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A workflow for improving estimates of microplastic contamination in marine waters: A case study from North-Western Australia☆

Frederieke Kroon [a, *], Cherie Motti [a], Sam Talbot [a], Paula Sobral [b], Marji Puotinen [c]

[a] Australian Institute of Marine Science (AIMS), Townsville, Qld, 4810, Australia
[b] Universidade Nova de Lisboa, Faculdade de Ciências e Tecnologia, Lisbon, Portugal
[c] Australian Institute of Marine Science (AIMS), Perth, WA, 6009, Australia

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ABSTRACT

Plastic pollution is ubiquitous throughout the marine environment, with microplastic (i.e. <5 mm) contamination a global issue of emerging concern. The lack of universally accepted methods for quantifying microplastic contamination, including consistent application of microscopy, photography, an spectroscopy and photography, may result in unrealistic contamination estimates. Here, we present and apply an analysis workflow tailored to quantifying microplastic contamination in marine waters, incorporating stereomicroscopic visual sorting, microscopic photography and attenuated total reflectance (ATR) Fourier transform infrared (FTIR) spectroscopy. The workflow outlines step-by-step processing and associated decision making, thereby reducing bias in plastic identification and improving confidence in contamination estimates. Specific processing steps include (i) the use of a commercial algorithm-based comparison of particle spectra against an extensive commercially curated spectral library, followed by spectral interpretation to establish the chemical composition, (ii) a comparison against a customised contaminant spectral library to eliminate procedural contaminants, and (iii) final assignment of particles as either natural- or anthropogenic-derived materials, based on chemical type, a compare analysis of each particle against other particle spectra, and physical characteristics of particles. Applying this workflow to 54 tow samples collected in marine waters of North-Western Australia visually identified 248 potential anthropogenic particles. Subsequent ATR-FTIR spectroscopy, chemical assignment and visual re-inspection of photographs established 144 (58%) particles to be of anthropogenic origin. Of the original 248 particles, 97 (39%) were ultimately confirmed to be plastics, with 85 of these (34%) classified as microplastics, demonstrating that over 60% of particles may be misidentified as plastics if visual identification is not complemented by spectroscopy. Combined, this tailored analysis workflow outlines a consistent and sequential process to quantify contamination by microplastics and other anthropogenic microparticles in marine waters. Importantly, its application will contribute to more realistic estimates of microplastic contamination in marine waters, informing both ecological risk assessments and experimental concentrations in effect studies.

1. Introduction

Plastic pollution is ubiquitous throughout the marine environment with an estimated hundreds of thousands of tons floating in the world's ocean (Cozar et al., 2014; Eriksen et al., 2014; van Sebille et al., 2015). Marine plastic pollution is prevalent in the water column, on the seabed and along shorelines (United Nations Environment Programme, 2009). These plastics are derived from a wide variety of marine and land-based sources, including (un-) intentional discard from shipping and fishing, and land disposal via industrial and urban discharges and river run-off (United Nations Environment Programme, 2009). The timeframe for complete mineralisation of plastic in the marine environment is unknown but is estimated to range from months to millennia (Barnes et al., 2009), depending on a combination of environmental factors and the chemical composition and properties of the polymer. Such persistence makes plastic pollution of the marine environment a long-term issue, particularly considering the projected exponential

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* Corresponding author.
E-mail address: f.kroon@aims.gov.au (F. Kroon).

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increase of plastic production (Lebreton et al., 2012).

Over the last decade, contamination of the ocean environment by microplastics (i.e. plastic debris <5 mm diameter) (Barnes et al., 2009; GESAMP, 2015) has received increasing attention as a specific aspect of marine plastic pollution (GESAMP, 2016). The terms 'primary' and 'secondary' microplastics refer to particles being either specifically manufactured for particular applications (e.g. resin pellets, cosmetic products, oil dispersants), or produced as a result of fragmentation from larger items, respectively. This distinction can be used to pin-point sources of marine plastic pollution and target measures for mitigation. Similar to marine plastic pollution in general, microplastic contamination of the ocean is widespread and has been reported for shorelines (Antunes et al., 2013; Browne et al., 2011; Gregory, 1978; Thompson et al., 2004), surface waters (Colton et al., 1974; Enders et al., 2015; Reisser et al., 2013; van Sebille et al., 2015), pelagic zone (Kukulka et al., 2012; Thompson et al., 2004) and deep sea habitats (Fischer et al., 2015; Van Cauwenberghe et al., 2013). Across these habitats, microplastic ingestion has been documented for a range of marine organisms (Nadal et al., 2016; Tanaka and Takada, 2016; Taylor et al., 2016), including species consumed by humans (Avio et al., 2015; Foekema et al., 2013; Neves et al., 2015; Possatto et al., 2011; Rochman et al., 2015; Wright et al., 2013).

Despite the increasing knowledge of the abundance, distribution and effects of microplastic contamination in the marine environment, the ecological risk posed by microplastics is still uncertain (GESAMP, 2016). Ecological risk is generally defined as the probability that adverse ecological effects will occur as a result of exposure to one or more stressors (U.S. Environmental Protection Agency, 1992). Hence, a critical component to assess the ecological risk of microplastics in marine environments is the availability of exposure data, including the concentration, type, size, and shape of microplastics (GESAMP, 2016). Obtaining such information is currently hampered by the lack of universally accepted methods for quantifying microplastic contamination (GESAMP, 2015; Hidalgo-Ruz et al., 2012; Lusher, 2015; Miller et al., 2017), including systematic procedures for polymer assignment using spectroscopy. Recently, Fourier transform infrared (FTIR) spectroscopy has been reported as the most reliable method to characterise the chemical composition of microplastics (Hidalgo-Ruz et al., 2012), because it is a chemically non-destructive, diagnostic, reproducible and simple to perform technique (Shim et al., 2017). However, confirmation by FTIR spectroscopy of particles that visually resemble plastics does not always occur (Graham and Thompson, 2009; Ivar do Sul et al., 2014; Stolte et al., 2015), or is conducted on only a subset of particles (Cole et al., 2014; Martins and Sobral, 2011; Reisser et al., 2013). Furthermore, spectral interpretation (e.g. comparison of diagnostic signals) and interrogation (e.g. comparison of overall spectra) is crucial to assign chemical type (Boruta, 2012; Neves et al., 2015; Shim et al., 2017; Workman and Weyer, 2012). However, most microplastic studies do not report on the exact process used to inspect FTIR spectral data (Frias et al., 2010; Imhof et al., 2017; Lusher et al., 2013; Reisser et al., 2013; Thompson et al., 2004). The lack of systematic procedures for chemical assignment using spectroscopy makes comparisons across studies difficult, and is ultimately contributing to unrealistic contamination information for marine environments.

In this study, we present an analysis workflow tailored to quantifying microplastic contamination in marine waters (Fig. 1). The specific aim of this workflow is to introduce a level of consistency in the step-by-step processing and associated decision making, thereby reducing bias in plastic identification and improving confidence in contamination estimates. In this tailored analysis workflow, we incorporate methods commonly used in processing and analysis of marine tow samples, namely stereomicroscopic visual sorting, microscopic photography and attenuated total reflectance (ATR) Fourier transform infrared (FTIR) spectroscopy (Miller et al., 2017). Specific processing steps include (i) the use of a commercial algorithm-based comparison of particle spectra against an extensive commercially curated spectral library, followed by spectral interpretation (and if necessary signal interrogation) to establish the chemical composition and to assign generic chemical type, (ii) a comparison against a customised contaminant spectral library to eliminate contaminants inadvertently introduced during collecting and processing, and (iii) final assignment of particles as either natural- or anthropogenic-derived materials, based on chemical type, a compare analysis of each particle against other particle spectra, and physical characteristics of particles. We demonstrate the application of this analysis workflow to recent surveys from the remote marine waters of North-Western Australia as a case study. Our results show that its application will contribute to more realistic estimates of microplastic contamination in marine waters, informing both ecological risk assessments and experimental concentrations in effect studies. The application of the analysis workflow further highlights potential improvements that could be made to specific processing steps and associated recommendations are made accordingly.

2. Material and methods

2.1. Study area

Surveys were conducted in remote areas not covered in previous studies (Reisser et al., 2013), namely along the coast of the Kimberley region and around several islands and shoals in Australia's Indian Ocean territory (Fig. 2). The Kimberley region in North-West Australia covers an area of 419, 558 km^2 and is sparsely populated (34,794 people in 2011) (Australian Bureau of Statistics, 2013). The climate is characterised by marked wet summers (November to April) and dry winters (May to October), with the majority of total rainfall (650–1200 mm) occurring from January to March (Bureau of Meteorology, 2012). The dominant oceanographic feature in the region, the Indonesian Throughflow (ITF), interacts with near-surface currents which flow northeast in summer and southwest in winter (Schiller, 2011). Currently, the main industries in the Kimberley are mining, tourism, retail, construction, agriculture, (pearl) aquaculture and fishing (Department of Regional Development, 2014). Offshore, the Browse and Bonaparte basins are two oil and gas bearing basins with fields first discovered in, and subsequently developed since the 1970s (Department of Regional Development, 2014; Zabanbark, 2010). Some of the offshore reefs and islands are fished by Indonesian fishermen under a 1974 Memorandum of Understanding between Australia and Indonesia (Stacey, 2007).

2.2. Microplastic sampling

The presence and abundance of microplastics were examined by replicate surface and subsurface tows at 14 locations along the remote North-Western coast of Australia during one transit voyage on the AIMS Research Vessel (RV) Solander (21st to 28th November 2015) (Table S1, Fig. 2). At 13 of these locations, four horizontal tows were conducted to sample the air-sea interface ($n = 2$) and the water column (depth of 5–10 m, $n = 2$), respectively. A neuston frame (dimensions: 74.5 cm diameter, 30.0 cm height) was used for surface tows, and a round frame (62.5 cm diameter) for subsurface tows; identical plankton nets (length 254 cm, 355 μm polyester (PES) with an open area of 50% and 150 μm thread thickness) were used on the neuston and round frames. Tows were conducted at <4 knots from a hydro winch on the starboard side of the vessel to