LOOKING AT THE MACRO IS NO LONGER ENOUGH: A PROTOCOL TO ADDRESS THE STUDY OF MICROPLASTIC INTAKE IN STRANDED CETACEANS

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FATE AND IMPACTS OF MICROPLASTICS: KNOWLEDGE AND RESPONSIBILITIES

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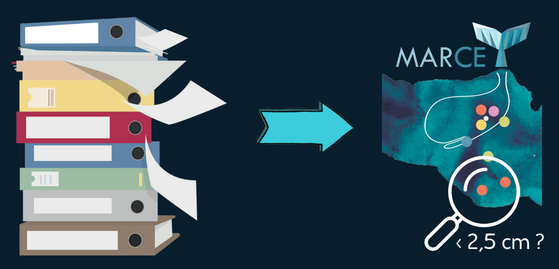
INTRODUCTION

Marine debris can impact biodiversity in a number of ways, and its effects may vary depending on the type and size of the debris and the organisms that encounter it [1]. Since the first evidence of a marine mammal's interaction with plastic intake, there have been **a number of studies on this subject**, together with alarming images of stomachs full of marine debris and a growing concern about it.



Which were the knowledge gaps found?

However, very little is known about the presence of microplastics in higher trophic level species such as cetaceans [2].

And moreover, up to more recently, they were primarly focused on the study of particles larger than 2.5 cm, and therefore failing to assess the microlitter presence, which remains a challenging task due to large gut content volumes and the difficulties of sampling following careful airborne contamination prevention protocols.

WHAT DID WE DO?

Working with stranded cetaceans, which represent a significant opportunity to study the interaction of marine fauna with plastic debris, we have validated a protocol for microplastic ingestion studies that serves to obtain samples from different multidisciplinary teams (i.e. veterinary and marine sciences schools), without interfering in the work of any of the parties [3].

THE PROTOCOL WORKFLOW

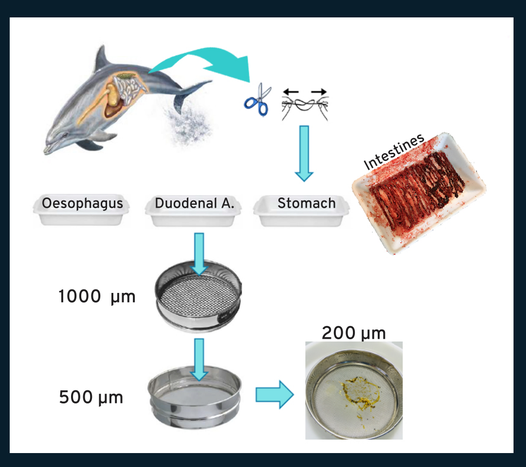
Before you start:
(i) Wear cotton clothes while manipulating the samples,
(ii) Clean all containers using distilled water prior to its reuse,
(iii) Perform blank controls filtering MilliQ water,
(iv) Place a clean petri dish with a filter paper close to the manipulation area to register possible airborne contamination,
(v) Cover all samples with aluminium foil after each single step and during the procedure.

Once the extraction of the digestion tract is performed and the necessary samples for anatomopathological, microbiological, parasitological and dietary studies are collected:

1. Separate each of the GIT compartments (oesophagus, duodenal ampulla, stomach and intestines) into different trays.
2. One by one, empty the digestive content of each GIT section on the table and rub the mucous membranes thoroughly until all the digestive remains have been washed away.
3. Carry out a macroscopic examination and take samples for histopathology and microbiology.
4. Rinse carefully so all the digestive content is retained in each of the sieves described above.
5. The filtered remains will be introduced into different containers for each organ (oesophagus, stomach and duodenal ampulla, and intestine) and will be stored for their subsequent laboratory study.

What were the keys to success?

Table design: A custom-made adaptation to the necropsy table was made connecting its drainage to a set of three stacked metal sieves (1000, 500 and 200 µm) where the washed stomach contents were retained after thorough gastrointestinal (GIT) rinses.



Team work: The protocol is simple and cost-effective and allows research teams to collect and analyse samples of the presence of microlitter in a comparable way, therefore reaching a more thorough understanding of the risk microplastics pose to marine mammals.

CONCLUSIONS

The successful table set up used for the extraction of microplastic particles from the gastrointestinal contents was proofed advantageous and applicable by any research group that already counts with the necessary facilities to perform cetaceans autopsy analysis, fulfilling the harmonisation needs as explicated by Panti et al. [4].

This approach is fully compatible with necropsy protocol in cetaceans [5], and at the same time complies with the recommendations for reporting ingested plastics in marine megafauna [6]. The proposed workflow allows the collection of valuable data for different interdisciplinary research teams, aiming to harmonize data, facilitate large-scale comparisons of plastic ingestion and also give scientific basis to future conservation policies.

Check out the complete protocol at:

SCAN ME

protocols.io

dx.doi.org/10.17504/protocols.io.bcfxitpn

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