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## **Book of Abstracts**



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# **Abstracts of Lectures**

## CAPILLARY ELECTROPHORESIS IN THE AGE OF ARTIFICIAL INTELLIGENCE

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Capillary electrophoresis (CE) has long been valued for high separation efficiency, short analysis time, low sample and reagent consumption, and compatibility with diverse analytical formats. These features make CE a particularly attractive platform for data-driven analytical chemistry as artificial intelligence and machine learning can reshape CE method development, from literature-guided selection of background electrolytes and separation conditions, through electropherogram processing, peak recognition and deconvolution, to predictive quality control and automated method comparison<sup>1</sup>.

A particularly illustrative example of this broader evolution is the connection between CE and microscale thermophoresis (MST), including online CE-MST and stop-flow MST platforms for studying physicochemical equilibria. The presentation will also discuss how artificial intelligence and generative AI can support the prospective evaluation of such emerging methodologies using the RGB\_ex-ante model combined with an AI-Delphi framework, in which independent experts evaluated the method while ChatGPT synthesized expert input and contributed to the consensus process<sup>2</sup>. A complementary case study on LLM-assisted ChlorTox estimation will be used to show the AI-driven chemical risk evaluation<sup>3</sup>.

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## ARE SAMPLE PRETREATMENTS NECESSARY IN FLOW CHEMICAL ANALYSIS?

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Flow analysis methods in chemical analysis have increasingly become essential tools across various fields of science and technology. These sophisticated instruments enable the acquisition of sensitive analytical data with high repeatability. Recent advancements have led to a growing demand for extracting information from more complex matrices, even beyond the conventional limits of quantitation.

Chelating resins are polymer-based materials bearing ligands that form stable complexes with metal ions and are widely used as solid-phase extraction (SPE) sorbents for trace metal separation and preconcentration. Conventional iminodiacetate (IDA)-type resins based on styrene/divinylbenzene copolymers suffer from reduced extraction efficiency in aqueous samples containing complex matrices. Consequently, hydrophilic methacrylate-based resins have been developed,<sup>1,2</sup> some of which are now commercially available. We proposed automated sequential injection SPE methods for trace metal determination in urine, in which NOBIAS CHELATE PA-1 was packed into a handmade minicolumn.<sup>3,4</sup> In this presentation, we report the synthesis of a novel chelating resin based on a hydrophobic polymer matrix and evaluate the effects of carboxymethyl functionalization on metal ion capture.

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## ADVANCING GLYCAN ANALYSIS BY CE/LIF

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Glycosylation analysis remains challenging due to the structural diversity of glycans and limitations in detection sensitivity, necessitating derivatization prior to capillary electrophoresis (CE). In this study, novel fluorescent labels based on a phenylpyridine scaffold bearing a hydrazide functional group were developed for sensitive glycan analysis. The synthesized tags were characterized using nuclear magnetic resonance, fluorescence spectroscopy, and mass spectrometry (MS). The hydrazide moiety enables efficient glycan labeling via hydrazone formation, eliminating the need for a reduction step, while the inherent positive charge of the labels supports both electrophoretic separation and MS detection in positive ion mode. Fluorescence excitation maxima in the 300–400 nm range allow compatibility with commonly available solid-state lasers and light-emitting diodes (LEDs) for fluorescence detection (LIF/LED-IF). To accommodate these properties, a commercial CE instrument was adapted using an external laser or LEDs coupled through the 3D-printed adapters, and optimal optical filters were selected based on signal-to-noise ratio. The optimized method achieved baseline separation of labeled oligosaccharides and detection limits in the nanomolar (MS) and picomolar (LIF) range. The approach was successfully applied to N-linked glycan profiling of glycoproteins, including monoclonal antibodies. This work demonstrates an effective labeling strategy and a practical framework for adapting CE/LIF systems to new fluorescent probes.

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# TAILOR-MADE MODULAR FLOW ANALYTICAL SYSTEMS FOR CLINICAL DIAGNOSTICS

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The widespread availability of electronic components, combined with numerous DIY fabrication options, has made it feasible to construct flow analysis systems in virtually any laboratory. A key advantage of this approach is the versatility of the developed systems, stemming from their adaptability to specific analytical challenges, enabling the creation of “tailor-made” solutions. In this presentation, the architecture of simple multicommutated flow analysis (MCFA) systems for iron determination<sup>1</sup> serves as a foundation for the development of more advanced MCFA systems, enabling complex analytical procedures related to iron metabolism parameters<sup>2</sup> and lactate dehydrogenase isoenzyme activity assessment<sup>3</sup>. By modifying the characteristics of optoelectronic detectors, it is also possible to perform immunoprecipitation-based determination of proteins essential for iron homeostasis<sup>4</sup>, as well as non-enzymatic lactate determination<sup>5</sup>.

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# CAPILLARY SURFACE FUNCTIONALIZATION FOR CONTROLLED ELECTROOSMOTIC FLOW AND ENHANCED SEPARATION PERFORMANCE

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Capillary electrophoresis (CE) coupled with capacitively coupled contactless conductivity detection (C<sup>4</sup>D) is a versatile platform for the rapid analysis of ionic species; however, its performance is fundamentally governed by electroosmotic flow (EOF), which is often difficult to control in a stable and reproducible manner. To address this limitation, capillary surface functionalization has emerged as an effective strategy for tailoring EOF and improving separation performance. In this work, we present an overview of covalent and polymer-based coating approaches for controlled EOF modulation, including aminosilane functionalization (e.g., APTES) and cationic polymer coatings such as polyallylamine (PAA). These surface modifications enable systematic tuning or suppression of EOF by manipulating surface charge and interfacial properties, thereby improving migration reproducibility and enhancing separation efficiency. By optimizing coating conditions, high-resolution separations can be achieved within short analysis times, even for complex mixtures containing multiple anionic species in real sample matrices. Furthermore, the increased robustness of the modified capillaries facilitates straightforward aqueous sample preparation, thereby reducing dependence on organic solvents and decreasing overall chemical consumption. This integrated approach emphasizes the importance of surface chemistry in enhancing CE performance and demonstrates its potential to promote sustainable analytical methodologies for food analysis.

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# GREEN WAY THROUGH A DIFFICULT TASK: A CE-UV METHOD FOR THE DETERMINATION OF CATECHOLAMINE METABOLITES IN COMPLEX SAMPLES

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The analysis of complex biological samples often requires labor-intensive and solvent-consuming preparation steps, which contradict the principles of Green Analytical Chemistry. In this work, a sustainable and practical approach based on capillary electrophoresis with UV detection (CE-UV) is presented for the determination of catecholamine metabolites in infant urine samples. A key aspect of the proposed methodology is the use of disposable diapers as non-invasive sampling devices, which significantly simplifies sample collection in neonatal diagnostics. The sample preparation step is reduced to a simple water extraction, eliminating the need for organic solvents and minimizing environmental impact. The extraction conditions were optimized using a Box–Behnken design, ensuring efficient recovery of the target analytes. The developed CE-UV method demonstrated satisfactory analytical performance, including limits of quantification at the low  $\mu\text{g mL}^{-1}$  level and good intra- and inter-day precision (RSD < 15%). The applicability of the method was confirmed through the analysis of real samples collected in a clinical setting. The greenness and practicality of the method were evaluated using AGREEPrep, GAPI, and BAGI tools, confirming that the proposed approach meets the criteria of sustainable and efficient analytical procedures. This study demonstrates that green analytical strategies can successfully address challenging analytical problems while maintaining robustness and applicability in routine laboratories.

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## PORTABLE LC WITH SAMPLE PREPARATION BASED ON BI AND SIC

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Chromatography is currently the spearhead of analytical chemistry. While research benefits from the flexibility and utmost resolution of commercial benchtop HPLC systems, that top performance is often an overkill for routine applications. Rather, many real-world applications would benefit from a compact, and thus portable, system able to operate at 100 bar with a dedicated detector and integrated sample preparation, like the sampling in remote locations, unstable analytes and time-critical decisions to name a few.

In this context, sequential injection chromatography (SIC) offers a suitable platform for flexibility and sufficient performance. This scheme comprises a bidirectional pump and a stream selector. The commercial advent of bidirectional pumps in the range of 50 to 200 bar, such as those from VICI, Analytical Scientific Instruments or Cetoni, enables commercial HPLC performance in this setup, allowing it to step out of the research environment into the real world. Additionally, bead injection (BI), the manipulation of solid phase extraction (SPE) sorbent slurries in closed manifolds around a stream selector, can be integrated for performing in-line SPE prior to SIC, enabling completely automated sample preparation prior chromatography.

BI and SIC synergize well, since they do not require additional hardware and the internal standard addition during SPE allows corrections that ease the chromatographic part. Only recently, custom valve flow-paths that improve the hyphenation of these techniques have been described in patents or scientific literature. The resulting system is very compact, and the portable proof-of-concept merely requires dedicated electronics and software. In this presentation, we will describe the fluidic part of such a portable LC system and how the optimized hyphenation can simplify the traditional benchtop instruments.

## CAPILLARY ELECTROPHORESIS IN FORENSIC DRUG ANALYSIS

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Rapid, reliable drug detection outside laboratory settings remains a major challenge in forensic science. This presentation demonstrates how capillary electrophoresis (CE) is evolving into a field-deployable solution while retaining laboratory-grade performance. We present recent advances in CE methodologies for the analysis of drugs of abuse in oral fluid and seized materials, including amphetamines, cocaine, opioids, tramadol<sup>1</sup>, and emerging semi-synthetic cannabinoids. Emphasis is placed on deep UV fluorescence detection and its role in achieving high sensitivity (low ng mL<sup>-1</sup>) and selectivity in complex matrices. A central focus is the development of compact, portable CE prototypes integrating simplified sampling, automated preparation, and miniaturized optical detection<sup>1,2</sup>. These systems demonstrate robust performance under real-world conditions and compliance with bioanalytical validation guidelines (ICH M10), supporting their applicability in roadside and field testing. Case studies illustrate rapid, quantitative drug detection, underlining the need for improved analytical tools in emerging drug markets. This work demonstrates that portable CE platforms can bridge the gap between laboratory accuracy and field deployment, offering a promising solution for next-generation forensic drug analysis.

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# FLOW-THROUGH ANALYTICAL SYSTEMS AND MICROSYSTEMS WITH ELECTROCHEMICAL DETECTION FOR MONITORING OF BIOLOGICALLY ACTIVE SPECIES.

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The presentation will focus on dissemination of results of IUPAC project focused on electroanalytical flow-through systems for monitoring of biologically active compounds (see IUPAC technical report <sup>1</sup>). It will address both current trends and future potential in this field for the development of new methods as well as fabrication and commercialization of the devices including miniaturization and portable assays realization under on-site, point-of-care, in-place, and field analyses. Attention will be paid to optimization and standardization of the flow-through systems and establishment of standardized testing protocols. Real application examples will demonstrate the benefits of these user-friendly systems. The above-mentioned topics will be discussed in the framework of activities of Division V (Analytical Chemistry) of IUPAC and its cooperation with EuChemS Division of Analytical Chemistry (DAC EuChemS). <sup>2</sup>

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## A MICROFLUIDIC PAPER-BASED ANALYTICAL DEVICE ( $\mu$ PAD) FOR FLUORIDE DETERMINATION

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A microfluidic paper-based analytical device ( $\mu$ PAD) for the determination of fluoride in aqueous samples was developed. The sensing mechanism was based on the suppression effect of fluoride ions on the formation of the intensely red-colored Fe(III)-thiocyanate complex because of the formation of the colorless Fe(III)-fluoride complex. Under optimal conditions, the  $\mu$ PAD is characterized by a linear detection range of 0.5 – 20 mg L, limit of detection of 0.22 mg L, and repeatability, expressed as relative standard deviation, of 3.42% (n = 75) for 8 mg L fluoride standards. The  $\mu$ PAD was successfully applied to the quantitative determination of the fluoride concentration in eight groundwater samples and the results obtained were statistically indistinguishable at the 95% confidence level from those obtained by a fluoride ion-selective electrode. Satisfactory recoveries were obtained by spiking the same samples with known fluoride concentrations. These results confirmed the suitability of the newly developed  $\mu$ PAD for fluoride determination in environmental waters.

# NEW APPROACH TO REFERENCE AND ION-SELECTIVE ELECTRODES IN FILTER PAPER MATRIX

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Although potentiometric detection is a well-established and widely applied technique in analytical chemistry, its implementation in microfluidic paper-based analytical devices ( $\mu$ PADs) remains comparatively limited. Typically constructed with glass bodies and containing internal electrolyte solutions, these electrodes are inherently fragile, relatively costly, and susceptible to leakage, which complicates their handling and transport. Moreover, their performance is influenced by external conditions such as temperature, humidity, and mechanical disturbances, leading to potential instability and signal drift in field applications.<sup>1</sup>

Developing dry, paper-based electrodes may help overcome several challenges associated with potentiometric measurements under environmental conditions. In this work, a novel approach to preparation a reference electrode, along with an ion-selective electrodes (ISEs), is presented. The reference electrode preparation is based on in-situ forming silver contact and silver chloride within a filter paper matrix. The stability of the electrodes and the independence of their potential from various solutions were evaluated. Moreover, the ISE dedicated to Cu(II) ions detection is proposed. The sensing membrane is based on polymer inclusion membrane selective toward copper(II) ions.<sup>2</sup> Both electrodes are integrated into a single potentiometric cell with a sample deposition zone. Upon deposition, the sample is distributed to both electrodes via capillary forces. The analytical performance of the developed system was evaluated through potentiometric measurements, demonstrating its applicability for Cu(II) detection.

*The study was supported by the Polish National Science Centre (Project OPUS NCN 2022/45/B/ST4/01463).*

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# LIQUID PHASE MICROEXTRACTION – CAPILLARY ELECTROPHORESIS FOR THE DIRECT ANALYSIS OF COMPLEX SAMPLES

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Analyses of complex samples are usually hampered by the presence of high concentrations of matrix components, which may interfere with the separation process and the detection of target analytes. Moreover, complex samples are often available in limited amounts, and concentrations of target analytes are much lower than those of matrix components. Separation methods, such as capillary electrophoresis (CE), are fit for very small sample volumes and often even for low concentrations of analytes. Nevertheless, the presence of matrix components constitutes the major limitation in CE analyses.

Sample pretreatment is, therefore, applied to complex samples, which is traditionally carried out by liquid-liquid extraction or solid phase extraction. However, they require additional instrumentation, are laborious, costly, waste-generating, and time-consuming, use far larger extractant volumes than needed for the actual injection, and are not always compatible with e.g. biological samples. Moreover, they are predominantly carried out manually and off-line with a risk of contamination during handling. Thus, greener and miniaturized liquid phase microextraction (LPME) techniques have been extensively developed and automated during the last two decades, which offer faster, cheaper, and safer sample pretreatment.

In this contribution, selected LPME techniques driven by both, passive diffusion and by electric field, will be presented and practical applications will be illustrated with emphasis on their automation and on-line coupling to CE. Novel concepts developed and designed in our laboratory will be demonstrated, including various in-vial sample pretreatment, sequential injection driven sample handling, as well as the direct extraction of dried biological materials coupled to CE. Critical evaluation and future outlooks will be also offered.

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# COMPARISON OF TWO FLOW METHODS FOR THE DETERMINATION OF ASCORBIC ACID IN PHARMACEUTICALS

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The pharmaceutical industry views as economically beneficial the replacement of manufacturing based on batch processes with continuous-flow manufacturing due to its flexibility. However, continuous manufacturing requires continuous on-line or in-line monitoring to ensure consistent production conditions. Ascorbic acid (AA), commonly known as Vitamin C, is a water-soluble compound that shows acidic and reducing properties due to the endiol group in its structure. Since it cannot be synthesized by the human body, AA must be ingested with food or food supplements. Consequently, there is a need to develop selective, simple and accurate methods for the determination of AA in pharmaceutical preparations. This paper compares two newly developed and validated on-line flow methods, based on Flow Injection Analysis (FIA) and Sequential Injection Analysis (SIA) principles, for the on-line determination of AA in pharmaceutical formulations. Both methods utilize a redox reaction where AA reduces a Cu(II)-bathocuproine disulfonate (BCS) complex, forming an orange Cu(I)BCS complex that is monitored spectrophotometrically at 483 nm. Under optimal conditions the FIA and SIA methods are characterised by linear ranges of 0.2  $\mu\text{M}$  - 40.0  $\mu\text{M}$  and 0.2  $\mu\text{M}$  to 10.0  $\mu\text{M}$ , and sampling rates of 120 and 72 samples/h, respectively. The LOD of both methods was determined as 0.06  $\mu\text{M}$ . The newly developed flow methods were successfully applied to the determination of AA in pharmaceutical products with good agreement with the standard method prescribed by Pharmacopoeia. In addition, recoveries experiments produced results in the ranges 99.6 % - 101.2 % and 97.1 % - 99.8 % for the FIA and SIA methods, respectively. These findings highlight the potential of these flow methods as a superior alternative to traditional batch-based assays for continuous quality control in pharmaceutical production

# REVISITING MULTI-WAVELENGTH DENSITOMETRIC MEASUREMENTS: A POWERFUL YET UNDERVALUED APPROACH TO HPTLC PLATE EXAMINATION

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Densitometric detection in TLC and HPTLC is based on spectrophotometric evaluation of chromatographic plates. In routine analysis, especially for single compounds, measurements are typically performed at one selected wavelength corresponding to the absorption maximum. This wavelength is usually determined using multi-wavelength scanning across a defined spectral range (e.g., 200-600 nm). However, this univariate approach becomes insufficient for complex mixtures, where multiple components exhibit overlapping but distinct absorption spectra. Relying on a single wavelength in such cases leads to a loss of valuable analytical information and limits reliable qualitative discrimination of samples.

An alternative strategy is proposed in which the multi-wavelength mode is used not just for wavelength selection, but as a source of high-dimensional data.<sup>1</sup> By retaining the full spectral dataset of HPTLC plates and applying multivariate analysis, more robust and generalizable classification models can be developed. The proposed approach is demonstrated using fountain pen inks as model systems - complex mixtures of azo and triphenylmethane dyes. The results show that multi-wavelength HPTLC densitometry significantly improves sample origin identification, highlighting its potential for advanced qualitative analysis.

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# HYPHENATED MICROEXTRACTION TECHNIQUES WITH FLOW SYSTEMS FOR ENVIRONMENTAL MONITORING OF PETROLEUM HYDROCARBONS

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Reliable determination of petroleum hydrocarbons underpins environmental forensics, ecosystem monitoring, agricultural land surveillance and ecological risk assessment. Solid-phase microextraction coupled to gas chromatography–mass spectrometry (SPME-GC-MS) now leads sample-preparation–detection workflows in this domain, displacing solvent-intensive Soxhlet and liquid–liquid extraction. Reports show that HS-SPME-GC-MS of total petroleum hydrocarbons in agricultural soils down to 0.1 mg / kg<sup>1</sup>, coupled SPME-GC-MS with untargeted chemometrics to discriminate crude oils of different geographical origin, yielding a forensic tool for source apportionment and spill investigation<sup>2</sup>. Integration with flow platforms (flow injection, sequential injection, lab-on-valve) further enables on-line conditioning, calibration and extraction–desorption cycling. Joint assessment by AGREEprep<sup>3</sup> (sample-preparation greenness) and BAGI<sup>4</sup> (method practicality) places SPME-GC-MS at a “golden mean” between solvent-heavy reference procedures and oversimplified rapid screens: it preserves quantitative compositional resolution while scoring markedly higher in greenness and applicability, as confirmed across forty-six TPH protocols in our benchmark.<sup>5</sup>

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# ENVIRONMENTALLY BENIGN FLOW INJECTION ANALYSIS USING NATURAL EXTRACTS AS GREEN REAGENTS FOR ANALYTICAL APPLICATIONS

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The increasing demand for sustainable analytical methods has driven the development of environmentally benign approaches that reduce hazardous reagents and waste. This study presents an eco-friendly flow injection analysis (FIA) system that employs plant-derived extracts rich in polyphenols, flavonoids, and anthocyanins as green chromogenic reagents for the online determination of aluminum and ammonia, replacing conventional synthetic chemicals. Aluminum is quantified via complex formation between  $Al^{3+}$  and anthocyanins in red galangal flower extract, producing a purple complex with maximum absorbance at 555 nm. Optimal analytical performance was achieved at pH 6, with 3% (v/v) extract dilution, a 125 cm mixing coil, and a sample volume of 150  $\mu$ L. Ammonia determination relies on its conversion to  $NH_3$  in an alkaline donor stream, followed by diffusion across a gas-permeable membrane into an acceptor stream containing purple sweet potato extract, where a color change is measured at 604 nm proportional to ammonia concentration. Optimization of standard parameters for ammonia determination produced optimal results under 1.5 M NaOH, 10% extract, and 50 cm mixing coil with 300  $\mu$ L sample volume. This work highlights the potential of natural extracts as sustainable alternatives in FIA systems with high accuracy (95-100% recovery) and satisfactory precision (RSD <5%), offering a promising pathway toward greener, cost-effective, and efficient analytical techniques for determination of aluminum and ammonia.

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# CHARACTERIZATION OF LIPOSOME BEHAVIOR IN CARRIER AMPHOLYTE SOLUTIONS USING TAYLOR DISPERSION ANALYSIS

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Liposomes are widely used as biomimetic systems and drug delivery vehicles, yet their stability under analytical conditions remains insufficiently understood. In this study, Taylor dispersion analysis (TDA) was applied to investigate the effect of carrier ampholytes (CAs) on the integrity of lipid vesicles. Multiple liposome systems were examined, including vesicles prepared from liver total lipid extract, defined phospholipid mixtures (POPC:DOPE:Chol, POPC:DOTAP:Chol, POPC:PA:Chol), and a clinically relevant lipid nanoparticle formulation. TDA enabled simultaneous detection of vesicle-sized populations and small molecular species, providing detailed insight into ampholyte–liposome interactions. The results revealed that the impact of ampholytes on vesicle integrity is strongly dependent on liposome composition. While vesicles derived from liver total lipid extract exhibited destabilization in the presence of broad-range ampholytes, leading to aggregation or disappearance of vesicle-sized populations, liposomes prepared from defined lipid mixtures and lipid nanoparticles remained structurally stable across the investigated concentration range. These results demonstrate that ampholyte-induced destabilization is strongly dependent on membrane composition and cannot be explained solely by electrostatic interactions. TDA proved to be a sensitive method for detecting early structural changes that are not observable by conventional means.

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# BEHIND THE SCENES OF BIOLOGICAL SAMPLE RESEARCH: BENEFITS OF INTEGRATING FLOW ANALYSIS WITH INDUCTIVELY COUPLED PLASMA MASS SPECTROMETRY (ICP-MS)

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Inductively coupled plasma mass spectrometry (ICP-MS) is an exceptionally sensitive and accurate technique that has found wide application in the elemental analysis of biological materials. Samples such as blood, urine, or cerebrospinal fluid constitute an important source of diagnostic information on a patient's health status. Their analysis enables, among other things, the identification of metabolic pathways and the investigation of transport mechanisms of foreign compounds, i.e. xenobiotics.

However, the complex composition of the biological matrix poses significant analytical challenges. Accurate determination of elements by ICP-MS requires appropriate sample preparation, which often involves multiple dilution steps, removal of matrix components, extraction of analytes, and their preliminary preconcentration. These procedures not only extend the analysis time and increase its costs, but also raise the risk of both systematic and random errors. Furthermore, clinical laboratories often process very large numbers of samples, many of which are available only in limited volumes. In this context, the use of flow-based techniques, enabling automated sample preparation and direct introduction into the spectrometer, can significantly improve ICP-MS analysis, making it more efficient and reproducible. This presentation discusses selected examples of the application of various flow analysis techniques coupled with ICP-MS in the investigation of biological samples. The examples presented are discussed with particular emphasis on the benefits arising from the use of flow-based techniques in combination with ICP-MS [1].

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## AUTOMATED SYSTEMS FOR MONITORING TESTS

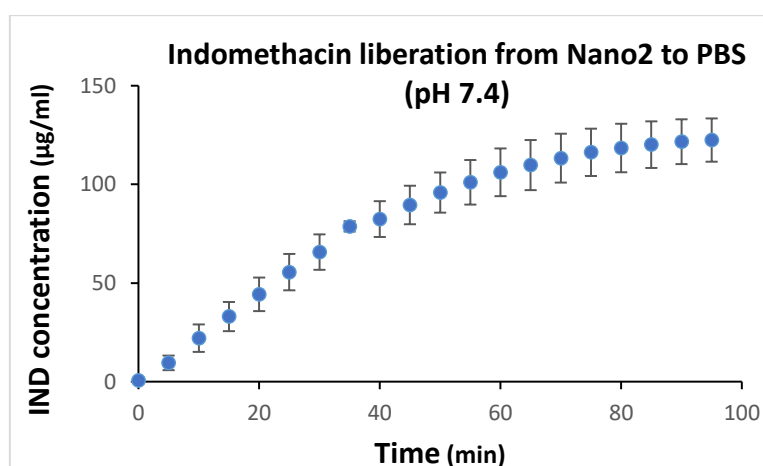
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The automated low pressure flow system, based on flow or sequential injection analysis that allows easy and quick manipulation with low volumes of liquids, enables application in different monitoring tests including liberation of active substances from different topical pharmaceutical formulations. Example of such test will be presented that corresponds to the liberation of indomethacin from 1% nanocrystal suspensions prepared to increase solubility of this lipophilic substance and thus increase the bioavailability.

Sequential injection system was on-line connected to the Franz cell where the sampling was accomplished in short time intervals, and the content of indomethacin was monitored in real time. The liberation profiles were measured for the original crystal form, two nanocrystals of different diameters (100 and 350 nm), and commercially available 1% indomethacin gel in two liberation media – acetate buffer of pH 4.5 and phosphate buffer of pH 7.4. The optimization of the flow system together with the obtained results evaluated from the perspective of pharmaceutical technology will be discussed in detail.

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# NANOMATERIALS AND NANOTECHNOLOGIES IN NOVEL DETECTION SCHEMES OF JANUS KINASE INHIBITORS AND ANTIOXIDANTS

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Janus kinase inhibitors (JAKis) represent a relatively new class of drugs approved for the treatment of inflammatory and autoimmune diseases by blocking Janus kinase (JAK) enzymes. Monitoring precise drug concentration ensures therapeutic effect while minimizing the risk of toxicity. In parallel to a key reference method of liquid chromatography–mass spectrometry (LC-MS), sensitive electrochemical methods for the JAKis detection are developed. This work addresses both current trends and future potential in the development of novel electrochemical sensors and procedures directed to a real-time point-of-care analysis enabling personalized therapeutic drug monitoring.<sup>1</sup>

Approaches for a prediction of antioxidant capacity in real samples based on bio-specific assays of antioxidant – pro-oxidant interaction in the presence of a selected (bio)discriminator (enzyme, nanozyme, DNA, lipid, etc.) under *in vitro* conditions are presented as well.<sup>2</sup> Utilization of effective electrochemical detection platforms and procedures in flowing systems is discussed.<sup>3</sup>

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## ON-SITE SCREENING AND QUANTIFICATION OF CANNABINOIDS IN CANNABIS PRODUCTS USING CAPILLARY ELECTROPHORESIS – DEEP UV FLUORESCENCE

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Hexahydrocannabinol (HHC) and tetrahydrocannabioctyl (THC-C8) are semi-synthetic cannabinoids (SSCs) that are increasingly added to cannabis products. These substances are often found in mislabelled and poorly regulated products. Although frequently marketed as safe alternatives to  $\Delta^9$ -tetrahydrocannabinol ( $\Delta^9$ -THC), psychotropic effects and potential health risks of SSCs are often underestimated and determination requires laboratory settings as there are no on-site screening methods. Thus, the aim of the study was to develop and validate a screening method for the rapid identification and determination of emerging SSCs, such as HHC and  $\Delta^9$ -THC-C8 in cannabis products that are suitable for use outside of traditional laboratory settings. With additional decarboxylation step, the method provided possibility to determine the total content of natural  $\Delta^9$ -THC and cannabidiol (CBD). For this purpose, a portable, in-house-built device based on capillary electrophoresis coupled with a deep-ultraviolet fluorescence detector (CE–FD) was used. The developed methodology met the validation criteria. Accuracy and precision were within 20% at the lower limit of quantification (LLOQ) and within 15% at other quality-control levels. Processed samples remained stable for at least three days, with reinjection reproducibility also within 15%. Carry-over was observed in samples with high analyte concentrations, but this was mitigated by improving the between-run rinsing protocol. Matrix effects remained within the range of 89.8 – 106.6%. The applicability of the developed and validated method was successfully demonstrated on 16 cannabis products. In most cases, the declared cannabinoids were detected; however, the measured concentrations were significantly lower than those stated by the manufacturers.

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# A NEW CALIBRATION FRAMEWORK TO STUDY AND REDUCE INTERFERENCE IN CHROMATOGRAPHIC DETERMINATION OF ASCORBIC ACID

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This study aimed to develop an innovative methodological approach based on analytical calibration to eliminate interference effects of various natures: multiplicative, additive, and nonlinear, leading to improved accuracy of analytical results. The study involved combining three calibration methods into a single procedure: the dilution method (DM), the integrated calibration method (ICM), and the H-point standard addition method (HPSAM). The performance of this approach (DM-ICM-HPSAM) was tested by chromatographic analysis of ascorbic acid in juice samples. High-performance liquid chromatography with diode array detection (HPLC-DAD) was used to determine this analyte. After optimization of the chromatographic method, the method was validated, and appropriate measurement conditions were selected for the HPSAM approach (multiple wavelengths). First, the DM-ICM-HPSAM was checked in analyses conducted on a sample with a known AA concentration, and then on several juice samples purchased from local stores. The reference method involved oxidizing ascorbic acid to dehydroascorbic acid in an acidic environment using the standard blue dye 2,6-dichlorophenolindophenol (DCIP) to a colorless leucocompound. The proposed approach allowed compensation for multiplicative effects (via comparisons of ICM estimates), additive effects (in accordance with the HPSAM), and nonlinear effects (thanks to the DM). Accurate results were obtained by extrapolating to zero degree of dilution. The types of interference effects present in the juice samples were identified. The RGB model was used to evaluate the developed approach in the context of White Analytical Chemistry, but the AGREE tool enabled an assessment of environmental friendliness. The method was shown to be consistent, coherent, and applicable to various analytical systems

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# PUSHING THE LIMITS OF SURFACE-ENHANCED RAMAN SPECTROSCOPY (SERS) IN FLOWING SYSTEMS

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Surface-enhanced Raman spectroscopy (SERS) is a highly sensitive analytical technique based on plasmonic nanostructures, typically silver nanoparticles, capable of amplifying Raman signals by several orders of magnitude. However, its application to complex mixtures remains limited due to insufficient selectivity and spectral complexity. These challenges can be mitigated by coupling SERS with separation techniques and developing advanced SERS substrates.

To address some of these limitations, we synthesize polystyrene microspheres decorated with silver nanoparticles (PS@AgNPs) that serve as an efficient SERS substrate. These hybrid structures provide improved suspension stability while preserving accessibility of the metallic surface. Their dimensions enable efficient manipulation using acoustofluidic forces, allowing controlled concentration and positioning. Indeed, we employ acoustofluidic manipulation of PS@AgNPs in microfluidic channels to create a dynamic, flow-through SERS detection zone. This “SERS cuvette” concept offers potential for integration with complementary analytical methods. Ongoing work focuses on coupling SERS with microfluidic capillary electrophoresis and developing a device enabling simultaneous SERS-MS detection.

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# NON-INTUITIVE EFFECTS OF IONIC STRENGTH IN COUPLED ACID-BASE EQUILIBRIA

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Ionic strength is commonly treated as a background parameter in analytical chemistry, expected to induce predictable and uniform shifts in equilibrium systems. However, when multiple acid-base equilibria are coupled, such as in buffer-indicator systems, the resulting analytical response may become non-intuitive. Importantly, changes in ionic strength affect only charged species, meaning that the direction and magnitude of the effect depend on whether the acid or the base form is charged.

In such systems, ionic-strength-induced shifts in the dissociation constant of the buffer lead to subtle changes in the actual pH of the solution, even when nominal conditions remain unchanged. Although these pH shifts are often too small to be measured directly, they propagate further into other coupled equilibria, including that of the analytical probe. As a result, the overall analytical response may reflect a combination of direct and indirect effects, which can either reinforce or counteract each other.

In this work, ratiometric UV-Vis spectrophotometry was used to investigate model systems under controlled ionic strength conditions. The results demonstrate that the analytical signal is not governed by ionic strength alone, but strongly depends on the nature of the coupled equilibria. Regimes of amplification and cancellation of the analytical response are observed, depending on how individual equilibria respond to ionic strength.

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# A WHITE ANALYTICAL CHEMISTRY-BASED APPROACH TO OPTIMIZING CAPILLARY ELECTROPHORESIS METHODS FOR ORGANIC ACID ANALYSIS IN FUNCTIONAL FOODS SAMPLES

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Short-chain fatty acids (SCFAs) are organic compounds containing aliphatic chains of one to six carbon atoms. These metabolites play an important role in metabolic, immune, and neurological homeostasis, and their concentrations have been associated with a wide range of disorders, including gastrointestinal diseases, cancers, and neurodegenerative conditions. Fermented plant-based foods, such as pickled vegetables, are natural sources of these metabolites and may serve as functional components of gut health-oriented diets.<sup>1</sup>

Considering the physicochemical properties of SCFAs, including their low molecular weight and high water solubility, capillary electrophoresis is a suitable technique for their determination. One of the most critical steps in the development of a capillary electrophoresis method is the optimization of the background electrolyte composition. The aim of this study is to optimize the composition of the background electrolyte in accordance with the principles of White Analytical Chemistry and the RGB fast model.<sup>2</sup> This concept integrates three key aspects of analytical performance: analytical quality (red), environmental safety (green), and practical and economic efficiency (blue). The selection of the optimal buffer composition was supported by a mathematical model developed to ensure a more objective and comprehensive decision-making process. The final method was validated, and the impact of the introduced changes on analytical quality, operational efficiency, and toxicological profile was assessed. In addition, the applicability of the developed method was verified using real samples of fermented food products.

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## AUTOMATED DES IN-SYRINGE EXTRACTION OF ANTIBIOTICS

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Antibiotics are widespread in aquatic environments in range from ppt to ppb.<sup>1</sup> In line with the new EU Urban Wastewater Treatment Directive, there is a need for rapid, cost-effective, and environmentally friendly methods for their determination. However, analysis usually requires sample preparation and preconcentration. A recent approach involves deep eutectic solvents (DES), which are considered green alternatives to conventional organic solvents due to their low toxicity and favorable physicochemical properties.<sup>2</sup>

We developed an automated dispersive liquid–liquid microextraction method using the Lab-In-Syringe (LIS) technique coupled online with HPLC. The method was applied to the determination of tetracycline, oxytetracycline, doxycycline, chlortetracycline, sulfamethoxazole, and trimethoprim in environmental waters. The extraction procedure, including system cleaning, takes 8 minutes and requires only 200  $\mu\text{L}$  hydrophobic DES and 1.7 mL sample. The method showed good linearity, detection limits of 9.9–30.3  $\mu\text{g L}^{-1}$ , and high precision (RSD < 4.3%), with satisfying recoveries. This demonstrates the potential of DES-based automated extraction for improving pharmaceutical analysis in environmental samples.

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# A MODULAR MICROFLUIDIC IR FLOW-CELL FOR INLINE AND IN-SITU REACTION MONITORING

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Microfluidic platforms offer significant advantages for chemical synthesis and screening, including reduced reagent consumption, enhanced safety, and improved control over reaction parameters<sup>1,2</sup>. However, their full potential is often limited by the lack of suitable analytical tools capable of real-time, in-line monitoring at microliter scales. Therefore, we present a microfluidic infrared (IR) flow-cell for the inline and in-situ monitoring of chemical reactions under continuous-flow conditions.

The microfluidic chip was fabricated in-house from fused-silica (FS) using high-precision selective laser-induced etching (SLE). The resulting flow-cell features a total internal volume of only 3  $\mu\text{L}$  and is designed for direct integration with a commercial IR probe (*Metler Toledo*) via a custom 3D-printed holder. The modular design also enables seamless incorporation into wide range of microfluidic workflows.

The analytical performance of the system was evaluated by comparing measurements obtained under microflow and conventional batch conditions using the same IR probe, demonstrating comparable calibration behavior and limits of detection. The applicability of the flow-cell for reaction monitoring was further demonstrated using a model imine formation reaction between benzaldehyde and aniline under continuous-flow. Additionally, the optical transparency of fused-silica enables in-situ investigation of photochemical transformations: This was demonstrated through the monitoring of a Paterò-Büchi cycloaddition under UV-irradiation in both continuous- and stopped-flow configurations. Overall, this work establishes a versatile microfluidic IR flow-cell as a powerful tool for real-time reaction analysis in microscale systems.

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## BEAD INJECTION AS A TOOL FOR ONLINE SOLID PHASE EXTRACTION

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Sequential Injection Analysis is a flexible and programmable platform for executing various analytical tasks in a flow. The key features are high precision, automation, miniaturization, operation speed, and long-term durability. These enable the development of methods for sample pretreatment, direct determination, or online hyphenation of multiple analytical instruments.

Online Solid Phase Extraction can be automated by Sequential Injection Analysis using either reusable commercial or lab-made (packed) cartridge columns or automatically disposable columns (Bead Injection). The extraction utilizes a broad range of commercial or experimental sorbents (silica, polymers, polysaccharides, or diatomaceous earth) in particulate, monolith, or nanofibrous morphologies.

Sequential Injection Analysis significantly accelerates method development by enabling continuous online detection at the column outlet to understand critical parameters during sample loading, matrix elimination, and analyte elution, maximizing method recovery and minimizing time and consumable demands.

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## MODERN FLOW APPROACHES FOR LPME AUTOMATION

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Sample preparation is a key step in the analytical process that decides on sensitivity, reliability, and selectivity of the analytical method. Above all, sample compatibility with the instrumental technique aimed for analyte quantification must be achieved. Sample preparation is a major error source, e.g., by contaminating the sample during treatment. Procedural automation and coupling sample preparation directly to the intended detection instrument can reduce these risks and often comes with a gain in cost- and time efficiency, reproducibility, and reliability.

While today, versatile autosampler systems are mostly employed for this purpose, flow techniques can show interesting advantages or add positive features to such online connection to the processing instrumentation, e.g. HPLC. Thus, getting familiar with the concepts and potential of handling samples in flow for analyte preconcentration and matrix clean-up can be more than just useful background knowledge for the analytical chemist.

In this seminar, selected approaches of flow-automating liquid phase (micro)extraction (LPE/LPME) procedures should be presented including specific challenges. Furthermore, the potential, characteristics, and differences in operation and performance of flow approaches should be given focusing on Sequential Injection Analysis and the Flow Batch technique Lab-In-Syringe (LIS), an approach overlapping with autosampler automation.

In this sense, required material for flow systems and their production by 3D printing will be shown. This will include the conception and 3D printing of a cost-efficient autosampler presenting an indispensable device for true procedural automation, at least for feeding sample solutions the analyzer system. In this sense, an overview of recent developments of LIS automated sample preparation and related instrumental developments will be given.

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# Abstracts of Posters

# AUTOMATED SOLID PHASE EXTRACTION OF CARVEDILOL USING AN AGAROSE– MWCNTS MONOLITHIC SORBENT

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Beta-blockers are widely prescribed pharmaceuticals used to treat cardiovascular diseases and other conditions<sup>1</sup>. These compounds are of particular concern as they are not removed during water treatment processes and can therefore be found in the environment<sup>2</sup>. The aim of this work was to develop an automated solid phase extraction (SPE) method for the determination of carvedilol (CAR) as a target compound.

In this context, a PTFE column was packed with a monolith sorbent composed of a biopolymer, agarose (AG), and multi-walled carbon nanotubes (MWCNTs). The monolith was prepared by mixing AG, MWCNTs and Tween 80/cyclohexane at 60°C, followed by gelation at 4°C for 24 h. The material was then subjected to ultrasound-assisted washing with a 1:1 (v/v) ethanol-water solution followed by drying at 40°C for 15 h.

The extraction procedure consisted in activating the sorbent with isopropanol and equilibrated with a solution of NaOH pH 9.5. Then, the sample containing CAR (pH 9.5) was loaded. After that, the column was washed with water and the analyte was eluted using acetonitrile (ACN). For the extraction stage, parameters such as sample pH (9.5), type and volume of eluent (ACN, 1.3 mL), elution flow rate (0.50 mL min<sup>-1</sup>), and sample volume and flow rate (10.00 mL, 0.39 mL min<sup>-1</sup>) were evaluated and optimized to achieve the best conditions for the SPE procedure.

*The study was supported by Universidad Nacional del Sur (PGI 24/Q119, 24/Q135)*

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# BRINGING SERS INTO FLOW: ACOUSTOFLUIDIC CONTROL FOR ENHANCED DETECTION

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Surface-enhanced Raman spectroscopy (SERS) is an analytical technique that utilizes metallic nanoparticles to amplify the Raman signal. The integration of SERS into flow-based systems enables real-time and in-line analysis. However, its practical implementation remains limited by poor signal reproducibility, insufficient control over nanoparticle aggregation, short interaction times and memory effects.

In this work, we aim to develop a system capable of SERS detection under flow conditions. For this purpose, we designed a novel type of substrate that can be manipulated within a flowing liquid medium using acoustofluidic techniques. The substrate consists of polystyrene microparticles coated with silver nanoparticles. Three types of polystyrene microparticles were synthesized, each differing in the length of the stabilizing polymer chain, namely Polyvinylpyrrolidone (PVP). We systematically evaluated the effect of PVP length on the suspension properties, including particle size, uniformity, and mass fraction, and investigated the coating of silver nanoparticles using electron microscopy. These observations were subsequently correlated with SERS sensitivity of adenin as a model analyte using 2D mapping of dried samples, providing a basis for subsequent investigations of substrate focusing under flow conditions in a capillary.

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# COMPARISON OF SEPARATION METHODS FOR ANALYSIS OF DRUGS CONTAINING ACETYLSALICYLIC ACID

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Acetylsalicylic acid (ASA) is one of the longest-used drugs for treating inflammation, reducing fever, and relieving pain. For this reason, a number of analytical techniques for its determination are being developed, both in pharmaceutical products for quality control and in biological samples. However, this process is complicated by the fact that in a basic conditions, this molecule undergoes hydrolysis to salicylic and acetic acids, which can ultimately lead to false-negative results.<sup>1</sup>

The main aim of our work was to compare various separation methods, including IEC, HPLC, GC-MS and ITP, in terms of their suitability for the hydrolytic study of acetylsalicylic acid. The time traces dependencies of ASA hydrolysis were measured at various pH levels and it was found out that hydrolysis proceeds most rapidly in a basic conditions (pH > 10), although gradual hydrolysis also occurs at lower pH. For this reason, the results ASA hydrolysis followed by the determination of reaction products was employed for founding of optimal experimental conditions which were subsequently utilized in the analysis of real samples of pharmaceuticals containing acetylsalicylic acid.<sup>2</sup>

*The study was supported by Masaryk University (project MUNI/A/1682/2025)*

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# NANOFIBROUS POLYAMIDE-BASED SORBENTS: A NOVEL APPROACH FOR EXTRACTION OF POLYCYCLIC AROMATIC HYDROCARBONS FROM RIVER WATER

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In this study various polyamide (PA) homopolymers and copolymers nanofibers with different alkyl chain lengths have been investigated as novel sorbent materials for on-line solid phase extraction of polycyclic aromatic hydrocarbons from river water. PA4/6 copolymer nanofibers showed the highest retention of tested analytes and comparable performance with commercial C18 sorbent. Developed on-line solid phase extraction method coupled with ultra-high performance liquid chromatography with fluorescence detection showed excellent analytical performance, with linearity ranging from 0.01 – 0.5  $\mu\text{g L}^{-1}$  for acenaphthene, anthracene, pyrene, chrysene, and 0.05 – 0.3  $\mu\text{g L}^{-1}$  for benzo(k)fluoranthene, benzo(a)pyrene, dibenzo(a,h)anthracene, indeno(c,d)pyrene and benzo(g,h,i)perylene, achieving determination coefficients ( $R^2$ ) between 0.9989 – 0.9999. The limits of detection and quantification were 0.003–0.01 and 0.01–0.05  $\mu\text{g L}^{-1}$ , respectively. The recoveries were between 92% and 105%, with RSD 1.5 – 6.7 %. The total time for the extraction and analysis of nine analytes was 14 minutes and was done automatically. That saves the time and limits the operator error. The polyamide-based sorbent requires no modifications and can be inexpensively prepared as homogenous mats by electrospinning process. It can be cut and manually packed into the column, allowing for easy column replacement, when needed, with inter-columns repeatability of RSD < 13%. In addition, due to the excellent resistance of polyamide nanofibers to HPLC conditions, a single extraction column can be reused for the preconcentration of multiple samples.

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# NOVEL STRATEGIES IN COUPLING SEQUENTIAL INJECTION TO CAPILLARY ELECTROPHORESIS FOR AUTOMATIC HANDLING, PRETREATMENT, AND ANALYSIS OF COMPLEX SAMPLES

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The coupling of flow-based sample preparation with capillary electrophoresis (CE) offers an effective route toward automated analysis of complex samples. In this work, an autonomous platform integrating Sequential Injection Analysis (SIA) with CE instrumentation is presented. The system is controlled by custom Python-based software enabling synchronized operation of pumps, valves, sample handling modules, and the CE unit.

The developed strategy allows automated sample transport, reagent addition, elution, homogenization, transfer of treated samples into the CE system, sequence execution, and unattended processing of multiple samples. The software also supports calibration and optimization of extraction/elution times.

As a model application, the platform was tested for processing dried blood spot (DBS) samples collected by volumetric microsampling (10  $\mu$ L). Automated DBS elution, mixing, and subsequent CE analysis were performed without manual intervention. The system was further evaluated using CE–MS detection for determination of Sotalol in DBS samples, achieving detection limits in the  $10^{-9}$  mol L<sup>-1</sup> range.

The poster will present the hardware and software architecture, control scripts, optimization outputs, and representative analytical data. The proposed approach reduces manual labor, improves repeatability, and expands possibilities for autonomous CE-based bioanalytical systems.

*The study was supported by: GA ČR 26-22325S, Automation of integrated and microscale clean-up, preconcentration and analysis of complex samples and from the Czech Academy of Sciences (Institute Research Funding RVO:68081715)*

# DUAL-FUNCTION PRUSSIAN BLUE MICROTEST FOR SELECTIVE SUCROSE DETECTION

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Prussian blue (PB) films are widely used in enzymatic biosensors as selective transducers for hydrogen peroxide generated in oxidase-catalyzed reactions<sup>1,2</sup>. Here, we demonstrate that enzymatically modified PB layers can play a dual role, acting both as sensitive detection elements and as anti-interference (scavenger) components. This functionality is particularly valuable in polyenzymatic systems, where intermediate species can affect selectivity. We exploit this concept in a microfluidic paper-based optical biosensor designed for the selective determination of sucrose.

The proposed device employs Prussian white as a chemosensitive element, which is oxidized to PB in the presence of hydrogen peroxide, producing an optical signal<sup>3</sup>. Glucose is detected via glucose oxidase, while sucrose is hydrolyzed by invertase to glucose in a bienzymatic cascade<sup>4</sup>. Crucially, the PB-based scavenging layer suppresses interference from glucose formed during sucrose hydrolysis, enabling accurate differentiation between the two sugars. The biosensor is simple, low-cost, and suitable for instrument-free analysis, demonstrating satisfactory analytical performance.

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# APPLICATION OF GC/MS METHOD FOR QUANTIFICATION SHORT-CHAIN FATTY ACIDS IN HUMAN MILK SAMPLES

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Short-chain fatty acids (SCFAs) are organic compounds characterized by a short carbon chain length (C1 – C6). These compounds are essential for the digestive system and help regulate metabolic processes in young children. WHO and UNICEF define breast milk as a “gold standard” of infant nutrition, as it provides everything a baby needs to grow properly. The SCFAs present in human milk play a key role in health prevention by reducing the risk of inflammation, atopic disease, and becoming overweight. Using gas chromatography-mass spectrometry (GC-MS) allows for the precise measurement of SCFAs, but a successful analysis depends on optimizing the sample preparation. The need to remove proteins and other interfering substances requires effective methods for cleaning and concentrating on the samples.

The study aimed to develop an effective method to measure eight SCFAs in human milk using GC-MS. The method was validated by testing its linearity, precision, trueness, limit of detection and quantification. The extraction process was optimized using the Box-Behnken design. This method enabled the assessment of the influence of specific factors and their interactions. Methyl tert-butyl ether (MTBE) was used for extraction, acting as both a solvent and protein precipitant. Acidifying the sample with HCl was essential to keep the analytes in a non-dissociated state, improving extraction efficiency. Centrifugation was performed at 4°C to prevent evaporation of volatile organic compounds and ensure clear separation of the layers. An efficient sample preparation procedure for the analysis of SCFAs in human milk was finally tested and validated.

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## AUTOMATION OF SERUM PROTEIN ANALYSIS FROM DRIED PLASMA SPOTS

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Serum protein electrophoresis (SPE) is a well-established analytical technique that enables the resolution of serum proteins into five to six distinct fractions—albumin,  $\alpha_1$ -,  $\alpha_2$ -,  $\beta$ -, and  $\gamma$ -globulins. Quantitative and qualitative alterations in these fractions are strongly associated with a wide range of pathological conditions, including inflammatory disorders, hepatic and renal dysfunction, and multiple myeloma.<sup>1</sup> Dried plasma spot (DPS) sampling represents an emerging microsampling strategy that facilitates the collection of minute blood volumes (in the microliter range). Following application, blood undergoes filtration through a size-selective membrane, effectively removing cellular components and potential interferents such as hemoglobin. The resulting plasma collected on a disc is analysed. This approach offers several advantages, including minimal invasiveness, reduced sample volume, enhanced biosafety, and the potential for decentralized or at-home sampling.<sup>2</sup>

In this study, we investigated the integration of DPS sampling with SPE analysis. The coupling strategy was systematically optimized across three DPS platforms, including one commercially available device and two in-house developed prototypes. The entire analytical workflow was highly automated, encompassing solution handling, analyte elution, and electrophoretic separation, all conducted using the Agilent Capillary Electrophoresis 7100 system.

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# FULLY AUTOMATED ONLINE BEAD INJECTION COUPLED TO SEQUENTIAL INJECTION CHROMATOGRAPHY FOR LIPOSOMES ANALYSIS IN FACIAL SERUM

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Nowadays, liposomal cosmetic formulations are increasingly used to enhance the stability and bioavailability of active compounds through encapsulation <sup>1</sup>. In this study, an automated sample preparation method combining Bead Injection and Sequential Injection Chromatography (BI-SIC) was developed to quantify an active ingredient (Niacin) and preservative (2-phenoxyethanol) in facial serum. The entire preparative procedure was automated using a special prototype valve <sup>2</sup>: an extraction microcolumn was formed in situ by packing Oasis HLB particles from a suspension into the injection loop. During the extraction step, both liposomes and analytes were retained on the sorbent. Subsequently, the analytes were selectively eluted and transferred to the separation column, while the sorbent and the retained liposomes were removed by flow reversal to the waste. The valve switching time of 24 s was sufficient to transfer the analytes to the separation column using 80 µL of eluent, critical for achieving maximum analyte recovery without interference. After elution, the used sorbent was discarded, and chromatographic separation was proceeded with 25 % acetonitrile. The developed method requires only 10 µL of sample and approximately 4 mg of sorbent per extraction. The total analysis time, including sorbent packing, conditioning, matrix cleanup, sorbent unpacking, and chromatographic separation, was 7.5 min (8 samples per hour). The method's applicability was demonstrated by analyzing a commercial liposomal facial serum. This research was co-funded by the European Union under the ATEBIO project (Advanced Techniques for Biomedical Diagnostics, Project ID CZ.02.01.01/00/23\_020/0008535), bilateral Mobility project (Mobility CZ-AT), SVV 260 782, and GAUK project no. 206623.

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# SWITCHABLE DEEP EUTECTIC SOLVENTS FOR GREEN AND SEPARATION-FRIENDLY MICROEXTRACTION

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There is a growing demand for alternative extractants in separation science, specifically in microextraction techniques to develop green sample preparation methods. To achieve this goal, various new solvent systems have been reported as alternatives to traditional hazardous organic solvents. Deep eutectic solvents (DESs) are a class of green solvents that can be used to extract either hydrophilic or hydrophobic compounds and can be tuned to offer various types of analyte interaction. However, a single DES is unfit to extract analytes of widely different polarity.

The introduction of switchable deep eutectic solvents (SDESs) can fill this gap and be used effectively to extract analytes of different natures from the same sample by sequentially toggling their polarity. SDESs have proven promising solvents for liquid-phase microextraction approaches, given their combined advantages of polarity and water-solubility switching, high extraction efficiency through various interactions, tunability, and biodegradability (hence eco-friendliness).<sup>1</sup> Driving forces for switching the hydrophilic-hydrophobic nature of SDES include changing sample pH, sample purging with CO<sub>2</sub>, or change in temperature.<sup>2</sup> This poster overviews the switching mechanisms of SDES with the above-mentioned driving forces, dissociation and reformation of SDESs based on switchable constituents, commonly reported hydrogen bond donor components, details their numerous advantages, and shows the possibility of integration of SDES into automated sample preparation strategies.

*This study was supported by GAUK (Project No. 424126) and GAČR (Project No. 26-22325S).*

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# POTENTIOMETRIC SENSOR FOR DETERMINATION OF LITHIUM IN A FLOW SYSTEM

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Lithium salts are widely used in medicine and modern energy storage technologies. Lithium carbonate is used in the treatment of bipolar disorder as a mood-stabilising medication; however, due to its narrow therapeutic range, lithium blood levels must be regularly monitored. Moreover, the increasing use of lithium-ion batteries highlights the importance of lithium determination in environmental and technological samples. Consequently, there is a need to develop simple, accessible, and reliable analytical methods for determining lithium, which could serve as an alternative or complement to existing techniques.

This study aimed to develop a potentiometric sensor for lithium determination in a flow system. The research included the selection of suitable reference electrodes, both batch and adapted for flow-mode operation, optimisation of the ion-selective electrode composite composition, and determination of the appropriate measurement time. In the batch system, lithium was determined in model and pharmaceutical samples. The instrumental conditions for measurements performed in the flow system were optimised. Flow-system studies were conducted in stop-flow mode using two flow modules fabricated by 3D printing. Basic validation parameters were determined, and the proposed method was applied to lithium determination in pharmaceutical samples.

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# DETERMINATION OF GENTAMICIN USING GOLD NANOPARTICLES AND SMARTPHONE-BASED DETECTION

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Gold nanoparticles (AuNPs) have gained significant attention in chemical analysis in recent years due to their chemical stability and the ease with which their surfaces can be functionalised using compounds capable of interacting with target analytes. These characteristics make them suitable for the development of various analytical approaches, including colourimetric methods [1]. Aminoglycoside antibiotics, such as gentamicin, are commonly applied in the treatment of bacterial infections; however, their residues can persist in environmental matrices (e.g., water) as well as in food products (e.g., meat, milk, and honey), emphasising the need for simple and portable detection techniques.

In this study, AuNPs were applied to develop a smartphone-based detection method for gentamicin determination. This work involved optimisation of AuNP synthesis as well as analytical conditions, including reagent concentrations, reaction parameters, and sample preparation. Measurement parameters using a smartphone as a detection system were also optimised, particularly the selection of an appropriate colour space model and analytical channel. For the proposed method, analytical parameters, including limits of detection and quantification, linear range, precision, and trueness, were determined. The effect of potential interferents, including other antibiotics (notably aminoglycosides), was investigated. The method was verified by determining gentamicin in model samples. The proposed approach was applied for determining this antibiotic in pharmaceutical and milk samples, and the results were compared with those obtained by the spectrophotometric method.

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## SPIT, BREATHE, DETECT: BILE ACID SIGNATURES OF BARRETT'S ESOPHAGUS

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Identifying noninvasive biomarkers for Barrett's esophagus (BE) remains a major clinical challenge, particularly given the diverse biological matrices exposed to refluxed bile acids. In this study, we profiled fifteen bile acids across four matrices—gastric juice, saliva supernatant and mucus, and exhaled breath condensate (EBC)—in a cohort of 80 individuals with gastroesophageal reflux disease (GERD), BE, or healthy physiology.

Across all matrices, glyco-conjugated bile acids dominated the bile-acid pool. Concentration ranges differed markedly: 1–700 nM in saliva supernatant, 5 nM–2.5 µM in saliva mucus, 0–100 nM in EBC, and 0–0.4 mM in gastric juice. Saliva-based matrices showed only subtle diagnostic differences, with no significant univariate separation, although mean total bile acids were slightly higher in BE than in healthy controls. EBC provided minimal diagnostic value. In contrast, gastric juice exhibited a clear disease-associated signature, with significant differences in total and tauro-conjugated bile acids and multiple individual species distinguishing GERD, BE, and healthy subjects. Healthy individuals showed higher gastric bile-acid levels than BE patients, suggesting that BE development reflects esophageal exposure patterns and mucosal vulnerability rather than absolute gastric bile-acid load. These findings highlight a matrix-specific hierarchy of diagnostic information, with gastric juice providing the strongest discriminatory signal.

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## ITP ANALYSIS OF AMINE DERIVATIVES OF ADAMANTANE

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The amine derivatives of adamantane (Amantadine, Rimantadine, Memantine) are widely used as antiviral drugs against various strains of flu<sup>1</sup> and/or for treatment of Parkinson disease or SARS-Cov-2 infection, recently. SQ109 based on ethylenediamine-skeleton of adamantane is active against both drug susceptible and multi-drug-resistant tuberculosis bacteria while it is assumed that adamantane skeleton is responsible for biological activity as consequence of its high lipophilicity and hydrophobicity.

The development of determination of above-mentioned compounds is paid high attention.<sup>1</sup> The analytical methods of molecular spectroscopy (absorption, luminescence, NIR), flow-injection analysis and potentiometry with ISE were tested<sup>1,2</sup> while hyphenated chromatographic methods, mostly HPLC-MS, were employed for analysis of biological samples.<sup>1</sup> In addition, capillary electromigration techniques (electrophoresis, isotachopheresis - ITP) were utilized for analysis of these compounds in mixture<sup>2-4</sup> and their sufficient resolution in CZE was achieved by addition of cyclodextrines when inclusion complexes of high stability are formed.<sup>4,5</sup> The application of cyclodextrins as complexing agents in capillary ITP is less often than in CZE, therefore the new analytical method for determination of Amantadine and Rimantadine in mixture was developed.

*The study was supported by Masaryk University (project MUNI/A/1682/2025).*

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# ENVIRONMENTALLY FRIENDLY MATERIALS BASED ON DEEP EUTECTIC SOLVENTS AND THEIR APPLICATION IN ANALYTICAL CHEMISTRY

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Deep eutectic solvents (DESs), formed by hydrogen-bond donors (HBDs) and acceptors (HBAs), have emerged as a versatile class of designer solvents with melting points significantly lower than those of their individual components. Among them, natural deep eutectic solvents (NADESs), composed exclusively of naturally occurring metabolites, represent a particularly promising group of environmentally friendly media.<sup>1</sup> Their tunable physicochemical properties, including negligible vapor pressure, low toxicity, and high biodegradability, position DESs as attractive alternatives to conventional organic solvents in analytical chemistry.<sup>1</sup> Beyond their green credentials, the structural diversity of DESs enables fine control over extraction efficiency, selectivity, and compatibility, and enables their application in both liquid and solid forms. This adaptability facilitates their use as efficient extraction media and sorbent phases in modern microextraction techniques.<sup>2</sup> This work presents an overview of recent advances in DES- and NADES-based materials, with a focus on their application in analytical chemistry. Particular attention is given to their role as extraction media and sorbent phases in microextraction techniques, where they offer enhanced performance while reducing environmental impact. The discussed developments highlight the growing potential of DESs not only as sustainable substitutes for traditional solvents but also as functional materials that can redefine sample preparation strategies in modern analytical workflows.

*The study was supported by the Polish Ministry of Science and Higher Education.*

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## FLOW SYSTEM FOR VOLTAMMETRIC DETERMINATION OF CREATININE USING IN-FLOW COPPER NANOPARTICLES-MODIFIED ELECTRODE

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Creatinine is a key biomarker routinely used in clinical diagnostics to assess kidney function and monitor various metabolic disorders. Its determination in biological samples is therefore of considerable importance. Conventional methods for creatinine determination, including spectrophotometric (e.g. Jaffé-based) and chromatographic techniques, although well established, are often time-consuming and require complex instrumentation. In this context, electrochemical methods, particularly voltammetry, have attracted increasing attention due to their simplicity, sensitivity, and potential for miniaturisation and on-site analysis. The integration of such techniques with flow-based systems offers additional advantages, including automation, improved reproducibility, and reduced reagent consumption. Moreover, performing the electrode modification under flow conditions enables automation of this step and further simplifies the overall analytical procedure.

The aim of this study was to develop a flow system for the voltammetric determination of creatinine using a screen-printed carbon electrode modified with copper nanoparticles. Both the electrode modification and the creatinine determination were carried out within a single automated procedure. Preliminary studies involved the optimisation of conditions for the electrodeposition of copper nanoparticles onto a carbon electrode, as well as for creatinine determination. The analytical parameters of the proposed method were evaluated, and the results preliminarily confirm its suitability for the analysis of biological samples.

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# DETERMINATION OF CORTISOL USING A SMARTPHONE AS A DETECTION SYSTEM

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Cortisol performs several essential functions in the body, and its proper concentration is crucial for maintaining homeostasis. Abnormal cortisol levels – both deficiency and excess constitute one of the significant causes of many serious diseases. Dysregulation of cortisol concentration leads to a range of adverse physiological effects. Therefore, monitoring its levels, especially using rapid and non-invasive methods, is of great clinical importance. In clinical diagnostics, cortisol is most commonly measured in blood serum using standardised methods based on immunoassays, radioimmunoassays, or high-performance liquid chromatography coupled with tandem mass spectrometry. Although these methods offer numerous analytical advantages, their application requires specialised instrumentation. For this reason, there is growing interest in developing novel, easily accessible methods for cortisol determination in pharmaceutical and biological samples.

In this study, a method for cortisol determination using a smartphone as a detection system was developed. The reaction between cortisol and tetrazolium blue chloride in the presence of tetramethylammonium hydroxide was selected. As the cortisol concentration increases, the color of the solution changes from pale yellow to magenta, enabling spectrophotometric and colorimetric determinations. In the study, the PhotoMetrix PRO software was used for image analysis and processing. Optimal measurement conditions were established, and the most suitable RGB channel was selected. Validation of the method included determination of linear range, limit of detection, precision, and trueness. It was also applied to determine cortisol in a pharmaceutical product. The obtained results indicate the potential for practical application of the proposed approach, utilising a smartphone as a detection system.

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# OPTIMIZATION OF GOLD NANOPARTICLES SYNTHESIS FOR GENTAMICIN DETERMINATION

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Gentamicin belongs to the group of aminoglycoside antibiotics, which are commonly used in human and veterinary medicine, especially in the treatment of infections caused by Gram-negative bacteria. The extensive use of aminoglycosides in farm animals leads to their accumulation in the food chain and, consequently, in the ecosystem. It is therefore necessary to develop simple and reliable analytical methods for monitoring the concentration of aminoglycosides in pharmaceutical preparations, animal-derived products, and environmental samples. Chromatographic techniques and immunoenzymatic assays are the most frequently used methods for determining these analytes. However, due to the high costs of these analyses and complex sample-preparation procedures, novel approaches are being developed to determine aminoglycoside antibiotics. Recently, methods based on the use of gold nanoparticles for their determination have been increasingly developed, particularly using colourimetric and spectrophotometric methods.

This research concerned the optimization of the gold nanoparticle synthesis process with a view to their use in the determination of gentamicin. As part of the study, appropriate concentrations of reagents (tetrachloroauric acid and trisodium citrate) required for nanoparticle synthesis were selected, considering the possibility of their application for the determination of gentamicin in the widest possible concentration range. In a next stage, the most optimal conditions for the interaction of gold nanoparticles with gentamicin were determined, including the influence of the pH of the reaction medium, the concentration of the nanoparticles, and the volume of the added analyte solution. The analytical signal was recorded spectrophotometrically and using a smartphone camera, with the R, G, B, H, S, L, I, and V channels corresponding to the components of the relevant colour model. Additionally, TEM images were taken of both the synthesized gold nanoparticles and the product of their interaction with the analyte.

# AUTOMATED PREPARATION OF A MIMETIC MEMBRANE IN A SEQUENTIAL INJECTION SYSTEM FOR EVALUATING THE MEMBRANOTROPIC EFFECT OF LORATADINE

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Emerging contaminants are of increasing concern due to their potential impact on human health and ecosystem integrity. Liposomes as membrane models enable the assessment of their toxicity, particularly regarding bioaccumulation, while reducing the need for biological membranes and live organisms in experimental studies<sup>1</sup>. The present study focuses on the automated preparation of a mimetic membrane composed of 1,2-dipalmitoylphosphatidylcholine (DPPC) and the evaluation of the effect of a widely used antihistaminic, loratadine, on their permeability using the calcein fluorescence release assay. The model membrane was prepared by rapidly injecting 60  $\mu\text{L}$  of an ethanolic lipid solution ( $150 \mu\text{L s}^{-1}$ ) into 1940  $\mu\text{L}$  of a  $62.8 \text{ mg L}^{-1}$  calcein solution, previously heated for 120 s at  $60 \text{ }^\circ\text{C}$ . Air-assisted mixing was achieved through the introduction of air during 6 s at  $150 \mu\text{L s}^{-1}$ . Then, the final liposome solution was introduced inside a dialysis membrane to remove extra-liposomal calcein. After loading the solution into the dialysis membrane, it was immersed in a vessel containing ultrapure water, which was continuously renewed by an automated circulation system. The fluorescence intensity of the liposomal suspension was measured at  $25^\circ\text{C}$  ( $\lambda_{\text{ex}} = 452 \text{ nm}$  and  $\lambda_{\text{em}} = 513 \text{ nm}$ ). Then, 50  $\mu\text{L}$  of a  $400 \text{ mg L}^{-1}$  contaminant solution was added to the liposome, and the change in the fluorescent signal intensity was measured every 30 min for 2 hours. The procedure concluded with the addition of 10  $\mu\text{L}$  of 3% Triton X-100. The fraction of released calcein was calculated according to Seiichi et al<sup>2</sup>.

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NOT ONLY „MEOW MEOW” IN M-CAT – DETECTION OF 4-MMC IMPURITIES USING  
CE-DAD

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4-Methylmethcathinone (mephedrone, 4-MMC, 'meow meow', M-Cat) is a synthetic derivative of cathinone, structurally belonging to the  $\beta$ -ketoamphetamine group. Its synthesis can be carried out using several methods, with the most popular including the oxidation of 4-methylephedrine (one-step variant) and the bromination of 4-methylpropiophenone (4-MPP) followed by the reaction of 2-bromo-4'-methylpropiophenone (4-BMPP) with methylamine (two-step variant).<sup>1</sup> During the synthesis of 4-MMC using 4-BMPP, a characteristic impurity designated by the abbreviation TMMPI is formed.

The aim of the study was to develop a method enabling the detection of substances that may occur in 4-MMC obtained by the above methods. During the study, the synthesis of one of the analytes, TMMPI, was performed, followed by its characterization using <sup>1</sup>HNMR, <sup>13</sup>CNMR, and HRMS techniques. The optimization and validation of the method were carried out using the MEKC-DAD technique. The variability of migration times of analytes over four consecutive measurement days was less than 2,2%, while the variability of peak areas was less than 15% at each of the four concentration levels. The applicability of the method was confirmed by the analysis of the mephedrone street-drug sample, revealing an intense 4-MMC peak and two other, yet unidentified signal.

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# ON-LINE CHEMICAL CLEAVAGE OF PROTEINS FOR RAPID AND SEQUENCE-SPECIFIC LC-MS IDENTIFICATION OF DIAGNOSTICALLY CHALLENGING BACTERIA

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Rapid and reliable identification of bacterial pathogens is essential for effective antimicrobial therapy, particularly in the context of increasing antibiotic resistance. Current routine methods such as MALDI-TOF-MS provide fast, cost-effective analysis but lack sequence specificity and often fail to distinguish closely related species, whereas genomic approaches, although highly specific, remain time- and resource-intensive.

This work presents a proteomic workflow based on online acid cleavage of proteins (OLACP) coupled with liquid chromatography (LC) and high-resolution mass spectrometry (MS). The key innovation is protein cleavage performed directly in the acidic mobile phase, eliminating the need for time-consuming trypsin digestion, a major barrier to the application of LC-MS proteomics in clinical diagnostics. Bacterial cells are lysed and proteins extracted using formic acid, followed by cleavage within 35 s at 195 °C in a heated capillary installed between the autosampler and the chromatographic column. This enables rapid generation of sequence-specific peptides and their LC-MS analysis within a total turnaround time below 20 minutes.

The OLACP-LC-MS pathogen identification workflow provides a fast, sensitive, and automatable solution that overcomes the main bottleneck of proteomic sample preparation, while improving taxonomic resolution and the potential to detect protein markers of antibiotic resistance, compared to MALDI-TOF-MS.

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# SMARTPHONE AS A DETECTION SYSTEM FOR TWO-COMPONENT DETERMINATION OF SILICATE AND PHOSPHATE IONS

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Portable analytical systems are becoming increasingly important in the remote monitoring of environmental water, enabling the rapid and easy determination of key pollutants. Phosphate and silicate ions are important inorganic compounds that play a key role in biogeochemical cycles and the process of eutrophication, which is why it is essential to monitor them. The spectrophotometric method based on molybdenum blue is widely used for their determination due to its high sensitivity and well-known reaction mechanism. Under strongly acidic conditions, both substances react with molybdate(VI) ions to form heteropolyacids, which can then be reduced to intensely coloured molybdenum blue complexes, enabling optical detection. Accurate measurement of phosphate(V) and silicate(IV) ions requires careful control of parameters such as acidity, reagent concentration, reaction time, temperature and potential interfering substances.

In this study, the molybdenum blue method was modified to develop a simple, two-component approach for determining phosphate and silicate ions using smartphone-based detection. Ascorbic acid was used as the reducing agent. Calibration was performed according to a 2<sup>2</sup> factorial design. To distinguish the signals and simultaneously determine both analytes, reaction parameters were selected, including reagent concentrations, acidity and reaction time. Images of the resulting solutions were recorded using a smartphone and the PhotoMetrix PRO app. The method was validated and analytical parameters were determined. The practical utility of the developed method was confirmed through the analysis of water samples.

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## COMBINATION OF DILUTION AND INTEGRATED CALIBRATION METHODS FOR ACCURATE CHROMATOGRAPHIC DETERMINATION OF SUGARS IN HONEY

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A high-performance liquid chromatography method with evaporative light scattering detection (HPLC-ELSD) was adapted, optimized, and validated for the determination of eight sugars: fructose, glucose, sucrose, turanose, maltose, trehalose, melezitose, and raffinose in honey samples. The nonlinear dependence of the measured analytical signal on concentration, as well as the possibility of various types of interference effects arising from the complex honey matrix, which can distort the results, prompted the development of a completely new calibration method. The primary goal of this approach, which combines the dilution method (DM) and the integrated calibration method (ICM), is to improve the accuracy of the results. This novel calibration method, DM-ICM, allows compensation for nonlinear and multiplicative effects. High accuracy (RE<1%) and precision (CV<3%) of the results obtained from analyses of a synthetic sample were demonstrated. In the analysis of natural honey samples, the error was reduced by up to 80% compared to results obtained using external calibration with linear and polynomial fits. The developed analytical method and methodological approach were critically assessed in terms of white analytical chemistry, greenness, analytical efficiency, practical usability, social impact, sustainability, and innovation, with emphasis on their high analytical capabilities, particularly in the context of improved accuracy.

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# SOLID PHASE PRECONCENTRATION OF BIOLOGICAL THIOLS USING GOLD NANOPARTICLES GRAFTED TO ANION EXCHANGE SUPPORT

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Biological thiols, e.g. homocysteine, cysteine, and glutathione, are associated with numerous diseases.<sup>1</sup> However, their direct analysis from non-invasive samples such as exhaled breath condensate (EBC) is challenging due to their presence in the nanomolar to subnanomolar range; thus, a preconcentration approach is required.<sup>2</sup> Our novel approach for selective preconcentration of biological thiols on gold nanoparticles introduces a flow-based microcolumn preconcentration utilizing AuNPs grafted onto solid support microparticles. After thorough screening of various materials, the strong anion exchanger (SAX) microparticles yielded optimal parameters for AuNP grafting and detection compatibility. Preconcentrated biothiols were subsequently released using a minimal volume (20 µL) of 1 mM dithiothreitol desorption agent. The preconcentrated biothiols in the eluate were derivatized and analyzed by CE-LIF. The method exhibited good inter-column variability (6.1-9.7 %) and linearity (0.985-0.999), enabling quantification of endogenous thiols in EBC samples. This novel configuration reduces sample processing time, allows for larger sample volumes (1-10 mL) and enables easy integration into automated workflows.

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INFLUENCE OF D<sub>2</sub>O ON THE DNA SEPARATION USING CE-LIF

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Capillary electrophoresis with laser-induced fluorescence detection is a standard technique for the separation of DNA fragments. Several fluorescent dyes are used for the labelling of DNA. Deuterated water used as a solvent causes less effective fluorescence quenching and lower absorbance of the visible part of the spectra in contrast to normal water.<sup>1</sup> This can lead to a higher fluorescence signal in deuterated water-based electrolytes compared to the normal ones. Deuterated water also affects the CE separation. It is capable of decreasing electroosmotic flow and Joule heating, which improves the electrophoresis process.<sup>2</sup> This work studies the influence of heavy water on separation and detection limits for capillary electrophoresis of DNA.

Selected DNA dyes and their complex with DNA were characterized using a spectrofluorometer. Fluorescence intensities of the stained DNA in normal and deuterated water were compared to select the most affected dyes for further electrophoretic experiments. The poly(n,n-dimethyl acrylamide) gel and background electrolytes in normal and heavy water were prepared. Our experiments confirm observations<sup>2</sup> about longer separation times and higher resolution in heavy water-based solutions. Also, the increase in fluorescence signal of selected DNA-dye complexes was observed.

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# CAPILLARY ELECTROPHORESIS WITH UV DETECTION FOR THE DETERMINATION OF SHORT-CHAIN FATTY ACIDS IN BIOLOGICAL SAMPLES

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Short-Chain Fatty Acids (SCFAs) are carboxylic acids essential for overall gut health, as they are the primary source of energy for colon cells, they strengthen the intestinal barrier, and regulate the local immune system. In intestinal diseases (e.g. Inflammatory Bowel Disease, IBD), an imbalance in gut bacteria often leads to alterations in SCFA levels, thus their quantification in biological matrices is useful to monitor therapy efficacy. While these acids are determined in faeces using a variety of analytical techniques, capillary electrophoresis (CE) is particularly suitable, offering fast, efficient separation.

The aim of this study was to compare a commercial CE kit with a self-developed buffer set, including the method validation according to ICH Q2 guidelines, and the determination of SCFA content in faecal samples. During method optimisation, two background electrolyte (BGE) systems were compared for their analytical efficiency. They were comprehensively evaluated using multiple criteria, including White Analytical Chemistry (WAC) models. The developed CE method achieved high sensitivity ( $LOD \leq 0.9 \mu\text{g mL}^{-1}$ ;  $LOQ \leq 3.0 \mu\text{g mL}^{-1}$ ), a wide linearity range ( $2.5 - 100 \mu\text{g mL}^{-1}$ ), high trueness ( $\leq 10\%$ ), and excellent selectivity against interfering inorganic and organic anions.

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