Compressing the workflow of Infrared Microscopy with intelligent automation

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Infrared (IR) spectroscopy is widely adopted as a technique for the identification of materials. IR microscopy allows spectra to be measured on small samples down to a limit of a few microns, with this limit dictated by the diffraction limit of the infrared radiation. Spectra can be measured on an IR microscope using transmission, reflectance or ATR techniques. There are a wide range of applications for IR microscopy within the chemical and polymer industries, requiring identification of the individual materials present to fully characterise the product. IR imaging of samples can be performed, however, these systems are more expensive than standard IR microscopes and the experiments can be significantly longer and require significant data processing to extract the useful information about the sample. Automation of a series of IR microscopy experiments is described covering the most common types of usage of these systems.

An infrared microscopy experiment can consist of multiple measurements on a sample at multiple points on the sample and the regions of interest (ROI) will almost certainly be of different types, sizes and shapes. The most common ROIs encountered in an IR microscopy experiment are :

Particles - either discrete particles or contaminants
Layers - multilayer laminates or paint chips
Inclusions - embedded materials, contaminants or product defects

Using a completely manual IR microscope the user could move to each of the points in turn and measure an IR spectrum at that point. This would be a slow and labor-intensive operation. The addition of an automated XY stage and the ability to select a series of points for measurement will speed up the experiment significantly. However, there may be a significant number of points to measure and the measurement at each point needs to be optimised to get the best quality data. The optimisation includes obtaining the best sensitivity for the ROI and avoiding stray light by non-optimum aperture settings. The PerkinElmer Spotlight 200i IR Microscope software contains intelligent software routines to assist the user in finding the ROI within a sample and optimising the measurement for those ROI. The experiment workflow will consist of the same steps, differing only in the Analyse Image step :

1. Collect a visible image (survey) of the sample
2. Analyse Image (Detect particles, find layers or find inclusions)
3. Collect and analyse IR data

The visible image can be the camera field of view or can be a visible image survey collected over a much larger area of the sample, up to several mm.

Detecting Particles and Inclusions

A typical sample will consist of many particles with a range of different sample shapes and sizes. To optimise the measurement for each particle requires the aperture size to be maximised avoiding

overaperturing as that would introduce stray light into the spectra. The Detect Particles routine will analyse the visible image and try to detect particles present. For each particle detected the software will determine the largest rectangle that could fit within the particle and set the aperture to that size. The software will then automatically scan backgrounds and then sample spectra for all the particles matching the appropriate apertures. The software can also automatically search against spectral libraries to identify the particles, color coding them according to their identity as shown in Figure 1. The only difference with an inclusion is that it is embedded within a sample matrix. In such cases, the software identifies a suitable region of matrix material and will automatically subtract the matrix spectrum from the inclusion + matrix spectrum.

Detecting Layers

A typical sample, such as a multilayer food packaging, will consist of multiple layers of varying thicknesses of different materials, each layer having a functional purpose. A common experiment would perform a linescan measuring spectra across the multilayers. The spacing of the measurements and the aperture size would need to be set to the minimum width of an expected layer, thus requiring a significant number of spectra to be collected in the experiment and with sub-optimum apertures. The Detect Layers routine will detect the layers present, optimise the aperture setting and collect a spectrum for each layer, with identification of the layers if required, see Figure 2. Layer thicknesses are also reported.

Conclusions

Use of an automated IR microscope and intelligent ROI detection routines generates high quality spectra and results for samples significantly faster than manual operation or complete sample imaging. The routines optimise the measurements giving the best sensitivity without allowing spectral artefacts. The system allows automated spectral measurement in any of the sampling modes; transmission, reflectance or ATR. IR microscopy that has been previously considered to be complex requiring skilled operators to perform the experiments is now operable as a routine instrument.

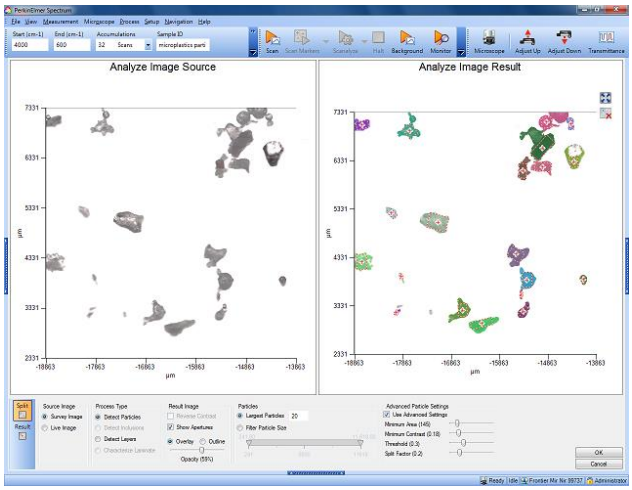

Figure 1. Use of Detect Particles to find microplastics on a membrane filter

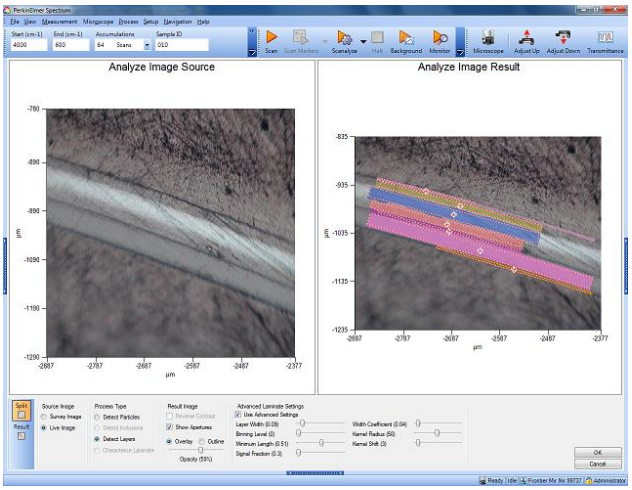

Figure 2. Detection of layers in a multilayer food packaging material