DeFishGear

MICRO 2015

Seminar on Microplastics Issues

BOOK OF ABSTRACTS

Piran, May 4-6 2015

DeFishGear project

Derelict fishing gear management system in the Adriatc region

Proceedings of the

MICRO2015

Seminar on microplastics issues

Piran, May 4-6 2015

Seminar was organized by National Institute of Chemistry within the **DeFishGear project** (Derelict fishing gear management system in the Adriatic region) and in cooperation with other project partners.

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DERELICT FISHING GEAR MANAGEMENT SYSTEM IN THE ADRIATIC REGION

Coordinated actions for a litter-free Adriatic

PROJECT OVERVIEW

Marine litter - any persistent, manufactured or processed solid material discarded, disposed of or abandoned in the marine and coastal environment-poses a major threat to wildlife and ecosystems, as well as, to humans and their livelihoods. Marine litter is a complex and multi-faceted issue with environmental, economic, safety, health and cultural implications. Marine litter knows no boundaries and represents a pervasive and persistent problem that expands beyond borders away from the source of origin. Coordinated and multi-sectoral action is key to combating marine litter.

Photo:Thomais Vlachogianni

The DeFishGear project aims to facilitate efforts for integrated planning to reduce the environmental impacts of litter - generating activities and ensure the sustainable management of the marine and coastal environment of the Adriatic Sea. Ultimately, the DeFishGear project will provide a strategic input to regional efforts in successfully achieving good environmental status in the Mediterranean Sea.

The DeFishGear activities are implemented by a multi-disciplinary team comprising academia, research institutes, national and local authorities and NGOs from all seven countries of the Adriatic Sea, reinforcing and strengthening cooperation and fostering joint and harmonized actions towards a litter-free Adriatic.

MAIN LINES OF ACTIVITY

- Carrying out a comprehensive assessment of the status (amounts, composition, impacts) of marine litter (macro-litter & micro-litter) in the Adriatic through harmonized and coordinated monitoring activities;
- Development of recommendations and policy options based on sound-scientific evidence and knowledge to meet regional and national objectives regarding marine litter (Marine Strategy Framework Directive, Regional Action Plan on Marine Litter Management in the Mediterranean, Ecosystem Approach, etc.);
- Establishment of a Regional Network of Experts on marine litter;
- Development of capacities to monitor marine litter in a harmonized way through reinforced exchange of experiences, techniques and know-how;
- Setting up of a system to collect and recycle derelict fishing gear and implementation of 'fishing for litter' activities, in an environment-friendly way;
- Targeted awareness raising for fishermen, policy makers, educational community and others on the impacts of marine litter and the types of action needed to effectively address this issue.

Phto: Ante Žuljević

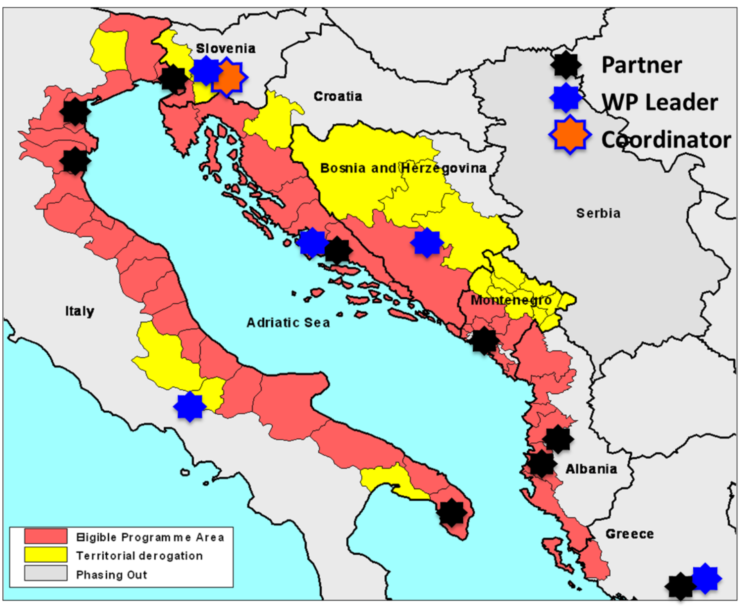
MAIN EXPECTED RESULTS

- Improved knowledge base on the occurrence, amounts, sources and impacts (including socio-economic impacts) of all types of marine litter including micro-plastics in the Adriatic;
- Harmonized marine litter monitoring activities in the Adriatic and enhanced collaboration as a building block for future actions;
- A strengthened & reinforced science-policy interface on marine litter;
- Joint, coordinated and/or complementary schemes to manage human activities generating litter in the Adriatic Sea and strengthened implementation of relevant policy frameworks (MSFD, ICZM, ECAP, MSP, etc.) at the regional level.

Phto: Mojca Trkaj

Project duration November 2013 - March 2016

Project value 5.353.756 €



DeFishGear
project map

PROJECT PARTNERSHIP

LEAD PARTNER

National institute of Chemistry, Ljubljana, Slovenia

Photo: Pierpaolo Giordano

PROJECT PARTNERS

- Italian National Institute for Environmental Protection and Research, Chioggia, IT
- Ca' Foscari University of Venice, IT
- Mediterranean Consortium, Rome, IT
- Regional Agency for Environmental Protection in the Emilia-Romagna Region, Cesenatico, IT
- Institute for Water of the Republic of Slovenia, Ljubljana, SI
- University of Nova Gorica, the Laboratory for Environmental Research, Nova Gorica, SI
- Institute for Oceanography and Fisheries, Split, HR
- Hydro-Engineering Institute of the Faculty of Civil Engineering, Sarajevo, BA
- Institute of Marine Biology, University of Montenegro, Kotor, ME
- Agricultural University of Tirana, Laboratory of Fisheries and Aquaculture, Tirana, AL
- Regional Council of Lezha, Lezha, AL
- Mediterranean Information Office for Environment, Culture and Sustainable Development, Athens, GR
- Hellenic Centre for Marine Research (with participation of the Institute of Oceanography and Institute of Marine Biological Resources and Inland Waters), Anavyssos, GR
- Public Institution RERA SD for Coordination and Development of Split Dalmatia County, Split, HR
- Euro-Mediterranean Centre on Climate Change, Lecce, IT

ASSOCIATES

- PlasticsEurope AISBL, Brussels, BE
- Ministry for Agriculture and Environment, Ljubljana, SI
- Italian Ministry of Environment, Land and Sea, Rome, IT
- Croatian Environment Agency, Zagreb, HR
- Fishing League, Rome, IT
- Agency for Watershed Area of Adriatic Sea Mostar, Mostar, BA

Photo: Ante Žuljević

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IPA Adriatic Cross-border Cooperation Programme

The IPA Adriatic Cross-border Cooperation Programme (hereinafter called Programme) is the result of joint programming work carried out by the relevant participating countries (Italy, Greece, Slovenia, Croatia, Montenegro, Albania, Bosnia and Herzegovina and Serbia) and is part of the cooperation process in the Adriatic area. The Programme draws its strength and incisiveness from the wide experience, gained during the previous Programme period producing concrete results from the studies and analysis financed in the past.

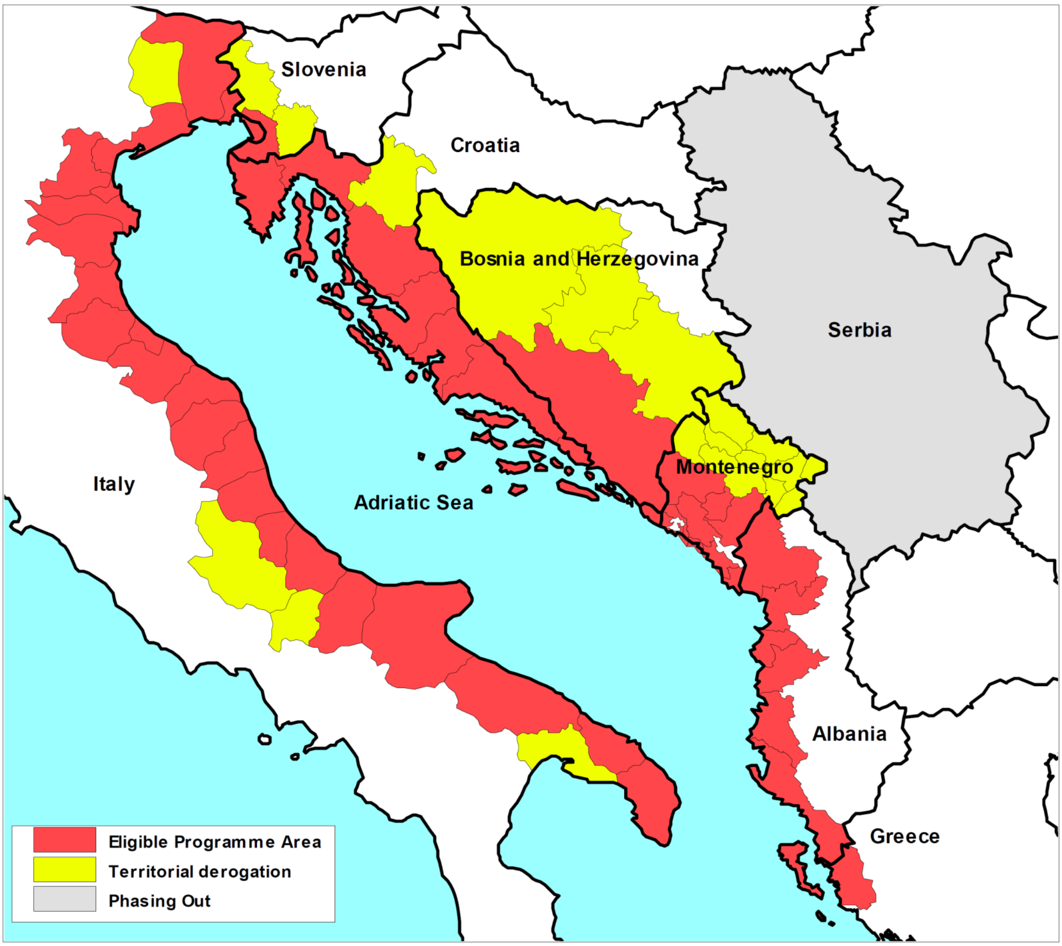
IPA stands for Instrument for Pre-accession Assistance and the IPA Adriatic Programme supports Candidate and Potential Candidate Countries for their progressive alignment with the standards and policies of the European Union, including where appropriate the acquis communautaire, with a view to membership.

Many factors make cooperation in the Adriatic area important today, particularly from a political and economic point of view:

1. Factors connected to the political stability of the area. Following ten years of conflict, the area is now moving towards progressive integration both "vertical" (within European and International institutions) and "horizontal" through the creation of a free trade area;
2. Factors connected to geographic and cultural proximity which make possible the intensification of multilateral relationships among Adriatic coastal regions to support local processes of harmonious growth, sustainable development and unity among peoples.

Eligible area:

Three EU Member States (Italy, Slovenia and Greece), one Candidate Country (Croatia) and three Potential Candidates Countries (Bosnia and Herzegovina, Montenegro and Albania) are participating in the Programme. Additionally, a phasing out participation by Serbia is envisaged for joint projects in the field of institutional co–operation.



More info: www.adriaticipacbc.org

Dear MICRO 2015 participants,

It is a great honor and pleasure to welcome you to the MICRO 2015 seminar. While microplastics may themselves be small and can easily be overlooked, they are a growing problem for our oceans and seas, rivers and streams – mostly as a consequence of the enormous amounts of plastics that we make, use and discard. Current research is providing shocking evidence of both the omnipresence and abundance of microplastics and, although their long-term effects on these watery environments are still unknown, their effects are slowly being documented.

This seminar is devoted to the exchange of the most recent findings on all aspects of microplastics from its sources to its effects on biota, its pollutant transfer role in - both in marine and freshwater environments. It is clear that all of these aspects must be understood and addressed if we are to have a chance of mitigating the negative effects of microplastics in our environment.

An important goal of the DeFishGear project is to increase awareness about microplastics pollution and encourage activities to reduce their emmisions in the Adriatic Sea and the countries that surround it. Therefore regional participation in this seminar is a very important element of this event. Namely, we hope that all participants will further spread understanding of the issues associated with microplastics and act as local leaders in reducing microplastics in our seas and oceans.

Only together can we keep our shared waters, wherever we live, in good health for the creatures and plants that live there, for the food they provide us, and for the sheer beauty and wonder they can give us.

Welcome!

Dr. Andrej KRŽAN
DeFishGear Coordinator and MICRO 2015 Chairman

Programme

Monday, May 4

18:00 – 20:00 Registration and networking event

Tuesday, May 5

8:00 – 8:45 Registration

8:45 – 9:00 Opening

Occurrence of microplastics in the environment

9:00 – 9:40	**IL1**	**GALGANI Francois**	**The Mediterranean Sea: From litter to microplastics**
9:40 – 11:00	OL 1	Palatinus	Surface microplastic quantities and distribution in the Slovenian part of the Trieste bay
	OL2	Smodlaka	Marine plastic debris in the north-eastern Adriatic
	OL3	Mazziotti	Sea surface microplastics distribution on Emilia-Romagna coast (Northwestern Adriatic sea - Italy): DeFishGear project preliminary results
	OL4	Suaria	Neustonic microplastics in the Southern Adriatic Sea

11:00 – 11:30 Coffee break

11:30 – 12:10	**IL 2**	**TWEEHUYSEN Gijsbert**	**Sampling River Litter**
12:10 – 13:30	OL5	Vianello	Occurrence and distribution of floating microplastics in the North Adriatic Sea: preliminary results.
	OL6	Mori	Water Flow Resistance of Plankton Net
	OL7	Liubartseva	Modeling the transport of floating plastic debris in the Adriatic Sea
	OL8	Berden Zrimec	Seabed mapping with remote sensing

13:30 – 15:00 Lunch break and Poster session

Characterization of microplastics

15:00 – 15:40	**IL3**	**LAFORSCH Christian**	**Microplastics in freshwater ecosystems**
15:40 – 16:40	OL9	Pojana	An integrated approach for the classification of Plastic Large Micro Plastics (LMP) in beach sand
	OL10	Horvat	Techniques useful for characterization of microplastics (ATR IR, NIR, Optical microscopy and IR microscopy)
	OL11	Pojana	Characterization of Plastic Large Micro Particles (LMP) from the beach sand of Lido Island (Venice, Italy)

16:40 – 17:10 Coffee break

Microplastics as vectors for biological and chemical contaminants

17:10 – 17:50	**IL4**	**KARAPANAGI-OTTI Hrissi**	**Hazardous Chemicals and Microplastics in Coastal and Marine Environments**
17:50 – 18:30	OL12	Kržan	Influence of Plastics Weathering on Adsorption of Pollutants
	OL13	Turk	Biofouling community and degradation rate of bioplastic in the marine environment

19:30 - Conference Dinner (optional)

Wednesday, May 6
Impacts on biota

9:00 – 9:40	**IL5**	**FOSSI Maria Cristina**	**Do fin whales (Balaenoptera physalus) feed in microplastic impacted areas in the Pelagos Sanctuary**
9:40 – 10:20	OL14	Bajt	Microplastics in sediments and fish of the Gulf of Trieste
	OL15	Regoli	Ecotoxicological risk of microplastics in marine organisms

10:20 – 11:00 Coffee break

Prevention and mitigation

11:00 – 11:40	**IL6**	**VLACHOGIA-NNI Thomais**	**Plastics & microplastics pollution in the marine environment: Challenges and solutions**
11:40 – 12:40	OL16	Mosotti	LIFE+ MERMAIDS PROJECT: Mitigation of Microplastics Release Caused by Textile Washing Processes
	OL17	Raddadi	Petroleum-based plastics biodegradation by anaerobic marine consortia

12:40 – 14:00 Lunch break and poster session

Discussion & Conclusions Chair: ANDREJ KRŽAN
14:00 – 14: 30 Panel statements 6 x 5 min
14:30 – 15:30 Discussion + Conclusions

15:30 – 16:30 Coffee break and networking

Content

Invited lectures

Oral contributions

Posters

INVITED LECTURES

The Mediterranean Sea: From litter to microplastics

Francois Galgani

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Human pressures on the oceans have increased substantially in recent decades and coastal and marine human activities generate considerable quantities of waste. Much of this litter will persist in the sea for years, decades or even centuries. The occurrence of litter has been demonstrated worldwide, in oceanic gyres, on shorelines, in sediments and in the deep sea but the Mediterranean Sea is one of the main affected area and, although they vary between countries; some of the largest amounts of Municipal Solid Waste are generated annually per person (208 – 760 kg/Year, http://www.atlas.d-waste.com/). Plastic, which is the main litter component, may comprises up to 95% of waste accumulated on shorelines, the surface or sea floor. Surveys conducted to date show considerable spatial variability with accumulation rates varying widely as influenced by many factors, such as the presence of large cities, shore use, hydrodynamics and maritime activities. A majority of these materials decompose slowly. It may take centuries for physical, chemical and biological processes to degrade plastics to microplastics and nanoplastics where they also occur ubiquitously at the surface, in sediments and on beaches, and have been detected in marine organisms. Persistence is a key characteristic of plastics and our knowledge on degradation processes need to consider many aspects such as abrasion (mechanical), photo degradation, thermal, chemical and biological degradation. As a consequence, microplastics comprise a very heterogeneous group, varying in size, shape, colour, chemical composition, density and other characteristics. Most of the studies focus on sampling the sea surface and/or water column and intertidal sediments. In the Mediterranean Sea, Mean sea surface plastic was found in concentrations up to 80 000 -250000 particles / km² giving estimated weights of 600 - 3000 tons for the whole basin. At this scale, the spatial distribution is irregular, with a patchy plastic pattern that may be related to the variability in the Mediterranean surface circulation disabling the formation of permanent accumulation areas. Beach survey on the Mediterranean (Spain, France, Malta, and Greece) revealed an abundance of pellets on all of the studied beaches reaching 1,000 pellets/m2 with a highly variable abundance of virgin pellets. Micro plastic pollution has also been demonstrated into sediments from the deep Mediterranean Sea.

Little has been 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, because of their physico-chemical properties (high surface to volume ratio), flotation, their size close to membrane permeability and their potential to be directly ingested by the smallest marine species. There is however a lack of validated research methodologies and data on environmental concentrations and impacts.

To support actions to be taken in order to minimize impacts on the marine environment, the European Commission developed the Marine Strategy Framework Directive (MSFD), establishing a framework within which Member States must take action to achieve or maintain Good Environmental Status (GES). Microplastics is one of the indicators (indicator 10.1.3) listed in Annex I of the MSFD for determining GES and has been defined as "trends in the amount, distribution and, where possible, composition of microparticles (in particular microplastics)". If methodological protocols in Europe are now available for their assessment, harmonization is necessary for common and comparable approaches. Definition of baselines and targets is still under progress to support not only the implementation of monitoring but also reduction measures to be taken in 2016.

Finally, research has become critical in order to better support public policies. It will need to focus on degradation processes, the distribution and densities of microplastics and related effectors (human factors, hydrodynamics, geomorphology etc.), novel methods and automated monitoring devices and finally the rationalization of monitoring including the management of data.

Sampling River Litter

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River litter is in its essence an emission into the environment of products that have been produced or used for economical purposes, either as a resource, a finished product or a semi-finished product. They have lost almost all of their economical value and remain in the environment as a polluting substance: litter.

A river concentrates these products and transports them from the watershed to the seas. Understanding the caracteristics of litter and how litter behaves in riverine conditions is critical to assess the abundance of litter in a river and to estimate the contribution of a river to the pollution of the marine environment.

At present very limited experience is available on the methods with which the presence of riverine litter can be measured. Many studies have been done with the use of a sampler with a mesh size of 0,3 mm (manta nets) but mostly in a marine environment or in larger fresh water lakes. Waste Free Waters has developed a sampler with a mesh size of 3,2 mm especially suited for sampling in riverine conditions. Both methods collect different fractions of the litter and could be regarded as complementary. In the presentation these differences will be discussed.

In 2014 a European project is finished where the presence of riverine litter in 4 European rivers was measured by a number of different methods.

The study focussed on the identification and assessment of riverine input of marine litter. Main challenges of the study included selection of effective sampling methods, taking into account riverine and meteorological circumstances, select relevant sampling locations in each river and select relevant methods for analysis and litter characterization. The project brought new insights on the behavior and occurrence of litter and how to measure and report the results in a coherent way. The project results will be discussed in European Technical Committees.

This presentation will discuss some of the findings and will present methods for sampling and characterization of different types of riverine litter with different methods, their drawbacks and their potential.

Microplastics in freshwater ecosystems

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Among marine litter, plastic waste is of growing concern, as nowadays it has become ubiquitous in the oceans, be it packaging material, household items or industrial waste. Floating fragments accumulate in pelagic habitats forming great garbage patches. Non-floating debris accumulates on the seafloor and in beach sediments, posing risk to the biota. Biofouling and mechanical abrasion cause the formation of microplastic particles making them more prone to consumption by aquatic organisms. These particles may then be incorporated into the tissue of organisms and consequently accumulate in higher trophic levels. Next to mechanical impairments of swallowed plastics mistaken as food, many plastic associated chemicals have been shown to be carcinogenic, endocrine-disrupting or acutely toxic. Moreover, the polymers can adsorb toxic hydrophobic organic pollutants and transport these compounds to otherwise less polluted habitats. Along this line, plastic debris can act as vector for alien species and diseases. Although there is a degree of awareness for the potential risks, the impact of plastic particles on aquatic ecosystems is far from being understood.

A large portion of the plastic waste is produced onshore and then enters the marine environment via water discharge to the river systems. Hence, the oceans are considered as the main sink of plastic debris. However, there is a considerable gap of knowledge about the contamination of freshwater ecosystems with plastic particles. Recent studies report not only the presence of microplastics in freshwater ecosystems, but show that contamination is as severe as in the oceans. Therefore, freshwater ecosystems do not only act as a source of plastic particles for the oceans, they also act, at least temporarily, as a sink. This may come along with all the associated harmful consequences that have been reported previously for marine ecosystems.

Hazardous Chemicals and Microplastics in Coastal and Marine Environments

Hrissi K. Karapanagioti

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The term microplastics describes small pieces of plastic found in the ocean, commonly defined as < 5mm in diameter. Two different types of microplastics, primary (industrial or production) and secondary (consumption), are considered. Primary microplastics are virgin plastic pellets that reach the marine environment due to accidental releases and poor handling. Secondary microplastics are produced as a result of fragmentation from large items. Microplastics often contain chemicals added during manufacture (the secondary ones) and can sorb and concentrate pollutants from the surrounding seawater. Plastic pellet type abundance is similar to that of the production of plastics. This means that the majority of the beached plastic pellets are made of polyethylene. The low-density ones that are also the most abundant float on the sea surface, and end up on beaches. High-density microplastics have been found and identified in sediments. While they float on the sea surface, they interact with the surface layer of the sea that is enriched in pollutants. Near ports the sea surface is polluted with organic pollutants that are sorbed by plastic pellets. This results in different concentrations of chemicals on the various plastic pellets depending on the sampling point. There are several studies that demonstrate the variability of pollutant concentrations in the globe as well as in smaller scales (e.g. Greek Seas). Also, a high variability of the type of chemicals found on plastic pellets exist including polyaromatic hydrocarbons, polychlorinated diphenyls, chlorinated pesticides, perfluoroalkyl substances, etc. So far, plastic pellets have been used as a means of monitoring organic pollutants. However, since many organisms interact with microplastics, there is an increasing concern for the fate of the sorbed chemicals once the microplastics are inside the body of marine organisms. Currently, there is emerging evidence of transfer of chemicals from ingested plastics into tissues. In addition to the mechanical break down, when in the marine environment due to various processes and especially UV light, plastics degrade, and their surface is transformed. Beached and marine plastics have been found to interact not only with organic chemicals but also with metals and especially, with different types of microbes. This causes plastics to be denser and to sink to the bottom of the sea and thus, to interact with more chemicals. Microplastics are dynamic materials that are transported, transformed, and sources of pollutants and microbes to the coastal and marine environment.

Do fin whales (*Balaenoptera physalus)* feed in microplastic impacted areas in the Pelagos Sanctuary?

*Maria Cristina Fossi[1] *,*
Cristna Panti[1], Letizia Marsili[1], Daniele Coppola[1], Matteo Baini[1], Matteo Giannetti[1,2], Ilaria Caliani[1], Maria Grazia Finoia[4], Simone Panigada[5], Fabrizio Rubegni[6], Roberta Minutoli[3]

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The emerging issue of microplastics (plastic fragments smaller than 5 mm) in marine environment is recently raising increasing attention. The impacts of microplastics on baleen whales, which potentially undergo to the ingestion of micro-litter by filtrating feeding activity, are largely unknown (1,2). Here we present the case study of the Mediterranean fin whale (*Balaenoptera physalus)*, exploring the toxicological effects of microplastics on mysticetes investigating two sub-groups foraging in two areas of the Pelagos Sanctuary (SPAMI-Mediterranean Sea), Ligurian and Sardinian Sea and in three period of sampling: July (J), August (A) and September (S). The work is implemented through three steps: 1) collection/count of microplastics in Pelagos Sanctuary; 2) detection of phthalates in superficial neustonic/planktonic samples; 3) detection of phthalates, organochlorines and biomarkers responses (CYP1A1, CYP2B, lipid peroxidation) in skin biopsies of fin whales collected in the Pelagos Sanctuary (n=30). High occurrence of plastic particles have been detected in superficial neustonic/planktonic samples collected in the Pelagos Sanctuary areas investigated (mean value 0.62 items/m^3) with high concentration of phthalates (DEHP and MEHP), used as tracers of plastic derivates. The GIS data elaboration from the study areas, highlights how microplastics appear mainly distributed in the pelagic environment instead of in neritic areas, suggesting the presence of transient convergence areas during the summer. Moreover our results show, for the first time, an overlap between pelagic areas with high microplastic density and the whales feeding grounds, suggesting the exposure to microplastics threat during their foraging period, as confirmed by the increasing toxicological stress from July to September. Our study was successful in identifying temporal and regional ecotoxicological differences supporting the hypothesis of fin whale as wide-scale indicator of the impact of microplastics and related contaminants in marine environments.

Acknowledgements
This project was supported by the Italian Ministry of Environment, Territory and Sea (prot n.39752/III-17). We would like to thank the Italian Navy for support in microplastic sampling in the Ligurian Sea.

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Plastics & microplastics pollution in the marine environment: Challenges and solutions

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Microplastics pollution is an emerging threat, widespread and ubiquitous throughout the marine environment.[1] Microplastics are part of the overall plastic marine litter problem, generated by our plastic-addicted societies and the mismanagement of plastics and plastic waste. Whilst the societal benefits of plastics are undoubtedly remarkable and far-reaching, the downsides of our 'plastic age', which include environmental and socio-economic impacts, need to be urgently tackled.[2,3]

Some of the key challenges and hindrances that hamper down the effective closure of the plastic life cycle related loopholes that lead to an increasing introduction of plastics and microplastics –from primary and secondary sources- into the coastal and marine environment include: the limited and fragmented understanding of the problem due to the lack of accurate, coherent, reliable and comparable scientific data; the misconceptions and misunderstandings related to possible solutions, i.e. the case of bio-degradable or bio-based plastics [4] or the not viable option of microplastics removal from habitats [5]; the reluctance of countries to commit themselves to reaching ambitious targets via comprehensive programmes of measures; the fact that marine litter is often considered as someone else's problem inhibiting coordinated, diversified and multi-level actions; the poor exploitation of the full potential that marine litter initiatives and projects render for capitalization, replication and collective learning.

Over the past decade, increased scientific interest has produced an expanding knowledge base for plastics and microplastics pollution and with knowledge comes greater responsibility.[1,6] Perhaps it is too utopian to dream of zero waste societies, but from a very pragmatic point of view we can prevent marine litter, including plastics and microplastics reaching our coasts and seas if we all take our share of responsibility and embrace a full systemic change towards a circular economy. After decades of 'training' ourselves and our societies on consuming more and throwing things away, we need to find innovative ways to do more with less; ways to move away from single-use and superfluous products; ways that will allow us to move up the waste hierarchy.

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ORAL CONTRIBUTIONS

Sea surface microplastic quantities and distribution in the Slovenian part of the Trieste bay

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Results presented in this study were one of the first attempts to evaluate floating microplastic pollution of the Slovenian sea. Between December 2012 and August 2014 four sampling events took place. 17 sea surface samples were taken with epineuston net of 300 µm mesh size according to published methodologies in the entire Slovenian sea area. Analysis of the samples was done by visual identification of microplastic particles (300 µm – 5 mm) with the help of binocular magnifier (under 40x magnification). The average number of microplastic particles was 471,903 per square kilometer. The highest concentration observed was with 3,098,765 particles per square kilometer and the lowest with 13,888 particles per square kilometer. The chemical analysis of 14 % of all particles found (6086) showed polyethylene to be the predominant type of plastic with over 80%. The amount of microplastic particles found in this research is higher than in any other similar studies conducted on sea-surface samples and in lakes.

Marine plastic debris in the north-eastern Adriatic

M. Smodlaka Tanković, V. Stinga Perusco, J. Godrijan, D. Marić Pfannkuchen, M. Pfannkuchen*

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In our society plastic is one of the go to materials and it has a major commercial, industrial, medicinal and municipal applications. Plastic production has increased dramatically from the 1950s, and following this plastic waste has accumulated in the environment at an increasing rate. There it is subjected to wind and river-driven transport, ultimately reaching the marine environment. Data on amounts of plastic litter in the Mediterranean is generally limited and shows great variations. All sources report high amounts of plastics, in both floating and beach litter, as well as in litter on the sea floor. Major concerns have been raised about microplastics and its impact on marine organisms. Due to its small size, microplastics may be ingested by the fauna, with uncertain consequences for the health of the organism.

The objectives of this study were to (i) assess the presence of plastics in marine litter, (ii) identify presence of microplastics in the water column (iii) examine the bioaccumulation of microplastics, via a proxy - common pandora (*Pagellus erythrinus*), a popular food fish in Mediterranean countries.

A frequented beach was surveyed in April, June and November 2014, following the methodology suggested through the Marine Framework Directive. Marine litter was classified into 8 main categories: plastic, rubber, cloth, paper, wood, metal, glass and other. Major contribution was from cigarette buds and filters, plastic pieces 2.5 cm > < 50cm, polystyrene pieces 0 - 2.5 cm and other plastic/polystyrene items. The presence of microplastics in the water column was investigated on a transect across the northern Adriatic. Ingestion of microplastics was investigated on common pandora by stomach content analysis. This is the first investigation of this kind in the north-eastern Adriatic Sea, and the collected data justify a growing concern about microplastics and its impact on the marine environment.

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Sea surface microplastics distribution on Emilia-Romagna coast (Northwestern Adriatic sea - Italy): DeFishGear project preliminary results

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Northern Adriatic Sea and the coastal stretch in the Emilia-Romagna region of Italy is under the direct influence of the inputs of the river Po [1]. The influence and the effect of the river inputs of the basin of the Po valley on the coastal area are evident from the salinity values, which noticeably fall along the coastal area in comparison with the open sea. Here we report the results of three transects through the Northwest Adriatic sea (13.77 total nautical miles) carried out in October-December 2014. Microplastics samples were collected at 11 sites using a manta trawl lined with a 333 µn mesh.
The average abundance and mass was 63175 particles km^{-2} and 27.29 g km^{-2} respectively. The highest value of 128841 particles km^{-2} occurring in front of Cesenatico (3 km off shore). Only in front of Porto Garibaldi the maximum value was detected in the offshore site (20 km), while in the other transects this are not observed.
The Fragments (G103, G104, G105, G106) represent the micro litter category more frequently observed (80%). Concerning the plastic items transparency, 68% of the microplastics analyzed were opaque.

Acknowledgements. The authors thank the crew of the oceanographic M/S Daphne II, Mr Dino Pagan and Mr Marco Papperini.

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Neustonic microplastics in the Southern Adriatic Sea

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Neustonic micro-plastic abundance and polymeric composition were determined after a cruise conducted in the Southern Adriatic Sea between May 9th and 17th 2013. Plankton samples were collected using a Neuston net (200 µm mesh size) which sampled the first 50 cm of the sea surface at a speed of ~2 kts for 5-6 minutes. Samples were then stored in ethanol 70% and in the laboratory micro-plastics were hand-picked using a dissecting stereomicroscope, counted, weighed and split into 7 different size classes. On a subset of collected particles (> 0.7mm) FT-IR analyses were performed to characterize the polymeric composition of the items. All 29 surface tows contained plastic particles of various typologies (e.g. filaments, fragments, thin plastic films), colours and sizes. A total of 5940 plastic particles were collected during the survey, the vast majority of which were hard plastic fragments (78.5%) or synthetic fibers and filaments (19.2%). Most particles were white (27.8%), transparent (22.5%) or black/grey (21.4%). 98.2% of all the particles were < 5 mm and plastic abundance markedly increased with decreasing size (i.e. 52.8% of all the particles were smaller than 0.5 mm), indicating very high fragmentation rates. Overall, an average concentration of 1.05 ± 1.13 particles/m2 and 442.88 ± 1145.96 g/km^2 was observed throughout the study area, with micro-plastic densities ranging from 0.10 particles/m2 to a maximum of 4.86 particles/m2. FT-IR analyses indicated polyethylene as the predominant polymer (41%), followed by polyester and paint (12%), polypropylene (10%), polystyrene and polyimide (5%), polyammide (3%), paraffin (4%) and 1% bioplastic (i.e. polycaprolactone). In addition, 7% of the items were characterized as non-plastic materials (i.e. minerals, cellulose and cotton fabric), suggesting a potential bias when visually sorting for micro-plastics. On the whole, very high levels of plastic pollution have been found in our study area. Despite any clear geographical pattern in plastic distribution was identified, the conspicuous spatial heterogeneity in plastic abundances and polymeric compositions seem to confirm the existence of multiple pollution sources insisting on the Adriatic Sea.

Occurrence and distribution of floating microplastics in the North Adriatic Sea: preliminary results.

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A preliminary monitoring survey of floating microplastics (MPs) in the North-Adriatic Sea was carried out, as part of activities for the "Protocols MATTM - Regions, Marine Strategy implementation - LD. 190/2010". Data were collected during two sampling campaigns (winter and spring) conducted in 2014 along two transect, one in front of the Lagoon of Venice (Pellestrina Island – North) and another in front of the mouth of the Po River (Po della Pila – South). Microplastics (MPs) samples, collected using a Manta trawl, were processed following MATTM operating protocols and then imaged and quantified by stereo-microscopy; a percentage of each sample was analyzed by µFT-IR (Micro Fourier Transform Infrared Spectroscopy) to determine the chemical composition of collected MPs. Results showed a MPs average concentration of 12,19±20,13 particles m^{-3} (winter sampling) and 1,01±1,02 particles m^{-3} (spring sampling). µFT-IR analysis showed that the most widely diffuse polymers were polyethylene, polypropylene, ethylene vinyl acetate and polystyrene (91,2% of total analyzed MPs). Pelagic habitats were also investigated, measuring hydro-chemical parameters, the abundance of phytoplankton (average conc. 5809000 cell. dm^{-3} – winter and 2876520 cell. dm^{-3} – spring) , zooplankton (average conc. 4522 ind. m^{-3} - winter and 3693 ind. m^{-3} - spring). A comparison between MPs and zooplankton abundance was carried out in order to evaluate the availability of MPs which could be potentially ingested from plankton-feeders organisms. MPs: zooplankton average ratio for both transects resulted 0,01 in winter and 0,001 in spring. Presented data are courtesy of Italian Ministry for the Environment, Land and Sea and the Veneto Region.

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Water flow modelling through plankton net

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The majority of microplastics monitoring studies in marine and riverine environments are conducted using plankton trawls. The most common type used is a manta trawl with a fixed geometry of the water inlet opening. Recommendations for trawling speed as well as monitoring of net clogging are empirical and do not have any theoretical background.

Preliminary results of a numerical modeling of water flow through manta trawl are presented. Water flow rate through the trawl is observed with respect to changing flow resistance and trawl velocity. Flow resistance simulates the effect of clogging due to accumulation of debris that should decrease water flow rate through the trawl. The volume of water sampled is generally considered to be the product of trawl's inlet cross-section area, relative trawl-to-water velocity and sampling time. We define the relative flow rate as the ratio of actual water volume flow rate at the trawl inlet and the product of trawl velocity and cross-section area of the trawl's inlet (theoretical flow rate). Relative flow resistance is a multiplication factor of flow resistance coefficients that define flow resistance through the net. The study was conducted in two steps: first a numerical model of a small section of 300 μm net was set up to obtain reference flow resistance parameters, where trawl velocity was varied from 0,25 to 2 m/s. The results were used to set the reference flow resistance in the subsequent 2D numerical model on a full trawl geometry in which varying trawl velocity and flow resistance is studied with respect to relative flow rate through the trawl.

Results are presented as relative volume flow rate through the trawl for different trawl velocities and flow resistances. Additionally pressure distributions and velocity vectors are presented for better understanding of flow phenomena through and around the trawl. It is shown that higher flow resistance of the trawl's net (more debris accumulated on the net) results in significant flow deflection and decreased volume of sampled water. The actual sampled volume is thus considerably smaller than theoretical even with a 'clean' net. Accumulation of debris on the net can further reduce the sampled volume by up to 20 %. An interesting result of the study is finding that higher trawl velocity produces a higher pressure difference on the net, which forces slightly larger amounts of water through the trawl and results in slightly increased volume of sampled water.

Our study suggests that reported concentrations of microplastics per volume of water might need to be corrected to take into account deviations due to trawling velocity and clogging of the net.

Modeling the transport of floating plastic debris in the Adriatic Sea

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Great threat of marine plastic pollution to human health and negative economic effects on coastal communities stimulate monitoring [1] and modeling [2] the transport of FPD (Floating Plastic Debris) in the Mediterranean Sea.

In the present work, the simulations of FPD distribution are carried out by a Lagrangian model MEDSLIK-II [3] combined with the operational service from the Adriatic Forecasting System [4] in order (1) to identify areas of maximum accumulation of FPD, (2) to quantify which shores are the most prone to the FPD deposit; (3) to understand the donor-acceptor interconnections among the Adriatic sub-regions.

Evolution of FPD concentration is calculated by the *methodology developed for* satellite-tracked Lagrangian drifters [5]. Majority of FPD is supposed to originate from 68 biggest Adriatic rivers, 45 cities with a *population* of more than 20 000, and 76 most congested shipping lanes.

Analysis of statistics obtained by the simulations of multiple FPD released during 2010-2014 has shown that, in contrast to the global ocean [5], the Adriatic Sea is a highly dissipative system; the main FPD sink is the coastline; the mean FPD half-life time is about 45 days. The average FPD path is about 605 km in the Adriatic.

On long-term time-mean scales, distinctive "hot spots" are found in (1) the southern tip of Corfu Island (Greece), (2) Durres (Albania), (3) **Dubrovnik archipelago including** *Mljet Island (Croatia), and (4)* the *Marche region and Gargano Peninsula (Italy). These areas can be recommended for thorough marine litter monitoring and the most frequent cleanup.*

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Seabed mapping with remote sensing

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In 2014, European Commission stated that marine based economy is held back by a lack of information about the sea, the seabed and the life it supports [1]. Increased knowledge of our seas will promote growth; in the case of seafloor, knowledge about habitats for fish nurseries, commercial shellfish as well as seabed integrity which supports biodiversity and functioning of the ecosystem. In this scope, the impact of plastic waste on the environment has an important role in its health and functioning [2]. Many of this impacts are not clear yet and must be studied in more detail.

By remote sensing, plastic waste and its impact on the surrounding environment can be monitored frequently and in large areas. The resulting prompt and relevant information is important for several blue economy sectors (tourism, coastal management, etc.) as well as scientific studies.

Harpha Sea (http://www.harphasea.com) has been active in the field of surveying and geoinformatics for over 20 years, with development of bathymetric models and underwater archaeology surveys, terrestrial surveys, LIDAR, aerial recording, GIS services (field surveys, GIS maintenance, spatial analysis, map making), and 3D modelling. We are specialised in coastal water surveys with research boats, automatic research vessels and drones, using top quality survey instruments (multibeam and sidescan echosounders, subbottom profilers, underwater cameras, etc.) as well as a system for data elaboration and visualisation in 2D or 3D virtual environments (Fig. 1).

Our system for geo-referenced habitat classification from boats and drones/planes enables monitoring of large areas, providing detailed information on seabed habitats, including bathymetry. The data are processed with commercial and optimized software, enabling cost-effective and relatively rapid seabed mapping as well as monitoring.

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An integrated approach for the classification of Plastic Large Micro Plastics (LMP) in beach sand

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Contamination of coastal areas by plastic litter represents an emerging issue of concern because of the increasing use of polymeric disposable items, and there is a lack of information about sources, sinks, and pathways of microplastics into these related compartments. Microplastics embedded in beach sand result from combined contributions of various contamination and degradation processes from terrestrial and marine sources. An integrated approach, involving size, shape and color analysis, as well as chemical characterization, for the classification of plastic particles in the 1-5 mm size range, the so-called Large Micro Plastics (LMP), was developed and is here presented. Such size range was moreover selected as a potential subject of study in order to understand the composition and genesis of smaller microplastics (<1 mm) and nanoplastics (<300 μm). Moreover, LMP can be useful markers about ongoing processes of degradation and fragmentation of larger plastic debris. An optimized high resolution photographic procedure coupled with image analysis was developed in order to determine bidimensional size, shape and true color of collected LMP. An instrumental, automated tridimensional particle size and shape analysis was applied for the physical characterization of extracted LMP. The use of Near Infrared Spectroscopy (NIR) and Raman Spectroscopy coupled with Optical Microscopy (OM-RS) allowed the chemical identification of extracted particles. The main aim of the this work was to develop an integrated approach which could allow to correctly define physical information, such as tridimensional size, shape and color of LMP, which are still underestimated [1]. Such approach meets the need to fill the gap between awareness of the LMP presence in marine litter and the evaluation of their transformation pathways from sources to potential environmental targets. The developed approach allowed a detailed physical and chemical characterization of LMP collected from August 2014 to February 2015 on the Lido Island (Venice, Italy), in a coastal protected area never investigated before.

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Techniques usefull for characterization of microplastics

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Identification of microplastic items collected during monitoring activities is a challenge due to 1) small particle size, 2) brittleness of samples and 3) high rate of weathering of the material due to the mechanical and photo degradation as well as hydrolysis. Sample manipulation is challenging and time consuming. One of the aims of the DeFishGear project was to develop a fast and efficient compilation of methods for identification of microplastic particles.

At NIC we employ 4 methods for identification of microplastics: 1) optical microscopy, 2) ATR-FTIR spectroscopy, 3) NIR spectroscopy and 4) IR microscopy.

Optical microscopy

Optical microscopy is used for size, shape and color determination of microplastic particles. A Leica DMS 1000 microsystem with an automatic stage is used. No special sample preparation is needed. Obtained scaled digital image are further processed to determine the size, color and shape of the samples. Sample manipulation is the major bottleneck.

ATR-FTIR spectroscopy

ATR-FTIR spectroscopy is the most common used characterization technique for MP. Material composition and in some cases (PE, PP) also the degradation rate with the calculation of carbonyl index may be determined. Time consuming manual manipulation of samples and particle size limitations are key issues.

NIR spectroscopy

A modified near infra-red (NIR) spectrometer sIRoGran (GUT GmbH, Stuttgart, Germany) NIR, automatic scanning facility and spectral database was adapted for fast characterization of microplastics. Characterization efficiency for particles above 1 mm is above 80 %. Characterization of expanded plastics and dark plastic particles is not possible.

IR microscopy

Bruker LUMOS FTIR microscope was adopted for identification of microplastic particles smaller than 1 mm. The microscope is most often used in ATR mode although transmission and reflectance modes are possible. Sample preparation is not complex (placing particles on a glass slides). Microscope can characterize samples in micrometer scale.

The 4 techniques are complementary and offer a plastics characterization solution suitable for most encountered cases.

Characterization of Plastic Large Micro Particles (LMP) from the beach sand of Lido Island (Venice, Italy)

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The presence of Large Micro Particles (LMP) in coastal beach sediment is extremely variable and affected by seasonality and overtime events. A robust discussion on such a topic for a given site would require a representative amount of collected samples (and corresponding data) in space and time. In general the proven chemical identification of microplastic debris is a relatively easy task and it's usually obtained almost effortless with user friendly spectroscopic analyzers such as Near Infra Red – NIR and Infrared (FT-IR). The real challenge is to perform a representative sampling and a qualified physical characterization in terms of size and shape.

A summary of the very first sampling and analysis of LMP in the sediment of the Alberoni beach of the Lido island (Venice, Italy), in a coastal protected area never investigated before, is here presented with a critical discussion of the on-the-field and subsequent in-the-lab hitches encountered.

A seasonal comparison of two sampling campaigns, October and December 2014, have been performed in terms of LMP abundance, chemical composition, color and item category mostly according to European guidelines [1]. Chemical analysis was performed with through NIR spectroscopy and Raman coupled to microscopy: a comparison of the two techniques in terms of size capability (i.e. macro vs. micro) is presented along with the corresponding microphotographs of extracted plastic samples and the role of the adsorbed organic pollutants on their surface.

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Influence of Plastics Weathering on Adsorption of Pollutants

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Microplastics (MP) can be found in virtually all marine and other aqueous environments. Due to the large specific surface and relative hydrophobicity MP adsorb organic pollutants (POPs) such as PCBs, PAHs etc. [1]. The danger is that upon ingestion by organisms adsorbed pollutants may enter the food chain. It is indicated that environmental ageing increases the tendency of MP to adsorb pollutants [2].

In our study we performed controlled laboratory ageing of polyethylene terephthalate (PET), polyethylene (PE) and oxidatively degradable PE (oxo-PE) by exposure to UV light, heat and water. Treated materials were exposed to two model pollutants: triclosan (5-choro-2-(2,4-dichorophenoxy)phenol, TC) and methyl parabene (methyl 4-hydroxybenzoate, MP). The adsorbed model pollutants were quantitatively determined after desorption.

Results showed that PET weathering significantly increased TC adsorption. PE oxidation as indicated by the carbonyl index (CI) resulted in increased adsorption: TC adsorbed with a simple, positive correlation to CI, whereas PB adsorption was dependent on TC presence, indicating an important synergy. This showed, for the first time, that adsorption of pollutants may be dependent on the pollutant mix MP are exposed to, illustrating the complex, multifaceted nature of the process. oxo-PE showed higher adsorption compared to PE.

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Biofouling community and degradation rate of bioplastic in the marine environment

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The impact of bioplastic on the marine environment is not known well, since many different types of bioplastic exist and cannot be treated equally. The European standard EN13432 determine by what time and to what extent a bioplastic must degrade under controlled conditions in an industrial composting system. The biodegradation of polyethylene compounds in the presence of different microbes has been studied in landfills [1], but not in the marine environment.

A series of laboratory and *in situ* experiments was set up to evaluate the rate of degradation of one type of bioplastic in the marine environment and to determine dominant microorganisms in biofilms. Three different types of experiments were established to follow the succession in the degradation of the bioplastic material: i) in microcosms with seawater and different sized fractions of the microbial community with incubation under controlled laboratory conditions, ii) in an aquarium filled with different macro-organisms (potential consumers) and iii) in the coastal sea environment (in the water column at approximately 3 m depth) incubated under natural environmental conditions with ambient assemblages of organisms. Microbial community dynamics were followed using microscopic and molecular techniques to determine the microbial succession rates and community composition. In order to determine changes in the microbial community structure during the degradation process, the bacterial community DNA was extracted and 16S rRNA bacterial gene clone libraries were constructed.

Our results on community composition and microscope observations suggest a fast biofouling and emphasize the importance of complex living communities, composed of bacteria and eukaryotes, for efficient bioplastic degradation. The preliminary results show that the degradation process was on a time scale from weeks to months. In the aquarium experiment a very fast and complete degradation (less than 14 days) was observed, even when the material was protected by a net, in order to prevent the consumption by fish and crabs. In the coastal sea the degradation of the bioplastic material took several months. The FTIR and elemental analyses showed the degradation of bioplastic. The analyses of 16S rRNA gene clone libraries revealed great differences in bacterial community structure between ambient seawater and microcosm population.

Acknowledgements
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Microplastics in sediments and fish of the Gulf of Trieste

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Production of plastics has increased extensively over the last decade; from 204 Mtons in 2002 to 299 Mtons in 2013. As a consequence, a fast increase in the quantity of plastic materials released in the natural environment was observed. These materials are very resistant and can persist in their original form for many decades. Microplastics are also very widespread in the marine environment and they most probably result from the degradation of larger particles. The investigated area is a part of the northern Adriatic Sea where the coast is highly urbanized and industrialized. The marine environment along the coast is affected by pollution from different sources, namely industrial waste waters, waters from the sewage treatment plants, intensive maritime traffic, tourism, freshwater inputs by rivers and agriculture. The aim of the present study was to examine the presence of microplastics in the marine environment in the Gulf of Trieste. For this purpose, the content of microplastic particles in surficial sediment samples and in stomachs of four commercially important fish species was determined.

Plastic particles from the sediment were separated in a saturated NaCl solution. After sieving and washing the obtained sample was examined under a light and fluorescence microscope. Selected particles were collected for FTIR characterization. The size of particles varied from 200 to 600 µm, while some particles exceeded 3 mm. Different types of particles were observed. The major part was composed of fibres of different colours, but some fragments and films were also detected. According to the FTIR spectra, the composition was very heterogeneous, but polyethylene and polypropylene were the most abundant materials. Some of the fibres, even coloured, were of polysaccharidic composition, most probably cotton. The surface of these fibres was in general not smooth. Therefore we assumed, that as a factor to discriminate between plastic particles and natural materials, the smoothness of the particles may be used. Many transparent fibres were also detected, but their composition still has to be determined. The concentration of particles in the dry sediment was between 3 and 87 particles per 100 g. In general, the offshore areas were less contaminated than the inshore areas. The distribution of particles in inshore areas was not uniform, and was showing higher content in more closed areas, e.g. marinas and small harbours.

Fish caught in the Fisheries Reserve were analysed for their stomach content and several plastic pieces were found in 15% of the stomachs. Microplastic particles found were fibres, pieces of spherical shape, and wires, all determined as polyamide or polyethylene using FTIR and most ranging bellow 500 µm size. Microplastics were mostly found in the stomachs of *Dicentrarchus labrax* and *Sparus aurata* (40% of stomachs), rarely in *Pagellus erythrinus*, but never in the stomachs of *Liza aurata*, although this species is the most abundant in the area. As the microplastics were not found in species *Lisa aurata* which feeds on the sediment, we hypothesise that microplastics are mostly ingested in the water column, where they are expected to be even more abundant than in the sediment, or possibly the predator fish (*Dicentrarchus labrax*, *Sparus aurata*) consume prey which is already containing microplastics. Most microplastics are assumed to be floating in the upper few meters of the water column. This is also supported by the result of this study that more microplastics were found in sediments close to the shore where it is being deposited in shallow waters.

Ecotoxicological risk of microplastics in marine organisms.

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Microplastics represent a growing environmental concern for the oceans due to their potential capability to adsorb different classes of pollutants, thus representing a still unexplored source of exposure for aquatic organisms. Although several organisms can ingest microplastics with potentially adverse effects on their health, a clear evidence of their presence in wild organisms is still lacking.

In this study, different micro-polymers were shown to efficiently bind polycyclic aromatic hydrocarbons with a time and dose-dependent trend, and transfer these chemicals to filter feeding mussels *Mytilus galloprovincialis*. Tissue localization of microplastics revealed their presence in haemolymph, gills and especially digestive tissues where a marked accumulation of PAHs was also observed. A wide spectrum of molecular, biochemical and cellular effects was assessed, including the analysis of transcriptomic profile, immunological responses, lysosomal destabilization, peroxisomal proliferation, antioxidant and neurotoxic effects, onset of genotoxicity. The use of this multidisciplinary approach demonstrated significant alterations in gene expression and in functional responses at cellular level in mussels exposed to both virgin and contaminated microplastics.

The presence of microplastics was also evaluated in wild commercial fish from the Adriatic sea. After a validation of a new extraction protocol with an extraction yield higher than 90%, microplastic items were observed in 30% of analyzed specimens, with higher occurrence in benthic compared to pelagic species. Polyethylene was the predominant polymer (65%) in the gastrointestinal tract of fish.

The overall results showed clear ecotoxicological effects of microplastics and their occurrence in biota compartment, underlying the potential risk of these emerging compounds for the marine environment.

LIFE+ MERMAIDS PROJECT: Mitigation of Microplastics Release Caused by Textile Washing Processes

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Among the multiple human pressures on aquatic ecosystems, the accumulation of plastic debris is one of the most obvious but least studied. While plastics generate remarkable societal benefits, there are downsides to our ‘plastic age’. Durability, unsustainable use, and inappropriate waste management cause an extensive accumulation of plastics in natural habitats. 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. The issue of (micro) plastics (MP) connects to several European water policies. The European Marine Strategy Framework Directive (MSFD, 2008/56/EC) addresses the issue of marine litter, including plastics. In particular, decision 2010/477/EU describes the “Criteria to be used by the Member States to assess the extent to which good environmental status is being achieved”. Considering the lack of knowledge about the microplastics and the few methods to characterize them and diminish their environmental impact, a consortium of research institutes (CNR IPCB, CNR ISMAC, LEITAT), industry (Polysistec) and a foundation that fights against the increasing plastic contamination of our seas (Plastic Soup Foundation), have presented the MERMAIDS project that started on July 1st 2014 with the financial contribution of EU LIFE+ Environment Program. In particular the project aims to reduce the release of microplastics, deriving from the washing processes of clothes, by means of textile treatments or new detergent additives. Indeed recent studies [1] highlighted that a major source of microplastics derives from textiles substrates containing synthetic fibres such as polyester, polypropylene, polyamide, polyacrylic. In this presentation the project objectives and the results already obtained are explained.

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Petroleum-based plastics biodegradation by anaerobic marine consortia

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About 299 million tons of plastics were produced worldwide in 2013 and EU contributed with about 57 Mtonne. Most of plastic demand and production is for polyethylene (PE), polypropylene (PP), polyvinyl chloride (PVC) and polystyrene (PS). Due to their stability and chemical and biological inertness, these plastics have been accumulating in the environment leading to pollution and health concerns. Specifically, marine litter is actually a big issue both for the physical impact as well as the toxic effect on marine biodiversity. Environmental biotechnology can offer promising tools/strategies for mitigating the environmental impact associated with plastic waste. To date there are very few reports on the biodegradation of petroleum-derived plastics and especially on those entering the sea.

The aim of this work was to assess the ability of anaerobic marine microbial communities to degrade PE, PP, PVC and PS. This was performed by first enriching anaerobic microbial communities in three different media targeting nitrate-reducing bacteria, sulphate-reducing bacteria and methanogenic archaea, and further evaluating their biodegradative capability. For this purpose, enriched consortia were subcultured in the presence of target plastics films as major carbon source. Microbial growth was monitored by gas and ion chromatography and biofilm formation on plastic films. Plastics biodegradation was evaluated by thermogravimetrical analysis (TGA), gel permeation chromatography (GPC) and determination of gravimetric weight loss percentage.

After 7 months of incubation, little growth was observed in all enriched consortia. Adhered protein concentration (of up to 200 mg/g of plastic polymer) confirmed the formation of biofilms on plastics surface. A lower thermal stability of PP, PE and PVC polymers incubated with eight different microcosms was detected by TGA indicating the occurrence of biodegradation. Moreover, GPC analyses performed on PVC films revealed up to 7% decrease in mean molecular weight (M_n) compared to abiotic control. A second sampling point was considered for the eight most promising consortia after 10 months of incubation. TGA showed different thermal stability profiles compared to abiotic controls. Furthermore, gravimetric weight loss percentage of up to 12% and a decrease of up to 11% in M_n were observed in the case of PVC films.

In conclusion, in this work we enriched marine anaerobic consortia with the ability to degrade petroleum-derived plastics. Microbial communities' analysis is in progress to identify the microorganisms involved.

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POSTERS

Microplastic pollution on the Slovenian sea and coast – the first results of the DeFishGear project

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The marine microplastic pollution results in the Slovenian sea was first researched through the collaboration between Institute for Water of the Republic of Slovenia and Univeristy of Nova Gorica and is now continued by the DeFishGear project. In 2014 one sea surface (including riverine outflow) and one beach sampling were done. The sampling of sea surface was made by using manta net with 308 µm pore size in August 2014, where 4 samples on the sea were sampled and 2 samples near the river Dragonja outflow were sampled. Beach sediment was sampled in September 2014 when samples for large microplastic particles (LMP) and small microplastic particles (SMP) were collected on the sandy Lazaret beach (near Debeli rtič). The microplastic particles were separated from all samples by the use of stereomicroscopes and analysis of particle size and number was done by the image analysis program (Axio Vision, Carl Zeiss). Among sea surface samples and SMP beach sediment samples fibers were the most common microplastic particles, followed by fragments. Granules, foams and pellets are very rare in both kinds of samples. While the most common microparticles among LMP sediment samples are non-plastic from category other (cheramic, glass, metals). From the first DeFishGear results we can conclude that fibers are the most common microplastic category and also the category for which material characterisation is most difficult and is not possible only by performing visual observation by the stereomicroscope. On the other side the chemical analysis of material by FT-IR microscope takes a lot of time, therefore the chemical analysis of all these particles is impossible. For this reason the development of new statistical methodology is very important.

Determination of POPs adsorbed on plastic pellets

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Plastic pellets are industrial raw material, usually in the shape of small cylindrical granules with a diameter of a few mm. They can be unintentionally released to the environment during manufacturing as well as transport and can eventually reach the marine compartment. Due to their environmental persistence, they are widely distributed in the oceans and on beaches [1]. Several studies have revealed the presence of organic contaminants at concentrations from sub ng g^{-1} to mg g^{-1} on/into plastic pellets found in coastal environment worldwide [1, 2, 3]. These compounds are either plastic additives (e.g. PBDEs) or hydrophobic chemicals (e.g. PCBs, organochlorine pesticides) which adsorb from the surrounding seawater. Among these chemicals, some are recognized as POPs (Persistent Organic Pollutants) because of their persistence, bioaccumulation and potential environmental and health adverse effects. Thus, in order to better assess the impact of plastic pellets in coastal environmental, it is necessary to determine the level of associated organic pollutants.

The present study was carried out in the frame of DeFishGear project, which focuses on marine litter and microplastics issues in Adriatic region. This investigation aimed at developing an experimental protocol allowing the quantification of POPs: 11 organochlorine pesticides and 25 PCB congeners. The plastic pellets, sampled on beaches from the Adriatic Sea, were first sorted by colour. The POPs were extracted from the plastic matrix (0.5-1 g) in a pressurized fluid extractor (50°C, 100 bar). Prior to evaporation, the extract was cleaned on Florisil sorbent in a solid-phase extractor (SPE). The concentrated extract were qualitatively analysed on GC-MS. The quantification of the POPs was performed on GC-ECD. The preliminary investigations made on Greek pellets showed the presence of organochlorine pesticides such as DDEs and HCHs.

Acknowledgements:
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Plastic microfiber emissions from domestic washing

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Textiles and garments made from synthetic fibers have become an integral part of modern life. Their artificial structures can offer astounding properties such as bulletproofing, water resistance, insulation, etc. Their omnipresence however is also a potential source of microplastic particles entering the environment [1].

In our study we quantified the emissions of plastic particles from washing and drying a fleece blanket made of microfibers in domestic washing and drying machines. The blanket used was made of polyethylene terephthalate – a non-degradable polyester, and was in the form of fibers with a 12 µm diameter. A short, 15 minute, low temperature (30 °C) washing cycle was used. Ten successive washings were carried out each followed by tumble-drying. Three series of experiments were carried out: without additives, with detergent, and with detergent and softener. Microfiber emissions were monitored by means of an external filtering setup using a stainless steel mesh with 200 µm openings. The collected fibers were dried and weighed.

Results show that each washing or drying run causes microfiber emissions. Starting with a new blanket first washing/drying runs give the highest emissions, which then decrease in successive runs and eventually reach a stable constant level. Stable long-term emissions were reached in washing whereas in drying ten runs were still insufficient to reach a stable level of emissions. Our experiment shows that 0,001 wt.% of loose fibers is emitted into wastewater during each washing. The built-in filter in the washing machine is not an obstacle for microfibers. The amount of microfibers that pass the 200 um filter is very low showing that one filtering step could significantly reduce microfiber emissions from washing.

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Microplastic occurrence and diet composition in red mullet, *Mullus surmuletus*, in eastern Adriatic sea

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Microplastic fish ingestion has not yet been subject of scientific investigations in the Croatian part of the Adriatic Sea. Red mullet, *Mullus surmuletus,* has been chosen as a study organism as it is opportunistic feeder and represents benthic important commercial species. Diet composition and microplastic occurrence in red mullet stomachs were investigated. Stomach contents of red mullets (6.40 – 32.90 cm TL; N=203) from the central part of Eastern Adriatic were sampled on a monthly basis from December 2011 to October 2012. Commonly accepted procedures were followed during the diet composition inspection and standard keys were used for food items determination. Animal species were preferential prey with predomination of crustaceans (~30%). During the visual inspection of stomach contents, the microplastic particles were separated and counted. From the 203 examined specimens, microplastic items were found in 63 stomachs. Ingested microplastics were mostly filaments in different colors. Approximately 31% of processed fish ingested plastic, averaging 8.5 pieces per fish. Our results indicated widespread occurrence and ingestion of microplastic particles in red mullet.

Abundance and characterization of floating microplastics along the Tuscany coast (Italy): the first application of the MSFD monitoring protocol

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Due to the increasing use of plastic and its dispersion in the marine environment and accumulation in all habitats, the issue of plastic debris needs to be deeply investigated [1]. In particular, despite the Mediterranean sea is one of the hot spot area in the world for plastic debris accumulation [2], the knowledge on distribution and occurrence of floating microplastics is still lacking. Microplastics can affect marine biota increasing the likelihood of ingestion of plastics by marine organisms entering the food web. For this reason the European Union has promoted the Marine Strategy Framework Directive (MSFD) with the aim of reaching the "Good Environmental Status" by 2020.

This work has been carried out as part of implementation of the Descriptor 10.1.3 [3] of the MSFD in Tuscany (Italy), with the aim to gain information on abundance and distribution of microplastics. Sampling has been realized in two seasons (winter and spring) using a manta trawl (330 μm mesh size). Samples were collected along 4 transects 100 km far from each other, located from the estuary of Arno river to the promontory of Argentario. Each transect was divided into 4 stations located at increasing distance from the coast (20 km, 10 km, 5 km, 0.5 km). The analysis were performed according to the MSFD protocol. All data was normalized to the total volume of water filtered and expressed as items/m^3 and microplastics were characterized by colour, shape and size. A total of 2670 microplastics were isolated in the 72 samples, white was the predominant color; the majority of items are fragments and the most of microplastics fall in the measured from 1 to 2.5 mm size class. In the winter, the highest values of microplastics have been found in the station at 20 km from the coast with a gradient decreasing in the stations closer to coast; whereas in spring, the highest was found in the station at 10 km. This work represents the first application of the MSFD protocol in the monitoring of microplastics in Tuscany and will allow to understand the distribution and abundance of microplastics in the Tuscany coastal waters.

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Methods of extracting microplastic particles from fish digestive track.

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This poster presents the potentials and ongoing research on chemical digestion in extracting the microplastic particles from fish digestive tract. Main goal of this research is to determine a method that will be strong enough to dissolve the organic material and at the same time to have none or as little as possible impact on microplastic particles. The research is also focusing on other aspects of possible protocols such as the environmental impact of waste solvents, health treats to the researchers and the price of required materials and solvents.

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Mussel culture farm as possible source of microplastic litter for the coastal environment

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At the present time, offshore mussel culture farms (MCFs) can be considered as one of the major drivers affecting both the structure and functioning of the coastal area in the Northern Adriatic Sea (NAS), having important biogeochemical, biological, ecological and socio-economic linkages with other factors. They acts as no-take zones and fish aggregating devices [1]; furthermore they are expected to be a potential source of impact on bottom sediments, and to contribute to marine litter [2]. Moving from this, we focalized the attention on a preliminary characterization of the bottom under a mussel farm located on the NAS West coast, near the Sile river mouth. In order to obtain a map of possible impacts, two different methods have been combined, an acoustic seafloor mapping (by using multibeam device) and a sampling activity along the entire perimeter of the farm (by using a rapido trawl gear). The detailed topography and acoustic backscatter of the bottom below and around the MCF allowed us to identify areas exposed to higher biodeposits accumulation, whereas the trawling samples allow us to quantify the litter. The highest concentration of litter was found close to the south side of the MCF, because of the morphological and hydrological features of the farm. Almost all of the litter collected is represented by plastic (polypropylene), extruded, tubular nets used in the off-shore mussel farming. This kind of litter could be a source of microplastic for the coastal ecosystem.

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