

PRESENCE, BEHAVIOR AND RISK ASSESSMENT OF PHARMACEUTICALS IN MARINE ECOSYSTEMS (PHARMASEA)



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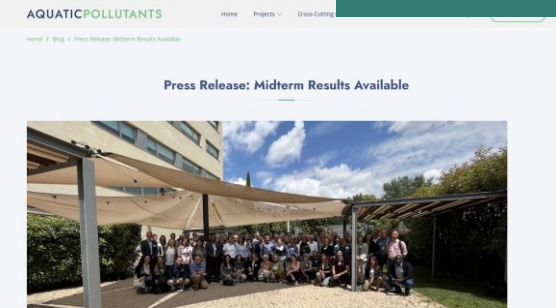
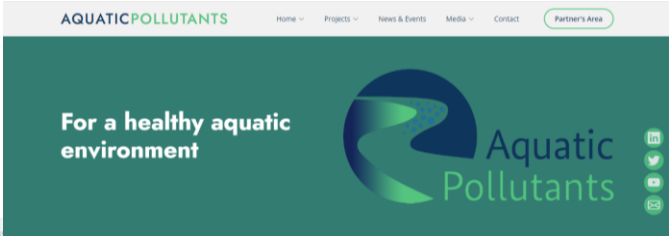


Funded by the Horizon 2020
Framework Programme of the
European Union



PHARMASEA

PRESENCE, BEHAVIOR AND RISK ASSESSMENT OF PHARMACEUTICALS IN MARINE ECOSYSTEMS



18 projects funded by the **AquaticPollutants ERA-NET**

Risks caused by Contaminants of Emerging Concern (CECs) and pathogens present in water resources.



UNIVERSITÀ
POLITECNICA
DELLE MARCHE

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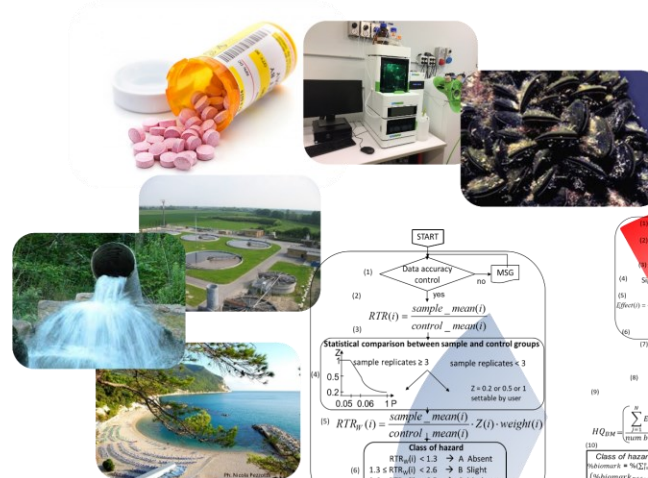
PI: Prof.
Victor E. Leon

MAIN OBJECTIVES

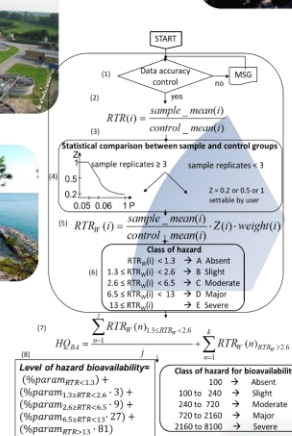
- Qualitative and quantitative characterization of the presence, distribution, fate and behavior of APIs in seawater, sediments and biota
- Mechanisms of action and ecotoxicological adverse effects from molecular to organism level
- Harmonization of comprehensive and site-specific procedure for APIs Environmental Risk Assessment
- Raise awareness on the environmental relevance of APIs in aquatic ecosystems; Promote participative approach and citizens engagement

**NEED OF
EFFECT-BASED RISK ASSESSMENT
PROCEDURES TO SUPPORT THE
IMPLEMENTATION OF RELEVANT EU
POLICIES**

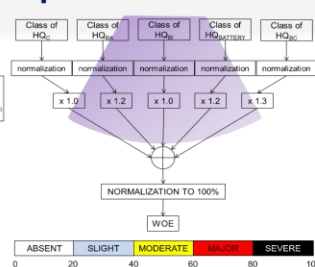
WP1 Pharmaceuticals in European coasts



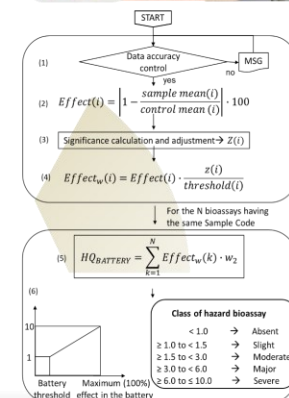
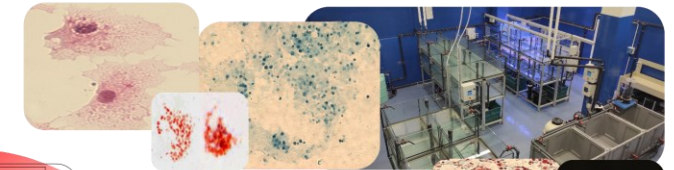
WP5 Project and data management



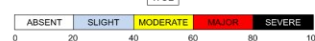
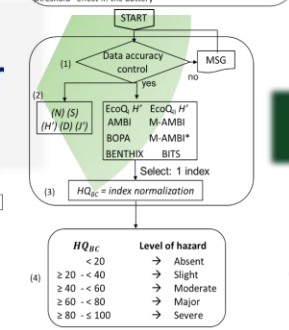
WP3 Weight of evidence model for pharmaceuticals

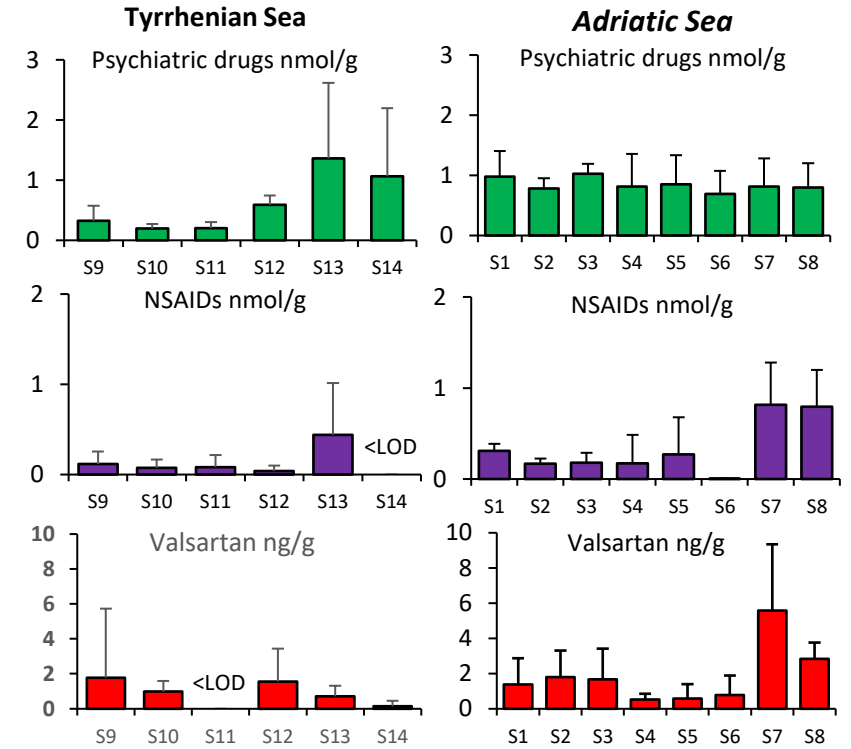
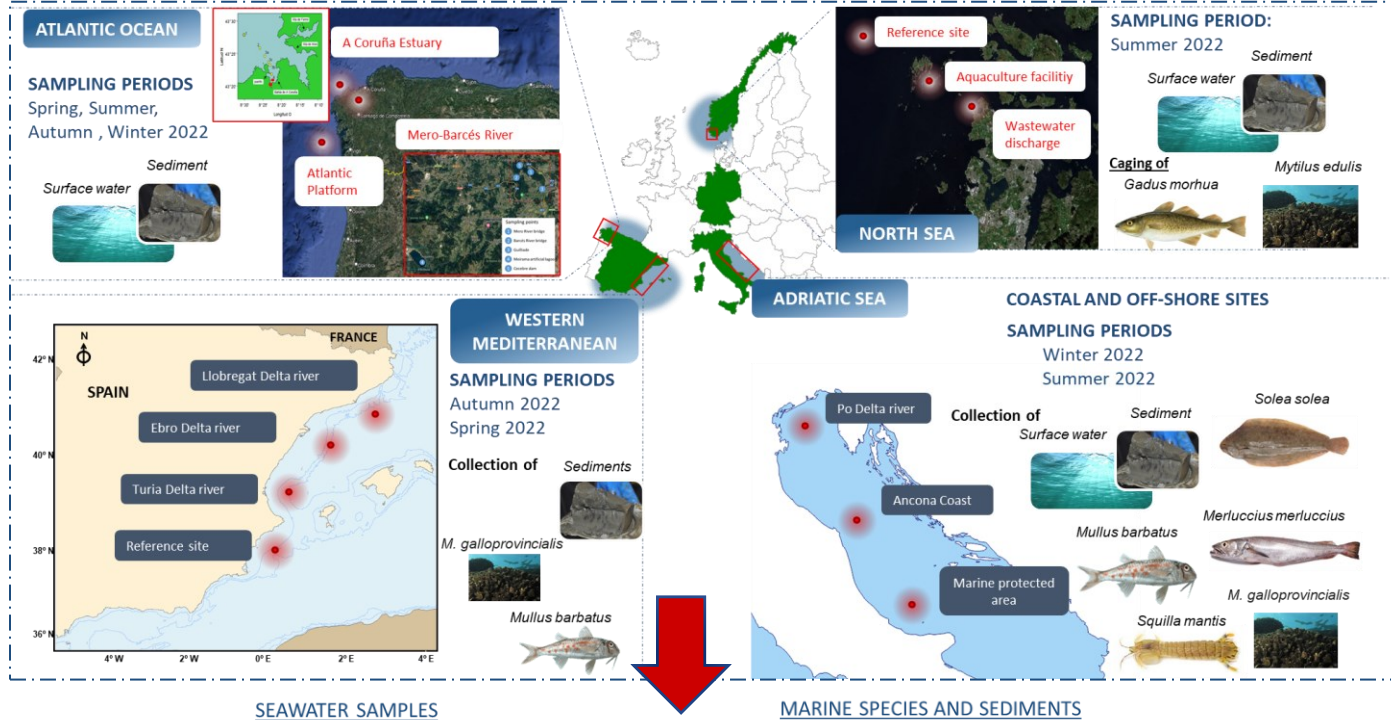


WP2 Effects of pharmaceuticals in aquatic species

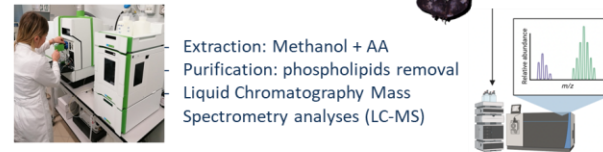
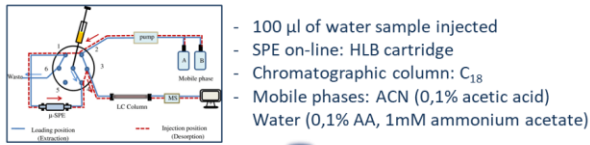


WP4 Dissemination and stakeholder engagement





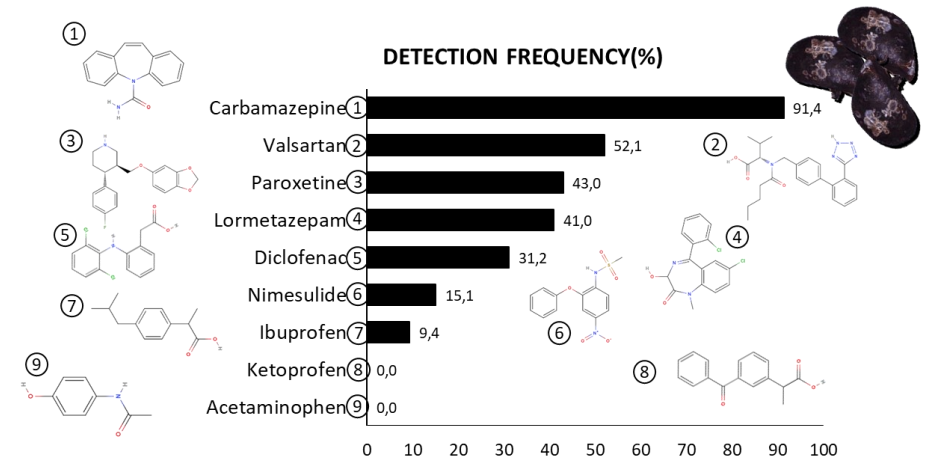
SEAWATER SAMPLES
 ANALYTICAL METHODOLOGY BASED ON ON-LINE μ SPE HPLC-MS-MS



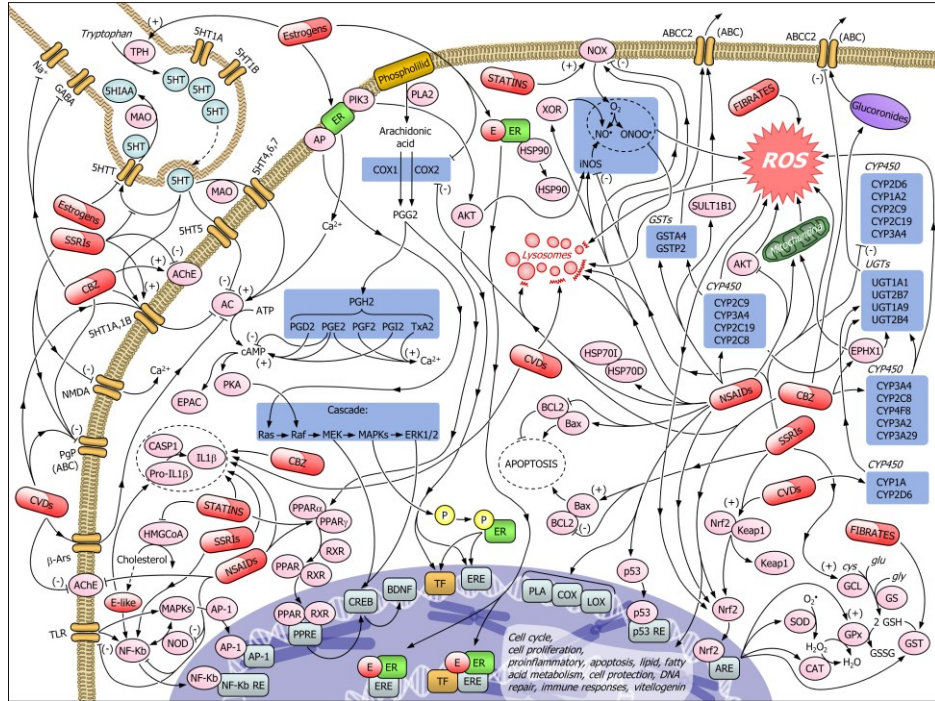
56 PHARMACEUTICALS BELONGING TO 7 THERAPEUTIC CLASSES:

MUSCULOSKELETAL	NERVOUS SYSTEM	CARDIOVASCULAR
Analgesics: Acetaminophen, 4-formilantipyrine, 4-Acetamidoantipyrine, Antypirine, Propyphenazone, Xylazine, NSAIDs: Diclofenac, Ibuprofen, Ketoprofen, Naproxen, Nimesulide,	SSRIs: Citalopram, Fluoxetine, Paroxetine, Sertraline NSRIs: Duloxetine, Venlafaxine, Desmethylvenlafaxine Antiepileptics: Carbamazepine, 10,11,dih- 10, 11diOH carbamazepine, Primidone	Calcium-antagonists: Amlodipine, Diltiazem, β- blockers: Atenolol, Bisoprolol, Carazolol, Metoprolol, Propanolol, Sotalol ACE-inhibitors: Ramipril, Verapamil Sartans: Valsartan Diuretics: Furosemide, Hydrochlorothiazide Antidiabetics: Metformine
ALIMENTARY TRACT Omeprazole, Pantoprazole	ANTIBIOTICS Azythromycine, Clarithromycin, Erythromycin, Erythromycin anhydrate, N4-acetyl sulfamethoxazole, Roxithromycin, Sulfadimethoxine, Sulfamerazine, Sulfamethoxazole, Trimethoprim, Tylosin	LIPID REGULATORS Fibrates: Bezafibrate, Clofibrac acid, Fenofibrate, Gemfibrozil, Statins: Atorvastatin, Rosuvastatin,
SYSTEMIC HORMONAL L-tyroxine		

MUSCULOSKELETAL	NERVOUS SYSTEM	CARDIOVASCULAR
Analgesics: Acetaminophen NSAIDs: Diclofenac, Ibuprofen, Ketoprofen, Naproxen, Nimesulide	SSRIs: Fluoxetine, Paroxetine, Sertraline NSRIs: Venlafaxine, Desmethylvenlafaxine Antiepileptics: Carbamazepine, 10,11,epoxyde carbamazepine Anxiolitics: Lorazepam, Lormetazepam	β- blockers: Atenolol, Propanolol ACE-inhibitors: Ramipril, Verapamil Sartans: Valsartan
ANTIBIOTICS Ciprofloxacin, Sulfamethoxazole		LIPID REGULATORS Fibrates: Gemfibrozil Statins: Atorvastatin

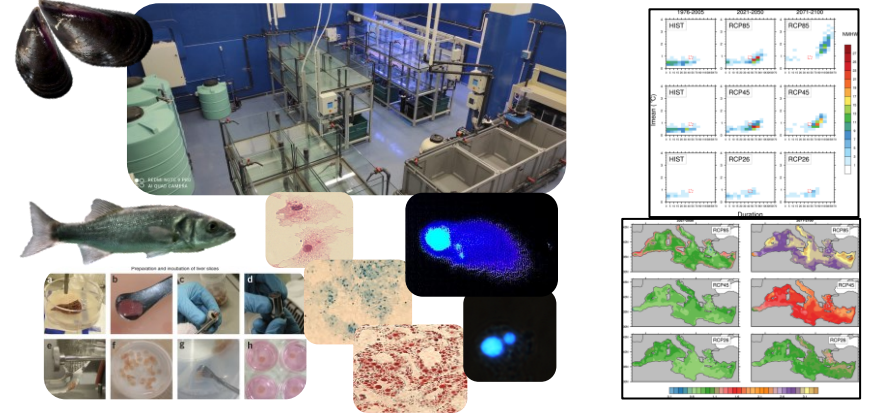


PRIORITY 1- CORE GROUP OF INTEREST

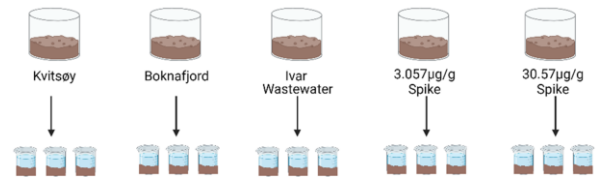
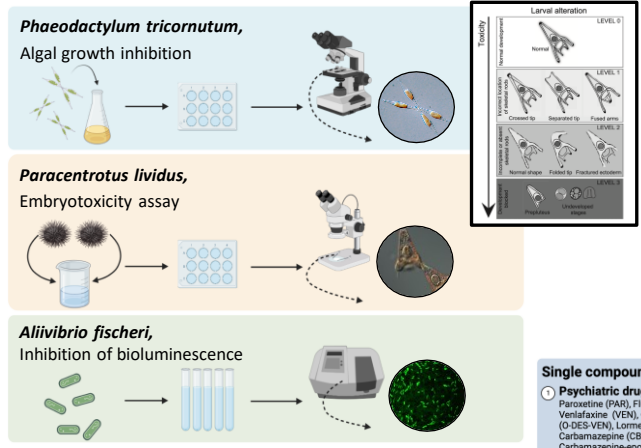


IN VIVO LABORATORY EXPERIMENTS, *Mytilus galloprovincialis*:

PSYCHIATRIC DRUGS
 CARDIOVASCULAR DRUGS
 LIPID LOWERING AGENTS
 NSAIDs

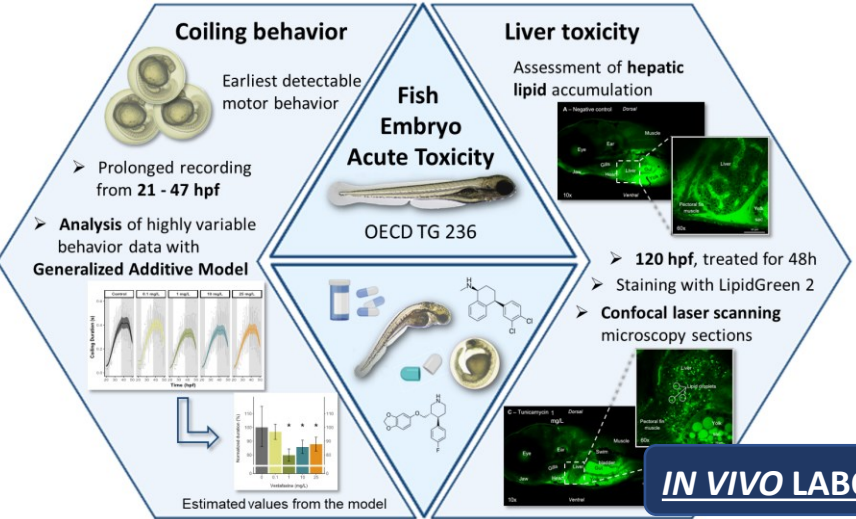
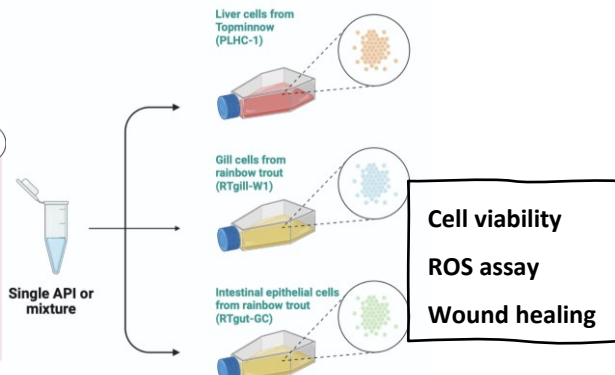


IN VIVO Ecotoxicological bioassays



IN VITRO EXPERIMENTS: Fish cell lines

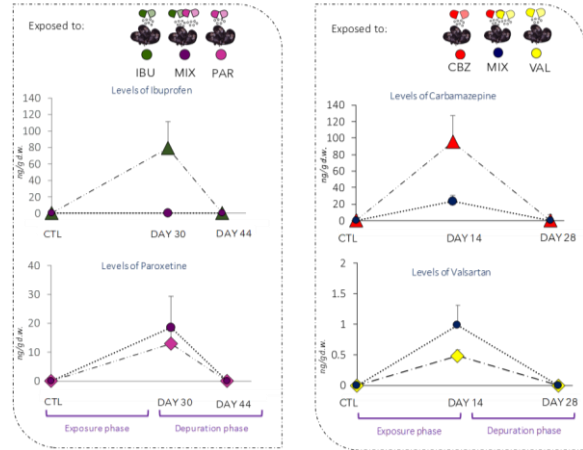
- Single compounds**
- 1 **Psychiatric drugs:** Paroxetine (PAR), Fluoxetine (FLU), Venlafaxine (VEN), O-Desmetil-Venlafaxine (O-DES-VEN), Lormetazepam (LORM), Carbamazepine (CBZ), Carbamazepine-epoxide (CBZ-EP)
 - 2 **Cardiovascular drugs:** Atenolol (ATE), Ramipril (RAM), Valsartan (VAL)
 - 3 **Lipid lowering agents:** Gemfibrozil (GEM)
 - 4 **Non Steroidal-Antinflammatory drugs:** Diclofenac (DIC), Ibuprofen (IBU), Naproxen (NAP), Nimesulide (NIM)
- Mixtures**
- 1 **Mixture 1:** IBU + PAR
 - 2 **Mixture 2:** CBZ + VAL
 - 3 **Mixture 3:** VEN + NAP
 - 4 **Mixture 4:** IBU + PAR + CBZ + VAL
 - 5 **Mixture 5:** IBU + PAR + CBZ + VAL + LORM + DIC + NIM
 - 6 **Mixture 6:** VAL + ATE + GEM + RAM



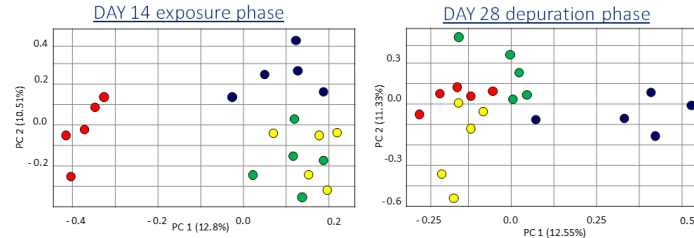
IN VIVO LABORATORY EXPERIMENTS, *Danio rerio*:

Cell viability
 ROS assay
 Wound healing

DRUG ACCUMULATION



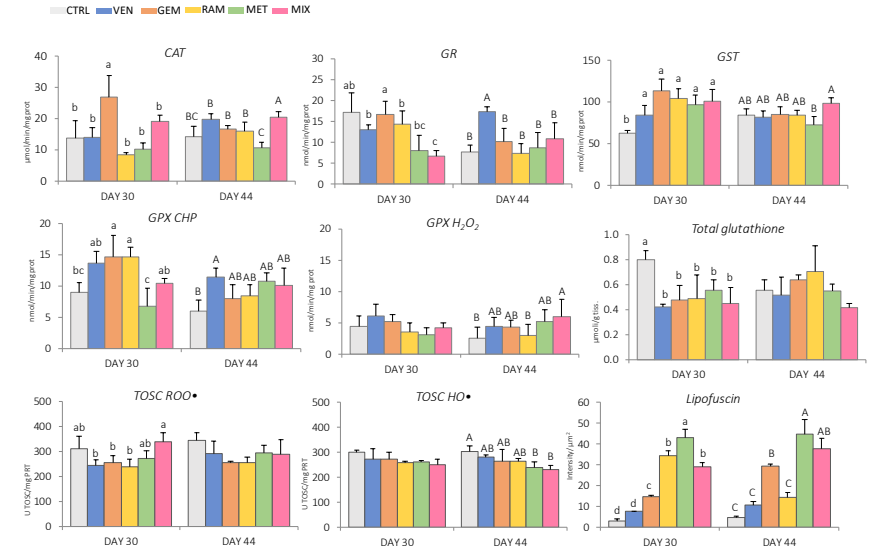
CHANGES IN TRANSCRIPTIONAL PROFILE



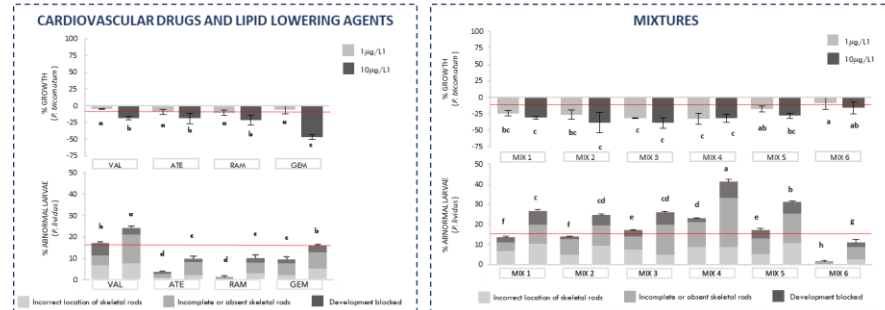
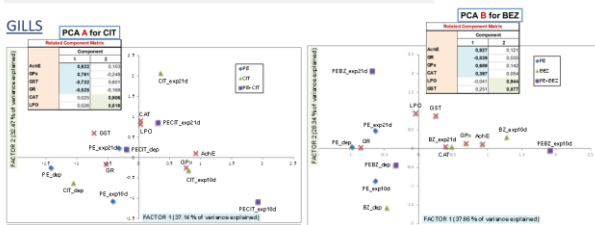
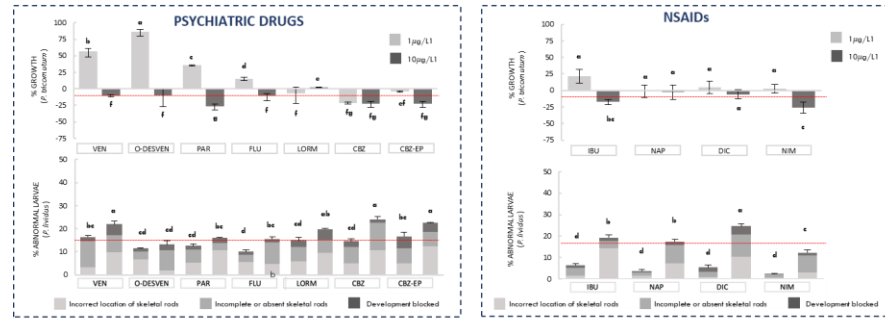
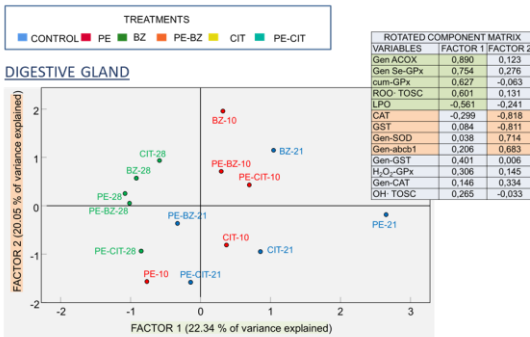
MODULATED PATHWAYS

- Neurological responses (red dot)
- Cell cycle (blue dot)
- Drug metabolism (red dot)
- Immune system (blue dot)
- Oxidative metabolisms (red dot)
- Immune system (yellow dot)
- Oxidative metabolism (yellow dot)
- Immune system (blue dot)
- Oxidative metabolism (blue dot)
- Lipid metabolism (blue dot)

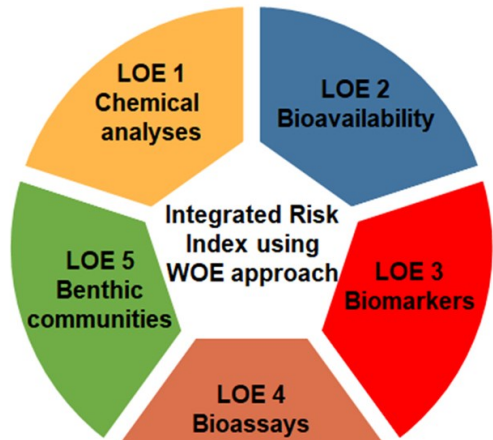
BIOMARKERS



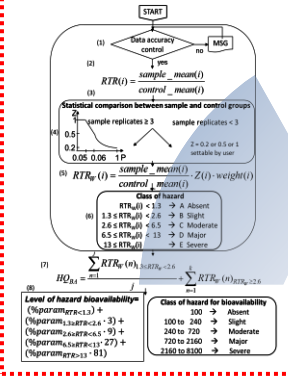
ECOTOXICOLOGICAL BIOASSAYS



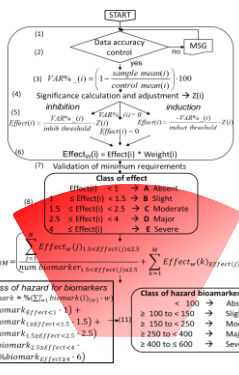
- Competing mechanisms in mixture-exposed organisms
- Metabolism/excretion processes
- Immune parameters as the most sensitive in response to pharmaceuticals and mixtures.
- Impaired lipid metabolism and redox homeostasis.
- Modulation of neurotoxicity and DNA damage.
- Effects still evident after the end of depuration period
- Low acute toxicity



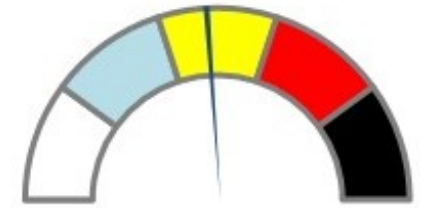
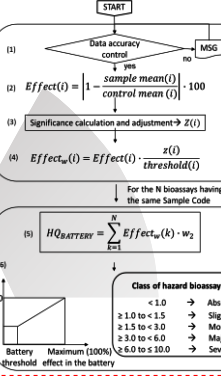
LOE BIOAVAILABILITY



LOE BIOMARKERS



LOE BIOASSAYS



SYNTHETIC RISK INDEX



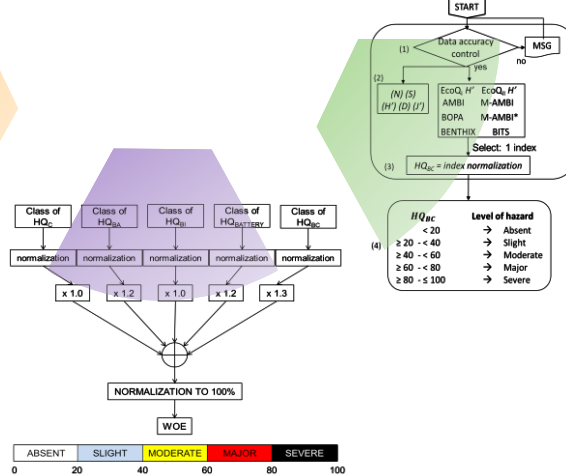
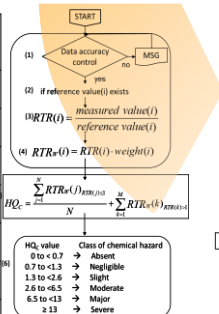
EXPOSURE PHASE



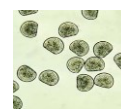
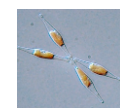
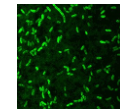
DEPURATION PHASE

Treatment	Loe 2 Bioavailability	Loe 3 Biomarker	WOE INTEGRATION	Level of Risk
Cbz	Severe	Moderate	MAJOR	
Val	Moderate	Moderate	SLIGHT	
Cbz + Val	Major	Slight	SLIGHT	
Ibu	Major	Moderate	MODERATE	
Par	Major	Moderate	MODERATE	
Ibu + Par	Major	Moderate	MODERATE	
Ven	Ongoing	Moderate		
Gem	Ongoing	Moderate		
Ram	Ongoing	Moderate		
Met	Ongoing	Moderate		
Mixture	Ongoing	Moderate		

Treatment	Loe 2 Bioavailability	Loe 3 Biomarker	WOE INTEGRATION	Level of Risk
Cbz	Absent	Moderate	SLIGHT	
Val	Absent	Slight	ABSENT	
Cbz + Val	Absent	Slight	ABSENT	
Ibu	Absent	Moderate	SLIGHT	
Par	Absent	Moderate	SLIGHT	
Ibu + Par	Absent	Absent	SLIGHT	
Ven	Ongoing	Moderate		
Gem	Ongoing	Moderate		
Ram	Ongoing	Moderate		
Met	Ongoing	Moderate		
Mixture	Ongoing	Moderate		



Sample	Level of hazard	Sample	Level of hazard	Sample	Level of hazard	Sample	Level of hazard
VEN 1 µg/L	ABSENT	VAL 1 µg/L	ABSENT	IBU 1 µg/L	ABSENT	MIX 1 1 µg/L	ABSENT
VEN 10 µg/L	ABSENT	VAL 10 µg/L	ABSENT	IBU 10 µg/L	ABSENT	MIX 1 10 µg/L	SLIGHT
O-DESVEN 1 µg/L	ABSENT	ATE 1 µg/L	ABSENT	NAP 1 µg/L	ABSENT	MIX 2 1 µg/L	ABSENT
O-DESVEN 10 µg/L	ABSENT	ATE 10 µg/L	ABSENT	NAP 10 µg/L	ABSENT	MIX 2 10 µg/L	MODERATE
PAR 1 µg/L	ABSENT	RAM 1 µg/L	ABSENT	DIC 1 µg/L	ABSENT	MIX 3 1 µg/L	SLIGHT
PAR 10 µg/L	SLIGHT	RAM 10 µg/L	ABSENT	DIC 10 µg/L	ABSENT	MIX 3 10 µg/L	MODERATE
FLU 1 µg/L	ABSENT	GEM 1 µg/L	ABSENT	NIM 1 µg/L	ABSENT	MIX 4 1 µg/L	SLIGHT
FLU 10 µg/L	ABSENT	GEM 10 µg/L	MODERATE	NIM 10 µg/L	ABSENT	MIX 4 10 µg/L	MODERATE
LORM 1 µg/L	ABSENT					MIX 5 1 µg/L	ABSENT
LORM 10 µg/L	ABSENT					MIX 5 10 µg/L	SLIGHT
CBZ 1 µg/L	ABSENT					MIX 6 1 µg/L	ABSENT
CBZ 10 µg/L	SLIGHT					MIX 6 10 µg/L	ABSENT
CBZ-EP 1 µg/L	ABSENT						
CBZ-EP 10 µg/L	SLIGHT						

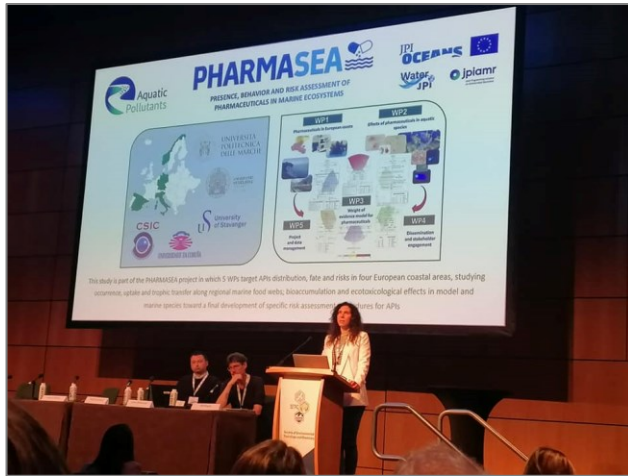


- Chronic conditions caused a level of risk between “SLIGHT” and “MODERATE”
- Evidence of interaction in Mixtures treatments
- Decrease of risk level after the depuration period

- Lower acute toxicity compared to chronic exposure conditions
- Higher risk of mixtures compare to single pharmaceuticals
- Lack of clear dose-dependent effects

SCIENTIFIC COMMUNITY

Participation to 10 Scientific Conferences
more than 30 contributions Platform/Posters



Website and social media



PharmaSea
@PharmaSeaAquatPoll

CITIZENS

Science meeting, exhibitions
Citizen engagement



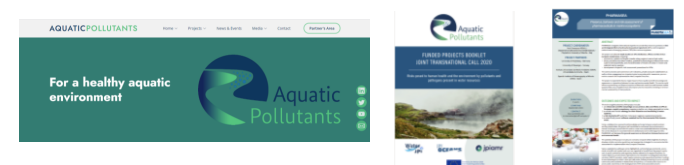
TV programs,
Newspapers, Magazines



Survey for citizens and Scientific Community (SETAC)

- Increase awareness
- Counteract wrong infos
- Disseminate good practices
- Mitigation strategies

Collaboration with Aquatic Pollutants Transfer Project



Environmental Pollution 300 (2022) 119076
Contents lists available at ScienceDirect
Environmental Pollution
journal homepage: www.elsevier.com/locate/envpol

Marine heatwaves hamper neuro-immune and oxidative tolerance toward carbamazepine in *Mytilus galloprovincialis*^a
Alessandro Nardi ^{a,1}, Marica Mezzelani ^{a,1}, Silvana Costa ^a, Giuseppe d'Errico ^a, Maura Benedetti ^a, Stefania Gorbi ^a, Rosa Freitas ^a, Francesco Regoli ^a

Environmental Research
journal homepage: www.elsevier.com/locate/envres

Bioaccumulation and fate of pharmaceuticals in a Mediterranean coastal lagoon: Temporal variation and impact of a flash flood event

Science of the Total Environment
journal homepage: www.elsevier.com/locate/scitotenv

Mixtures of environmental pharmaceuticals in marine organisms: Mechanistic evidence of carbamazepine and valsartan effects on *Mytilus galloprovincialis*
Marica Mezzelani ^a, Luca Peruzzo ^b, Giuseppe d'Errico ^a, Massimo Milan ^b, Stefania Gorbi ^a, Francesco Regoli ^{a,b}
M. García-Pimentel ^a, J.A. Campillo ^a, J.M. Castaño-Ortiz ^a, M. Llorca ^c, V.M. León ^a



Pharmaceuticals in marine environment

PHARMASEA 

Aquatic Pollutants | OCEANS | Water JPI | Jpiamr | 

The main source for water contamination by pharmaceuticals is represented by: *

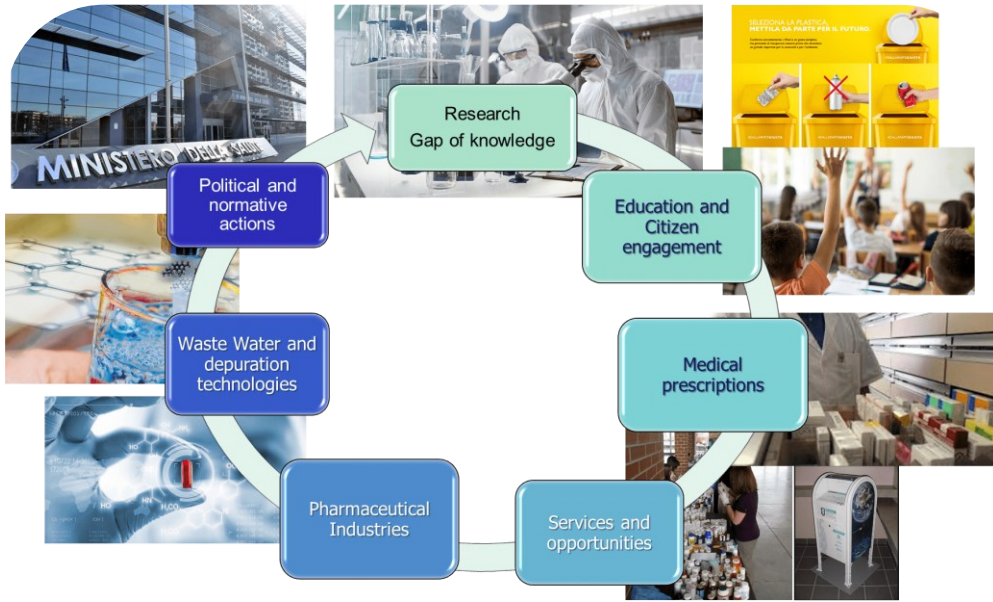
- Landfills, in which large amount of toxic leachates are formed during rainy days, lastly ending up in water bodies
- Humans and animals who take drugs, which are largely eliminated as parental compounds or metabolites in an active form in urine and faeces, lastly ending up in sewages
- Tourists, who do not dispose drugs in the appropriate bins in seaside resorts
- The pharmaceutical industry, which throws expired or unsold drugs into rivers lastly ending up in coastal areas

Are urban sewage treatment plants able to remove drug residues? *

- yes, with a removal efficiency of 100%
- only with a removal efficiency of 50%

PRIVATE COMPANIES

REGULATORY BODIES

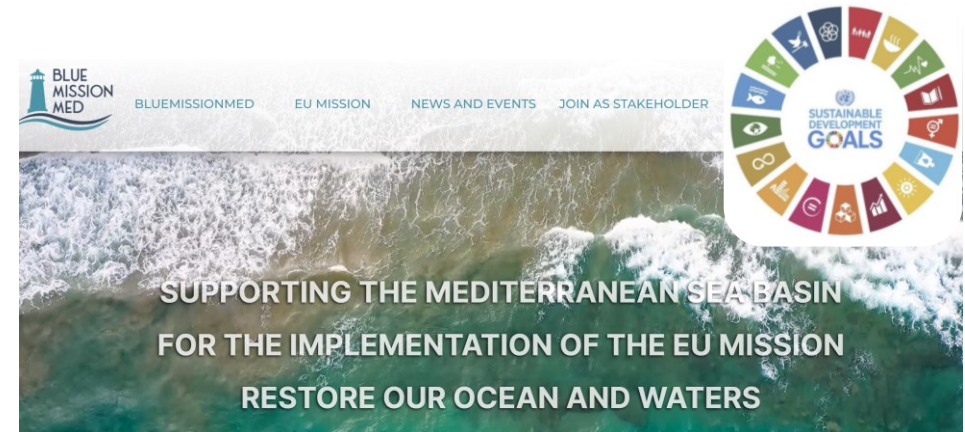
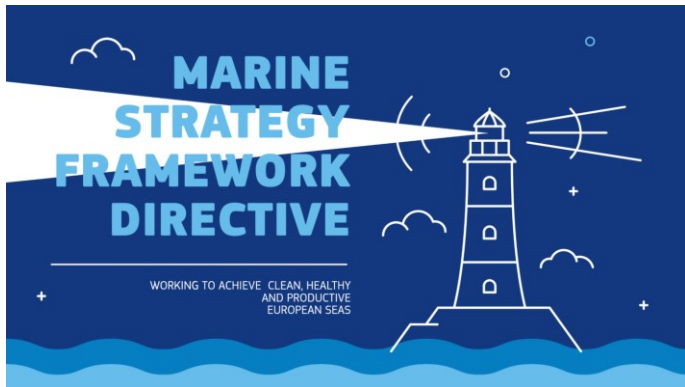
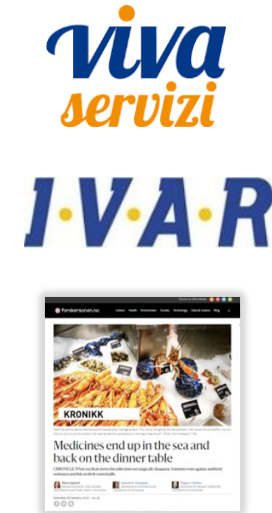


CIRCULAR KNOWLEDGE APPROACH

Pharma industries



WWTPs companies



A special thank to Aquatic Pollutants, JPIs and National Funding Agencies



Funded by the Horizon 2020
Framework Programme of the
European Union



MINISTERO DELL'ISTRUZIONE, DELL'UNIVERSITÀ E DELLA RICERCA



**PLEASE, FILL OUT OUR
SURVEY!**

YOUR OPINION MATTERS!



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