Use and development of the Microparticle Sample Preparation Kit

Isolating microplastics and other microparticle contaminants for spectroscopic identification

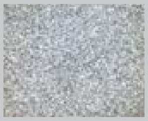
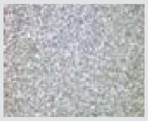
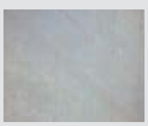
The isolation and identification of particles, specifically microplastic particles, is an area of increasing interest. The discovery of microplastic particles in sea life, natural waterways, and in consumable products like bottled water and other beverages has sparked concern for the potential environmental and toxicological impact of these contaminants. The isolation of particles from liquid suspensions involves filtration and identification of the resulting particles is accomplished using either FTIR or Raman microscopy, or both. While there are a wide variety of choices for filter materials, it is important that the filter material be compatible with vibrational spectroscopic methods. This report provides details on the development and use of the Thermo Scientific™ Microparticle Sample Preparation Kit for dilute suspensions of particles in liquids such as water.

The first step of the development process was to select a filter material that would allow for both FTIR and Raman analysis. The characteristics of the filter material that were considered most important for this evaluation were the following:

1. The filter material should allow for FTIR microscopic analysis preferably in transmission as well as reflectance modes and should be suitable for Raman analysis as well.
2. The filter material should provide for a clear visual image of the particles. The surface texture cannot interfere with analysis routines that use visual image analysis for targeting potential particles for spectroscopic identification as well as using the visual image for determining the size and shape.
3. The filter needs to lay flat for analysis. While there are ways to mount more flexible filter materials to get them flat and there are ways to accommodate uneven filter surfaces these are just complications that, if they can be avoided, simplify the analysis.
4. Any spectroscopic contributions from the filters should be very small or limited and well defined so they do not interfere with the analysis of the particles of interest.
5. The filter material should be reasonably non-reactive with the liquid matrixes of interest.
6. The filters should be readily available and not prohibitively expensive.

Results from a recent study using common types of filters can be seen in Table 1.

Table 1. Summary of advantages and disadvantages of different filter materials

Filter type	Advantages	Disadvantages	FTIR	Raman
Gold-coated polycarbonate 	• Readily available • Can be used with filter apparatus without gaskets	• Does not lie flat • Highly reflective surface may hinder contrast (particle recognition) • Expensive	Good choice for reflection	• Possible to see polycarbonate peaks through gold • Some broad baseline offset with some lasers
Silver 	• All metal • Less expensive than gold coated	• More rigid than gold-coated PC • More of a textured surface at high magnification • More reactive – reported problems with pH of carbonated water	• Reasonable for reflection – less reflective than gold	• Some spectral artifacts from filters themselves
Al_2O_3 	• Readily available • More rigid • Transmitted light possible if intense enough • Less expensive option	• Delicate – easily broken • Visual images – contrast an issue – surface not clearly defined. Some features on surface that might be detected as particles	• Can be used in transmission but limited to > 1,250 cm^{-1} • Some spectral peaks and some variation in peaks over the filer. • Reflection weak	• Some Raman spectral contributions from the filters – broad features • Baseline offsets • Laser light transmits through
Silicon 	• Rigid • Good visible images	• Square • Needs gasket development • Fragile • Expensive	• Transmission • Some variation across filter (filter background: (Si-O)) – broad baseline offset • Reflection not as good as gold but possible	• Silicon peaks

We chose the silicon filter because it has the majority of the desired traits. Silicon allows for transmission analysis with the FTIR microscope and while silicon has strong Raman spectral features, they are reasonably sharp, well defined, and can be accounted for during Raman analysis. A Raman spectrum of the filter itself can be subtracted from all of the particle spectra in the map. That removes the spectral features of the silicon filter from the particle spectra but there may be some residual spectral features that remain due to incomplete subtraction. The spectral regions containing these residual features can be avoided when doing spectral searching; to be sure they do not interfere with particle identification.

Since the silicon filters are square and rigid it was necessary to adapt a standard filtration apparatus designed for 13 mm diameter circular filters. Gaskets were used to adapt the square filters to the apparatus designed for larger circular filters, to prevent leaks, and to provide some cushion for the brittle silicon filters to prevent breakage. The filter apparatus used for this investigation is shown in Figure 1.

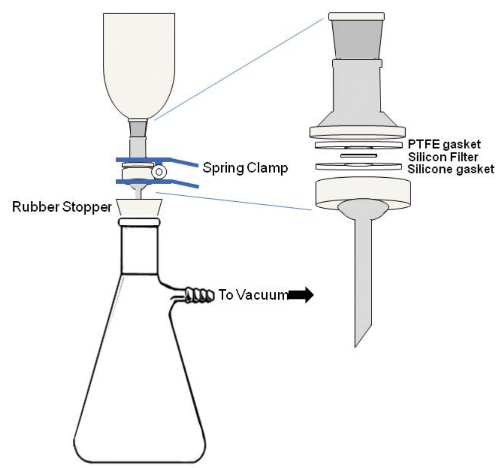


Figure 1. Apparatus for filtering microplastic particles from drinking water for vibrational spectroscopic analysis

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Two types of gaskets were used. A polytetrafluoroethylene (PTFE) gasket (1-inch diameter with an 8 mm diameter hole) was used on top of the silicon filter. These gaskets were sonicated for 50 minutes in ultra-filtered deionized water in an attempt to remove any particles from the filter itself. The gasket underneath the silicon filter was silicone. The use of the silicone gasket (1-inch diameter and 9 mm diameter hole) provided a better seal and the filter was less likely to break compared to using two PTFE gaskets. However, care must be taken to center the silicon filter on the silicone gasket to avoid contact between the backside of the silicon filter and the silicone gasket in the area exposed by the hole in the PTFE gasket. The reason for this is that the silicone gasket can leave some residue on the backside of the filter and if the filter is analyzed in transmission mode on an FTIR microscope these residues can be detected. Figure 2 shows a transmission map of the silicone filter and defines the usable area of the silicon filter that was not in contact with the silicone gasket. If the gasket and filters are properly centered then the residue should be outside the area occupied by the filtered particles (8 mm circular area). This is not an issue when analyzing the filter using Raman microscopy or FTIR microscopy in reflectance mode.

To be sure all the particles on the filter are coming from the bottled water it is important to avoid introducing additional particles from either the glassware or the environment. This was only partially successful, and particles could be detected on new filters after re-filtering previously filtered water. Controlling the conditions and careful treatment of the apparatus is crucial for reproducible and meaningful sampling.

The particles on the silicon filters can then analyzed using a Thermo Scientific™ Nicolet™ iN10 MX FTIR Imaging Microscope or a Thermo Scientific™ DXR2 Raman Microscope, or both. These instruments both have software options specifically designed for automated particle analysis. The Nicolet iN10 MX FTIR Imaging Microscope uses the Particle Wizard option in the Thermo Scientific™ OMNIC™ Picta™ Software and the DXR2 Raman Microscope uses the Particle Analysis option that is part of the Thermo Scientific™ Omnic™ Atlus Software and the Thermo Scientific™ Omnic™ for Dispersive Raman Software.

Summary

The Microparticle Sample Preparation Kit was developed to allow for the isolation of particles from dilute suspensions in liquids and subsequent vibrational identification of the particles using FTIR and Raman spectroscopy. The silicon filters were chosen to provide good visual images and allow for both transmission and reflectance FTIR analysis, as well as Raman analysis. A commercially available filter apparatus designed for 13 mm circular filters was adapted to use these rigid, square filters using gaskets to provide leak-free operation and protection for the thin, brittle silicon filters. Together with the iN10MX FTIR Imaging Microscope and the DXR2 Raman Microscope this kit provides a streamlined path for identifying a wide variety of particle contaminants.

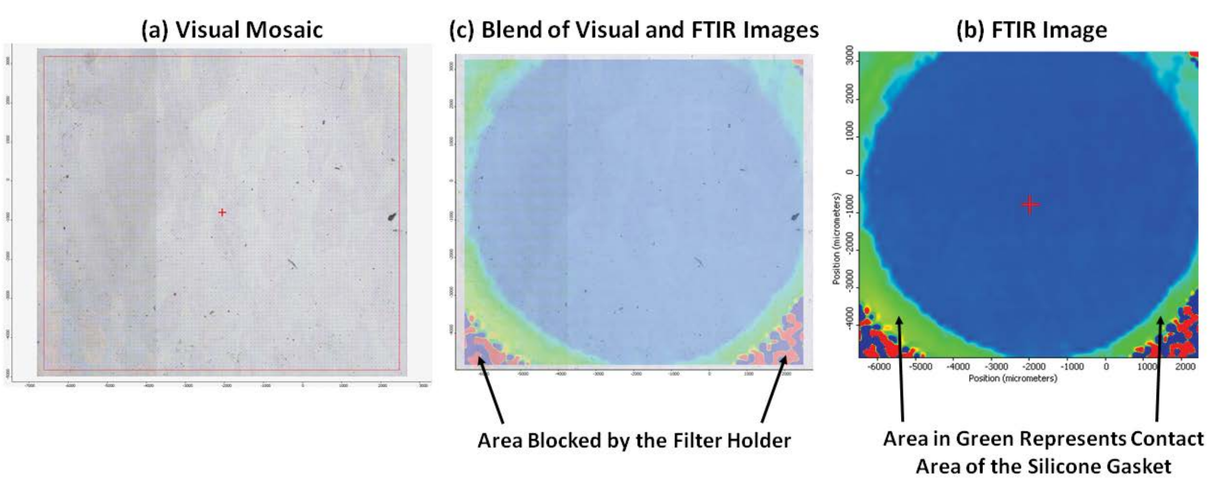

Figure 2. Illustrating the usable filter area with an FTIR transmission image of the filter. (a) The visual mosaic image, (b) The FTIR transmission image, and (c) A blend of the visual and FTIR images. The lighter blue area in the center of the image represents the usable filter area for transmission FTIR analysis.

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