

INTERNATIONAL CONFERENCE ON MICROPLASTIC POLLUTION IN THE MEDITERRANEAN SEA



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**MED** INTERNATIONAL CONFERENCE ON MICROPLASTIC POLLUTION IN THE MEDITERRANEAN SEA



# **BOOK OF ABSTRACTS**

# INTERNATIONAL CONFERENCE ON MICROPLASTIC POLLUTION IN THE MEDITERRANEAN SEA CAPRI SEPTEMBER 26<sup>TH</sup>- 29<sup>TH</sup> 2017 HOTEL LA RESIDENZA - CAPRI (NA) - ITALY

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Paul Connett François Galgani	Zero Waste Movement Founder, <b>USA</b> French Institute for Research and Exploitation of the Sea, <b>France</b>	
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Gunter Pauli	Zero Emissions Research and Initiatives, Belgium	
Lola Rodriguez	Leitat Technological Center, <b>Spain</b>	
<b>Richard C. Thompson</b>	University of Plymouth, <b>UK</b>	
Maria Westerbos	Plastic Soup Foundation, The Netherlands	

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# ABOUT

Following the conclusion of the Project LIFE13 ENV/IT/001069 – Mermaids: "Mitigation of microplastics impact caused by textile washing processes", the Organizing and Scientific Committees are pleased to invite the scientific community and stakeholders to the "International Conference on Microplastic Pollution in the Mediterranean Sea" ( $\mu$  MED) hosted in Capri (NA), Italy, on September 26-29<sup>th</sup> 2017.

The conference will focus on one of the most actual and dangerous source of marine pollution. In fact, even though plastic pollution is a well-known worldwide problem, the discovery of the presence of microplastics or even nanoplastics in seas and oceans represents an unexpected and very alarming turn in this environmental challenge. Microplastics are defined as plastic fragments typically smaller than 5 mm, able to pass through wastewater treatments plants, accumulating on shorelines and eventually reaching marine ecosystems. There, such fragments remain unaltered, representing a serious risk not only for marine flora and fauna, but also for human health.

Regarding the impact of these pollutants, recent studies have proved that the Mediterranean Sea is one of the most worrisome region of the world, showing a microplastic concentration even higher than that of the Nord Pacific area (Plastic soup vortex).

Against this background,  $\mu$ MED conference aims to share current knowledge, identify issues and propose solutions and actions to face and mitigate this environmental threat.

# TOPICS

#### THE CONFERENCE WILL BE FOCUSED ON THE FOLLOWING TOPICS:

- Microplastics and marine environment
- Microplastics in the Mediterranean Sea
- Sources of microplastics
- Degradation of microplastics in marine environment
- Impact of microplastics on marine life
- Microplastics: from marine pollution to human food chain
- Socioeconomic impacts of microplastics
- Mitigation of microplastic impact
- Biodegradable microplastics
- Solutions and future steps

# PROGRAMME

#### 26/09/2017 - AFTERNOON 5.00-7.00 PM Registration - Welcome - Wine & Cheese 27/09/2017 - MORNING SESSION 9.00-9.30 AM Registration Institute for Polymers, Composites Maurizio Avella and Biomaterials - CNR, Italy **Department of Chemical Sciences and** Maurizio Peruzzini Materials Technologies - CNR. Italy 9.30-10.00 AM Institute for Polymers, Composites Opening, welcome and introduction **Cosimo Carfagna** and Biomaterials - CNR, Italy Institute for Macromolecular Studies -Salvatore lannace CNR. Italy Federico di Penta Marevivo, Italy MICROPLASTICS AND MARINE ENVIRONMENT Chairman: Mario Malinconico Getting to the front end of microplastic 10.00-10.30 AM Paul Connett Zero Waste Movement, USA pollution of the sea 10.30-11.00 AM Cyrill Gutsch Parley for the Oceans, USA The air we breathe 11.00-11.30 AM **COFFEE BREAK** Euro-Mediterranean Center on Climate Change and Impacts on the 11.30-12.00 PM Antonio Navarra **Climate Change, Italy** Mediterranean Basin To what extent microplastic from the open 12.00-12.15 PM Alexandra Ter Halle Université de Toulouse, France ocean are weathered? Wintertime microplastics in different depth 12.15-12.30 PM Lehtiniemi Maiju SYKE Marine Research Centre, Finland layers in the northern Baltic Sea Challenging the microplastic extraction from 12.30-12.45 PM Mikaël Kedzierski Université Bretagne Sud, France sandy sediments Are our synthetic fabrics released into the marine environment? Evidences on 12 45-1 00 PM Raquel Villalba Leitat Technological Center, Spain microplastics pollution in waste water

1.00-2.30 PM **LUNCH** 

27/09/2017 - AFTERNOON SESSION

#### MICROPLASTICS IN THE MEDITERRANEAN SEA Chairwoman: Lola Rodriguez

coming from our laundry

How concerned should we be about Richard C. Thompson Plymouth University, UK 2.30-3.00 PM microplastics? Foundation Euro-Mediterranean Towards 3D modeling the plastic marine 3 00-3 15 PM Svitlana Liubartseva debris in the Mediterranean Centre on Climate Change, Italy Institute for Coastal Marine Microplastics in the Mediterranean Sea and Giuseppe Andrea 3.15-3.30 PM de Lucia Environment - CNR, Italy impacts on biota

			PROGRAMME
3.30-3.45 PM	Stefania Gorbi	Polytechnic University of Marche, Italy	Distribution of microplastics in Adriatic food webs
3.45-4.00 PM	Manuela Piccardo	University of Trieste, Italy	Preliminary assessment of microplastic accumulation in wild Mediterranean fish species
4.00-4.30 PM	COFFEE BREAK		
4.30-5.00 PM	François Galgani	Institut Français de Recherche pour l'Exploitation de la Mer, France	An overview of plastic pollution in the Mediterranean Sea: the importance of microplastics
5.00-5.15 PM	Noam Van Der Hal	University of Haifa, Israel	A temporal survey of microplastics ingested by herbivorous Eastern Mediterranean siganids
5.15-5.30 PM	Natascha Schmidt	Aix Marseille Université, France	Zooplankton and plastic additives – insights into the chemical pollution of the low-trophic level of the Mediterranean marine food web
5.30-5.45 PM	Mel Constant	Université de Perpignan, France	Floating microplastics in the NorthWestern Mediterranean Sea: strong temporal and spatial heterogeneities
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#### M POSTER SESSION

### 28/09/2017 - MORNING SESSION

### IMPACT OF MICROPLASTICS ON HUMAN AND MARINE LIFE / DEGRADATION OF MICROPLASTICS IN MARINE ENVIRONMENT

Chairwoman: Raffaella Mossotti

9.30-10.00 AM	Maria Cristina Fossi	University of Siena, Italy	Impact of microplastics on Mediterranen biodiversity
10.00-10.15 AM	John van den Hoff	Australian Antarctic Division, Australia	Size selective feeding by mesopelagic fish can change sea-surface microplastic abundance: impact of marine life on microplastic abundance
10.15-10.30 AM	Jean-François Ghiglione	Observatoire Océanologique de Banyuls, France	Impact of microplastics on marine microbial life
10.30-10.45 AM	Stefania Piarulli	University of Bologna, Italy	Microplastics occurrence in the gastrointestinal tract of key marine invertebrates at different trophic levels from the North Adriatic coast
10.45-11.00 AM	Dror Angel	University of Haifa, Israel	Microplastics as ephemeral and possibly preferred substrates for planktonic cnidarian planulae
11.00-11.30 AM	COFFEE BREAK		
11.30-12.00 PM	Roberto Danovaro	Stazione Zoologica Anton Dohrn, Italy	Impact of microplastic on the deep sea
12.00-12.15 PM	Filipa Bessa	Universidade de Coimbra, Portugal	Microplastics in juvenile commercial fish from a natural estuarine environment
12.15-12.30 PM	Christophe Brunet	Stazione Zoologica Anton Dohrn, Italy	Dynamics and fate of microplastics in pelagic ecosystem: a large mesocosms integrated- ecosystem approach
12.30-12.45 PM	Maria Luiza Pedrotti	Université Pierre et Marie Curie, France	TARA Mediterranean expedition: Assessing the impact of microplastics on Mediterranean ecosystem
12.45-1.00 PM	Ludovic Hermabessiere	Agence Nationale de Sécurité Sanitaire de l'Alimentation, France	Leaching of a common antioxidant (Irgafos 168 ®) from microplastics using in vitro enzymatic model
1.00-2.30 PM	LUNCH		

Capri (NA), Italy, September 26<sup>th</sup>-29<sup>th</sup> 2017

PROGRAMME

#### 28/09/2017 - AFTERNOON SESSION

### IMPACT OF MICROPLASTICS ON HUMAN AND MARINE LIFE / DEGRADATION OF MICROPLASTICS IN MARINE ENVIRONMENT

Chairwoman: Maria Westerbos

2.30-3.00 PM	Tamara Galloway	University of Exeter, UK	Impacts of microplastics on humans and marine life
3.00-3.15 PM	Ika Paul-Pont	Institut Universitaire Européen de la Mer, France	Nanopolystyrene inhibits the allelopathic activity of Alexandrium Minutum
3.15-3.30 PM	Lucia Pittura	Polytechnic University of Marche, Italy	Long term exposure of mytilus galloprovincialis to benzo(a)pyrene-contaminated LD-PE microparticles
3.30-3.45 PM	Costanza Scopetani	University of Florence, Italy	PBDEs in Talitrus Saltator (Montagu) (crustacea, amphipoda) and the trade-off effect of microplastics
3.45-4.00 PM	Kévin Tallec	Institut Français de Recherche pour l'Exploitation de la Mer, France	Nanoplastics impacts on gamete quality of Pacific oyster, Crassostrea Gigas
4.00-4.30 PM	COFFEE BREAK		
4.30-5.00 PM	Pietro Ferraro	Institute for Applied Sciences and Intelligent Systems - CNR, Italy	New imaging tools for detection and sorting of microplastics in marine environment
5.00-5.15 PM	Dannielle Green	Anglia Ruskin University, UK	Microplastics affect the ecological functioning of an important biogenic habitat
5.15-5.30 PM	Evdokia Syranidou	Technical University of Crete, Greece	Degradation of microplastics by indigenous marine communities
5.30-5.45 PM	Kathrin Oelschlagel	Fraunhofer Institute of Ceramic Technologies and Systems, Germany	Imitating the weathering of microplastics in the marine environment
5.00-5.15 PM 5.15-5.30 PM	Dannielle Green Evdokia Syranidou	Anglia Ruskin University, UK Technical University of Crete, Greece Fraunhofer Institute of Ceramic	Microplastics affect the ecological functioning of an important biogenic habitat Degradation of microplastics by indigenous marine communities Imitating the weathering of microplastics in the

6.30 PM DEPARTURE TO RESTAURANT "IL FARO" IN ANACAPRI FOR THE SOCIAL DINNER

#### 29/09/2017 - MORNING SESSION

#### SUSTAINABILITY, SOLUTIONS AND NEXT STEPS Chairman: Cosimo Carfagna

9.30-10.00 AM	Gunter Pauli	Zero Emission Research Initiative	From nuisance to resource - the design of the next generation of polymers
10.00-10.15 AM	Francesco Degli Innocenti	Novamont, Italy	Biodegradable plastics – Marine biodegradability and potential mitigation of plastic littering and microplastic formation
10.15-10.30 AM	Ester Kentin	Leiden University, The Netherlands	Banning microbeads in cosmetic products: legal challenges
10.30-10.45 AM	Giorgio Zampetti	Legambiente, Italy	The first study about the widespread presence of plastic pellets along the Italian coast. An assessment of the threat of microplastics marine pollution from an environmental NGO's point of view
10.45-11.00 AM	Miriam Weber	Hydra Institute for marine sciences, Italy	Assessing marine biodegradability of plastic – towards an environmentally relevant international standard test scheme
11.00-11.15 PM	Christian Lott	Hydra Institute for Marine Sciences, Italy	Marine fate of biodegradable plastic – substitution potential and ecological impacts

11.15-11.45 PM COFFEE BREAK

 			PROGRAMME
11.45-12.15 PM	Gaetano Leone	UNEP - Mediterranean Action Plan	Final remarks
12.15-12.30 PM	Maurizio Avella	Institute for Polymers, Composites and Biomaterials - CNR, Italy	Conclusions - Best poster award ceremony

# **Microplastics and Marine Environment**

# Getting to the front end of microplastic pollution of the sea

### Paul CONNET

Zero Waste Movement, USA

In this presentation, Paul Connett will outline the "Ten Steps to Zero Waste", as part of an overall strategy to move towards a sustainable future:

- 1) Source Separation;
- 2) Door-to-door collection;
- 3) Composting;
- 4) Recycling;
- 5) Community reuse, repair and research centers;
- 6) "Save as You Throw" economic incentives;
- 7) Other residual reduction initiatives;
- 8) Residual Separation and organic stabilization facilities;
- 9) Zero Waste Research for Better Industrial Design, and
- 10) Interim landfills to accept currently non-recyclable materials

In each of these ten steps Connett will offer educational and practical steps that can be taken to minimize the release of microplastics into the environment, without resorting to problematic and unsustainable practices such as incineration and other resource destruction technologies.

### The air we breathe

### **Cyrill GUTSCH**

Parley for the Oceans, USA

As scientific understanding of marine plastic pollution deepens, public awareness grows. Although the threat has risen in the global consciousness, solutions remain elusive. Recognizing the complexity of the issue, Parley for the Oceans set forth a new approach: a strategy driven by creative collaboration and eco innovation, based on the fact that every second breath we take is generated by the oceans. The strategy - Parley AIR: Avoid, Intercept, Redesign - is shifting the conversation and driving change across industry, government, and in the creative communities that mold reality.

Designer and strategist Cyrill Gutsch founded Parley in 2012 to provide a collaboration network where creators, thinkers and leaders come together to raise awareness for the beauty and fragility of our oceans and collaborate to end their destruction. Parley is known for renaming sustainability into 'Eco Innovation,' a concept realized through high-caliber collaborations and the introduction of Ocean Plastic<sup>™</sup>, a range of premium materials for the sports, fashion and luxury industries made from upcycled plastic debris collected on high seas, beaches and in remote coastal communities.

Parley understands current plastic is a design failure, seeing the long-term solution for marine plastic pollution not in recycling, but in the redesign of the harmful material, processes and thinking. As a catalyst innovation, Ocean Plastic<sup>™</sup> provides an immediate replacement for new, virgin plastic that raises awareness of the issue while also providing a funding mechanism that allows for the implementation of the Parley AIR Strategy in four key areas: Communication and Education, Direct Impact, Research and Development, and Eco-Innovation.

### **Climate Change and Impacts on the Mediterranean Basin**

### Antonio NAVARRA

Euro-Mediterranean Center on Climate Change, Italy

In this contribution, the results of the investigations on the principal modes of climate variability on interannual and decadal scale through statistical models, numerical simulation and simplified models will be presented. Moreover, results of the simulation and evaluation of climate change and its impact on the Mediterranean Basin will be discussed.

# To what extent microplastic from the open ocean are weathered?

# <u>Alexandra TER HALLE<sup>1</sup></u>, Lucie LADIRAT<sup>1</sup>, Marion MARTIGNAC<sup>1</sup>, Anne Françoise MINGOTAUD<sup>1</sup>, Emile PEREZ<sup>1</sup>, Olivier BOYRON<sup>2</sup>

<sup>1</sup>Université de Toulouse; UPS/CNRS; IMRCP, 118 route de Narbonne, F-31062, Toulouse Cedex 9, France

<sup>2</sup> C2P2 - LCPP Group, UMR CNRS 5265, Université de Lyon, ESCPE Lyon, Bat 308F, 43 Bd du 11 novembre 1918, 69616 Villeurbanne, France

The importance of plastic marine debris becomes increasingly assessed and recent studies have revealed the large extent of this problem, estimating a minimum of 5.25 trillion visible plastic particles in the oceans. Another serious concern is the occurrence of micrometric and nanometric plastic debris not visible to the naked eye. At this point, thorough studies are necessary to quantify and characterize the plastic debris. Indeed, although degradation of polymers has been studied for a long time, the weathering conditions occurring in a complex environment such as oceans lead to masses of questions. Which types of polymers are present? What is the extent of their degradation? What is the degradation process? What is the influence of the biofilm and possible additives or adsorbed pollutants? In order to answer to some of these questions, in this work, plastic debris was collected in the North Atlantic sub-tropical gyre during the Expedition 7<sup>th</sup> Continent sea campaign and analyzed with a wide variety of techniques: electronic microscopy, infrared spectroscopy, size exclusion chromatography, calorimetry. For polyethylene samples, this showed a strong influence of the weathering on the crystallinity and the molar mass, proving a strong degradation. The global analysis of plastic debris including micro-sized particles (below 0.3 mm) revealed the presence of various polymers, going further than the expected polyethylene and polypropylene.

# Wintertime microplastics in different depth layers in the northern Baltic Sea

#### Maiju LEHTINIEMI, Juha FLINKMAN, Erika ZIDBECK, Outi SETALA

SYKE Marine Research Center, Helsinky, Finland

The distribution and number of microplastics in different water layers in the northern Baltic Sea were studied in the open sea areas of the northern Baltic Sea in January 2017. The sampling covered different basins: Gulf of Finland, Archipelago Sea, Bothnian Sea and Bothnian Bay. Altogether 7 stations were sampled during a monitoring expedition. Ca. 30 samples were collected from 2-3 distinct water layers separated by temperature and salinity gradients. Samples were taken with a MultiNet plankton sampler equipped with 100  $\mu$ m mesh size nets and the common manta trawl with a 330  $\mu$ m net. Samples were further processed in the laboratory and organic material digested prior to microscopy. Differences in the type and number of microplastics were observed between the samples taken with the two devices as well as between the water layers. The results are discussed from the viewpoint of the water column microplastic monitoring.

### Challenging the microplastic extraction from sandy sediments

# <u>Mikaël KEDZIERSKI</u><sup>1</sup>, Véronique LE TILLY<sup>1</sup>, Patrick BOURSEAU<sup>1,2</sup>, Hervé BELLEGOU<sup>1</sup>, Guy CESAR<sup>3</sup>, Olivier SIRE<sup>1</sup>, Stéphane BRUZAUD<sup>1</sup>

<sup>1</sup>Université Bretagne Sud, IRDL FRE CNRS 3744, 56100 Lorient, France <sup>2</sup>Université de Nantes, CNRS GEPEA UMR 6144, 44602 Saint-Nazaire, France <sup>3</sup>SERPBIO, Université Bretagne-Sud, 56321 Lorient, France

Plastics are one of the wastes the most commonly observed on beaches. This pollution needs a constant survey and a chemical identification of the smallest fractions of these plastic wastes also called microplastics. Some of the methodologies used are fast but destructive, others as Raman or IR spectroscopies require more time but preserve the samples for further analyses. These latter methods indeed involve sand-plastic separation pretreatment procedures. Methods of extracting microplastics display two main constraints: the cost and the plastic extraction rates. Based on the example of two extraction technics, the sodium iodide (Nal) density separation and the elutriation system, the present communication aims to show i) how to decrease the microplastic extraction cost? And ii) how to extract the densest microplastics? Firstly, the extraction cost is mainly due to the expensive price of Nal used in the experiments. To decrease this cost, based on a new protocol, the recyclability of NaI have been monitored during ten life cycles. The results have shown that the density of NaI solutions remains relatively stable along the cycles implying that the solution can be reused at least ten times without plastic extraction decreased and that the extraction cost is significantly decreased. Secondly, based on calculations and the study of the density of the plastic products in Europe, it can be shown that 70 to 75 % of these plastics can be extracted with the elutriation system proposed by Kedzierski and colleagues. The numerical modeling of the elutriation developed allows calculating a new column dimensioning and changes in the protocol to greatly increase the elutriation efficiency up to 98% of European plastics product. These results will help to build the next generation of elutriation system.

# Are our synthetic fabrics released into the marine environment? Evidences on microplastics pollution in waste water coming from our laundry.

### Raquel VILLALBA, Àngels ROVIRA, Laura GELABERT

LEITAT Technological Centre, Barcelona, Spain

Microplastic particles of synthetic clothes coming from laundry wastewater have been encountered in marine sediments, ecosystems and runoff and sewage waters. The *"Accumulation of Microplastic on Shorelines Worldwide: Sources and sinks"* states that *"the source of the microplastic fibres in the sewage treatment plants is most likely to be from washing machine wastewater as the mixture of fibres found in synthetic textiles is similar to the mixture of microplastic fibres found in beaches at disposal sites and in the wastewater of sewage treatment plants"*. On average, more than 1900 fibres of microplastics can be released by a synthetic garment during one wash.

MERMAIDS project promoted the mitigation of impact caused by micro and nanoplastic particles resulting from laundry wastewater on European seas' ecosystems, by demonstrating and implementing innovative technologies and additives for laundry processes and textile finishing treatments. Existing technologies and products were analyzed to determine the main factors involved in the fibres release during laundry process to further improve them. Mitigation measures were proposed for each step in the production of synthetic fibres and also from a detergency approach.

The project results are illustrated in the *Handbook for zero microplastics from textiles and laundry, Good practice guidelines for consumers* and a set of *Policy recommendations* to support policy makers in their future decisions on the review of the EU regulatory framework that could directly or indirectly address microplastics release coming from wastewater of laundry activities.

A reduction of 24% of the total microplastic release in laundry wastewater was demonstrated for standard fabrics by means of the improvements achieved. However additional research is needed to better define the many different parameters involved to propose further mitigation measures.

*Further Information* http://life-mermaids.eu/es/

22

# **Microplastics in the Mediterranean Sea**

### How concerned should we be about microplastics?

#### **Richard C. THOMPSON**

Plymouth University, UK

Microplastics are small fragments of plastic debris. The smallest fragments to be separated from the natural environment are around 20  $\mu$ m in diameter. This material has been reported on shorelines and in the water column on a global scale, its abundance appears to be increasing and there are concerns that it may present hazards to wildlife and to human health.

Plastic products bring many societal benefits and as a consequence, annual global production has increased from 5 million tonnes in the 1950s to over 250 million tonnes today. However, because of their disposable nature substantial quantities of plastic items are discarded to the natural environment and to landfill every year. This material will not readily biodegrade, however it will fragment and so the abundance of microplastic is likely to increase over the next few decades.

Laboratory experiments have shown that microplastics are ingested by filter feeders, deposit feeders and detritivores and there is concern that ingestion of this material could present a physical hazard to wildlife, for example by compromising the ability to feed. In addition, there is evidence, that small fragments of plastic could facilitate the transfer of toxic substances to wildlife. Two routes have been suggested: (1) the release of chemicals incorporated during manufacture as plasticisers, flame retardants and antimicrobials, and (2) the release of persistent organic pollutants (POPs) that have arisen in the environment from other sources and have sorbed to plastic debris in seawater.

This presentation will summarise current scientific understanding about the accumulation and potential environmental consequences of microplastic debris.

# Towards 3D modeling the plastic marine debris in the Mediterranean

# Giovanni COPPINI<sup>1</sup>, Svitlana LIUBARTSEVA<sup>1</sup>, Rita LECCI<sup>1</sup>, Sergio CRETÌ<sup>1</sup>, Giorgia VERI<sup>1</sup>, Emanuela CLEMENTI<sup>2</sup>, Nadia PINARDI<sup>1,3</sup>

<sup>1</sup> Foundation Euro-Mediterranean Centre on Climate Change (CMCC), Via Augusto Imperatore 16, 73100 Lecce, Italy

<sup>2</sup> Istituto Nazionale di Geofisica e Vulcanologia (INGV), Via Donato Creti, 12, 40100 Bologna, Italy
<sup>3</sup> Università degli Studi di Bologna, Viale Berti-Pichat 6/2, 40127 Bologna, Italy

The Copernicus Marine Environment Monitoring Service (CMEMS) provides multivariable data flow for addressing many cross-cutting marine environmental issues including plastic pollution in the Mediterranean Sea.

Recently, we have implemented a 2D Markov chain model to calculate the floating debris concentrations at the sea surface and the fluxes onto the coastlines in the Adriatic Basin over 2009–2015. The model is based on combining terrestrial and maritime plastic litter inputs with the Lagrangian model MEDSLIK-II, forced by the Adriatic Forecasting System (AFS) ocean current simulations with horizontal resolution of 1/45° and ECMWF wind analyses on 1/8°. At the lateral open boundary, AFS is nested within the parent 1/16° resolution model of the entire Mediterranean, which outputs are available through CMEMS. With a relatively short particle half-life of 43.7 days, the Adriatic Sea is defined as a highly dissipative basin where the shoreline is the main sink of floating debris. Our model results show that the coastline of the Po Delta receives a plastic flux of approximately 70 kg (km day)<sup>-1</sup>. The most polluted sea surface area (>10 g km<sup>-2</sup> floating debris) is represented by an elongated band shifted to the Italian coastline and narrowed from northwest to southeast. Impact matrices are computed for the Adriatic subregions to quantify complex source-receptor relationships in the basin.

Currently, we are working on the 3D plastic marine debris tracking in the Mediterranean Sea. A new Lagrangian transport scheme uses CMEMS ocean currents and Stokes drift components obtained directly from the WaveWatch-III model implemented in the Mediterranean. Plastic bio-fouling, loss of buoyancy and gravitational sinking are taken into consideration. The main objective of our activity is searching for "hot spots" (1) on the coastline, (2) at the sea surface, and (3) at the seabed of the basin.

### Microplastics in the Mediterranean Sea and impacts on biota

### **Giuseppe Andrea DE LUCIA**

IAMC-CNR Institute for Coastal Marine Environment – National Research Council, Torregrande (Oristano-Sardinia), Italy

Marine litter entering the oceans has been estimated to be 4.8-12.7 million tons per year. Mechanical stress, UV radiation, chemical and biological action cause the constant degradation and break down of plastic objects into smaller fragments. The Mediterranean Sea is considered one of the main polluted areas in the world since is a closed basin and therefore presents complex hydrodynamics, that need to be understood to determine the movement of plastic objects. The Marine Strategy Framework Directive (European Commission, 2008/56/EC) demands Member States to monitor descriptor 10 (Marine Litter). Therefore increasing knowledge on abundance, distribution and its impacts on the marine environment, in order to achieve or maintain the Good Environmental Status (GES) for the marine environment by 2020. Microplastics are ubiquitous and accumulate on the surface of the sea and in the sediments. The most common types of Microplastics encountered in the marine environment are spheres, pellets, fragments and fibres. Due to their small size, MP are considered bioavailable to organisms and potentially release chemicals, such as additives. Plastic litter density has been demonstrated to be correlated with human population, but different works show that this correlation may not subsist in every Mediterranean areas.

# Distribution of microplastics in Adriatic food webs

Carlo Giacomo AVIO<sup>1</sup>, Lucia PITTURA<sup>1</sup>, Stefania GORBI<sup>1</sup>, Serena ABEL<sup>1</sup>, Sonia AMORELLO<sup>1</sup>, Francesco REGOLI<sup>1,2</sup>

<sup>1</sup> Dipartimento di Scienze della Vita e dell'Ambiente (DiSVA), Università Politecnica delle Marche, Ancona, Italy

<sup>2</sup> Consorzio Interuniversitario per le Scienze del Mare, CoNISMa, ULR Ancona, Ancona, Italy

Microplastics (MPs) are widely diffused in the marine environment and their ingestion by marine organisms is being demonstrated in a growing number of laboratory and field studies. Although several organisms can ingest MPs with potentially adverse effects, a clear picture on their presence and distribution in wild organisms and trophic webs is still lacking.

In this study, the distribution of MPs was characterized in several species collected from two areas of the North and Central Adriatic Sea, respectively. According to a recently validated protocol, MPs were extracted from gastrointestinal tracts of fish and soft tissues of invertebrates. Particles were characterized in terms of size, shape and polymer typology trough microscopy and  $\mu$ FT- IR analyses. The results indicated the occurrence of MPs in approximately the 30% of 362 analysed specimens; particles were represented by fragments, lines, films and pellets while polyethylene, polystyrene and polyamide were the dominant polymers. A certain variability was observed among the different species, particularly in terms of percentage of specimens with ingested MPs. On the other hand, similar results were obtained when comparing the two Adriatic areas, both in terms of species. Invertebrates typically exhibited a lower frequency of microplastics in soft tissues in respect to gastrointestinal tract of fish, but with a higher potential of particle transfer to human consumers.

In conclusion, this study provides new insights on the presence, distribution and typology of MPs in the Adriatic food webs, representing an important baseline assessment on this kind of emerging contaminants in the Adriatic biota.

### Preliminary assessment of microplastic accumulation in wild Mediterranean fish species

# <u>Manuela PICCARDO<sup>1</sup></u>, Serena FELLINE<sup>2</sup>, Maria Giulia LIONETTO<sup>2</sup>, Roberto CARICATO<sup>2</sup>, Cosimino MALITESTA<sup>2</sup>, Antonio TERLIZZI<sup>1</sup>

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Because of their abundance, persistence and ubiguity, microplastics represent a major environmental problem, recognized even by European Union which calls for further investigation. Most studies have shown ingestion of microplastics in laboratory organisms, but only the analysis of wild specimens can represent the real problem. In this study, we carried out microplastic extraction on several specimens of Mullus barbatus, using the vertical laminar flow hood during all technical procedures, in order to limit external contamination. Our results showed high unprecedented concentrations of microplastics in gastrointestinal tract (14,6  $\pm$  4,61 items/fish), far above the European concentration. Even the frequency of occurrence (92%) was higher than that recorded in the Adriatic Sea (64%) and the Iberian peninsula (18,8%). Furthermore, for the first time, we found microplastics in wild fish gills (34,25 ± 9,89 items/fish) with a frequency of occurrence of 100%. Fragments and lines were the most abundant types of microplastics in both tissues (44% and 39% for fragments in gastrointestinal tract and gills, respectively; 44% and 55% for lines in gastrointestinal tract and gills). The dimensional distribution analysis showed the dominance of particles between 100-500  $\mu$ m (49%) in gills, while, pieces lower than 10 µm (76%) represented the most abundant class in gastrointestinal tract. The presence of a such high levels of microplastics could cause possible negative effects on wild fish health. Impacts can be multiple, but the main ones are physical and toxicological (intestinal blockage, reduction of intestinal absorption and starvation, changes in hepatic cell) which, will be investigated more thoroughly even in relation to human consumption. Finally, the use of vertical laminar flow hood during all procedures, has solved the huge problem of airborne contamination by allowing the count of fibers generally ignored.

# An overview of plastic pollution in the Mediterranean Sea: the importance of microplastics

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Because of an highly populated and closed basin, high amounts of solid waste that are generated and 30% of the maritime traffic, the Mediterranean Sea is one of the most affected sea, with more than 700 tons of plastic entering the basin every day. Distribution is influenced by the presence of large cities, shore use, hydrodynamics and maritime activities. Due to various degradation processes, microplastics comprise a very heterogeneous group, varying in size, shape, colour, chemical composition and density. Despite higher densities in some hydrodynamic structures, up to 65 million particles/km<sup>2</sup>, mean sea surface plastic range from 100000 to 400000 particles/km<sup>2</sup> giving estimated weights of 600 - 3000 tons for the whole basin. At this scale, the spatial distribution is irregular, with pattern related to the variability in the surface circulation. Beach survey revealed an abundance of pellets, reaching 1000 pellets/m<sup>2</sup> and micro plastic pollution has been recently demonstrated into sediments from the deep Mediterranean Sea.

Little is known about the extent of the damage caused by microplastics. Amongst known effects described for marine litter, mechanical effect (tissues/cells injury, alteration of digestive functions), release of chemical and the transport of species have been described as the most adverse.

To support actions to minimize impacts on the marine environment, the UNEP/MAP Regional Plan and the Marine Strategy Framework Directive (MSFD) have established frameworks within which Member States must take action to reach a Good Environmental Status (GES). Microplastics is one of the indicators for these initiatives (indicator 10DC2 of the MSFD and common indicator 23 of the UNEP/ECAP RPML). However, in order to better support these initiatives, research on protocols for large scale assessments, harmonization, definition of baselines/targets, and data management schemes have become critical.

### A temporal survey of microplastics ingested by herbivorous Eastern Mediterranean siganids

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Microplastics have been recognized in recent years, as a growing problem in marine ecosystems throughout the world. However, it is not clear how long this problem has existed. Siganids (rabbitfish) are marine herbivores that appear to consume microplastics readily, as shown in a recent study. The findings reported here are an additional study that was undertaken to explore whether abundances of microplastics in siganid guts have changed over the years.

Digestive tracts from rabbitfish collected over the past 50 years in Israeli coastal waters (stored at the National Zoological Collections at Tel Aviv University and the Hebrew University) were examined for presence of microplastics. There was a gradual increase in the presence of microplastic particles in the fish guts, from the early 1970s, through the 1980s, peaking recently with 92% of all siganids examined (2016) containing microplastics. Also, types of microplastics were found to vary along this timeline.

The temporal rise in the number of particles per fish gut and the percentage of fish with ingested microplastics suggests that marine microplastic pollution is an increasingly growing problem. Where as microplastics are often consumed by animals that confuse these with their natural food items, microplastics may also adhere to marine algae and plants, causing herbivores and omnivores to consume these at higher rates than would otherwise be expected. Therefore, presence of microplastics inside algae and marine plants are being investigated as well. Microplastics have a variety of impacts on the biota that consume them and on the food web in general and strategies to reduce their abundances must be considered. These and other findings will be described and discussed in this presentation.

# Zooplankton and plastic additives – insights into the chemical pollution of the low-trophic level of the Mediterranean marine food web

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Plastic materials such as microplastics pose a variety of problems once they reach the environment via improper waste disposal or spills, among others. While microplastics are often ingested by marine organisms, marine life is not only threatened by the physical damage plastic items can cause, but also by the possible chemical pollution resulting from the leaching of plastic additives or other adsorbed molecules on the plastics surface during long range transport. The demonstrated toxicity of some of these molecules has led to national and international legislations limiting or banning their use. However, a wide variety of substances are still found in plastic products and their impact on the marine and terrestrial environment is yet to be assessed. Marine ecosystems could be particularly at risk since they act as recipients for plastic pollution from terrestrial sources via river runoffs, wastewater treatment effluents or air-borne transportation. Representing the base of the marine food web, zooplankton plays a vital role in the ecosystem functioning, yet the actual impact of microplastic on these organisms and the occurrence and magnitude of potential resulting chemical contamination have been poorly studied so far.

In this presentation we hence investigate the possible exposure of zooplankton to plastic additives, including phthalates, organophosphate esters and bisphenols. Samples originate from three different sites of the Bay of Marseille, France, with one sampling point being situated at the effluent zone of the local wastewater treatment plant. Sample collection occurred once every two months using a neuston net (mesh size 150  $\mu$ m); here the results from four sampling campaigns will be presented. To complete this data, organic contaminants were also analyzed in seawater from the same sampling points. Plastic additives were analyzed using ASE (accelerated solvent extraction) and SPE (solid phase extraction) followed by LC and GC coupled with mass spectrometry.

### Floating microplastics in the NorthWestern Mediterranean Sea: strong temporal and spatial heterogeneities

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Mediterranean Sea is one of the most sampled areas for floating microplastics (MPs). However, only few investigations have been conducted at small spatial and temporal scales in coastal areas. To fill this gap, MPs (< 5 mm) were collected off the mouth of two contrasted rivers: the Rhône River, the largest source of freshwater and sediments into the Mediterranean Sea and a typical small Mediterranean coastal river, the Têt River. Close surface seawater transects were performed using a manta trawl (>300  $\mu$ m), at different seasons in the Rhône area and every month during one year in the Têt area. After removal of organic matter, MPs were examined under a dissecting stereo-microscope. Preliminary results show highly variable MPs concentrations even at small scale. Indeed, concentrations ranged from 0.05 to 0.6 items·m<sup>-3</sup> for the Têt area with an average of 0.2 items·m<sup>-3</sup> and from 0.1 to 0.5 items·m<sup>-3</sup> with an average of 0.3 items·m<sup>-3</sup> for the Rhône area.

Concentrations can change by a factor of 4 between two consecutive days at the same location, and by a factor of 3 between two consecutive trawls on the same day. Fibers are the most abundant shape (40-90%), followed by fragments (0-50%). Foams and films are less represented (0-20%). FTIR analysis indicates that fragments and films were mostly polyethylene (PE) and polypropylene (PP), while foams were essentially made of polystyrene (PS). Fiber analysis is on going. Occasional presence of lint composed of hundreds of fibers can partly explain the high differences observed at small scales, as well as fast changing river inputs. These extended observations of floating MPS in the NW Mediterranean coastal environment underlines the necessity of performing replicate sampling to get a better insight into the spatial and temporal distribution patterns of these worrying pollutants.

Impact of microplastics on human and marine life Degradation of microplastics in marine environment

### Impact of microplastics on the Mediterranean biodiversity

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The Mediterranean Sea has been recently described as one of the most affected areas by marine litter, including microplastics (plastic fragments smaller than 5 mm), in the world. Although effects of plastic litter on marine biodiversity have been recently investigated in several oceanic areas, poor information are still available from the Mediterranean Sea. Recent studies in the different regions of the Mediterranean basin suggested that some areas are affected by important concentration of microplastics and plastic additives (phthalates), representing a potential risk for marine endangered species (baleen whales, sea turtles, filter feeders sharks) and for the all Mediterranean biodiversity. To cover the current knowledge gaps on this issue a harmonised methodological approach for the assessment of the marine litter and microplastics impact on Mediterranean biodiversity is needed.

Here we propose marine organisms as bioindicator species, applying a new integrated monitoring tool to implement future mitigation actions at basin scale. The quantification of microplastics in the marine environment can depend on several environmental factors and change according to multiple oceanographic features, and therefore, cannot reflect the potential impact on organisms and ecosystems. The information obtained by biondicator species could better integrate the spatial and temporal presence of marine litter/microplastics in the marine environment. In addition, the use of biondicators can allow to measure not only the occurrence of marine litter in the species and its environment but also the threat posed to organisms by the evaluation of contaminants (associated/sorbed to plastic litter) accumulation and any related biological effect (biomarkers).

The identification of the most representative bioindicator species for marine litter/microplastics in the Mediterranean environment is one of the main goals of the multidisciplinary strategy developed in the Plastic Busters SDSN-MED, UfM project (http://plasticbusters.unisi.it/).

# Size selective feeding by mesopelagic fish can change seasurface microplastic abundance: impact of marine life on microplastic abundance

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The frequency distribution of sea-surface plastic particles are thought to be basically uniform across the major oceans of the world. However, in an area with locally abundant and extremely size selective predators, losses within specified size ranges could be expected. Given the difficulties in sampling the Southern Ocean we assessed the size distribution of plastic particles recovered from a west-facing beach on a sub-Antarctic island, Macquarie Island. The resulting frequency distribution showed losses from certain microplastic size ranges.

On the same island we also collected scats from fur seal species (*Arctocephalus* sp.). Identification of prey remains recovered from the scat samples showed their dominant (*ca.* 90%) food were lantern fish *Electrona subaspera*, a size selective zooplankton predator. Notably, a proportion of seal scats contained plastic particles thought to originate from the seals' prey. The size distribution of plastic particles found in the seal scats examined for this study overlap the largest losses in size classes of the beach recovered material.

Of the total amount of plastic particles circulating in the world's oceans, a significant proportion may therefore be bound up in the digestive tracts of vertebrate predators such as fish and seals. Estimates of total plastic loading from net sampling are therefore likely to be underestimated.

### Impact of microplastics on marine microbial life

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Plastic marine debris (PMD) is polluting at an alarming rate our oceans and leads to a worldwide threat to aquatic wildlife. Recent studies indicated that various bacteria rapidly colonize these surfaces (called "plastisphere"), but their impact on the global marine microbial life remain to be investigated. Indeed, bacterioplankton play key roles in marine ecosystems (regulation of major biogeochemical cycles, depollution, ...) and the recent input of large quantities of plastic may alter their diversity and activities. The large set of samples taken during the TARA-Mediterranean expedition revealed for the first time a clear niche partitioning between PMD and organic particle-attached (PA) or free-living (FL) bacteria. A marked originality of the PMD fraction was observed, with higher bacterial cell density and diversity compared to PA and FL lifestyles. In particular, higher evenness in PMD indicated a favorable environment for a very large number of bacterial species. Cyanobacteria were particularly overrepresented in PMD, together with essential functions for biofilm formation and maturation. Potential pathogenic species were identified, but further analyses are needed before waving alarmist conclusions on the potential role of PMD as vector for their dispersal. The consistent distinction of community structure between the three lifestyles, which exceeded the large-scale geographical variation in the Mediterranean basin, support a new trade-off for the maintenance of a large diversity of bacterioplankton in the Oceans. 'Plastic specific bacteria' recovered only on the PMD represented half of the OTUs, thus forming a distinct biota that should be further considered for understanding microbial biodiversity in changing marine ecosystems.

### Microplastics occurrence in the gastrointestinal tract of key marine invertebrates at different trophic levels from the North Adriatic coast

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Plastic debris represent a large ubiquitous component of marine litter in the marine environment recognised internationally as a matter of increasing concern. Microplastics (plastic particles < 5 mm) are increasingly documented in marine organisms, including species involved in human consumption such as molluscs, crustaceans and fishes. Physiochemical properties (size, density, shape, chemical composition) of MPs might influence their capability to be ingested by organisms. Once ingested the fate of MPs appears various depending on their properties and might be very specie-specific. This work aimed to provide evidence on the spatial distribution of MPs along different trophic levels in coastal lagoon saltmarshes within the Po Delta, discriminating various typologies of MPs. Representative marine species characterized by different trophic position and feeding strategy (herbivores, detritus- and deposit-feeders, filter feeders, carnivores) were sampled during June-July 2016 in 3 coastal lagoons (20-40 km apart) potentially characterized by the presence of MPs derived from the effluents of a huge amount and typology of human pressures. Within each lagoon 3 sites have been selected (about 100-200 m apart) where 10 individuals for each target species were collected in order to explore spatial scales of variations in the distribution of microplastics. Microplastics were extracted from the gastrointestinal tract of sampled specimens, microscopically described (number, size, color, shape), and chemically characterized in terms of polymer typology using the ITR-FTIR technique. Obtained results are expected to provide knowledge of which are the trophic levels where MPs are present, whether specific typologies of polymers tend to accumulate more at certain trophic levels than others, and whether microplastics accumulation is consistent at different spatial scales

## Microplastics as ephemeral and possibly preferred substrates for planktonic cnidarian planulae

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Plastic pollution is one of our many concerns with respect to the health of our seas. Another concern that affects many of our interests in the coastal zone is that of jellyfish (and especially nuisance jellyfish) blooms. These blooms may impact fisheries, the integrity and functioning of marine ecosystems, coastal industries, shipping, tourism and coastal recreation, desalination plants, and more. There is a sense, among some scientists and the public that the frequency and intensity of jellyfish blooms is increasing in recent years, though this trend is debateable.

Many cnidaria have complex life cycles that include both free-swimming and attached (not necessarily benthic) stages. Experimental work has shown that certain cnidarian planulae prefer artificial over natural substrates and in an increasingly plasticized ocean, the chances of planulae settling on plastic and microplastic particles is also on the rise. Scientists have therefore wondered if there is a relationship between the increasing abundances of plastic substrates in the sea and the frequency of jellyfish blooms. Microplastic particles sampled by means of Manta net tows in the eastern Mediterranean Sea were examined to explore this question and results from these will be presented.

### Impact of microplastic on the deep sea

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The deep ocean (>200 m depth) encompasses 95% of the global ocean volume, and is both the largest and least explored biome on Earth. Global change and several anthropogenic impacts, including bottom fisheries, increasing exploitation of hydrocarbons and seabed minerals, are threatening the deep-sea ecosystems. Recent studies have also revealed the presence of plastic debris in the deep sea floor and within organisms, but the extent to which (micro)plastics can affect this remote ecosystem is still very limited. Some physical and biological processes (e.g., oceanographic processes, marine snow, microbial biofilm, fouling) have been suggested to promote the transport/sinking of plastics to the seafloor, and increasing relevance of this contamination in the deep sea could be expected due to the continuous plastic input into the sea from several sources. In addition, deep-sea ecosystems are characterized by more stable chemical-physical conditions than the sunlight zone, such as lack of light radiation and very low temperatures. These characteristics are expected to favour the persistence of (micro)plastics over time, thus determining important consequences for deep-sea trophic webs and ecosystem functioning. In the present talk, the potential threats and implications of (micro)plastic debris on deep-sea life will be presented in order to identify the possible future strategies to investigate and mitigate the negative effects of (micro)plastics on the largest biome of our biosphere.

## Microplastics in juvenile commercial fish from a natural estuarine environment

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Microplastics are considered a widespread environmental contaminant in the marine environment. Microplastic ingestion has been reported by a range of marine species but less known is the level of microplastic contamination by these species when they live in transitional ecosystems such as estuaries. The aim of this study was to assess the ingestion of microplastics by three important commercial fish species: the sea bass (Dicentrarchus labrax), the common two-banded seabream (Diplodus vulgaris) and the European flounder (Platichthys flesus) from a transitional coastal habitat (Mondego estuary, Portugal). Anthropogenic debris was extracted from the gastrointestinal tract of fish using two approaches: the visual inspection and a 10% KOH solution and were characterized under a stereomicroscope. The amount, type, colour, and polymer type of ingested microplastics from 150 fish collected in two areas (downstream, close to the mouth of the estuary, and upstream close to the river), were quantified. A total of 157 microplastics were extracted from the gastrointestinal tract of all fish, with an average of  $1.67 \pm 0.27$  (SD) particles per fish. Plastic particles were identified as fibers (96%) and fragments (4%), of variable size and color. A sub-sample of the retrieved particles was analysed under a FTIR spectrometer revealing that the most dominant plastic polymer types found were Polyethylene (PE), Polypropylene (PP), Polyester (P) Nylon and Rayon. No significant spatial difference was found between the occurrences of microplastics in fish along the estuary, but the higher number of fibers (80%) was found in the GI tract of the commom two-banded seabream (Diplodus vulgaris), a very voracious and fast species. We report some of the first findings on presence of this emerging pollutant in juvenile commercial important fish raising concerns regarding the potential negative effects on fish health condition.

## Dynamics and fate of microplastics in pelagic ecosystem: a large mesocosms integrated-ecosystem approach

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Microplastics may affect marine biota in several ways: physically, by blocking or disturbing vital organs (e.g., feeding apparatus); chemically, by releasing toxic pollutants; or acting as hard substrates for the small plankton communities. However, little is known on the pelagic ecosystem response to microplastics pollution. Here, we present the first results of an integrated pelagic ecosystem study (INPUT) aiming to investigate the effects of microplastics on the trophic web and coastal ecosystem functioning during a phytoplankton bloom. The experiment consisted in the deployment of six mesocosms ( $\approx$  53 m<sup>3</sup> each, 3 m diameter at surface and 15 m depth; June 2016, Gulf of Naples), all enriched with macronutrients (phosphate and silicate) inducing a microalgal burst, while three were also amended by microplastics (five different types and densities, diameter ranged between 20 and 1000  $\mu$ m). The experiment lasted 15 days, with a daily sampling program at three depths (0.5, 4.5 and 9.5 m) carrying out a multidisciplinary and end-to-end study (from viruses to mesozooplankton). Taxonomic and functional diversity (virus, bacteria, phytoplankton, microzooplankton and mesozooplankton) was explored through conventional taxonomic measurements together with metabarcoding and metatranscriptomics approaches. The fluxes in the trophic web were assessed through measurements of photosynthesis, bacterial and viral production, egg production and grazing by mesozooplankton. Particulate and dissolved matter in

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the water (polysaccharides, CDOM, metals, nutrients, carbon, microplastics) was measured daily all along the duration of the experiment. Preliminary results show that mainly two kinds of microplastics shape the prokaryotic community dynamics and its enzymatic activities, and thus being responsible for nitrogen recycling modifications. Moreover, females of some copepods species ingest microplastics. In synthesis, the first set of data from the INPUT experiment shows that microplastics pollution modulates the ecology of the pelagic system.

## TARA Mediterranean expedition: Assessing the impact of microplastics on Mediterranean ecosystem

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The accumulation of plastic debris on the surface of the oceans is widely recognized as a newly emerging problem for worldwide marine systems. To better understand the impact of plastic on the Mediterranean ecosystem, TARA-Méditerranée expedition crossed the entire Mediterranean Sea in 2014 to study the distribution of plastic fragments and zooplankton. Surface samples were collected with a 330  $\mu$ m Manta net and plastics were sorted from 124 samples and digitally imaged with the Zooscan system. Particle characteristics as size, surface area and circularity were determined and the distribution and concentration of floating microplastics were compared to the abundance of the neustonic zooplankton.

The first global biogeography map of the 98 dominant zooplankton groups and 6 categories of plastic particles in the basin were established. Results showed that plastic fragments were present in all Mantatows. The plastic samples analyzed contained mainly fragments but different typology described as foam, filaments, polystyrene and row pellets and fibers were also present. The average surface debris concentration over the entire survey was  $2x10^5$  particles/km<sup>2</sup> with values varying from  $2x10^3$  particles/km<sup>2</sup> in the Eastern basin, to more than  $2x10^6$  particles/km<sup>2</sup> in the Western basin. Coastal zones of Naples, Corsica and Marseille were clearly identified as areas of particularly high plastic concentration. The high ratio of plastic abundance to zooplankton abundance may suggest a potential impact of microdebris on various taxa and their incorporation into the neustonic food web, with consequences on the pelagic biota in the most polluted areas.

The next step is to link the basin wide distribution of plastic fragments to surface circulation models in order to identify hotspots of plastic accumulation and predict the pathways and fate of plastic debris in the Mediterranean Sea. Ecological implications of plastic fragments in the Mediterranean is the subject of work in progress conducted by the 20 partner institutions of TARA-Med Consortium.

## Leaching of a common antioxidant (Irgafos 168 <sup>®</sup>) from microplastics using *in vitro* enzymatic model

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Plastics debris, including microplastics, are nowadays recovered in every marine compartments. Many studies have been conducted on the leaching of adsorbed pollutants on microplastics but few have focused on plastic additives. If leaching of some plastic additives has been recently demonstrated in water, no work has examined the leaching of plastic additives when ingested by marine organisms and exposed to digestion, which could have potential adverse effects on the host. In this work, the development of an *in vitro* enzymatic model to study the leaching of a common plastic additive, an antioxidant, found in polyethylene (PE) has been performed. Home-made PE microplastics (20-100 µm) loaded with Irgafos 168 <sup>®</sup> (PE-Ir) were used in the present study. The *in vitro* enzymatic model mimics the main enzymes in the stomach and intestine of vertebrates: pepsin and trypsin respectively. Moreover, an intestinal enzymatic cocktail including amylase, lipase and trypsin represented by pancreatin was also used. PE-Ir particles were exposed to the three digestive conditions for 8h at two different temperatures (20 and 37°C) representing fish and human models, respectively. The leaching of Irgafos 168 <sup>®</sup> and enzyme specific activities were recorded over time using GC-MS/MS and enzymatic assays, respectively. Leaching of Irgafos 168 <sup>®</sup> will be compared with blank made of buffer without enzyme and is expected to be modulated by the presence of digestive enzymes.

This work will be used to evaluate the transfer of a common plastic additive to marine organisms and to the upper food chain through risk assessment evaluation.

### Impacts of microplastics on humans and marine life

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There is no doubt that plastics provide many societal benefits, offering inexpensive, lightweight and durable items for consumer products, food packaging and construction. Multi- million tonnes production rates, coupled with indiscriminate disposal have allowed plastic wastes to infiltrate ecosystems on a global scale, posing uncertain risks to ecosystems and to human health. Of particular concern is the reported presence of microscopic plastic debris (< 1mm in diameter and with no lower size limit = micro- and nano-plastic) in aquatic terrestrial and marine habitats. Microplastics present a risk to health because their small size overlaps with prey items ingested by many marine organisms, with the potential for entry into the marine food web. Once ingested, microplastics may cause harm through particle toxicity or by releasing persistent, bioaccumulating or toxic contaminants absorbed to their surface from surrounding waters, or leached from within the plastic polymer itself. In addition, microplastics may act as vectors for the uptake of pathogenic organisms.

In this presentation, the routes of exposure and risks presented by plastics polymers found in marine debris will be discussed. The contribution of microplastics as a vector for human contamination compared with other routes of exposure to contaminants will be considered. Finally, the potential for green chemistry approaches to formulate safer, more sustainable polymers will be considered as an avenue for reducing the ecological and public health impacts of plastic waste.

### Nanopolystyrene inhibits the allelopathic activity of *Alexandrium minutum*

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Formation of nanoplastics (NP,  $\leq$  100nm) upon solar light degradation of marine plastic debris was recently demonstrated, highlighting the importance to evaluate their environmental concentrations, and their impacts on organisms and communities. Indeed, interactions between organisms are influenced by a wide range of environmental factors including pollutant. This work aims to understand the potential effects of NP on the allelopathic interactions between *Alexandrium minutum* and a diatom *Chaetoceros* sp.

*A. minutum* is a dinoflagellate reported in worldwide oceans and responsible for production of toxins including unknown extra-cellular compounds exhibiting allelopathic activities. Allelopathy is defined by the interaction between photosynthetic organisms through production of chemical compounds. Allelopathic impacts on phytoplankton species are widely reported, with the diatom *Chaetoceros* sp. being particularly sensitive. To investigate the interactions between NP and *A. minutum* allelochemicals on *Chaetoceros* sp., diatom cultures (control) were exposed to *A. minutum* filtrate (A) or nanopolystyrene beads (NPS; 5.7 µg mL<sup>-1</sup>) alone, and to *A. minutum* filtrate preliminarily incubated with NPS (ANPS). Toxicity was assessed through the measurement of maximum quantum yield (QY) used as a proxy of photosynthesis activity using a Pulse Amplitude Modulated fluorimeter.

Results confirmed toxicity of *A. minutum* allelochemicals on *Chaetoceros* as suggested by the decrease in *Chaetoceros* QY by 20, 90, 96 and 100% after 1, 2, 3 and 24h of exposure to *A. minutum* filtrate, respectively. Conversely, in control, NP and ANPS conditions the QY remained stable during the first hours and increased at 24h, probably due to diatom growth. These results suggest that NPS

#### IMPACT OF MICROPLASTICS ON HUMAN AND MARINE LIFE. DEGRADATION OF MICROPLASTICS IN MARINE ENVIRONMENT

seemed to have mitigated *A. minutum* allelopathic activity. We hypothesize that adsorption of allelochemicals on NPS could have modified their toxicity or availability for diatoms. Further experiments are ongoing to clarify the interactions between NPS, *A. minutum* ECC and diatoms, and the role of particle charge, nature and size.

## Long term exposure of *Mytilus galloprovincialis* to benzo(a)pyrene contaminated LD-PE microparticles

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The ubiquitous presence of microplastics (MPs) in the marine environment has been demonstrated, raising concern for their interactions with marine organisms. These particles efficiently adsorb persistent organic pollutants from the surrounding environment and, due to the small size, they are easily available for ingestion at all trophic levels. Once ingested, MPs can induce mechanical damage, sub- lethal effects and biological responses, further modulated by possible release of adsorbed chemicals or additives.

In this study, ecotoxicological effects of MPs and their interactions with PAHs were investigated in Mediterranean mussels, *Mytilus galloprovincialis*. Organisms were exposed for 28 days to 10 mg/L of low-density polyethylene (LD-PE) microparticles (size range 20-25  $\mu$ m), both virgin and pre-contaminated with benzo(a)pyrene. Ingestion of microplastics and their tissue translocation were evaluated through a quantitative extraction protocol and histological analyses. Bioaccumulation of B(a)P was determined and integrated with the analysis of a wide range of cellular biomarkers at both biochemical and molecular levels, including immunological and antioxidant systems, neurotoxic responses, genotoxic damages and lipid peroxidation. Obtained results showed a different susceptibility of analysed pathways, depending on time of exposure, tissue and treatment typology, offering new insights on mechanisms of microplastics toxicity for bivalves.

## PBDEs in *Talitrus saltator* (Montagu) (Crustacea, Amphipoda) and the trade-off effect of microplastics

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The supralittoral amphipod *Talitrus saltator*, is an established POPs (Persistent Organic Pollutants) biomonitor in coastal environments and it is able to swallow microplastics (MPs) in natural condition.

Since plastic pollution has become an environmental concern, *T. saltator* was selected to study the role of MPs in the transfer of organic pollutants and to investigate if ingested MPs, could either transfer contaminants to biota or clean it adsorbing pollutants taken from the diet.

To this purpose, two laboratory experiments were performed.

In the first lab-experiment, *Sandhoppers* were fed with fish food mixed with polyethylene MPs contaminated with a labelled brominated diphenyl ether (<sup>13</sup>C-labelled BDE-47). The results showed that *T. saltator* is not only able to swallow MPs, but also to bio-accumulate BDE-47C13 from MPs.

In a second lab-experiment, *T. saltator* was fed with food spiked with <sup>13</sup>C-labelled BDE-47 and successively with food mixed with clean MPs to evaluate if MPs are able to sorb the pollutant ingested by *T. saltator*. The data obtained through GC-NCI-MS (Gas chromatography–mass spectrometry operating in negative ionization mode) and X Ray Micro-CT (Micro-Computed Tomography) analysis indicated that *T. saltator* accumulates <sup>13</sup>C-labelled BDE-47 from food and that MPs, once ingested, may be able to act like "scavengers" for <sup>13</sup>C-labelled BDE-47, adsorbing pollutants from *T. saltator*.

Based on the preliminary results, it appears that MPs could play a potential double role, both as source and sink of contaminants.

## Nanoplastics impact on gamete quality of pacific oyster, *Crassostrea gigas*

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Ocean plastic pollution is a major environmental concern due to the everincreasing plastic production worldwide. Plastic debris breakdown in the form of microplastics (<5 mm) in oceans and recent studies also demonstrated the formation of nanoplastics (defined as particles smaller than 100 nm) upon photodegradation and mechanical degradation. While little information is available in regards to their environmental concentrations and toxicity, the few experimental published studies demonstrated hazardous effects on marine organisms. Investigating nanoplastics environmental levels and toxicity is of major importance as they have different properties than microplastics, with higher reactivity and ability to pass across biological barriers due to their nano-size. Here, we investigated the impacts of nano-sized polystyrene particles (100nm) coated with carboxylic (PS-COOH) and amine groups (PS-NH<sub>2</sub>) on spermatozoa and oocyte of pacific oyster, Crassostrea gigas, one of the most important aquaculture species worldwide. A dose-response experiment using several concentrations (0.1, 1, 10 and 100  $\mu$ g/mL) was performed, and toxicity was investigated on several end-points (cells viability and reactive oxygen species production, sperm motility and oocytes shape) using flow cytometry and microscopy.

PS-COOH demonstrated greater toxicity than PS-NH<sub>2</sub> with a significant decrease in spermatozoa and oocytes viability at the highest concentration. A significant dose-dependent oxidative stress was also observed in spermatozoa that demonstrated altered motility. For oocyte, no signs of oxidative stress were observed while a strong decrease of oocyte viability (-30%) was observed only at the highest PS-COOH concentration (100  $\mu$ g/mL). Our results demonstrated an effect of nanoplastics on some characteristics of oyster's gametes which could have consequences on the overall oyster development, especially fertilization success and embryo-larval development. Thus, our next goals will explore effects of nanoplastics smaller than 100 nm on gamete quality for embryo development to improve knowledge of nanoplastics' risk for oyster.

## New imaging tools for detection and sorting of microplastics in marine environment

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Digital holographic microscopy (DHM) has proved to be a powerful imaging tool for identifying, analysing and reconstructing the 3D shape of small organisms and objects in their natural environment. In fact, DHM has the advantage, compared to other imaging techniques, to be a non-intrusive, non-destructive and label-free method for in situ measurements. This makes holography the most suitable tool for underwater imaging, where many of the species under investigation are very fragile and can be damaged. In particular, we built up an optofluidic platform based on DHM able to perform such analysis in microfluidic environment, i.e. in dynamic conditions and also in case of a turbid medium.

In this work, we take advantage of this technique to identify, sort and reconstruct the morphology of different classes of microplastics dispersed in water (e.g. PVC, PET, PP, ecc.) and to provide an effective assessment of their abundance. By adopting special algorithms for numerical processing of the acquired images, we try to separate the plastics from other materials, such as organic debris (shell fragments, animals parts, diatoms, etc.) and other items (metal paint coatings, tar, glass, etc.).

### Microplastics affect the ecological functioning of an important biogenic habitat

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Biological effects of microplastics on the health of bivalves have been demonstrated elsewhere, but ecological impacts on the biodiversity and ecosystem functioning of bivalve-dominated habitats are unknown. Thus, we exposed intact sediment cores containing European flat oysters (Ostrea edulis) or blue mussels (Mytilus edulis) in seawater to two different densities (2.5 or 25 µg L<sup>-1</sup>) of biodegradable or conventional microplastics in outdoor mesocosms. We hypothesised that filtration rates of the bivalves, inorganic nitrogen cycling, primary productivity of sediment dwelling microphytobenthos, and the structure of invertebrate benthic assemblages would be influenced by microplastics. After 50 days, filtration by *M. edulis* was significantly less when exposed to 25 µg L<sup>-1</sup> of either type of microplastics, but there were no effects on ecosystem functioning or the associated invertebrate assemblages. Contrastingly, filtration by O. edulis significantly increased when exposed to 2.5 or 25 µg L-1 of microplastics, and porewater ammonium and biomass of benthic cyanobacteria decreased. Additionally the associated infaunal invertebrate assemblages differed, with significantly less polychaetes and more oligochaetes in treatments exposed to microplastics. These findings highlight the potential of conventional or biodegradable microplastics to impact the functioning and structure of sedimentary habitats and show that such effects may depend on the dominant bivalve present.

### Degradation of microplastics by indigenous marine communities

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Plastic debris represents a significant problem among the various problems facing the marine environment. In this work, we explore the ability of two marine indigenous communities to degrade secondary microplastics. Polyethylene (low density as well as high density polyethylene) films were exposed to UV irradiation until they were fragmented to microplastics under mild mechanical stress. Next, 50mg of sterile microplastics with size 2 mm-250  $\mu$ m were added in sterile flasks and were incubated separately with these two pelagic microbiomes. At the end of every month, the aqueous medium containing the microplastics and the associated microbiome were filtered and the filtrate was stored in -20°C in order to perform various analysis such as measurement of proteins (EPS), carbohydrates (EPS), viscosity and viable cell concentration. ARISA PCR was performed in order to monitor the community succession of free cells as well as of the adhered assemblages. The weight loss of the collected microplastics was measured and the degradation efficiency of each community was estimated.

A significant decrease in the weight of the polyethylene microplastics was observed along the experimental period, implying the potential ability of indigenous communities to *in situ* degrade secondary microplastics.

## Imitating the weathering of microplastics in the marine environment

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The existence of microplastic particles (polymer particles < 5mm) in our environment has already been proven by several studies. Scientists know that the interactions of meteorological (sun radiation [UV] and air temperature) and marine effects (water temperature, salinity and turbulences) lead to changes in physico-chemical properties of plastic products or briefly to their degradation and fragmentation. By understanding the individual steps of the weathering process and the respective consequences for material properties (size, shape, density, crystallinity and surface properties), it is possible to predict the distribution, behavior and interactions of the particles in the environment and finally to assess their risk towards nature.

The aim of this work is to mimic the weathering of microplastics in the marine environment under laboratory conditions and to evaluate its influence on material properties. At first the study is focusing on abiotic weathering processes including UV radiation and mechanical stress. For weathering processes data concerning the initial state of microplastics, the energy source and input during weathering and the state of microplastics after weathering will be presented. Powders of low-density polyethylene (LDPE) and polyethylene terephthalate (PET) are used as raw materials for the tests. The powders are dispersed in artificial sea water and then subjected to weathering. Material properties including particle size, shape, concentration, crystallinity and density are measured. After 28 days of UV exposure no changes in particle size or shape for both materials can be detected by dynamic imaging (5-1700  $\mu$ m). The appearance of submicron-scaled particles is determined by nanoparticle tracking analysis NTA (several nanometers – 500 nm). For LDPE a decrease in particle size and an increase in particle concentration is measured with NTA indicating the formation of nanoparticles. The crystallinity determined by differential scanning calorimetry and the density measured by helium pycnometry of LDPE and PET is increasing during UV exposure.

### Sustainability, solutions and next steps

### From nuisance to resource - the design of the next generation of polymers

#### **Gunter PAULI**

#### Zero Emission Research Initiative Network

The world of plastics has transformed our lives. Waves of innovations have permeated our societies and took a place in every niche of production and consumption. This era of chemical engineering brought wealth and comfort, thriving on science and innovations. However, the scholars behind this breakthrough focused more on functionality and performance than on the behavior of these polymers after their useful life. First of all, many polymers demonstrate a disconnect between the "half-life" and "performance". How can one explain that a one way plastic water bottle made from PET only needs to provide a useful packaging service for weeks, at most months, where as the half life can be calculated in decennia sometimes even in centuries?

While this disconnect in design has been widely debated, the industry still remained ignorant about degradation of plastics its framework of molecular structuring and the blending of cocktails until the plastic islands emerged in the oceans. Most focus of degradation studies centered around biodegradation: the aerobic decomposition of polymers in soil. The presence of micro-organisms in this layer of earth facilitates the break-down of plastics. However, these micro-organisms are not present when ultraviolet rays regulate the environment, and as a result, plastics designed for biodegradation all too often do not photo-degrade and accumulate.

The situation gets worse when plastics that over (a long) time degrade in the soil, and perhaps in light, ultimately end up in water as is estimated to be the case for 7% of all polymers produced. The soup of micro-organisms in water is totally different, and the worst imaginable shift is found in the sea which could be described as a soup of bacteria. Somehow we forgot that salt is a preservation agent, thus the bacteria that thrive in this ecosystem have life conditions that are totally different than in soil. Therefore, plastics that end up in the sea only wither down, but hardly degrade unless these have been designed to degrade in soil, light, fresh water and the especially in the sea.

The understanding of this quadruple level of complexity opens up a new world for chemical engineers who now must understand the ecosystems that will receive polymers, realizing that UV-blocking agents may turn objects stable in light and in

water contributing to the expansion of the plastic islands made of micro-beads. This drama of today must lead to a better understanding of the real world and therefore it is an exceptional opportunity to redesign the whole system of polymers from the monomers up to the matrix of polymers and the full cycle in the life of a molecule. If the chemical industry were to embrace this reality, it can rethink its critical role in society while making a major leap forward towards sustainability.

### Biodegradable plastics – marine biodegradability and potential mitigation of plastic littering and microplastic formation

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The problem of plastic marine debris is not new; careless waste management requires a serious investment in awareness, prevention, and recovery programs at global scales. The bioplastics industry does not consider biodegradability as a license for littering in the environment for several reasons that follow. Packaging and consumer products must have the potential to be recovered in some way at their end of use. In certain contexts, biodegradability allows recovery through organic recycling. This option is contemplated by the European Directive on Packaging and it is beneficial whenever packaging is mixed with kitchen waste (biowaste). To avoid misleading communications, it is critical that the term biodegradable only be associated with the relevant degradation environment (where) and its associated conditions (how much and how long). In agriculture, tests specific to soil define mulch film biodegradation because this depositional environment is microbiologically different from composting. Similarly, tests specific to the marine environment are now under development at ASTM and ISO level. Some biodegradable plastics showed high level of biodegradation (as CO<sub>2</sub> evolution) in less than 1 year using these test methods. How should we interpret these very promising biodegradation data? Generally speaking, the environmental risk depends on the concentration of the environmental stressor and on its residence time in the environment. The lower the concentration and the shorter the residence time, the better. Bioplastics do not immediately disappear upon exposure to the sea. However, biodegradability is a factor that reduces the risk by reducing the stressor's residence time. Therefore, on one hand the idea of solving the problem of plastics in the ocean just by shifting to bioplastics is unfounded (bioplastics do not disappear by magic). On the other hand, for those applications where accidental release is certain or very probable, biodegradability can become a means of decreasing the environmental risk. Materials that show full and relatively fast biodegradation may be suitable for plastic products known to wear down or become stranded (for example, fishing gear) and scatter into the sea. Bioplastics hold promise for aquaculture professional applications (e.g. nets for mussels farming) where the disposal of plastic waste is an inevitable outcome. Bioplastics can be the right solution for specific products, if properly applied.

### Banning microbeads in cosmetic products: legal challenges

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Although a small part of the microplastics in the Mediterranean Sea originate from cosmetic and personal care products, these microplastics can only be prevented from entering the environment if they are no longer added to these products. There are several legal routes to ban microplastics in cosmetic products. First of all, national legislation can be adopted. The Microbead-Free Waters Act of the United States was the first national ban in 2015. France followed while Taiwan, South Korea and New Zealand also have proposed bans. Second, regional regulations, such as within the European Union, in particular the Cosmetics Regulation and REACH Regulation, could also suit to ban microbeads in cosmetic and personal care products. Third, voluntary agreements with and between cosmetic manufactures could be concluded to phase out microbeads. And fourth, labelling initiatives, by the cosmetics manufactures, NGOs and government, can help consumers to identify products without microbeads.

All routes face serious legal challenges. First, the present national bans only restrict the use of microbeads in rinse-off cosmetic products, while leave-on products pose an equally threat to the environment. Second, a limited definition of microplastics is used in national bans as well as in labelling initiatives. Third, national bans but also EU regulation may limit the possibility to adopt more stringent regulation. The US Microbead-Free Waters Act prohibits federal states to ban microbeads in other cosmetic products, such as leave-on, and also other phases of microplastics. Fourth, banning microplastics as substances in products in the EU requires amendment of the REACH Regulation to first of all include polymers in the regulation at all. Fifth, voluntary agreements are restricted by EU and national competition rules, and can only be concluded if they comply with strict conditions. Sixth, the WTO TBT Agreement is applicable to national and regional bans of microbeads, as a ban is qualified as a 'technical barrier to trade'. Justification has to be sought in the protection of the environment and human health. International standards may be used as guidelines, so setting the 'right' standards is crucial.

### The first study about the widespread presence of plastic pellets along the Italian coast. An assessment of the threat of microplastics marine pollution from an environmental NGO's point of view

#### **Giorgio ZAMPETTI**

#### Legambiente Onlus

62

Microplastics marine pollution is a global threat and Italy is not immune to it. The potential solutions need to take into consideration the changing global attitude towards plastics; moreover, legislative and preventative measures must also be considered in order to reduce the dangerous impact of microplastics pollution on the entire ecosystem. We expect governments take the lead in applying these measures to private and public businesses, especially those who are dealing with plastic production and transformation. The European plastic industry produces around 58 million tonnes of raw plastic material a year. Pathways of loss to the environment of this material are poorly understood but pre-production microplastic pellet loss has been identified as one of the largest source of microplastic pollution affect EU water. Plastic pellets (or nurdles), when handled irresponsibly, reach drains, rivers and the sea, with several consequences. Legambiente, as part of the European Coalition to End Plastic Pellet Loss, is working for defining the problem and solutions in Italy. On the occasion of this international conference, Legambiente will present for the first time a preliminary study about the problematic presence of nurdles all over Italian coastal locations; a research conducted with the help of citizens and volunteers' reports. Understanding problems, on a local and global scale, is the first step to raising awareness among stakeholders and policy makers, which can then contribute to solutions and citizens taking direct action, just as Legambiente has done over the last several years. This issue truly requires full-scale engagement, it's necessary to develop new thinking about the way we prevent, design, produce, consume and dispose of plastics. To do this, a strong network between countries needs to drive and foster awareness campaigns and joint policies, as well as pave the way for the decisive transition towards a circular global economy.

### Assessing marine biodegradability of plastics - towards an environmentally relevant international standard test scheme

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In the process of becoming independent from fossil hydrocarbon resources biobased plastic is one option, being also favoured by programmes of the European Commission. To achieve maximum sustainability bio-based polymers that are also biodegradable are entering the market, with nationally differing regulation. It is wise to assess the risk of these new materials especially in fields where their use is intrinsically linked to a loss to the environment (e.g. by conversion to micro-plastics through wear), or where unintentional littering is probable. Thus, standard tests (e.g. ISO) are needed to validate the claim of a material being "biodegradable" for consumer safety and environmental impact. Here we give an overview of current marine standard tests and present results from the EU-funded project Open-Bio on the development of tests of biodegradability under marine conditions. We present feasible field test systems for three coastal scenarios: plastic being buried in intertidal beach sand (eulittoral test), plastic floating in the shallow water column (pelagic test) and plastic sunken to the sandy sublittoral (benthic test). The field tests were optimized to a state that the test material could be observed in situ over a time of up to 2 years without loss of larger fragments, and marine disintegration could be followed for common biodegradable polymers. Degradation rates for the tested polymers are given. A set of mesocosm experiments simulating the same three habitats supported the field research in a semi-controlled setting of environmental conditions, and allowed to critically evaluate field and lab test results, leading to an environmentally relevant test scheme. Our outlook shows the next steps of test development needed to provide a comprehensive toolset to cover the majority of conditions in which plastic is found in the marine realm.

### Marine fate of biodegradable plastic - substitution potential and ecological impacts

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Biodegradable plastic is gaining attention, also through market regulation by a growing number of countries. The substitution of conventional plastic by these new materials is discussed as one mitigation measure to the ever-growing global problem of marine plastic litter. Based on available market data of plastic production, littering risk estimations and meta-analyses of the marine litter in the global oceans we present mathematical and conceptual models to assess the substitution potential of biodegradable plastics for conventional polymers, and evaluate the possible role as a mitigation strategy against marine plastic litter accumulation. We estimate substitution potentials from 0.3 to 80 % for different scenarios, which highlights the importance to better understand the ecological impact of biodegradable materials when entered the marine environment. Currently, our knowledge on the fate of conventional plastic and these new materials in the ocean is based on laboratory tests, analysis of materials collected in the field, observational field data, and only a few in-situ studies so far. Functional aspects as the interaction of biofilm and fouling communities with the polymer surface and the metabolism of these biota are largely unknown. Since five years we have been investigating the formation of the microhabitat of marine plastic in consecutive field experiments addressing the shortterm (weeks) biofilm formation and the mid-term (up to one year) fouling succession in two environmental coastal settings, a sublittoral sandy seafloor and the water column in the warm-temperate climate of the Mediterranean Sea. We also report the preliminary results of comparative *in-situ* experiments currently in progress in the tropical ocean of NE Sulawesi, Indonesia. Data of biofilm and fouling development and composition, metabolic rates and degradation rates of common biodegradable polymers and blends serve as a baseline for further studies towards a comprehensive understanding of the ecological impact of plastic in the marine environment.

### POSTER SESSION

POSTER SESSION	
P1	Smart solutions
	Claudia ALTAVILLA, Andrea SORRENTINO, Lorena AFFATATO,
	Salvatore IANNACE, Maria GELLI
P2	Microplastics from waste-water treatment plants.
	Preliminary data
	Ricardo GOUVEIA, Joana ANTUNES, Paula SOBRAL, Leonor AMARAL
P3	Polystyrene debris determination in seawater by
	fluorescence measurements
	Gianfranco CAROTENUTO, Sergio DE NICOLA
P4	Preliminary data on the impact of microplastics on blue-
	red shrimp (Aristeus antennatus) in the Mediterranean
	Sea
	Ester CARRERAS-COLOM, Joan E. CARTES, Francesc PADRÓS, Anna
	SOLER, María CONSTENLA, Maite CARRASSÓN
P5	Isolation and chemical characterization of microplastics
	and their degradation by-products in coastal sediments
	Alessio CECCARINI, Andrea CORTI, Francesca ERBA, Sabrina
DC	BIANCHI, Jacopo LA NASA, Francesca MODUGNO, <u>Valter CASTELVETRO</u>
P6	Degradation of biodegradable plastic buried in sand
	Mariacristina COCCA, Francesca DE FALCO, Gennaro GENTILE,
	Roberto AVOLIO, Maria Emanuela ERRICO, Emilia DI PACE, Maurizio AVELLA
P7	Biodegradable coatings of synthetic fabrics to retain
	microplastic release
	Francesca DE FALCO, Mariacristina COCCA, Vincenzo GUARINO,
	Gennaro GENTILE, Veronica AMBROGI, Luigi AMBROSIO, Maurizio
	AVELLA
P8	Eco-sustainable finishing treatment of polyamide fabrics
	to reduce the release of microplastics during washing
	processes
	Francesca DE FALCO, Maria Pia GULLO, Gennaro GENTILE, Roberto
	AVOLIO, Maria Emanuela ERRICO, Emilia DI PACE, Veronica
	AMBROGI, Maurizio AVELLA, Mariacristina COCCA

68 μ MED International Conference on Microplastic Pollution in the Mediterranean Sea

P9	Microplastic abundance and polymer types in a
	Mediterranean environment
	Nikoletta DIGKA, Catherine TSANGARIS, Helen KABERI, Argyro
	ADAMOPOULOU, Christina ZERI
P10	Biodegradable poly(butylene succinate)-based
	composites for food packaging
	Salvatore MALLARDO, Valentina DE VITO, Mario MALINCONICO,
	Maria Grazia VOLPE, Gabriella SANTAGATA, <u>Maria Laura DI LORENZO</u>
P11	Testing and identification of plastics debris collected from
	mediterranean beaches
	Luca FAMBRI, Denis LORENZI, Riccardo CECCATO, Claudia GAVAZZA
P12	Ingestion of virgin microplastics by adult fish is not
	causing apparent harm
	Boris JOVANOVIĆ, <u>Kerem GÖKDAĞ</u> , Olgaç GÜVEN, Yilmaz EMRE,
	Elizabeth M. WHITLEY, Ahmet Erkan KIDEYS
P13	Removal of heavy metal ions from wastewater using bio
	and nanosorbents
	Anita GROZDANOV, Katerina ATKOVSKA, Kiril LISICKOV, Gordana
	RUSESKA, T. Aleksandar DIMITROV
P14	MWCNT/Polyaniline nanocomposites used for pH
	nanosensors of marine waters
	Anita GROZDANOV, Aleksandar PETROVSKI, Perica PAUNOVIK,
	T. Aleksandar DIMITROV
P15	Analytical approach for the detection of microfibers from
	textile laundry
	Jasmin HAAP, Edith CLASSEN
P16	Microplastic exposure assessment from the perspective
	of nanoparticle research
	Thorsten HÜFFER, Antonia PRAETORIUS, Stephan WAGNER, Frank
	VON DER KAMMER, Thilo HOFMANN
P17	Statistical methodology for identifying microplastic
	samples collected during TARA-Mediterranean campaign

POSTER	SESSION
	Mathilde FALCOU-PREFOL, <u>Mikaël KEDZIERSKI</u> , Marie Emmanuelle
P18	KERROS, Amanda ELINEAU, Maria Luiza PEDROTTI, Stéphane BRUZAUD
P10	From coral triangle to trash triangle - How the hotspot of
	global marine biodiversity is threatened by plastic waste
	Markus T. LASUT, Miriam WEBER, Christian LOTT
P19	Nanoplastic impact on human health: A 3D intestinal
	model to study the interaction with nanoplastic particles
	Roman LEHNER, Alke PETRI-FINK, Barbara ROTHEN-RUTISHAUSER
P20	Study on microplastic release from fishing nets
	Alessio MONTARSOLO, Raffaella MOSSOTTI, Alessia PATRUCCO,
	Marina ZOCCOLA, Rosalinda CARINGELLA, Claudio TONIN
P21	Mitigation of the impact caused by microfibres released
	during washings by implementing new chitosan finishing
	treatments
	Raffaella MOSSOTTI, Alessio MONTARSOLO, Alessia PATRUCCO,
	Marina ZOCCOLA, Rosalinda CARINGELLA, Claudio TONIN
P22	Experimental nuclear applications in marine plastics
	research

François OBERHÄNSLI, Peter SWARZENSKI, Inmaculada TOLOSA, Bruno DANIS, Marc METIAN

#### Microplastic bacterial communities vary over time, P23 space and polymer nature in coastal ecosystem Laura FRERE, Lois MAIGNIEN, Morgane CHALOPIN, Emmanuel RINNERT, Hilary MORRISSON, Christophe LAMBERT, Arnaud HUVET, Julie **REVEILLAUD, Ika PAUL-PONT**

- Plastic soles: microplastic litter in the gastrointestinal P24 tract of Solea solea from the Adriatic Sea Giulio PELLINI, Alessio GOMIERO, Tomaso FORTIBUONI, Gianna FABI, Fabio GRATI, Nora TASSETTI, Piero POLIDORI, Carmen FERRA VEGA, **Giuseppe SCARCELLA**
- First record of levels and distribution of microplastics in P25 offshore sediments of the Adriatic Sea

Alessio GOMIERO, Guido BRUNO, Marco GIRASOLE, Valentina MUSSI,

Giulio PELLINI, Gianna FABI

P26 Non-conventional ultra-sound assisted extraction of alginates from Sargassum seaweeds: from coastal wastes to novel polysaccharide source

<u>Gabriella SANTAGATA</u>, Giorgio GRILLO, Barbara IMMIRZI, Silvia TABASSO, Giancarlo CRAVOTTO, Mario MALINCONICO

P27 Sub-basin scale heterogeneity in the abundance and polymeric composition of microplastics in the Mediterranean Sea

<u>Giuseppe SUARIA</u>, Carlo Giacomo AVIO, Francesco REGOLI, Stefano ALIANI

- P28 **A research on microplastic presence in outdoor air** Meral YURTSEVER, Ahmet Tunahan KAYA, Senem CİFTÇİ BAYRAKTAR
- P29 Commonly used disposable plastic bags as a source of microplastic in environment

Meral YURTSEVER, Ulaş YURTSEVER

### **Smart solutions**

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Recycling initiatives are became a vital part of municipal-sustainability strategies. If adequately designed, they can be opportunities for new markets, creating valuable impressions, maximizing sales opportunities and driving deeper tourist loyalty.

ECOPUNTI.CNR is an initiative developed by the IPCB of CNR, within the OR.C.HE.S.T.R.A. project. The main objective of is to develop an integrated and environmentally sound waste management system, by means of innovative technologies and recycling techniques, supported by an awareness campaign and a communication program. An incentive based recycling system to promote environmentally sustainable practices has been developed by using an innovative technology platform. Reverse Vending Machines (RVM), appositely developed, are placed across the historic center of Naples and inside the boat of Lauro's line. The RVM offer a modern and automated deposit system for collecting, sorting, and handling the return of used beverage containers for recycling or reuse. Each reverse vending machine is equipped with a display screen, internet connectivity and an upgraded user interface and experience. Consumer product companies, local stores or other organizations can sponsor a machine, with contests and prizes to promote their products or services.

When tourists or inhabitants returning their empty containers using the RVM, they benefit from instant rewards accumulated through vouchers produced by the machine. They can then redeem their points for rewards by going in one of the private local stores that precipitate to the initiative. Tourists and citizens will be exposed to a communication program promoting environmental awareness using the RVM as a platform. A website has been designed to integrate the different components of the initiative. This will provide a set of additional benefits, namely: e-learning tools, applications, knowledge sharing. Local commerce will benefit from hosting this initiative and will therefore be provided with a non-conventional way to create a wide network of customer.

# Microplastics from waste-water treatment plants. Preliminary data

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Waste water is a known source of various types of microplastics but their inputs are seldom studied. In this work, we are investigating the amounts and types of microplastics in the waste water of 2 plants serving different communities, one bigger treating mixed domestic and industrial waste water and the other smaller and mostly treating domestic waste water. Samples are taken at the entrance of the plant and from the treated effluent flowing to the environment, treated with a NaCl saturated solution and filtered through GFC/C filters (~1 $\mu$ m) in a laminar flow chamber. Contamination by airborne microplastics is prevented at all times during manipulation and observation under the stereoscopic microscope.

So far we have been able to detect large quantities of microparticles of various sizes especially floating in the treated effluent but also embedded in the particles that make up the sediment fraction of the incoming wastewater. Particles will be sorted by type (fragments, beads, fibers, color and shape), counted, measured, and analysed with FTIR to sort out their natural or non-natural origin. This is a work in progress as part of a master thesis at FCT-NOVA.

# Polystyrene debris determination in seawater by fluorescence measurements

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Microplastics are synthetic polymer debris of different size, shape and density based on their chemical composition. There is an outstanding evidence that microplastics are present in all environments and can reside in the environments for a long time, contributing significantly to pollution. Microplastics comprise a variety of macromolecules like polyethylene, polyethylene terephthalate, polypropylene, polystyrene, polyvinyl chloride, etc. However, polystyrene is very slow to biodegrade and it is increasingly abundant as a form of litter in the outdoor environment, particularly along shores and waterways, especially in its foam form, and also in increasing quantities in the Pacific Ocean. Foamed polystyrene constituted almost 30% of the total debris (ranging from <10% in Mayaguez, PR, to almost 50% in Tacoma, WA). Owing to the presence of phenyl rings into the polymeric structure, polystyrene fluorescence is very strong (in fact, it is used as plastic scintillator). This has implications in diagnostic applications where it is necessary to detect polystyrene debris within a complex environment such as the non-fluorescent seawater by measuring the intensity of visible light (blue) emitted under UV-light exposition. For example, pure solid polystyrene shows a blue emission peak at wavelength of ca. 425 nm, for an excitation wavelength of 365 nm. The principle we have studied for monitoring polystyrene pollution in the seawater environment is schematically shown in Figure 1. It operates in light scatter mode to collect the fluorescence under UV excitation using matched filter and detecting the optical signal with a photodetector.

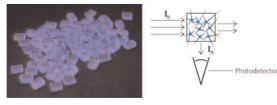


Fig. 1 - Very strong visible (blue) fluorescence of amorphous polystyrene pellets under UV light (365 nm) and schematic representation of the apparatus used for polystyrene pollution measurements.

# Preliminary data on the impact of microplastics on blue-red shrimp (Aristeus antennatus) in the Mediterranean Sea

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Microplastic pollution is currently one of the most concerning threats for wildlife. Ingestion of microplastics has been confirmed in a wide variety of marine species. However, their potential effect remains unclear and only a few studies have addressed this issue in natural environments. The present study aims to assess the presence and potential effects of microplastics in the shrimp Aristeus antennatus, with a particular focus on diet. A total of 29 individuals were collected from three different localities in the western Mediterranean Sea in 2011 and their gut contents were examined in order to determine their diet composition and presence of microplastics. The percentage of occurrence of microplastics varied among localities, with no individuals contaminated in Cap Salou and 40% of occurrence in individuals near the mouth of Ebro's river. Most of the microplastics found were fibres which were tangled up in bundles sometimes. No relationships were found between the presence of microplastics and the condition of the shrimps (condition factor and hepatosomatic index) or the fullness of their stomachs. A multivariate analysis of the gut content showed marginally significant differences between those individuals with microplastics and those without. Detailed analysis of the ecological category of the items (planktonic, hyperbenthos, epibenthos and endobenthos) showed that the contribution of endobenthos to gut content was significantly higher in those individuals that contained microplastics. This trend could be related to natural differences on the feeding habits of Aristeus antennatus. Individuals with preferences for endobenthic invertebrates would have been more exposed to plastic debris deposited on the sediment and would have incorporated more fibres through their diet. Once inside, the complex morphology of the shrimp's gut would have favoured its accumulation. More studies are needed to discuss this issue and explore the potential effects of microplastics upon a larger sample of shrimps.

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# Isolation and chemical characterization of microplastics and their degradation by-products in coastal sediments

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The environmental pollution by plastics debris is attracting increasing attention for the harmful effects that either directly or subtly they exert on living organisms, especially in marine habitats. Indeed the hazard related to entanglement and ingestion by marine species has been the subject of many investigations and institutional projects. Most past programs concerning the marine litter issue have been focused the monitoring of abundance, distribution, size and typology of polluting plastics fragments floating in open sea or deposited in coastal marine sediments, and involved the collection of plastics debris down to millimetre or, less frequently, even sub-millimetre scale size, all of which are often referred to as "microplastics". The classification by size is usually based on mechanical (sieving) or density sorting procedures, poorly effective and not exhaustive in the evaluation of debris of truly micron-sized scale, particularly in the presence of solid matrices. This holds particularly for the analysis of smaller plastics particles deriving from the mainly photo-oxidative degradation processes of larger fragments, resulting in the generation of oxidized functional groups and carboncarbon chain scissions with consequent molecular weight reduction. The combination of photo, thermal, and hydrolytic degradation promotes significant variations of the physical (e.g. density) and chemical characteristics of plastics debris; in particular, the surface chemical composition is affected, thus modifying the behaviour of such microplastics in different environmental compartments. In the present study a procedure aimed at improving the analytical determination of microplastics and microplastics degradation products from shoreline sediments, based on selective solvent extraction and chemical characterization by FT-IR, Pyrolysis-GC-MS and Gel Permeation Chromatographic analyses is described. The results obtained so far, concerning samples collected from a northern Tuscany beach, revealed the dominant presence of poly(styrene) and polyolefin by-products as well as significant differences in their distribution along transepts spanning from the intertidal to the supralittoral zone.

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### Degradation of biodegradable plastic buried in sand

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The growing usage of plastics, mostly in the packaging sector, is causing serious problems concerning their disposal. Many reports highlighted that plastic litter end up in the oceans causing serious environmental problems. In this respect, several attempts have been focused on the usage of biodegradable plastics representing a possible solution to this problem. Biodegradable plastics include either bio-based plastics synthesized from renewable resources, such as poly(lactic acid) (PLA) and polyhydroxyalkanoates (PHA), or plastics produced from fossil resources, including aliphatic polyesters such as polycaprolactone (PCL) and polybutylene succinate (PBS). Although these polymers are considered environmental friendly materials, they also have some drawbacks, such as their limited degradation in marine environment. In fact, recently, PCL was identified as floating particles in Mediterranean Sea.

The purpose of this study was to investigate the degradation of biodegradable polymers such as PLA, PCL, polyhydroxybutyrate (PHB) and polybutylene succinate adipate (PBSA) buried in sand, to verify the behaviour of these polymers in habitat where plastic waste can be stranded when carried by the sea. The degradation of the selected biodegradable polymers was followed by gravimetric methods, Fourier transformed infrared spectroscopy (FTIR), thermal analyses, and scanning electron microscopy (SEM).

# Biodegradable coatings of synthetic fabrics to retain microplastic release

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Synthetic fibres made of polyesters, polyamides, polyacrylics and polypropylenes, represent about the 60 % of the global fibres used for apparel, and such value is steadily increasing year after year. Despite the advantages of the use of synthetic fabrics, such materials have an unpredictable impact on the environment. Recent researches on the sources of microplastic pollution, have discovered that the washing processes of synthetic clothes significantly contribute to the amount of microplastics released in marine environment. In fact, the washing damages the fabric, which releases microfibres in the water. A promising approach to mitigate such release of microfibres, could be the application of a coating on the textile surface, in order to protect the fabric from the stresses of a washing process. Of course, such coating should be homogeneous and not affecting the hand of the textile. For this purpose, among the different techniques currently available, the present work proposes to investigate electrofluidodynamics as a highly versatile and cost effective process to functionalize polymer based textile substrates. Two biodegradable polymers from biosources - i.e., polylactic acid (PLA) and polybutylene succinate adipate (PBSA) – were applied on woven polyamide fabrics. Coating morphology was analysed by scanning electron microscopy (SEM) and further characterizations were performed through attenuated total reflectance (ATR) Fourier Transform InfraRed (FT-IR) spectroscopy and thermogravimetric analysis (TGA). Finally, washing tests were performed to evaluate microplastics release from uncoated and coated fabrics. The experimental results revealed that both PLA and PBSA coatings are able to efficaciously protect the fabrics during the washing process, with a decrease of ca. 80% of released microfibres respect to uncoated fabrics



# Eco-sustainable finishing treatment of polyamide fabrics to reduce the release of microplastics during washing processes

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The washing processes of synthetic clothes have been lately identified as a main source of microplastic pollution in marine environment. During a common washing process, synthetic fabrics undergo mechanical and chemical stresses that induce the detachment of microfibers from the main yarns. Such micro fragments remain in the wastewater, eventually reaching marine ecosystems where they represent a serious threat for the flora and fauna. A possible solution that could mitigate such source of microplastic pollution, is the application of functional finishing treatments able to protect fabrics during washings, reducing the amount of microfibres released. The present work proposes an innovative finishing treatment of polyamide fabrics by using pectin, a natural polysaccharide extracted from fruits. Pectin was chemically modified by reaction with glycidyl methacrylate (GMA), whose vinylic groups were exploited to graft pectin on the surface of the polyamide fabric, creating a coating on the fibres. The effectiveness of the surface treatment was assessed by using the following characterization techniques: scanning electron microscopy (SEM), solid state nuclear magnetic resonance spectroscopy (NMR), attenuated total reflectance Fourier transform infrared spectroscopy (ATR-FTIR) and thermogravimetric analysis (TGA). Furthermore, washing tests of untreated and treated fabrics were carried out to assess the release of microplastics. The washing effluents were filtered and the filters were analysed by SEM to evaluate the amount of microfibres released. The obtained results showed that the application of the pectin-based coating, could reach a reduction of about the 70% of the number of microplastics released by untreated polyamide fabrics during a domestic washing process.

P09

# Microplastic abundance and polymer types in a Mediterranean environment

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Microplastics have become a more and more dominant threat to marine ecosystems. The ubiquity of microplastics is one of the major problems: from the sea surface and water column to the beach and seabed sediment or even ingested by marine organisms, small plastic particles have been found. The present work reports microplastic abundance and composition in sea surface water, beach sediment and biota in the Corfu island area (Northern Ionian Sea). Microplastics detected in samples from water, sediment, fish gut or mussel tissue were stereoscopically observed, categorized by shape, size, colour and their polymer type was identified by FTIR analysis. Microplastic items in sea surface water ranged from 0 particles/km<sup>2</sup> to 1,619,658 particles/km<sup>2</sup>. Microplastics average abundance in beach sediments ranged from 17 to 95 items/m<sup>2</sup>. Out of all fish and mussels tested, the percentage of individuals detected with microplastics was 51.25% and 66.25% respectively, while the average abundance of microplastic detected in fish and mussels was 0.89 particles/fish and 1.25 particles/mussel. The majority of microplastics in all environmental compartments were identified as polyethylene. Relations in size classes and polymer type among environmental compartments are investigated. Results describe a holistic image of the pollution caused by microplastics in the study area and can be useful for an integrated microplastic monitoring.



# Biodegradable poly(butylene succinate)-based composites for food packaging

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Biodegradable and biobased poly(butylene succinate)-based composites containing a β-cyclodextrin/D-limonene inclusion complex were investigated as novel materials for food packaging applications. The composites were prepared by twin screw extrusion, then compression molded to thin films. Formation of the inclusion complex (IC) was proved by infrared spectroscopy, nuclear magnetic resonance, and X-ray diffraction. The addition of CD inclusion complex into the polymeric matrix represented a crucial strategy to preserve D-limonene from evaporation during melt processing of the composites, as shown by TGA analysis. FTIR and TGA analysis evidenced that D-limonene was efficiently encapsulated inside  $\beta$ -CD cavities, since the inclusion complex obtained was thermally stable up to the onset of  $\beta$ -CD degradation temperature. Moreover, the CD-lim complex affected thermal stability of PBS, because of the hydrogen bonding occurring between polar groups of CD-lim and carbonyl groups of PBS, as evidenced by FTIR. Optical micrographs showed homogeneous distribution of the IC within the PBS matrix, possibly favored by interactions between the polar groups of the composite components. DSC analysis highlighted the anti-nucleating action of  $\beta$ -CD-lim complex towards PBS, evidenced by a significant delay in crystallization kinetics of PBS.

# Testing and identification of plastics debris collected from mediterranean beaches

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This presentation is a contribute to shed more light on the type of various plastics that can be retrieved near beaches. According to recent extimation, the main source of plastic marine litter is land-based. For this reason, a selection of plastics wastes has been collected from various beaches and shore-lines of Tyrrhenian Sea, Ionian Sea and Adriatic Sea, as precursor of microplastics in the Mediterranean Sea.

Testing characterization has been devoted to the i) identification of type of plastic and to the ii) determination of the grade of physico-chemical degradation after long term in marine environmental. In fact, a combined effect of sun-light exposure, marine water, micro-mechanical stresses and other factors determine not only a progressive cracking of physical integrity of various plastic items, but also a chemical modification of polymer chains.

Different marine samples of polystyrene PS (foamed and bulk), polypropylene PP, polyethylene PE, polyethyleneterephthlate PET, were firstly identified by Infrared spectroscopy (Medium FTIR - range 4000-600 cm<sup>-1</sup>).

Subsequently, thermogravimetry (TGA) and calorimetry (DSC) showed a specific in dependence on physical-chemical aging. In particular thermal stability, oxidative degradation, glass transition temperature of PS and PET debris, melting and crystallization behaviour of semicrystalline polymers (PE, PP, PET) were determined and compared with respect to correspondent virgin polymers.

Particular attention was also spent to the determination of OIT (Oxidation Induction Temperature) for PE, PP and PS specimens, as indication of preliminary oxidation/degradation occurred during natural aging. Information could be complementary evaluated with TGA and DSC results.

Moreover, Near Infrared (NIR) analysis is also presented and compared in order to allow a local identification of various plastic debris for direct sorting.



## Ingestion of virgin microplastics by adult fish is not causing apparent harm

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In parallel to the microplastic levels in the oceans, the literature on their impacts (as well as their potential toxicity) to all components of the food chain of marine organisms. Among aquatic organisms, fish are particularly susceptible to ingestion of microplastic particles due to microplastics appealing coloration, buoyancy, and resemblance of food. Growth, development, behavior, reproduction, and mortality of fish are identified as main negative effects of microplastic exposure. However, in past experimental setups, fish were usually exposed to microplastics either with concentrations which are unrealistically high or often contaminated on purpose with persistent organic chemicals, or larval stages of fish used. We have tested effects of virgin microplastics in adult fish (seabream Sparus aurita) after 45 days of dietary exposure to environmentally relevant concentration of 6 common types of microplastic. Tested parameters were, (a) the overall growth, (b) the levels of glucose, AST, ALT, LDH, and GGT in their blood, (c) tissue and cytomorphologic changes in the gastrointestinal tract, liver, pancreas, spleen, and mesentery and (d) histopathology analyses of the fish. Our study revealed that neither of the measured parameters differed significantly when the control was compared to the treatments for the adult sea bream.

# P13

# Removal of heavy metal ions from wastewater using bio and nanosorbents

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The rapid industrial development and urbanization have intensified environmental pollution and caused deterioration of eco-systems by accumulation of many pollutants, especially heavy metals. Most of the heavy metals are toxic and their ions are not biodegradable with the tendency to accumulate in the soil, water resources and the living organisms, hence they are significant environmental pollutants. Therefore, the treatment of the heavy metal ions and their elimination from water and wastewater is very important for environmental protection, and thus the public health.

In the frame of this work, the adsorption abilities of natural and nanosorbents, particularly of natural peanut husks, expanded perlite and graphene, to remove Ni(II), Pb(II) and Fe(II) ions from water systems, were investigated.

The influence of the pH (4-8) of the solution, the amount of adsorbent (0.5 -5.5 g/L), the initial metal ion concentration (0.3 - 2.0 mg/L) and the contact time (5-180 min.) on the efficiency of removal of metal ions, was investigated. Thus, the optimal conditions for achieving maximal effectiveness for heavy metals removal, were determined.

The characterization of the sorbents was performed utilizing the following techniques: SEM, TGA, TEM, Raman spectroscopy. Adsorption equilibrium of the systems was analyzed using the following isotherms: Langmuir, Freundlich, Langmuir - Freundlich and Redlich - Peterson.

The maximal adsorption capacity of the peanut husks, perlite and bentonite for Ni(II), Fe(II), Pb(II) was obtained and the percentage of removal was determined.

A comparative analysis for the efficiency of all used sorbents for Ni(II), Pb(II) and Fe(II) ions removal from the three component systems was conducted at the end.

The expanded perlite gave the best results for the removal of Ni (II) and Pb (II) ions, while graphene proved to be excellent adsorbent for Fe(II) ions with an efficiency of 100%.



## MWCNT/Polyaniline nanocomposites used for pH nanosensors of marine waters

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Polymer nanocomposites of conductive polymer, polyaniline (PANI) with Multi Wall Carbon nanotubes (MWCNT) have gained a great interest for their application in environmental and water quality monitoring (where pH value becomes one of the reliable data). Compared to the inorganic counterparts, conducting polymers have advantage in achieving high sensitivity and selectivity by virtue of their chemical and structural diversity.

In the framework of FP7 project COMMON SENSE (OCEAN 2013.2-614155) screen printed electrodes as a pH nanosensors based on nanocomposites of conductive polymer matrix-PANI and MWCNT were prepared by electrochemical polymerization. Characterization was performed by several spectroscopic techniques and electrical measurements.

Electrochemical characterization by means of cyclic voltammetry and steady state polarization was performed in order to determine conditions for electropolymerization of the aniline to PANI. Electropolymerization of the PANI based composites was performed at 0.75 V vs. saturated calomel electrode (SCE) for 40 and 60 minutes. Morphological and structural characteristics of the produced composites were studied using scanning electron microscope (SEM) and Raman spectroscopy, while thermal stability was determined using thermal gravimetric analysis (TGA) and differential thermal analysis (DTA). According to the morphological and structural study, fibrous and porous structure of PANI based composites was detected well embedding MWCNTs. Also, strong interaction between quinoidal structure of PANI with carbon nanostructures via  $\pi$ - $\pi$  stacking was detected by Raman spectroscopy. TGA coupled with DTA showed the increased thermal stability of composites.

The obtained nanocomposites exhibited a high value of conductivity which attributed to the synergy effect of the conductive polymer matrix and carbon nanostructure. Resistivity (i.e. conductivity) changes were measured at different pH (4 to 10) as well as in different marine regions have shown that the MWCNT/PANI nanocomposite exhibited higher resistivity changes.

# Analytical approach for the detection of microfibers from textile laundry

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Sewage from textile laundry processes has been identified as a potentially important source of synthetic fragments in the environment. However, there is still a gap of knowledge regarding the extend of fiber discharge and the main influence factors. Detailed investigations on the release of micro- sized fibers from laundry require precise and reproducible analytic methods. Reported analytics for microplastic fiber detection in sewage are often based on filtration of the effluent combined with visual analysis of the particles e.g. microscopy. This workflow is time consuming and only allows an estimation and extrapolation of the fiber content per wash. Commonly applied particle detection systems like light scattering or laser diffraction are limited due to the non-spherical shape of the fiber or the turbidity of the sewage medium.

In this study, the dynamic image analysis is applied to analyze suspended textile fibers and particles in sewage. This optical detection system allows a fast and non-destructive measurement of fibers and particles in a broad size range (10 - 3500  $\mu m$ ). Furthermore, it enables the statistical analysis regarding various fiber characteristics like size, shape, elongation etc. and gives quantitative information about the number of fibers per wash and the size distribution.

We show applications of the dynamic image analysis to sewage samples from laundry and address the possibilities as well as the challenges in analyzing microsized fibers in complex medium like wastewater.



## Microplastic exposure assessment from the perspective of nanoparticle research

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Although marine littering has been controlled since the 1970s, the debate on the occurrence and the consequences of plastic particles with sizes between 1  $\mu$ m and 5 mm, so-called microplastics, in the environment has received increased attention only in the recent decade. There is a rapidly growing body of published research into the impacts of microplastics on various ecosystems. Yet, our understanding of the occurrence, behavior and transport of microplastics in aquatic systems and to ultimately acquire the ability to assess their risks, remains insufficient. The open research questions and challenges to close these knowledge gaps for microplastics are very similar to topics and challenges for the research on potential environmental implications of engineered nanoparticles (ENPs).

In this contribution current challenges and open questions regarding the fate and exposure assessment of microplastics are related to lessons learned over a decade of studying ENPs in the environment. First, the similarities between both materials are used to identify the transferrable knowledge to microplastics exposure assessment. For example, to understand the transport and transformation processes, laboratory and mesocosm studies as well as environmental fate models for ENPs have been developed in recent years, with varying levels of detail, realism, and spatial resolution. Many of these approaches could serve as a starting point to study microplastics.

This is then followed by a critical discussion of differences requiring specific adjustments for microplastics. In order to quantify the release of secondary microplastics, quantification of mismanaged plastic waste, which is released in the aquatic environment, and determination of the fragmentation rates of the

#### **POSTER SESSION**

released plastic waste in the aquatic environment are required. The aim of this work is to support a more rapid development of the tools and methods required to advance microplastics exposure and ultimately risk assessment to take prompt and necessary regulatory actions.



## Statistical methodology for identifying microplastic samples collected during TARA-Mediterranean campaign

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The goal of this project is to investigate new methods of compiling data coming from samples which have been selected from an internal library containing more than 40,000 samples of microplastics taken during the 2014 TARA cruise in the Western Mediterranean basin. The poster will present the techniques best adapted to investigate large numbers of plastic particles, through a methodological approach which represents a major advance compared to current practices involving considerable human resources. In order to do this, we have implemented a robust statistical methodology to subsample the whole library dataset, leading to a highspeed structural analysis by combining vibrational microspectroscopy and image recognition.

These techniques have been applied to a library of microplastics (a total of 43,000 particles collected from 129 samplings, each comprising between 11 and 8,000 samples) brought together during the TARA Mediterranean campaign (2014). The extraction of microplastics sampled at sea has allowed us to feed the database continuously. In keeping with previous works carried out on the characterization of microplastic samples using the infrared spectroscopy, this work has been carried out jointly by the IRDL and the Villefranche Oceanography Laboratory (UPMC: Université Pierre et Marie Curie). The chemical nature of the matrix of the plastic particles has been characterized using techniques and approaches specifically developed for the study of microplastics (Raman and infra-red microspectrophotometries and multivariate statistical analysis).

# From coral triangle to trash triangle - How the hotspot of global marine biodiversity is threatened by plastic waste

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Southeast Asia harbours the highest marine diversity of our planet. At the same time the countries in the so-called Coral Triangle have the highest potential/risk of plastic pollution to the marine environment. Biodiversity research is still struggling with the sheer inventory of biota, as many marine organisms already are under risk of becoming extinct by human influence. Manado (North Sulawesi, Indonesia) is a case study in the very heart of the Coral Triangle. It is a booming city with a growing population >500.000, right next to Bunaken, one of the iconic world-class destinations of diving tourism, and a national park. Manado is origin to a stream of mismanaged waste entering the ocean every day with an estimated volume of 330 m<sup>3</sup> d<sup>-1</sup> in 2016, with drastic effects to the nearby coastal environment, especially coral reefs and mangroves. Waste management strategies are rudimentary, baseline data on sources, quantity and quality of plastic waste are lacking, micro-plastic in special is hardly addressed, thus urgently needed risk assessment and mitigation concepts lack fundamental knowledge. Here we provide observational data as beach clean-up reports, ROV seafloor surveys, and gut content analyses of commercially caught fish, and accidental findings as the plastic gut content of the "living fossil" Latimeria menadoensis, the iconic coelacanth, as the first collection of available information for this region. To overcome this paucity of knowledge we propose an interdisciplinary action plan for the Manado area, that can be extended and adapted to the wider region of the Coral Triangle.

The Mediterranean Sea also has a high biodiversity, parts of which are already lost or highly threatened. Plastic pollution has a long history although the problem has been addressed by large only recently. The Coral Triangle could profit from expertise and concepts developed in the Med and give back a region where due to the relative short history of plastic waste, and still spatially restricted heavy urbanization, the impact on near-pristine marine ecosystems can be studied in order to find global solutions.

# Nanoplastic impact on human health: A 3D intestinal model to study the interaction with nanoplastic particles

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Much attention has been paid in recent years to the fate of microplastics in the environment. Several studies have shown that microplastics can be taken up by a variety of organisms (e.q. fish, mussels, zoo plankton, sea urchin, birds) and thus can cause adverse effects such as death due to ingestion and entanglement, as well as pro-inflammatory responses. However, recent studies point out that the process of plastic degradation progresses beyond the formation of microplastics. Therefore, microplastic particles are not the end of the chain but a continued disintegration into nanoplastic particles has to be expected. However, up to now, the knowledge of nanoplastic is very sparse since there are only a handful of scientific publications on the subject published focusing on aquatic organisms. Very limited data is available regarding the impact of these plastics on human health, although their transfer to humans via the food chain is increasing. Cellular uptake, accumulation in gastric intestinal tissue and inflammatory reactions by nanoplastics could be shown by in vitro studies. Particles in the nano range show changes in their chemical-physical properties, which is of specific concern regarding in vivo distribution as well as cellular uptake of the particles. As seen with microplastics, nanoplastics can also adsorb hydrophobic chemicals such as polychlorinated biphenyls, thus influencing their potential biological and toxicological impacts to the environment.

In our project, we therefore aim to demonstrate the effects of nanosized plastic particles on the human intestine tissue barrier, since the gastrointestinal tract represents the main possible uptake route of nanoplastic by the humans.

### Study on microplastic release from fishing nets

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In the marine environment, plastics of various size, classes and origins are ubiquitous and affect numerous species that become entangled in or ingest plastics.

Under environmental conditions, larger plastic items degrade to so-called microplastics (MP), fragments typically smaller than 5 mm in diameter. CNR-ISMAC started in 2016 the study of the release of microplastics debris coming from fishing nets abandoned in the marine ecosystem, the so-called ghost nets. From a macroscopic point of view, the phenomenon represents a serious threat for the marine environment, but under ageing conditions (UV radiation, waves action, etc) the nets can further degrade into microplastics. These fragments can be a vehicle for persistent organic pollutants and they can be ingested by aquatic organisms, especially filter feeders.

The morphological analysis performed by SEM (Scanning Electron Microscopy) demonstrated that the ghost nets recovered by the North Adriatic Sea are characterized by the presence of polymeric fragments (fibrils) that protrude from the surface of the polymeric filaments; these plastics fragments are able to generate microplastics in the marine environment. Moreover, in order to quantify the microplastics release from abandoned fishing nets, an internal method was set up and experiments with Linitest equipment were performed, merging the conditions reported in standard methods UNI EN ISO 105-E02, UNI EN ISO 105-E03 (used in textile field to assess fastness to sea water) and UNI EN ISO 105-C06 (fastness to washing). The quantification of release was performed by SEM with a statistical counting method. This work can be a contribute to the study of the formation of microplastics from abandoned fishing gear, that nowadays are considered among the main sources of this environmental issue.

# Mitigation of the impact caused by microfibres released during washings by implementing new chitosan finishing treatments

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Chitosan is a material of natural origin obtained by the deacetylation of chitin, widely commercially available and of low cost, because deriving from wastes of food industry. Chitosan has the advantages of being non-toxic, biocompatible and completely biodegradable, with a chemical structure similar to that of cellulose. It has been already applied in the textile industry to natural fibers. In this study it was applied to synthetic fibers to form a coating with the aim of reducing microplastics release from textile washings processes, that is an emerging environmental problem. Solutions of chitosan in citric acid were prepared in different concentrations and applied to polyester standard fabrics by means of a lab scale Foulard equipment. Washing cycles were performed according to a standard method (UNI EN ISO 105 C06) with a Linitest equipment to test the resistance of the coating and the efficiency of the treatment to decrease microplastics release from polyester fabrics. Scanning Electron Microscope (SEM) was used to check the presence of the coating before and after washing processes. Washing effluents were filtered by means of a peristaltic pump on filters with a porosity of 5 µm. The filters were observed with SEM analysis and microfibers were counted with a statistical method. The results achieved indicated a good behaviour of chitosan treatment to reduce the microplastics release from polyester fabrics. This achievement obtained at lab scale needs further studies to obtain a scale up of the treatment at industrial level.

### Experimental nuclear applications in marine plastics research

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Most plastics or 'polymers' are based either on the carbon or silicon atom, and can link to a hand-full of other elements (i.e., H, N, O, Cl and S) that define its physicochemical character and ultimate end-use. As they often form molecules that consist of 1000s of bound monomers, plastics typically have high molecular weight occurring in many forms, including some of minute size (microplastics), which are generally thought to represent an important threat to coastal and marine ecosystems.

Most plastics are inherently buoyant in water, and are thus efficiently dispersed around the world by wind, waves, and currents. The surface layer of the world's oceans today is thought to contain more than 5 trillion (5 x  $10^{12}$ ) pieces of mostly microplastics (< 5 mm) which can persist for decades. Plastics have been found to efficiently absorb various types of contaminants, acting as both source and longrange vectors for these compounds. In turn, these plastic-bound compounds have the capacity to enter trophic networks through different pathways, potentially enhancing bioaccumulation and/or biomagnification efficiencies.

The global proliferation of (contaminated) marine plastics has led to the recognition that uptake and loss kinetics of microplastics by marine organisms and resultant trophic network transfer processes are critical new research areas that need to be addressed using both experimental and observational methods. In this context, IAEA Radioecology Laboratory is developing new approaches that apply nuclear techniques to delineate degradation and transport kinetics of microplastics on select marine organisms in controlled, environmentally-relevant conditions. This presentation will provide an overview of such techniques in marine plastics research and draw relevance to policy implications.



# Microplastic bacterial communities vary over time, space and polymer nature in coastal ecosystem

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For the past decade several field studies have demonstrated that plastic debris and microplastics provide a novel substrate for habitat and transport of a wide range of marine organisms including prokaryote micro-organisms. The aim of the present study was to investigate microplastic-associated bacterial communities in coastal ecosystem (Bay of Brest, Brittany, France) using 16S rRNA gene amplicon sequencing on the Illumina MiSeq platform. Bacterial communities were analyzed on floating microplastics collected at two contrasted sites (a recreational marina vs. the center of the bay where most of the mixing of fresh and oceanic water occurs) and at two sampling times (October vs. December) in order to identify: (i) the taxa associated to microplastics compared to those found in the surrounding seawater, and their spatiotemporal variation; and (ii) potential differences as a function of polymer nature and size range of the collected microplastics.

Results showed that microplastic bacterial biofilm assemblages differed from

#### **POSTER SESSION**

surrounding seawater, and that the Vibrio genus was enriched in the microplastic matrix. Spatial and temporal differences were observed in microplastic bacterial community assemblages, which was not necessarily the case for the surrounding seawater communities harboring the colonizing pool. Polystyrene (PS) exhibited distinct bacterial community assemblages than polyethylene (PE) and polypropylene (PP) but no effect of particle size was demonstrated. All together, these data show that distinct taxon are selected and enriched from the colonizing pool as a function of the substrate and site. This study provides the groundwork for future work focusing on i) the substrate specificity (plastic polymer vs. other types of organic and mineral suspended particles) and ii) the identification of environmental parameters shaping the microplastic bacterial communities over space and time that will improve our understanding of the ecological consequences of microplastic contamination in marine ecosystems.



## Plastic soles: microplastic litter in the gastrointestinal tract of Solea solea from the Adriatic Sea

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Large amounts of tiny debris from plastic objects in the world's oceans represent a serious threat to human health and marine ecosystems. Once released into the aquatic environment plastic litter is broken down to smaller pieces through mechanical and physical action of UV light, waves, wind, etc. The resulting particles may become so small that they are readily taken up by fish, crustaceans, and mollusks in the aquatic environment. The mounting evidence of the occurrence of plastic particles in marine organisms that are part of the human food chain urgently calls for further and deeper investigations.

Therefore, this work was aimed at investigating for the first time the occurrence, amount, typology and spatial distribution of microplastic (MP) litter in the gastrointestinal tract of Solea solea in northern and central Adriatic Sea.

The digestive tract contents of 533 individuals collected in fall during 2014 and 2015 from 60 sampling sites were examined for MP. These were recorded in 95% of sampled fish, with more than one MP item found in around 80% of the examined specimens, for a total of 4,566 items, which means  $1.73 \pm 0.05$  items per fish in 2014 data and  $1.64 \pm 0.1$  in 2015. The most commonly found polymers were polyvinyl chloride, polypropylene, polyethylene, polyester, and polyamide, 72% as fragments and 28% as fibers. A peculiar spatial distribution was observed while assessing the abundance and the relative composition, with two hot spots: the Gulf of Trieste and off the Po river mouth.

# P25

# First record of levels and distribution of microplastics in offshore sediments of the Adriatic Sea

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Plastic waste is of increasing concern in the aquatic environment. A large portion of the plastic waste is produced onshore and reaches the marine environment, which is considered the main sink of plastic debris. Floating plastic particles accumulate in pelagic habitats. However, due to the biofilm formation they eventually sink and accumulate on the seafloor together with non-buoyant by design plastic particles posing risk to the benthic communities. There is, however, a considerable lack of standardized methods for micro plastic particles occurrence and composition characterization. In the present study, a benchmark among the best available extraction and detection technologies is presented. Average amount and polymeric composition of plastic particles in two offshore areas located in the northern Adriatic Sea were compared with two coastal sites. Small microparticles (< 500 µm) were extracted from 2 kg sediment samples and analyzed by sequentially visual microscopic inspection, SEM, EDS, FTIR and  $\mu$ -Raman microscopy and partly by thermal desorption pyrolysis gas chromatography/mass spectrometry. Detected plastic particles were identified as polypropylene, polyethylene, polyethylene terephthalate, polyvinylchloride, polystyrene and polyamide. An attempt to identify and quantify the organic plastic additives was performed being 1,2benzenedicarboxylic acid, dimethyl phthalate, diethylhexyl phthalate, dibutyl phthalate, diethyl phthalate and tert-butylphenol the most recurrent compounds. The preliminary results indicate that mean number of plastic particles was 1.2 per kg wet sediment at the two offshore sampling areas and 12.3 per kg wet sediment in the coastal sites.



### Non-conventional ultra-sound assisted extraction of alginates from Sargassum seaweeds: from coastal wastes to novel polysaccharide source

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Sargassum algae, belonging to the brown seaweeds class, are diffused both in tropical and temperate regions. During the last years, they underwent an uncontrolled diffusion mainly caused by eutrophication of hydric bodies, bringing to huge stockpiles of toxic biomass spreading in waters and beaches.

Seaweeds are particularly rich of salts (Ca, Na) of alginates, a known class of biodegradable and biocompatible polysaccharides with stabilizing and gelling properties. Cross-linked sodium alginate (SA) has many applications, from drug delivery to herbicides, microorganism immobilization and as a food-stabilizing agent. In this work, an eco-sustainable enabling extraction method based on ultrasound technique has been used to obtain SA from Sargassum seaweed wastes, the novel raw material has subsequently used as a polymeric matrix for agricultural mulching films.

US can dramatically affect biomass structure maximizing mass transfer in liquid suspensions and inducing the wall cell rupture. US-assisted extraction of Sargassum Algae has been compared with the conventional protocol, observing a huge reduction of time and volumes. A one-step extraction protocol may cut down waste and chemicals disposal. Isolated SA was structurally and thermally characterized with GPC, TGA, FTIR-ATR and NMR analyses, and compared with commercial product. Finally, a cast film was obtained.

In conclusion, this work proposes a new and sustainable waste valorisation protocol, starting from a negative-cost biomass and leading to the production of an exploitable bio-derived polymer.

# Sub-basin scale heterogeneity in the abundance and polymeric composition of microplastics in the Mediterranean Sea

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The Mediterranean Sea is one of the most polluted areas of the world with regard to microplastics. However, detailed knowledge about the spatial variability in microplastics abundance and composition is lacking. We present here the result of a large-scale survey of microplastic pollution in Mediterranean surface waters, demonstrating sub-basin scale heterogeneity in the abundance and in the polymeric composition of these floating particles, which is likely the ultimate result of a complex interplay between pollution sources, sinks and residence times of different polymers at sea. A total of 74 neuston samples were collected in the central-western Mediterranean during two consecutive cruises carried out in 2013. In the laboratory, all samples were sorted at the stereomicroscope and all plastic particles were handpicked using laboratory tweezers, counted, weighed and classified according to their size, shape and colour. All particles >700µm were characterized through FT-IR analysis (n = 4050 particles). Sixteen different classes of synthetic materials were identified. Low-density polymers such as polyethylene and polypropylene were the most abundant compounds, followed by polyamides, plastic-based paints, polyvinyl chloride, polystyrene and polyvinyl alcohol. Less frequent polymers included PET, polyisoprene, PVS, EVA, polyepoxide, paraffin wax and polycaprolactone. Overall, plastic particles were significantly (p=0.002) less abundant in the Adriatic Sea (467.79  $\pm$  1133.88 g/km<sup>2</sup>) than in the western Mediterranean (811.08  $\pm$  1769.75 g/km<sup>2</sup>). Significant differences between sub-basins were found also in the relative occurrence of different polymers. The composition of western Mediterranean samples was dominated by low-density polymers such as polyethylene and polypropylene, probably indicating longer residence times of particles at sea. Adriatic samples instead were more heterogeneous and rather characterized by a higher presence of high-density polymers such as paint chips, PS, PVC, PVA and PAs, suggesting a closer proximity to pollution sources and probably reflecting the distinctive hydrological features of the Adriatic basin.

### A research on microplastic presence in outdoor air

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Microplastics (MPs) are among major micro-pollutants which can be found in water sources and air in substantial quantities, and which still are not covered by standard sorting and analysis procedures. Even though researchers often come across larger "MP fragments and films" in water sources under review, it is understood that the rather more common microplastics in the WWTPs and the atmosphere are of the "MP fiber" variety.

Microplastic fibers from a number of sources can settle on the ground, only to be lifted into the air thanks to wind and air flows; remain suspended in the air for some time; and find its way to the lungs of people through the respiratory system, just like air pollutant emissions. The objective of the present study is to make an assessment of the volume of microplastic fibers finding their way to soil, compared to those freely floating in the air due to their light weight and micro-nano-size. For this purpose, the amount of MP in the samples taken through half an hour of vacuum suction (0.3 m<sup>3</sup>/min) at the intercity terminal located on the entrance to the university campus in Sakarya province, as well as of MP fallouts in the soil samples taken from the same location, were assessed. The samples taken in the period December 2016 - May 2017 revealed different amounts of MP fibers in each month, not to mention a number of MP particles, while dark blue, white, transparent and brown were the most frequently observed MP colors. The factor leading to the variation in MP volumes from one month to the next is the change in weather. For instance, samples taken after rainfall revealed substantial falls in microplastic counts. The volume of MP in circulation in the atmosphere can vary substantially along the axes of time, space, and seasonal conditions. That is why it is difficult to specify a clear figure for MP volumes a location is expected to have; however, it is possible to claim that atmosphere certainly contains some amount of MP, albeit in varying quantities. That is why microplastics in the air should be taken into consideration among PMs (as coarse, fine, or ultra-fine particles) which are considered major air pollutants, alongside the emissions of SOx, NOx and COx which had been under close supervision for guite some time, and thus should be studied in depth in terms of their impact on health.

# Commonly used disposable plastic bags as a source of microplastic in environment

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The use of disposable plastic bags, one of the leading plastic products encountered in daily life, have been mostly impossible to prevent, even though a falling trend is observed in the light of taxes and fines introduced in a number of countries. The term microplastics often recalls the fission of larger plastics into smaller "plastic fragments". Yet, MP fibers which are torn apart from synthetic textiles, and MP films which are produced as plastic bags crumble away in time, should also come to one's mind. The smaller the size of a plastic, the larger the number and range of species it could affect and harm.

The present study compares some characteristics of aged and new LDPE plastic bags. In this context, physical properties (such as durability, flexibility, surface morphology and fragmentation) of three aged bags (grocery bag, shopping bag and blue thin garbage bag) which were kept under normal conditions in a dark cupboard for 4 years, after approximately 3 months of exposure to sunlight outdoors in springsummer (annual average relative humidity is 73%) were compared against those of new bags. The deforming impact of UV in sunlight is well known; the bags exposed to sunlight for a specific term, followed by an extended period of no intervention, were found to be pretty fragile, fragmenting with even the softest of touches. Scanning Electron Microscope (SEM) images were taken to be able to get a better definition of the surfaces of those bags. The images reviewed revealed that aged bags were in the form of fragmented MP films with sharp edges and fractures, while new bags resisted tear when pulled with forceps, producing flexible, stretches, and thinned edges. In a nutshell, complete degrading and dissolution of plastic bags exposed to natural elements in the environment is not a very realistic expectation (in the last period of the degradation process), as they unavoidably turn into MPs, which, essentially, is a greater source of environmental pollution. That point will be even more clear, considering the plastics floating in water sources and oceans, and the biota exposed to this form of pollution.