MICRO meeting
Ostend, September 2014

Microplastics as a vector in the marine ecosystem

2 Mers Seas Zeeën
INTERREG IV A
FRANCE - ENGLAND - VLAANDEREN - NEDERLAND

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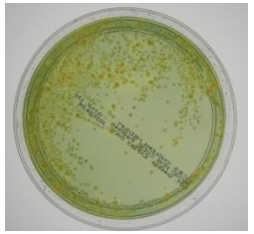
INTRODUCTION

Transport of Invasive/ Alien species

Additional effect?

Colonisation by Microbial species



Leaching and Adsorption of Additives & Environmental contaminants

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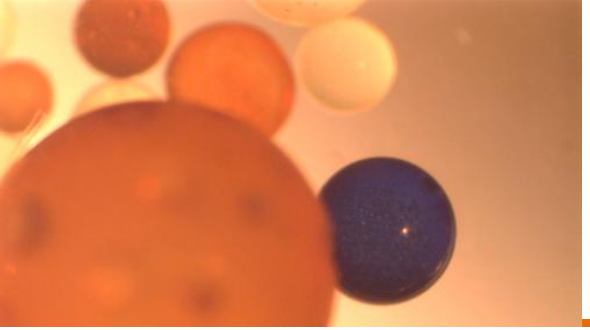

MAIN QUESTIONS

Which chemicals are present on plastic litter?

What are their main impacts of these chemicals on marine life?

Which bacteria are present on plastic litter?

What are the consequences for ecosystem health and human activities?

The main question here is whether microplastics are a threat to the ecosystem and possibly human health, including the risk of infection and contamination.

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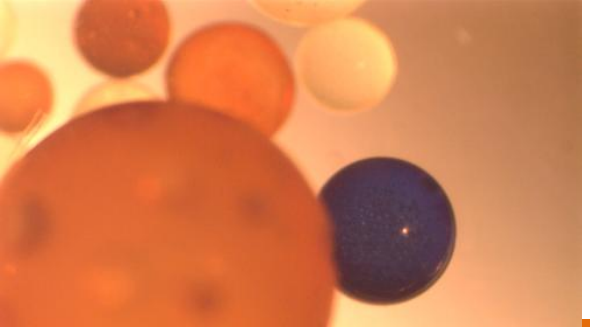

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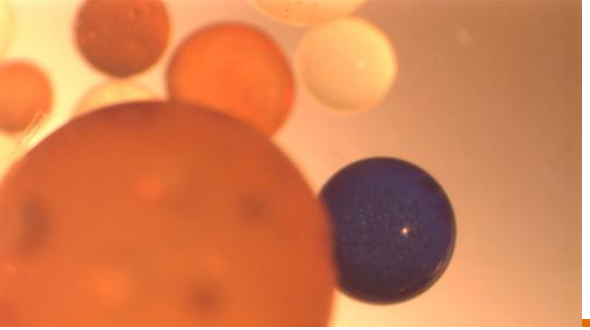

CHEMICAL COMPOUNDS ON PLASTIC LITTER

→GC-MS screening Marine litter

>450 chemicals identified

Plastic production process (general, precursors, intermediary,…)
Alkylated aromatic hydrocarbon (monocyclic)
2,4-Bis(1-methyl-1-phenylethyl)phenol
4,4'-ethylenebis (2,6-di-tert-butyl-phenol)
Benzoic acid, 5-chloro-2-(((2-chlorophenyl)imino)phenylmethoxy)-, methyl ester
Bisphenol A
Bisphenol A diallyl ether
Butanedioic acid
Dihydroxyacetophenone
Dimethyl-benzaldehyde
Dimethyl-phenyl-phosphonate
Dodecanedioic acid
Dodecanediol
Glycerol
Hexanedioic acid
Hexanedioic acid, alkyl ester
y-propanoic acid
alic acid
alic acid, alkyl ester

Fragrance (in cosmetics, medicins, perfumes,…) and their precursors
1,1,3-Trimethyl-3-phenyl-2,3-dihydro-1H-indene
Benzoic acid alkyl ester
Buten-one, trimethyl-cyclohexen-yl-
Galaxolide
Heptanoic acid
Isomethylionone
Lilial
Octanol
Phenylmeth
Phytol
Tonalide

Cosmetics (general, thickeners, bactericides,…)
Chloro-dimethyl-phenol
Docosanol
Ethanol, 2-phenoxy
Glycerol monostereate
Heptadecanol
Hexadecanol
Methyl-hydroxy-benzoic acid
Pentadecanol

CHEMICAL COMPOUNDS ON PLASTIC LITTER

Chemical Additives

- Pigments, dyes
- UV filters
- Stabilizers
- Solvents

Plastic Production

- (Branched) alkane/alkene
- Antioxidants
- Plasticizers
- Flame retardant
- Solvents
- Degradation products

CHEMICAL COMPOUNDS ON PLASTIC LITTER

Petrogenic & Pyrogenic compounds

- Polycyclic aromatic hydrocarbons (PAH)
- Alkylated PAHs
- Oxygenated PAHs

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Other POPs

- Pesticides
- PCBs

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Cosmetic Compounds

- UV filters
- Fragrance
- Thickeners

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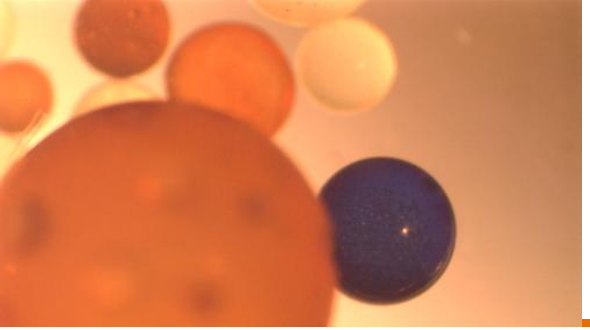
Biofilm & algae compounds

- Sesquiterpenes
- Fatty acids
- Sterols and steriods



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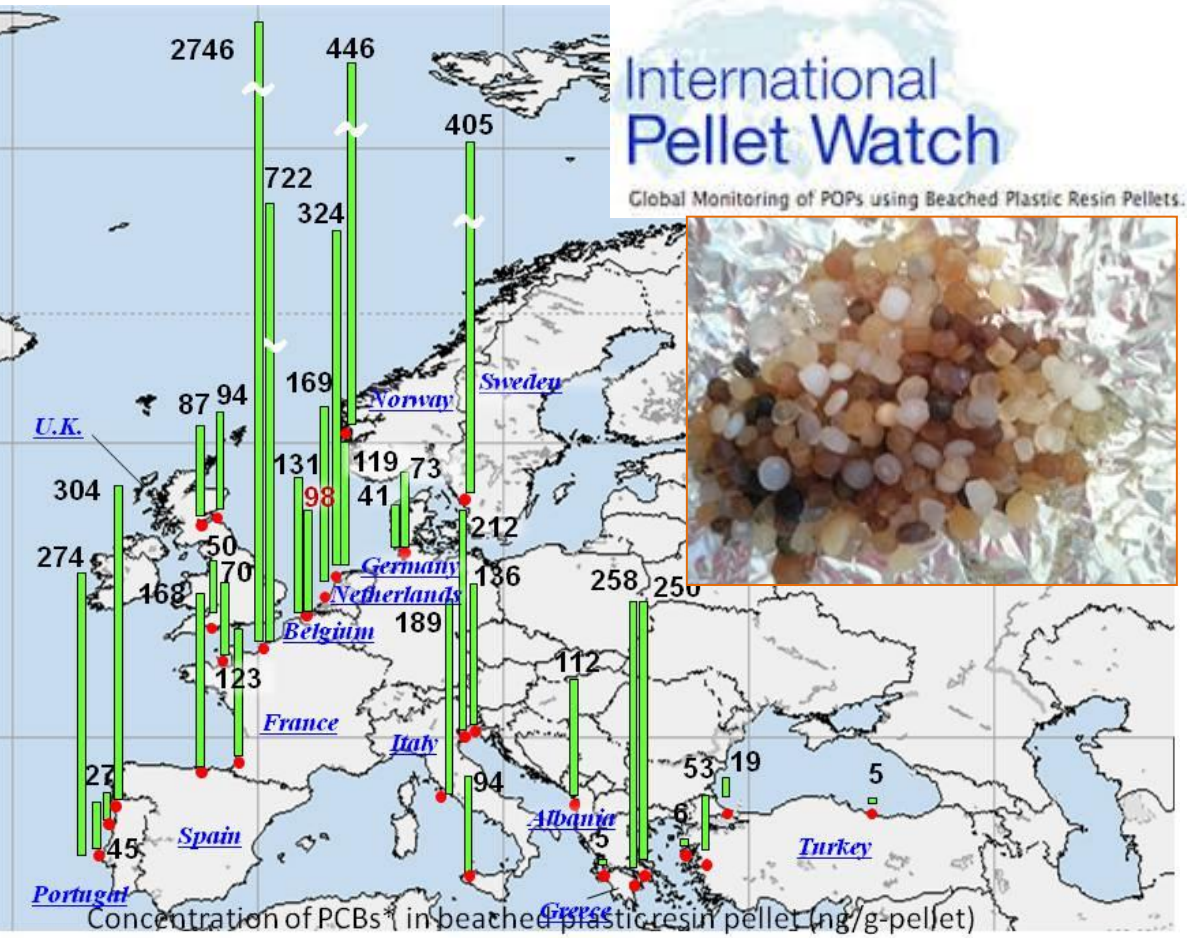


MICRO: sampled beach pellets in the InterReg region.

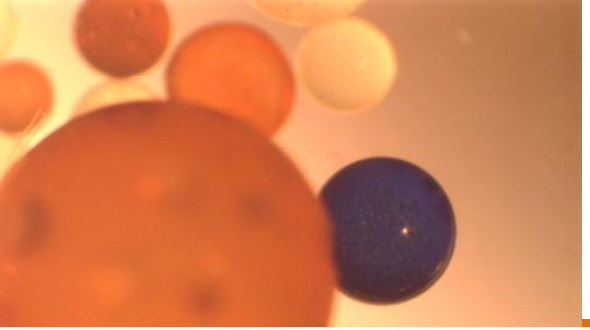
Belgium samples **(98 ng/g)** = means local pollution source of PCBs in the area.

These concentrations were lower than the neighbor countries (i.e., France : **~ 1000 ng/g**; Netherlands : ~ **300 ng/g**).

Why?

- Usage of PCBs in Belgium in the past may have been smaller than the other European countries.
- High PCB levels on pellets close to PCB sources, e.g. rivers (Seine)

→ **Pellets: indicator for pollution**



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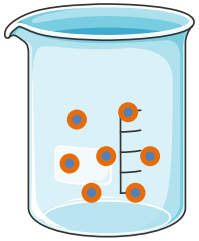
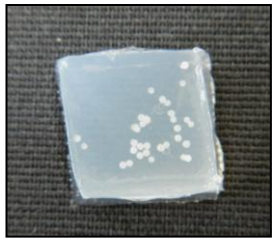
IMPACT OF PLASTIC-ADSORBED CONTAMINANTS



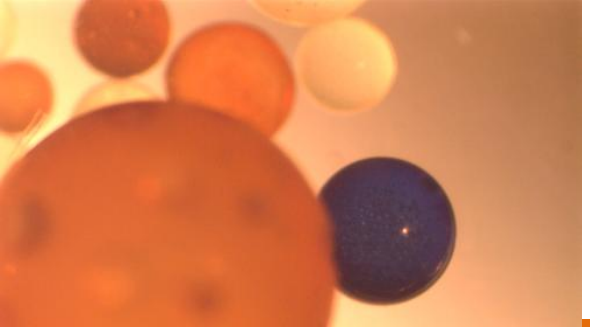
Microspheres (500-600µm) + PCBs



Gelatin feed

Norway lobster

- 3 weeks of exposure
- 1 week without feed (egestion)
- PCB levels in tissues are quantified
- sum of the 7 ICES PCBs: µg/kg fat.



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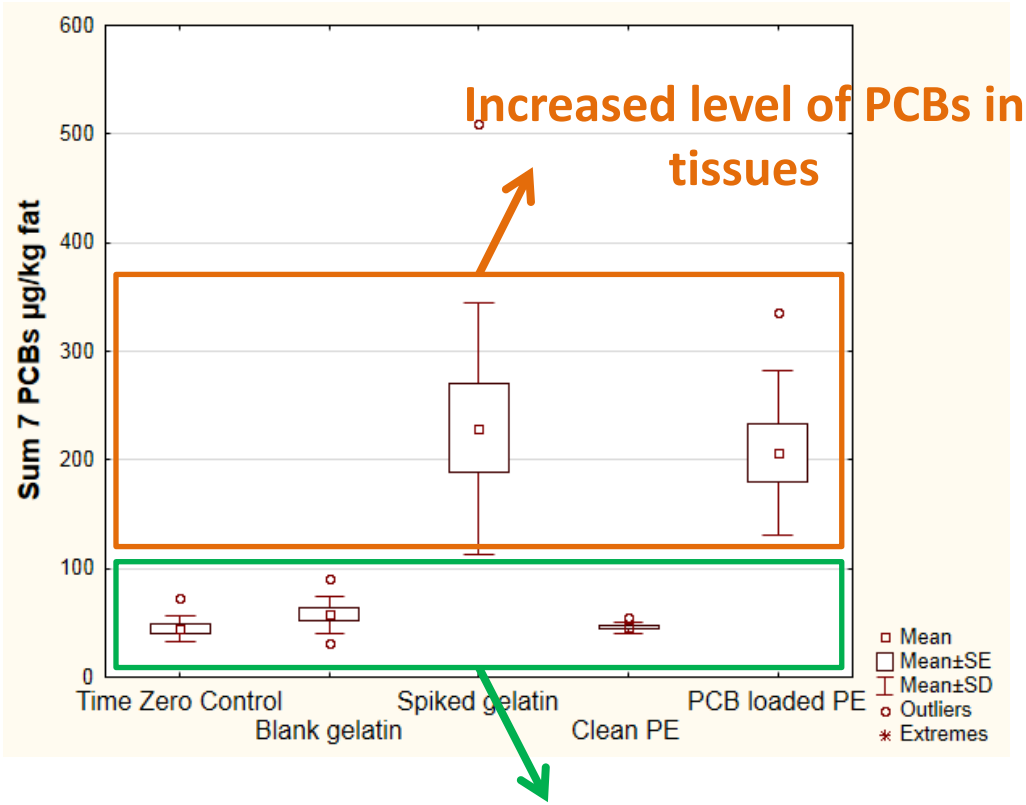
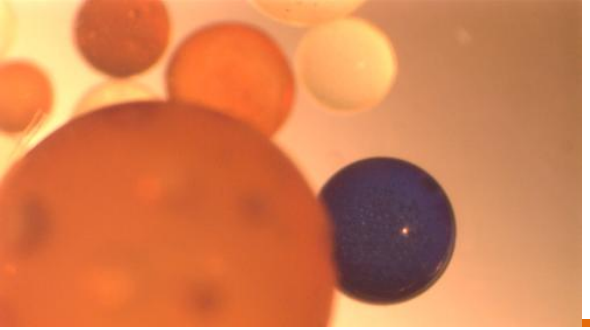


- Short period in the digestive tract will be sufficient to release PCBs
- Significant increase of PCB in tissues
- Small % uptake of PCB in tissues (0.07 – 0.17%)
- No specific effect of microspheres

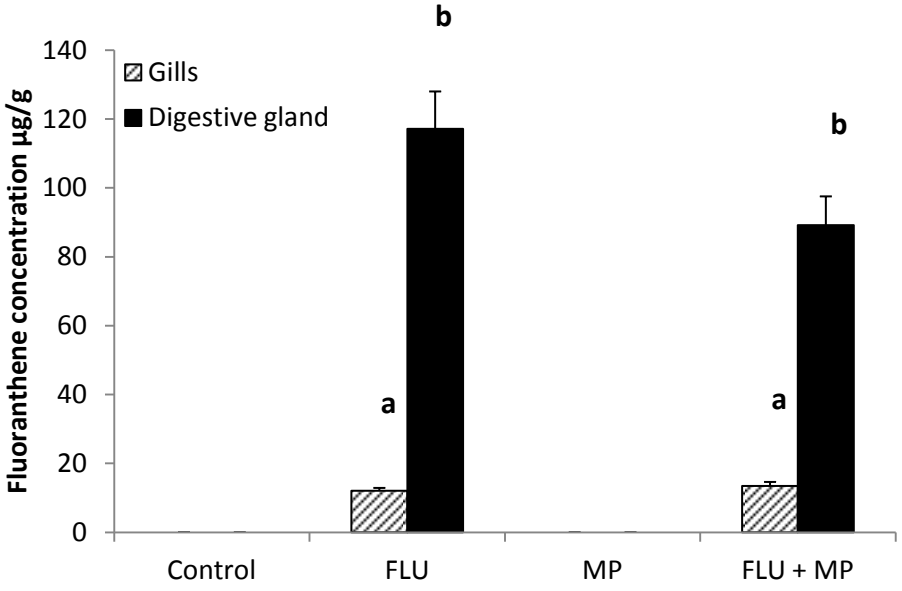


IMPACT OF PLASTIC-ADSORBED CONTAMINANTS

Main impact of MP coupled with Fluoranthene on mussels (a2)

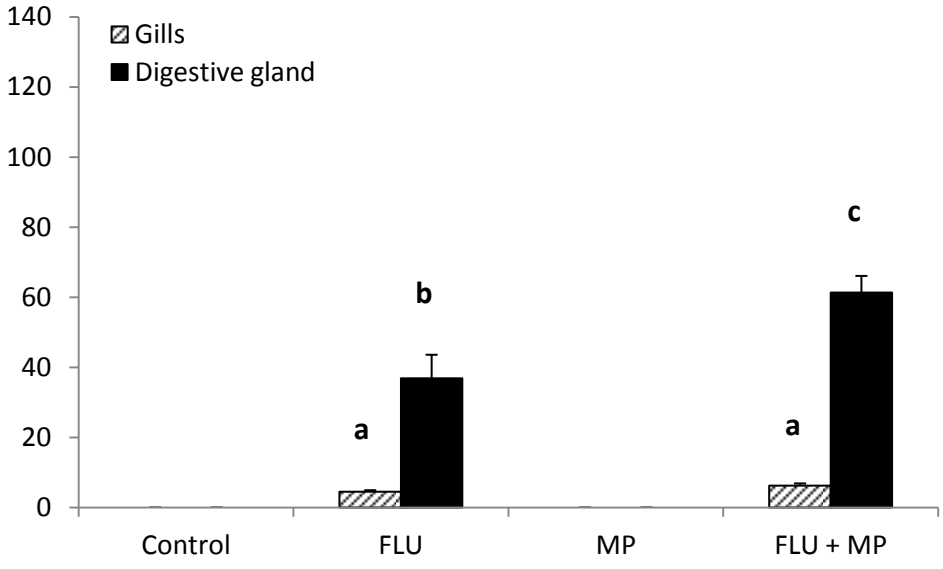
Impact of MP on FLU accumulation / depuration

- No significant effect of MP on FLU accumulation in mussels tissues
- Significant reduction of the depuration rate – linked to impact of MP on animal metabolism / activity of enzymes involved in PAH metabolism / transformation (Cyt-P450, GST?)



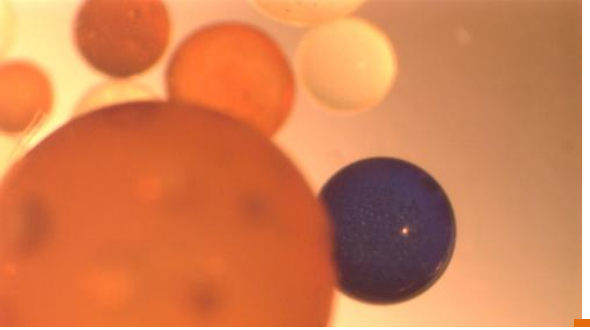
T7 – Exposure

ANOVA, p > 0,05



T14 - Depuration

ANOVA, p < 0,001



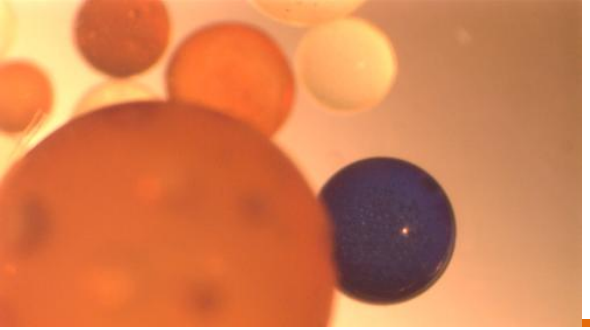
IMPACT OF PLASTIC-ADSORBED CONTAMINANTS

Interactive effects of MP and FLU on immune responses

- Significant impact on hemocyte viability – still observed after one week of depuration
- Induction of an oxidative stress – not found in the mussels exposed to FLU + MP
- Modification of phagocytosis response to FLU

Gene expression analysis

- Most of the effects observed at the end of the depuration phase (T14)
- No/few effects of MP on genes involved in PAH metabolism (enzyme activities?)
- Effects of MP on:
  - Digestive enzyme: Isocitrate dehydrogenase
  - Antioxidant defenses: catalase, super oxide dismutase
  - Multi drug transporter: P-glycoprotein
  - Glycolysis: Pyruvate kinase
  - Tumor suppressor: p53



IMPACT OF PLASTIC-ADSORBED CONTAMINANTS

MICRO exposure: only small % uptake of PCB in tissues (0.07 – 0.17%)

→Additional effect of PCBs due to microplastics ingestion may be minimal.

Koelmans et al. 2013, Besseling et al. 2013: experiments with PCB-containing sediments suggest a marginal increase of bioaccumulation of PCB in the tissues of *Arenicola marina* due to the presence of microplastics

→Additional effect of additives due to microplastics ingestion?

→Gastric conditions: increase the bioavailability of additives?

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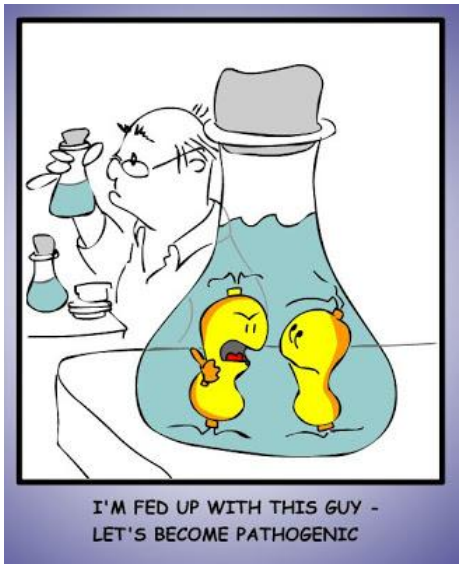

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BACTERIAL COMMUNITIES ON MARINE LITTER

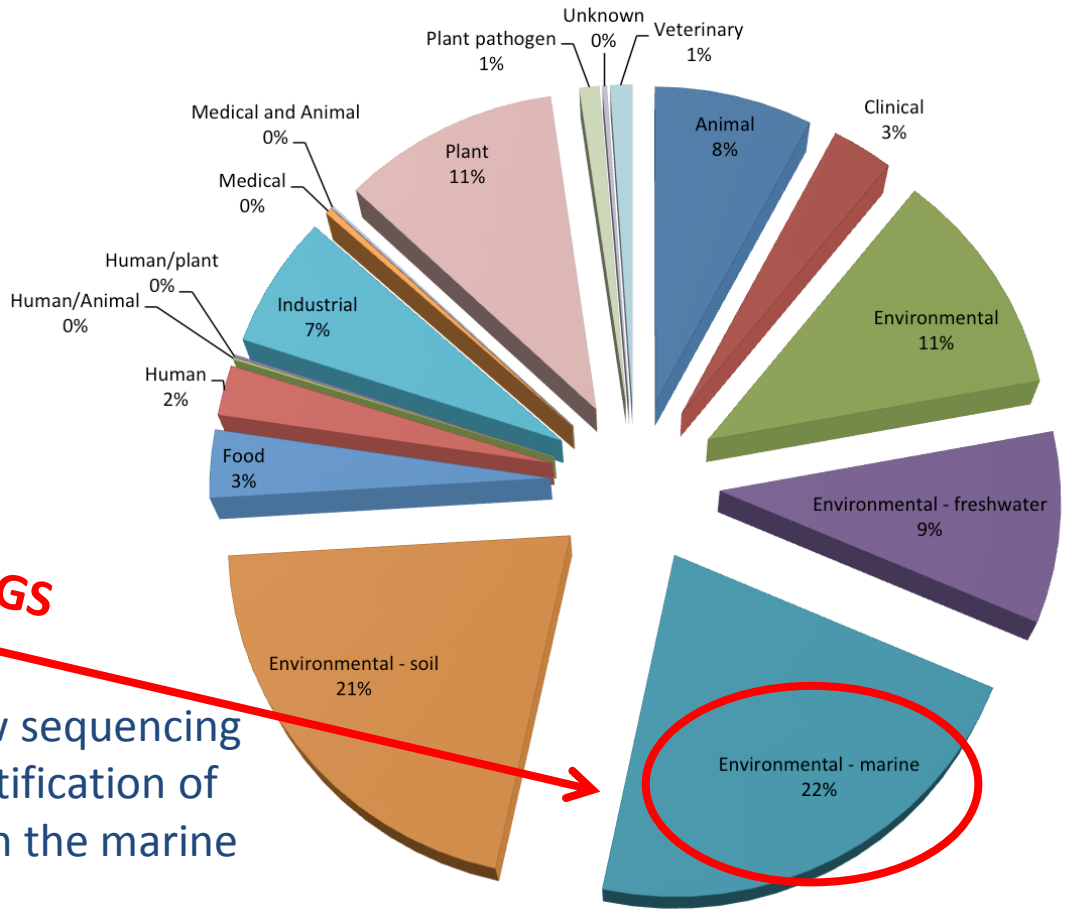
Identification of bacterial communities on plastic using Next Generation Sequencing

NGS

The use of new sequencing methods: identification of new bacteria in the marine environment

New Taxa

Sources of taxa, July 2013 to June 2014



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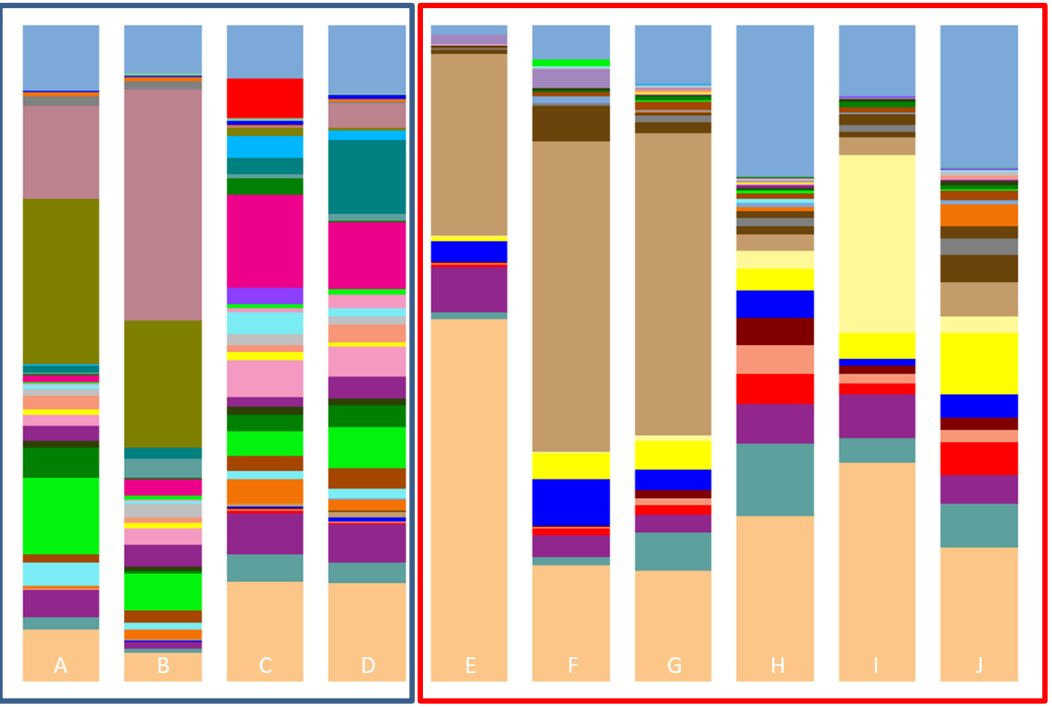
BACTERIAL COMMUNITIES ON MARINE LITTER

Amplicon sequencing – Family level:



High bacterial biodiversity

High diversity between samples!

Significant distinction between Beach pellets and Marine litter

'Uncultured' bacteria

Beach pellets

Marine liter

Potential pathogenic bacteria?

BACTERIAL COMMUNITIES ON MARINE LITTER

- **Potential pathogens? Yes indeed!!!**

Bacteroides thetaiotaomicron (infections)
Escherichia coli (fecal contamination, infections)
Shewanella putrefaciens (open lesions, sepsis)
Bacillus cereus (diarrhea)
Aliivibrio wodani (open lesions)
Stenotrophomonas maltophilia (infections)
Pseudomonas anguilliseptica (septicemia)
Escherichia fergusoni (open wound)
….

→ **A lot of potential pathogenic genera are observed**, such as Vibrio, Aeromonas, Enterobacter, Halomonas, Mycobacterium, Photobacterium, Pseudomonas, Rhodococcus, Shigella…

→ **Seawater? Sediment?**

→**The genus Vibrio is of main importance for the aquaculture of commercial bivalves** such as oyster and mussel, as some of the Vibrio species are well known pathogens for bivalves

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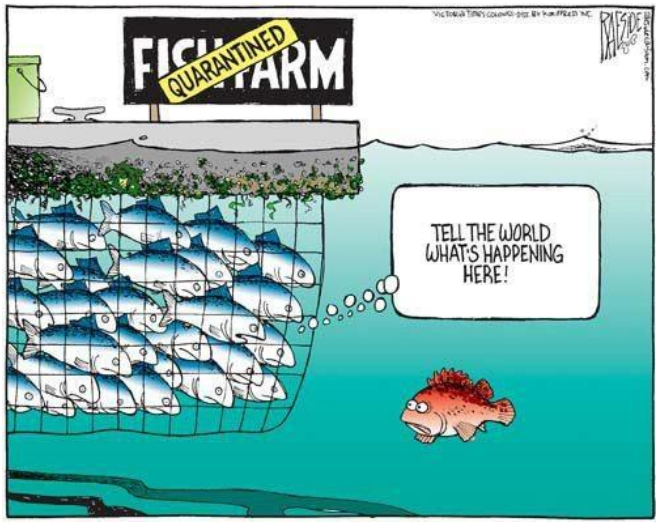
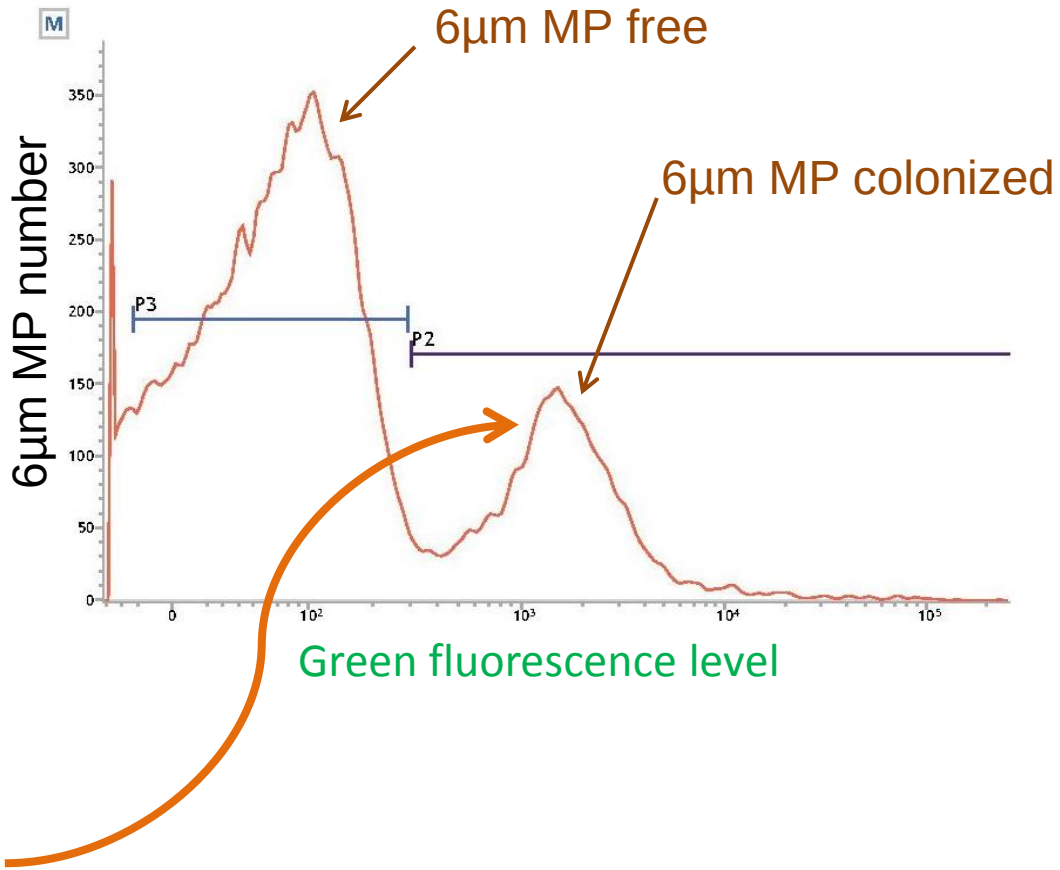


IMPACT OF PATHOGENIC BACTERIA ON MICROPLASTIC

Vibrio aestuarianus **is a pathogenic bacteria for adult oyster** ***Crassostrea gigas***

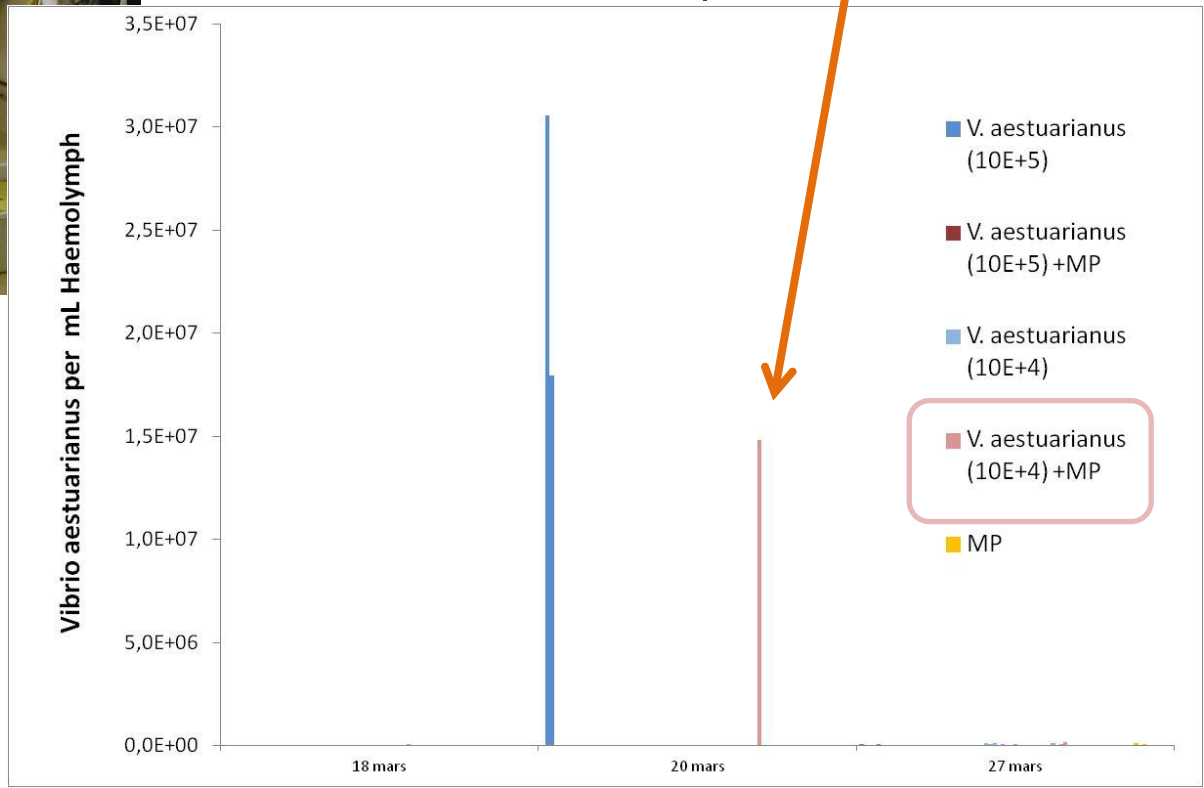
6 µm polystyrene MP are colonized within some hours by *Vibrio aestuarianus* *GFP* *(fluorescent green)*

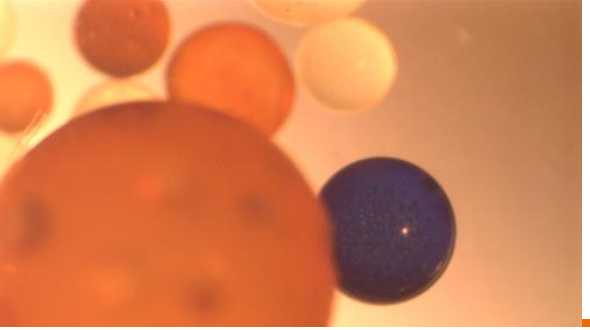


IMPACT OF PATHOGENIC BACTERIA ON MICROPLASTIC

Experimental infection design:

Vibrio aestuarianus **has been found in the haemolymph of oyster infected (bathing) by a mixture of 6 µm polystyrene MP colonized by the bacteria and free bacteria.**





TAKE HOME MESSAGES

Which chemicals are present on plastic litter?

→ *MICRO: a lot of diverse compounds are present, even harmful chemicals*

What are their main impacts of these chemicals on marine life?

→ *MICRO: microplastics are ingested,* but additional effect of PCBs due to microplastics ingestion may be minimal.

Which bacteria are present on plastic litter?

→ *MICRO: high bacterial biodiversity, even potential pathogenic bacteria*

What are the consequences for ecosystem health and human activities?

→ *MICRO: indication that plastics act as a vector for pathogenic bacteria*

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RESEARCH TOPICS

Additional effect of additives & chemicals due to microplastics ingestion?

Properties of identified marine bacteria: Functional diversity

Biodegradation pathways: Are bacteria able to biodegrade marine plastic litter?

Effect of climate change on the bacterial communities e.g. *Vibrio spp.*

Are bacteria on microplastics able to infect seafood?

Thank you