Biophotonics and optofluidic

Department of Microphotonics

Institute of Scientific Instruments
The Czech Academy of Sciences

THEMATIC RESEARCH FOCUS

Research area
- Optics
- Laser spectroscopy
- Biophotonics
- Nanophotonics
- Lab-on-a-chip

Excellence
- Applications of focused laser beams (optical tweezers, optical cell sorters)
- Raman micro-spectroscopy combined with optical manipulations (Raman tweezers) for detection and identification of bacteria using techniques of AI
- Analysis of the relationship between a bacteriophage and its host bacterium
- Manufacturing of tailored microfluidic chips

Mission
Our main goal is to design and develop analytical techniques based on microfluidics and Raman spectroscopy to solve emerging tasks targeted on basic and applied biological research ranging from microorganisms to enzymes.

UP-TO-DATE ACTIVITIES

Research orientation
- Optical trapping
- Characterization of living microorganisms (e.g. bacteria, yeast and algae cells) using Raman microspectroscopy, Raman tweezers and microfluidic chips
- Optical monitoring of chemical reactions running in emulsion droplets and lab-on-a-chip

Main capabilities
Basic research
- Determination of selected chemical content in living microorganisms using Raman microspectroscopy combined with optical micromanipulation and microfluidic techniques

Applied research
- Design and manufacturing of microfluidic chips
- Monitoring of polymers inside the cells with industrial biotechnological applications
- Identification of bacteria with clinical applications

Innovations
- Tailored microfluidic chips for clinical investigations

Subfields of group activities
- Optical microscopy
- Microtechnology, nanotechnology
- Biophotonics

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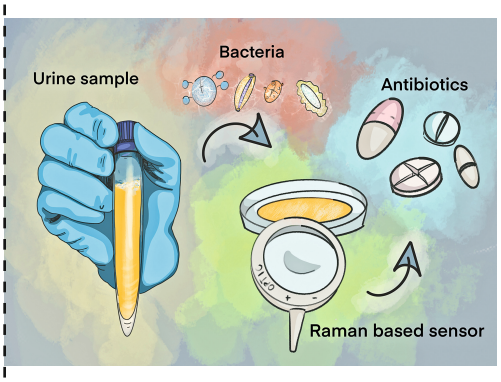


Imagine this wishful scenario in a clinical practice: A sick person enters a hospital emergency ward and the doctor collects a urine specimen from this person. Consequently, in an ideal case, pathogens are identified quickly and appropriate antibiotics are prescribed. Thus, we aim to introduce Raman spectroscopy as potential method of choice for POC measurement which excels in speed and sensitivity in clinical setting.

- Cell biology
- Biochemistry
- Laser spectroscopy
- Microuidics
- Lab-on-a-chip systems
- Microbiology

KEY RESEARCH EQUIPMENT

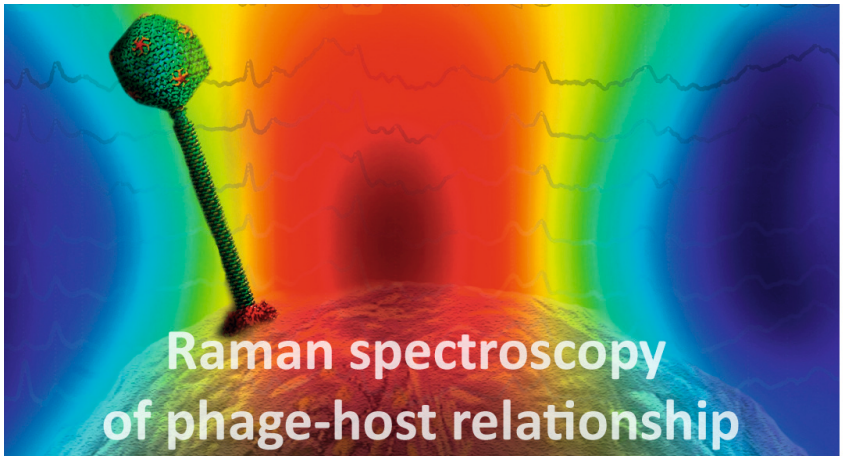
List of devices

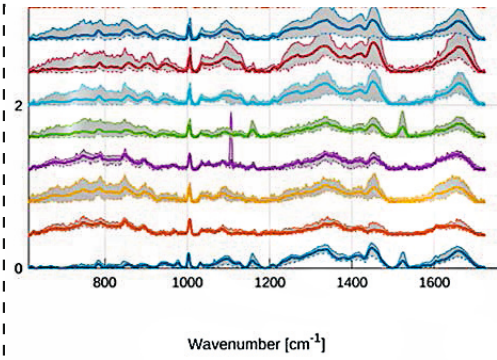
- Renishaw In Via Reflex Raman microspectrometer
- Raman tweezers made by the research team
- Optical cell sorters made by the research team
- Fast CCD cameras (thousands fps)

SELECTED PUBLICATIONS

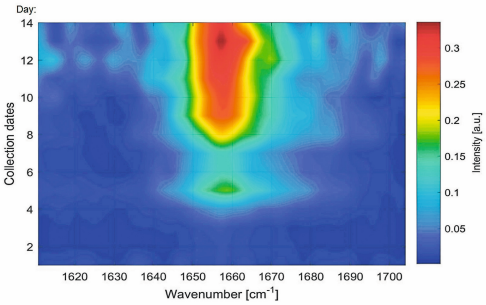
Identification and diferentiation of bacteria, bacteriophage, and yeast

- K. Rebrošová, S. Bernatová, M. Šiler, M. Uhlirova, O. Samek, J. Ježek, V. Holá, F. Růžička, P. Zemánek : *"Raman spectroscopy—a tool for rapid differentiation among microbes causing urinary tract infections."* Analytica Chimica Acta, **1191**, 339292 (2022).
- O. Samek, S. Bernatová, F. Dohnal: *"The potential of SERS as an AST methodology in clinical settings."* Nanophotonics, 2537–2561 (2021).
- S. Bernatová, K. Rebrošová, Z. Pilát, M. Šerý, A. Gjevik, O. Samek, J. Ježek, M. Šiler, M. Kizovský, T. Klementová, V. Holá, F. Růžička, P. Zemánek: *"Rapid detection of antibiotic sensitivity of Staphylococcus aureus by Raman tweezers."* Eur. Phys. J. Plus, **136**, 233 (2021).
- A. Němcová, D. Gonová, O. Samek, M. Sipiczki, E. Breierová, I. Márová: *"The Use of Raman Spectroscopy to Monitor Metabolic Changes in Stressed Metschnikowia sp. Yeasts."* Microorganisms, **9**, 277 (2021).
- Z. Pilát, A. Jonáš, J. Pilátová, T. Klementová, S. Bernatová, M. Šiler, T. Maňka, M. Kizovský, F. Růžička, R. Panůček, U. Neugebauer, O. Samek, P. Zemánek: *"Analysis of bacteriophage-host interaction by Raman tweezers."* Analytical Chemistry, **92**, 12304–12311 (2020).
- K. Rebrošová, M. Šiler, O. Samek, F. Růžička, S. Bernatová, J. Ježek, P. Zemánek, V. Holá: *"Identification of ability to form biofilm in Candida parapsilosis and Staphylococcus epidermidis by Raman spectroscopy."* Future Microbiology, **14**, 509–517 (2019)
- S. Bernatová, Donato M. Grazia, J. Ježek, Z. Pilát, O. Samek, A. Magazzu, OM. Maragò, P. Zemánek, PG. Gucciardi: *"Wavelength-Dependent Optical Force Aggregation of Gold Nanorods for SERS in a Microfluidic Chip."* J. Phys. Chem. C, **123**, 5608–5615 (2019)



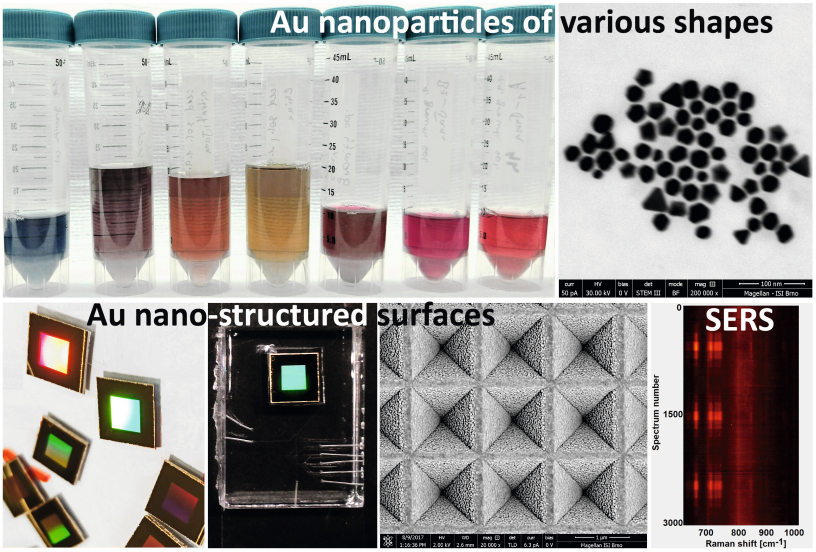


Selected Raman spectra of Staphyloccocus species from our spectral library used for identification employing techniques of artificial inteligence.



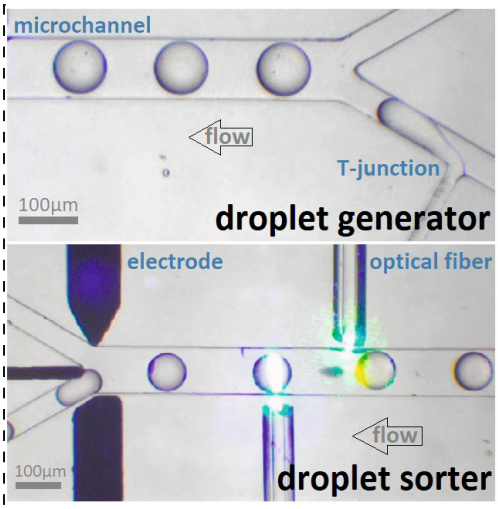
Spectral map of Metschnikowia andauensis, plotted using Raman signal intensities of unsaturated oil indicators during 14 days of cultivation. The map monitors the unsaturated oil content within the cells.

We have a system for Raman spectroscopic analysis of the relationship between a bacteriophage and its host bacterium. Raman spectroscopy combined with optical tweezers provides real-time, non-invasive measurement of the biochemical changes that occur in a single bacterium during the various phases of the phage infection.



We create Au nanoparticles of various sizes and nanostructured surfaces optimized for surface-enhanced Raman spectroscopy (SERS). We use SERS for highly sensitive detection of extremely dilute analytes, such as pesticide residues, secondary metabolites, or individual bacteria.

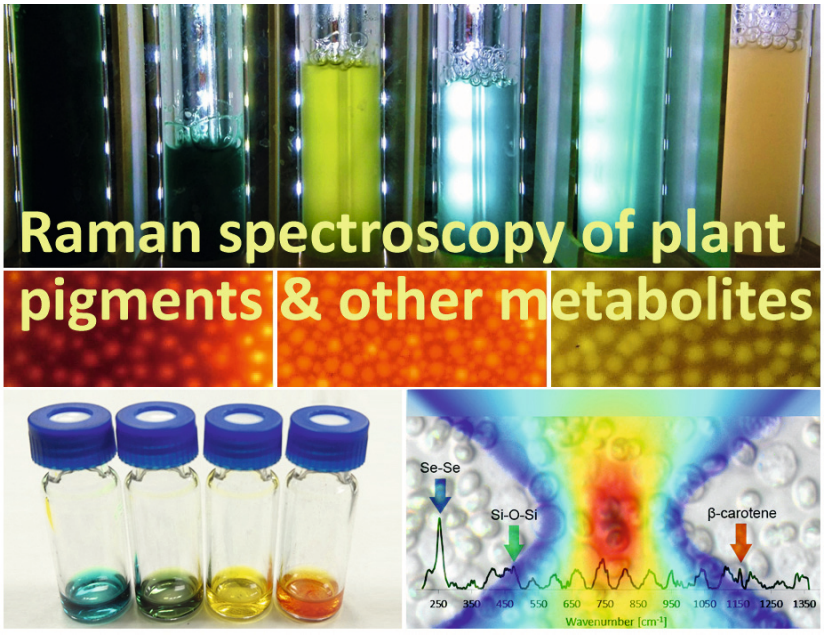
- O. Samek, K. Mlynáriková, S. Bernatová, J. Ježek, V. Krzyžánek, M. Šiler, P. Zemánek, F. Růžička, V. Holá, M. Mahelová: *"Candida parapsilosis biofilm identification by Raman spectroscopy"*. Int. J. Mol. Sci. **15**, 23924–23935, 2014.
- K. Mlynáriková, O. Samek, S. Bernatová, F. Růžička, J. Ježek, A. Haroniková, M. Šiler, P. Zemánek, V. Holá: *"Influence of culture media on microbial fingerprints using Raman spectroscopy"*. Sensors **15**(11), 29635–29647, 2015.
- S. Bernatová, O. Samek, Z. Pilát, M. Šerý, J. Ježek, P. Jákl, M. Šiler, V. Krzyžánek, P. Zemánek, V. Holá, M. Dvořáčková, F. Růžička: *"Following the mechanisms of bacteriostatic versus bacericidal action using Raman spectroscopy"*. Molecules **18**(11), 13188–13199, 2013.



In digital microfluidics, emulsion droplets are used as miniature test tubes, each with a separate chemical reaction, or a living cell. The droplets are generated in the T-junction, incubated, analyzed via optical fiber and automatically sorted (based on fluorescence or Raman spectra) into different output channels by electric pulses. We use droplets for analysis of enzymatic reactions involving dehalogenases.

Applications to microorganisms containing polymers and algae

- M. Kizovský, Z. Pilát, M. Mylenko, P. Hrouzek, J. Kuta, R. Skoupý, V. Krzyžánek, K. Hrubanová, O. Adamczyk, J. Ježek, S. Bernatová, T. Klementová, A. Gjevik, M. Šiler, O. Samek, P. Zemánek: *"Raman Microspectroscopic Analysis of Selenium Bioaccumulation by Green Alga Chlorella vulgaris."* Biosensors, **11**, 115 (2021).
- S. Obruča, P. Sedláček, F. Mravec, V. Krzyžánek, J. Nebesářová, O. Samek, D. Kucera, P. Benešová, K. Hrubanová, M. Milerová, I. Márová: *"The presence of PHB granules in cytoplasm protects non-halophilic bacterial cells against the harmful impact of hypertonic environments"*. New Biotechnology **39**, 68–80, 2017.

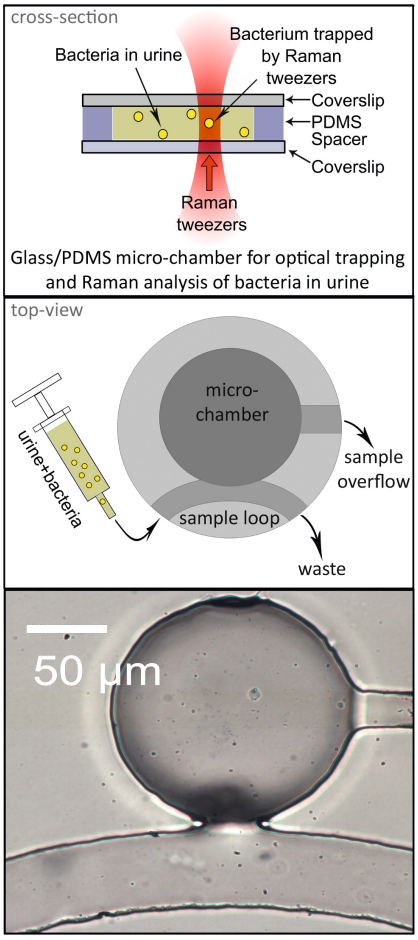


We have know-how in Raman spectroscopy of algal and plant pigments and various other metabolites and biomolecules in general. We specialize in development of complex instrumentation hardware, software and methods for specific Raman-based analytical tasks.

– Z. Pilát, A. Jonáš, J. Ježek, P. Zemánek: *"Effects of Infrared Optical Trapping on Saccharomyces cerevisiae in a Microfluidic System".* Sensors **17**, 2640, 2017.
– O. Samek, S. Obruca, M. Siler, P. Sedlacek, P. Benesova, D. Kucera, I. Marova, J. Jezek, S. Bernatova, P. Zemanek: *"Quantitative Raman Spectroscopy Analysis of Polyhydroxyalkanoates Produced by Cupriavidus necator H16".* Sensors **16**, 2016

Optofluidic applications

– Z. Pilát, M. Kizovský, J. Ježek, S. Krátký, J. Sobota, M. Šiler, O. Samek, T. Buryška, P. Vaňáček, J. Damborský, Z. Prokop, P. Zemánek: *"Detection of chloroalkanes by surface-enhanced Raman spectroscopy in microfluidic chips."* Sensors, **18**, 3212 (2018).
– T. Buryška, M. Vašina, F. Gielen, P. Vaňáček, L. Van Vliet, J. Ježek, Z. Pilát, P. Zemánek, J. Damborský, F. Hollfelder, Z. Prokop: *"Controlled oil/water partitioning of hydrophobic substrates extending the bioanalytical applications of droplet-based microfluidics."* Analytical Chemistry, **91**, 10008–10015 (2019).
– A. Jonáš, Z. Pilát, J. Ježek, S. Bernatová, P. Jedlička, M. Aas, A. Kiraz, P. Zemánek: *"Optically transportable optofluidic microlasers with liquid crystal cavities tuned by the electric field."* ACS Applied Materials & Interfaces, **13**, 50657–50667 (2021).
– A. Jonáš, Z. Pilát, J. Ježek, S. Bernatová, T. Fořt, P. Zemánek, M. Aas, A. Kiraz: *"Thermal tuning of spectral emission from optically trapped liquid-crystal droplet resonators."* J. Opt. Soc. Am. B, **34**, 1855–1864 (2017).
– M. Šiler, J. Ježek, P. Jákl, Z. Pilát, P. Zemánek: *"Direct measurement of the temperature profile close to an optically trapped absorbing particle."* Optics Letters, **41**, 870–873 (2016).
– Z. Pilát, S. Bernatová, J. Ježek, J. Kirchhoff, A. Tannert, U. Neugebauer, O. Samek, P. Zemánek: *"Microfluidic cultivation and laser tweezers Raman spectroscopy of E. coli under antibiotic stress."* Sensors, **18**, 1623 (2018).



We use microfluidic systems for optical trapping and Raman spectroscopy of individual bacteria in clinical samples (e.g. urine) or in cultivation media. The cells can be automatically identified, sorted and separated based on their Raman spectra.

M AIN COLLABORATING PARTNERS

Collaboration with academic partners

– Brno University of Technology (Brno, CZ)
– Consiglio Nazionale delle Ricerche (Messina, IT)
– Institute of Experimental Physics, Slovak Academy of Sciences (Košice, SK)
– Masaryk University (Brno, CZ)
– Universität für Bodenkultur (Wien, Austria)
– University of Graz (Graz, Austria)
– University of Life Sciences (As, Norway)
– University of Naples Frederico II (Naples, IT)
– University of Jena (Jena, Germany)
– IPHT Jena (Jena, Germany)

Collaboration with companies

– Photon Systems Instruments (Drásov, CZ)
– Meopta (Prerov, CZ)

EXPECTATIONS

Offers

– We offer collaboration in the areas of our expertise
– Partnership in international projects
– Custom manufacturing of microfluidic chips

Requirements

We would like to cooperate with academic partners as well as companies in the fields of optics, biophotonics, biotechnology, microtechnologies and applications of microfluidic chips.