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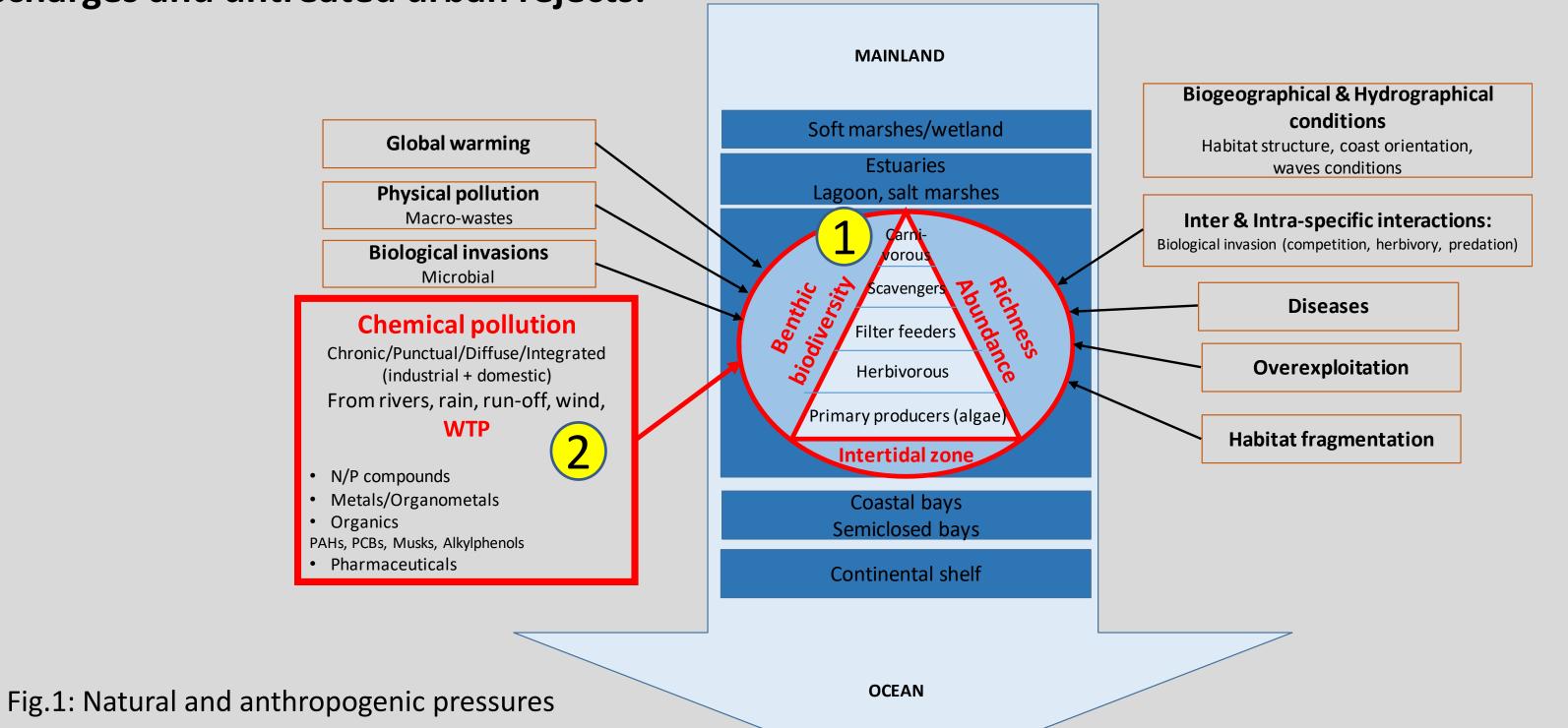
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#### Introduction:

Intertidal rocky shores are exposed to natural and human pressures (fig.1). Current works within the EU Water Framework Directive about the Good Ecological Status (GES) of water bodies show deficiencies on how biological indicators respond to each pressure in particular coastal wastewater treatment plant (WTP) discharges and untreated urban rejects.



In order to identify potential effects of sewage discharges on the benthic diversity of intertidal rocky shores along the Basque coast, this study (1) analyses communities on locations impacted and not impacted to WTP discharges and (2) identify priority and emerging pollutants at the outlet of each WTP.

Spain France PE: 27 500 eq/inhab Flow: 5 930 m<sup>3</sup>/day PE: 58 000 eq/inhab Flow: 8 200 m<sup>3</sup>/day PE: 45 000 eq/inhab Flow: 7 350 m<sup>3</sup>/day © Ortho Littorale V2 - MEDDE - L. Huguenin, UPPA - UPV/EHU, IPREM Intertidal impacted location Intertidal control location

Fig.2: Sampling locations

Methodology: 5 locations (3 impacted and 2 control) divided into 3 sites according to a distance gradient Sampling campaign from March to June 2017

Bakio (Control)

Erromardie (Exposed)

Hendaye (Exposed)

Ondarroa (Exposed)

Socoa (Control)

Locations

Bakio (Control)

Socoa (Control)

+ Control

Erromardie (Exposed)

Hendaye (Exposed)

Ondarroa (Exposed)

## Biological approach

### At each location Biological sampling (n=417 quadrats) Site 1 Site 2 Site 3 Lower mediolittoral zone Area 2 Area 1 Area 2 Area 1 Upper mediolittoral $0.1 \, \mathrm{m}^2$ Fig.3: Sampling design used at each location for biological samples

according to a distance gradient from the outfall (Sites 1, 2,3)

MDS compares the relative similarity of benthic assemblages of each location (fig. 4)

- **Distinctions:**
- between French and Spanish locations (due to habitat structure).
- between "control" and impacted locations in both countries.
- high and low variabilities in impacted locations (Ondarroa and Hendaye –vs– Erromardie). The same occurs in control locations (Socoa –vs– Bakio).
- > Response of benthic assemblages to WTP outfalls is different between locations.
- > It is then more wise to analyze separately these both countries (eg. French control location –vs– French impacted locations) even if these differences have to help considered in analyses.
- Pairwise tests could not identify an effect of distance from outfalls (Kruskalmc; p>0.05) on mean taxonomic richness (mTR) excepted for Erromardie (Site 1 ≠ 3) (French impacted location).
- -> Erromardie: maybe linked to the large size and flow of the WTP (58 000 eq/inhab and 8 200  $m^3$ /day).
- Only one higher significant mTR (Kruskalmc; p<0.05) between control (Socoa) and impacted (Erromardie) locations.
- Spatial variations (but not significant) inside control locations in France and Spain.
- Macroalgae TR > macrofauna TR
- -> competition & other environmental factors explain this unbalanced abundance.
- Red macroalgae TR > other macroalgae

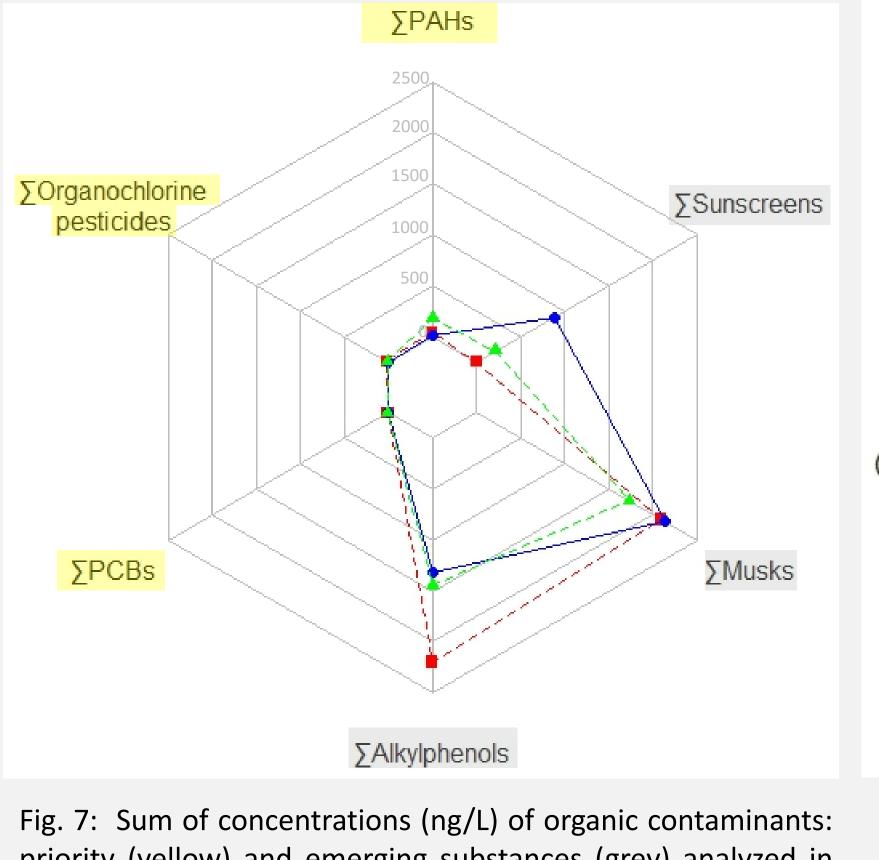
-> contrary to what happens in northern French regions (Ar Gall et al. 2016).

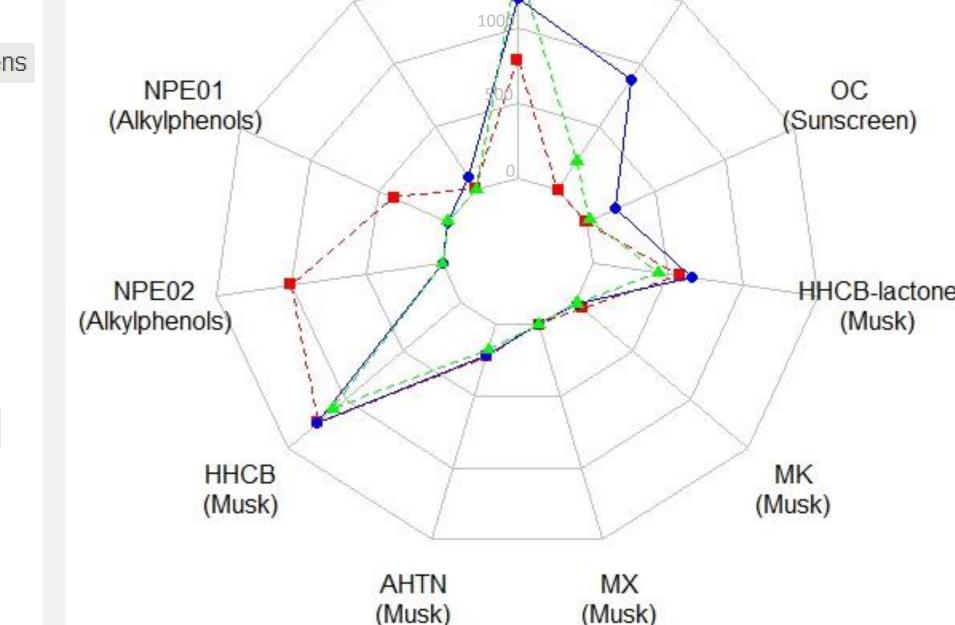
# Chemical approach

### Wastewater sampling

Sampling at the **outlet of the WTP** with glass bottles

Analyze of micropollutants: priority and emerging pollutants (PAHs, PCBs, synthetic musks, Alkylphenols (by GC-MS))





priority (yellow) and emerging substances (grey) analyzed in the outlet of WTP in Hendaye (blue line), Erromardie (dotted green line) and Ondarroa WTP (dotted red line).

Fig. 8: Concentrations (ng/L) of emerging organic contaminants analyzed in the outlet of WTP in Hendaye (blue line), Erromardie (dotted green line) and Ondarroa WTP (dotted red line).

Carbamazepine

(Pharmaceuticals)

(Musk)

- These radar charts allow to characterize the rejects of each WTP (Hendaye, Erromardie and Ondarroa) by presenting the sum of organic micropollutants (fig. 7) and concentrations of each emerging substances (fig. 8).
- No major difference was noticeable between each WTP.
  - Major micropollutants detected in wastewaters are especially Alkylphenols, Musks and Sunscreens (eg. NP, HHCB, HHCB-lactone and Carbamazepine). No or few priority contaminants (PAHs, pesticides and PCBs) were identified.



Fig. 4: Non-metric multi-dimensional scaling plot (MDS) of benthic

taxon assemblages at impacted and control locations.

On Rocky platforms in intertidal zone

Macroalgae and fixed macrofauna identified

Mobile macrofauna counted in abundance

France

Spain

Stratified random sampling design

and counted in percentage cover

Fig. 5: Mean richness of taxonomic groups (green, brown and red macroalgae and macrofauna) of control and impacted locations (tested with a Multiple comparison test after a Kruskal-Wallis test; Kruskalmc).

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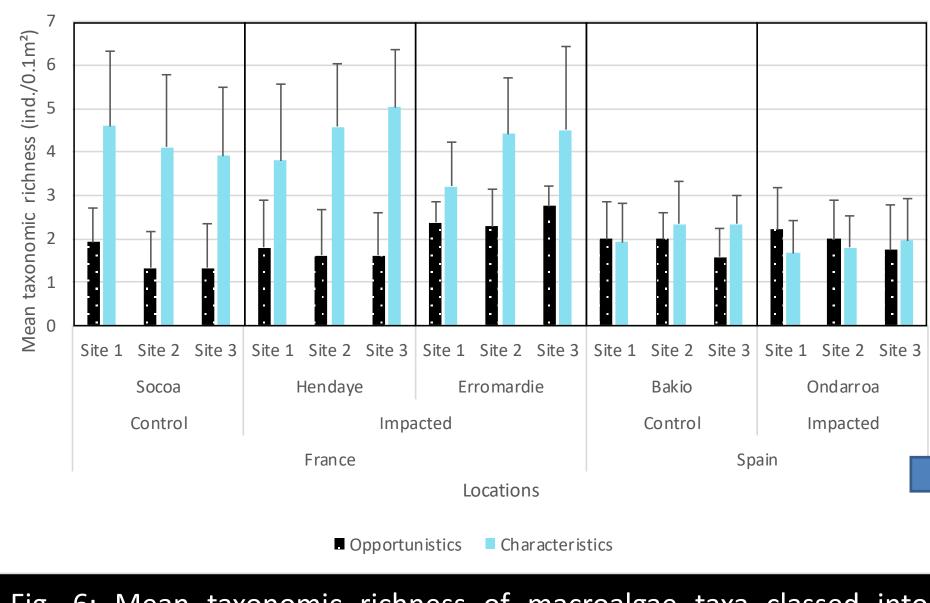


Fig. 6: Mean taxonomic richness of macroalgae taxa classed into 3). functional groups (characteristic -vs- opportunistic) according to Ar Gall & Le Duff (2016) (France) and Juanes (2008) (Spain) in control and impacted locations (tested with a Multiple comparison test after a Kruskal-Wallis test; Kruskalmc).

Allows to identify symptoms of anthropogenic disturbances (fig. 6):

### In France

- characteristic taxa > opportunistics (excepted for the most impacted location Erromardie (sites 1 and 3; *Kruskalmc*; *p*<0.05).
  - **In Spain** (visually, *Kruskalmc*; p>0.05):
- characteristic taxa > opportunistics on the ontrol location (Bakio).
- opportunistic taxa > characteristics on the impacted location (Erromardie) excepted on the site
- mTR of characteristic taxa in France > in Spain (*Kruskalmc*; *p*<0.05).

Conclusion: This study allowed to have a first insight of benthic diversity from "Control" locations and others impacted to WTP outfalls in French and Spanish Basque coast. Each WTP was also characterized by the analysis of the micropollutants at each discharge. A clear distinction was noticeable between French and Spanish diversity while WTP do not present major differences in term of micropollutants quantity and identity. To explain this distinction, other metrics (eg. for WFD) and factors such as habitat structure (geomorphology) and hydrography would be considered. Next steps will be to analyze the bioaccumulation of contaminants in organisms living on these study locations to highlight potential links between biological and chemical approaches.

