

# ***Tumor cell identification using Raman spectroscopy in combination with optical trapping and microfluidics***

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The progress of the research initiative “Jena Cell Identification Group” (JenZIG) is presented. Scope is the combination of biophotonic tools such as Raman spectroscopy with micromanipulation and microfluidic environment. A model system consists of cells that can be found in peripheral blood of tumor patients. Lymphocytes and erythrocytes were extracted from blood samples. Breast carcinoma derived tumor cells (MCF-7, BT-20) and acute myeloid leukemia cells (OCI-AML3) were grown in cell cultures.

Raman images were collected from dried cells on calcium fluoride slides<sup>1</sup>. Support vector machines (SVM) classified 99.7% of the spectra to the correct cell type. Subsequently, the model was applied to identify cells from a mixed population. The classification was validated by fluorescence staining after Raman measurements.

Raman spectra were collected from cells in aqueous buffer<sup>2</sup>. A laser emitting 785 nm light was used for optical trapping of single cells and for excitation of the Raman spectrum. SVM distinguished 1210 spectra of tumor and normal cells with a sensitivity of >99.7% and a specificity of >99.5%. The correct cell types were predicted with an accuracy ≈92%.

A novel microfluidic glass chip was designed to inject single cells, modify the flow speed, accommodate fibers of an optical trap and sort single cells after Raman based identification<sup>3</sup>. The optical trap was realized by two 1070 nm single mode fiber lasers. Whereas the integrated microchip setup used 514 nm for excitation of Raman spectra, a quartz capillary setup excited spectra with 785 nm laser wavelength. Classification models were trained using linear discriminant analysis with accuracies that were comparable with previous Raman experiments<sup>1,2</sup>.

Modulated wavelength Raman spectroscopy has been applied to suppress fluorescence background in Raman spectra of single cells in aqueous buffer [4]. The laser excitation wavelength of 785 nm was modulated with a frequency of 40 mHz by 0.6 nm. Altogether 840 spectra were collected at several days to compensate day-to-day variations of the system and the progressive fixation of cells. The classification accuracy of a SVM was 93.3% for cancer vs. normal cells and 81.2% for the recognition of the cell type.

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## References

- [1] U. Neugebauer, J.H. Clement, T. Bocklitz, C. Krafft, J. Popp, *J. Biophoton.* **3**, 579-587 (2010).
- [2] U. Neugebauer, T. Bocklitz, J.H. Clement, C. Krafft, J. Popp, *Analyst* **135**, 3178-3182 (2010).
- [3] S. Dochow, C. Krafft, U. Neugebauer, T. Bocklitz, T. Henkel, G. Mayer, J. Albert, J. Popp, *Lab Chip* **11**, 1484-1490 (2011).
- [4] A.C. de Luca, M. Mazilu, A. Riches, C.S. Herrington, K. Dholakia, *Anal. Chem.* **82**, 738-745 (2010).