



BIOSENSORS

Twelfth International Workshop

01 – 03 October 2026 / Fez, Morocco

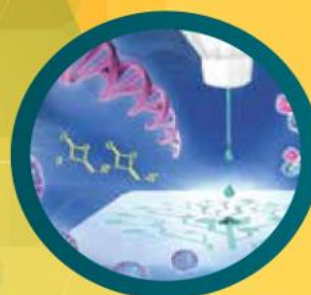


**The Twelfth International Workshop
on Biosensors**

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Workshop
Fez
2026
Biosensors



**01 – 03 October 2026
08:30 - 18:00**



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Twelfth International Workshop on Biosensors 2026



biosensors



PL 1: ULTRA-SOUND BASED 3D PRINTING MICROELECTRODE SENSING ARRAYS for BRAIN-COMPUTER INTERFACE

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Abstract:

A semi-invasive brain-computer interface (BCI) needs to simultaneously achieve high signal-to-noise ratio (SNR) neural signal acquisition and low immunological rejection risk. Existing electrode fabrication processes struggle to balance implantation precision and mechanical matching with soft brain tissue. Unlike conventional non-contact electroencephalogram (EEG) caps and fully implantable microelectrode arrays, we propose a novel fabrication strategy based on ultrasound contact-free printing of conductive hydrogels, which enables AI-regulated preparation of microelectrode arrays. This approach significantly improves the sensitivity of EEG recording, and successfully captured alpha, beta, and gamma waves through the thick pig skull in our experiments, forming a comprehensive research platform with both clinical translational potential and fundamental research value.

In this study, we used ultrasound contact-free printing to in situ fabricate conductive hydrogel electrode arrays, achieving high-precision controllable deposition of electrodes on the brain tissue surface, which addresses the issues of large implantation trauma and poor interfacial matching in traditional fabrication processes. We conducted in vivo validation on a live pig model, where the novel semi-invasive electrode successfully acquired stable, high-quality EEG signals, with mechanical compatibility and long-term stability significantly outperforming conventional rigid electrodes. On this basis, we further embedded the conductive hydrogel electrodes into brain organoid chips to construct an "on-a-chip brain" system. This platform can not only record electrophysiological signals of brain organoids in real time, but also synchronously detect neurochemical signals such as dopamine, providing multi-dimensional observation capabilities for brain development mechanism research, neuropathological analysis, and drug screening.

This study breaks through the bottlenecks of traditional processes for semi-invasive BCI fabrication. The constructed on-a-chip brain platform supports both fundamental brain science research and clinical translation of BCI technology, providing a brand-new tool for innovative development in this field.

PL 2: CATALYTIC AND AFFINITY-TYPE BIOLOGICAL RECOGNITION ELEMENTS TO USHER IN THE NEXT ERA OF IN VIVO BIOSENSORS

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Abstract:

Following the current most successful electrochemical biosensors, the sensors for continuous glucose monitors (CGMs) for diabetes management¹, the development of biosensors for continuous sensing of biomarkers and drugs, is of increasing interest to both researchers and clinicians². Since the characteristics of biosensors are highly dependent on biological recognition elements (BREs), the development of BREs, specifically designed for continuous monitoring system is the most challenging and the game-changer. BREs are categorized into two types; biocatalytic type BREs (BioCat-BREs), and bioaffinity type BREs (BioAff-BREs).

The representative BioCat-BREs are enzymes, especially oxidoreductases. Among the varieties of BioCat-BREs, the oxidoreductases which are capable of direct electron transfer with electrodes are ideal for the development of electrochemical principle based biosensors for continuous monitoring, although the number and variation of DET-type enzymes are limited³. Therefore, the creation of engineered BioCat-BREs capable of DET with electrode is an emerging challenge. For the detection of protein/peptide biomarkers in picomolar (pM) to nanomolar (nM) concentrations, current biosensor principles use BioAff-BREs such as antibodies and aptamers. The advantages of using such highly sensitive and selective BioAff-BREs are at the same time the disadvantages in using them to create a continuous monitoring system. In fact, the development of the technologies to regenerate BioAff-BREs is the most challenging task to realize biosensors for continuous in vivo monitoring of peptides/proteins^{4,5}.

In this presentation, I will discuss the challenges and strategies involved in developing DET-type BioCat-BREs and regenerable BioAff-BREs to usher in a new era of biosensors for continuous in vivo monitoring systems.

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KN 1: BIOLUMINESCENT AND COLORIMETRIC NANOBIOSENSORS FOR PORTABLE AND WEARABLE DIAGNOSTICS

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Abstract:

The development of biosensors that combine sustainability, high analytical performance, and compatibility with smartphone-based readout remains a significant challenge in modern diagnostics. Different biorecognition elements, including enzymes, whole cells, and cell-free transcription-translation systems, can be integrated into sustainable, low-cost supports to produce fit-for-purpose biosensors, including wearable formats such as skin patches and facemask-integrated devices.

This work presents the implementation of multiple strategies and sensing platforms aimed at enhancing the performance of optical biosensors based on bio-chemiluminescent and colorimetric detection, in combination with bioinspired nanomaterials. These approaches address diverse diagnostic challenges, including cystic fibrosis and skin cancer. The integration of bioluminescence with nanomaterials (e.g., metal-organic frameworks and gold nanoparticles) and with paper-based, 3D-printed, facemask-embedded, and microfluidic thread-based analytical devices (μ TADs) is reported.

Cell-based and cell-free optical biosensors developed using synthetic biology tools and interfaced with smartphone detection are described. Modular biosensing architectures employing orthogonal bioluminescent and colorimetric readouts were designed for multiplexed analysis and embedded into wearable platforms. Facemask-integrated biosensors were developed for monitoring cystic fibrosis biomarkers, specifically nitrate and quorum-sensing molecules associated with *Pseudomonas aeruginosa* infection, in exhaled breath condensate. These devices combine colorimetric detection based on the Griess reaction with bioluminescence to enable multiplexed sensing.

KN 2: From Laser to Sensor: Engineering Laser-Induced Graphene for Advanced Sensing and Biosensing

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Abstract:

Laser-induced graphene (LIG) has become a versatile material and manufacturing method for creating advanced electrochemical sensors and biosensors. It is produced by directly converting carbon-rich substrates using a laser into three-dimensional porous graphene, providing high electrical conductivity, a large electroactive surface, flexibility, and compatibility with quick, mask-free, and scalable fabrication. Its main advantage, however, is the capacity to tailor its surface and interfaces for targeted analytical uses.

This keynote lecture will present our research journey from the fundamental electrochemical characterization of LIG to the development of functional sensing and biosensing devices. Different strategies for tailoring LIG will be discussed, including surface activation, metal nanostructuring, conducting-polymer modification, and integration with molecularly imprinted polymers and aptamers. Representative applications will include the detection of electroactive molecules, pharmaceutical compounds, and clinically relevant biomarkers associated with cancer and acute myocardial infarction. Attention will be given to gold-nanostructured LIG platforms for point-of-care diagnostics, molecularly imprinted interfaces for HER2 cancer biomarker detection, and flexible, reagent-free MIP sensors for therapeutic drug monitoring. The relationships between laser-processing conditions, porous architecture, surface chemistry, recognition-layer design, and electrochemical performance will be highlighted.

Remaining challenges, including fabrication reproducibility, biofouling, long-term stability, selective biofunctionalization, and validation in complex samples, will also be examined. Finally, emerging opportunities involving wearable formats, multiplexed detection, wireless readout, and smart data analysis will be discussed. These developments demonstrate how LIG can evolve from a laser-written carbon material into an adaptable foundation for accessible, personalized, and next-generation biosensing technologies.

Keywords : Laser-induced graphene; electrochemical biosensors; molecularly imprinted polymers; nanostructured interfaces; flexible sensors; point-of-care diagnostics

KN 3: BREATH AMMONIA SENSING : CHALLENGES AND OPPORTUNITIES

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Abstract:

The potential to be able to perform non-invasive analysis by using exhaled breath as a source of physiological information remains a compelling possibility. Several trace breath gases have already found useful clinical and other applications, such as nitric oxide, acetone, isoprene and others. Breath ammonia has long been identified as an exhaled breath gas with such potential in areas including chronic liver disease (CLD), chronic kidney disease and the diagnosis of *Helicobacter pylori* (*H. pylori*) infection. However, it has faced particular challenges, both analytical and physiological. It has proven challenging to measure at clinically relevant concentrations with anything other than highly specialised instrumentation such as selected-ion flow-tube mass spectrometry (SIFT-MS). This has severely limited the number and scale of studies. Sensor-based approaches would be ideal for large-scale studies. However, these have also proved challenging to develop due to analytical demands of sensitivity, selectivity, matrix interference, etc.

Since 2005, a part of my research has been focused on the development of a breath ammonia measurement sensor system based on an impedimetric polyaniline nanoparticle sensor. An early version of this system was shown to demonstrate correlations with breath ammonia in haemodialysis patients [1]. More recently, the system was further refined and validated against SIFT-MS in healthy subjects [2]. However, in a study using patients with CLD, who often have high blood ammonia, no correlation could be established with breath ammonia [3]. These findings are consistent with previous studies that showed interference from salivary urea metabolism by oral bacteria. However, recently a large-scale study using photoionization ion mobility spectrometry has demonstrated that nose-exhaled ammonia could be used to identify *H. pylori* infection [4]. This re-opens the possibility for the application of breath ammonia sensor systems for *H. pylori* screening and diagnosis.

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KN 4 : IMPEDIMETRIC PEPTIDE-ARRAY SENSORS FOR THE DETECTION OF IGGS ASSOCIATED WITH PEANUT AND HAZELNUT ALLERGENS

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Abstract:

Food allergies can cause a broad range of adverse clinical manifestations. Food-specific immunoglobulin G (IgG) profiling at the peptide-epitope level may provide complementary information for characterizing allergen-specific immune responses. In this study, we developed a silicon-based microchip functionalized through click chemistry with allergen-derived peptides identified by the Fraunhofer Institute for Cell Therapy and Immunology. The platform was designed to investigate peptide-antibody interactions and select peptides with high specificity toward peanut- or hazelnut-associated IgGs.

The immunosensor was fabricated by electrografting diazotized aminophenylacetic acid, followed by the covalent attachment of dibenzocyclooctyne–amine (DBCO-NH₂), enabling copper-free click immobilization of azide-modified peptides. Surface functionalization was characterized by energy-dispersive X-ray spectroscopy, while sensor performance was evaluated by electrochemical impedance spectroscopy and supported by fluorescence microscopy.

Seven allergen-derived peptides were investigated using rabbit sera. The electrochemical responses identified peptides exhibiting the strongest and most selective reactivity toward anti-peanut or anti-hazelnut IgGs. The immunosensor enabled detection over serum dilutions ranging from 1:5000 to 1:500, consistent with the reference ELISA range. Microchips functionalized with peanut peptides T12 and EO13 and hazelnut peptides S4 and EO14 specifically detected nut-related IgG responses. The remaining peptides also showed promising discriminatory capabilities for developing a multivariable electrochemical fingerprint of allergen-specific IgGs. Overall, the platform provides a versatile approach for peptide screening and multiplexed characterization of immune responses associated with peanut and hazelnut allergens.

Keywords: Food allergens, immunosensor, copper-free click chemistry, rabbit serum, IgG, electrochemical impedance spectroscopy.

ACKNOWLEDGMENTS:

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KN 5 : CLINICALLY DRIVEN BIOSENSING FOR MALARIA DIAGNOSIS AND PROGNOSIS

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Abstract:

Our research develops point-of-care biosensing technologies starting from the identification of a clinically relevant unmet need and progressing through biomarker selection, assay development and clinical validation. Malaria provides a representative example of this approach. In imported malaria, patients frequently present with non-specific symptoms, creating a need not only for rapid diagnosis but also for early identification of individuals at risk of severe disease. To address this, we are developing biosensing strategies targeting both parasite- and host-derived biomarkers associated with infection, parasite burden and disease severity. Lateral flow assays are used not only as qualitative diagnostic tools, but increasingly as quantitative platforms, combining optimized assay design with digital readout to extract clinically meaningful biomarker information and support severity assessment. In parallel, electrochemical aptamer-based sensors are being explored for applications requiring more precise quantification, with particular attention to bioreceptor engineering, sensor architecture and signal transduction. Across these platforms, analytical performance is considered in the context of the intended clinical question, sample matrix and operational setting, and promising approaches are progressively evaluated using clinically relevant samples. Using malaria diagnosis and prognosis as a model, this work illustrates how clinically driven biosensor development can connect biomarker discovery with quantitative point-of-care measurement and, ultimately, create technologies with potential for translation beyond the laboratory, including through emerging commercialization initiatives.

KN 6 : DNA-BASED SYNTHETIC GENES AND CELLS FOR SENSING APPLICATIONS

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Abstract:

DNA nanotechnology uses synthetic DNA (or nucleic acids) as a versatile material to rationally engineer devices, switches and structures with different applications (e.g., in-vivo and in-vitro diagnostics, drug delivery, genetic circuits etc.). During this presentation I will introduce the field of DNA nanotechnology and I will show how to exploit the “designability” of DNA to fabricate DNA-based genes and synthetic cells that display programmable features and can be used for sensing and diagnostic applications.

KN 7 : PAPER-BASED ELECTROCHEMICAL (BIO)SENSORS AS SUSTAINABLE AND SMART DEVICES

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Abstract:

As reported in my recent perspective entitled “Paper as a Sustainable Material for Smart Electrochemical (Bio) sensors with Unprecedented Features: A Perspective” published in Analytical Chemistry [1] “The use of paper in electrochemical devices not only provides additional features to the electrochemical devices such as the environmentally friendless, ease multiplexed analysis, and three tridimensional structures by folding and unfolding operations but has broken down barriers for delivering measurement without (i) addition of reagents, (ii) sample treatment for liquid, aerosol, and solid samples, and (iii) any additional pump for microfluidics”. In this lecture, I will report on the roadmap research activity carried out in my group in the last 10 years related to the development of paper-based electrochemical (bio)sensors for delivering smart lab-on-a-chip-on-paper with unprecedented features, overcoming also the limitation of polyester-ceramic-based printed (bio)sensors and exploring new sensing applications, including electrochemical paper-based devices for periprosthetic joint infection diagnosis.

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Acknowledgements:

Financial support is recognized under the National Recovery and Resilience Plan (PNRR), Missione 6 – Salute, Componente 2 – Innovazione e ricerca sanitaria, Investimento 2.1 – Valorizzazione e potenziamento della ricerca biomedica del SSN, Avviso pubblico n. 377, finanziato dall'Unione Europea – NextGenerationEU – The diagnostic pillar for diagnosis of infections in the orthopedic field: Smart and sustainable paper-based electrochemical point-of-care device assisted by artificial intelligence– PNRR-POC-2023-12378291 CUP E83C23003450007– Decreto di Assegnazione BANDO PNRR M6/C2 2023 26 marzo 2024.



OC 1 : FUNCTIONAL NANOPORE SENSING AND SEQUENCING FOR DIAGNOSTICS

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Abstract:

Biological nanopores are proteins with transmembrane pore that can be embedded in lipid bilayer. With the development of single-channel current measurement technologies, biological nanopores have been reconstituted into membranes and used for single-molecule sensing of various analytes and events such as single-molecule DNA sensing and sequencing. We deploy various strategies to engineer nanopore proteins and develop sensing strategies to improve the sensitivity for specific analytes, including small-molecule drugs, amino acids and neuron transmitters, with potential applications in nanopore protein sequencing and nanopore-based liquid biopsy, and we constructed a copper(II)-functionalized *Mycobacterium smegmatis* porin A (MspA) nanopore with the N91H substitution, which enables direct identification of all 20 proteinogenic amino acids when combined with a machine-learning algorithm.

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Zhang M, Tang C, Wang Z, Chen S, et al, Chen L*, Geng J*. Real-time detection of 20 amino acids and discrimination of pathologically relevant peptides with functionalized nanopore. *Nature Methods*. 21(4):609-618 (2024)

OC 2: DUAL ELECTROCHEMICAL IMMUNOSENSOR FOR FLUOROQUINOLONE DETECTION IN MEAT BASED ON AN ELECTROACTIVE DERIVATIVE OF DIFLOXACIN

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Abstract:

The use of veterinary drugs has become essential in animal husbandry and the agro-industrial sector for disease prevention, treatment, and growth promotion. However, their application may lead to the presence of residues in animal-derived products such as meat, milk, eggs, and honey, thereby posing potential health risks to consumers. Although low levels of contamination may not represent a significant public health concern, the extensive and improper use of these drugs can contribute to the emergence of microbial resistance. Among veterinary antibiotics, quinolones are widely used for both therapeutic and prophylactic purposes. Therefore, the detection and quantification of even trace levels of these residues are of major importance to ensure food safety.

Enzyme-linked immunosorbent assays (ELISA) are widely employed for antibiotic analysis because of their high sensitivity, rapid throughput, and low sample volume requirements. Nevertheless, these assays also present several limitations related to enzyme activity measurements, including the presence of natural inhibitors, nonspecific enzyme binding, and temperature-dependent variations in enzyme activity. In this study, an innovative and sensitive immunosensor based on an electrochemically active quinolone derivative was developed.

Difloxacin (6-fluoro-1-(4-fluorophenyl)-7-(4-methylpiperazin-1-yl)-4-oxo-1,4-dihydroquinoline-3-carboxylic acid) was chemically modified with a ferrocene group at its carboxylic function to enable competitive detection. Polyclonal IgG antibodies were immobilized on a carbon screen-printed electrode via carbodiimide coupling following diazonium activation of the sensor surface. The fabrication process of the immunosensor was characterized using electrochemical impedance spectroscopy (EIS), differential pulse voltammetry (DPV), and cyclic voltammetry (CV).

A one-step competitive assay was then established for the determination of four quinolone compounds in buffer solution. The method consisted of pre-mixing 50 μ L of sample with 50 μ L of ferrocene-modified difloxacin. The analytical performances obtained were consistent with the maximum residue limits established by the European Union (100 μ g/kg). Finally, the efficiency of the immunosensor and the suitability of the extraction protocol were validated through the analysis of 5,400 meat samples.

Keywords: Immunosensor, electroactive derivatives, Enrofloxacin, Fluoroquinolones, Competitive assay, Screen-printed dual carbon electrodes (SPdCEs), Meat analysis.

OC 3 : ELECTROCHEMICAL BIOSENSORS FOR FOOD CONTAMINANTS DETECTION: FROM QUATERNARY AMMONIUM COMPOUNDS TO OCHRATOXINE A

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Abstract:

Food safety remains a major global challenge due to contaminants that may affect human health and compromise product quality. Food contamination can arise from biological hazards, physical pollutants, and chemical contaminants, including pesticides, disinfectants, heavy metals, and mycotoxins. The increasing complexity of food production chains requires analytical tools that are rapid, sensitive, cost-effective, and suitable for routine monitoring.

Among the available analytical approaches, electrochemical biosensors have emerged as promising alternatives to conventional laboratory methods. By combining a biological recognition element with an electrochemical transducer, they enable rapid and selective detection of target analytes. Their advantages include high sensitivity, portability, low-cost fabrication, and suitability for on-site analysis. Recent advances in bioreceptor engineering and sensor miniaturization have expanded their applications in food safety.[1]

This presentation highlights recent developments in electrochemical biosensing for food contaminant detection. Particular attention will be paid to monitoring quaternary ammonium compounds (QACs), such as benzalkonium chloride (BAC) and didecyltrimethylammonium chloride (DDAC), widely used as disinfectants in the agri-food sector. Enzymatic biosensors based on cholinesterase inhibition represent a promising approach for detecting these compounds at concentrations relevant to food safety control.[2]

In addition, the design of a peptide-based electrochemical biosensor for the detection of ochratoxin A (OTA) will be presented. OTA is a widespread mycotoxin frequently found in cereals, coffee, dried fruits, wine, and other food products. Due to its nephrotoxic, immunotoxic, and potentially carcinogenic effects, its monitoring remains a major concern. The proposed strategy uses synthetic peptides as recognition elements, offering high stability, ease of synthesis, tunable affinity, and lower production costs than conventional bioreceptors. Their integration into electrochemical platforms opens new opportunities for rapid OTA screening in food matrices. These examples illustrate the versatility of electrochemical biosensors and the potential of emerging bioreceptors for next-generation food safety monitoring.

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OC 4 : COMBINING PIEZO- AND CHEMO-RESISTIVE NANOCOMPOSITE SENSORS TO MINIMIZE PRESSURE ULCERS DEVELOPMENT

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Abstract:

Early detection of bedsores (pressure ulcers) is expected to improve the health of disabled patients and save hospital costs [1]. We are presenting an original strategy that combines the different smart properties of conducting polymer nanocomposites (CPC) for the monitoring of the pressure applied on skin with piezo-resistive sensors and the volatiles emitted by bedsores as a function of their evolution with chemo-resistive sensors. On the one hand quantum resistive pressure sensors (pQRS) [2], were developed by building a graphene / carbon nanotubes conducting architecture into a thermoplastic poly(urethane) matrix. Sixteen of these pQRS were subsequently assembled into an array to map the surface pressure applied to skin. On the other hand, an artificial nose (e-nose) of quantum resistive vapour sensors (vQRS) [3], fabricated by spray layer by layer from nine types of functionalized carbon nanotubes, was designed to be sensitive to three volatiles, identified as biomarkers of bedsores [4]. Both types of sensors were found highly sensitive respectively to pressure (some kPa) and vapours (some 100 ppb) suggesting that these promising results represent a credible first step towards the prevention of bedsores that will require further data collection and analysis to optimise an effective diagnostic algorithm.

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OC 5 : HPV TESTING: A SOLVED PROBLEM?

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Abstract:

HPV DNA testing is a well-established tool complementing Pap smears in cervical cancer screening. PCR-based testing allows for high-risk HPV (HR-HPV) DNA detection and direct genotyping of the most common high-risk strains. However, in many countries, PCR-based tests remain too expensive for regular preventive screening and most commercial assays are certified exclusively for cervical smears. Our latest studies focus on non-invasive HPV detection from urine samples for cervical cancer screening, and from oral samples and plasma for oropharyngeal cancer disease monitoring. From a methodological point of view, we use isothermal amplification and electrochemical readout, with the aid of qPCR and digital PCR as validation methods. Cervical cancer remains a highly prevalent oncological diagnosis worldwide. Although screening and vaccination programs have reduced incidence, many at-risk women do not undergo regular gynecological examinations. Non-invasive urine testing represents a promising alternative, offering the potential for home self-sampling. This approach could significantly increase screening uptake and coverage among these underserved populations [1].

Recently, HPV has also gained interest in head and neck oncology. HPV-related oropharyngeal tumors present a more favorable prognosis and higher treatment sensitivity than HPV-negative cases. [2]. Given the higher radiosensitivity of HR-HPV-positive oropharyngeal cancers, radiotherapy doses can be safely de-escalated to mitigate severe adverse effect, as radiation dermatitis [3]. For these applications, we build upon our previous experience with cervical precancer detection [4] by developing new detection strategies combining (i) recombinase polymerase amplification (RPA) as an isothermal amplification reaction (ii) amperometry as a common end-point technique and (iii) electrode chips for rapid and parallel measurement of multiple sample.

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OC 6 : ELECTROCHEMICAL DETECTION OF HUMAN CYTOMEGALOVIRUS IN GLIOBLASTOMA: A COMPARATIVE MULTI-METHOD STUDY

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Abstract:

Human cytomegalovirus (hCMV) acts as an oncomodulator in glioblastoma IDH-wildtype (GBM), promoting tumor progression by altering oncogenic signaling pathways, suppressing anti-tumor immunity, and remodeling the microenvironment. While targeting hCMV via adjuvant antivirals improves patient survival, determining true viral presence remains a challenge due to the lack of highly sensitive, standardized tools validated for oncological tissue. This study aims to combine multi-level biological analysis (DNA, RNA, and protein) with a novel electrochemical (EC) bioassay to reliably map viral presence and overcome the limitations of traditional diagnostic standards.

We developed an EC biosensor using loop-mediated isothermal amplification (LAMP). During isothermal amplification, biotinylated dUTPs are incorporated into the viral amplicons. These target strands are selectively hybridized to sequence-specific capture probes immobilized on carboxylic magnetic beads. Following magnetic washing, the complex is enzymatically labeled with a streptavidin-peroxidase polymer (SPP). Amperometric readout is performed via hydroquinone/H₂O₂ mediation on an 8-electrode carbon array chip.

The clinical utility of the EC bioassay was applied on a cohort of 113 GBM patient samples, directly comparing performance across formalin-fixed paraffin-embedded (FFPE) and fresh-frozen tissues evaluated by standard qPCR, IHC, and CISH. The findings of this comprehensive comparative analysis aim to identify optimal approaches for hCMV screening in solid tumors and thereby contribute to the development of personalized medicine with the aim of prolonging patient survival.

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OC 7 : NEW NANOMATERIAL AND POLYMER FILM MODIFIED ELECTRODES FOR SENSORS AND BIOSENSORS PREPARED IN TERNARY DEEP EUTECTIC SOLVENTS

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Abstract:

Modification of carbon-based electrodes by nanomaterials can lead to enhanced sensor and biosensor characteristics owing to electrocatalytic effects and to increased active surface area. This can be improved even more by further modification with coatings of electroactive polymer coatings. Recent research in this regard has involved preparing polymer coatings arising from the electropolymerisation of redox dyes, such as phenazines or triarylmethanes. Nanomaterials that have been employed include carbon nanotubes (MWCNT), graphene quantum dots and metal or metal oxide nanoparticles [1].

Binary and ternary deep eutectic solvents (DES) have been evaluated as media for polymerisation to prepare such polymer-modified electrodes, and compared with synthesis in aqueous solution, since it is generally found that use of DES leads to better sensor and biosensor analytical parameters [1]. Amongst the DES, choline chloride has almost always been used as the hydrogen bond acceptor (HBA). Hydrogen bond donors (HBD) have focused on ethylene glycol that yields the relatively non-viscous DES, ethaline. DES with two HBD, ternary DES, have been found to influence the polymer film voltammetric behaviour according to the relative amount of each HBD. The ternary DES usually include ethylene glycol as one HBD and different percentages of urea, oxalic acid, acetic acid or fructose as second HBD.

Studies of modified electrode platforms based on poly(neutral red), poly(brilliant cresyl blue), poly(cresyl violet) and poly(methylene blue) on gold nanoparticles [2], MWCNT [3,4] or graphene quantum dots [5] plus investigating the influence of water in the DES on the polymer film properties [6] have been chosen to illustrate the approach taken. Characterization was by electrochemical and surface analysis techniques. Better performance when using ternary rather than binary DES in the application of these modified electrodes as enzyme biosensors and as sensors and is achieved in terms of sensitivity, selectivity and detection limits, shown for the determination of pharmaceuticals and in food quality monitoring.

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OC 8 : INHIBITION OF AMYLOID- β PEPTIDES FIBRILIZATION BY CURCUMIN: AFM AND ELECTROCHEMICAL CHARACTERISATION

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Abstract:

Alzheimer's disease (AD) disrupts the brain's ability to function by impairing the communication between neurons, primarily caused by the build-up of amyloid beta ($A\beta$) plaques. Structurally, the $A\beta$ peptides contain only three electroactive amino acid residues, tyrosine Y¹⁰, three histidines H⁶, H¹³ and H¹⁴, and methionine M³⁵. Atomic force microscopy (AFM) and differential pulse voltammetry (DPV) have been recognised as fruitful approaches to study the $A\beta$ peptides' structure and aggregation mechanisms [1, 2]. The naturally occurring polyphenol curcumin [3, 4] found in turmeric has strong affinity for binding to $A\beta$ peptides. Its structure allows curcumin to interact with the hydrophobic regions of $A\beta$ peptides, which are crucial for β -sheet formation, inhibiting $A\beta$ aggregation and fibril formation, destabilizing existing plaques, and reducing neurotoxicity. The time-dependent structural modifications and electrochemical behaviour of human $A\beta_{1-40}$ and $A\beta_{1-42}$ peptides following their interaction with curcumin, using AFM and DPV, was investigated. The results showed that curcumin disrupts and slows the fibrillization process by binding to specific aromatic residues of the $A\beta$ peptides, via curcumin methoxy and hydroxy groups. This dual ability of curcumin to slow fibril formation and induce the formation of unconventional $A\beta$ monomeric, oligomeric, prefibrillar, and flexible fibrillar species, may reduce the toxicity associated with soluble oligomers and fibrillar $A\beta$ aggregates, highlighting curcumin's potential as a therapeutic agent for AD.

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OC 9 : ELECTROCHEMICAL GENOSENSORS FOR HEATHER HONEY DNA AUTHENTICATION USING SWCNT-MODIFIED ELECTRODES

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Abstract:

Honey is a natural sweetener with a rich nutritional profile and several health-promoting properties that are strongly linked to its botanical origin. With the growing number of food fraud cases, high-quality products such as honey are susceptible to fraudulent practices, e.g., the mislabeling of its botanical and/or geographical origin. These practices not only compromise consumer confidence, product quality, and fair market competition, but are also difficult to detect using conventional analytical methods. Therefore, there is a need to develop a reliable analytical tool capable of proving the honey's botanical authenticity and distinguishing between the authentic and adulterated honey products. In this study, an electrochemical DNA-based biosensor was developed for the detection of heather DNA (a prominent honey plant species in Portugal) in honey samples. The sensing strategy is based on a sandwich hybridization assay between a heather-specific DNA target sequence and its complementary probe immobilized on screen-printed carbon electrodes (SPCEs). To enhance sensor's performance, the complementary probe was divided into two shorter sequences: one modified with an amine group (the DNA-capture probe) and the other with a fluorescein isothiocyanate label (the DNA-signaling probe). Additionally, the electrode surface was modified with single-walled carbon nanotubes (SWCNTs), increasing the electroactive surface area, as well as improving the electron transfer efficiency. Both the surface modification and sensor assembly were optimized to maximize the electrochemical signal response. The optimized biosensor presented an improved electrochemical signal following each optimization step, consequently, demonstrating the effectiveness of the proposed approach. Overall, the developed genosensor represents a promising alternative to the conventional analytical techniques for the authentication of heather species-specific DNA in honey samples.

Acknowledgements: This work received financial support from the PT national funds (FCT/MECI) through the project UID/50006/2025 DOI 10.54499/UID/50006/2025 - Laboratório Associado para a Química Verde - Tecnologias e Processos Limpos and from the project "Challenging the Analytical Diagnosis of Neurodevelopmental Disorders: Pioneering Neurochemical Devices", funded by FEDER funds and PT funds by FCT/MECI, operation no. 15875, with reference COMPETE2030-FEDER-00699000. Stephanie Morais (2023.028929.BD) is grateful to FCT and the European Union (EU) for their grants financed by POPH-QREN-Tipologia 4.1-Formação Avançada, funded by Fundo Social Europeu (FSE) and Ministério da Ciência, Tecnologia e Ensino Superior (MCTES). M.F.B. thanks the TENURE – FCT Tenure Program – 1 st Edition (Reference 2023.15797.TENURE.001) with financial support from FCT/MCTES through national funds.

OC 10 : ADVANCEMENTS IN BIOSENSORS FOR EARLY DETECTION OF TARGET PROTEINS

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Abstract:

A molecular wire refers to a molecular chain with extended electronic conjugation, enabling strong coupling between the donor and acceptor, such as molecules or electrodes attached to the chain ends. Peptide backbones can serve as effective molecular wires for electrochemical biosensors if they fulfill three main criteria: efficient charge transport, enhanced chemical stability, and high conformational flexibility. The electron transfer mechanism is influenced not only by the chain length but also by factors such as the extent of secondary structure, dipole orientation, hydrogen bonding, redox potential, and specific amino acid residue.¹

Herein, we present a multifunctional sensor platform that utilizes a peptide molecular wire as a conductive support for immobilizing ligands in affinity-based bioassays. A short helical peptide sequence with alternating polar and ionizable side chains was synthesized and functionalized with a redox marker, Methylene Blue (MB), at one end. It was then anchored to the gold surface through gold/thiol chemistry². The modified peptide acted as a molecular wire and facilitated a two-step electron transfer process from the redox marker to the gold surface³. The electrode reaction of MB at the peptide-supported layer, represented by the typical peak-shaped voltammetric curves characteristic of MB signaling, has been studied in the context of the two-step electron transfer (EE) mechanism. The kinetic data have been estimated by comparing the experiment with numerical simulations under conditions of square wave voltammetry while approximating the electrode reaction with a diffusionless, surface-confined electrode process in the absence of significant lateral interactions.

The developed platform successfully detected two targets, the anti-tumor-associated carbohydrate antigen-antibody and the growth hormone secretagogue receptor, via affinity interactions with their small ligands grafted onto the peptide wire. Our findings demonstrate the efficacy of the peptide molecular wire in electrochemical biosensors and imply potential applications in biomedicine and biosensing.

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OC 11 : MULTIFUNCTIONAL POLYPYRROLE COMPOSITE INTERFACES FOR WEARABLE BIOELECTRONICS AND SENSING

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Abstract:

Flexible and wearable bioelectronic systems require multifunctional materials that combine electrical conductivity, mechanical flexibility, structural stability, and adaptability to diverse sensing environments. In this work, we present the development of multifunctional polypyrrole (PPy)-based composite interfaces designed for wearable bioelectronics and sensing applications. The materials were developed by incorporating various functional fillers and polymeric additives into conductive PPy matrices, allowing the tuning of electrical, mechanical, and surface properties according to the intended application. The developed composite films exhibit flexible architectures with improved conductivity, enhanced mechanical stability, and good conformability to soft and dynamic surfaces, making them suitable for wearable sensing platforms. Particular attention was dedicated to the relationship between composition, morphology, and functional performance, highlighting the role of conductive and structural components in modulating the sensing response.

The bioelectronic potential of the materials was demonstrated through electrocardiographic (ECG) sensing experiments, where the flexible PPy-based interfaces enabled stable acquisition of physiological electrical signals under wearable conditions. Beyond ECG monitoring, the multifunctional nature of the composites opens perspectives for broader applications including piezoresistive motion sensing, smart conductive textiles, and protective wearable systems requiring both electrical functionality and flame-retardant behavior.

The presented approach emphasizes the versatility of conductive polymer composites as adaptable platforms for next-generation wearable bioelectronics. By combining tunable physicochemical properties with flexible and lightweight architectures, PPy-based multifunctional interfaces represent promising candidates for the development of integrated sensing systems, soft biointerfaces, and advanced wearable technologies.

OC 12 : PROGRESS IN THE DEVELOPMENT OF ELECTROCHEMICAL (BIO)SENSORS FOR ORGANIC CHEMICAL CARCINOGENS IN FOOD MATRICES AND FOOD QUALITY CONTROL ACCORDING TO THE LEGISLATION

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Abstract:

The International Agency for Research on Cancer (IARC) of WHO identifies 122 human carcinogens and 93 probable human carcinogens classified into 4 groups of evidence for the carcinogenicity. Together with traditional food safety testing methods performed in sophisticated labs, rapid detection methods and use of sensors for the identification and *in-situ* and real-time *on-site* monitoring of potential food contaminants becomes crucial. Electrochemical (EC) biosensors meet the required parameters offering simple operation, fast response, portability, sensitivity, selectivity, low cost, easy miniaturization and ability of integration into automatic systems without compromising analytical characteristics [1-3]. In this work, progress in the EC (bio)sensors for the detection of WHO indicated carcinogens including mycotoxins is treated. Novel building components of the biosensors, mostly as the nanotechnology products such as nanocomposites, metal-organic frameworks (MOFs), zeolitic imidazolate frameworks (ZIFs), and covalent organic frameworks (COFs) are categorized together with novelties in immune-, aptamer- and MIP-based recognition schemes, dual-detection procedures and ratiometric measurement. The EC sensing devices reach relevant concentration levels stated by the European Commission and EFSA legislation. Advantages and disadvantages of the EC approaches for assays in real food samples are critically evaluated. To facilitate the development of more reliable, robust and in food quality control applicable EC techniques, emerging trends and recommendations are stated.

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OC 13 : CLAMPING THE LAMP: A PHOTOELECTROCHEMICAL PLATFORM FOR SELECTIVE KRAS MUTATION DETECTION THROUGH WILD-TYPE SUPPRESSION

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Abstract:

KRAS mutations are among the most clinically relevant oncogenic alterations in colorectal, lung and pancreatic cancer. However, their accurate detection remains challenging due to the overwhelming abundance of wild-type (WT) DNA typically present in clinical samples. Here, we present a photoelectrochemical (PEC) biosensing platform that integrates clamp-inhibited loop-mediated isothermal amplification (C-LAMP) with enzyme-free singlet oxygen (¹O₂)-driven PEC transduction for selective *KRAS* mutation analysis.^[1]

The strategy employs locked nucleic acid (LNA) clamp probes to selectively suppress WT amplification during isothermal amplification, thereby enriching mutant alleles and improving single-nucleotide variant discrimination. Following amplification, *KRAS* mutation-specific products are captured on magnetic microbeads and hybridized with photosensitizer-labelled detection probes. Upon visible-light illumination, the photosensitizer generates ¹O₂, which drives hydroquinone/benzoquinone redox cycling and produces a photocurrent proportional to the amount of mutant amplicons, eliminating the need for enzymatic substrates and simplifying the analytical workflow.

The integrated C-LAMP/PEC platform achieved a limit of detection of 35 copies/μL (58 aM) and enabled detection of variant allele frequencies as low as 4.8%. Compared with conventional LAMP, incorporation of LNA clamp probes substantially enhanced mutant-to-WT discrimination by effectively suppressing non-target amplification. While quantitative PCR (qPCR), droplet digital PCR (ddPCR) and next-generation sequencing (NGS) remain the clinical standards for *KRAS* genotyping, the proposed C-LAMP/PEC approach offers a simplified isothermal workflow with reduced instrumentation requirements and compatibility with decentralized testing. The methodology was successfully validated using cancer cell lines and patient-derived fresh frozen tissues, showing complete concordance with ddPCR and Nanopore sequencing within the evaluated cohort.

These results demonstrate that combining molecular WT suppression with enzyme-free PEC transduction provides a robust, sensitive and instrumentation-simplified strategy for clinically relevant *KRAS* mutation detection, highlighting the potential of photoelectrochemical biosensing for decentralized molecular diagnostics.

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OC 14 : SINGLET OXYGEN-BASED PHOTOELECTROCHEMISTRY FOR INFECTIOUS DISEASE DIAGNOSTICS

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Abstract:

The global incidence of arbovirus outbreaks, e.g. dengue, has been rising at an alarming rate. Such viruses are transmitted by mosquitoes in tropical and subtropical regions and are responsible for a significant public health burden. Importantly, population growth, urbanisation and climate change led to an increase in the spread of arboviral diseases. Hence, diagnosing these viral infections is crucial to reducing the spread and disease burden. However, conventional diagnostic tools are often expensive and time-consuming (i.e. reverse-transcription polymerase chain reaction) or lack accuracy and sensitivity (i.e. rapid diagnostic tests).

To overcome such challenges, we developed an innovative singlet oxygen (1O_2)-based photoelectrochemical biosensing platform for detecting viral RNA related to infectious diseases. Our technology combines (i) photosensitisers that produce 1O_2 upon illumination, (ii) a redox reporter, and (iii) magnetic beads for immobilisation and enhancing sensitivity.^{1,2} Moreover, the technology allows direct RNA detection without conversion to DNA³ and eliminates matrix effects because of a light on/off switching mode. This ensures that the technology is rapid and easy to use, allowing its use at the point of care.

Here, we present an overview of the work regarding infectious disease diagnostics. This covers various challenges unique to this application: (i) the direct detection of viral RNA which contains stable secondary structures and requires rational probe design, and (ii) the detection of the viral genome that spans thousands of nucleotides by employing DNazymes. Additionally, we developed a multiplexing strategy that allows the analysis of multiple nucleic acid sequences in one sample by using multiple photosensitisers. This makes the photoelectrochemical technology suitable for developing an on-site test that can detect multiple pathogens simultaneously.

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OC 15 : Direct single-molecule detection of CoA-SH and ATP by the membrane proteins TMEM120A and TMEM120B

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ABSTRACT

Human TMEM120A and its homolog TMEM120B are members of the human transmembrane protein family 120. Recent studies have implicated them in pain perception, resistance to Zika virus infection, and modulation of lipid metabolism. With the elucidation of their protein structures, both proteins have been found to bind coenzyme A (CoA).

In this study, using single-channel electrophysiological patch-clamp techniques, we demonstrated that both proteins possess the ability to conduct ions when reconstituted into artificial lipid membranes, and characterized their potassium channel activities. At the single-molecule level, we verified the binding capacity of both proteins to CoA and performed single-molecule kinetic analyses of the binding events. We observed that both proteins can bind adenosine triphosphate (ATP), leading to substantial conformational changes in the channels. We further explored the effects of different cationic species on the interactions of the two proteins with CoA and ATP, finding that Mg^{2+} enhances the interaction between ATP and the channel proteins while attenuating CoA binding. Given that the interactions of these two channel proteins with CoA and ATP can be directly detected at the single-molecule level, we developed these proteins into single-molecule biosensors for CoA and ATP detection.

In summary, this study fills a gap in single-molecule-level research on TMEM120 transmembrane proteins and provides methodological insights for further elucidating the complex physiological functions and signaling pathways involving these two proteins. Given that CoA and ATP concentrations play important guiding roles in the clinical diagnosis of various diseases, these biosensors hold promise as novel detection tools for precision clinical diagnostics, adjunctive medication guidance, and intraoperative point-of-care testing in the future.

Keywords:

TMEM120A/TMEM120B; Biosensor; Single-molecule detection

OC 16 : RESEARCH ON TUMOR CELL CFDNA DETECTION BASED ON THE AUTOCATALYTIC EFFECT OF THE CRISPR/CAS12A SYSTEM

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Abstract:

The CRISPR-Cas system, derived from the adaptive immune system of bacteria, possesses dual functionalities of precise target nucleic acid recognition and highly efficient nucleic acid cleavage, thereby demonstrating broad application prospects in biosensing and clinical diagnostics. However, existing CRISPR-Cas-based detection strategies still suffer from several limitations, including redundant detection architectures, limited signal amplification efficiency, and the requirement for complex nucleic acid structural designs. To more effectively exploit the dual capabilities of Cas12a in nucleic acid recognition and cleavage, this study introduced two innovative mechanisms: programmable activation of Cas12a using split single-stranded DNA (ssDNA) activators and site-specific regulation of Cas12a trans-cleavage activity through locked nucleic acid (LNA) modification. Based on these strategies, we developed a novel programmable nucleic acid circuit termed CRISPR-Cas Autocatalysis Amplification driven by LNA-modified Split Activators (CALSA), which enables self-catalytic positive-feedback cascade signal amplification mediated by Cas12a. Using the CALSA platform, synthetic genomic DNA targets were employed to systematically evaluate its signal amplification capability, detection sensitivity, and nucleic acid recognition specificity. Compared with conventional direct CRISPR/Cas12a detection methods, CALSA achieved an approximately three-orders-of-magnitude enhancement in signal amplification efficiency, reached femtomolar-level sensitivity within less than one hour, and exhibited ultrahigh specificity capable of discriminating single-base mutations. Building upon these findings, the CALSA strategy was further applied to the real-time detection of cell-free DNA (cfDNA) secreted by different tumor cell lines, enabling accurate quantitative identification of the breast cancer cell line MCF-7. These results not only demonstrate the significant potential of the CRISPR/Cas12a-based CALSA nucleic acid circuit for point-of-care diagnostics, but also highlight its substantial value in the development of programmable logic circuits and advanced biosensing technologies owing to its remarkable programmability.

OC 17 : SURFACE PLASMON RESONANCE SCREENING STUDY OF MANNOSE-FUNCTIONALISED GOLD NANOPARTICLES ON LECTINE INNOVATING BIOINTERFACE

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Abstract:

Gold Nano-Particles (GNP) were designed for combining several medical imaging modalities with therapeutic activities. In order to better exploit their potential, we are exploring different ways of functionalization for active targeting. There is therefore a real need to develop efficient test for checking their ability for active targeting. The aim of the project is to establish an original biointerface able to test the ability of mannose-functionalised gold nanoparticles (GNP-Man) to specifically interact with the lectin, Concanavalin A (ConA).

We started from naked gold SPR chips by performing in situ functionalisation of self-assembled monolayer (SAM) of a thioctic acid derivative, characterized by an endocyclic disulfide bond, linked to the macrocycle DOTAGA, named TADOTAGA. The monitoring of SAM formation has been characterized by SPR leading to efficient and reproducible process enable to provide robust chemical layer bearing anchor sites for upstream biofunctionalization. Such a platform has enabled the covalent grafting of mannose under optimized conditions determined on the basis of qualitative and quantitative interactions with ConA. Due to the tetrameric structure of the lectin, the mode of immobilisation offers a smart ligand-based biointerface with good stability, strong regenerative capacity and available sites for analyte-based mannose as GNP-Man. Different batches of GNP-Man have been produced and deeply characterized by TEM, DLS and spectroscopic absorbance in UV-visible range. 5 populations of GNP-Man have been produced named as GNP-xMan with x= 10, 20, 50, 100 and 200 depending of molar ratio "Mannose/GNP" during the sugar functionalisation step. A dose-response SPR study has been conducted with the GNP-10Man on fresh ConA biointerfaces between 15 µg/mL to 300µg/mL that validates the complete architecture of the supramolecular assembly and the methodology. From this study, we launched the screening method for all the populations at 30µg/mL and performed a comparative analysis that demonstrated a good correlation and specificity of the response signal in function of the theoretical density of mannose per GNP and with the control GNP-0Man.

To conclude, we have developed a biomolecular architecture on SPR gold chips that acts as a screening platform for batches of GNPs coated with varying densities of mannose, demonstrating its effectiveness in distinguishing targeting properties. Further experiments are engaged to deeper study the kinetics of the interactions and the targeting properties of GNP-xMan in order to contribute to the selection of the best hit for theranostic purposes.

OC 18 : A MULTIPARAMETRIC ELECTROCHEMICAL SENSOR PLATFORM FOR POINT-OF-CARE DIAGNOSIS OF PERIPROSTHETIC JOINT INFECTION

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Abstract:

Periprosthetic joint infection (PJI) is a clinically complex complication of joint arthroplasty arising from the interplay among microorganisms, the prosthetic implant, and the host immune response. Early diagnosis is essential to prevent severe complications and progression to chronic infection. To this end, standardized diagnostic criteria identify synovial white blood cell (WBC) count as a key diagnostic marker, while emerging biomarkers, including pH and lactate, have been proposed as complementary parameters⁽¹⁾. In this work, we present a paper-based, screen-printed electrochemical platform for the multiparametric detection of PJI-associated biomarkers. The system integrates paper-based microfluidics to enable the simultaneous analysis of WBC count, pH, and lactate. It includes an impedance-based sensor for WBC quantification by electrochemical impedance spectroscopy (EIS), a potentiometric pH sensor based on a carbon black/polyaniline (CB/PANI) composite, and an amperometric lactate sensor based on a carbon black/Prussian blue (CB/PB) -modified electrode functionalized with lactate oxidase immobilized through EDC/NHS coupling chemistry. Data analysis is supported by chemometric approaches, including partial least squares (PLS) and principal component analysis (PCA). The analytical performance of each sensing component was initially evaluated in standard solutions. WBC measurements were performed in phosphate-buffered saline (PBS) containing 10 μm beads as WBC-mimicking particles. The resulting calibration curve was $y = (25.65 \pm 2)x - (48.11 \pm 7)$, with $R^2 = 0.925$. The pH sensor exhibited a linear response within the physiological range, described by $y = (-0.044 \pm 0.005)x + (0.310 \pm 0.004)$, with $R^2 = 0.970$. For lactate detection, the corresponding calibration curve was $y = (1.18 \pm 0.07)x - (2.15 \pm 0.08)$, with $R^2 = 0.947$. Preliminary measurements on synovial fluid samples were consistent with reference laboratory methods, supporting the feasibility of a low-cost, multiparametric point-of-care platform for rapid PJI assessment.



Acknowledgements: Financial support is recognized under the National Recovery and Resilience Plan (PNRR), Mission [Missione 6 – Salute, Componente 2 – Innovazione e ricerca sanitaria], Investimento 2.1 – Valorizzazione e potenziamento della ricerca biomedica del SSN, Bando [Avviso pubblico n. 377], finanziato dall'Unione Europea – NextGenerationEU – Titolo Progetto [The diagnostic pillar for diagnosis of infections in the orthopedic field: Smart and sustainable paper-based electrochemical point-of-care device assisted by artificial intelligence]– PNRR-POC-2023-12378291 CUP E83C23003450007– Decreto di Assegnazione BANDO PNRR M6/C2 2023 26 marzo 2024.

Reference: ^[1]Müller, Dirk, et al. "Synovial pH, lactate, and glucose as adjunct biomarkers to exclude periprosthetic joint infection in hip and knee arthroplasty: a prospective study." *Journal of Bone and Joint Infection* 10.4 (2025): 293-301.

OC 19 : PAPER-BASED ELECTROCHEMICAL (BIO)SENSOR ARRAY FOR REAL-TIME CONTINUOUS MONITORING INTO ORGAN-ON-CHIP PLATFORMS

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Abstract:

The demand for physiologically relevant in vitro models has driven the rapid advancement of Organ-on-Chip (OoC) technology, which offers a powerful alternative to traditional monolayer cell cultures and animal models. While various analytical approaches have been explored to monitor these platforms, there remains a critical need for integrated, cost-effective, and sustainable solutions that enable continuous, real-time monitoring of the cellular microenvironment without requiring invasive sampling or destructive end-point analyses. In this context, electrochemical (bio)sensors have emerged as a promising technology, offering high sensitivity and portability; however, their seamless integration into versatile, low-cost microfluidic architectures continues to be a key objective for next-generation biomedical devices.

In this work, we present a novel screen-printed paper-based electrochemical (bio)sensor array for the simultaneous monitoring of seven key metabolic biomarkers relevant to cell metabolism and tissue function. The platform employs a tailored functionalization strategy for each analyte detection: iridium oxide film for pH; carbon black–Prussian blue nanocomposite for glucose and lactate, via N-(3-Dimethylaminopropyl)-N'-ethylcarbodiimide and N-Hydroxysuccinimide (EDC-NHS) covalent immobilization of oxidase enzymes; carbon black nanoparticles for lactate dehydrogenase, dissolved NO, and dissolved O₂; and ion-selective membrane for nitrate.

Analytical performance was evaluated in both standard solutions and cell culture medium, demonstrating high reproducibility (RSD% 2–8%) and accuracy (recovery 95–105%). By harnessing paper as a multifunctional material, a paper-based passive microfluidic platform was integrated with the (bio)sensors. The potentiometric sensors exhibited good sensitivity, equal to $-0.080 \pm 0.002 \text{ V pH}^{-1}$ and $-0.054 \pm 0.002 \text{ V } (\log a_{\text{NO}_3^-})^{-1}$, recovery values of 95%

and 96% with bias% range within -6% and -4% for pH and nitrate, respectively. For amperometric detection, the glucose (bio)sensor displayed a sensitivity equal to $0.18 \pm 0.02 \mu\text{A mM}^{-1}$, a LOD equal to 0.3 mM and 86% recovery. Lactate and lactate dehydrogenase were also characterized using microfluidic platform, yielding sensitivities equal to $0.26 \pm 0.01 \mu\text{A } (\log \text{mM})^{-1}$ and $-1.13 \pm 0.08 \mu\text{A } (\text{U mL}^{-1})^{-1}$, respectively.

These results demonstrate the feasibility of embedding sustainable, high-performance sensors into unconventional microfluidic devices, toward the paper-based application within the OoC world, providing a scalable strategy for real-time metabolic monitoring.

Acknowledgements: PHOENIX-OoC Project, funded under the European Union's Horizon Europe EIC 2023 Pathfinder Open program, Grant Agreement No. 101130395

OC 20 : THIOL-MEDIATED ELECTROCHEMICAL SENSOR FOR THE BREAST CANCER BIOMARKER CA 15-3 AS A FIRST STEP TOWARDS MULTI-ELEMENT BIOMARKER DETECTION

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Abstract:

Cancer antigen 15-3 (CA 15-3), a circulating fragment of the MUC1 glycoprotein, is widely used for monitoring breast cancer progression. In this study, we report a label-free, receptor-free electrochemical detection strategy based on adsorption-mediated modulation of gold surface oxidation. Pencil graphite electrodes (PGEs) were modified with electrochemically deposited gold nanoparticles (AuNPs) using a reproducible multi-electrode fabrication system. The nanostructured AuNP surface exhibited enhanced electroactive area and improved electron-transfer properties, as confirmed by ferri/ferrocyanide characterization. Analytical detection was performed in phosphate-buffered saline by monitoring the intrinsic gold oxidation peak. Following incubation with CA 15-3, a concentration-dependent decrease in the gold oxidation current was observed, attributed to adsorption-induced surface passivation of electrochemically active gold sites. The platform demonstrated a linear response across the tested concentration range with a calculated limit of detection of 1.2×10^{-4} U/mL. Measurements in spiked plasma samples confirmed the feasibility of detecting CA 15-3 in complex matrices under controlled conditions. This work establishes a chemically driven, adsorption-based electrochemical sensing strategy that exploits gold surface chemistry for label-free biomarker detection. We will share our first designs for in house screen printing strategies with potential for multi element analysis in the context of breast cancer screening.

OC 21 : REAL TIME DIAGNOSIS OF CARDIAC DISEASES WITH IMPLANTABLE SENSORS

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Abstract

Nanotechnology and nanomedicine is cutting-edge field that is growing, providing new solutions in different areas. These new technologies in medicine cover many possibilities for the study, treatment and diagnosis of different diseases in a more efficient and personalized way. Thanks to this technology, a key tool recently developed in biomedical engineering research, are implantable sensors.

The development of miniaturized implantable biosensors in the human body has revolutionized the field of medicine in terms of diagnosis, and monitoring of numerous conditions and diseases, such as cardiovascular disorders and metabolic biomarkers. One of the great advances that these sensors have introduced is their ability to monitor clinical analytes practically in real time, obtaining records of the body's biophysical and biochemical parameters in a continuous way and for extended periods. But despite the great progress made, implantable sensors still face many challenges before they can be implemented as a standard method.

This talk will present new technologies in implantable sensors for its implantation in blood vessels, which allow for continuous monitoring and early detection of cardiac diseases. We will show the developments achieved in this area by our research group for different applications validated in in vivo models, such as monitoring ischemia in fetuses for detection of fetal growth restriction and the detection of biomarkers of heart disease for early diagnosis. Future trends and the advantages and limitations of this technology will be discussed.

OC 22 : ULTRA-SENSITIVE SINGLE-MOLECULE FLUORESCENT QUANTIFICATION OF DNA EPIGENETIC MODIFICATIONS

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Abstract:

DNA epigenetic modifications contribute critical regulatory functions to the underlining genetic sequence. 5-Methylcytosine (5mC), the fifth base and 5-Hydroxymethylcytosine (5hmC), the sixth base are the two major DNA epigenetic modifications in mammalian genome and synergistically play crucial roles in development and pathogenesis. They display tissue and cell type specific distribution in our genomes and aberrant 5mC/5hmC changes are well-accepted hallmarks of many diseases including cancers. Detection and quantification of 5mC and 5hmC in clinical samples provide important information for diagnosis and prognosis purposes. The current methods to quantify DNA epigenetic modifications such as HPLC-MS, antibody dot blot/immunostaining and HPLC-MS either are semi-quantitative or require large amount of DNA, thus limiting their use in clinical setting. We have developed a series of new detection and quantification approaches for DNA epigenetic modifications at the single molecule level [1-4]. By using single-molecule fluorescent imaging, one can quantify one modification or two modifications simultaneously on various biological samples. We further quantified the ratio of 5hmC modified cell-free DNA (cfDNA) in the circulating blood of volunteers at different ages using our ultrasensitive single-molecule immunofluorescent imaging method.

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OC 23 : NANOBODY-BASED PHOTOELECTROCHEMICAL SENSING FOR POINT-OF-CARE DETECTION OF AFRICAN TRYPANOSOMIASIS

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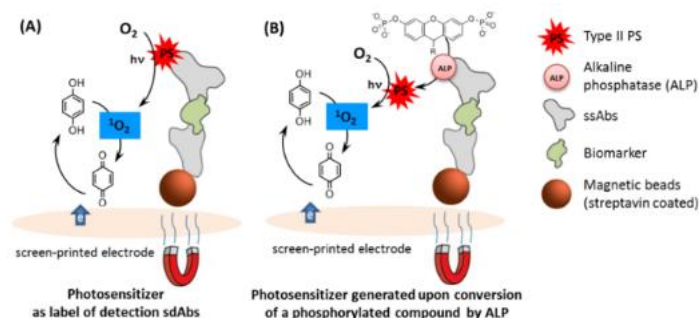
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Abstract:

African trypanosomiasis is a neglected tropical disease endemic to Africa, Asia, and South America



and frequently found in Europe. The World Health Organization (WHO) has prioritized the elimination of this disease by 2030, emphasizing the need for improved surveillance and diagnostic tools.¹ Current antibody-based methods cannot differentiate between active and past infections. Biomarkers like *T. congolense* pyruvate kinase (TcoPYK), which indicate active infection, offer a more targeted diagnostic approach. This work explores the use of camelid single-domain antibodies in a photoelectrochemical (PEC) platform for TcoPYK detection. While these nanobodies have shown promise in ELISA assays, these are time-consuming and require specialized personnel. Here, we propose a sensitive, user-friendly, and point-of-care-compatible PEC device integrating nanobodies through two distinct detection strategies. Both approaches rely on photosensitizer-mediated singlet oxygen (1O_2) generation for target quantification.² In the indirect method, nanobodies are fused with alkaline phosphatase, which enzymatically converts fluorescein diphosphate into active fluorescein a type II photosensitizer upon target recognition. In the direct method, fluorescein is site-specifically conjugated to the detection nanobody via sortase labeling in a sandwich-type assay format. Under light illumination (470nm 45mW), fluorescein generates 1O_2 , which reacts with the redox mediator to produce an amplified photocurrent, enabling rapid and sensitive PEC-based biomarker detection.² To enable selective target capture and improve performance in complex matrices, the capture nanobody is functionalized with biotin and immobilized on streptavidin-coated magnetic beads, facilitating easy sample handling and reducing matrix effects. This dual-strategy PEC platform underscores the versatility of nanobodies in biosensing and advances the development of rapid, robust, and deployable diagnostics for African trypanosomiasis. The assay enables clear signal discrimination and maintains target recognition and enzymatic activity, supporting its further development as a portable, sensitive diagnostic tool for detecting active African animal trypanosomiasis infections.

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OC 24 : DUAL-FUNCTIONALISED AMPLICONS ENABLE EXTRACTION-FREE PHOTOELECTROCHEMICAL DIAGNOSIS OF CHIKUNGUNYA VIRUS

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Abstract:

RNA extraction remains a major bottleneck preventing molecular diagnostics from reaching decentralised and resource-limited settings.¹ This challenge is particularly acute for emerging arboviral diseases such as chikungunya virus (CHIKV), which require rapid diagnosis for outbreak surveillance and patient management. Reverse transcription-polymerase chain reaction (RT-PCR) provides excellent analytical performance, but its reliance on laboratory infrastructure and extensive sample processing limits decentralised use. Developing extraction-free molecular strategies that maintain high sensitivity and specificity in minimally processed clinical samples therefore remains an unmet need. Here, we introduce a molecular encoding strategy for extraction-free CHIKV diagnosis that integrates mild detergent-mediated lysis, one-pot reverse transcription-recombinase polymerase amplification (RT-RPA) and singlet oxygen-mediated photoelectrochemical (PEC) detection. In this platform, RT-RPA is performed using a biotinylated primer and a chlorine 6-functionalised primer, generating amplicons that are simultaneously encoded with capture capability and signal-generating functionality. Following amplification, these photoactive amplicons are captured onto streptavidin-coated magnetic beads, enabling purification, concentration and direct PEC readout on disposable screen-printed electrodes. Upon visible-light illumination, the incorporated photosensitiser generates singlet oxygen, producing a photocurrent proportional to the target concentration.^{2,3} The platform enables direct detection of CHIKV RNA from crude serum lysates without RNA extraction, eliminating one of the major bottlenecks in decentralised nucleic acid testing. The assay achieves attomolar sensitivity within one hour and demonstrates excellent specificity against seven clinically relevant co-circulating alphaviruses. Testing on clinical patient samples shows agreement with RT-PCR, despite eliminating the extraction step entirely. Overall, this work establishes molecular encoding of amplification products as a general strategy for extraction-free diagnostics, directly coupling amplification and signal generation for sensitive PEC detection in complex biological matrices.

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2. Trashin, S. et al. Singlet oxygen-based electrosensing by molecular photosensitizers. *Nat. Commun.* 8, 16108 (2017).
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OC 25 : EMERGING STRATEGIES TO PRODUCE PAPER-BASED AND 3D-PRINTED (BIO)SENSORS BASED ON LASER-NANOSTRUCTURED GRAPHENE

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Abstract:

Paper-based (PADs) and Additive Manufacturing 3D-printed devices (AMDs) are compelling advances in modern (bio)electroanalytical science, offering rapid prototyping and design freedom and providing a unique route to on-demand, low-cost (bio)analytical platforms; however, several challenges remain. In this context, integrating/printing Nanostructured Films into lab-made PADs and AMDs, despite strong promise, remains underexplored. Herein, the production of functional graphenic nanofilms via CO₂ laser plotter and their integration into PADs and AMDs will be presented, along with their use for sensing and enzymatic biosensing. Attention will be paid to both laser-assisted assembly of nanostructured surfaces and the fabrication of disposable devices using low-cost, sustainable substrates via affordable benchtop microfabrication technologies. In particular, (i) a paper-based graphene bioelectrode enabling third-generation fructose dehydrogenase sensing of inulin will be presented. Polyimide-derived Laser-Induced Graphene (LIG) was integrated into office, recycled, and industrial by-product-containing papers, and the bioelectrocatalytic properties were examined. The complete LIG biosensors assembled on bamboo 'tree-free' paper (FB) proved more effective and were used to enzymatically detect inulin in urine (LOD = 0.3 mg L⁻¹) and serum (LOD = 1.1 mg L⁻¹), at levels relevant to clinical glomerular filtration rate testing. FB-LIG biosensor-based inulin analysis requires 10 min and yields satisfactory recoveries (90–111%; RSD ≤ 7.9%, n = 3), establishing it as the first third-generation paper-based biosensor. (ii) Moreover, new strategies for integrating laser-produced graphenic nanofilms onto 3D-printed models will be presented. These approaches enable on-demand printing of graphenic sensing circuits onto various 3D substrates via Fused Filament Fabrication. Using this rapid and scalable approach, complete sensors and enzymatic biosensors were produced in series. Their morpho-chemical and electrical/electrochemical features were then thoroughly examined, and (bio)electroanalytical potential was demonstrated by detecting a range of analytes in diverse agrifood and biological samples. Reproducible data with nano- to micromolar detection limits and satisfactory recoveries were obtained across all applications; analytical data are not reported because the studies are still ongoing. This presentation demonstrates how CO₂-laser plotter technologies, combined with benchtop manufacturing approaches, enable the production of on-demand, sustainable (bio)analytical devices that address diverse analytical challenges.

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OC 26 : CARBONIC ANHYDRASE-BASED PAPER-BASED COLORIMETRIC DEVICE FOR ACETAZOLAMIDE DETERMINATION IN BIOLOGICAL SAMPLES

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Abstract:

Acetazolamide (ACZ) is a drug commonly used to treat edema from heart failure, epilepsy, and glaucoma; moreover, ACZ, due to its diuretic activity, is also employed as a masking agent for doping. Several methods for analyzing ACZ have been reported in the literature, but there remains a lack of smart analytical devices capable of evaluating ACZ in biological samples with intra-sulphonamide selectivity. In this presentation, a carbonic anhydrase-based, paper-based colorimetric device (cPAD) is presented for the direct determination of ACZ in urine and serum. The cPAD, which operates via ACZ-driven inhibition of carbonic anhydrase, was fully designed and fabricated in-house using benchtop instruments. The cPAD resulted in an origami setup composed of the ‘sampling zone’ devoted to sample pretreatment via paper filtration and the ‘colorimetric zone’ housing the enzyme and the corresponding colorimetric reaction. The colorimetric signal relies on the carbonic anhydrase-catalyzed conversion of 4-nitrophenyl acetate (4-NA) to p-nitrophenol, resulting in a yellow color on paper. The sensing principle is based on ACZ inhibition of enzyme activity in a concentration-dependent manner, resulting in a proportional color ‘switch-off’. The paper-device operating strategy consists of: (i) sample loading onto the dedicated zone, (ii) overlaying with the ‘colorimetric zone’, and (iii) the 4-NA loading followed by a 10-min incubation. Eventually, the colorimetric signal was collected using a smartphone. The cPAD paper components for sample collection and enzymatic-assay hosting were carefully optimized with respect to paper type and operating geometries. The enzymatic reaction was carefully optimized to maximize assay sensitivity and the selectivity toward other sulphonamides. The proposed cPAD allowed the determination of ACZ in both urine and serum using matrix-matched calibration in the range of 0.05-0.5 ppm for urine ($R^2=0.994$; LOD = 0.011 ppm) and 0.1-0.8 ppm for serum ($R^2=0.993$; LOD = 0.024 ppm), proving for both matrices good reproducibility ($RSD \leq 13\%$, $n=3$). Eventually, the cPAD exploitability was successfully demonstrated for ACZ determination in urine and serum samples, yielding satisfactory recoveries (86-121%; $RSD \leq 14\%$, $n=3$). Summing up, the proposed cPAD enables smartphone-based ACZ determination at clinically relevant concentrations within 20 minutes, requiring only 10 μ L of sample, representing a step toward the sustainable expansion of point-of-care devices.

Acknowledgements:

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OC 27 : PAPER-BASED LASER-INDUCED GRAPHENE OBTAINED VIA UV LASER FOR APTAMER-BASED INSULIN DETECTION

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Abstract:

Laser graphitization of cellulosic substrates offers a sustainable method to directly produce functional carbon nanostructured films for electrochemical devices. Given the paper's nature, obtaining conductive tracks suitable for sensing requires targeted optimization, which is often difficult to achieve. In this context, despite their advantages, such as higher resolution and power control, UV lasers remain underexplored compared to CO₂ lasers, and paper-based graphitized sensors haven't been used for aptasensor development despite their potential.

Herein, integrated self-supporting sensors were produced in series via UV laser processing starting from common-use paper, and their applicability was extended to the fabrication of paper-based aptasensors designed for insulin detection. Since UV-laser conversion of paper into laser-induced graphene (LIG) surfaces had not been explored, it was initially investigated carefully. Laser parameters were comprehensively studied, and the resulting conductive tracks were characterized electrically and electrochemically, revealing morphochemical features. In brief, chromatographic paper was pretreated with sodium tetraborate and yellow wax, and then converted to porous paper-derived LIG, demonstrating how laser parameters affect the final properties. Using optimal UV-laser settings, paper-LIG-based three-electrode systems (working, counter, reference) were implemented, modified with AuNPs to serve as bioelement anchoring points, and used to build aptamer-based insulin biosensors. Aptamers, as alternatives to antibodies, were immobilized on the gold nanoparticle-modified LIG surface, demonstrating their ability to detect insulin in model and biological solutions via a label-free approach. Numerical data aren't reported in this abstract while studies are ongoing, but preliminary results indicate that sensitivity and reproducibility are sufficient to detect this biomarker in clinical settings. In summary, the first paper-based UV-laser-produced aptasensor is introduced, unveiling this fabrication method for sustainable affinity biosensing.

Acknowledgements:

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OC 28 : LUCIFERASE–AuNP BIOCONJUGATES FOR PLASMON-ENHANCED BIOLUMINESCENCE: TOWARD MINIMALLY INVASIVE ATP SENSING

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Bioluminescence (BL), the emission of light in organisms such as bacteria, fungi, insects and marine species, arises from an enzyme-catalyzed reaction in which luciferase oxidizes luciferin in the presence of molecular oxygen and, for firefly luciferases, adenosine 5'-triphosphate (ATP). BL biosensing offers major analytical advantages, including low optical background, high signal-to-noise ratio and the absence of external excitation, making luciferases highly attractive reporters for bioanalytical assays and imaging both in vitro and in vivo. Engineered luciferase mutants with shifted emission maxima enable tunable sensing platforms (1), which can be further enhanced by coupling to plasmonic nanostructures.

In this work, citrate-stabilized gold nanoparticles (AuNPs) synthesized via the Turkevich method are conjugated with green-, orange- and red-emitting luciferases (3) (BgLuc, BoLuc, BrLuc) to obtain bioluminescent nanotags. The localized surface plasmon resonance (LSPR) of AuNPs amplifies the local electromagnetic field and increases the apparent radiative efficiency (2,4) of the luciferase emitter, resulting in a bioluminescence enhancement of about 140–155% for all three mutants under optimized conjugation conditions. A preliminary ATP dose–response curve with BgLuc–AuNP nanotags yields a limit of detection of 10 picomoles, highlighting the potential of plasmon-enhanced BL nanotags for ultrasensitive ATP quantification.

The implementation of the obtained plasmon-enhanced BL nanotags into hydrogel-based microneedle (MN) patches could provide minimally invasive ATP sensors. Swellable, crosslinked hydrogel MNs can pierce superficial skin layers and absorb dermal interstitial fluid (5), delivering ATP extracted from interstitial fluid to luciferase–AuNP conjugates. Preliminary studies are also reported to optimize fluid uptake, mechanical robustness and analyte diffusion, enabling compatibility with compact optical readout modules for real-time, on-patch monitoring of tissue ATP levels.

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OC 29 : BOOSTING BIOSENSING WITH NOVEL BIOLUMINESCENCE CELL-BASED PLATFORMS

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Microfluidic-based devices, including paper-based, 3D-printed and thread-based analytical devices, are receiving significant attention as platforms for point-of-care (POC) diagnostics, environmental monitoring, and food safety control thanks to their advantageous properties in terms of portability, low-cost, and sustainability. Bioluminescence (BL) provided a formidable manifold system relying on different luciferases and luciferin analogues with a wide range of applications spanning from molecular imaging to biosensing. The availability of new BL proteins with tuned properties, both in terms of emission wavelength, kinetics and protein stability, is highly valuable in the bioanalytical field, with the potential to improve the sensitivity and analytical performance of the currently used methods for ATP detection, whole-cell biosensors, and viability assays among others. Thanks to reporter gene technology a BL reporter protein can be expressed under the regulation of a target promoter sequence or enhancer elements, thus enabling correlation of reporter protein expression, measured as light signal, and transcriptional regulation. The unprecedented technological evolution of portable light detectors opened new possibilities to implement bioluminescence detection into miniaturized devices. A portfolio of cell-based BL biosensors implementing novel luciferases and novel supports, such as threads and nanostructured polymers, will be presented with a look at the current challenges and the different strategies used to convert current laboratory methods into portable cell-based platform for biosensing.

OC 30 : MICRONEEDLE ELECTROCHEMICAL PLATFORMS FOR DETECTING BIOMARKERS ASSOCIATED WITH NEURODEGENERATIVE DISEASES

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Abstract:

Minimally invasive technologies for monitoring biomarkers in skin interstitial fluid (ISF) have attracted growing attention in recent years, both in academic research and in the commercial sector. They are performed by microneedle-based electrochemical biosensors (MNs) either directly involving the target analyte (e.g. for the detection of dopamine or further catecholamine derivatives) or through an enzymatic reaction (e.g. for glucose and lactic acid) [1,2]. Beside these classes of biomarkers, proteins are also particularly relevant for the diagnosis of many diseases, such as neurodegenerative disorders. Amyotrophic Lateral Sclerosis (ALS) is associated with clinical manifestations such as muscle atrophy, dysarthria, dysphagia, and respiratory impairment. Despite the lack of established biomarkers, recent studies have shown that ALS progression is linked to increasing levels of specific cytokines, including Interleukin-18 (IL-18) and Interleukin-6 (IL-6), which can also be measured in ISF [3]. Thanks to our recent experience in observing the advantages in the application of an electrochemical immunoassay for the identification of ALS progression by IL-18 quantification [4], we present here the development of a microneedle-based electrochemical biosensor for the detection of this biomarker in ISF. The device consists of three rows of 3D-printed MNS, coated with gold by sputtering to provide electrical conductivity and functionalized with primary antibodies to enable selective recognition of IL-18. Electrochemical detection is carried out using an impedimetric approach, which was evaluated in matrices of increasing complexity. In artificial ISF, the sensor achieved a detection range of 31.2–250 pg mL⁻¹, covering the clinically relevant concentrations needed to differentiate healthy subjects from patients at both early and advanced stages of the disease [3].

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OC 31 : DEVELOPMENT OF AN ELECTROCHEMICAL BIOSENSOR FOR PROTEASE DETECTION

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Proteases are enzymes that degrade proteins and peptides into smaller fragments or amino acids by hydrolysis of peptide bonds. Proteases participate in numerous biological processes and due to their great diversity differences in catalytic activity and specificity, these enzymes are strictly regulated - even minor alterations in their regulation can lead to development of various diseases (i.e. cancers) [1], [2]

Here, we developed an electrochemical biosensor on co-immobilized L-amino acid oxidase from *Crotalus atrox* and horseradish peroxidase by glutaraldehyde cross-linking, enabling real-time measurement of protease activity through the detection of amino acids released from dipeptide probes. The biosensor was characterized by using L-leucine as a model analyte and showed a sensitivity of 1033.49 ± 66.29 nA/mM, an LOD of 0.029 ± 0.002 mM, an LOQ of 0.089 ± 0.006 mM, and a CV of 6.41%. Protease activity was evaluated using different concentrations of Pronase E from *Streptomyces griseus* at a fixed concentration of 1 mM of the L-Leu-Leu dipeptide. Activity was calculated from the initial reaction rates obtained by chronoamperometry, yielding an LOD of 0.74 ± 0.17 µg/ml, an LOQ of 2.244 ± 0.499 µg/ml, and a CV of 10.89%. The biosensor also successfully monitored endogenous serum proteases and exogenous Pronase E activity in diluted human serum. The results demonstrate feasibility of the platform for sensitive, real-time protease activity monitoring and a potential clinical application.

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OC 32 : MAGNETIC CARBON BLACK MODIFIED WITH A MOLECULARLY IMPRINTED POLYMER FOR THE SELECTIVE EXTRACTION AND DETERMINATION OF CARVACROL

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Abstract:

Essential oils are complex mixtures of bioactive volatile compounds widely used in pharmaceutical, food, cosmetic, and agricultural industries. Among their constituents, thymol, a monoterpene phenol commonly found in carvacrol and oregano, has attracted significant attention due to its strong antimicrobial and antioxidant properties. However, the selective and sensitive determination of carvacrol in complex essential oil matrices remains challenging. In this work, a multifunctional magnetic carbon black-based molecularly imprinted polymer (MCB-MIP) was developed as a selective platform for carvacrol extraction and detection. The hybrid material integrates the high surface area, excellent adsorption capacity, and electrical conductivity of carbon black with the magnetic property of iron oxide nanoparticles, enabling rapid magnetic separation from complex samples. The incorporation of molecular imprinting technology provides specific recognition sites complementary to carvacrol, enhancing selectivity and binding affinity. The synergistic combination of magnetic separation and selective molecular recognition results in enhanced sensitivity, rapid extraction, and efficient quantification of carvacrol. The proposed MCB-MIP system demonstrated strong potential as a robust and reliable analytical platform for the determination of carvacrol in essential oils and other complex matrices.

This work is part of a project that has received funding from the European Union's Horizon Europe research and innovation program under the grant agreement No. 101058570 (RELIANCE).

OC 33 : DUAL-SELECTIVE WEARABLE PLATFORM FOR SELF-REFERENCED POTENTIOMETRIC MONITORING OF CORTISOL AND POTASSIUM IN SWEAT

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Abstract:

Low-concentration neutral biomarkers remain a major challenge for wearable technologies, as generally relying on unscalable recognition frameworks along with energy-demanding detection principles, and real-time correction for dynamic biofluid variability [1-3]. The combination of molecular-imprinting technology with potentiometric transduction represents an effective strategy to enable cost-efficient, reagent-free, and robust operation [4], while minimizing the power consumption within continuous on-body applications. Considering this context, we developed a dual-readout potentiometric platform for wearable tracking of sweat cortisol and potassium ions under physical exercise. The (bio)sensing module includes a cortisol-selective electrode based on a cortisol-imprinted polymer integrated with a potassium-selective membrane, where cortisol binding changes the permeability of endogenous sweat potassium ions, producing a measurable potentiometric signal. A potassium-selective electrode was further incorporated to provide real-time background correction and account for inter- and intra-subject sweat variability, while also allowing the monitoring of electrolyte balance during physical activity. Cortisol binding (4 min) and continuous sweat renewal were achieved using a custom-designed paper-based microfluidics. Potassium and cortisol levels were monitored through both periodic and continuous on-body measurements during treadmill and stationary bike exercise in seven volunteers, and the results showed remarkable agreement with corresponding reference methods ($r = 0.999$ and $r = 0.993$, respectively).

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OC 34 : DIGITAL PHOTOCORROSION QUANTUM SEMICONDUCTOR BIOSENSOR FOR BACTERIA AND BACTERIAL SPORES

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Abstract:

Monitoring pathogenic bacteria in anthropogenic and natural water reservoirs remains a significant challenge for health authorities, as it requires preferably (semi)autonomous biosensors located remotely and providing sensitive data at attractive cost. Our research focused on digital photocorrosion (DIP) of GaAs/AlGaAs quantum semiconductors, with photoluminescence employed for monitoring the flow of electric charge from/to bacteria demonstrated attractive detection sensitivities for *E. coli* [1], *L. pneumophila* [2], and *Bacillus cereus* group spores [3]. Meanwhile, the application of samples with stacks of numerous ~ 20 nm thick GaAs/AlGaAs nanolayers has allowed us to introduce the innovative concept of a regenerable photonic aptasensor [4]. The results suggest the potential of sustainable detection with over 30 pairs of GaAs/AlGaAs nanolayers, each designed for completing an individual biosensing run.

I will discuss the challenges and perspectives of commercialization of DIP biosensors for quasi-continuous monitoring of selected water reservoirs with a workstation comprising water sampling module for automated collecting of samples and pre-concentrating bacteria [5].

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OC 35 : A novel Sonogel-Carbon based amperometric biosensor integrated into a continuous-flow system for glucose determination in untreated blood serum and plasma

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Abstract:

The integration of alternative sinusoidal electrochemical growth methods with advanced polymer-based transducers and microfluidic architectures represents a significant trend in improving clinical biomonitoring systems. A new amperometric biosensor integrated into a 3D-printed microfluidic platform has been developed and evaluated for the determination of glucose in untreated human serum and plasma. The sensing platform employs a Sonogel-Carbon as the electrochemical transducer substrate which was sequentially modified through a layer-by-layer assembly. Initially, a film of Prussian Blue (PB) was deposited onto the surface to act as an efficient redox mediator. Subsequently, glucose oxidase (GOx) was immobilized by entrapment during the electrochemical growth of a conductive polymer network (PEDOT) by using sinusoidal voltages. Finally, it incorporates a multipolymeric diffusion membrane containing polyurethane and Nafion that limits the access of glucose to the enzyme. Under continuous flow conditions, a high sensitivity of $0.39 \mu\text{A mM}^{-1} \text{cm}^{-2}$ was obtained for this electrode in a linear range of 1.5 – 45 mM ($R^2 = 0.9951$), with a low detection limit (LOD) of 0.36 mM. This biosensor also showed good repeatability (RSD = 2.74%, $n = 4$) and reproducibility (RSD = 4.52%, $n = 5$). Additionally, it exhibited high selectivity (no observation of significant interferences by interfering species such as ascorbic acid, uric acid, urea, dopamine, BSA, lactate, NaCl, CaCl₂ and KCl). Finally, the platform was successfully validated through the real-time monitoring of glucose in both synthetic and untreated human clinical samples using the additively manufactured polyethylene terephthalate glycol (PETG) microfluidic platform, featuring an optimized microfluidic volume about 280 μL . The analytical results demonstrated an excellent correlation with standard hospital reference methodologies, confirming the system reliability. These findings underscore the potential of this microfluidic electrochemical tool as a robust, precise, and cost-effective solution for near-patient biomedical monitoring.

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OC 36 : ADVANTAGES OF THE EMPLOYMENT OF 3-OCTYLTHIOPHENE OVER SONOGEL-CARBON NANOTUBES FOR ION-ELECTRON-TRANSDUCER IN MONITORING OF Na⁺ AND K⁺ FOR PHYSIOLOGICAL SAMPLES

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Clinical analyses provide indispensable information on a patient's condition, but traditional centralized testing often delays critical decision-making. Enabling rapid, efficient analysis at primary care centers or the patient's home is vital for the decentralization of digital healthcare (e-Health). This shift requires the miniaturization of analytical systems, making portable, selective, and low-cost devices highly sought after. Among these, potentiometric solid-contact ion-selective electrodes (SC-ISEs) stand out for their sensitivity and selectivity in monitoring critical parameters like Na⁺ and K⁺ in intensive care units. Previously, our group developed potentiometric sensors using a Sonogel-Carbon Nanotube composite as an ion-electron transducer. While these devices showed excellent sensitivity and lifetime, their calibration-free potential was limited, requiring electrochemical re-treatment after sequential measurements. To address this, conducting polymers like poly(3-octylthiophene) (POT) have been used as promising transducers due to their hydrophobicity, which hinders detrimental water layer formation, and low electroactivity, which minimizes side reactions. However, POT reproducibility remains a challenge in literature. In this work, Sonogel-Carbon-based potentiometric sensors for Na⁺ and K⁺ determination were fabricated by electrodepositing a POT layer as the ion-electron transducer. The resulting devices exhibited excellent figures of merit, particularly outstanding repeatability and reproducibility, successfully achieving calibration-free operation. In addition, an electrochemical process reduced conditioning time of the resulting sensor. The sensors also displayed high selectivity and were successfully validated in real serum and plasma samples under continuous flow using a 3D-printed microfluidic system.

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OC 37 : A Novel Immunosensing Platform for NT-proBNP Quantification in Complex Matrices toward Early Heart Failure Diagnosis

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Efficient functionalization of gold surfaces remains a key challenge in biosensor development, as conventional immobilization strategies often lead to low coupling efficiency, loss of activity, and random antibody orientation. Here, we report a novel strategy for antibody immobilization on gold electrodes for the detection of N-terminal pro-brain natriuretic peptide (NT-proBNP), a key biomarker for early heart failure diagnosis. The approach combines Light-Activated Site-Specific Conjugation (LASIC) for pre-functionalization of antibodies with copper-free strain-promoted azide-alkyne cycloaddition (SPAAC) for oriented surface attachment with the thiol-azide present on the electrode, providing a simplified and green, two-step functionalization procedure that reduces the number of steps compared to conventional methods. Successful formation of the self-assembled monolayer and antibody immobilization were confirmed by infrared spectroscopy, Sodium dodecyl sulfate–polyacrylamide gel electrophoresis SDS-PAGE, and contact angle measurements. The immunosensor was implemented on a four-gold electrodes biosensor and evaluated in complex matrices including artificial plasma. Electrochemical impedance spectroscopy (EIS) and cyclic voltammetry (CV), coupled with the standard addition method, enabled monitoring of antigen recognition and quantification of NT-proBNP. The sensor exhibited high sensitivity with a detection limit of 0.1 pg/mL, a linear range of 0.1–1.2

pg/mL, and measurable concentrations up to 5 pg/mL in PBS, while maintaining selectivity against other interfering HF biomarkers Interleukin 10 (IL-10) and Tumor Necrosis Factor-alpha (TNF- α). Analysis of an unknown 600 pg/mL NT-proBNP sample diluted 1000 fold in artificial plasma and detection in a saliva sample demonstrated the method's potential for rapid, accurate, and green biosensing applications.

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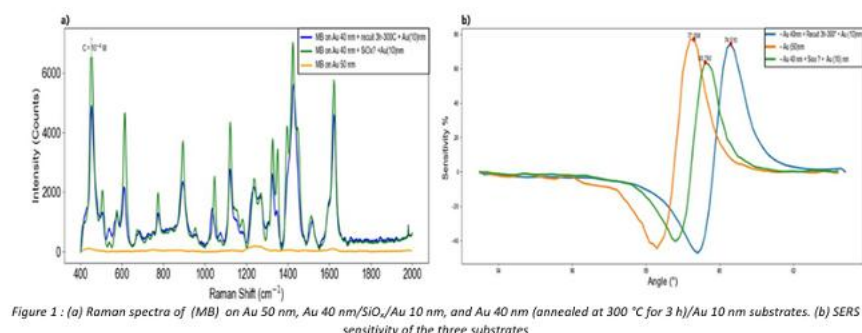
OC 38 :DEVELOPMENT OF A BIOCHIP ENABLING SURFACE-ENHANCED RAMAN SPECTROSCOPY (SERS) AND SURFACE PLASMON RESONANCE (SPR) FOR THE CHARACTERIZATION OF EXTRACELLULAR VESICLES.

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ABSTRACT:

Extracellular vesicles (EVs) represent highly promising biomarkers whose comprehensive characterization necessitates the use of complementary analytical techniques such as surface plasmon resonance imaging (SPRi), surface-enhanced Raman scattering (SERS), and atomic force microscopy (AFM). However, the design of a single substrate that is simultaneously compatible with all three methods remains challenging. SPRi requires a continuous gold film of optimal thickness (50 ± 5 nm), whereas SERS relies on metallic nanoparticles whose size, shape, and surface density must be precisely controlled. The objective of the present work is to develop a multifunctional plasmonic substrate that provides an effective compromise among these different constraints, thereby enabling multimodal characterization of extracellular vesicles. Starting from a 40 nm evaporated gold film on the glass, we investigated two strategies to achieve a smoother surface with reduced roughness. Specifically, we systematically examined (i) thermal annealing at 300 °C for 3 hours to form terrace-like gold nanostructures [1], and (ii) deposition of a thin SiO_x overlayer, which has previously been shown to enhance SPRi sensitivity [2]. Both treatments produced a modified, flatter substrate. For SERS characterisation, a 10 nm nanostructured gold overlayer was sputter-deposited (0.15 Å, 32 mTorr) onto either a 40 nm thermally evaporated gold film or a 40 nm gold film additionally coated with a SiO_x layer, using deposition parameters optimised for SERS performance. This multilayer architecture preserved the required plasmonic resonance while generating a high density of electromagnetic “hot spots”, thereby enabling efficient Raman signal enhancement. Combining the 40 nm gold underlayer with the 10 nm nanostructured top layer resulted in an effective gold thickness of 50 ± 5 nm, which is well suited for SPRi operation. Under these optimised conditions, the plasmonic chip produced strong SERS signals while retaining approximately 75% of the SPRi sensitivity, closely comparable to the 77% sensitivity measured for a conventional SPRi reference chip. These results demonstrate that precise control over both the gold film thickness and its nanoscale morphology enables the fabrication of a single, multifunctional platform that supports SERS, SPRi, and AFM, with only a modest compromise in optical performance. Perspectives: To evaluate this substrate across a range of biological samples and to investigate the effectiveness of surface functionalization by antibodies.

Mots-Clés: Spectroscopie Raman, Plasmonique, Résonance plasmonique, Vésicules extracellulaires.



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OC 39 : FROM CLINICAL DIAGNOSTICS TO ENVIRONMENTAL MONITORING: 3D-PRINTED MICROFLUIDIC CELLS FOR SIMULTANEOUS MULTI-ANALYTE DETECTION

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Abstract:

The growing demand for rapid, sensitive, and reliable analytical tools has promoted the development of new technologies able to simultaneously quantify multiple analytes in complex matrices. In the clinical field, the COVID-19 pandemic highlighted the importance of robust diagnostic strategies based on the detection of several biomarkers in order to improve diagnostic accuracy and cut down the risk of false-positive and false-negative results. Similarly, in the environmental field, contamination arising from industrial activities, particularly heavy metal pollution, remains a major concern due to its adverse effects on ecosystems and human health.

In this context, multiplexed analytical devices capable of simultaneously determining several analytes within a single sample has emerged as a highly attractive approach. Such platforms provide more comprehensive information about the sample, reduce analysis time and costs, minimize sample consumption, and improve the reliability of analytical results. Moreover, the simultaneous determination of multiple biomarkers or contaminants enables cross-validation of results, enhancing diagnostic accuracy and reducing the likelihood of erroneous interpretations.

Therefore, the present work focuses on the development of 3D printed microfluidic platform for the simultaneous determination of sodium, potassium, glucose and chloride anions and nitrite in serum/plasma and water samples, respectively. The proposed microfluidic systems showed good analytical performance and was successfully applied to real samples, highlighting its potential as a versatile and cost-effective tool for multi-analyte determination in clinical and environmental applications.

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OC 40 : CHARACTERIZATION OF MESENCHYMAL STEM CELL-DERIVED EXTRACELLULAR VESICLES IN THE CONTEXT OF CELL-FREE BIOTHERAPY.

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Abstract:

Once viewed as cellular waste, extracellular vesicles (EVs) have emerged as a revolutionary "cell-free" solution in regenerative medicine. Derived from mesenchymal stem cells (MSCs), these vesicles encapsulate the therapeutic paracrine activity of their parent cells without the safety risks associated with whole-cell transplantation [1]. However, their clinical translation remains a significant challenge due to their inherent heterogeneity and the complexities involved in their isolation and characterization, which vary depending on culture conditions and biogenesis pathways [2-3]. Each characterization technique has its own strengths and limitations. Therefore, integrating complementary analytical approaches is essential to obtain a more comprehensive and reliable characterization. In this work, we implemented a multimodal characterization strategy for secretome derived from human adipose tissue MSCs (AD-MSCs) maintained in a 3D culture system in order to qualify vesicular populations in both crude and size-exclusion chromatography (SEC) processed secretome. In-solution analysis, utilizing Interferometry Light Microscopy (ILM/VideoDrop) and Nanoparticle Tracking Analysis (NTA) to measure extracellular vesicles concentration and size distribution, complemented by protein assays and SDS-PAGE, were first realized. Then an on-chip analysis was engaged, employing Surface Plasmon Resonance imaging (SPRi) in a multiplexed format, to capture EVs based on membrane proteins, followed by Atomic Force Microscopy (AFM) inside the spots, to determine topography and enable fine discrimination of subpopulations. We demonstrate that while AD-MSC bioproduction in 3D systems produced increasing quantity of particles over time, the secretome undergoes a critical compositional shift from a highly homogeneous population of small extracellular vesicles (sEVs) to a more heterogeneous mixture. By implementing this multimodal approach, we established a definitive hierarchical phenotypic signature (CD44 > CD81 > CD9 > CD63) that acts as a quality fingerprint, coherent with the MSC origin and the vesicular nature of the samples. Furthermore, the high-resolution coupling of SPRi and AFM successfully identified intact sEVs even when they co-eluted with proteins during chromatography, providing a level of proof that single-technique analyses miss. In conclusion, this robust framework provides the "missing link" to transform unpredictable biological "cocktails" into standardized, reproducible pharmaceutical bioproducts ready for the next phase of developing cell-free regenerative medicine.

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PO 1: Transition Metal Dichalcogenide based Photodetectors and Nano-heterostructures for Catalytic Applications

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Abstract:

Transition metal dichalcogenides (TMD) have emerged as an important class of materials for several applications. Their two-dimensional nature renders them useful in many applications ranging from optoelectronics to batteries, spintronics and catalysis.

In this talk, I will discuss the possibilities of formation of nano-heterostructures based on MoSe₂ – a 2D TMD and their applications for optoelectronics and catalysis, in particular water splitting. Interestingly, the TMDs can also be used as a Selenium source to passivate lead halide perovskites rendering them highly stable.

Further, the TMDs have been shown to exhibit charge separation and therefore are useful for applications in photodetectors and catalysis, in particular electrocatalysis for reactions such as the hydrogen evolution reaction or the oxygen evolution reaction. Due to the layered nature of the TMDs these can accommodate ions between the layers and therefore are very advantageous for use as capacitor or battery applications.

I will discuss a few of these possibilities.

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PO 2 : INTEGRATION OF PAPER-BASED ELECTROCHEMICAL (BIO)SENSORS FOR LACTATE DEHYDROGENASE AND NITRATE INTO ORGAN-ON-CHIP DEVICES

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Abstract:

Organ-on-Chip (OoC) technologies are emerging as physiologically relevant in vitro models capable of reproducing key features of human tissues. Their full potential, however, relies on the integration of analytical tools able to continuously monitor biomarkers associated with cellular metabolism, inflammation, and tissue damage. In this context, paper-based electrochemical sensors represent a promising strategy for developing low-cost and sustainable OoC platforms with real-time monitoring capabilities. In this work, two paper-based electrochemical sensors are developed for the detection of nitrate (NO_3^-) and lactate dehydrogenase (LDH). These (bio)sensors are part of an integrated OoC platform for osteoarthritis simulation, designed to integrate 7 electrodes for simultaneous monitoring of 7 targets. Osteoarthritis is a degenerative disease characterized by cartilage degradation, chronic inflammation, and oxidative stress. LDH serves as an indicator of cytotoxicity released by damaged chondrocytes, while nitrates, stable catabolites of nitric oxide (NO), allow assessment of inflammatory status and oxidative stress. The sensors were fabricated via screen-printing technology on a wax-patterned paper substrate. The architecture employs Ag/AgCl as the reference electrode, alongside graphite for working and counter electrodes. The working electrodes were modified with a carbon black (CB) dispersion [1]. Nitrate determination was performed by casting an ion-selective membrane onto the working electrode, whereas LDH detection was performed via an amperometric technique with a response time of 60 s, based on electrochemical oxidation of NADH to NAD^+ at the CB-modified electrode surface [2]. The nitrate sensor demonstrated a Nernstian response with a sensitivity of $-0.052 \pm 0.001 \frac{\text{V}}{\log(a)_{\text{NO}_3^-}}$ ($y = (-0.052 \pm 0.001)x + (0.0463 \pm 0.001)$, $R^2 = 0.991$) in distilled water and $-0.059 \pm 0.004 \frac{\text{V}}{\log(a)_{\text{NO}_3^-}}$ ($y = (-0.059 \pm 0.004)x + (0.14 \pm 0.001)$, $R^2 = 0.959$) in α -MEM cell culture medium. In both matrices, a linear dynamic range from 10^{-4} to 1M was achieved, with limits of detection (LOD) of $6 \times 10^{-5} \text{ M}$ and limits of quantification (LOQ) (LOQ) of $2 \times 10^{-4} \text{ M}$ in standard solution while, in α -MEM a LOD of $7 \times 10^{-5} \text{ M}$ and a LOQ of $2 \times 10^{-4} \text{ M}$, along with high precision in the biological matrix ($\text{RSD}\% = 3\%$). The LDH biosensor exhibited a linear response in the range of 0.1 - 0.35 U mL^{-1} in both phosphate buffer and α -MEM with sensitivities of -8.8 ± 0.03 ($y = (-8.8 \pm 0.03)x + (3.85 \pm 0.007)$, $R^2 = 0.970$) in standard and $-0.66 \pm 0.07 \mu\text{A (U mL}^{-1})^{-1}$ ($y = (-0.66 \pm 0.07)x + (0.56 \pm 0.02)$, $R^2 = 0.905$) in -MEM. In the standard solution, a LOD of 0.04 U mL^{-1} a LOQ of 0.1 U mL^{-1} , and good repeatability ($\text{RSD} = 7\%$) were obtained. Combining potentiometric and amperometric approaches demonstrates versatile, portable, cost-effective, and eco-friendly paper-based sensors for OoC integration.

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Acknowledgment: Phoenix-OoC project, funded under the European Union's Horizon Europe EIC 2023 Pathfinder Open program, grant agreement No 101130395.

PO 3 : A PAPER-BASED ELECTROCHEMICAL OXYGEN SENSOR FOR INTEGRATION INTO NEXT-GENERATION PAPER-BASED ORGAN-ON-CHIP PLATFORMS

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Abstract:

Organ-on-chip (OoC) technology is emerging as a powerful alternative to conventional in vitro models by recreating physiologically relevant organ microenvironments within microfluidic platforms.¹ Herein, we present a sustainable paper-based electrochemical sensor for dissolved oxygen monitoring, designed for integration into paper-based OoC devices. Continuous monitoring of dissolved oxygen is essential for maintaining physiologically relevant microenvironments in OoC systems, where oxygen levels critically influence cellular metabolism and function. The sensor was fabricated on a wax-patterned paper substrate by screen printing, and the working electrode was modified with 6 μL of carbon black dispersion² to enhance electrochemical performance and significantly improve signal stability, reducing the RSD from 81% to 9% in oxygen-saturated solutions. The device enables chronoamperometric detection of dissolved oxygen in both phosphate buffer and cell culture media. The sensor showed a linear response in phosphate buffer (0.05 M; pH 7.4) ($R^2 = 0.980$) and good agreement with a commercial oximeter (Pearson's $r = 0.943$), confirming the reliability of the proposed platform. The dynamic performance of the sensor was further investigated using controlled O_2/N_2 gas mixtures to evaluate hysteresis under physiologically relevant oxygen conditions. Comparable forward and backward calibration sensitivities (0.60 ± 0.06 and $0.59 \pm 0.05 \mu\text{A } \% \text{O}_2^{-1}$, respectively) together with linearity ($R^2 = 0.924$ and 0.941) demonstrated negligible hysteresis and signal reversibility. Preliminary experiments performed in cell culture medium (α -MEM) further confirmed the platform's applicability, yielding a linear response to oxygen modulation ($R^2 = 0.981$; sensitivity = $0.73 \pm 0.06 \mu\text{A } \% \text{O}_2^{-1}$). Overall, the proposed paper-based oxygen sensor represents a low-cost, sustainable platform for continuous dissolved oxygen monitoring, with strong potential for integration into next-generation paper-based Organ-on-Chip systems for real-time biological monitoring.

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Acknowledgement:

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PO 4: BIOINSPIRED MATERIALS FOR ADVANCED PORTABLE AND MINIATURIZED BIOSENSOR

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Abstract:

Bioinspired materials are increasingly employed in the development of advanced portable and miniaturized biosensors due to their ability to reproduce key biological functions while offering enhanced stability, reduced cost, and facile integration into analytical devices. Among them, Molecularly Imprinted Polymers (MIPs) are synthetic materials containing selective recognition sites generated by polymerization in the presence of a template molecule and subsequent template removal, enabling antibody-like molecular recognition [1]. At the same time, nanozymes are nanomaterials exhibiting enzyme-like catalytic activity that combine high catalytic efficiency with remarkable chemical stability and robustness compared with natural enzymes [2]. Owing to these complementary characteristics, both MIPs and nanozymes have emerged as promising bioinspired materials for the fabrication of sensitive, selective, and portable biosensing platforms. In this work, two detection strategies based on paper-based screen-printed electrochemical sensors are proposed. The first strategy involves the modification of electrodes with molecularly imprinted polymers (MIPs) for the selective quantification of clinically relevant biomarkers, such as lactate, which can be used as an indicator of infectious processes in the orthopedic field. The second strategy relies on the use of nanozymes, synthesized by coprecipitation and immobilized onto the electrode surface, for the optical-colorimetric detection of target compounds, with quantification achieved through analysis of the RGB color components of the acquired image.

Acknowledgements:

Financial support is recognized under the National Recovery and Resilience Plan (PNRR), Missione 6 – Salute, Componente 2 – Innovazione e ricerca sanitaria, Investimento 2.1 – Valorizzazione e potenziamento della ricerca biomedica del SSN, Avviso pubblico n. 377, finanziato dall'Unione Europea – NextGenerationEU – The diagnostic pillar for diagnosis of infections in the orthopedic field: Smart and sustainable paper-based electrochemical point-of-care device assisted by artificial intelligence– PNRR-POC-2023-12378291 CUP E83C23003450007–BANDO PNRR M6/C2 2023 26 marzo 2024.

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PO 5 : ELECTROCHEMICAL MIRNA-BASED SENSING: A NOVEL LABEL-FREE APPROACH FOR DIAGNOSING NEURODEVELOPMENTAL DISORDERS

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Abstract:

Attention-Deficit/Hyperactivity Disorder (ADHD) and Autism Spectrum Disorder (ASD) are neurodevelopment disorders that currently pose a major global public health concern, but lacking accurate and/or fast diagnostic tools since the conventional detection methods, like RT-qPCR are expensive and time-consuming, driving the demand for innovative, point-of-care screening alternatives. Nevertheless, recent evidence highlights miRNA-151a-3p as a promising and highly stable circulating biomarker, accessible through non-invasive biofluids such as saliva and plasma, that are directly linked to the development of these disorders. In this work, screen-printed gold electrodes (SPGEs) functionalized with a specific DNA-151a-3p capture probe were employed as a promising strategy for the direct detection of miR-151a-3p. Cyclic Voltammetry (CV) and Electrochemical Impedance Spectroscopy (EIS) were used to monitor the biosensor's assembly. To maximize the electrochemical optimal response, key experimental parameters were evaluated, including a comparison between different electrode composition, alongside the concentration and incubation periods of the different assembly steps and reagents. In the end, the optimal parameters were selected based on the Charge Transfer Resistance (R_{ct}) values, where the highest R_{ct} indicated a superior surface coverage and more efficient immobilization, ensuring the most accurate device performance. Preliminary results demonstrate that this label-free biosensor exhibits high selectivity and sensitivity, successfully distinguishing the target miR-151a-3p from the non-complementary sequences within biologically relevant concentration ranges. Future work will focus on constructing the calibration curves to determine the device's linear range and the selectivity and robustness will be evaluated in biological matrixes, thereby operating as a more cost-effective, fast, and portable diagnostic tool than traditional assays.

Acknowledgements:

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PO 6 : A RAPID, COST-EFFECTIVE SMARTPHONE-BASED FLUORESCENT BIOSENSOR FOR HISTAMINE DETECTION USING A MOLECULARLY IMPRINTED MEMBRANE

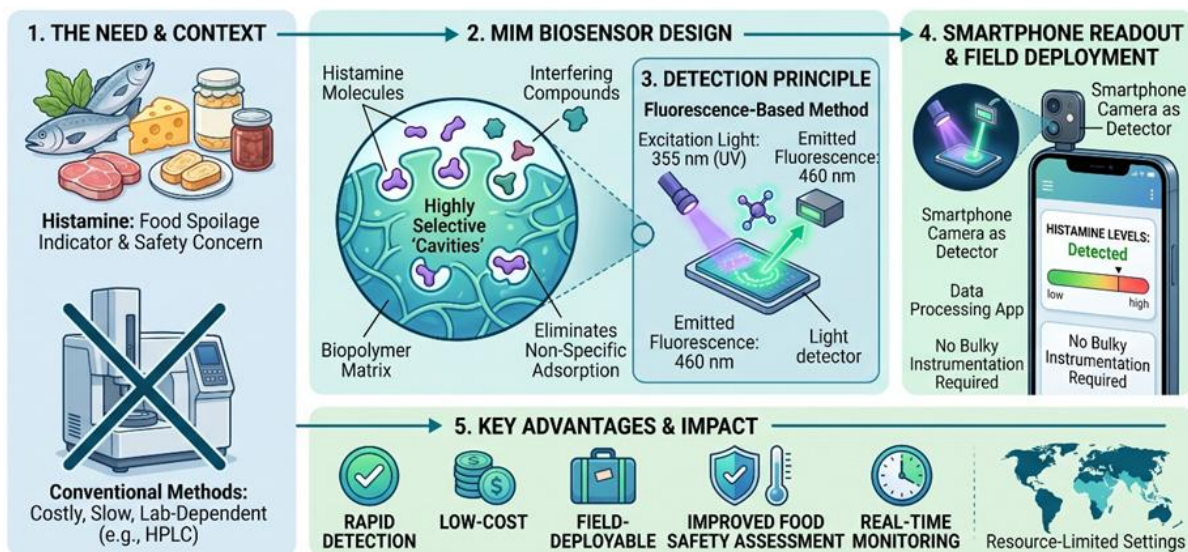
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Abstract:

Histamine is a critical indicator of food spoilage and a major safety concern in fish and fermented products, where elevated levels can trigger scombroid poisoning and other adverse health effects. Conventional quantification methods, such as HPLC, are highly accurate but costly, time-consuming, and reliant on specialized laboratory equipment, limiting their applicability for rapid, on-site screening. To address this gap, we developed a molecularly imprinted membrane (MIM) biosensor fabricated entirely from green materials. Utilizing a sustainable biopolymer matrix, this eco-friendly sensor enables the selective, fluorescence-based detection of histamine while effectively eliminating non-specific adsorption. Coupled with a smartphone-based fluorescence readout, the system removes the need for bulky instrumentation. Fluorescence intensity was recorded at an excitation wavelength of 355 nm and an emission wavelength of 460 nm. The calibration curve demonstrated excellent linearity in the micromolar range, achieving a limit of detection (LOD) of 5.1 μM and a limit of quantification (LOQ) of 15.5 μM . This approach offers a rapid, low-cost, environmentally sustainable, and field-deployable alternative to conventional techniques, demonstrating strong potential for real-time histamine monitoring and food safety assessment, particularly in resource-limited settings.



PO 7 : UTILIZING MICRO-IMMUNOELECTRODES TO MEASURE SYNAPTIC ACTIVITY-INDUCED CHANGES OF AMYLOID- β IN AN ALZHEIMER'S DISEASE MOUSE MODEL

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Abstract:

The overproduction and aggregation of the amyloid- β (A β) peptide is one of the defining features of Alzheimer's disease (AD) pathology. A β is secreted into the extracellular space, where under normal physiological conditions it is cleared from the brain interstitial fluid (ISF). In AD, A β accumulates in the ISF and gives rise to pathological species. We have developed a novel micro-immunoelectrode (MIE) to rapidly measure A β levels in the ISF using electrochemical techniques (Prabhulkar et al., 2012; Yuede et al., 2016). The MIEs consist of a carbon fiber electrode to which we covalently attach antibodies against the peptide of interest. We then use amperometry to measure the oxidation of a tyrosine residue within human A β and correlate the resultant current to the concentration of A β present. As tyrosine oxidation is irreversible, each oxidized A β peptide will only be measured once, so will not contribute to MIE signal over time and we can specifically measure changes in the ISF pool. With this technique, we have been able to measure longitudinal changes in A β_{40} , A β_{42} , and A β oligomers every 60 seconds in vitro and in vivo in mouse models of AD. Now, we are utilizing MIEs to measure rapid changes in A β levels in ex vivo slice physiology following electrical and pharmacological manipulation of synaptic activity in an AD mouse model. With the development of this technique, we can now study acute changes in A β levels to better understand the pathological mechanisms of AD.

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PO 8 : Development of a Novel Electrochemical DNA Biosensor for the Assessment of Heavy Metal-Induced Genotoxicity in Olive Mill Wastewater

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Abstract:

Heavy metal contamination originating from olive mill wastewater constitutes a major environmental concern in Mediterranean regions, where large volumes of this effluent are generated seasonally. Conventional detection methods—atomic absorption spectrophotometry, flame emission spectrometry, molecular spectrophotometry, and chromatographic techniques—remain largely confined to laboratory settings due to their cost and operational complexity. Electrochemical approaches offer a promising alternative, combining high sensitivity, simplicity, rapid analysis, and compatibility with on-site applications.

In this work, a novel electrochemical DNA biosensor was developed to investigate heavy metal–DNA interactions and assess their genotoxic potential. Well-defined oxidation signals corresponding to guanine and adenine were clearly resolved, with peak currents increasing proportionally with DNA concentration, confirming the sensor's responsiveness and analytical reliability. Upon incubation with Pb^{2+} and Cd^{2+} , a significant enhancement of these oxidation currents was observed, consistent with a direct interaction with DNA and progressive destabilization of the double helix, thereby increasing the accessibility of purine bases. Prolonged exposure to Pb^{2+} further suggested oxidative damage to DNA bases, pointing to a genotoxic mechanism beyond simple electrostatic binding.

The developed biosensor was subsequently applied to real samples derived from olive mill waste treatment processes. The results showed that the sample matrix strongly influences the electrochemical response, likely due to competitive interactions and complexation effects that modulate metal bioavailability and DNA accessibility. Based on these findings, this biosensor is proposed as a promising tool for evaluating the toxicity of water and biochar within the framework of the 3D-STELLAR project, which aims to treat and valorize toxic wastewater generated from olive oil extraction units, in line with the principles of green and sustainable chemistry and a circular economy approach.

Keywords :

Electrochemical DNA biosensor; Heavy metals; Genotoxicity; Olive mill wastewater; DNA–metal interaction

PO 9 : DNA IS A ROPE, NOT A WIRE: PROBE DENSITY GOVERNS ELECTRON TRANSFER IN MB-DNA ELECTROCHEMICAL BIOSENSORS

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Abstract:

Electrochemical DNA hybridization biosensors employing methylene blue (MB)-labeled probes are widely used for nucleic acid detection. However, the origin of their signal remains unresolved, particularly whether electron transfer occurs through the DNA base π -stack or through oligonucleotide conformational flexibility that brings the redox label to the electrode surface. Here, we address this question by systematically controlling the density of MB-labeled DNA probes on gold electrodes and evaluating its effect on the electron transfer mechanism.

Key preparation conditions were first optimized, including probe immobilization, hybridization time and target concentration. Thiolated and non-thiolated DNA controls, together with aqueous washing, distinguished covalently attached probes from nonspecifically adsorbed oligonucleotides. Gold electrodes were then functionalized with MB-labeled DNA across a 1000-fold concentration range (0.53-30.30 pmol/cm²), while actual surface coverage was independently quantified by RuHex chronocoulometry.

Cyclic voltammetry at varying scan rates revealed a clear probe density dependent transition. In low-density single-stranded DNA systems, electron transfer was consistent with a surface-confined, adsorption-controlled process, whereas higher probe densities pronounced redox peak separation and diffusion-like charge transfer. After hybridization with complementary targets, this diffusion-controlled response was observed regardless of the initial probe density, most likely due to the reduced double-stranded DNA flexibility.

Targets containing one or two mismatches produced MB signals practically indistinguishable from fully complementary sequences, even under varying ionic strength conditions. These results indicate that base mismatches do not significantly affect electron transfer in our model system. Instead, the signal is primarily determined by probe density, DNA flexibility and MB accessibility to the electrode surface, establishing probe surface density as a key design parameter for MB-based DNA biosensors.

PO 10 : AN RPA-ENHANCED PHOTOELECTROCHEMICAL BIOSENSING PLATFORM FOR THE DETECTION OF *KRASG12C* MUTATION IN CANCER

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Abstract:

Cancer remains a leading cause of mortality worldwide, with approximately 20 million new cases and 9.7 million deaths reported in 2022 and a projected rise to 35 million cases by 2050.¹ Despite advances in diagnosis and therapy, relapse and metastasis remain major clinical challenges and key drivers of cancer-related mortality, underscoring the need for sensitive and reliable tools for early detection and disease monitoring. Current cancer diagnostics rely heavily on molecular techniques for biomarker detection and patient stratification. *KRAS* is one of the most frequently mutated oncogenic driver genes and is associated with several fatal cancer types including lung, pancreatic and colorectal cancers. *KRASG12C* mutation detection is particularly relevant as it is currently the only *KRAS* variant with available targeted therapies.² Current gold-standard molecular diagnostic techniques for *KRAS* mutation detection include targeted next-generation sequencing (NGS) and polymerase chain reaction (PCR)-based methods, although they are highly specific and sensitive, they still require expensive instrumentation, trained personnel and long assay times.³ To overcome these current diagnostic limitations, this work focuses on developing a novel singlet oxygen (¹O₂)-based photoelectrochemical (PEC) sensing platform integrating recombinase polymerase amplification (RPA) for sensitive *KRASG12C* detection. The incorporation of RPA enhances analytical sensitivity by enabling rapid isothermal amplification (without thermal cycling as PCR) of *KRASG12C* within approximately 30 minutes. Importantly, this RPA strategy uses allele-specific primers optimized for amplification efficiency and specificity to discriminate *KRASG12C* from the wild type. Moreover, the use of labeled RPA primers allows direct functionalization of the amplicons: a biotin-labeled primer enables immobilization onto streptavidin-coated magnetic beads, while a Rose Bengal-labeled reverse primer serves as the photosensitizer for singlet oxygen generation upon illumination at 550 nm (Fig. 1). Overall, this approach enables rapid, low-cost *KRASG12C* screening, offering a faster and more affordable alternative to current methods.

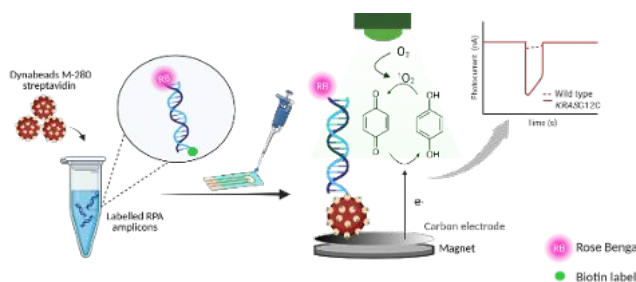


Figure 1. An illustration of the RPA-based PEC platform for sensitive *KRASG12C* detection.

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PO 11 : HYBRIDIZATION CHAIN REACTION-AMPLIFIED SINGLET OXYGEN-BASED PHOTOELECTROCHEMICAL BIOSENSING FOR NUCLEIC ACID DETECTION

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Abstract:

Nucleic acid biomarkers enable early, sequence-specific detection of cancer-associated mutations. However, the established tools for reading them, such as PCR and next-generation sequencing, remain costly, complex, and dependent on trained personnel and centralized infrastructure, limiting their use at the point of need.

Our group addressed these challenges by developing a singlet oxygen ($^1\text{O}_2$)-based photoelectrochemical readout for nucleic acid detection.^{1,2} A photosensitizer conjugated to a detection probe generates $^1\text{O}_2$ under illumination, which reacts with a redox-active reporter in solution to produce a measurable current. Magnetic beads concentrate the assay components at the electrode, simplifying sample handling, while a light on/off switching mode suppresses matrix effects at their source. As a label-based readout, its sensitivity scales with the number of photosensitizer labels a single target brings into the detection volume.

To increase this number without enzymes or thermal cycling, we integrate hybridization chain reaction (HCR), an isothermal, enzyme-free amplification strategy.³ A single recognition event triggers the self-assembly of photosensitizer-labeled hairpins into a long DNA concatemer, accumulating many labels per target. Since this concatemer is the structure the readout interrogates, HCR is embedded directly in signal generation rather than appended as a separate amplifier. Integrating HCR also introduces label positioning as a new design variable, absent in a conventional sandwich format where the label occupies a single fixed site. Using the KRAS G12C oncogenic mutation as a model target, we show that HCR-driven label accumulation yields a clear photocurrent enhancement over non-amplified controls, consistent with results obtained by gel electrophoresis. By systematically varying the number of labeled hairpins, as well as reaction parameters such as hairpin concentration and reaction volume, we find that labeling architecture strongly affects amplification efficiency, with toehold-proximal labeling impairing HCR polymerization. These findings provide a rational basis for designing hairpin probes optimized for $^1\text{O}_2$ -based readout.

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PO 12 : AI-DRIVEN BIOSENSING FOR PERSONALIZED HEALTHCARE: WEARABLE MONITORING AND RAPID POINT-OF-CARE DIAGNOSIS

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Abstract:

Artificial intelligence (AI) is expanding the capabilities of wearable sensors and point-of-care testing (POCT), creating new opportunities for continuous health monitoring, rapid diagnosis, and personalized clinical decision support. We reviewed the applications of machine learning and deep learning in biosensor data processing, sensor fusion, automated sample analysis, disease classification, and clinical decision-making (1). AI-enabled wearable sensors support continuous and potentially non-invasive monitoring of physiological signals and biomarkers, while intelligent POCT platforms enable rapid analysis at or near the site of patient care. Applications include cardiovascular and neurological monitoring, cancer screening, infectious disease testing, chronic disease management, and the analysis of complex biomarker profiles. AI can improve the interpretation of multivariate sensor outputs, reduce noise and interference, integrate data from multiple sensors, recognize clinically relevant patterns, and facilitate portable or smartphone-assisted diagnostics. Key advances include TinyML-supported electrochemical sensing, machine-learning analysis of urinary biomarker panels, deep-learning-assisted ultrasound interpretation, and AI-controlled microfluidic immunoassays. These advances highlight AI potential to transform conventional sensing platforms into intelligent systems that produce clinically meaningful information. However, clinical translation remains constrained by challenges related to data quality, algorithm validation, model interpretability, sensor reproducibility, privacy and cybersecurity, regulatory approval, interoperability, cost, and user acceptance. Explainable AI, representative clinical datasets, prospective validation, secure data-management frameworks, and user-centered device design are therefore essential for responsible implementation. Overall, the convergence of AI, wearable sensing, and POCT could support a transition from episodic laboratory testing toward continuous monitoring and rapid, personalized diagnostic support.

Keywords:

Artificial intelligence; wearable sensors; biosensors; point-of-care testing; machine learning.

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PO 13 : ADDITIVE MANUFACTURING FOR BIOELECTROANALYTICAL DIAGNOSTICS AND SENSING

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Abstract:

Additive Manufacturing (AM), commonly known as 3D printing, has emerged as a powerful technology in electrochemistry, offering rapid prototyping, design freedom, and the ability to produce integrated devices, providing a unique route to on-demand, low-cost (bio)analytical platforms. AM enables the integration of hybrid architectures within a single device, in which 3D printing provides the structure, fluidic system, and mechanical frame, and functional components, manufactured using complementary technologies (e.g., paper-based sensors, stencil-printed electrodes, immunochemical elements), can be readily integrated as needed. From an analytical perspective, this approach enables the automation of analytical procedures, yielding fully integrated analysis systems on board 3D devices. Herein, two representative 3D-printed electrochemical platforms are presented. (i) A 3D-printed analytical device integrating functional paper components for the direct determination of amitraz in apicultural matrices. The platform combines a 3D-printed microfluidic architecture with interchangeable paper-based electrochemical sensors and a 'hydrolysis-paper' component that enables in situ conversion of amitraz into its electroactive metabolite, 2,4-dimethylaniline (2,4-DMA). This architecture enables sample processing and sensing on a single device, achieving sensitive, reproducible, and low-cost detection of amitraz in honey and beeswax within a few minutes, without the need for sample pretreatment. (ii) An integrated 3D-printed electrochemical microfluidic device for immunocapture and electrochemical assessment of transferrin saturation (TSAT) in human serum. The device, entirely produced via AM, comprises a platform that incorporates a rotary-valve-controlled microfluidic network, an on-device immunoassay module, and an electrochemical cell fabricated by combining conductive filaments (CB-PLA) and non-conductive filaments (PLA). The system enables selective isolation of transferrin (Tf) and simultaneous electrochemical detection of Tf and Tf-bound iron directly from untreated human serum, providing clinically relevant information with significantly reduced analysis time. This presentation positions 3D printing as a pivotal technology for next-generation electrochemical systems for on-field and Point-of-Care testing, enabling the seamless integration of heterogeneous functional components into all-in-one analytical devices that encompass entire analytical processes.

Acknowledgement:

This research was funded by the progetto centro di ricerca per l'innovazione nel settore agroalimentare – addendum “Completamento del Centro Europeo AGRIBIOSERV” – C43C22002360001 a valere sulla sub misura B 4.1 del Programma unitario di intervento per le aree del terremoto del 2009 e 2016 PNRR/FONDO COMPLEMENTARE.

PO 14 : DEVELOPMENT OF DNA APTAMER BIOSENSING PLATFORM FOR TETRACYCLINE RECOGNITION: A COMBINED EXPERIMENTAL AND IN SILICO STUDY

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Abstract:

The widespread use of tetracycline has led to its increasing occurrence in environmental compartments, raising concerns about ecological safety and potential risks to human health. In this study, we report a novel sensing strategy based on the repurposing of a single-stranded DNA stem-loop sequence, initially designed for hybridization-based recognition, into an aptamer-like sequence capable of binding tetracycline. Preliminary experimental investigations indicated that this DNA architecture can interact with tetracycline molecules. Subsequently, computational analyses were performed to elucidate the structural interactions between the aptamer and tetracycline.

Experimentally, the selected sequence was integrated into a gold nanoparticle (AuNP)-based colorimetric assay for tetracycline detection. The sensing mechanism relies on target-induced aggregation of AuNPs, resulting in a visible color change and a quantitative absorbance response (A₆₅₀/A₅₂₀). The developed platform exhibited a limit of detection of 10⁻⁵ M. Selectivity was assessed in the presence of structurally related and biologically relevant interfering molecules, with tetracycline consistently producing a significantly stronger signal.

To strengthen the structural basis of this design, two complementary approaches were employed for three-dimensional structure prediction. A thermodynamics-based method using Mfold was first applied to generate multiple thermodynamically favorable conformations, followed by structural selection to identify representative candidates. In parallel, a deep learning-based modeling approach provided an alternative 3D structure. This combined strategy improved structural reliability and yielded robust starting configurations for subsequent molecular modeling analyses.

To elucidate the recognition mechanism at the molecular level, molecular docking followed by molecular dynamics simulations was performed. Structural analyses, including root mean square deviation (RMSD), radius of gyration (Rg), and solvent-accessible surface area (SASA), and MM/PBSA calculations confirmed the high stability of the aptamer-tetracycline complex, in agreement with experimental observations. These results highlight the robustness and potential applicability of the proposed sensing platform for tetracycline detection.

Keywords:

Tetracycline, DNA aptamer, molecular docking, molecular dynamic, Colorimetric biosensor.

PO 15 : SIMULTANEOUS PROFILING AND QUANTIFICATION OF TAU AND ITS PHOSPHORYLATED ISOMERS USING ENGINEERED FRAC NANOPORES

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Abstract:

Tau aggregation and hyperphosphorylation are key pathological hallmarks and early diagnostic biomarkers of Alzheimer's disease (AD), but simultaneous profiling and quantification of Tau and its phosphorylated isoforms remain technically challenging. We engineered a high-resolution FraCG6DG13F nanopore by introducing an acidic aspartate residue at the conserved G6 site of FraCG13F. Optimized low-pH and high-salt conditions reduced electroosmotic flow, improving temporal resolution for polypeptide detection. Combined with machine-learning algorithms, our workflow enabled high-precision peptide analysis and positional phosphorylation isomer discrimination. Using Tau (2N4R) as a model—focusing on S400 and S402 phosphorylations relevant to early neurodegeneration—12 tryptic peptides were identified with >91.1% accuracy. Furthermore, positional phosphorylation isomers were precisely resolved and quantified in mixed digests with >93% accuracy, showing sensitivity comparable to microgram-level mass spectrometry. This study demonstrates advances in the analysis of phosphorylated Tau in nanopore-based single-molecule methods, highlighting its potential for high-resolution characterization and accurate quantification of proteins with PTMs.

PO 16 : RAPID ULTRASOUND-ASSISTED SYNTHESIS OF A MOLECULARLY IMPRINTED POLYMER FOR DUAL-MODE OPTICAL DETECTION OF PROMETHAZINE

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Abstract:

Molecularly imprinted polymers (MIPs) have emerged as versatile synthetic recognition platforms, offering tailor-made binding sites for target molecules and enabling selective chemical sensing¹. Herein, we report a MIP for promethazine (PMZ), a phenothiazine-based pharmaceutical compound², developed through a rapid ultrasound-assisted synthesis strategy. The imprinted material was obtained using probe sonication within 15 min, providing a straightforward and time-efficient route to the formation of the recognition matrix. Beyond molecular recognition, the resulting material exhibited a distinctive dual-mode optical response, combining colorimetric and fluorescence signals upon interaction with PMZ. A characteristic red–pink coloration accompanied by fluorescence under UV irradiation at 365 nm was observed, while variations in the optical response were associated with changes in PMZ loading and release. The recognition properties were further assessed through adsorption isotherm studies, yielding an imprinting factor of 8.66 and a maximum adsorption capacity of 80.72 mg g⁻¹. These preliminary findings demonstrate the potential of combining rapid ultrasound-assisted molecular imprinting with dual-mode optical and redox-assisted signal generation for the development of selective PMZ recognition platforms. Further investigation of the underlying optical and redox mechanisms will be required to establish the stimulus-responsive behavior of the material.

Keywords:

Molecularly imprinted polymer; Promethazine; Dual-mode optical detection; Molecular recognition; Fluorescence; Colorimetric sensing.

PO 17 : CYCLIC ALKYL(AMINO)CARBENE-BASED ORGANIC RADICALS STABLE IN ACIDIC, BASIC, AND BIOLOGICAL MEDIA, AND FLUORIDE SENSORS

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Abstract:

Compared with N-heterocyclic carbenes (NHCs), cyclic (alkyl)(amino) carbenes (CAACs) and their derivatives exhibit enhanced nucleophilicity (higher HOMO energy and greater σ -donating ability) and electrophilicity (lower LUMO energy and enhanced π -accepting ability). Consequently, several highly reactive and otherwise unstable intermediates have been successfully isolated using CAACs that would be inaccessible through conventional approaches or with NHCs. Furthermore, numerous transition-metal catalysts supported by CAACs have demonstrated improved catalytic performance compared with their NHC analogues. Motivated by these intriguing electronic and steric properties, our research has focused on the isolation of air- and moisture-stable organic radicals using CAACs. This presentation will discuss the synthesis and properties of two stable, structurally characterized glyoxal radical cations, 1^{++} and 2^{++} (Figure 1), stabilized by cyclic (alkyl)(amino)carbene (CAAC) and bicyclic (alkyl)(amino)carbene (BICAAC) ligands, respectively. Notably, 1^{++} and 2^{++} remain stable under harsh conditions, including exposure to strong acids and bases, mild oxidizing and reducing agents, and even bovine serum. Remarkably, 2^{++} represents the first isolated radical stabilized by a BICAAC ligand. The presentation will also highlight the potential applications of carbene-derived low-valent phosphorus compounds as fluoride-sensing agents.

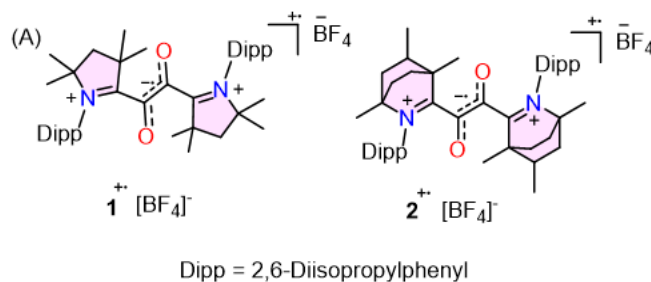


Figure 1: Molecular structures of 1^{++} and 2^{++}

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- (3) C. Majumder, A. Sharma; B. Das, R. Yadav, S. Kundu, *J. Am. Chem. Soc.*, 2025, **147**, 6905–6913.

PO 18 : BIOSENSOR FOR ELECTROCHEMICAL NUCLEASE ACTIVITY DETECTION

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Abstract:

Nucleases, enzymes that cleave nucleic acids, have recently emerged as potential biomarkers for various diseases, particularly cancer. Studies indicate that the activity of nucleases is abnormal in cancer [1]. Nevertheless, the detection of nucleases remains challenging. Existing methods, such as “DNaseAlert™,” are used in research and development but have not been widely adopted for diagnostic purposes. The aim of this research was to develop a nuclease activity-based electrochemical biosensor capable of differentiating endonucleolytic and exonucleolytic activities using a single assay. A methylene blue (MB)-labelled ssDNA probe was immobilized on a gold electrode through a thiol linker. In the absence of target DNA, the surface-confined MB label produced a characteristic redox signal. Hybridization with an ssDNA substrate containing self-complementary regions that form defined dsDNA restriction sites altered the local environment and electron-transfer behaviour of MB, resulting in a change in the voltammetric response, reflected by a shift in the MB oxidation peak potential. In the absence of nuclease activity, the intact ssDNA substrate partially hybridized with the electrode-bound probe and produced an intermediate oxidation peak shift. Incubation with the endonuclease BsuRI released fully complementary ssDNA fragments, enabling more efficient probe hybridization and producing a markedly increased potential shift. In contrast, digestion with exonuclease I degraded the ssDNA substrate and removed sequences required for hybridization, resulting in a response close to the baseline of the unhybridized MB-labelled probe. Thus, the system provides a proof of concept for distinguishing endonucleolytic and exonucleolytic DNA cleavage activities using a single electrode and a single electrochemical readout.

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PO 19: LASER-INDUCED SYNTHESIS OF METAL-OXIDE NANOZYMES ON ACTIVATED CHARCOAL MEMBRANE AS OXIDASE MIMICS FOR COLORIMETRIC VERTICAL-FLOW ASSAYS

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Abstract:

Natural enzymes are highly efficient and selective biocatalysts with broad applications in medical diagnostics, environmental monitoring, and industrial catalysis. However, their practical use can be limited by high production costs, poor stability, and limited reusability. Nanozymes, nanomaterials capable of mimicking enzyme-like catalytic activities, have therefore attracted considerable interest as more robust and tunable alternatives. Nevertheless, conventional nanozyme synthesis often involves multistep procedures, chemical reagents, and solvent-intensive processing.

In this work, we propose a one-step, in situ laser-assisted strategy for the synthesis of metal-oxide nanozymes directly on activated charcoal membranes. The resulting catalytic membranes are designed for integration into a vertical-flow analytical platform with a dual colorimetric readout: quantitative spectrophotometric measurements for laboratory validation and smartphone-based RGB analysis for potential equipment-free applications. A preliminary screening of transition-metal precursors based on Fe, Co, Ni, and Mn was first conducted to identify the most promising catalytic system. The Fe precursor was subsequently optimized by investigating laser power, resolution, and pattern-filling geometry, including crossfill, grid, zig-zag, and diagonal patterns. The effects of precursor amount (0.1–10 mmol) and reaction-medium pH (3.5–5.5) were also evaluated. Particular attention was given to the balance between catalytic activity and membrane integrity, since increasing laser intensity can enhance material transformation while simultaneously compromising the mechanical properties required for reliable flow-through operation.

These results demonstrate the potential of laser-assisted synthesis for the direct fabrication of catalytic membranes and provide a basis for the development of integrated nanozyme-based vertical-flow analytical devices. Ongoing work is focused on optimizing the membrane architecture and analytical response toward rapid colorimetric detection of target analytes.