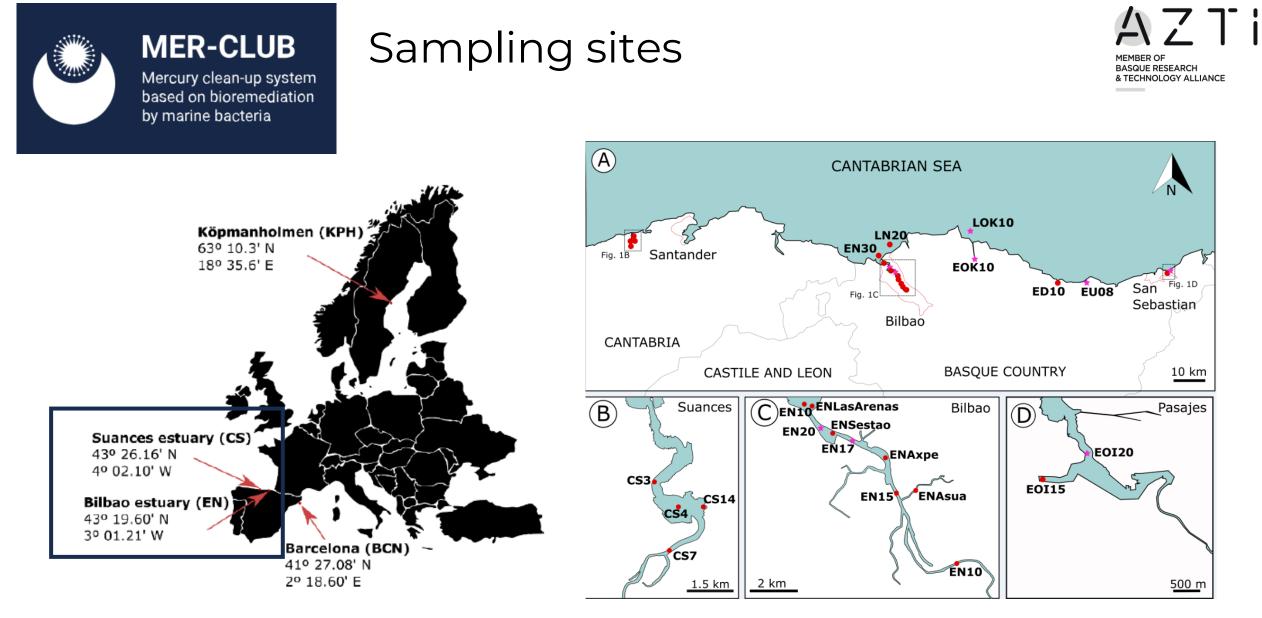
Identification of bioindicators of Hg pollution Identification of bacteria harbouring Hg-related genes



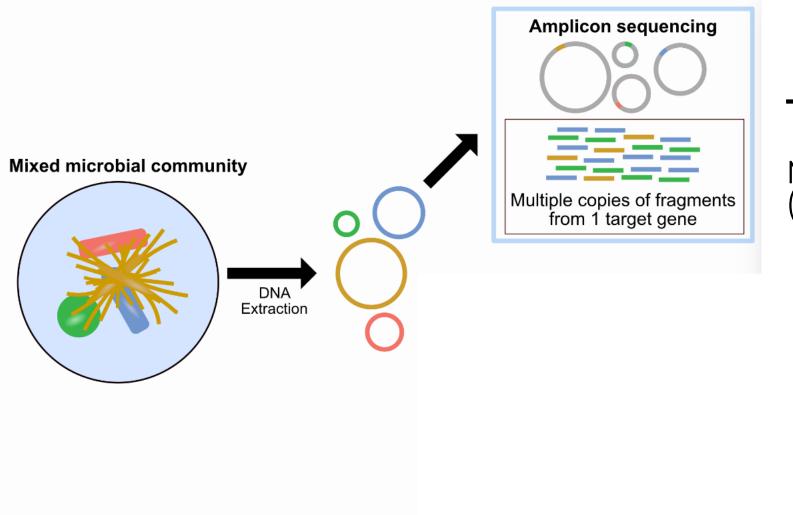


Adapted from https://astrobiomike.github.io/misc/amplicon\_and\_metagen





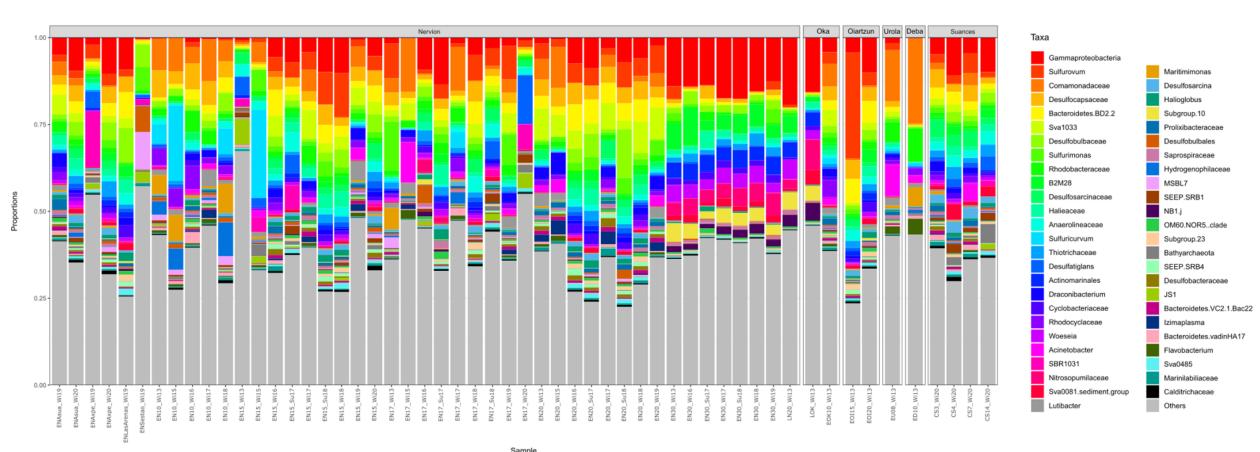
High organic matter content (5-7% dry weight) Anoxic conditions



### Taxonomy

Metabarcoding (16S rRNA genes)

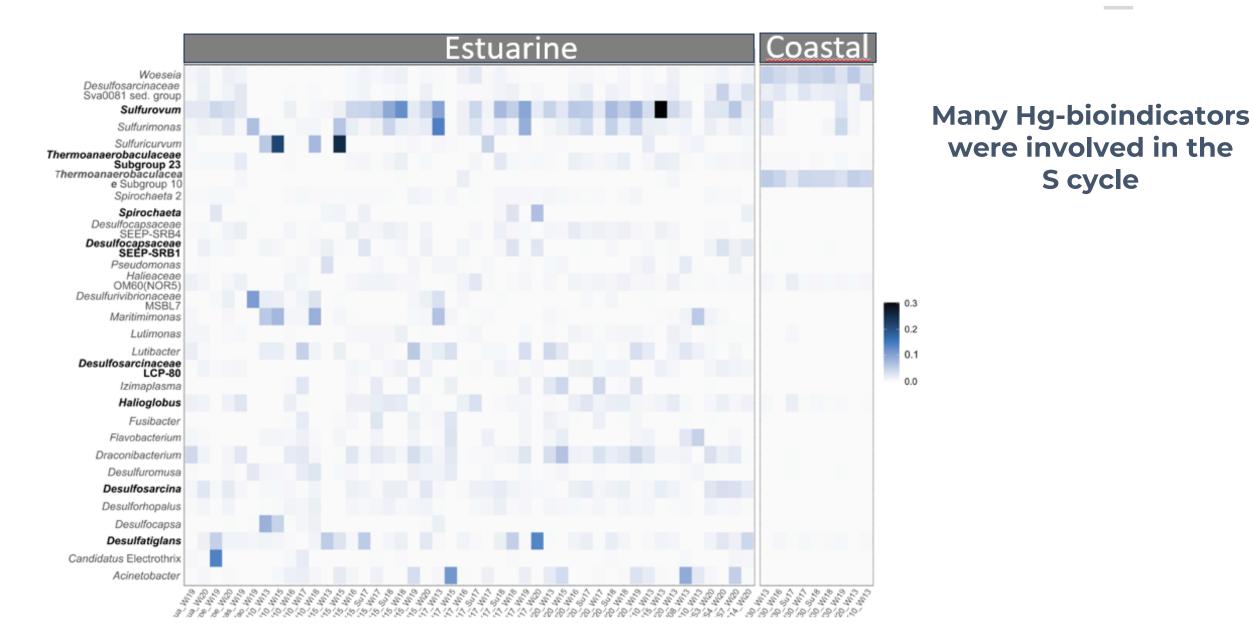
# Taxonomic diversity of sediment bacteria by metabarcoding (16S rRNA genes)



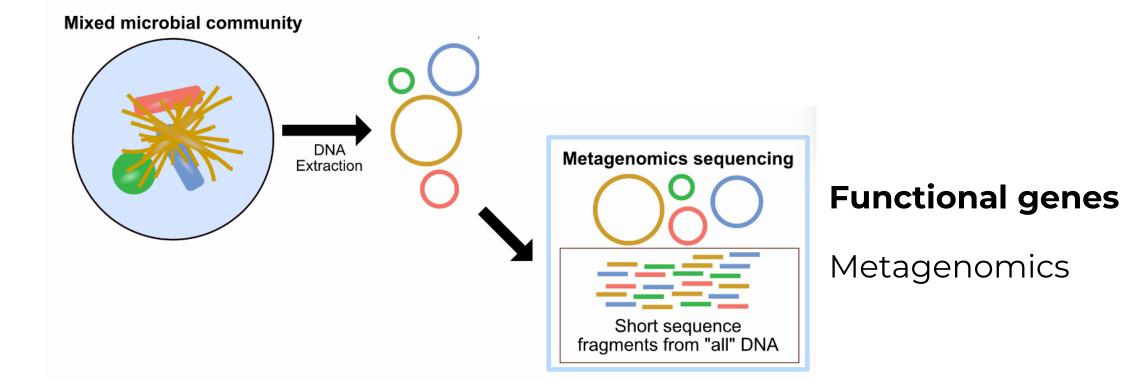
AZII MEMBER OF BASQUE RESEARCH & TECHNOLOGY ALLIANCE Top-abundant taxa in the samples and Hg-bioindicators as identified by TITAN (Threshold Indicator Taxa Analysis, Baker & King 2010)



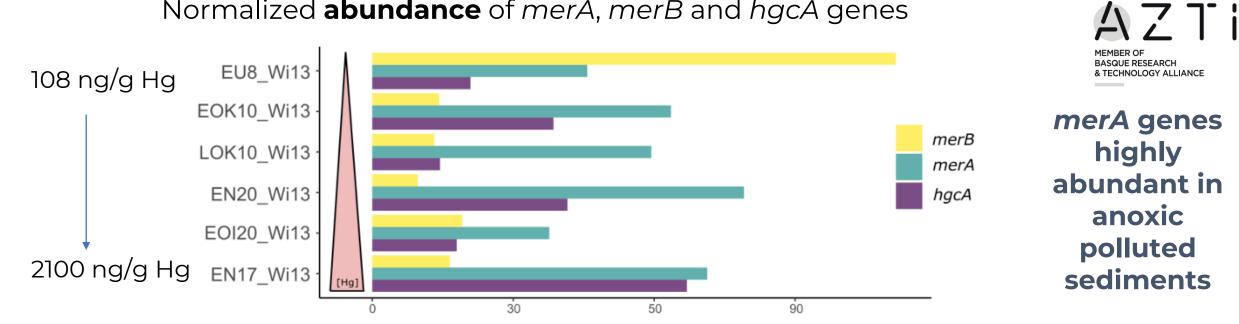
**S** cycle



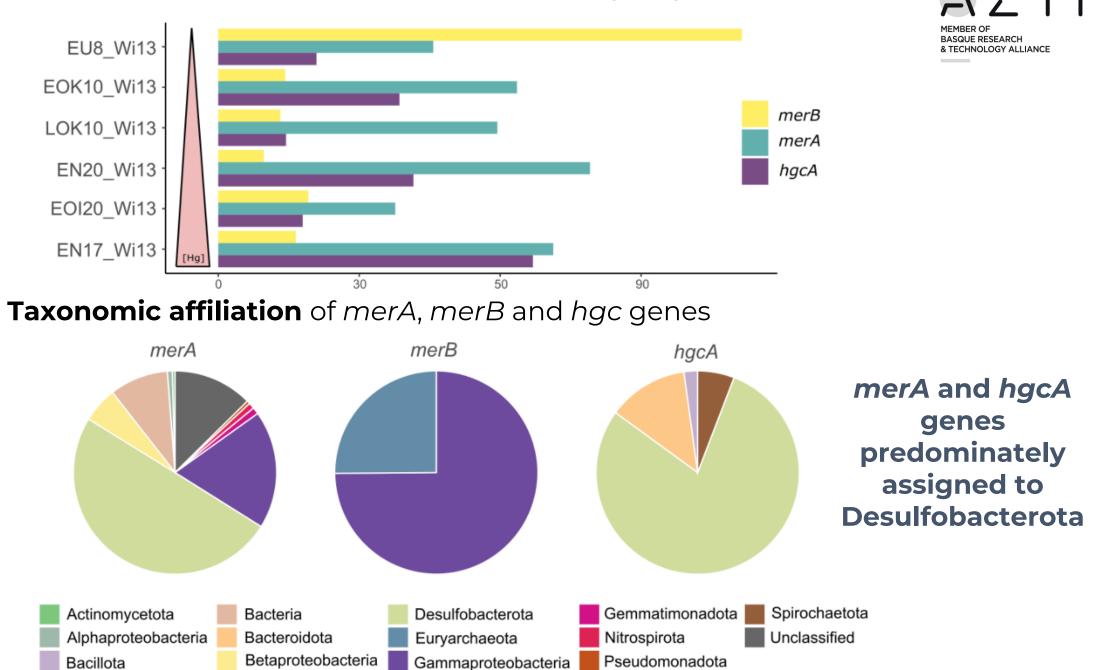
## Methodology



### Normalized **abundance** of *merA*, *merB* and *hgcA* genes

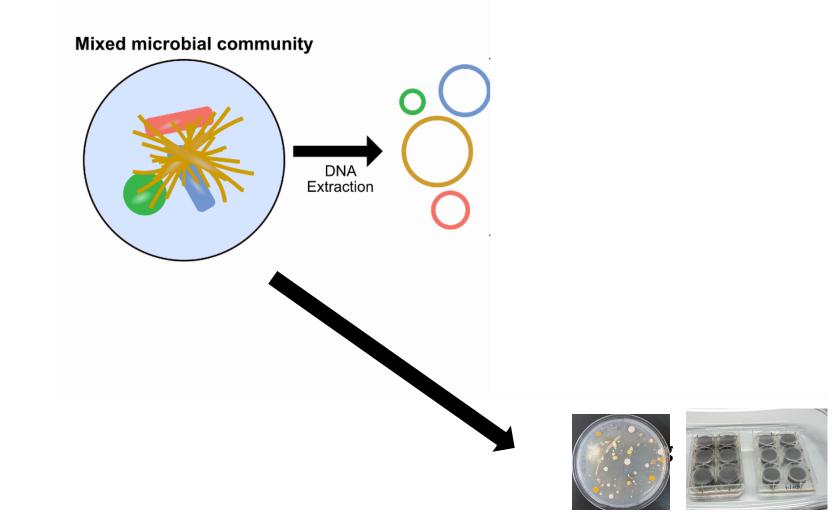


Normalized abundance of *merA*, *merB* and *hgcA* genes



## Which bacteria did we retrieve in culture?

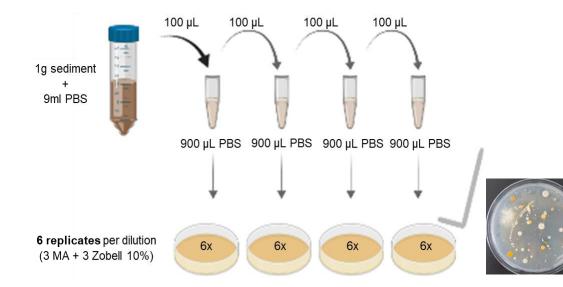
MEMBER OF BASQUE RESEARCH & TECHNOLOGY ALLIANCE



Cultures



### Direct plating in agar plates



### Soil-substrate membrane

### System Rasmussen et al., 2008

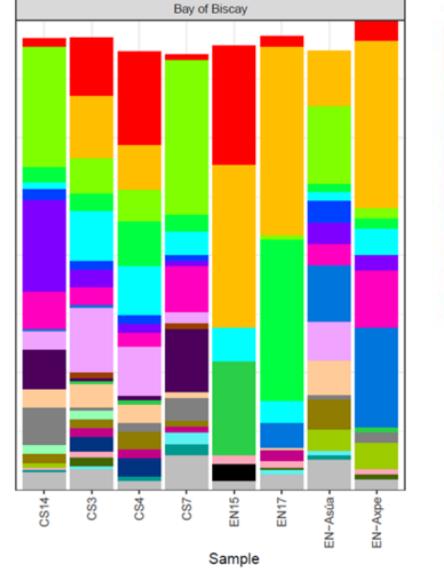




• Culture collection with > 1000 bacterial isolates





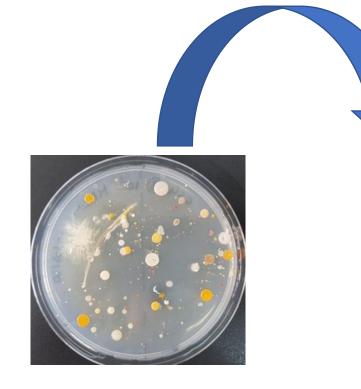




### Most isolates were affiliated with Gamma-proteobacteria



- Culture collection with > 1000 bacterial isolates
- >50 isolates with *merA* genes



PCR screening of merA genes 1002 1003 1007 1073

Thalassospira Pseudomonas Bacillus Shewanella Arthrobacter Vibrio Acinetobacter Aeromonas Pseudoalteromonas Xanthomonas

Marinobacter

### Phylogenetic tree of merA sequences

EFD92905 1 MAG Mera Can-

11486 2 Geopsychrob

EU8.26675 2 llumatobacter nonamiensir

EOK10.26628 2 Desulfobacterales

EN20.45428 8 Desulfobacterales

EU8.27591 3 Desulfobacterales

EOK10.4999 2 Desulfopila sp

EOK10.11513 5 Desulfobacterales

EU8.9905 2 De EN17.1090 EN17.20031 2 Dev

E0120-1 EOK10.1204

2305 5 Desulfoc

EN20.31182 2 unclassified

11664 5 Desulfobacterota EOK10.13632 3 Desulfobacteraceae

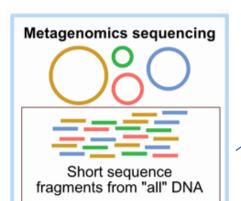
EOK10.15743 2 Desulfobacterales

LOK10.29840 2 Gammaproteobacte

E0120

merA genes from cultures and metagenomes do not overlap

LOK10.





Isolate 1 (av \* marinobacter sp.\* CSA Isolate 1019 4 metrimetrinobacter sp.\*

ENAxpe isoliate 1322 2 Marinobacter sp\* CS3 Isolate 1555 3 Marinobacter sp 🖈

EN17 Isolate 1328 1 Marinobacter sp.\*

EN15 Isolate 1220 2 Neptuniibacter sp.\*

EN17 Isolate 1327 3 Marinobacter sp. 🜟

EN15 Isolate 1210 3 Marinobacter sp. 🙁

EN15 Isolate 1203 1 Marinobacter sp. \*

-EN15 Isolate 1202 1 Marinobacter sp. 🜟

EN15 Isolate 1204 1 Marinobacter sp. 🜟

EN15 Isolate 1208 2 Marinobacter sp.\* EN15 Isolate 1209 1 Marinobacter sp. EN15 Isolate 1200 + Marinobacter sp. \* EN15 Isolate 14 17 Solate 1318 1 Marinobacter sp.

EN15 Isolate 1197 1 Alcanivorax sp

MEMBER OF

BASQUE RESEARCH & TECHNOLOGY ALLIANCE

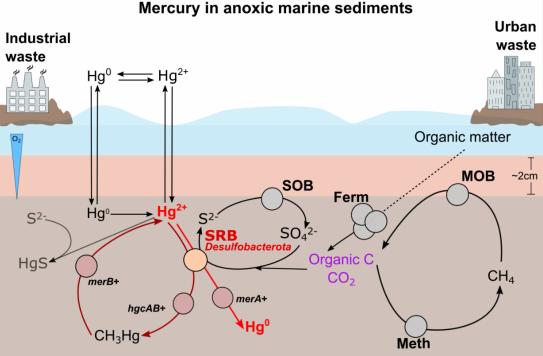
AZTI

### TAKE HOME MESSAGES



The current known diversity of environmental mercury reducers is highly biased by the fact that a large fraction of them originate from oxic culturing approaches

Sulfate-reducers (Desulfobacterota) may have a predominant role in mercury transformations in anoxic sediments, not only as mercury methylators but also as mercury reducers







## Acknowledgements



### MER-CLUB

Mercury clean-up system based on bioremediation by marine bacteria

Co-funded by the European Union

### The MER-CLUB research team!

Project funded by the European Climate, Infrastructure and Environment Executive Agency (CINEA) EMFF programme (Grant agreement 863584)



