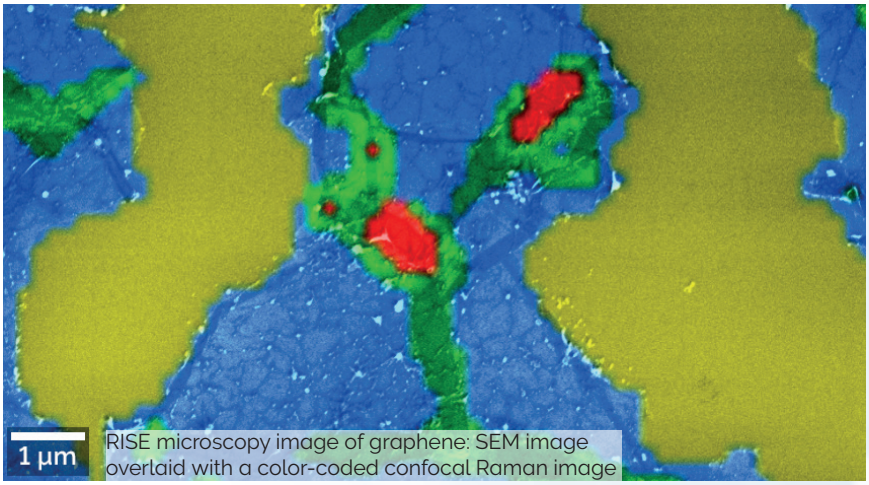
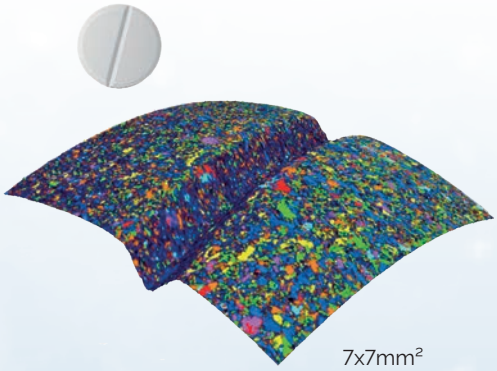
APPLICATION NOTE

Correlative Confocal Raman Microscopy

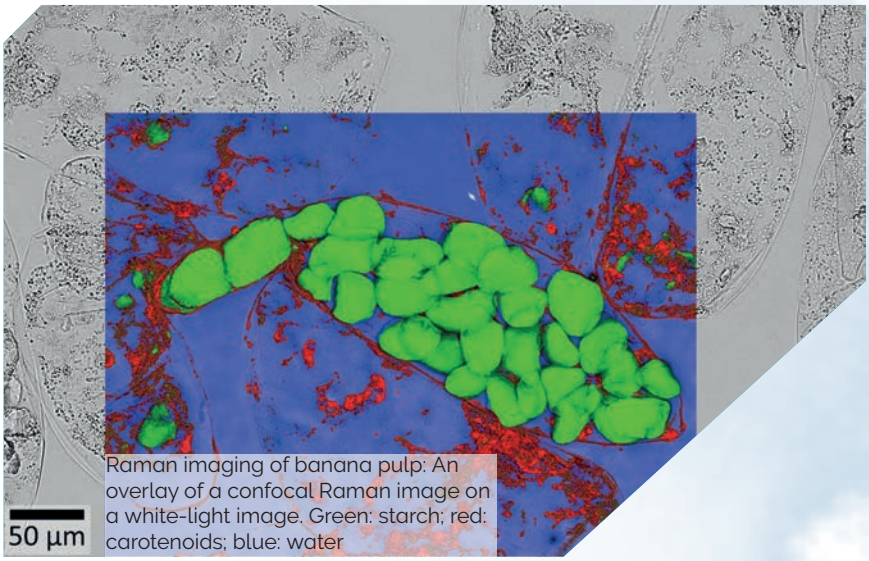
A powerful tool for non-destructive, label-free chemical and structural characterization



RISE microscopy image of graphene: SEM image overlaid with a color-coded confocal Raman image



Topographic Raman microscopy image of a pharmaceutical tablet



Raman imaging of banana pulp: An overlay of a confocal Raman image on a white-light image. Green: starch; red: carotenoids; blue: water

Confocal Raman microscopy is a uniquely effective method for analyzing the chemical composition of heterogeneous samples on the sub-micrometer scale.

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application note

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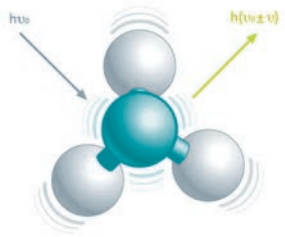
The Raman principle

The Raman effect is based on the inelastic scattering of light by the molecules of gaseous, liquid or solid materials. The interaction of a molecule with photons causes vibrations of its chemical bonds, leading to specific energy shifts in the scattered light. Thus, any given chemical compound produces a particular Raman spectrum when excited and can be easily identified by this individual "fingerprint."



Raman spectroscopy is a well-established, label-free and non-destructive method for analyzing the molecular composition of a sample.

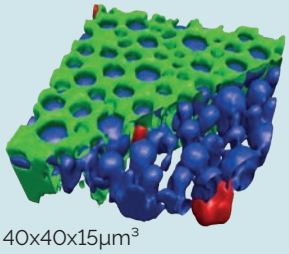
Raman imaging

In Raman imaging, a confocal microscope is combined with a spectrometer and a Raman spectrum is recorded at every image pixel. The resulting Raman image visualizes the distribution of the sample's compounds. Due to the high confocality of WITec Raman systems, volume scans and 3D images can also be generated.

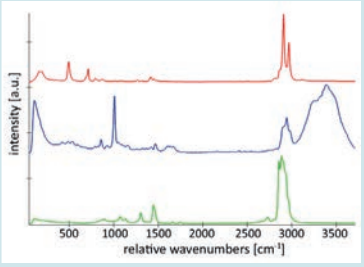
Technologies and Applications

Confocal Raman microscopy for 3D imaging

Volume scans and 3D images are valuable for providing information about the dimensions of objects or the distribution of a certain compound throughout a sample. Here, a 3D Raman image of a pharmaceutical emulsion consisting of water (blue), oil (green) and some impurities (red) is shown. The oil was partially removed from the image for a better sample overview.

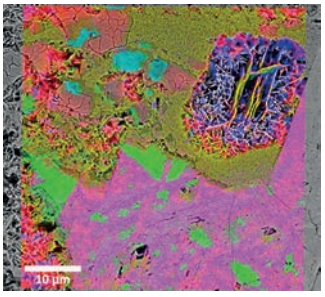


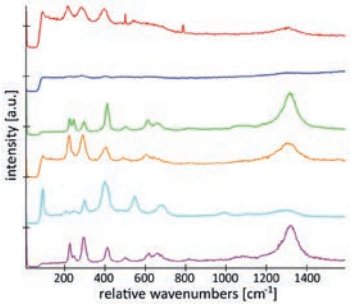
40x40x15µm³



Raman - SEM (RISE™) imaging in geology

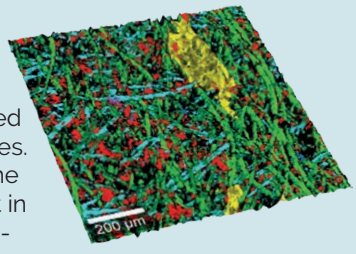
Geological samples are often very complex. With confocal Raman imaging, the spatial distribution and association of components or mineral phases can be evaluated. In this iron-rich sample, several crystal forms of hematite and goethite were identified by their Raman spectra. From these spectra, a Raman image was generated and overlaid with a scanning electron microscopy (SEM) image. Both measurements were performed with a combined Raman-SEM system (RISE = Raman Imaging and Scanning Electron microscopy).

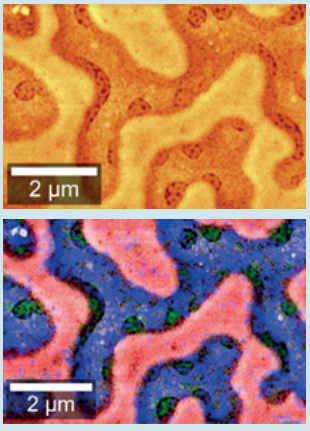




Topographic Raman imaging with TrueSurface™

With WITec's TrueSurface microscopy option confocal Raman imaging can be guided by the surface topography of even rough, inclined or irregularly-shaped samples. A sensor actively monitors the sample surface and keeps it in focus. As examples, topographic Raman images of a pain relief tablet (title page) and of paper are shown.





Correlative Raman-AFM imaging

Combining Raman imaging and atomic force microscopy (AFM) in one instrument makes it possible to obtain chemical and structural information from the same sample area at the highest resolution. The AFM phase image of a polymer blend (top) was overlaid with the Raman image. Thus, the sample's molecular components and its physical features could be correlated (bottom).