A NOVEL ANALYTICAL METHOD TO ASSESS MICROPLASTIC DIVERSITY, ABUNDANCE AND MASS IN THE MARINE ENVIRONMENT SAMPLES

Luis E. Medina, Tatiana Zaliznyak and Gordon T. Taylor

NAno-RAMAN Molecular Imaging Laboratory, School of Marine and Atmospheric Sciences Stony Brook University, Stony Brook, New York, USA

luis.medina@stonybrook.edu

Introduction

To date, research into marine microplastic (MP) pollution has focused on quantifying total number of MP particles from environmental samples using techniques suitable for particles >300 µm in diameter. Consequently, an important fraction of particles have likely been understudied, possibly leading to an underestimation of the ocean's plastic budget. Moreover, unlike total mass, particle number does not translate into total plastic for budget calculations. It is also not yet clear how oceanographic processes affect the breakdown of these particles nor how sampling methods and analytical techniques bias inventory estimates. These issues require measurement of MP mass as a determinant of total plastic loads.

Using Raman microspectroscopy coupled with 3-D spectral imaging, we have developed an automatable methodology for environmental samples to detect, identify, and quantify mass of micron-sized MPs directly from filters. This method can detect MP particles less than 20 µm in diameter and generate 3-dimensional images based on the Raman spectra of the polymer. The volume of an individual particle can be determined, and the mass calculated based on known MP densities. This methodology enables the estimation of MP mass, providing vital information for mass balance calculations of each type of MP in the ocean.

Methodology

1. Standards suspensions of MPs particles < 250 µm in diameter derived from different types of synthetic polymers (e.g., polypropylene, polyethylene) were prepared in seawater.

2. MP suspensions along with laboratory and field blanks were filtered through 0.2 µm pore size Anopore inorganic (Al_2O_3) filters (25 mm diam.).

3. To minimize non-plastic residue and phytoplankton pigment fluorescence, filters were rinsed with methanol.

Renishaw® inVia™ confocal Raman micro-spectrometer
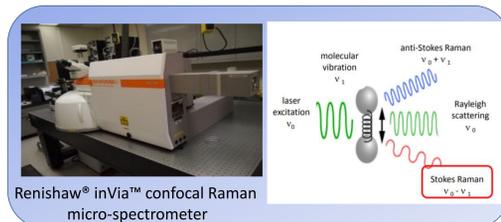
4. To identify MP particles on filters, full point maps are run on at least 10 filter sections. Motorized stage moves sample within a grid under laser in small increments enabling collection of spectra at various points (Fig 1.a and 1.b).

5. After identification, and based on the chemical spectra of each particle, a chemical image was generated from which the volume was calculated. Using the know density of each plastic, the mass of the MP particle was calculated.
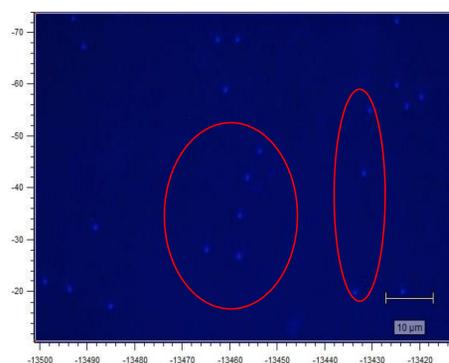
Figure 1.a. Polyethylene microbeads (0.46 µm) under epifluorescent illumination.
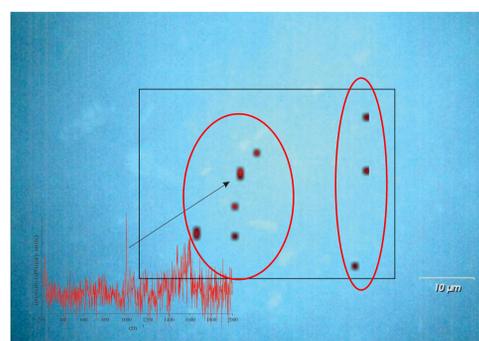
Figure 1.b. Polyethylene microbeads (0.46 µm) under bright-field illumination and identified by Raman spectra.

Results

MPs of polypropylene were generated as standards. As an example, the three-dimensional distribution of diagnostic polypropylene Raman spectral peaks were used to generate an image of an amorphous particle with dimensions varying between 10 and 20µm **(Fig. 2)**. Based on this 3-D spectral image, the particle volume was 7547 µm³, corresponding to a mass of 6 ng polypropylene (density 0.946 g/cm³). Time required to produce this chemical map was approximately 40 min. To validate this result, the hypothetical volume of the best-fit solid ellipse enclosing the particle was determined to be (7270 µm³), also corresponding to a mass of ~6 ng polypropylene.
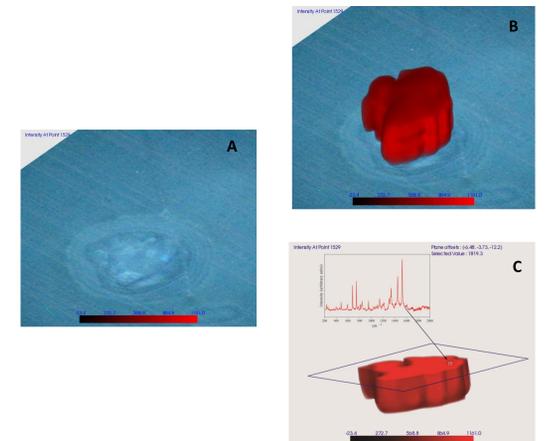
Figure 2A. 2-D Bright-field image showing a MP particle. 3B and 2C. 3-D chemical maps of diagnostic peak and Raman spectrum for polypropylene.
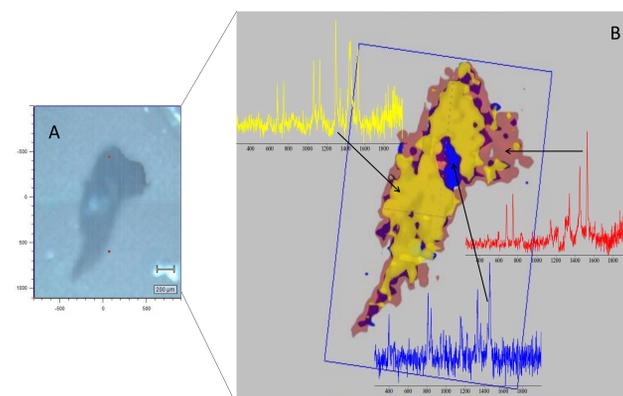
Figure 3A. 2-D Bright-field image of a MP particle. 3B. 3-D chemical map of diagnostic peaks and Raman spectra of polyprolylene (red), polyethylene (blue) and the mixture (yellow).

Figure 3B shows the 3-D chemical map of a MP particle generated based on the diagnostic peaks of polypropylene (red), polyethylene (blue), and a mixture of both (yellow). The amorphous particle has major and minor axes of 1450 and 510 µm respectively. Based on this 3-D spectral image, the volume of the particle was 37,456 µm³, corresponding to a mass of 1023 ng polypropylene (density 0.946 g/cm³) and 256 ng of polyethylene (density 0.975 g/cm³). Data acquisition time for this chemical map was approximately 180 min.

Analysis of an Environmental Sample

Applying this methodology, we started to analyze water samples collected from coastal Antartica by LDEO (Hugh Ducklow, Columbia University, NY). The first samples analyzed (0 m depth) revealed 35 MP particles/L. Particle diameters ranged between 6-31 µm.

Figure 4A is a bright-field image of the filter area mapped for the 6 µm particle. The 3-D chemical image of the same particle is presented in **Figure 4B**. In total, the particles contained 18 ng of polypropylene, corresponding to 200 ng/L.
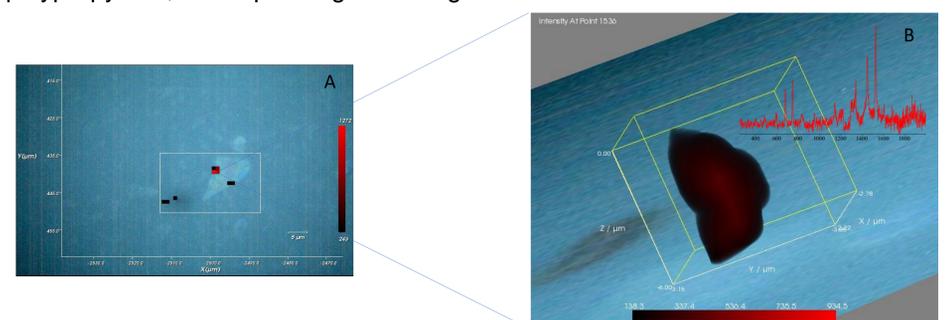
Discussion

Raman microspectroscopy has the capability to generate spatial chemical maps of plastic particles based on diagnostic spectral peaks of the polymers and thus allows for precise identification and quantification of MPs captured on Al_2O_3 filters at a spatial resolution down to 0.46 µm (equipment detection limit for MP). This capability facilitates detection of very small MP particles from environmental samples and allows for estimation of particle abundance and mass of each polymer. Armed with this information, a more realistic estimate of MP abundances can be obtained to ultimately generate a MP mass balance for the ocean.