PHD PROJECT DESCRIPTION

Project title:

Development of novel LDI-MS-based approaches for rapid lipid profiling

1.1. Project goals

The project aims to develop novel LDI-MS-based approaches for rapid lipid profiling, with particular emphasis on nanostructured NALDI plates synthesized by physical vapour deposition. The project addresses the need for faster analytical methods that could simplify or partially replace classical lipidomic workflows based on GC-MS and HPLC-MS, which often require time-consuming sample preparation, extraction, derivatization or chromatographic separation. The developed approach will focus on obtaining rapid lipid fingerprints from various chemical and biological matrices with minimal sample preparation. Particular attention will be paid to method sensitivity, reproducibility, selectivity and the ability to detect lipid-profile changes related to sample origin, quality, degradation, adulteration, biological stress or disease-related processes. The expected outcome is a flexible analytical platform for fast screening and comparative lipid profiling in food, microbial, cellular and milk-derived systems.

1.2. Outline

Lipid profiling is an important analytical approach in biomedical research, food quality control and technological process monitoring, as lipids are key structural and functional components of biological and food-related systems. They form cellular membranes, participate in energy storage, regulate signaling pathways and modulate inflammatory and metabolic processes. Therefore, changes in lipid composition may reflect oxidative stress, inflammation, microbial adaptation, response to drugs, antibiotics or toxic agents, as well as disease-related processes. In biological matrices, including cells, microorganisms and body fluids such as blood or milk, lipid profiling may support the detection of metabolic disturbances, infection, toxicity or potential disease markers. Milk is of particular interest, as it may be considered both a biological fluid reflecting physiological or pathological changes and an important raw material for food production. In food products, lipid profiles determine nutritional value, technological properties, stability and sensory quality, while their changes may indicate food ageing, oxidation, hydrolysis, rancidity or adulteration by replacement of high-quality fats with cheaper substitutes.
The development of rapid analytical methods for lipid profiling is of significant practical and scientific importance. Early detection of lipid-related changes may support screening of pathological processes, while rapid quality control during technological production may help prevent economic losses associated with product degradation, adulteration or process failure. Conventional lipid analysis based on GC-MS or HPLC-MS provides high analytical performance; however, it usually requires time-consuming sample preparation, extraction, derivatization and chromatographic separation. In this context, LDI-MS/NALDI-MS methods represent an attractive alternative, enabling rapid acquisition of lipid fingerprints with limited sample preparation. The potential of LDI-based techniques has already been demonstrated for profiling and differentiating lipid-rich

samples and other low-molecular-weight compounds, including approaches using nanostructured plates produced by techniques such as ALD and CVD. However, developing a comprehensive methodology for a selected group of samples still requires designing dedicated NALDI surfaces with optimized physicochemical properties. Physical vapour deposition offers a promising route for producing such nanostructured plates in a controlled and reproducible manner, which may improve sensitivity, selectivity and repeatability of lipid profiling.

1.3. Work plan

The first research task will focus on the synthesis of nanostructured NALDI plates by physical vapour deposition using selected coinage and noble metals, such as gold, platinum and copper, as well as mixed-metal systems, e.g. Au-Cu and Au-Pt. The influence of deposition conditions, metal composition, doping and surface morphology on plate properties will be assessed using microscopic, spectroscopic and surface-characterization techniques. In the first analytical step, the obtained plates will be systematically tested using lipid standards representing different lipid classes, in order to evaluate ionization efficiency, reproducibility, background signal, sensitivity and selectivity. Based on these results, the most effective surfaces will be selected and applied to more complex matrices, including milk, bacterial cultures and cell lines exposed to selected stress, toxic or stimulating factors. Selected plate variants may also be tested using clinical or tissue samples to verify their applicability in detecting biologically relevant lipid-profile changes. GC-MS, and optionally LC-MS/HPLC-based methods, will be used as control techniques for validation and comparison of the obtained lipid profiles.

1.4. Literature (max. 7 listed as a suggestion for a PhD candidate preliminary study)

a. Maślak E., et al. *Silver Nanoparticle Targets Fabricated Using Chemical Vapor Deposition Method for Differentiation of Bacteria Based on Lipidomic Profiles in Laser Desorption/Ionization Mass Spectrometry.* Antibiotics, 12(5), 874, 2023. DOI: 10.3390/antibiotics12050874**.**
b. Sibińska E., et al. *Advances in LDI-MS Analysis: The Role of Chemical Vapor Deposition-Synthesized Silver Nanoparticles in Enhancing Detection of Low-Molecular-Weight Biomolecules.* Journal of the American Society for Mass Spectrometry, 35, 2041-2055, 2024. DOI: 10.1021/jasms.4c00071.
c. Mametov R., et al. *Metabolic profiling of bacteria with the application of polypyrrole-MOF SPME fibers and plasmonic nanostructured LDI-MS substrates.* Scientific Reports, 14, 556, 2024. DOI: 10.1038/s41598-024-56107-0.

1.5. Required initial knowledge and skills of the PhD candidate

The PhD candidate should be scientifically curious, creative and able to work both independently and as part of a research team. Basic knowledge of analytical chemistry, mass spectrometry and lipid analysis is expected. Familiarity with sample preparation, chromatographic techniques such as GC or HPLC, and mass spectrometry-based methods, including MALDI/LDI-MS, will be particularly valuable. The candidate should also have basic knowledge of instrumental techniques used for material and surface characterization, as well as experience in experimental data processing and interpretation. English sufficient to read and discuss scientific literature is required.

1.6. Expected development of the PhD candidate's knowledge and skills

During the PhD project, the candidate will gain interdisciplinary knowledge and practical skills in the development of rapid LDI/NALDI-MS-based methods for lipid profiling. The candidate will develop competences in analytical chemistry, mass spectrometry, lipidomics, chromatographic analysis and characterization of nanostructured analytical surfaces. Practical skills in sample preparation, GC-MS and optionally LC-MS/HPLC analysis, LDI/MALDI-MS measurements, optimization of NALDI plates and statistical processing of lipid profiles will be strengthened. The obtained results will be presented in scientific publications and at domestic and international conferences and may contribute to the development of new analytical solutions with potential patent protection.