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2023

A LONG TERME, L'INGESTION DE MICROPLASTIQUES NUIT À LA CROISSANCE ET À LA REPRODUCTION CHEZ LES POISSONS

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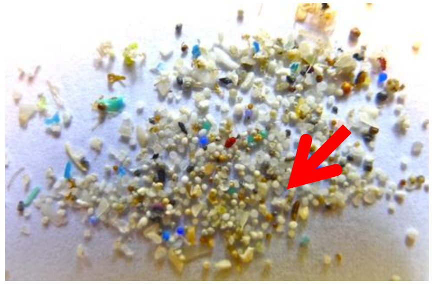
UMR MARBEC

Microplastics in the environment

- 1 nm < nano < 1 µm < microplastic < 5 mm < macro

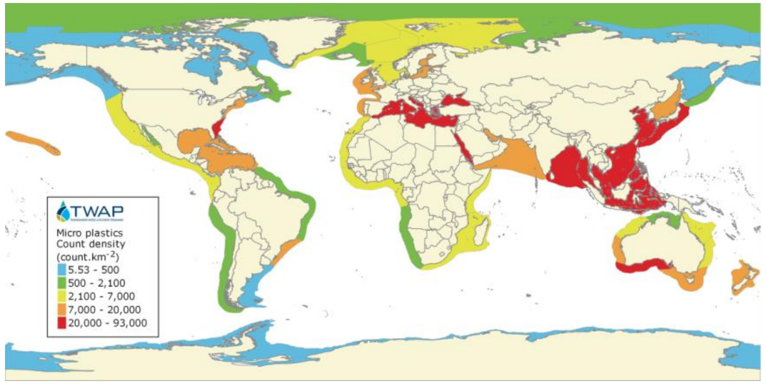


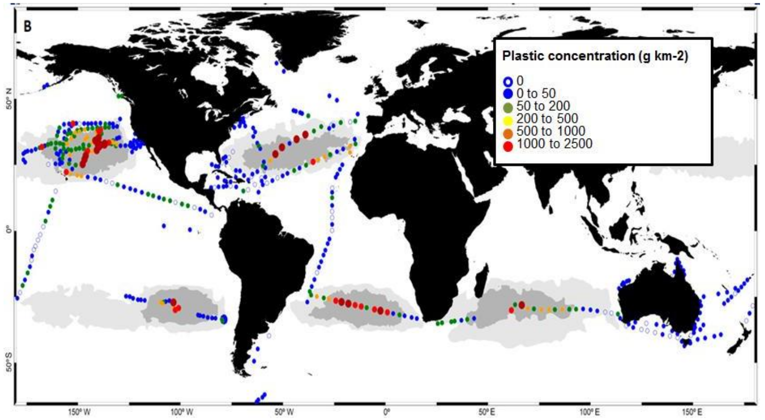


- MPs are very diverse
  - shapes (particles, beads, fibres…)
  - plastic types (HDPE, LDPE, PVC, PS…) + biodegradables
  - additives (flame retardant, dye…)

Microplastics in the environment

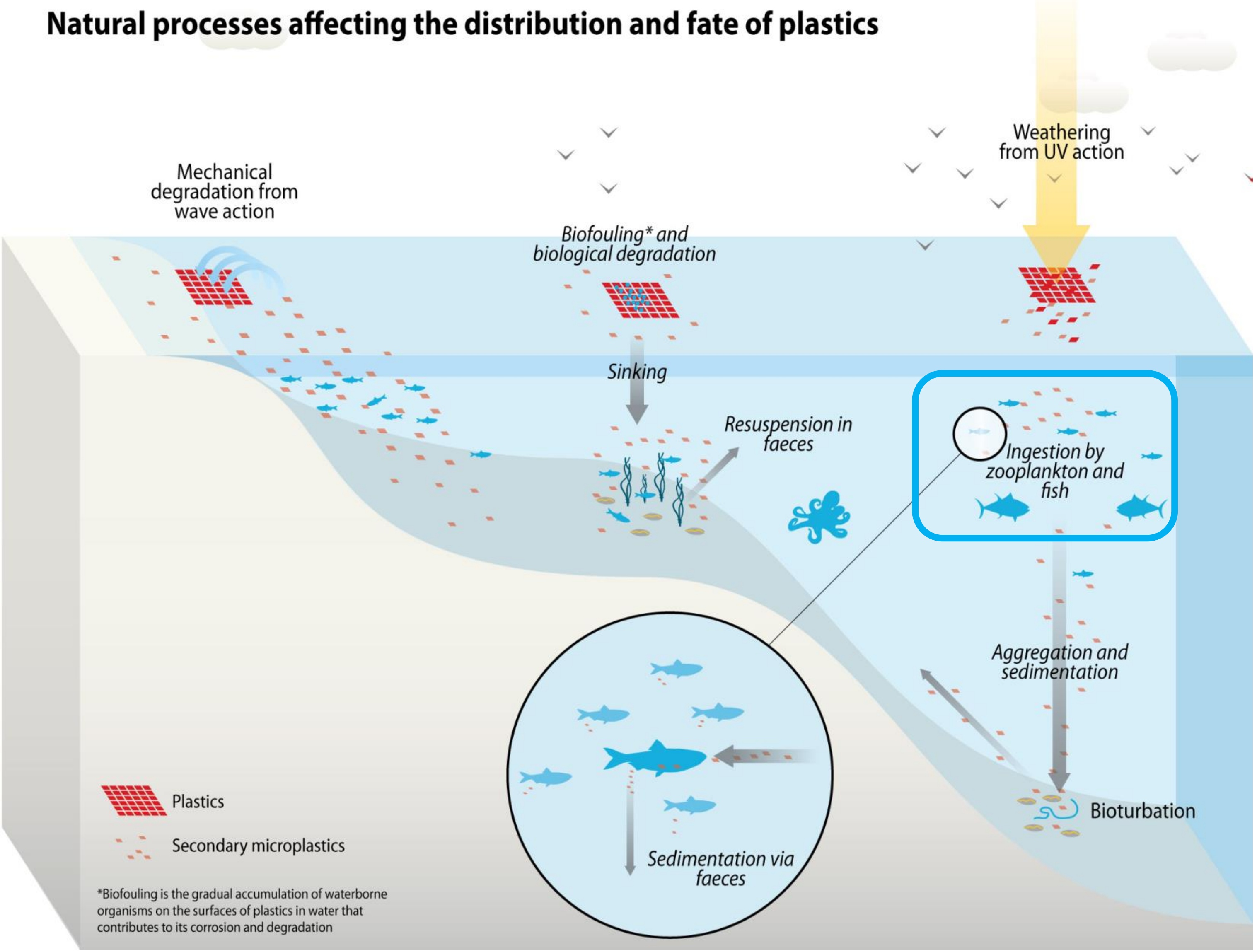
- Multiple sources
- Ubiquitous distribution in aquatic environment
- Most studies are focused on water compartment of marine environment but the same issues occurred in freshwater
- Sediment is an underestimated compartment





TWAP Large Marine Ecosystems Indicators (2015)
Cozar et al. (2014) *PNAS* **111**, 10239–10244
Browne et al. (2011) *Environ Sci Technol* **45**, 9175–9179

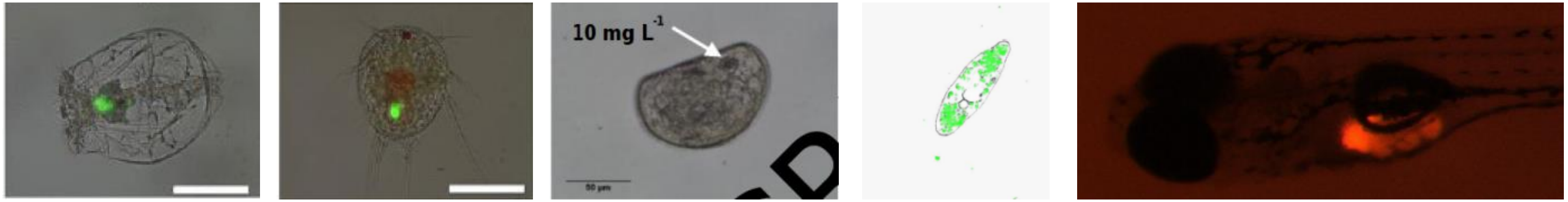
Microplastics in the environment



https://www.grida.no/resources/6911

Direct uptake of MPs

- All tested organisms were able to ingest MPs



  - 10-20 µm seems to be the upper limit for plankton
- No internalisation for cells in culture or microalgae
  - In the micrometer range
- Size, shape, weathering seem to influence uptake
- Egestion is often fast (within hours) and evidences for translocation are debated
  - Translocation seems to be limited to some organisms
  - Translocation seems limited to the smallest MPs or NPs

Importance/relevance of trophic transfer for MPs uptake?

- Preys can act as a funnel delivering high amounts of MPs
- and transport across compartments act as a new source of MPs (e.g. sediment → plankton → pelagic)
- May be important for some organisms or life stages (e.g. fish larvae)

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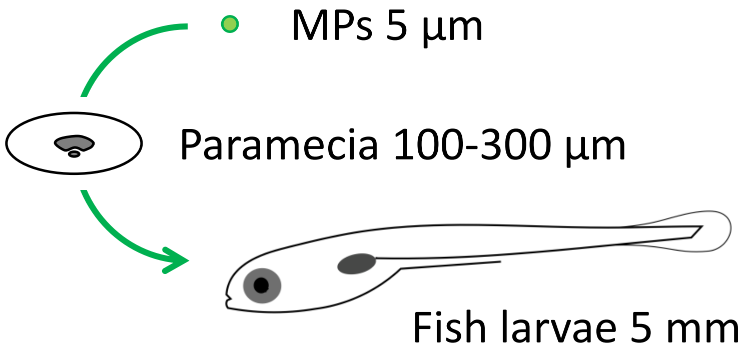
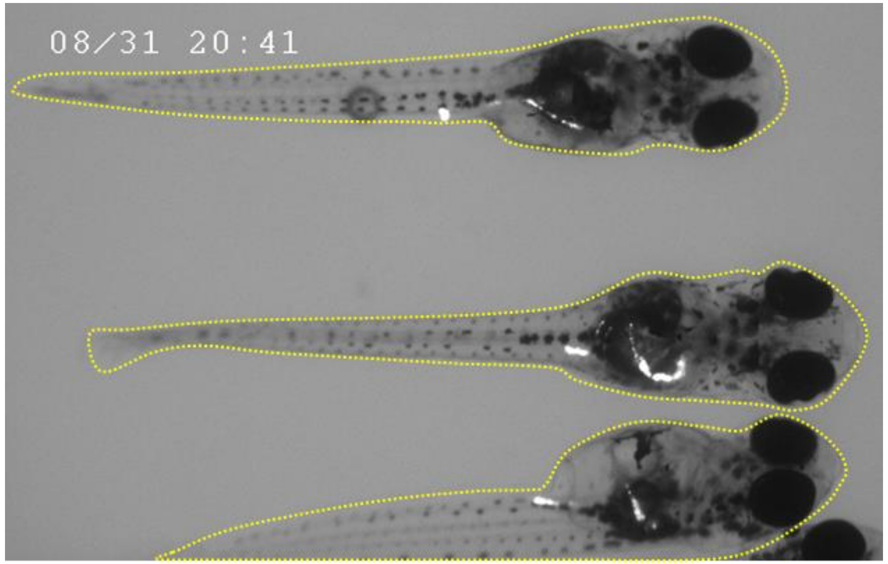
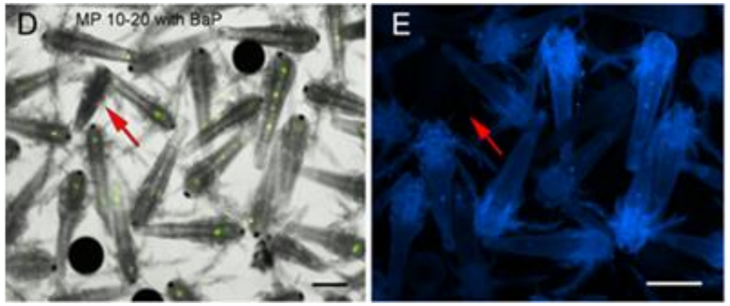


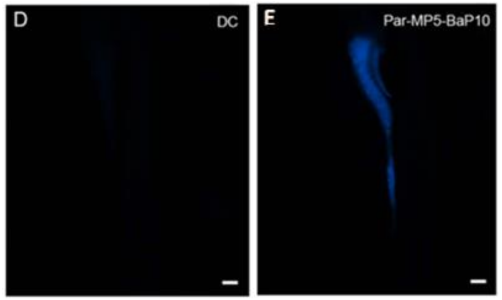


Can MPs act as vector for pollutants?

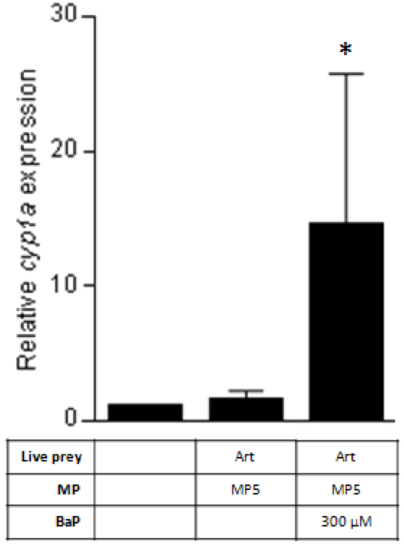
- Beyond additives, many chemicals can adsorb to MPs
- Mechanisms underlying adsorption and desorption inside organisms remain largely unknown
- Chemicals can be delivered to organisms by MPs
- Chemicals may be "pre"-desorbed in preys



BaP desorption in prey



BaP transfer to larvae through food chain



BaP is bioavailable in fish larvae

Batel et al. (2016) *Environ Toxicol Chem* **35**,1656-66.
Cousin et al. (2020) *Mar Environ Res* **161**, 105126

Regulatory (acute) toxicity tests

Organisms		Methods	Size (µm)	LOEC (mg/L)
Rotifer	*Brachionus plicatilis*	ASTM	1-4	1
			4-6	>10*
Copepod	*Tigriopus fulvus*	ISO 14669	1-4	1
			4-6	>10*
Copepod	*Acartia clausi*	ISO 14669	4-6	>30*
Mussel	*Mytilus galloprovincialis*	ISO 17244	1-4, 4-6, 6-8.5, 11-13	>100*
			<63	>100*
Sea urchin	*Paracentrotus lividus*		4-6, 11-15	100*
			<40	>100*
Fish	*Oryzias melastigma*	OECD 212	4-6	>10*

* Maximum tested concentration

→No toxicity using acute tests
→No toxicity at environmental concentration

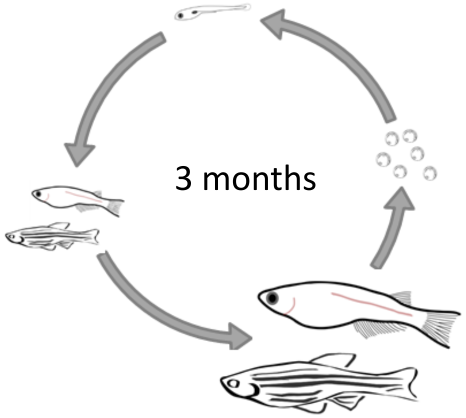
Beiras et al., (2018) *J Hazard Mat* **360**, 452-460

Evaluation of chronic toxicity

- Industrial microplastics
  - LDPE 2-10 µm (Micropowders Inc.)
  - PVC 80-200 µm (Fainplast Srl.)
- Spiked with three chemicals

Plastic types	Control	+ perfluorooctane sulfonic acid	+ benzo[a]pyrene	+ benzophenone-3
PE	PE-MP	PE-PFOS	PE-BaP	PE-BP3
PVC	PVC-MP	PVC-PFOS	PVC-BaP	PVC-BP3

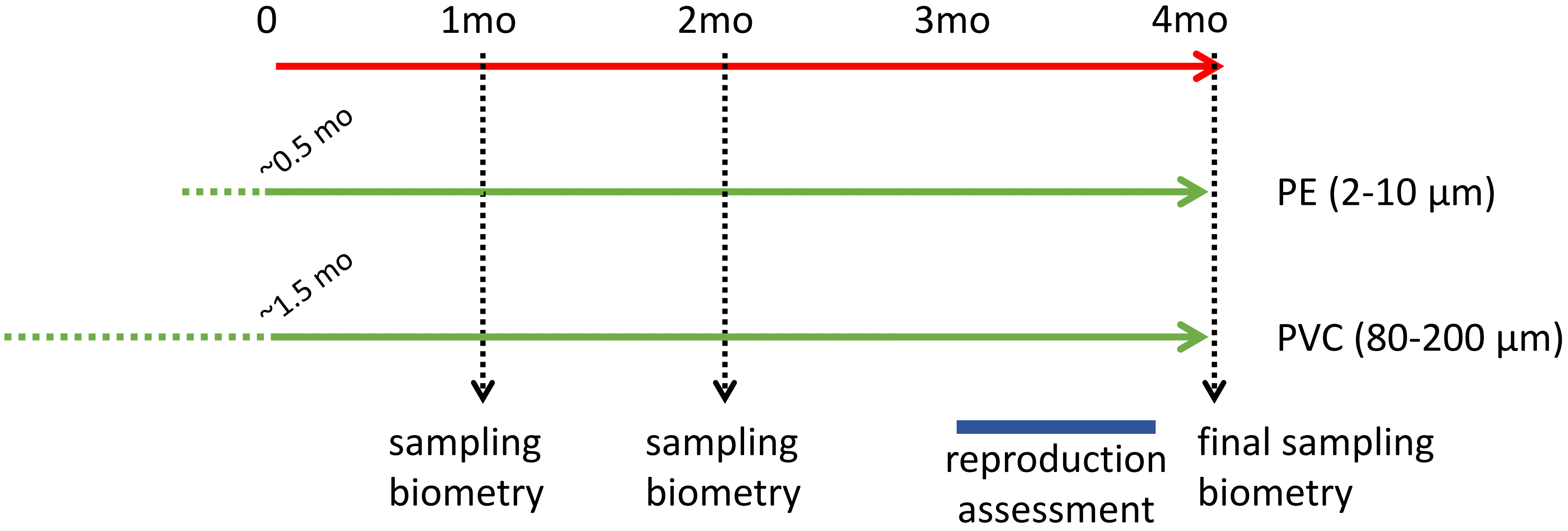
- Two short-life cycle model species
  - Zebrafish (*Danio rerio*)
  - Marine medaka (*Oryzias melastigma*)



Cormier et al., (2021) *J Hazard Mat* **415**, 125626.

Long-term exposure

- Dietary exposure over 4 months
  - Spiking of diet (food pellets 100-400 µm) at 1% w/w
  - Exposure schedule according to MPs size



Long-term exposure

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  - Spiking of diet (food pellets 100-400 µm) at 1% w/w
  - Exposure schedule according to MPs size

This means:

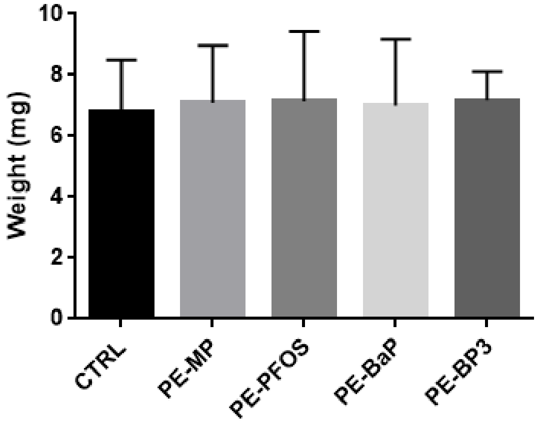
- **1.5-3.5 particles** of 330x330x330 µm per individual per day for adults

Automated flow-through system with 30% of daily water renewal

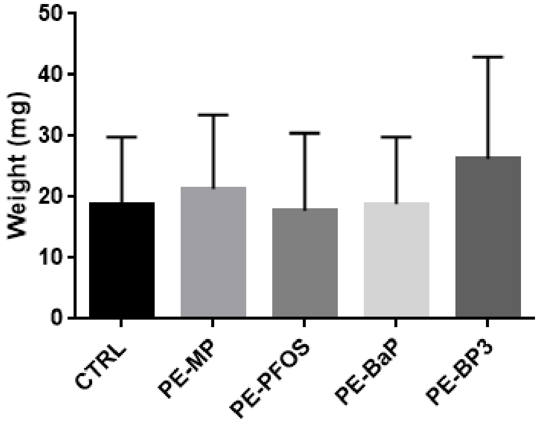
No or weak early disruption of growth

→1 month of exposure	→2 months of exposure	

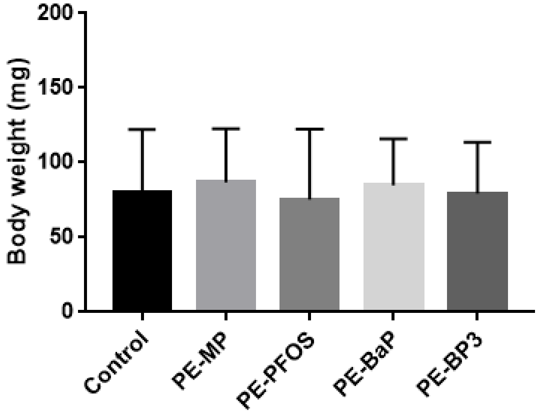
→ medaka /PE



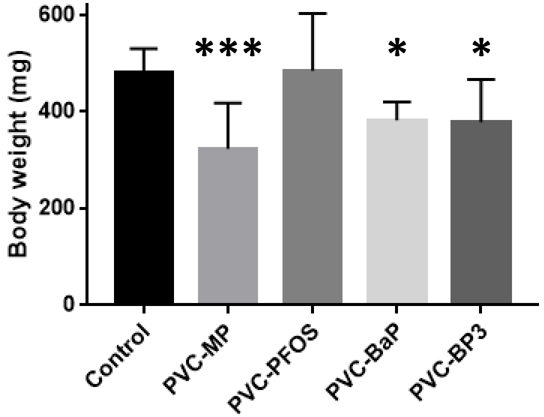
→ medaka /PE



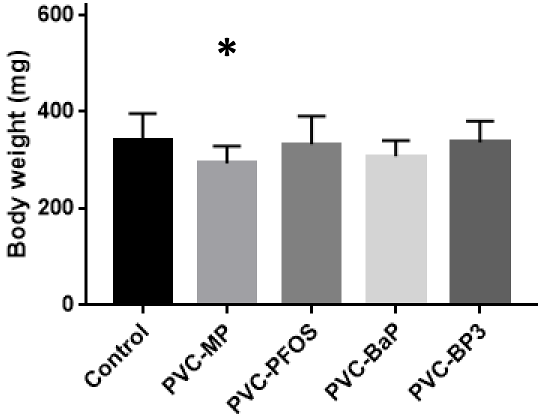
→ zebrafish /PE



→ zebrafish / PVC / female



→ zebrafish /PVC / male



→ No or weak early effects on growth
→ No difference between plastic types and pollutant

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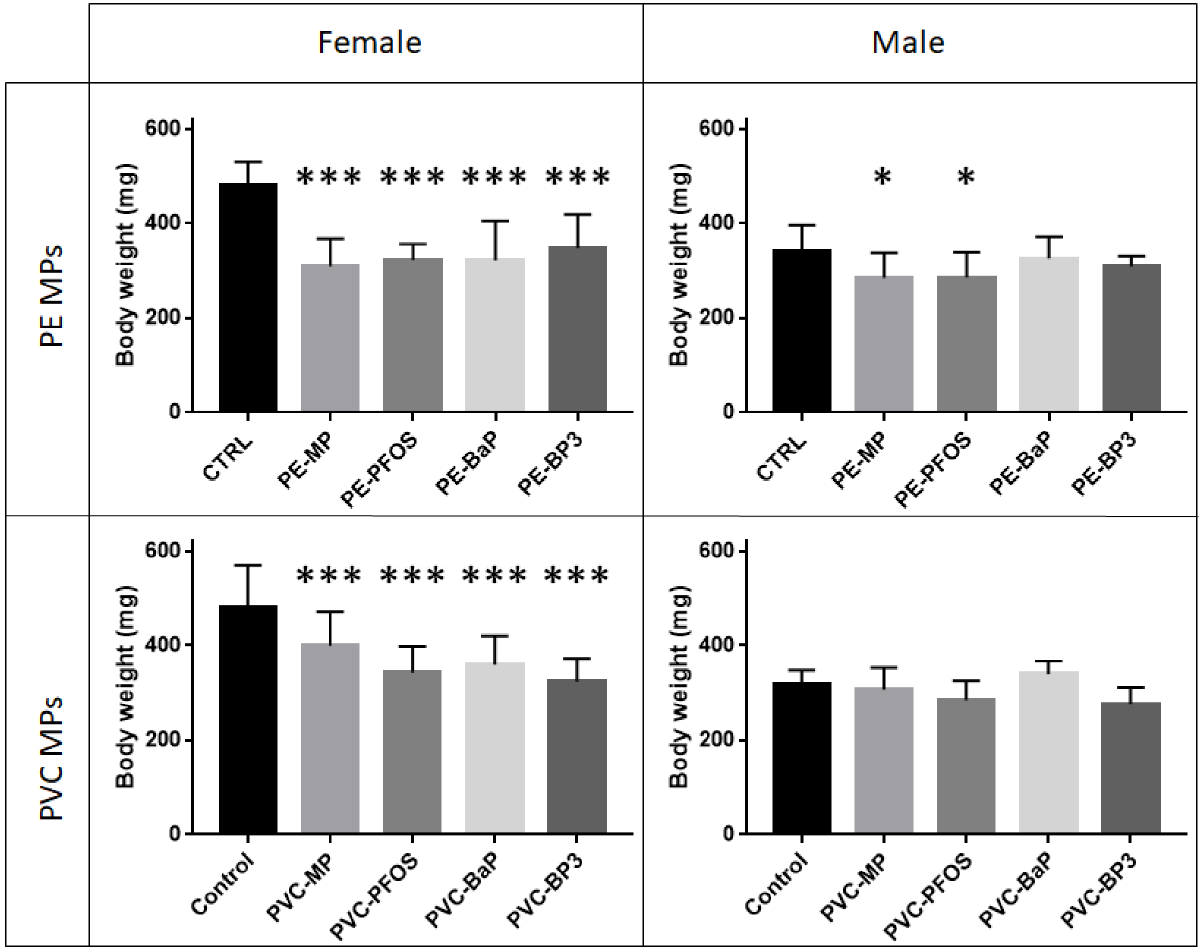
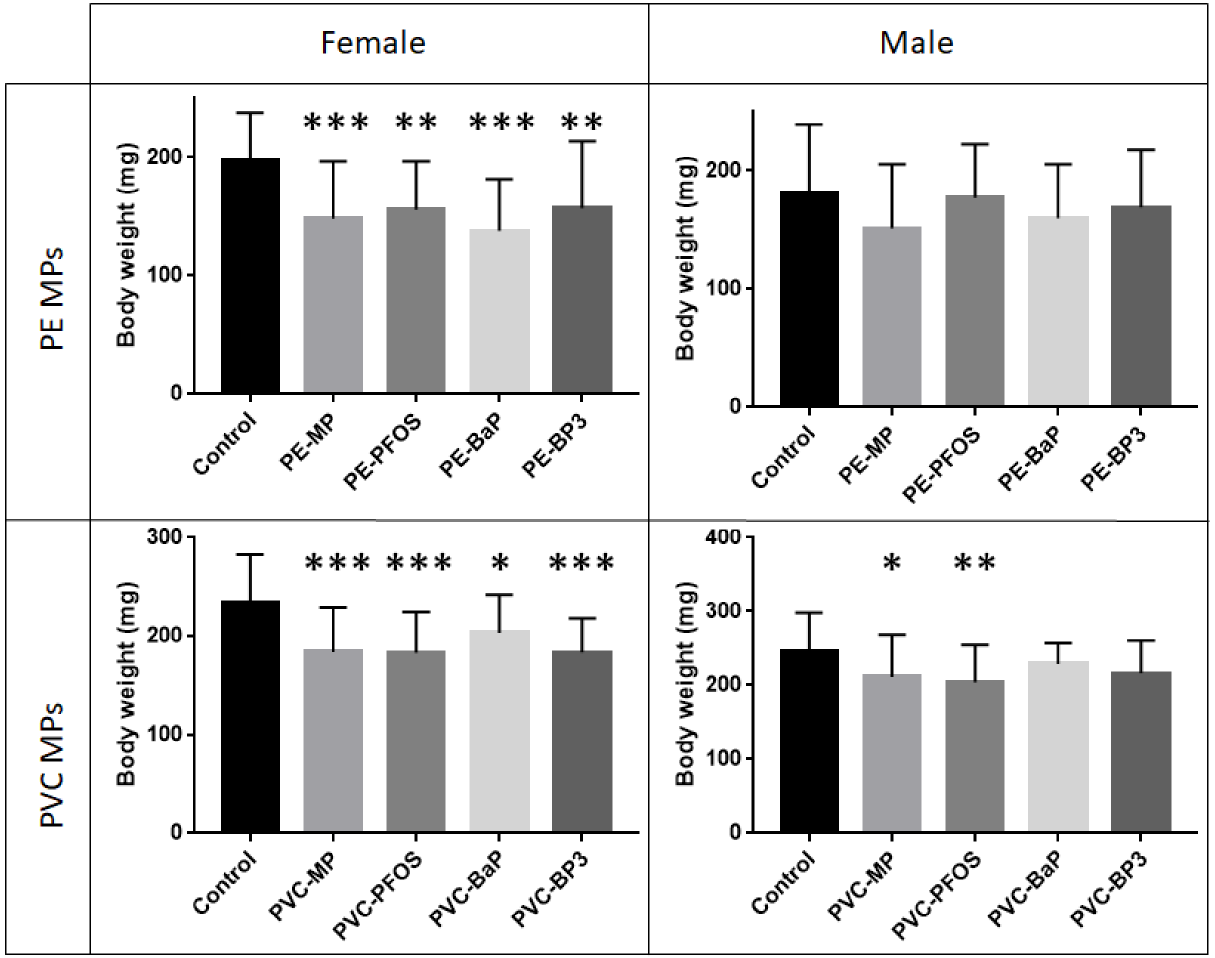
Disruption of growth after 4 months

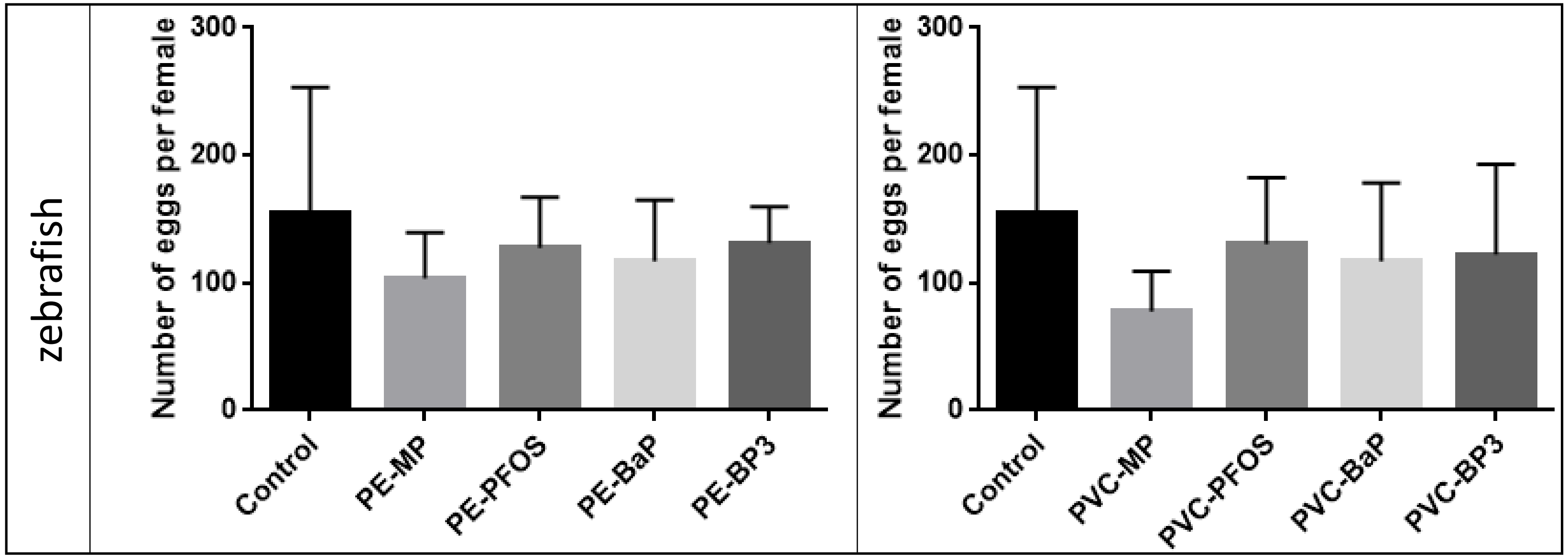




→ Long term decrease in females (20-35%) in both species
→ No difference between MPs

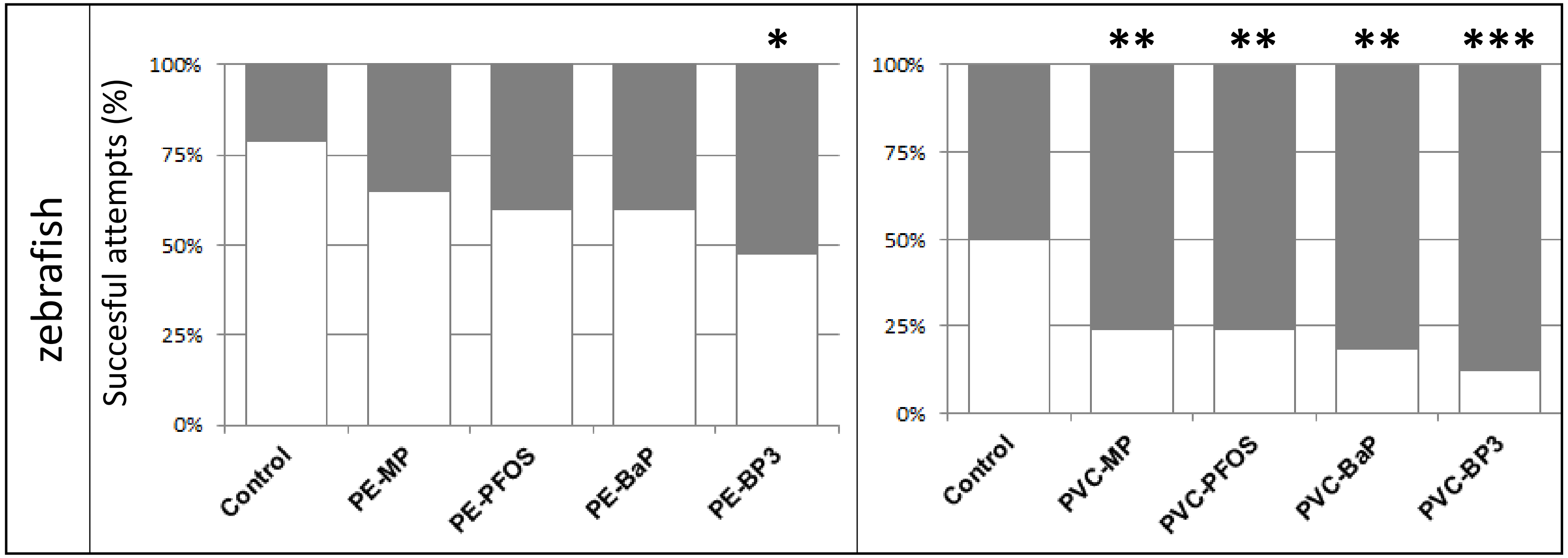
Cormier et al., (2021) *J Hazard Mat* **415**, 125626.

Consequences on reproduction - zebrafish



→ No change in number of eggs per female = spawn size

Consequences on reproduction - zebrafish



→ Massive diminution of the number of spawns
  → Decrease in reproductive output

→ Difference depending on plastic type
→ Difference depending on chemicals

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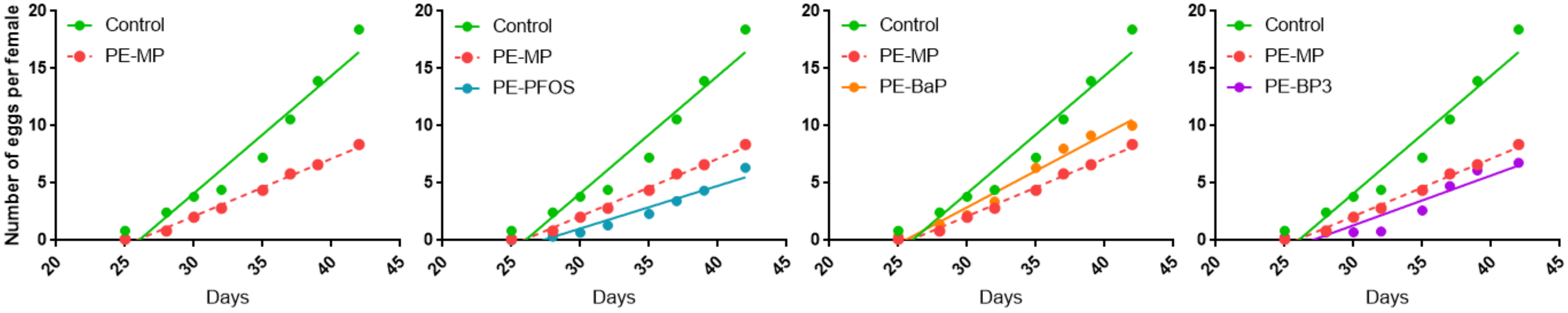
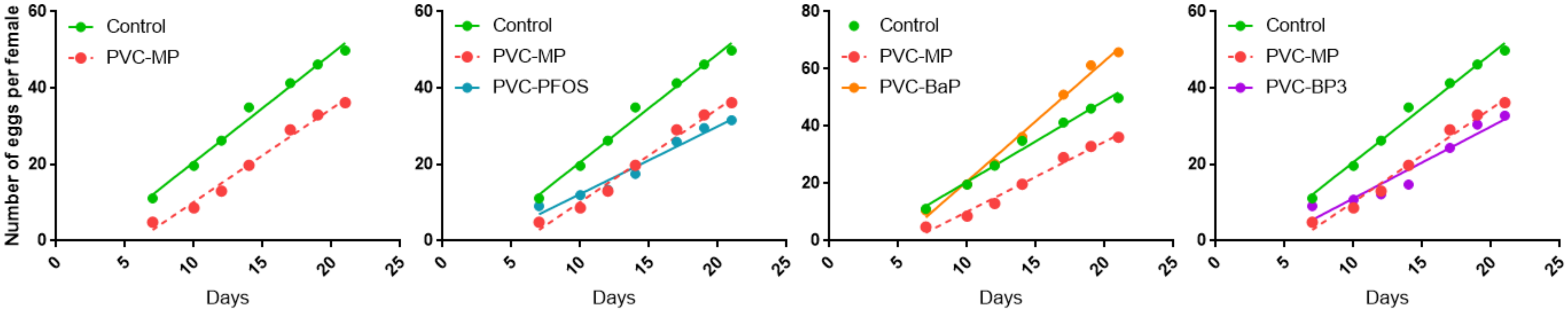
Consequences on reproduction - medaka



PE-MP ↘↘ PE-PFOS ↘ ↘ ↘ PE-BaP ↘ PE-BP3 ↘ ↘



PVC-PFOS ↘ PVC-BaP ↗ PVC-BP3 ↘

→ Significant decrease in eggs number spawned by females (reproductive output)

→ Differences between plastics and pollutants

What about environmental MPs?

➢ MPs sampled on Guadalupe beaches

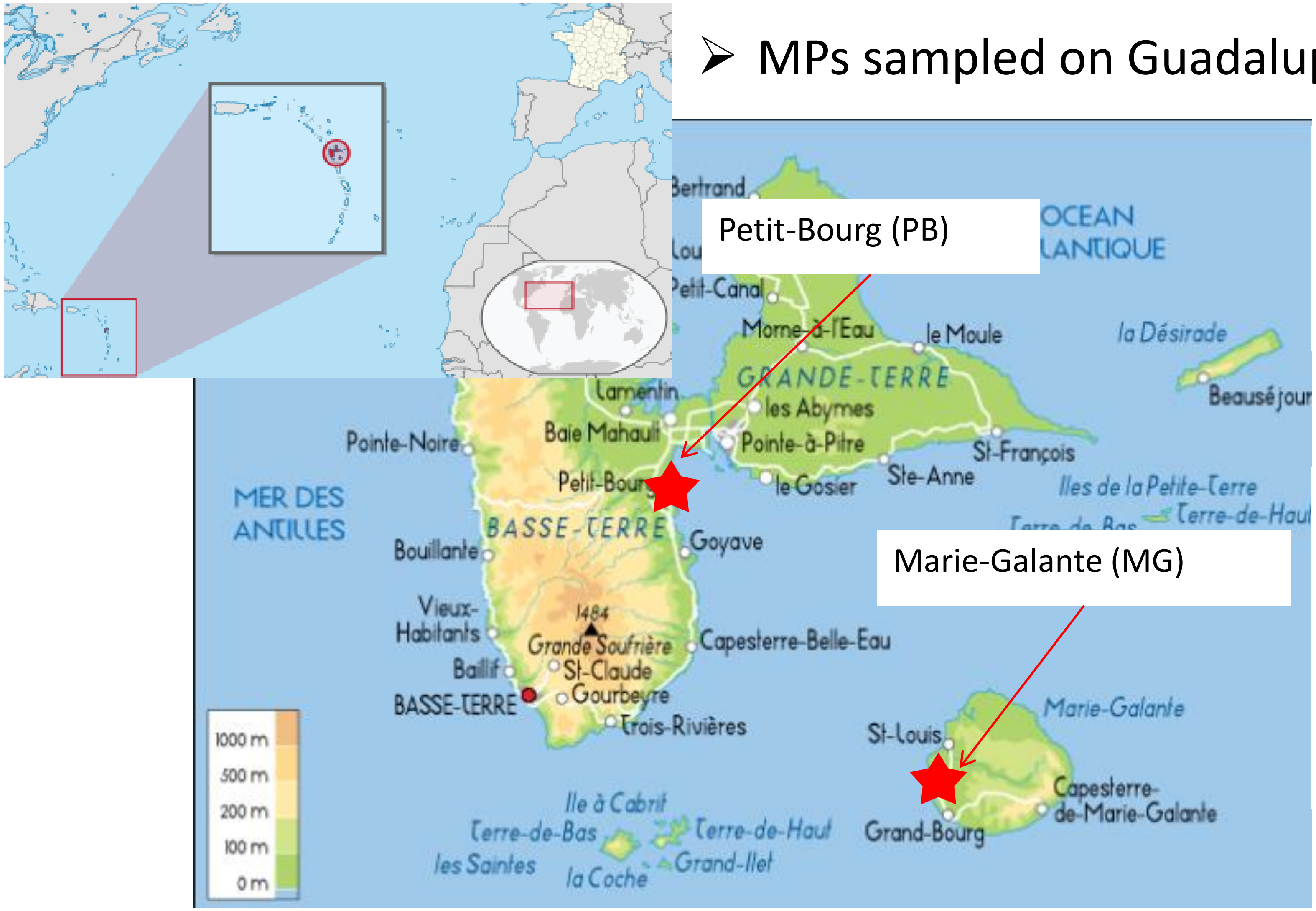


Table 1
Polymer composition of microplastic samples from two different beaches in Guadeloupe archipelago (% in mass).

MPs composition	Marie-Galante	Petit-Bourg
Polyethylene	78.3	74.6
Polypropylene	21.2	24.8
Polystyrene	0.1	0.4
Polyvinyl acetate	0	0.2

→ Similar composition

Table 3
Toxicity of leachates from microplastic samples (<250 µm) of a "virgin" poly-ethylene resin obtained from reference PE (PE Rotogal), Petit-Bourg (PB), Marie-Galante(MG) assessed using the sea urchin (48 h), the jellyfish (48 h) and the zebrafish (96 hpf) embryo-larval assays. Concentrations are expressed in equivalent g of MPs per liter. n.c.: not calculable (EC_{50} >1 g/L).

	MPs sample	NOEC	LOEC	Toxicity units (TU = $1/EC_{50}$)
Sea urchin	PB	1.00	3.33	0.21 (0.18–0.25)
	MG	3.33	10.0	<0.1
Jellyfish	PB	<0.033	<0.033	2 (1–3)
	MG	0.033	0.10	n.c.
Zebrafish	PB	>50.0	>50.0	<0.02
	MG	>50.0	>50.0	<0.02

→ No acute toxicity

Cormier et al., (2021) *Ecotox. Environ. Safe.* **208**, 11665

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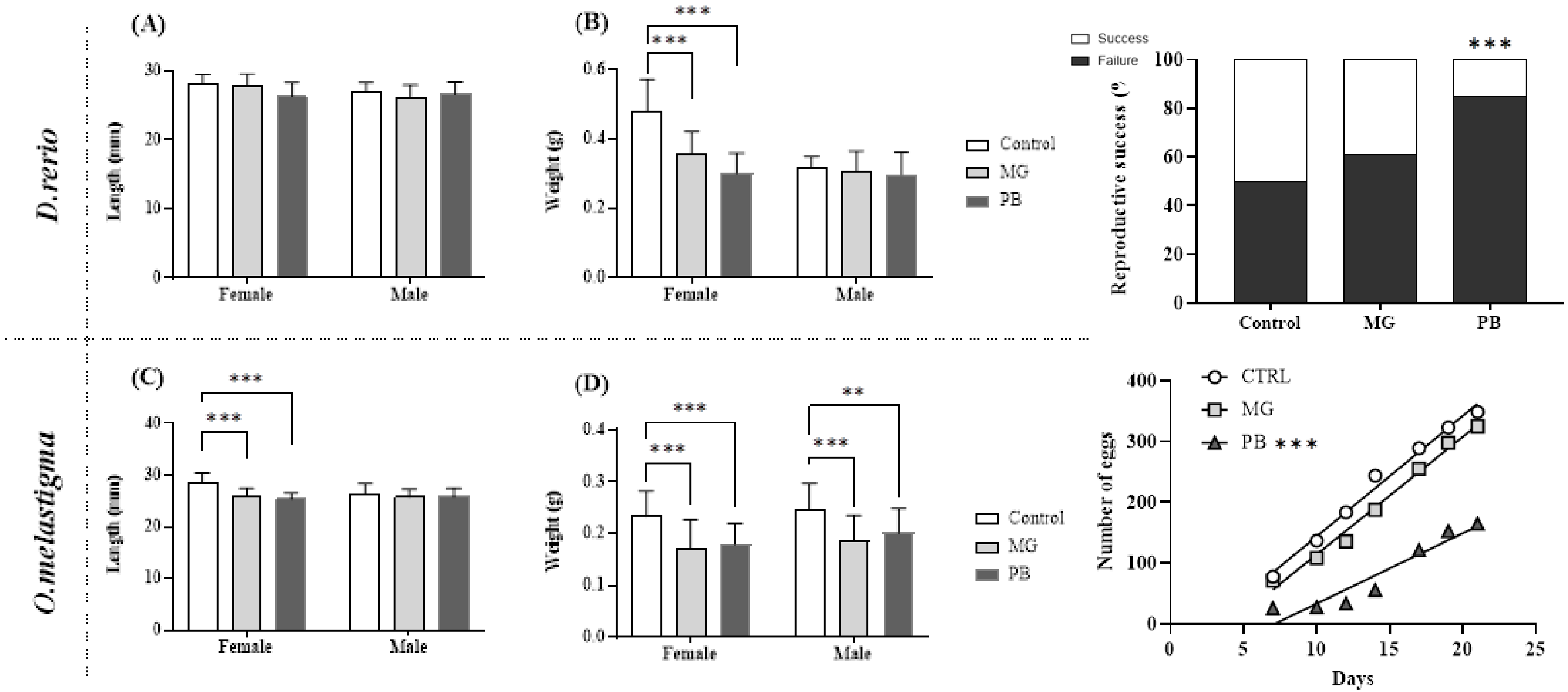
Chronic exposure to Guadalupe MPs



→ Long-term disruption of growth especially in females and independent of the sampling site

→ Disruption of reproduction in fish exposed to PB MPs

Cormier et al., (2022) *Environmental Pollution* **308**, 119721

Chemical load of MG and PB MPs

Non target screening of organic chemicals qualitative results

Organic compounds	MG	PB
tribromophenol	+	+
tribromoanisole	+	+
dichlorinated PCB	+	+
trichlorinated PCB	+	+
dichlorobenzene	+	+
pentachlorobenzene	+	+
chloroacenaphthylene	+	+
trichlorobenzene	**++**	+
bumetrizole	**++**	+
octabenzone	**++**	+
hydrocarbons	+	**++**
alkanes	+	**++**
phenol with an alkane chain	+	**++**
octadecanoic acid	+	**++**

Concentration of trace metal in MG and PB samples.
Results are expressed in µg/ g

Metals	MG	PB
^{63}Cu	31	**85**
^{206}Pb	**102**	18
^{208}Pb	**102**	18
^{66}Zn	26	**292**
^{111}Cd	**222**	9
^{52}Cr	**47**	4.9

→ Hydrocarbons, alkanes and Cu may explain higher toxicity of PB MPs

Conclusions on toxicity

- Depends on the way it is assessed
- **Regulatory tests** often **failed** to reveal toxicity
  - Too short, too insensitive and not necessarily suitable for MPs (e.g. buoyancy, aggregation)
- There are evidences of **toxicity after long-term exposure**
  - Growth (→ particles?) and reproduction (→ chemicals?)
  - Energy unbalance?
  - Potential consequences at population level
- There are evidences of toxicity due to adsorbed pollutants

Still needed - Perspectives

- Validation of the energy unbalance hypothesis
  - Interference with microbiota
- Evaluation of smaller sizes
  - Combination with translocation potential
- Evaluation of other plastic types
  - e.g. fibres, polymers including biodegradable ones
- Interaction with pathogens
  - Role in disease transmission and health issues
- Evaluation of ecological relevance for chemical transfer
- High-throughput but relevant toxicity tests

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Let's keep our fish alive !

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