application note

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Food Analysis with Confocal Raman Microscopy

In the food industry, various ingredients, additives and bio-polymers such as emulsifiers, stabilizers, carbohydrates or thickeners are commonly used in order to optimize the texture or the flavor of food. The distribution and the microstructure of the ingredients strongly determine the properties of the final product. Therefore, research and development as well as quality control require powerful analytical tools for studying the distribution of the various compounds in the food.

As Confocal Raman Microscopy provides the ability to nondestructively image the chemical composition of a sample and requires only minimal sample preparation, it can be used for three-dimensional analysis of emulsions, suspensions or solids. In this study, Confocal Raman Microscopy is used to investigate an instant gravy thickener, an emulsion, and white chocolate.

Confocal Raman Microscopy

Raman spectroscopy has become widely used for the characterization of materials in terms of chemical composition. Optical microscopy, on the other hand, is capable of providing spatial resolution down to 200 nm using visible light excitation. In a confocal microscope, only light from the focal plane is detected while out of focus light is rejected, thus providing depth resolution and a strongly reduced background signal. Images are recorded point by point and line by line while scanning the sample through the excitation focus. With this technique, the specimen can be analyzed in steps along the optical axis and even a depth profile or 3D image can be generated. As the images consist of tens of thousands of spectra, very short acquisition times for a single Raman spectrum are essential. With the Confocal Raman Microscope alpha300 R used for this study, the acquisition time for a Raman spectrum is in the range of 10 - 100 milliseconds.

Experiment and Results - Instant Gravy Thickener

In the first experiment, instant gravy thickener particles have been imaged with a scan range of 50 x 50 µm and 150 x 150 pixels (22,500 spectra, 70 ms/spectrum) using a 532 nm Nd:Yag laser for excitation. Fig. 1a shows the color-coded Raman image obtained by evaluating dedicated peak characteristics of the corresponding spectra shown in fig. 1b. The red region shows the distribution of starch surrounded by an excipient (blue) whereas lipids (yellow) are located between the starch grains together with other additives (green).

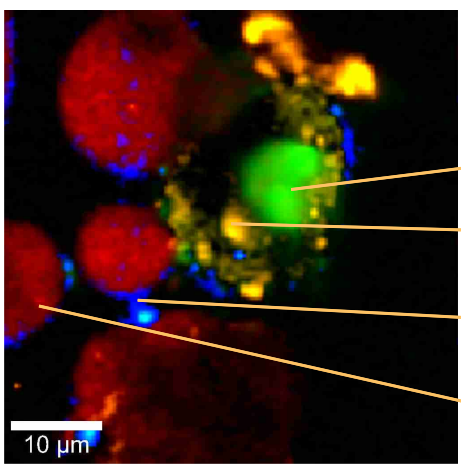

Fig. 1a: Raman image of instant gravy thickener particles. (scan range: 50 x 50 µm, 150 x 150 pixels, 22500 spectra, 70 ms/spectrum, excitation: 532 nm Nd:Yag)

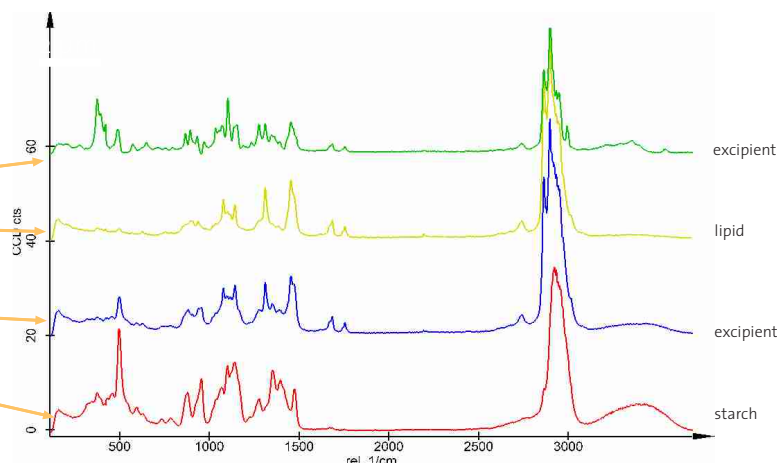

Fig.1b: Corresponding Raman spectra.

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Emulsions for food production

In a second experiment, an emulsion used in food production was imaged with a scan range of 30 x 30 µm and 120 x 120 pixels (14,400 spectra, 30 ms/spectrum). Such emulsions are added to the food mixture in order to introduce or adjust dedicated properties in the final products. The emulsion contains a fatty matrix, an aqueous phase and the emulsifier polyglycerol polyricinoleate (PGPR). After analyzing the spectral characteristics of the components, figure 2 shows their distribution, clearly revealing that the emulsifier PGPR (yellow) aggregates as an interface between the water droplets (blue) in the fatty matrix (green & red).

Chocolate

In the final experiment, white chocolate was investigated. The scan range was 50 x 50 µm with 150 x 150 pixels (=22,500 spectra) and the measurement used an integration time of 40 ms for each spectrum. One can clearly see a distinct phase separation in the chocolate material. Saccharose (blue) and additive particles (red) are embedded in a fatty matrix (green). The size of the saccharose particles vary between 650 nm and 10 µm.

Conclusion

Confocal Raman Microscopy is a powerful tool for the analyis of the chemical composition of heterogeneous samples on the sub-µm scale. In this application note it was demonstrated that the distribution of various chemical compounds in instant gravy thickener, emulsions and chocolate products could be successfully visualized, leading to a more thorough understanding of the product and the production process.

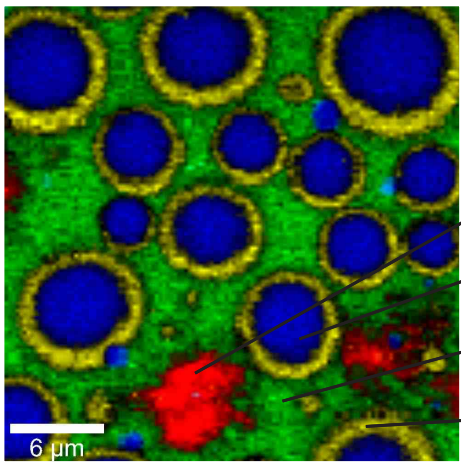

Fig. 2a: Raman Image of an emulsion used in food production. Scan range: 30 µm x 30 µm, 120x120 pixels (=14400 spectra, 532 Nd:Yag Laser for excitation, 100 x oil objective (NA=1.25). Red + green: fatty matrix, blue: aqueous phase, yellow: emulsifier PGPR

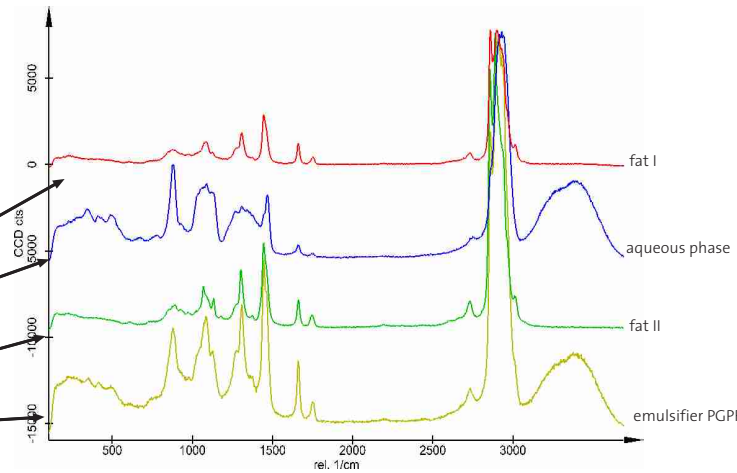

Fig. 2b: Corresponding spectra of the compounds contained within the emulsion.

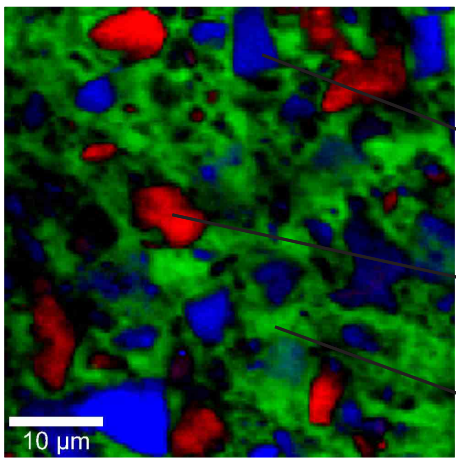

Fig. 3a: Raman image of white chocolate, (scan range: 50 x 50 µm, 150 x 150 pixels, 22500 spectra, 40 ms/spectrum, excitation: 532 nm Nd:Yag), . Saccharose (blue) and additives (red) particles are embedded in a fatty matrix (green).

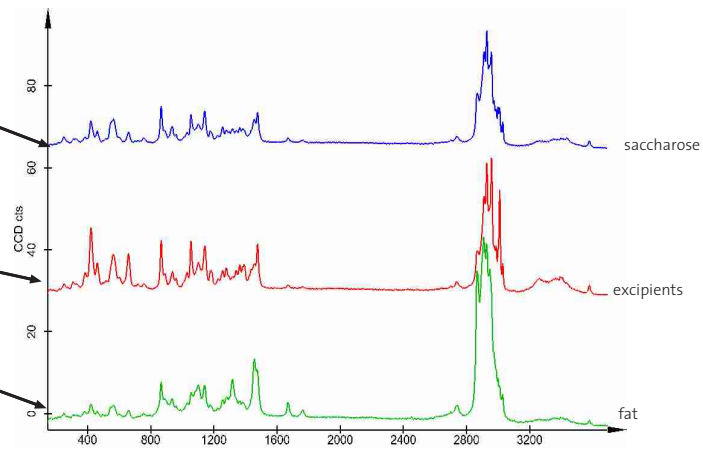

Fig. 3b: Corresponding spectra