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Analysing microplastics with the inVia™ confocal Raman microscope

Microplastics in the environment

Plastics are extensively used in products and packaging and, unfortunately, make their way into the environment, causing significant issues for wildlife. In addition to concerns about bulk plastic contamination, there has been much interest in microplastics. These are small particles or fibres, typically less than 5 mm in diameter, sometimes found at high levels in the environment [1,2,3]. They are classified as: primary microplastics, which are intentionally manufactured; and secondary microplastics, which are fragments derived from the breakdown of larger plastic debris.

Microplastics can accumulate in the tissues of many organisms, enter the food-chain, and adversely affect ecosystems, biodiversity, and human health[4]. Consequently, there is much research into the levels of contamination and how microplastics propagate in the environment.

What can Raman spectroscopy offer?

Raman spectroscopy, an optical scattering technique, has much to offer researchers investigating plastics, as it can identify them rapidly and non-destructively.

Renishaw's inVia Raman microscope is ideal for studying microplastics:

- You can analyse particles with sizes ranging from many millimetres across down to as small as 1 µm
- Its measurements can be automated, saving you time, and giving consistent, reliable results
- It has an extensive suite of analysis tools you can use to identify particles (using spectral lookup databases) and generate statistics on their composition, size, and shape
- It has advanced focus-tracking, and can analyse particles collected on a wide range of substrates

The Renishaw inVia Qontor confocal Raman microscope

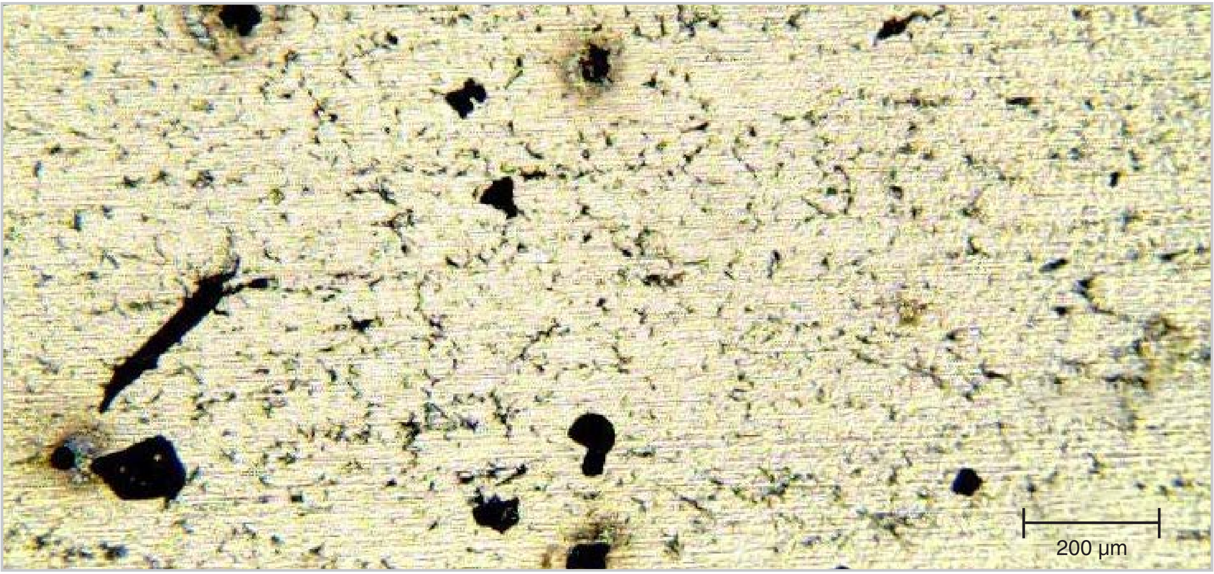
Automated analysis on the inVia microscope

The inVia microscopes software provides great flexibility in designing automated workflows that enable fast, non-labour-intensive analysis.

The following is an example workflow for the analysis of microplastics:

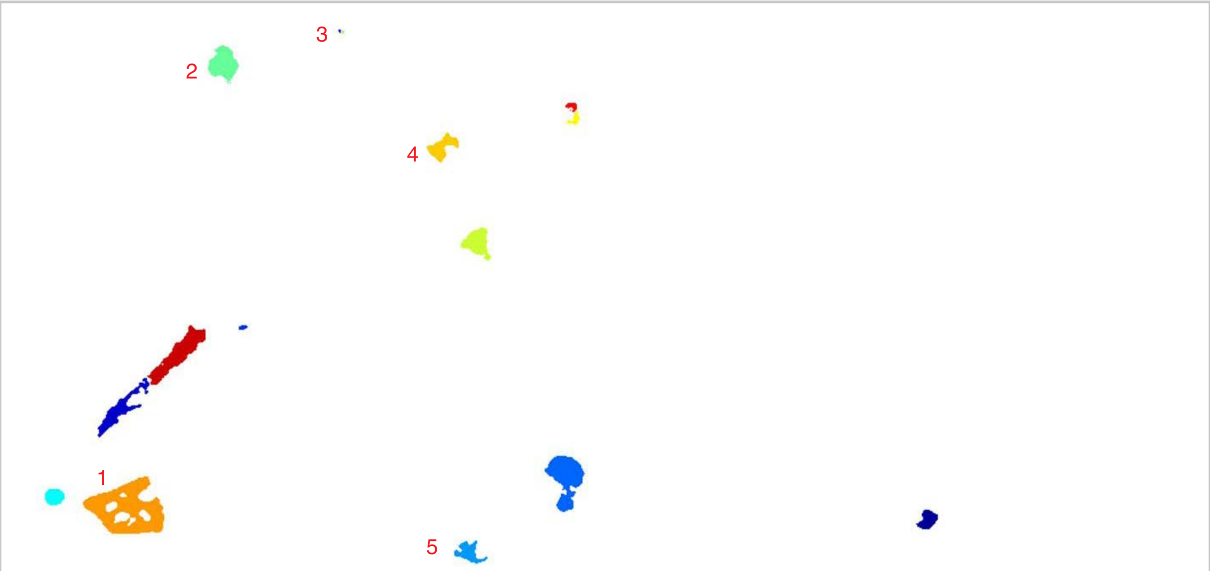
- Deposition. Filtered microparticles are deposited onto a substrate
- Loading. The substrate is placed on the microscope stage of the inVia system
- Optical imaging. The inVia microscope generates a high-resolution optical microscope image of the substrate by moving it, acquiring multiple high-magnification images, and tiling them together. Where optical contrast is limited, laser mapping enables candidate plastic regions to be rapidly determined
- Particle determination. The software analyses the image, determining the positions of the particles
- Raman analysis. Each particle is then automatically analysed, either by taking a single spectrum from the particle centre, or by mapping the whole particle (the latter would indicate if the particle had multiple polymer types within it). Depending on the sample, LiveTrack™ automated focus-tracking may or may not be needed
- Analysis. This involves matching the spectra to Renishaw's polymer database (possibly augmented with user-created custom databases)

In the following example, this workflow is used to analyse particles extracted from sea water. These were deposited on a Renishaw mirror-polished stainless-steel slide, an ideal substrate as it affords excellent optical contrast, and it does not interfere with the polymer spectrum.



Optical imaging

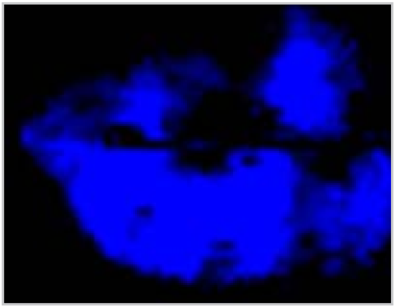
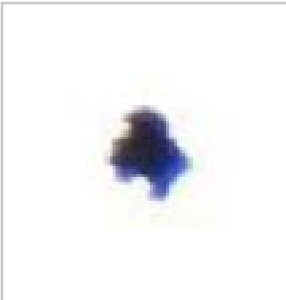
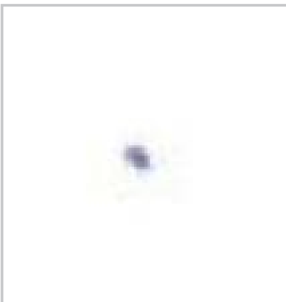
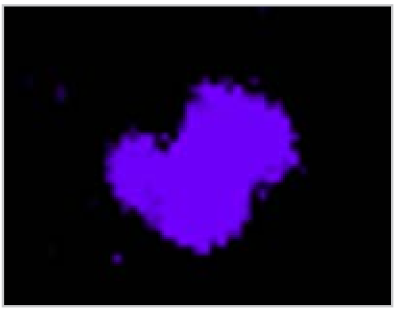
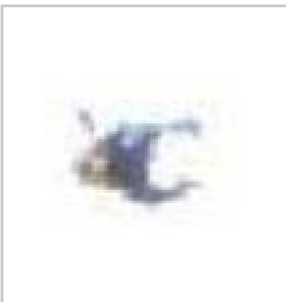
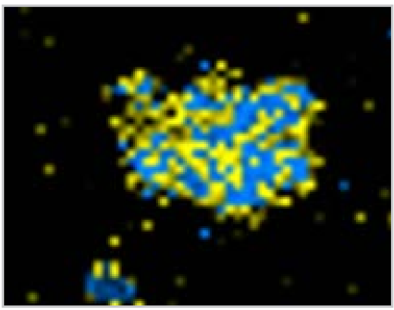
The high-resolution optical microscope image of the substrate (1.8 mm × 1.1 mm). Typically, large domains are contaminated debris, and so are more likely to be of interest. In this study, these particles were selected for further analysis. If a more detailed investigation is required, smaller domains can be analysed.



Particle determination

The software determines the locations of the particles and highlights them in distinct colours. Five have been assigned numbers to aid later discussion.

In this workflow, each particle was mapped, the spectral data then compared with Renishaw's database of polymeric and inorganic materials, and images produced. For brevity, we give details of just five particles.

Particle	Optical microscope image	Raman image	Conclusion
1			The bulk of the particle is identifed as polypropylene (coloured blue in the Raman image)
2			The particle is a combination of hematite (magenta) and an other iron-based inorganic (green)
3			The particle is anatase (green) and quartz (orange)
4			The particle is silica (purple)
5			Particle comprises an intimate mix of PTFE (blue) and an unidentified flourescent material (yellow), likely a dye.

Analysis of all the particles indicated that 2 of the 14 were microplastic fragments (polypropylene and PTFE). However, the remaining microparticles could still be identified based on the Raman analysis. These additional microparticles were largely inorganic and mineral-based species including: hematite, pyrrhotite, anatase, quartz and silica.

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Conclusions

This document has shown that the inVia confocal Raman microscope is an ideal tool for the analysis of microplastics. You can use it to locate fragments, identify the polymers present, and output statistics on particle size and composition. In the example, we used a flat substrate, but with LiveTrack focus-tracking the methods shown here can be applied to a broad range of substrates.

References

[1] Richard C. Thompson, Ylva Olsen, Richard P. Mitchell, Anthony Davis, Steven J. Rowland, Anthony W. G. John, Daniel McGonigle, Andrea E. Russell (2004). “Lost at Sea: Where Is All the Plastic?” Science 07 May 2004: Vol. 304, Issue 5672, pp. 838 DOI: 10.1126/science.1094559

[2] Shim, W. J. and Thompson, R.C., (2015). Microplastics in the Ocean. Archives of Environmental Contamination and Toxicology, 69, 3, 265 - 268

[3] Arthur, C., J. Baker and H. Bamford (eds) (2009). Proceedings of the International Research Workshop on the Occurrence, Effects and Fate of Microplastic Marine Debris. NOAA Technical Memorandum NOS-OR&R-30.

[4] http://www.independent.co.uk/news/science/microplastic-waste-this-massive-tiny-threat-to-sea-life-is-now-in-every-ocean-9602430.html

A range of related Renishaw literature is available. Please ask your local Renishaw representative for more information.

Renishaw. The Raman innovators

Renishaw manufactures a wide range of high performance optical spectroscopy products, including confocal Raman microscopes with high speed chemical imaging technology, dedicated Raman analysers, interfaces for scanning electron and atomic force microscopes, solid state lasers for spectroscopy and state-of-the-art cooled CCD detectors.

Offering the highest levels of performance, sensitivity and reliability across a diverse range of fields and applications, the instruments are designed to meet your needs, so you can tackle even the most challenging analytical problems with confidence.

A worldwide network of subsidiary companies and distributors provides exceptional service and support for its customers.

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