LEACHATES FROM VIRGIN AND BEACHED PLASTIC ITEMS: ASSESSMENT OF CHEMICAL COMPOSITION AND ECOTOXICITY TO MARINE ORGANISMS

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INTRODUCTION

Occurrence of marine plastic debris is a well-known issue, which worsens in basins with limited water exchanges and densely populated coasts. For this reason, Adriatic Sea is predicted as a preferential area of plastics accumulation [1]. Plastic is estimated to degrade in hundred years [2]. Floating, drifting or sinking in the sea, plastic can move for long distances, transporting inner additives [3] and adsorbs chemicals on its surface, which can be released again in the environment [4,5,6]; thus interacting with different ecological niches and different species, both pelagic and benthic and posing potential risks to marine biodiversity.

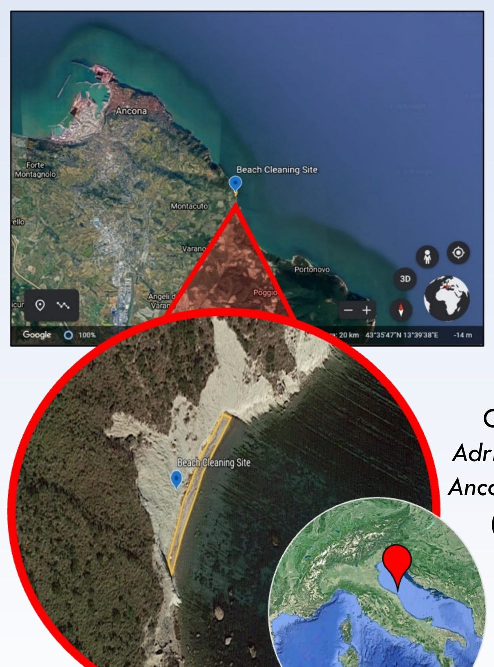
AIM

This study aims to explore the chemical characteristics of leachates from both beached and virgin plastic items and evaluate their toxicity to marine organisms of different trophic levels, using a battery of ecotoxicological bioassays.

EXPERIMENTAL PLAN

COLLECTION AND CHARACTERIZATION OF BEACHED PLASTIC ITEMS

SAMPLING AREA



Central Adriatic Sea, Ancona Coast (Italy)

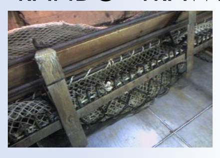
SAMPLING ACTIVITY

SELECTED ITEMS

PLASTIC POLYMER IDENTIFICATION (FT-IR)

1 Heterogeneous Fragments (HF)
2 Polystyrene Boxes (PB)
3 Mussel Nets (MN)
4 "Rapido" Trawls Rubber (RTR)
5 Drink Bottles (DB)

"RAPIDO" TRAWL

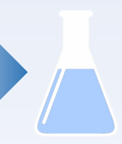


typical fishery targeting flatfish in the western side of the north-central Adriatic

LEACHATES PREPARATION AND ANALYSYS

Items are cutted in fragments ≤ 1cm^2

Seawater *Leaching Time* (LT) 72h



100 g/L Plastic-Water Ratio (except for PB and DB)

'VIRGIN': new store-bought material
'BEACHED': naturally weathered material collected on the beach

PLASTIC LEACHATE FOR CHEMICAL ANALYSIS

HPLC Technique for **polycyclic Aromatic Hydrocarbons (PAHs)**

Gas Chromatography With Flame Ionization Detector (FID) for **Aliphatic Hydrocarbons (AHs)**

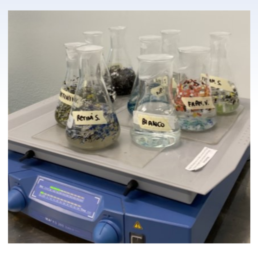
Atomic Absorption Spectrophotometry Techniques for the determination of **metals**

SECOND WASHING SOLUTION

ORGANIC COMPOUNDS
methanol (LT 4h)

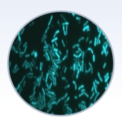
METALS
nitric acid 20% (LT 4h)

PLASTIC LEACHATE FOR ECOTOXICOLOGICAL BIOASSAYS

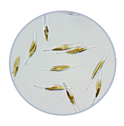


Leachate is **filtered** with a Minisart syringe filter 0.45 μm *to avoid mechanical disturbance of plastic particles*

Tested at different dilutions (100; 33; 20; 10 g/L)



Inhibition of bioluminescence in ***Aliivibrio fischeri*** (UNI EN ISO 11348-3:2019)



Growth Inhibition of the microalga ***Phaeodactylum tricornutum*** (UNI EN ISO 10253:2016E)

RESULTS

PLASTIC POLYMER IDENTIFICATION

Identification of plastic polymers was performed using Fourier transform infrared spectroscopy (FT-IR) technique in attenuated total reflectance (ATR). IR spectra were acquired with a resolution of 4 cm^{-1} in the mid-infrared region. Sample identification was achieved by comparing the IR spectrum with standard spectrum libraries (PerkinElmer®). Polymers corresponding to the reference spectra for more than 70% have been validated.

Category	Polymer	Percentage
HF	polypropylene (PP)	73%
	polyethylene (PE)	6.66%
	polystyrene (PS)	6.66%
	polyethylene terephthalate(PET)	6.66%
	ethylene_propylene (EP)	6.66%
MN	polypropylene (PP)	67%
	polyethylene (PE)	33%
PB	polystyrene (PS)	100%
DB	polyethylene terephthalate (PET)	100%
RTR	undefined polymer (rubber)	
	polyamide fiber (PA)	

Tab. 1 Polymer composition of samples expressed in percentage.

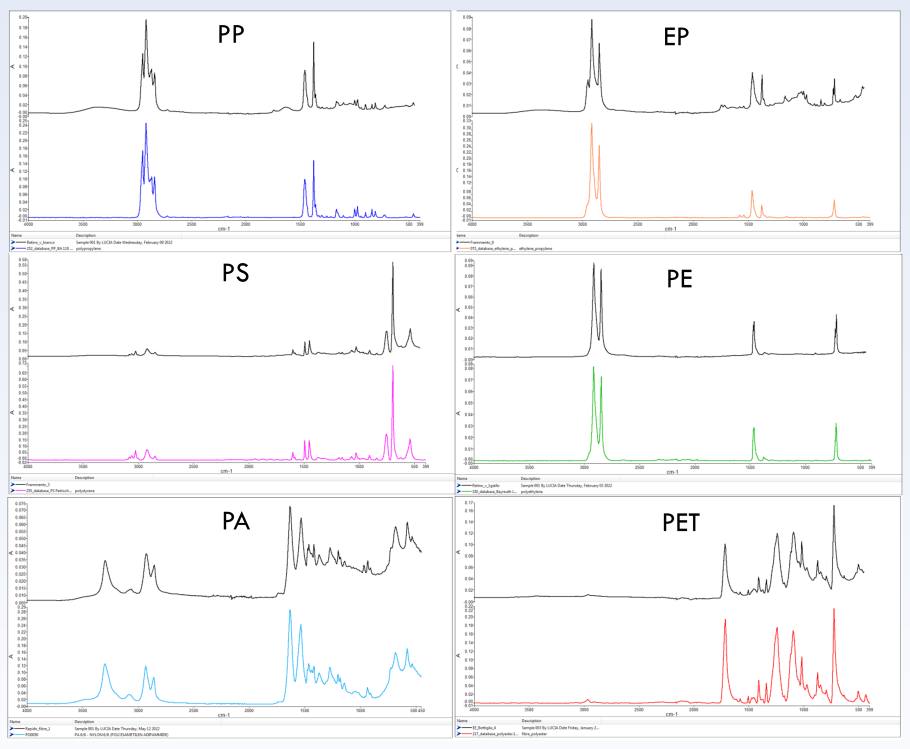


Fig.1 Spectra expressed in absorbance obtained from analysed samples by FT-IR technique.

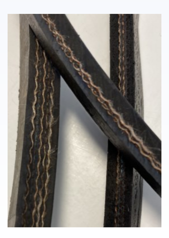


Fig. 2 RBR virgin plastic sample consists of two distinct parts: a) black rubber material, whose polymer cannot be analysed using FT-IR spectroscopy, due to its chemical-physical characteristics; b) fibrous material of polyamide.

CHEMISTRY

Chemical analyses revealed that both virgin and beached plastic leachates contain AHs in a range of 20-285µg/g and PAHs in a range of 34-1070 ng/g, while metals' concentrations in beached plastics ones proved to be more heterogeneous.

In general, beached plastic leachates revealed higher concentrations of AHs and PAHs than virgin plastic ones, except for RTR category which showed equivalent concentrations of AHs between virgin and beached plastics. In particular, PB leachates revealed higher concentrations of AHs, PAHs and metals than other samples.

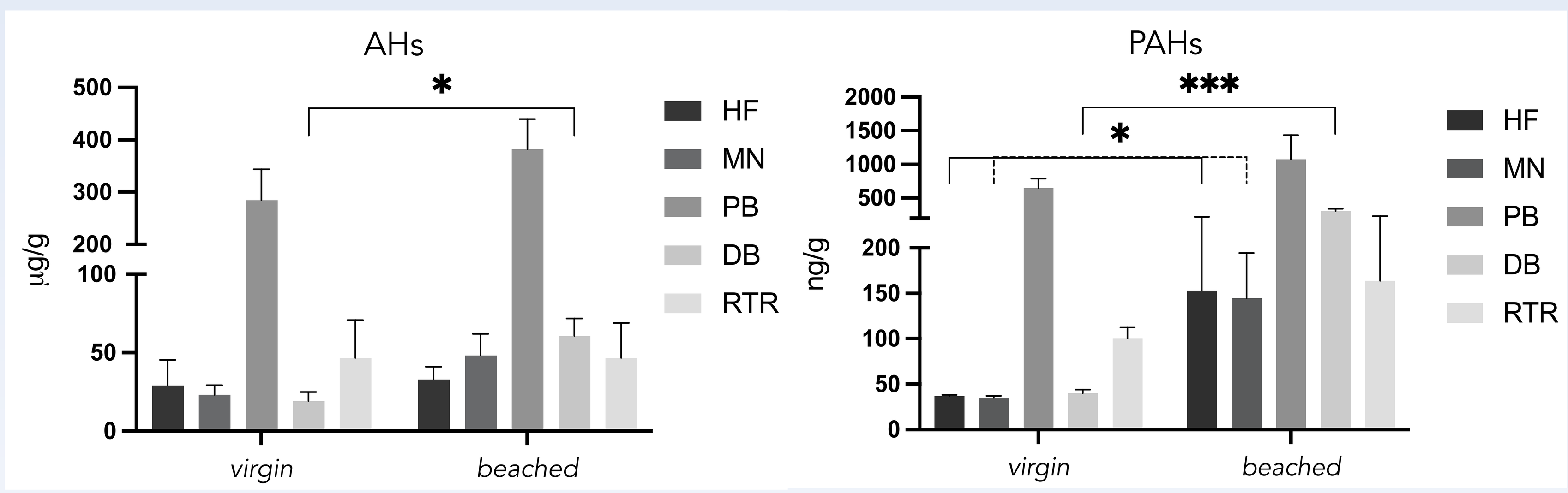


Fig. 4 Concentration (mean ± st. dev.; n = 3) of AHs (µg/g) and PAHs (ng/g) measured in virgin and beached plastic leachates of HF (heterogeneous fragments); MS (mussel nets); RTR ("rapido" trawl rubber); DB (drink bottles) and PB (polystyrene boxes);
T-test between virgin and beached plastic leachates (* $p<0.05$; *** $p<0.001$).

µg/g (p.s)	Element	HF mean	HF s.d.	MN mean	MN s.d.	PB mean	PB s.d.	DB mean	DB s.d.	RTR mean	RTR s.d.
	Al	6.54	4.852	35.781	12.858	13.88	5.811	2.465	2.093	3.318	1.956
	As	<LOD		0.123	0.022	0.137	0.082	0.038	0.029	0.018	0.007
	Cd	0.005	0.004	0.001	0	0.006	0.001	0.001	0	0.001	0
	Cr	0.047	0.069	0.164	0.034	<LOD		<LOD		<LOD	
	Cu	<LOD		0.071	0.088	<LOD		0.014	0.059	<LOD	
	Fe	22.07	12.1	79.16	6.59	24.74	38.12	2.6	2.77	2.97	2.5
	Hg	<LOD		<LOD		<LOD		<LOD		<LOD	
	Mn	4.32	1.75	11.77	4.27	22.47	10.72	<LOD		0.137	0.029
	Ni	<LOD		0.045	0.011	<LOD		<LOD		<LOD	
	Pb	0.048	0.044	0.279	0.069	0.059	0.032	0.016	0.005	0.014	0.004
	V	1.639	0.452	13.383	4.048	<LOD		0.046	0.012	<LOD	
	Zn	0.66	0.43	5.629	1.868	1.107	0.899	0.35	0.363	3.859	1.862

Tab. 2 Metals concentration (µg/g) measured in beached plastic leachates of HF (heterogeneous fragments); MS (mussel nets); RTR ("rapido" trawl rubber); DB (drink bottles) and PB (polystyrene boxes).

BIOASSAYS

For both the bioassays, virgin plastic leachates cause higher effect than beached plastic leachates. In particular, leachates derived from virgin RTR show the highest toxicological effect, even at higher dilutions.

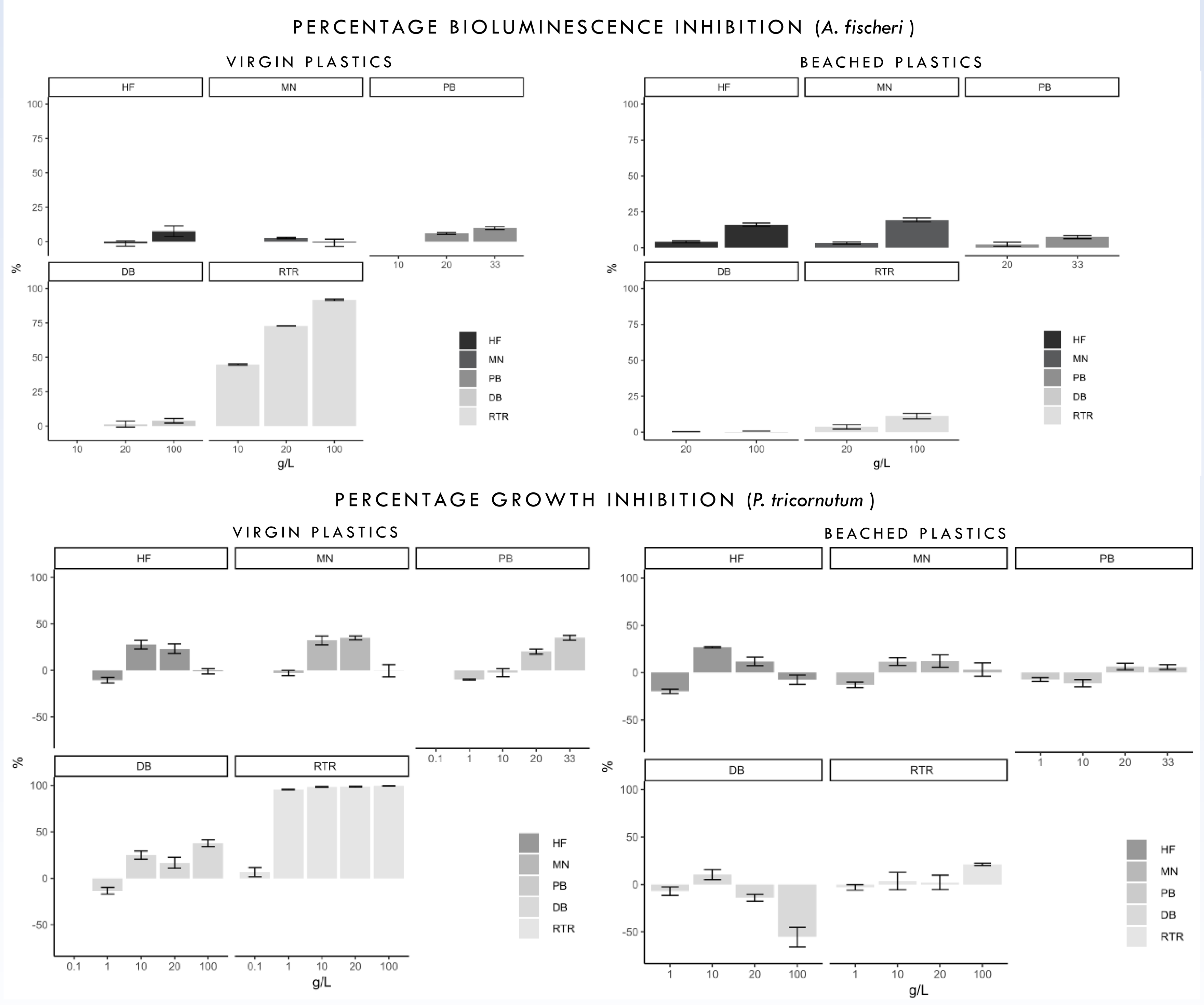


Fig. 3 Percentage (mean ± st. err.; n = 3) of *A. fischeri* bioluminescence inhibition and *P. tricornutum* growth inhibition exposed to different dilution of plastic leachates: HF (heterogeneous fragments); MS (mussel nets); RTR ("rapido" trawl rubber); DB (drink bottles); PB (polystyrene boxes).

CONCLUSIONS

- In general, virgin plastic leachates showed greater ecotoxicological effects than beached plastic ones, for both bacteria and algae bioassays. In particular, leachates derived from virgin RTR showed the highest ecotoxicological effect.
- PB leachates revealed higher concentrations of AHs, PAHs and metals than other samples, but obtained data are not directly related to the ecotoxicological effects measured with bioassays.
- *In progress*: analysis of metals in virgin plastics leachates and *Crassostrea gigas* embryotoxicity test, to implement the results obtained through an integrated ecotoxicological bioassays' battery.

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