

## *Strategies of FT-IR Spectroscopic Imaging*

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Spatially resolved Fourier Transform Infrared (FT-IR) microspectrometry of tissue thin sections in combination with digital imaging techniques show a great promise for in-vivo and ex-vivo medical diagnosis. This statement is based on the fact that infrared spectroscopic methods obtain objective information on the chemical composition of histological samples, and that diseases are linked to changes in the molecular composition of cells and tissues. False color images produced by infrared spectroscopic methods can be interpreted also by non-spectroscopists and are directly comparable to outcomes of standard histological staining protocols.

In the presentation we will discuss strategies of FT-IR spectroscopic imaging on the basis of experimental data which were obtained on histological thin sections of melanoma, colon adenocarcinoma, hepatocellular carcinoma, prostate carcinoma and hamster brain. For data acquisition either the infrared mapping methodology, i.e. utilizing an IR microscope equipped with a computer controlled xy stage, or the HgCdTe (MCT) focal plane array detector technique were applied. Both methods produce large hyperspectral data cubes which may be represented as a series of chemical images at distinct wavelengths or as a matrix of spectra as a function of spatial position.

For infrared spectroscopic tissue characterization, a great variety of different imaging methodologies has been proposed. The most simple and easy to interpret approach to extract the histologically relevant information from the spectral hypercubes is based on the idea to convert a defined spectral parameter (absorbance, band position) to color intensities and plot as a function of the spatial position (chemical mapping or functional group mapping). Spectral parameters can be identified and optimized by feature selection methods, however most of the spectroscopic information is lost in the univariate imaging procedure of chemical mapping. To circumvent this problem and to increase image contrast and certainty of the spectroscopic diagnosis, pattern recognition techniques such as cluster analysis, principal component analysis, or artificial neural network analysis were proposed to apply. These multivariate methodologies are based on the concept to derive for an entire spectrum one single parameter which describes the degree of similarity of spectral pattern. As in the example of chemical mapping, these parameters can be color scaled and plotted against the spatial coordinates. In the presentation we will give an overview of the distinct uni- and multivariate image reconstruction methods and discuss the suitability of these techniques for practical applications.