

Assessing the presence of microplastics in mussels in the Scottish coast

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The goals of this project were:

- a. Development and description of a procedure for extraction and quantification of microplastics (MPs) in marine mussels
- b. Evaluation and quantification the presence of MPs in mussels within the Mytilidae family: horse mussels, *Modiolus modiolus* (a subtidal filter feeder) and blue mussels *Mytilus edulis* (intertidal filter feeder), to assess the presence of MPs at sites on Scottish coast

Contribution: The MASTS Small Grant ref. BFSSG7 contributed to the development and the purchase of the necessary materials to build passive samplers and cages (Fig. 1) to display *M. edulis* mussels in the field, as well as to acquire equipment to measure field data such as pH, salinity and temperature upon cages collection. The MASTS Small Grant contribution has been acknowledged in the publications and presentations where this work has been shown and discussed (see list below).

Background:

The need for a standardised method to assess MPs from organisms, including mussels, has been highlighted by the International Council for the Exploration of the Sea (ICES) advice provided to the OSPAR Commission on plastic monitoring in organisms. Our goal was to develop a procedure for extraction and quantification of MPs in marine mussels. Our approach was first to optimise the digestion efficiency of *Mytilus edulis* soft tissues by three different methods: strong acid, strong base, and a new enzyme procedure. This new enzyme procedure uses industrial enzymes that are less expensive than other enzymes used for tissue digestion in laboratory research. Rates of soft tissue digestion were compared and the effects of each digestion method on polymer integrity were assessed by FT-IR analysis of extracted MPs from spiked samples. The FT-IR analysis was done in collaboration with Prof. Richard Thompson from Plymouth University. In addition, spike recovery rates of MPs were determined and airborne fiber contamination assessed.

Documentation of MPs presence and abundance in marine environments is an important initial step towards assessment of MPs effects on organisms. Particles have been found in most coastal areas where their presence has been investigated, but little is known about the abundance and types of MPs present on the Scottish coast. Our goal was to apply the standardised method developed by us to assess MPs presence at specific locations along the Scottish coast (Arisaig, New Haven, North Uist, Skye). The study included a dedicated field campaign (2015) to collect specimens of *M. edulis* in the estuary of the Forth River, at Port Edgar, Edinburgh. The goal was to assess the abundance and type of MPs present in caged mussels and relate results to sampling location and proximity to human activities. Passive samplers were also placed in addition to mussel cages to enable semi-quantitative assessment of MPs in the water column and compare results with particles found in mussels. The assessment of microplastics in subtidal horse mussels (*Modiolus modiolus*) from Orkney was conducted as an added value output of existing projects directed by Dr. Sanderson (Heriot-Watt University, HWU, Edinburgh). This project benefited from an ongoing collaboration with a monitoring programme undertaken at HWU.

Results and Discussion:

This work has contributed to the advancement of methodological procedures to better assess the presence of MPs in mussels (and bivalves in general) in coastal and subtidal environments. This work has led to two manuscripts (see details below), one accepted for publication in *Environmental Toxicology and Chemistry* and another in preparation for submission to *Environmental Pollution*. The work was presented in various prominent international meetings in the field, the most relevant of which were the Society of Environmental Toxicology and Chemistry (SETAC) meetings.

The number of particles found in field mussels from various locations in the Scottish coast is quite low and variable overall. Field collected *Mytilus* spp samples contained 5 ± 4.5 (mean \pm SE, $n = 35$) MPs per mussel. In caged *M. edulis*, the number of particles found per mussel was similar, 6 ± 3.7 , $n = 67$. Most (96%) particles found in mussels placed in Port Edgar were fibres, and only a low number were spheres or plastic films. 2 % each (Fig. 2). The number of particles found in passive samplers placed together with caged mussels was equally low (4 ± 2.7 , $n = 12$). Subtidal mussels *M. modiolus* from Orkney also presented with a low number of plastic particles (7 ± 3.9 , $n = 6$), and most of these could be attributed to airborne fibre contamination (procedural blanks 10 ± 3.9 , $n = 6$). Registered water temperatures in Port Edgar campaigns varied between $15.7\text{ }^{\circ}\text{C}$ in the summer (31/07/2015) and $5.6\text{ }^{\circ}\text{C}$ in the winter (14/12/2015). The measured pH value was always 7.9 and salinity varied between 31 and 33.

We have successfully tested various methods currently used on soft tissue digestion efficiency, and came up with a method, enzyme digestion, that is able to maintain MP integrity during sample processing, and has a high MP spike recovery. The enzyme digestion method developed by us in the present research is offered to become a standard method for MP quantification in mussels. We believe that our method provides not only a high MP recovery rate, but it also enables recovery of sensitive MPs without damage, and increases the utility of this extraction method for a more accurate estimation of the number of MPs present in bivalves.

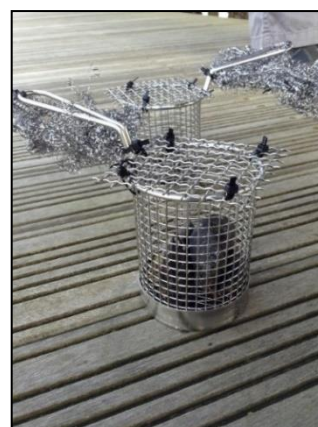


Figure 1. Example of cages used to quantify MPs found in *M. edulis* in Port Edgar, Edinburgh, in 2015 (scrubs worked as passive samplers)

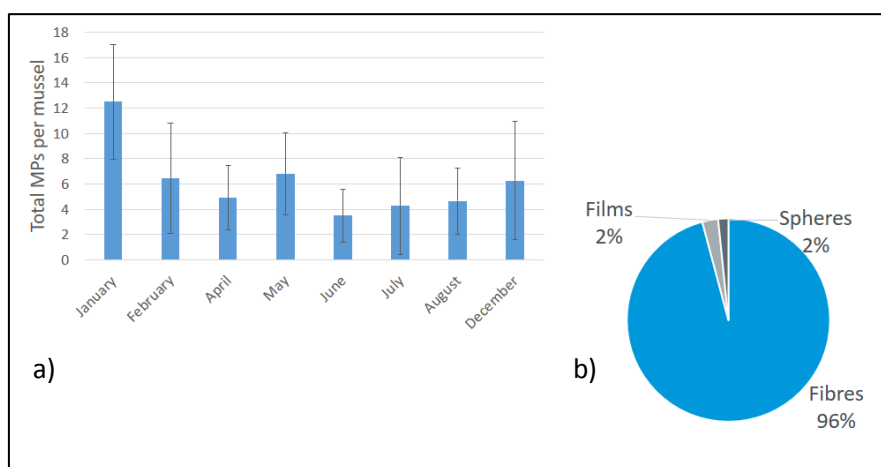


Figure 2. a) Mean number of MPs found in *M. edulis* placed in cages in Port Edgar, Edinburgh, in 2015; b) proportion of type of particles found in the analysed mussels

Publications:

- Catarino AI, Thompson R, Sanderson W, Henry TB. Development and optimization of a standard method for extraction of microplastics in mussels by enzyme digestion of soft tissues. *Environ Toxicol Chem.* 2016 Sep 1. doi: 10.1002/etc.3608
- Catarino AI, Macchia V, Sanderson W, Barras H, Thompson R, Henry TB. Assessment of microplastics present in mussels collected from the Scottish coast (in prep). *Environmental Pollution*

National and International Meetings/Conferences:

- “Assessment of Microplastics Present in Mussels Collected from the Scottish Coast” (Oral Presentation), Micro2016, 25-27 May 2016, Lanzarote, Spain, micro2016.sciencesconf.org
- “Quantification of Particles in Scottish Coastal Waters Using Mussels” (Oral Presentation), MASTS fifth Annual Science Meeting, 30 September – 2 October 2015, Technology & Innovation Centre, Glasgow, UK, www.masts.ac.uk/annual-science-meeting/
- “Distribution and quantification of microplastics present in mussels along the Scottish coast” (Poster), SETAC Europe 25th Annual Meeting, 3-7 May 2015, Barcelona, Spain, barcelona.setac.eu/?contentid=767