Using Tissue clearing for the analysis of ingested microplastic particles

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Abstract

The analysis of the ingestion of microplastics (MP) by biota is frequently performed through invasive procedures such as chemical digestion protocols or by histological analysis of thin sections. Nonetheless, both approaches lack spatial resolution and cannot offer information on regions in which MP particles potentially accumulate. Knowledge on accumulation zones of MP might enable the determination of a region of interest (ROI) for the analysis of effects or particle translocation into the surrounding tissue. A way of analyzing the digestive system of otherwise opaque organisms, is by rendering these organisms transparent. Different, promising approaches for the observation of ingested MP particles pose so called tissue clearing methods. They are currently applied to organs, tissue samples, or whole organisms, rendering the sample transparent and enable to look inside an otherwise opaque environment. Therefore, our goal was to adapt the CUBIC tissue clearing protocol (Clear, Un- obstructed Brain/Body Imaging Cocktails and Computational Analysis) for aquatic and terrestrial organisms of various functional feeding groups for the analysis of the uptake of fluorescent labeled microplastic (MP) particles. The adapted CUBIC method has led to transparency in all normally opaque organisms. It further offers a simple way of locating fluorescent labeled MP inside the digestive system of the different organisms while leaving them intact.

Keywords: Microplastics, CUBIC, Tissue clearing, Particle ingestion, Invertebrates, Anatomy

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