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MICRO 2018 Fate and Impacts of Microplastics: Knowledge, Actions and Solutions
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Preliminary study on microplastic assessment in the digestive system of marine mammals and turtles found stranded on Samos Island, Eastern Aegean Sea

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INTRODUCTION

The North-Eastern Aegean sea is characterized by the presence of a unique marine biodiversity, including cetaceans, turtles and monk seals, which occupy the top of the trophic chain and play an important role as bioindicators. To date, very few studies have been carried out for microplastic analyses in marine megafauna. However, previous research on the digestive tract of different marine species confirms the bioavailability of marine plastic debris.

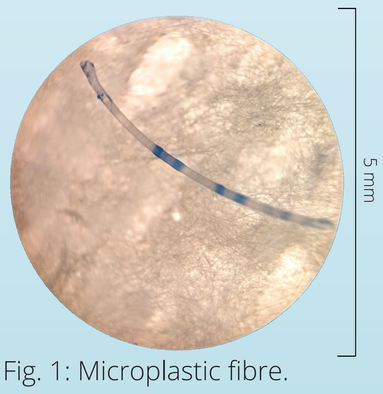

Fig. 1: Microplastic fibre.

MATERIALS AND METHODS

18 specimens of sea turtles (9 *Caretta caretta* and 9 *Chelonia mydas*), 4 of dolphins (2 *Delphinus delphis* and 2 *Stenella coeruleoalba*) and 2 Monk seals (*Monachus monachus*) of different sexes and sizes were found stranded on the shores of Samos Island between 2017 and 2018. During the necropsy, the compartments oesophagus, stomach and a random tract of the small and large intestines were isolated.
The content of each compartment was collected and the organs fully rinsed with distilled water.
The samples were passed through a stack of fractionating sieves (mesh sizes: 500 µm and 200 µm) using distilled water and a metal spatula.
Salt solution (1:1) saturation was added (24 hours) for density separation and the supernatant was mixed with H_2O_2 and CH_3COOH (1:1) to remove remaining debris.
The samples were filtered by vacuum through glass fibre filters (1.2 µm pore size). Plastic items were identified using the "hot needle test"[1], quantified and categorised into type (fiber; fragment), colour and size (SIZE A ≤ 0.2 mm; 0.2 < SIZE B ≤ 0.5 mm; 0.5 < SIZE C ≤ 1 mm; 1 < SIZE D ≤ 2.5 mm; 2.5 < SIZE E ≤ 5 mm; SIZE F > 5 mm) under x40 magnification.
A test for airborne contamination is conducted at random using moist filter papers in Petri dishes exposed to the laboratory setting as control blanks during all the examination process steps[2].

RESULTS

For all analyses, the environmental contamination blank controls did not indicate significant contamination levels in comparison to the levels found in the samples.

4 Dolphins
A total of 1306 microplastics was found.
Fibers were more abundant than fragments (χ^2 = 224.93, df = 1, p-value < 2.2e-16)
Blue was the predominant colour (χ^2 = 2322.4, df = 9, p-value < 2.2e-16)
The majority of the items found belonged to the size class B (0.2 - 0.5 mm) (χ^2 = 324.64, df = 5, p-value < 2.2e-16)
The main amount of plastic items found in the third stomach was higher than in the other compartments, although not significantly (Kruskal-Wallis χ^2 = 4.2041, df = 5, p-value = 0.5204). (Fig. 2)

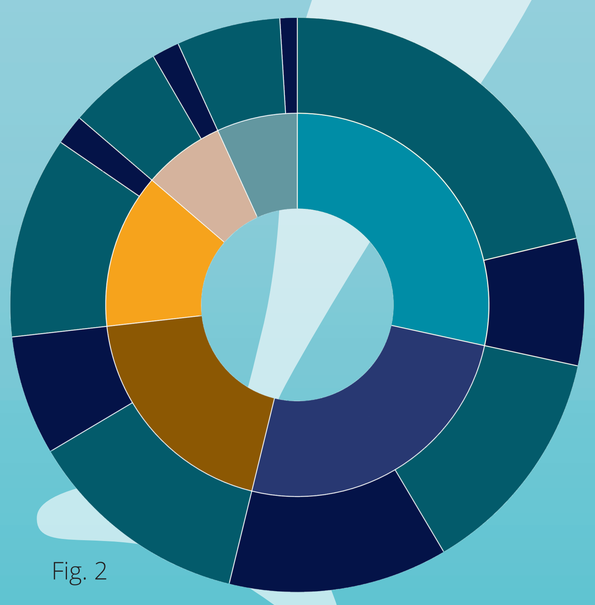
Fig. 2

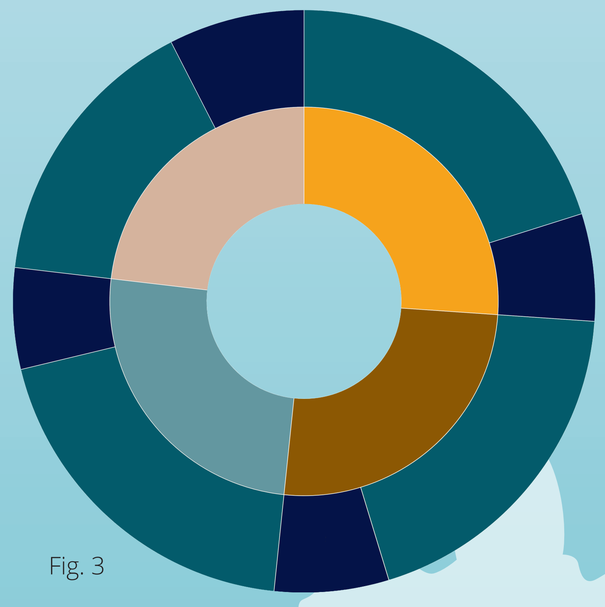
Fig. 3

18 Turtles
A total of 3554 microplastics was found
Fibers were more abundant than fragments (χ^2 = 857.77, df = 1, p-value < 2.2e-16)
Blue was the predominant colour (χ^2 = 4172.2, df = 8, p-value < 2.2e-16)
The majority of the items found belonged to the size class B (0.2 - 0.5 mm) (χ^2 = 1108.7, df = 5, p-value < 2.2e-16)
The main amount of plastic items found in the oesophagus was higher than in the other compartments, although not significantly (Kruskal Wallis Test: χ^2 = 2.5932, df = 3, p-value = 0.4587). (Fig. 3)

2 Monk seals
A total of 297 microplastics was found.
Fibers were more abundant than fragments (χ^2 = 128.03, df = 1, p-value < 2.2e-16)
Transparent was the predominant colour (χ^2 = 320.48, df = 8, p-value < 2.2e-16)
The majority of the items found belonged to the size class B (0.2 - 0.5 mm) (χ^2 = 152.96, df = 5, p-value < 2.2e-16).
The main amount of plastic items found in the stomach was higher than in the other compartments, although not significantly (χ^2 = 225.86, df = 1, p-value < 2.2e-16). (Fig. 4)

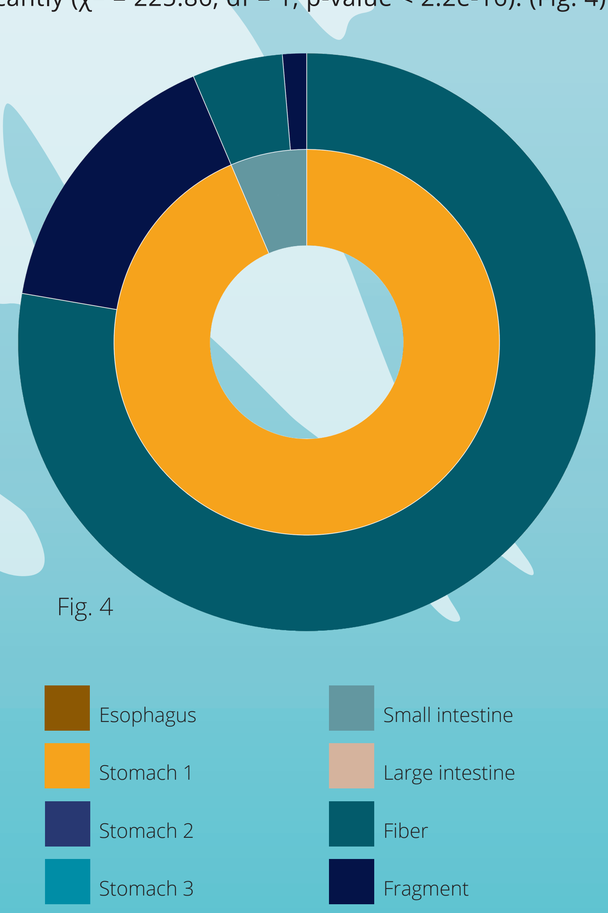

Fig. 4

DISCUSSION

A high concentration and variety of microplastics were found throughout each tract of the digestive systems, confirming the wide pollution of plastic affecting the top predators of the trophic chain.
The discovered trend showed that a greater amount of microplastics was found in the oesophagi of the turtle dataset, which can be explained by certain anatomical characteristics of the species. This finding highlights the importance of considering all compartments of the digestive tracts when conducting microplastic assessments.

The prevalence of specific colours and sizes needs further studies to identify the source.

CONCLUSION

Considering the number of samples and the species of turtles this study underlines for the first time the relevance of the oesophagus in microplastic analysis.
First data on microplastic contamination in Mediterranean Monk seal are reported in this research.

Analyses need to be conducted to understand whether there is a link between size and sex of the animals and the abundance of microplastics. Toxicological analyses are necessary to determine the potential impact on the health of these species in the Aegean Sea.

This study proposes an innovative, cost-efficient and targeted method, which maximises the results of a complete assessment of micro and macroplastic content in the digestive tracts of marine megafauna.

References
[1]De Witte B., Devriese L., Bekaert K., Hoffman S., Vandermeersch G., Cooreman K., and Robbens J. (2014). Quality assessment of the blue mussel (*Mytilus edulis*): Comparison between commercial and wild types. Marine Pollution Bulletin 85, 146-155.
[2]MSFD Technical Subgroup of Marine Litter. Guidance on Monitoring of Marine Litter in European Seas. JRC - Joint Research Centre. EUR 26113 EN (2013).

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