Fundamentals of Label-Free Spectromicroscopy

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The fundamentals of label-free spectromicroscopy will be reviewed. Spectromicroscopy is a combination of spectroscopy and microscopy used for going beyond simple imaging of samples by optical microscopy. This review includes the fundamentals of optical microscopy and its limitations as well as novel techniques going beyond, such as and X-ray microscopy, photoelectron microscopy, and atomic force microscopy-based techniques, including optical near-field microscopy, tip-enhanced Raman techniques, and photothermal expansion. Then, important fundamentals of spectroscopy are reviewed as a foundation for the combination with microscopy techniques. This includes the required radiation sources, i.e. lasers and synchrotron radiation, and the processes that are probed, including optical absorption, fluorescence, inelastic light scattering, as well as the emission of electrons and ions. It will also be explained what the difference between labeled and label-free approaches in spectromicroscopy is, so that this lecture serves as an introduction for applications of these techniques, which are elucidated in the subsequent lecture.

Applications of Label-Free Spectromicroscopy

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Important applications of spectromicroscopy will be presented, where preference is put on label-free approaches. It will be discussed where the strength and weaknesses of each approach are found and how to use spectromicroscopy in fundamental and applied research, depending on the intended use in materials research and life sciences. Emphasis will be put on those approaches that go beyond the diffraction limit of optical microscopy showing distinct details which cannot be visualized by optical microscopy, finally reaching the molecular scale. Detailed examples from X-ray microscopy, Raman-based approaches, optical near-field microscopy, photothermal expansion, and photoelectron microscopy are shown besides state-of-the-art results from fluorescence-based spectromicroscopy (super-resolution microscopy) beyond the diffraction limit.