High Resolution Imaging of Single Cells: Raman, FTIR, AFM and SNOM

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Endothelium plays an important role in cardiovascular system and regulates vascular homeostasis and its dysfunction is involved in several lifestyle diseases. The general aim of the research is to find biochemical features of endothelial dysfunction that cannot be found by other methods.

The comparison between various spectroscopic and microscopic techniques and their advantages as well as limitations in biomedical samples analysis is presented. In particular, *in situ* identification of selected compounds is discussed. Spectroscopic measurements are combined with AFM, SNOM, histochemical staining and fluorescence microscopy, if possible.

Confocal Raman spectroscopy and FT-IR imaging were used to monitor a molecular composition occurring in a single live human aorta endothelial cell. Based on Raman spectrum and using a chemometric approach it is possible to investigate biochemical changes induced by stress or pharmacological treatment. Generally, FTIR spectroscopy measurement in transmission is realized at the resolution of several microns, but with an objective of higher magnification the subcellular studies can be successfully performed.

Raman spectroscopy applied for cells isolated from the liver tissue enabled their specific characterization. In addition, this methodology can support other approaches for tracking changes at the cellular level due to pathology development. Here, an application of Raman confocal mapping for studies of cells isolated from healthy mice liver is shown. Hepatocytes, HSC, Kupffer and endothelial cells were identified and subjected for the further studies. With the use of chemometric tools we present a complex spectral characteristic of each investigated cell.

Atomic Force Microscopy (AFM) and Scanning Near-field Optical Microscopy (SNOM) are techniques providing images of structures (also biological materials) with nanometric optical resolution (about 50-60 nm) and topographic information at the same time. In this work the capabilities of both, AFM and SNOM in transmission configuration, to image endothelial cells are shown.

References

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