Program of the XXV International Symposium on Bioelectrochemistry and Bioenergetics of the Bioelectrochemical Society

26-30 May, 2019

Limerick, Ireland

B E S
The Bioelectrochemical Society

The Bioelectrochemical Society
Chemin du Closelet 2
1006 Lausanne
Switzerland

Printed in Ireland

Organising Committee

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Micheál Scanlon, *University of Limerick, Ireland*
Renata Bilewicz, *University of Warsaw, Poland*

Program

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B E S
The Bioelectrochemical Society

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19:00 – 19:30
Luigi Galvani Prize Lecture, KBG-12

Monday, 27 May 2019

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Poster Session

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18:00 – 20:00
Poster Session

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Program of the XXV International Symposium on Bioelectrochemistry and Bioenergetics

Sunday, 26 May - Afternoon

17:30 – 18:00

Opening Ceremony, KBG-12

Giulio Milazzo Prize Lecture

Room: KBG-12

Chaired by: L. Gorton

18:00 to 18:50

Wolfgang Schuhmann (Faculty of Chemistry and Biochemistry, Ruhr University Bochum, Bochum, Germany)

From nanobiosensors to bioelectrocatalysis and photobioelectrocatalysis

Luigi Galvani Prize Lecture

Room: KBG-12

Chaired by: L. Gorton

19:00 to 19:30

Nicolas Plumeré (Center for Electrochemical Sciences, Ruhr-University Bochum, Bochum, Germany)

Introducing Robustness into Fragile Bioelectrochemical Systems

Monday 27 May 2019-Morning

Plenary

Room: KBG-12

Chaired by: E. Lojou

09:00 to 09:45

Hagan Bayley (Department of Chemistry, University of Oxford, Oxford, United Kingdom)

Protein pores as nanoreactors for single-molecule covalent chemistry

S1 Biopower and Bioenergetics

Room: S2-005

Chaired by: R. Milton and S. Ferugia

09:50 to 10:10

Xinxin Xiao (Department of Chemistry, Technical University of Denmark, Kongens Lyngby, Denmark), Edmond Magner, Till Siepenkoetter, Jingdong Zhang

Flexible nanoporous gold film based enzymatic biofuel cells for wearable power sources

10:10 to 10:30

Julian Szczesny (Ruhr-University Bochum, Faculty of Chemistry & Biochemistry, Analytical Chemistry - Center for Electrochemical Sciences, Bochum, Germany), Felipe Conzuelo, Wolfgang Lubitz, Nikola Markovic, Inês A. C. Pereira, Nicolas Plumeré, Adrian Ruff, Wolfgang Schuhmann, Sónia Zacarias

High-Performance Gas Breathing H_2/O_2 Biofuel Cells Comprising a Hydrogenase/Polymer Based Bioanode

10:30 to 10:50

Isao Shitanda (Department of Pure and Applied Chemistry, Tokyo University of Science, Noda, Japan), Yoshinao Hoshi, Masayuki Itagaki, Tsutomu Mikawa, Kotaro Takamatsu, Seiya Tsujimura

Improving the Performance of Lactate/Oxygen Biofuel using MgO-templated Carbon coated Carbon Cloth Electrodes

10:50 to 11:10
Coffee Break

11:10 to 11:40 Keynote
Christopher Howe (Department of Biochemistry, University of Cambridge, Cambridge, United Kingdom)
Biophotovoltaics: exoelectrogenic activity of photosynthetic microorganisms

11:40 to 12:00
Darren Buesen (Center for Electrochemical Sciences, Bochum, Germany), Thomas Hoefer, Nicolas Plumeré, Huijie Zhang
A Kinetic Model for Redox-Active Film Based Photoelectrodes

12:00 to 12:20
Sergey Shleev (Biomedical Science, Malmoe University, Malmoe, Sweden), Magnus Falk, Juan Gonzalez-Martinez, Elena González-Arribas, Adrian Ruff, Wolfgang Schuhmann, Javier Sotres
Triple Hybrid Electric Power Biodevice for Harvesting and Storage of Chemical and Solar Energies

12:20 to 12:40
Marcos Pita (Biocatalysis / Catalysis Institute, CSIC, Madrid, Spain), Jose Carlos Conesa, Antonio L. De Lacey, Carmen Jarne, Logan Paul, Sergey Shleev
Underpotential Photoelectrooxidation of Water by SnS2-Laccase Cocatalysts on Nanostructured Electrodes with Only Visible Light Irradiation

S3 Sensing and Biointerfaces

Room: Room: KBG-12

Chaired by: W. Kutner and M. Fojta

09:50 to 10:10

Miroslav Fojta (Department of Biophysical Chemistry and Molecular Oncology, Institute of Biophysics of the CAS, Brno, Czech Republic), Zuzana Babkova, Stanislav Hason, Ludek Havran, Martina Outla, Hana Pivonkova, Jan Spacek

Adsorption and Electroreduction of Cytosine Oligonucleotide Blocks at the Mercury Electrode

10:10 to 10:30

Anna Banasiak (Applied Electrochemistry Group, Technological University Dublin, Dublin, Ireland), John Cassidy, John Colleran

DNA Biosensors as a Tool to Determine the Mode of Action and Nuclease Activity of Novel Bioinorganic Compounds

10:30 to 10:50

Marie Berthuel (Department of Molecular Chemistry, BEA/DCM/UGA, Gières, France), Serge Cosnier, Omar El Mazria, Chantal Gondran, Karine Gorgy, Stefan McMurtry

Electrochemical Biosensors based on Microdot Arrays

10:50 to 11:10

Coffee Break

11:10 to 11:40 Keynote

James. F. Rusling (Department of Chemistry, University of Connecticut, Storrs, USA), Ketki Bhalerao, Tianqi Chen, Karteek Kadimisetty, Mohammed Sharafeldin

Protein Biomarker Diagnostics for Cancer and Metastatic Cancer using Low Cost 3D-printed microfluidic arrays

11:40 to 12:00

Jesus Iniesta (Physico Chemical and Institute of Electrochemistry, Alicante, Spain), Craig Banks, Johnn Graham, Naiara Hernández-Ibáñez, Andrés Martín-Gómez, Vicente Montiel, Ignacio Sanjuán, Thies Thiemann

Fundamental insights into the electrooxidation of halocytosines at carbon based electrodes: mechanism, electroanalytical and computational studies

12:00 to 12:20

Michael Kitching (School of Chemistry, Dublin, Ireland), Aoife Morrin

Development of a DNA based electrochemical biosensor to determine the overall genotoxicity of multi-pesticide soil extract

12:20 to 12:40

Wlodzimierz Kutner (Institute of Physical Chemistry, Polish Academy of Sciences, Kasprzaka 44/52, Warsaw, Poland), Katarzyna Bartold, Silvia Cauteruccio, Francis D'Souza, Karolina Golebiewska, Emanuela Licandro, Wojciech Lisowski, Agnieszka Pietrzyk-Le

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S6 Emerging Topics in Bioelectrochemistry

Room: S2-006

Chaired by: L. Alfonta and A. de Lacey

09:50 to 10:10

Silvia Voci (Institute of Molecular Sciences/University of Bordeaux, Bordeaux, France)

Surface-Confined Electrochemiluminescence Microscopy of Cell Membranes

10:10 to 10:30

Wei Chung Sim (School of Chemistry, Suranaree University of Technology, Nakhon Ratchasima, Thailand), Panida Khunkaewla, Albert Schulte

A Three-electrode μL-cell for Green (Bio-)electroanalysis with Common Macro- and Microelectrodes

10:30 to 10:50

Antonio De Lacey (Instituto de Catálisis, CSIC, Madrid, Spain), Gabriel García-Molina, Iván López-Montero, Arturo Luis Meza-López, Paolo Natale, Ines Pereira, Marcos Pita, Marisela Vélez, Sonia Zacarias

Combining Membrane-Bound Enzymes on Electrodes for Electroenzymatic ATP Regeneration or Detection

10:50 to 11:10

Coffee Break

11:10 to 11:40 Keynote

Frank N. Crespilho (University of São Paulo, São Paulo, Brazil)
Operando, in situ and on-line techniques for real-time measurements in Bioelectrochemistry

11:40 to 12:00

Anne de Poulpiquet (Laboratory of Bioenergetics and Protein Engineering, Aix-Marseille University - CNRS, Marseille, France), Stéphane Arbaut, Laurent Bouffier, Bertrand Goudeau, Artemis Kosta, Hugo Le Guenno, Pauline Lefrançois, Elisabeth Lojou, Bastien Tassy
In Situ Fluorescence Microscopy for the Study of Electro-enzymatic Reactivity

12:00 to 12:20

Stephane Arbault (Institute of Molecular Sciences, CNRS UMR-5255, University of Bordeaux, Pessac, France), Bertrand Goudeau, Pauline Lefrançois, Jérôme Santolini
Electroanalysis of Enzymatic Activities at a Single Biomimetic Microreactor

12:20 to 12:40

Michael Füeg (Chemical Engineering Department, University of Alcalá, Alcalá de Henares, Spain), Abraham Esteve-Núñez, Pau Rodenas Motos
Towards the understanding how silver species influence electroactive Geobacter sulfurreducens biofilms

Monday 27 May 2019-Afternoon

S1 Biopower and Bioenergetics

Room: S2-005

Chaired by: S. Shleev and M. Di Lorenzo

14:00 to 14:30 Keynote

Daniel R. Bond (Department of Plant and Microbial Biology, University of Minnesota, Twin Cities, Saint Paul, USA)

Electron transfer out of Geobacter: most models are wrong, are some useful?

14:30 to 14:50

Gauthier Menassol (TIMC-IMAG, SYMMES-CEA, Grenoble, France), Lionel Dubois, Donald Martin, Abdelkader Zebda

Implantable Hybrid Biofuel Cell with Abiotic Biocathode and Enzymatic Bioanode

14:50 to 15:10

Richard Bennett (Biomolecular Electronics Research Laboratory, National University of Ireland Galway, Galway, Ireland), Dónal Leech

Enzymatic Biopower: Approaches for optimisation of enzyme electrode performance

15:10 to 15:30

Pankaj Vadgama (School of Engineering and Materials Science, Queen Mary Unversity of London, Mile End Road, London, United Kingdom), Jean-Pierre Alcaraz, Awatef Ben-Tahar, Francois Boucher, Philippe Cinquin, Donald Martin

Optimization of a biocompatible, implantable bioanodebased on glucose dehydrogenase

15:30 to 15:50 Invited Lecture

Tim Cummins (AltraTech Ltd, Ireland)

Bioelectrochemical Sensor Commercialisation

15:50 to 16:10

Coffee Break

16:10 to 16:30

Yolina Hubenova (Biochemistry and Microbiology, Plovdiv, Bulgaria)

New electrogenic bacteria Paenibacillus dendritiformis MA-72

16:30 to 16:50

Lo Gorton (Institute of Chemistry, Lund University, Lund, Sweden), Dónal Leech, Ross D. Milton, Shelley D. Minteer, Galina Pankratova

Wiring of Bacterial Cells to Electrodes through Redox Polymers

16:50 to 17:10

Xiaohong Chen (Department of Molecular Chemistry, Université Grenoble Alpes, Grenoble, France), Serge Cosnier, Fabien Giroud, Andrew Gross

Use of Alginate Hydrogel Coating to Improve Buckypaper-based Bioanode Stability

17:10 to 17:30

Alexander Kuhn (ENSCBP, University Bordeaux, Pessac, France), Peer Fischer, Sebastien Gounel, Nicolas Mano, Andrew Mark, Pascal Merceau, Jerome Roche, Emmanuel Suraniti

Helping enzymes to fight against thermodynamics: bioelectrocatalytic uphill production of hydrogen

17:30 to 17:50

Laura Wey (Department of Biochemistry, University of Cambridge, Cambridge, United Kingdom), Xiaolong Chen, Chris Howe, Joshua Lawrence, Jenny Zhang

Biological origins of cyanobacterial exoelectrogenesis

17:50 to 18:10

Fan Yang (College of Chemical and Biological Engineering, Zhejiang University, Hangzhou, China), Zhongjian Li, Bin Yang

Remediation of Cd Contaminated Soil based on Bioelectrochemical Reduction of Sulfate

S3 Sensing and Biointerfaces

Room: KBG-12

Chaired by: F. Lisdat and K. De Wael

14:00 to 14:30 Keynote

Benoit Limoges (Universite Paris Diderot / CNRS, Paris, France)
Molecular sensing through autocatalytic deprotection of a chemosensitive probe into a redox cycler and its promising use in bioelectroanalysis

14:30 to 14:50

Mihaela-Cristina Bunea (National Institute of Material Physics, Atomistilor 405A, Magurele, Romania), Teodor Adrian Enache
Redox behavior of azathioprine and it's interaction with DNA

14:50 to 15:10

David Novak (Department of Medical Chemistry and Biochemistry, Palacký Univerzity, Olomouc, Czech Republic), Krzysztof Stolarczyk, Jan Vacek, Jiri Vrba, Martina Zatloukalova
Electrochemical Cysteamine Assay as a New Approach for Studying the Reactivity of Bioactive Ligands

15:10 to 15:30

Tadas Penkauskas (Department of Bioelectrochemistry and Biospectroscopy, Vilnius University Life Sciences Center, Vilnius, Lithuania), Marija Jankunec, Gintaras Valincius
Tethered Bilayer Lipid Membranes – a Comprehensive Tool for Electrochemical Studies of Pore-Forming Toxins

15:30 to 15:50

Rima Budvytyte (Department of Bioelectrochemistry and Biospectroscopy, Life Sciences Center, Vilnius Univeristy, Vilnius, Lithuania), Aiste Gulla, Tadas Penkauskas, Julija Razumiene
The Interaction of Heat Shock Proteins with Lipid Membranes: a Novel Diagnostic Target

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Coffee Break

16:10 to 16:30

Fred Lisdat (Biosystems Technology, Technical University Wildau, Wildau, Germany), Wolfgang Parak, Marc Riedel

Light-triggered sugar detection using semiconductor materials

16:30 to 16:50

Graziela C. Sedenho (São Carlos Institute of Chemistry, University of São Paulo (USP), São Carlos, Brazil), Frank N. Crespilho, Ayaz Hassan

Confinement of Bilirubin Oxidase in Biogel Matrix for Bioelectrocatalytic Oxygen Reduction

16:50 to 17:10

Alonso Gamero-Quijano (Bernal Institute, Department of Chemical Sciences, University of Limerick, Limerick, Ireland), Pierre-Andre Cazade, Grégoire Herzog, Andrés F. Molina-Osorio, Micheál D. Scanlon, Damien Thompson

Interfacial Electron Transfer of Cytochrome c at an Electrified Liquid-Liquid Interface

17:10 to 17:30

Tautgirdas Ruzgas (Biomedical Science, Malmö University, Malmö, Sweden)

Plugging redox reactions into wireless devices: RFID-based biosensor tag

17:30 to 17:50

Robert Johnson (School of Chemistry, University of Lincoln, Lincoln, United Kingdom), Guzman Gil-Ramirez, Victoria Hugill, Adria Martinez-Avino

Building Better Biosensors: Controlling the Surface Density of Redox Probes on a Boron-Doped Diamond Electrode

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Sheida Esmail Tehrani (DTU Nanolab, Technical University of Denmark, Kgs. Lyngby, Denmark), Jenny Emnéus, Arto Heiskanen, Bettina Margrethe Jensen, Quang Long Nguyen, Tautgirdas Ruzgas, Stephan Sylvest Keller

Enzymatic Histamine Biosensor Based on Prussian Blue-Modified 3D Pyrolytic Carbon Microelectrodes

S6 Emerging Topics in Bioelectrochemistry

Room: S2-006

Chaired by: M. Pita

14:00 to 14:30 Keynote

Katharina M. Fromm (Department of Chemistry, University of Fribourg, Switzerland)

Ag+ as electron scavenger: bioinorganic aspects

14:30 to 14:50

Fangyuan Zhao (Analytical Chemistry – Center for Electrochemical Sciences, Ruhr University Bochum, Bochum, Germany), Felipe Conzuelo, Volker Hartmann, Marc Nowaczyk, Adrian Ruff, Matthias Rögner, Wolfgang Schuhmann

Improving the long-term Stability of Photosystem 1-based Photocathodes

14:50 to 15:10

Jan Vacek (Department of Medical Chemistry and Biochemistry, Faculty of Medicine and Dentistry, Palacky University, Olomouc, Czech Republic), David Novak, Michaela Pekarova, Martina Zatloukalova

Electrochemistry of Electrophilic Fatty Acids

15:10 to 15:30

Felipe Conzuelo (Analytical Chemistry - Center for Electrochemical Sciences, Ruhr University Bochum, Bochum, Germany), Volker Hartmann, Marc M. Nowaczyk, Adrian Ruff, Matthias Rögner, Wolfgang Schuhmann, Fangyuan Zhao

Photobioelectrodes with Anisotropic Electron Flow by Oriented Immobilization of Photosystem 1 Monolayers

15:30 to 15:50

Dmitri Ciornii (TH Wildau, Hochschulring 1, Berlin-Brandenburg, Germany), Fred Lisdat

Connecting photosystem I with electrodes by fullerens

Tuesday 28 May 2019-Morning

Plenary

Room: KBG-12

Chaired by: E. Magner

09:00 to 09:45 Keynote

Shana O. Kelley (Department of Biochemistry, University of Toronto, Toronto, Canada)

How Low Can We Go? Ultrasensitive Electrochemical Detection of Biomolecular Analytes

S1 Biopower and Bioenergetics

Room: S2-005

Chaired by: D. Bond

11:10 to 11:40 Keynote

Ann M. Rajnicek (University of Aberdeen, Aberdeen, United Kingdom)

Controlling Cell Migration and Differentiation using Extracellular Electric Fields

11:40 to 12:00 Invited

Stefano Freguia (Advanced Water Management Centre, University of Queensland, Brisbane, Australia), Jurg Keller, Pablo Ledezma, Juliette Monetti

Microbial electrochemistry enables nutrient recovery from urine while delivering benefits to urban wastewater networks

S3 Sensing and Biointerfaces

Room: KBG-12

Chaired by: P. Vadgama and A. Brett

09:50 to 10:10

Huangxian Ju (State Key Lab of Analytical Chemistry for Life Science, Nanjing University, Nanjing, China)

Electrochemical Catalytic Probes for Amplified Biosensing

10:10 to 10:30

Wanderson Silva (Departament of Chemistry, University of Coimbra, Coimbra, Portugal), Christopher M.A. Brett, Mariana Emilia Ghica

Novel Nanocomposite Based on Poly (Brilliant Cresyl Blue)-Deep Eutectic Solvent/Carbon Nanotubes for Biosensing Applications

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Muchun Zhong (School of Engineering and Materials Science, Queen Mary University of London, London, United Kingdom), Le Fang, Steffi Krause, Michael Watkinson

Zinc Ion Sensitive Light-addressable Potentiometric Sensor Based on a Self-assembled Monolayer

10:50 to 11:10

Coffee Break

11:10 to 11:40 Keynote

Martyn G Boutelle (Department of Bioengineering, Imperial College London, London, United Kingdom), Sally A. Gowers, Michelle L. Rogers, Isabelle C. Samper

Portable bioelectroanalysis systems for real-time patient and human tissue monitoring

11:40 to 12:00

Francisco Prieto-Dapena (Department of Physical Chemistry, University of Seville, Seville, Spain), Jacek Lipkowski, Manuela Rueda, Zhangfei Su

Photon-Modulation Infrared Reflection-Absorption Spectroscopy (PM-IRRAS) Study of Mixed Nucleolipid/Phospholipid Monolayers Supported on Au(111) Electrodes. Molecular Recognition Capabilities

S4 New Materials/Materials Chemistry for Bioelectrochemistry

Room: S2-006

Chaired by: W. Schuhmann and S. Cosnier

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Ana Maria Oliveira-Brett (Department of Chemistry, University of Coimbra, Portugal), Ana-Maria Chiorcea-Paquim, Teodor Adrian Enache

Amyloid beta peptides time-dependent structural modifications: AFM and voltammetric characterization

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Francisco Montilla (Instituto Universitario de Materiales, Universidad de Alicante, Alicante, Spain), Ana B. Delgado-Egea, Francisco Huerta, Emilia Morallón, César Quijada

Direct Electrochemistry of Cytochrome c eith Conducting Polymers

10:30 to 10:50

Serge Cosnier (Molecular Chemistry Department UMR CNRS 5250, CNRS-University Grenoble-Alpes, Grenoble, France), Redouane Borsali, Marie Carriere, Fabien Giroud, Karine Gorgy, Andrew Gross, Jules Hammond, Christophe Travelet

Redox Glyconanoparticles and Buckypapers for the Design of Enzymatic Fuel Cells

10:50 to 11:10

Coffee Break

11:10 to 11:40 Keynote

Kenji Kano (Graduate School of Agriculture, Kyoto University, 8116, Kyoto, Japan)

Direct Electron Transfer Performance of Protein Engineered D-Fructose Dehydrogenase

11:40 to 12:00

Tim Bobrowski (Analytical Chemistry – Center for Electrochemical Sciences, Ruhr University Bochum, Bochum, Germany), Andrzej Ernst, Adrian Ruff, Wolfgang Schuhmann

Long-term stable glucose determination using redox polymer/enzyme-based amperometric biosensors

Tuesday 28 May 2019-Afternoon

S1 Biopower and Bioenergetics)

Room: S2-005

Chaired by: D. Leech

14:30 to 14:50

Christopher Francois (Bristol Bioenergy Centre, T Block, Frenchay campus, Coldharbour Lane, Bristol, United Kingdom), John Andrews, Daniel Gapes, John Greenman, Ioannis A. Ieropoulos

Wet Oxidation Liquor Treatment using Microbial Fuel Cells

14:50 to 15:10

Mark Platt (Department of Chemistry, Loughborough University, Loughborough, United Kingdom)

Resistive Pusle Sensors for biosensors and imaging of nanomaterials

15:10 to 15:30

Mirella Di Lorenzo (University of Bath, Claverton Down, Department of Chemical Engineering, University, Bath, United Kingdom)

Towards Low-cost, Simple and Biodegradable Microbial Fuel Cells for Energy Harvesting and Sensing Applications

15:30 to 15:50

Iwona Gajda (Bristol BioEnergy Centre, University of the West of England, Bristol, United Kingdom), John Greenman, Ioannis Ieropoulos, Oluwatosin Obata, Maria-Jose Salar-Garcia

Long Term Bio-Power of Microbial Fuel Cells in Individual and Stacked Configurations

S3 Sensing and Biointerfaces

Room: KBG-12

Chaired by: C. Cristea and T. Ruzgas

14:00 to 14:30 Keynote

Bernd Giese (Department of Chemistry, University of Fribourg, Fribourg, Switzerland)

Ag+ as electron scavenger: EET rates in Geobacter sulfurreducens

14:30 to 14:50

Anca Aldea (National Institute of Materials Physics, Atomistilor 405A, Magurele, Romania), Victor C. Diculescu, Elena Matei

Flexible Ionophore-Based Sensors for Continuous Monitoring of Electrolytes

14:50 to 15:10

Matteo Tucci (Department of Environmental Science and Policies, Milan, Italy), Pierangela Cristiani, Matteo Grattieri, Shelley D. Minteer, Andrea Schievano

Bioelectrochemical herbicide sensor based on photocurrent inhibition of Anabaena variabilis

15:10 to 15:30

Haesik Yang (Department of Chemistry, Pusan National University, Busan, Korea), Jungwook Kwon

Rapid and Sensitive Detection of Aspergillus niger Using a Single-Mediator System Combined with Redox Cycling

15:30 to 15:50

Kelsey Cremin (OSS Lab/ Warwick Electrochemistry and Interfaces Group, University of Warwick, Coventry, United Kingdom), Bryn Jones, Gabriel Meloni, Orkun Soyer, Patrick Unwin, Christian Zerfass

Scanning Ion Conductance Microscopy (SICM) for Determining the Local Charge Environment of Gram-positive and Gram-negative Bacteria

15:50 to 16:10

Coffee Break

16:10 to 16:30

Cecilia Cristea (Department of Analytical Chemistry, Faculty of Pharmacy, Iuliu Hatieganu University of Medicine and Pharmacy, Cluj-Napoca, Romania), Florin Graur, Gheorghe Melinte, Mihaela Tertis

Detection of Interleukin 6 from Human Serum by Label Free Electrochemical Sensors

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Emmanuel Iwuoha (University of Western Cape Sensor Laboratories (SensorLab), Robert Sobukwe Road, Bellville, Cape Town, South Africa), Usisipho Feleni, Laura Pacoste

Phenotype-based Nanoamperometric Signalling of Breast Cancer Drug

16:50 to 17:10

Ankitha George (Chemical Engineering, Indian Institute of Technology Madras, Chennai, India), M. S. Amrutha, Ramanathan S, V.V.R. Sai, Sujatha Sunil

Development of electrochemical and optical biosensors for the detection of mosquito borne diseases

17:10 to 17:30

Nadica Ivosevic DeNardis (Division for Marine and Environmental Research, Rudjer Boskovic Institute, Zagreb, Croatia), Ruza Frkanec, Lucija Horvat, Galja Pletikapic, P.Thomas Vernier

Design and characterization of marine-based plasma membrane vesicles

17:30 to 17:50

Karolien De Wael (AXES Research Group, Department of Chemistry, University of Antwerp, Antwerp, Belgium)

Bio-inspired laser-induced electrochemical sensing strategies: antigen/nanobody based magnetosensing

S4 New Materials/Materials Chemistry for Bioelectrochemistry

Room: S2-006

Chaired by: A. de Poulpiquet and A. Brett

14:30 to 14:50

Bo Zhou (School of Engineering and Materials Science, Queen Mary University of London, London, United Kingdom), Qian Guo, Steffi Krause, Ana Sobrido

Photoelectrochemical imaging system using α-Fe_2O_3 nanorods as substrate

14:50 to 15:10

Huaiguang Li (Center for Electrochemical Sciences, Bochum, Germany), Darren Buesen, Erik Freier, Thomas Happe, Joerg Henig, Wolfgang Lubitz, Kallol Mukherjee, Stefanie Stapf, Rhodri Williams, Martin Winkler

Bioelectrode Engineering - Control of Catalytic Film Thickness for Enzymatic Fuel Cells

15:10 to 15:30

Tobias Vöpel (Center or Electrochemical Sciences, Ruhr University Bochum, Bochum, Germany), Herrmann Christian, Adam David, Buesen David, Müller Frank, Kopiec Gabriel, Marquitan Miriam, Plumeré Nicolas, Happe Thomas, Martin Winkler, Kaiser Wolfgang

Site Specific Avidity Binding Tags based on Intrachain Histidine Motifs

15:30 to 15:50

Cristina Carucci (ISM UMR CNRS 5255, Univ. Bordeaux, Bordeaux, France), Sabrina Bichon, Alexander Kuhn, Nicolas Mano, Lin Zhang

Porous electrodes for miniaturized biofuel cells

15:50 to 16:10

Coffee Break

16:10 to 16:30

Agnieszka Wieckowska (University of Warsaw, Pasteura 1, Warsaw, Poland), Renata Bilewicz, Maciej Dzwonek, Michal Kizling

Redox modified gold clusters for ET enhancement

16:30 to 16:50

Maciej Dzwonek (Faculty of Chemistry, University of Warsaw, Warsaw, Poland), Renata Bilewicz, Michal Kizling, Agnieszka Wieckowska

Size dependent catalysis of phosphine-modified gold nanoparticles

16:50 to 17:10

Victor Diculescu (National Institute of Materials Physics, Magurele, Romania)

Magnetic Electrodes and Nanoparticles for Direct Immobilization of Biomolecules through Magnetic Forces

17:10 to 17:30

Ewa Nazaruk (Faculty of Chemistry, University of Warsaw, Warsaw, Krakowskie Przedmiescie 26/28, Poland)

Controlling The Drug Release From Mesophase – Modifying Release Rates from Lipidic Cubic Phase Carriers by Modulating Electrostatic Interactions or Phase Transitions

Wednesday 29 May 2019-Morning

Plenary

Room: KBG-12

Chaired by: R. Bilewicz

09:00 to 09:45 Keynote

P. Thomas Vernier (Frank Reidy Research Center for Bioelectrics, Old Dominion University, Norfolk, Virginia, USA)

Transport Through Electrically Stressed Cell Membranes — Introduction to the Electropermeome

S2 Electroporation and Electrophysiology

Room: S2-005

Chaired by: E. Dempsey and J. Lowry

09:50 to 10:10 Invited

Jacek Lipkowski (Department of Chemistry, University of Guelph, Guelph, Canada), Fatemeh Abbasi, Julia Alvarez-Malmagro, J.Jay Leitch, Piotr Pieta, Zhangfei Su

The effect of amiloride an ion channel blocker on pore formation by alamethicin in negatively charged and zwitterionic bilayers - a molecular view

10:10 to 10:30

Peter Kramar (Laboratory for biocybernetics, University of Ljubljana, Faculty of Electrical Engineering, Ljubljana, Slovenia), Günter Allmaier, Katja Balantic, Federica Castellani, Damijan Miklavcic, P. Tom Vernier, Victor U. Weiss

Effect of calcium ions on the cell membrane models

10:30 to 10:50

Ann-Sofie Cans (Department of Chemistry and Chemical Engineering, Gothenburg, Sweden), Jenny Bergman, Jacqueline Keighron, Devesh Mishra, Karolina Skibicka, Yuanmo Wang

Ultra-fast glutamate biosensor recording in rodent brain slice reveal complex single exocytosis transients

10:50 to 11:10

Coffee Break

11:10 to 11:40 Keynote

Michal Cifra (Institute of Photonics and Electronics, Czech Academy of Sciences, Prague, Czech Republic)

How is Electromagnetic Field Generated in and Interacts with Organisms at Molecular level?

11:40 to 12:00

Stephane Marinesco (Lyon Neuroscience research center, University of Lyon, Lyon, France), Charles Chatard, Anne Meiller, Andrei Sabac

In Vivo Brain Monitoring Using Minimally-invasive microelectrode biosensors Based on Platinized Carbon Fibers

12:00 to 12:20

Ilaria Palchetti (Dipartimento di Chimica, Università di Firenze, Sesto Fiorentino (Fi), Italy)

Gold nanoparticle decorated graphene nanoflakes for electrochemical biosensing of clinically significant molecules

12:20 to 12:40

Renata Bilewicz (Faculty of Chemistry, University of Warsaw, Warsaw, Poland), Ewa Nazaruk, Martina Zatloukalova

Electrogenic Transport of Na^+/K^+-ATPase Incorporated in Lipidic Cubic Phase Films as Biomimetic Membranes

S3 Sensing and Biointerfaces

Room: KBG-12

Chaired by: M. Scanlon and L. Gorton

09:50 to 10:10

Julia Alvarez-Malmagro (Faculty of Chemistry, Pasteura 1, Warsaw, Poland), Renata Bilewicz, Dorota Matyszewska, Ewa Nazaruk

Incorporation of Phytantriol Cubosomes into Gold (111) Supported DMPC Biomimetic Bilayer: a Spectroelectrochemical Study

10:10 to 10:30

Damian Dziubak (Faculty of Chemistry, Warsaw University, Warsaw, Poland), Jan Pawlowski, Slawomir Sek

Thiolated chitosan layers adsorbed on gold surface as a system for deposition of lipid bilayer

10:30 to 10:50

Katarzyna Krukiewicz (Centre for Research in Medical Devices, National University of Ireland, Galway, Galway, Ireland), Manus Biggs, Dawid Janas, Catalina Vallejo-Giraldo

Flexible neural electrodes based on self-supporting carbon nanotube films

10:50 to 11:10

Coffee Break

11:10 to 11:40 Keynote

Aihua Liu (Institute for Biosensing, and College of Life Sciences, Qingdao University, Qingdao, China), Ren Han, Chongyang Liu, Ge Wang, Lu Yan

Phage Display Based Biosensing and Nanomedicine

11:40 to 12:00

Somjai Teanphonkrang (Science, Suranaree University of Technology, Nakhon Ratchasima, Thailand), Pimchai Chaiyen, Salome Janke, Panida Khunkaewla, Adrian Ruff, Wolfgang Schuhmann, Jeerus Sucharitakul, Wipa Suginta

Enzyme Biosensing of a Urinary Biomarker with Redox Polymer-Based Readout and Allosteric Activation

12:00 to 12:20

Britta Lindholm-Sethson (Dept. of Chemistry, Umeå University, Umeå, Sweden), Yaser Khani, Solomon Tesfalidet

Investigation of Ionic interactions at a lipid cubic phase with Electrochemical Impedance Spectroscopy

12:20 to 12:40

Eithne Dempsey (Department of Chemistry, Maynooth, Maynooth, Ireland)

Scanning electrochemical microscopy imaging of poly (3,4-ethylendioxythiophene)/thionine electrodes for lactate detection via NADH electrocatalysis

S5 Protein Electrochemistry and Protein Engineering

Room: S2-006

Chaired by: R. Ludwig and E. Lojou

09:50 to 10:10

Martin Jönsson-Niedziólka (Institute of Physical Chemistry, Polish Academy of Sciences, Warsaw, Poland), Paulina Jelen, Magdalena Kundys-Siedlecka, Emilia Witkowska Nery

Enzyme Inhibition in Continuous Flow Measurements in a Microfluidic System

10:10 to 10:30

Lucyano Macedo (São Carlos Institute of Chemistry, University of São Paulo, São Carlos, Brazil), Frank Crespilho, Ayaz Hassan, Graziela Sedenho

Operando X-Ray Absorption Spectroscopy Applied to Bioelectrochemistry

10:30 to 10:50

Elisabeth Lojou (BIP-CNRS-Aix Marseille University, Marseille, France), Romain Clément, Vivek Hitaishi, Marianne Ilbert

Interplay between Orientation at Electrodes and Copper Activation of Thermus thermophilus Laccase for O_2 Reduction

10:50 to 11:10 Coffee Break

11:10 to 11:40 Keynote

Lital Alfonta (Life Sciences and Chemistry, Beer-Sheva, Israel), Dror Aizik, Itay Algov, Mor Fridman, Eden Ozer

Genetically Encoding for Improved "Wiring" in Enzymes and in Microorganisms

11:40 to 12:00

Justin Weeks (Department of Chemistry, University of Oxford, Oxford, United Kingdom), Holly Reeve, Kylie Vincent

Electroenzymatic NADH Recycling for Selective In-Flow Biocatalytic Reductions

12:00 to 12:20

Jing Tang (Department of Chemistry, Kongens Lyngby, Denmark), Wei Huang, Silke Leimkuehler, Loredana Preda, Jens Ulstrup, Rebecka Werchmeister, Ulla Wollenberger, Xinxin Xiao, Jingdong Zhang, Zhiyong Zheng

Graphene-Sulfite Oxidase Bioanodes for Enzymatic Biofuel Cells

12:20 to 12:40

Ulla Wollenberger (Institute for Biochemistry and Biology, Potsdam University, Golm, Germany), Chantal Iobbi-Nivol, Paul Kaufmann, Silke Leimkühler, Biljana Mitrova, Armel T.F. Waffo

Biosensor for trimethylamine –N-oxide with an chimeric enzyme

Thursday 30 May 2019-Morning

Plenary

Room: KBG-12

Chaired by: L. Gorton

09:00 to 09:45

J. Emnéus (Technical University of Denmark, Lyngby, Denmark), A. Asifa, A-I. Buneaa, A Heiskanena, S. M. Kahna, J. Kajteza, S.S. Kellera, N.B. Larsena, A. Martinez-Serranob, T. Ramos Morenoc

D and 3D Bioelectrochemical Lab-on-a-Chip Systems For Environmental and Life Science Applications

S3 Sensing and Biointerfaces

Room: KBG-12

Chaired by: F. Lisdat and D. Leech

09:50 to 10:10

Karolina Schwarzova-Peckova (Department of Analytical Chemistry, Charles University, Faculty of Science, Prague, Czech Republic), Lenka Benešová, Kristýna Jelšíková, Jan Klouda, Karel Nesmerák, Polina Yershova, Michal Zelenský, Adéla Zárybnická

Towards Detection of Steroids: Electrooxidation on Bare Electrode Materials After Acid-Induced Dehydration

10:10 to 10:30

Rebeca Miranda-Castro (Universidad de Oviedo, c/ Julián Clavería 8, Oviedo, Spain), María Jesús Lobo-Castañón, Raquel Sánchez-Salcedo, Noemí de-los-Santos-Álvarez

Monitoring changes in the expression profile of long noncoding RNAs associated with prostatic cancer by using an electrochemical biosensing platform

10:30 to 10:50

Alexandra Verhaven (Faculté des Sciences, Université Libre de Bruxelles, Bruxelles, Belgium), Dan Bizzotto, Thomas Doneux

Study of electrochemical melting/hybridization of DNA self-assembled monolayers by Fluorescence Microscopy

10:50 to 11:10 Coffee Break

11:10 to 11:40 Keynote

Robert E. Gyurcsanyi (Department of Inorganic and Analytical Chemistry, Budapest University of Technology and Economic, Budapest, Hungary), Gyula Jagerszki, Istvan Makra, Soma Papp

Selective potentiometric sensing of ions and polyions by solid-state nanopore-based ion channels

11:40 to 12:00

Damion Corrigan (Department of Biomedical Engineering, University of Strathclyde, Glasgow, United Kingdom)

Fabrication and development of microelectrodes for label-free biosensing

12:00 to 12:20

Jan Klouda (Department of Analytical Chemistry, Faculty of Science, Charles University, Prague, Czech Republic), Jirí Barek, Karolina Schwarzova, Adela Zarybnicka

A Novel Approach to 7-dehydrocholesterol Determination in Smith-Lemli-Opitz syndrome Diagnosis

12:20 to 12:40

Stanislav Trashin (Chemistry, University of Antwerp, Antwerp, Belgium), Karolien De Wael, Francisco Morales-Yánez, Serge Muyldermans, Katja Polman, Saranya Thiruvottriyur Shanmugam

Enzymatic and Photocatalytic Amplification in Amperometric Immunoassays with Nanobodies

S5 Protein Electrochemistry and Protein Engineering

Room: S2-006

Chaired by: U. Wollenberger and M. Jönson-Niedziolka

09:50 to 10:10

Marta Meneghello (Institut de Microbiologie de la Méditerranée, CNRS, Marseille, France), Martino Benvenuti, Sébastien Dementin, Vincent Fourmond, Chloé Guendon, Christophe Léger

Exploration of the Biodiversity of CO Dehydrogenases using Protein Film Electrochemistry

10:10 to 10:30 Invited

Roland Ludwig (Biocatalysis and Biosensing Laboratory, Vienna, Austria), Alfons Felice, Lo Gorton, Roman Kittl, Su Ma, Stefan Scheiblbrandner, Christopher Schulz, Christoph Sygmund

Basics to Utilise Direct Electron Transfer Enzymes in Carbohydrate Biosensors

10:30 to 10:50

Gilbert Nöll (Chem.-Biol.-Dept., Organic Chemistry, Siegen University, Siegen, Germany)

Spectroelectrochemical Investigation of Cholesterol Oxidase from Streptomyces Lividans at Different pH

10:50 to 11:10 Coffee Break

11:10 to 11:40 Keynote

Pau Gorostiza (Catalan Institution for Research and Advances Studies (ICREA, Institute for Bioengineering of Catalonia, Barcelona, Spain)

Electrochemical tunneling spectroscopy to study charge transport in redox proteins

11:40 to 12:00

Barbara Jachimska (Jerzy Haber Institute of Catalysis and Surface Chemistry, Polish Academy of Sciences, Krakow, Poland), Sylwia Swiatek

Lactoglobulin as a platform for designing biologically active carriers

12:00 to 12:20

Djamel Eddine Chafai (Institute of Photonics and Electronics, Czech Academy of Sci, Chaberská 57, Praha 8 - Kobylisy, Czech Republic), Michal Cifra, Pavel Draber, Vadym Sulimenko

Alteration of Tubulin Binding and Microtubules Formation by Nanosecond Pulsed Electric Field

Poster Presentations

S1 Biopower and Bioenergetics (Microbial Electrochemical Cells, Enzymatic Fuel Cells, Photobioelectrochemistry)

s1-001

Myriam Ghodhbane (Univ. Grenoble Alpes, CNRS, Grenoble INP, LGP2, Institute of Engineering Univ. Grenoble Alpes, Grenoble, France), Mohamed Naceur Belgacem, Didier Chaussy, Lionel Du bois, Gauthier Menassol, Abdelkader Zebda

Optimization of the fabrication of an Abiotic Bioelectrode for Implantable Biofuel Cell

s1-003

Guillermo A. Huerta-Miranda (Institute of Renewable Energies, Autonomus University of Mexico, Temixco, Mexico), Margarita Miranda-Hernández

Electrochemical characterization of Geobacter sulfurreducens under fumarate-limiting condition, comparison between biofilm and chemical immobilization

s1-004

László Koók (RI on Bioengineering, Membrane Technology and Energetics, Veszprém, Hungary), Péter Bakonyi, Katalin Bélafi-Bakó, Nándor Nemestóthy, Gábor Tóth

The Interrelation Between Ion-Exchange Membranes and the Efficiency of Microbial Electrochemical Systems

s1-005

Soo Youn Lee (Gwangju Bio/Energy Research Center, Korea Institute of Energy Research, Gwangju, Korea), Taek Dong Chung, Hana Nur Fitriana, Sangmin Lee, Jin-Suk Lee, Sung Yul Lim

Bioelectrochemical Systems for Selective Conversion of Carbon Dioxide to Value-Added Chemicals

s1-006

Jiye Lee (Gwangju Bio/Energy Research Center, Korea Institute of Energy Research, Gwangju, Korea), Sun-A Choi, Hana Nur Fitriana, Jin-Suk Lee, Soo Youn Lee, You-Kwan Oh

Enhanced Production of Lipids and High-Value Products from Microalgae by Electrochemical Treatment

s1-007

Aihua Liu (Institute for Biosensing, and College of Life Sciences, Qingdao University, Qingdao, China), Lu Bai, Shuqin Fan, Aihua Liu, Lu Yan

Microbial Surface Displaying Enzyme Based Efficient Biofuel Cells

s1-008

Ross D. Milton (Department of Civil and Environmental Engineering) Stanford University, Stanford, USA), Jörg Deutzmann, Jürgen Koch, Jack Ruth, Seigo Shima, Alfred M. Spormann

Hydrogen and/or Formate Facilitate Flavin-Based Electron Bifurcation in Methanococcus maripaludis

s1-009

Mario Mitov (Chemistry, , Blagoevgrad, Bulgaria), Ivo Bardarov, Yolina Hubenova

Photo-induced Phenomena in Sediment Microbial Fuel Cells with Different Architecture

S2 Electroporation and Electrophysiology

s2-001

Kobi P. Bermingham (Maynooth University, Neurochemical Research Group, Maynooth University, Maynooth, Ireland), Fiachra B. Bolger, Michelle M. Doran, John P. Lowry

Characterisation of a Biosensor for the Real-Time Neurochemical Monitoring of L-Glutamate

s2-002

Djamel Eddine Chafai (Institute of Photonics and Electronics, Czech Academy of Sci, Chaberská 57, Praha 8 - Kobylisy, Czech Republic), Damijan Miklavcic

Detrended Fluctuation Analysis of Electroporation Current Signal from Cell Suspension and Tissue

s2-003

Michelle M. Doran (Department of Chemistry, Maynooth Univeristy, Maynooth, Ireland), Kobi P. Bermingham, John P. Lowry, Kenneth W. Pierce, Mark D. Tricklebank

The in-vivo Validation of a D-Serine Biosensor

s2-004

Tjaša Potocnik (Faculty of Electrical Engineering, University of Ljubljana, Ljubljana, Slovenia), Alenka Macek Lebar, Damijan Miklavcic

Decreased effectiveness of gene electrotransfer in acidic pH in vitro

S3 Sensing and Biointerfaces

s3-001

Indre Aleknaviciene (Vilnius University, GMC, Institute of Biochemistry, Sauletekio al. 7, Vilnius, Lithuania), Gintaras Valincius

EIS and SERS Analysis of Anchor Molecules for Tethered Bilayer Lipid Membrane Formation

s3-002

Simona Baluchová (Department of Analytical Chemistry, Faculty of Science, Charles University, Albertov 6, Prague, Czech Republic), Vincent Mortet, Karolina Schwarzová, Andrew Taylor

Dopamine Sensing by Boron-Doped Diamond Electrode Fabricated by Microwave Plasma Enhanced Chemical Vapour Deposition Process With Linear Antenna Delivery

s3-003

Lenka Benešová (Charles University, Faculty of Science, Albertov 6, Prague 2, Czech Republic), Jan Klouda, Karolina Schwarzová, Polina Yershova

Development of Non-enzymatic Electroanalytical Methods for Detection of Cholesterol

s3-004

William Cheuquepan (University Institute of Electrochemistry, Alicante, Spain), Juan M. Feliu, José Manuel Orts, Antonio Rodes

Spectroelectrochemical study of the adsorption and reactivity of 5-fluorouracil on Au and Ag electrodes.

s3-005

Alex da Silva Lima (Department of Fundamental Chemistry, São Paulo, Brazil), Mauro Bertotti, Flavia Carla Meotti, Litiele Cezar da Cruz, Carla Santana Santos

An Electrochemical Study of Urate Hydroperoxide Using Carbon Microelectrodes

s3-006

João Carlos de Souza (Department of Analytical Chemistry / São Paulo State University, São Paulo State University (UNESP) / Institute of Chemistry, Araraquara, Brazil), Maria Valnice Boldrin Zanoni, Ana Maria Oliveira-Brett

Electrochemical Oxidation Mechanism of the Hair Dye Reagent P-Toluenediamine at a Glassy Carbon Electrode

s3-007

Adrian Enache (National Institute of Materials Physics, Magurele, Romania), Victor Constantin Diculescu, Catarina Sofia Henriques de Jesus

Redox Mechanisms of Proteasome Inhibitors

s3-008

Bogdan Feier (Analytical Chemstry, Iuliu Hatieganu University of Medicine and Pharmacy, Cluj-Napoca, Romania), Adrian Blidar, Cecilia Cristea

Biological Samples Treatment for Electrochemical Analysis of Vancomycin

s3-009

Bogdan Feier (Analytical Chemstry, Iuliu Hatieganu University of Medicine and Pharmacy, Cluj-Napoca, Romania), Adrian Blidar, Cecilia Cristea

Electrochemical Fingerprint of Vancomycin

s3-010

Monica Florescu (Faculty of Medicine, Transilvania University of Brasov, Brasov, Romania), Melinda David, Adrian Serban

Rapid Screening of Levo-Thyroxine Using Nanoparticle-Based Sensor

s3-011

Július Gajdár (Department of Analytical Chemistry, Faculty of Science, Charles University, Albertov 6, Prague, Czech Republic), Zuzana Bábková, Jirí Barek, Marie Brázdová, Jan Fischer, Miroslav Fojta, Tomáš Gonec, Josef Jampílek, Konstantina Tsami

Relation Between Biological Activity, Structure and Voltammetric Behavior of Novel Antimycobacterial Agents

s3-012

Fernando Garrido Diaz (Nanotechnology, Cork City, Ireland), Alan O' Riordan

Towards a Point-of-Use Nanosensor System for on-Farm Disease Detection

s3-013

Carla Gonzalez-Solino (Department of Chemical Engineering, University of Bath, Bath, United Kingdom), Mirella Di Lorenzo, Pawan Jolly

Engineering Functional Enzyme Bioelectrodes with Conductive Polymeric Films for Sensing Applications

s3-014

Oana Hosu (Department of Analytical Chemistry, "Iuliu Hatieganu" UMP, Cluj-Napoca, Romania), Alina Adumitrachioaie, Cecilia Cristea, Mariagrazia Lettieri, Giovanna Marrazza, Robert Sandulescu

[beta]-Lactoglobulin Allergen Detection Based on an Electrochemical DNA-Assay

s3-015

Oana Hosu (Department of Analytical Chemistry, "Iuliu Hatieganu" UMP, Cluj-Napoca, Romania), Cecilia Cristea, Gheorghe Melinte, Robert Sandulescu

Aptamer-based Electrochemical Ferrocene-Labeled Biosensor for Tetracyclines Detection

s3-016

Marta Jarczewska (The Chair of Medical Biotechnology, Faculty of Chemistry, Warsaw University of Technology, Warsaw, Poland), Malgorzata Gagala, Elzbieta Malinowska, Anita Trojan

Studies on the affinity-based biosensors for electrochemical detection of HER2 protein

s3-017

Joanna Juhaniewicz-Debinska (Faculty of Chemistry, Univeristy of Warsaw, Warsaw, Poland), Slawomir Sek, Dagmara Tymecka

Mechanism of lipopeptide action on model lipid membranes

s3-018

Wesley B. S. Machini (Department of Chemistry, Faculty of Science and Technology, University of Coimbra, Coimbra, Portugal), Ana Maria Oliveira-Brett

In Situ Evaluation of DNA-Antidiabetic Drug Metformin Interaction Using a dsDNA-Electrochemical Biosensor

s3-019

Tanushree Mandal (School of Chemistry, National University of Ireland Galway, Galway, Ireland), Dónal Leech, Conan Mercer, James F. Rusling

Automated Microfluidics for Electrochemical Immunoassays for Prostate Cancer Staging and Grading

s3-020

Dorota Matyszewska (Faculty of Chemistry, University of Warsaw, Warsaw, Poland), Renata Bilewicz

Phospholipid layers as simple models to study the interactions with anticancer and cholesterol-lowering drugs.

s3-021

Anne Meiller (Lyon Neuroscience Research Center, University of Lyon, Lyon, France), Stephane Marinesco

Fluorinated Xerogel-Coated Carbon Fiber Microelectrodes for Brain Nitric Oxide Monitoring

s3-022

Fionn O'Maolmhuaidh (National Centre for Sensor Research, Dublin City University, Dublin, Ireland), Loanda Cumba, Robert Forster

Biocompatible Screen-Printed Electrodes: Cell Viability Investigations

s3-023

Michael D. Scanlon (Bernal Institute, Department of Chemical Sciences, University of Limerick, Limerick, Ireland), Alonso Gamero-Quijano, Grégoire Herzog

Bioelectrochemistry of Cytochrome c in a Closed Bipolar Electrochemical Cell with Immiscible Aqueous-Organic Electrolyte Solutions

s3-024

Stefan Scheiblbrandner (BOKU - University of Natural Resources and Life Sciences, Department of Food Science and Technology, Vienna, Austria), Roland Ludwig

Biosensor Development for SECM

s3-025

Christopher Schulz (DirectSens GmbH, Am Rosenbühel 38, Klosterneuburg, Austria), Alfons Felice, Roman Kittl, Claudia König, Roland Ludwig, Christoph Sygmund, Patrick Weinmann

The First Commercialized Third Generation Biosensor Series LactoSens® for the Detection of Lactose in Low Lactose Dairy

Samples

s3-026

Prabhakar Sidambaram (Applied Electrochemistry Group AEG, FOCAS Research Institute, Technological University Dublin, Dublin, Ireland), John Colleran

Micro- and Nanoelectrodes: Fabrication, Characterization and Application. The Detection of Silver and ROS/RNS in Biological Buffer

s3-027

Prabhakar Sidambaram (Applied Electrochemistry Group AEG, FOCAS Research Institute, Technological University Dublin, Dublin, Ireland), John Colleran

Single Cell Electrochemistry – Uptake of Bioinorganic Silver-based Drugs

s3-028

Stepánka Skalová (Department of Analytical Chemistry, Charles University, Faculty of Science, Hlavova 8, Prague 2, Czech Republic), Jirí Barek, Jan Fischer, Jan Krejcí, Tomáš Navrátil, Vlastimil Vyskocil

Voltammetric Detection of Anthraquinone-2-Sulfonate After its Separation by Means of a Microdialysis Catheter

s3-029

Mihaela Tertis (Analytical Chemistry Department, Faculty of Pharmacy, Iuliu Hatieganu University of Medicine and Pharmacy, Cluj-Napoca, Romania), Anida-Maria Babtan, Bianca Adina Bosca, Roxana Ioana Bordea, Cecilia Cristea, Claudia Nicoleta Feurdean, Aranka Ilea, Adela Cristina Lazar, Ioana-Codruta Mirica, Nausica Bianca Petrescu, Robert Sandulescu, Willi Uriciuc

Fast and Sensitive Detection of Methylglyoxal as an Important Advanced Glycation End Product in Oral Pathology

s3-030

Solomon Tesfalidet (Department of Chemistry, Umeå University, Umeå, Sweden), Paul Geladi, Britta Lindholm-Sethson, Sereilakhena Phal

Electrochemical Immitance Spectroscopy and Multivariate Data analysis for Analysis of a Therapeutic Drug - Methotrexate

s3-031

Sofiia Tvorynska (Department of Analytical Chemistry, Charles University, Prague, Czech Republic), Jirí Barek, Bohdan Josypcuk

Amperometric Choline Biosensor with Enzymatic Reactor in Flow Systems: Fabricating, Optimization and Application

s3-032

Veronika Urbanova (Palacky University - RCPTM, 17.listopadu 1192/12, Olomouc, Czech Republic)

Graphene derivatives and their application as platforms for aptasensors

S4 New Materials/Materials Chemistry for Bioelectrochemistry

s4-001

Mariola Brycht (Faculty of Science, Department of Analytical Chemistry, Charles University, Prague, Czech Republic), Ivan Dittert, Jan Krušek, Vincent Mortet, Karolina Schwarzová, Andrew Taylor, Jana Vosáhlová

Bio-Sensing of Dopamine and Its Precursors Using Boron-Doped Diamond Microelectrodes

s4-002

Andreea Cernat (Analytical Chemistry Department, Faculty of Pharmacy, Iuliu Hatieganu University of Medicine and Pharmacy, Cluj-Napoca, Romania), Andreea Bodoki, Cecilia Cristea, Maria-Bianca Irimes, Ioana-Ecaterina Pralea, Maria Suciu, János Györfi Szabolcs, Mihaela Tertis

Click Chemistry on Azide Functionalized Graphene Oxide

s4-003

William Cheuquepan (University Institute of Electrochemistry, , Alicante, Spain), Juan M. Feliu, Adolfo Ferre-Vilaplana, José M. Gisbert-González, Enrique Herrero

Why citrates shapes both gold and platinum nanoparticles: A fundamental insight for biomedical purposes

s4-004

Loanda Cumba (National Centre for Sensor Research, Dublin, Ireland), Robert Forster

SmartInks: Screen-Printing Platforms for the Development of Label-Free DNA/mRNA Assay

s4-005

Ales Danhel (Biophysical Chemistry and Molecular Oncology, Institute of Biophysics, Cezch Academy of Sciences, Brno, Czech Republic), Pavlina Havranova, Lukas Kejik, Filip Ligmajer, Peter Sebest

Spectroscopic Qualities of Silver Amalgam Particles

s4-006

Ana B. Delgado-Egea (Instituto Universitario de materiales, Universidad de Alicante, Alicante, Spain), Francisco Huerta, Francisco Montilla, Emilia Morallón, César Quijada

Thiophene-Based Conducting Polymers for Direct Electrochemistry of Cytochrome c

s4-007

Eithne Dempsey (Department of Chemistry, Maynooth, Maynooth, Ireland)

β-methylumbelliferone Surface Modification and Permeability Investigations at PENTEL™ Graphite Electrodes

s4-008

Pavlina Havranova (Institute of Biophysics of the CAS, v. v. i., Královopolská 135, Brno, Czech Republic), Ales Danhel, Filip Ligmajer

Electrodeposition of Silver Amalgam on Golden Plate Electrodes Using Double Pulse Chronoamperometry

s4-009

Jesus Iniesta (Physico-Chemical department and institute of Electrochemistr, Alicante, Spain), O.V. Alekhina, L. García-Cruz, A. Gerasimova, A.V. Melezhik, V. Montiel, A.G. Tkachev

A glance at the electroanalytical response of carbon materials based films doped with aminocumulene acting as dispersant and N-containing precursor

s4-010

Elzbieta Jablonowska (Faculty of Chemistry, University of Warsaw, Warsaw, Poland), Renata Bilewicz, Ewa Nazaruk

Incorporation of lipid liquid crystalline drug carriers containing doxorubicin into lipid model membranes

s4-011

Katarzyna Krukiewicz (Centre for Research in Medical Devices, National University of Ireland, Galway, Galway, Ireland), Beata Cwalina, Dominika Czerwinska-Glowka, Mieczyslaw Lapkowski, Wioletta Przystas, Malgorzata Skorupa, Sebastian Student, Daria Wieclawska, Ewa Zablocka-Godlewska

Electro-responsive nanocomposites as smart antibacterial surfaces

s4-012

Agata Krzak (Faculty of Chemistry, University of Warsaw, , Warsaw, Poland), Renata Bilewicz, Maciej Majdecki, Ewa Nazaruk

Cyclodextrin as a Carrier for Anticancer Drug-Temozolomide

s4-013

Ricardo Leote (National institute of Materials Physics, University of Bucharest, Magurele, Romania), Victor Diculescu, Monica Enculescu, Cristian M. Teodorescu

Nanostructured Samarium Oxide Electrodes for Biosensing Applications

s4-014

Ana Maria Oliveira-Brett (Department of Chemistry, University of Coimbra, Coimbra, Portugal), Isabel P.G. Fernandes

Lercanidipine Electrochemical Behaviour at Carbon Black Modified Electrodes

s4-015

Peter Sebest (Institute of Biophysics of the CAS, v. v. i., Department of Biophysical Chemistry and Molecular Oncology, Brno, Czech Republic), Ales Danhel, Veronika Ostatna

The Applicability of Pyrolytic Graphite Electrode Decorated by Silver Amalgam Particles in Bioanalysis

s4-016

Krzysztof Stolarczyk (Faculty of Chemistry, University of Warsaw, Warsaw, Poland), Marek Kubiszewski, Marta Laszcz, Andrzej Les, Katarzyna Sidoryk, Elzbieta U. Stolarczyk

Abiraterone Conjugated with Gold Nanoparticles for Anticancer Drug Delivery Application and Therapeutic Performance

s4-017

Jakub Vezník (Department of Chemistry, Faculty of Science and CEITEC, Masaryk University, Brno, Czech Republic), Karel Lacina, Libuše Trnková

Interplay of redox potential and pKa in aminoferrocene

s4-018

Jana Vosahlova (Faculty of Science, Charles University, Prague, Czech Republic)

Boron doped diamond microelectrodes: Fabrication, spectral and electrochemical characterization for estimation of coverage quality

S5 Protein Electrochemistry and Protein Engineering

s5-001

Itay Algov (Life-Sciences, Ben-Gurion University of the Negev, Beer-Sheva, Israel), Lital Alfonta, Raz Zarivach

Highly Efficient Flavin–Adenine Dinucleotide Glucose Dehydrogenase Fused to a Minimal Cytochrome C Domain

s5-002

Fred Lisdat (Biosystems Technology, Technical University of Applied Sciences Wildau, Wildau, Germany), Marc Riedel

Integration of bilirubin oxidase in transparent, inverse opal ATO electrodes for efficient oxygen reduction

s5-003

Alaa Oughli (Molecular nanostructures, Ruhr Üniversität Bochum, Bochum, Germany), James Birrell, Wolfgang Lubitz, Nicolas Plumeré, Olaf Rüdiger, Wolfgang Schuhmann, Marisela Vélez

Viologen-modified electrodes for protection of hydrogenases from high potential inactivation

s5-004

Libuse Trnkova (Department of Chemistry, Faculty of Science, Masaryk University, Brno, Czech Republic), Jaromir Hubalek, Jan Slavik

Interaction of Selenite with Metallothionein

S6 Emerging Topics in Bioelectrochemistry

s6-001

Stephane Arbault (Institute of Molecular Sciences, CNRS UMR-5255, University of Bordeaux, Pessac, France), Camille Colin, Jérôme Launay, Gabriel Lemercier, Fadhila Sekli Belaïdi, Neso Sojic, Pierre Temple-Boyer, Venkata S.R. Vajrala, Dodzi Zigah

Microwell Array Integrating Ring Nanoelectrodes for the Monitoring of Bioenergetic Responses from Single Mitochondria

s6-002

Eadaoin Carthy (School of Chemical Sciences, Dublin City University, Glasnevin, Dublin, Ireland)

Electrochemical Impedance Spectroscopy (EIS) Pathogen Detection in a Microfluidic Electronic Lab-on-a-Disc (eLoaD) Platform

s6-003

Marcin Szymon Filipiak (Nanomaterial-Based Biosensors Team, BioMed X GmbH, Heidelberg, Germany), Oscar Gutierrez-Sanz, Martin Jönsson-Niedziólka, Alexey Tarasov, Kishan Thodkar, Daniel Vetter

Observation of direct electron transfer from glucose dehydrogenase to single sheet graphene electrode

s6-004

Lukáš Fojt (Department of biophysical chemistry and molecular oncology, Institute of Biophysics of the CAS, v. v. i., Brno, Czech Republic), Miroslav Fojta, Bohumír Grüner

Study of different icosahedral carboranes as a novel electrochemical label

s6-005

Valentina Grippo (Department of Medical Chemistry and Biochemistry, Palacky University, Olomouc, Czech Republic), Jan Vacek, Martina Zatloukalova

Nitro-fatty acids redox transformations: electrochemical and EPR study

s6-006

Joohee Jeon (Department of Chemistry, Seoul National University (SNU), Seoul, Korea), Eun Joong Kim, Sun-Heui Yoon

Neural Interface via Artificial Synapse between Live Neuron and Solid Electrode

s6-007

Nikolina Mitreska (Institute for Biophysics, School of Medicine, University of Saarland, Homburg, Germany), Ivan Bogeski, Rubin Gulaboski, Markus Hoth, Reinhard Kappl, Valentin Mirceski, Ewa Slowik, Katerina Stankoska

Hydrogen Bonding and Ca^{2+} Affinity of Coenzyme Q_1 and Decylubiquinone

s6-008

Katerina Stankoska (University of Saarland, School of Medicine, Institute for Biophysics, Homburg, Germany)

Calcium Affinity of Coenzyme Q10 and its Hydroxylated Derivative

s6-009

Mihaela Tertis (Analytical Chemistry Department, Faculty of Pharmacy, Iuliu Hatieganu University of Medicine and Pharmacy, Cluj-Napoca, Romania), Anida-Maria Babtan, Bianca Adina Bosca, Dan Buhatel, Bianca Ciui, Cecilia Cristea, Claudia Nicoleta Feurdean, Aranka Ilea, Anca Ionel, Nausica Bianca Petrescu, Robert Sandulescu, Arin Sava, Willi Uriciuc

Electrochemical Detection of N^{ϵ}-(carboxymethyl)lysine in Raw Human Saliva

s6-010

Martina Zatloukalova (Department of Medical Chemistry and Biochemistry, Palacky University, Olomouc, Czech Republic), Jeanne Crassous, Martin Jakubec, Jan Storch, Jan Vacek

Electrochemical and Spectral Characterization of Flavo[6]helicene and its Application in Bioanalysis and Materials Science

s6-011

Dimitrios Zouraris (Chemical Engineering, National Technical University of Athens, Athens, Greece), Maria Dimarogona, Antonis Karantonis, Anthi Karnaouri, Evangelos Topakas

Study of H_2O_2 kinetics of an immobilized Lytic Polysaccharide Monooxygenase with FTacV

Abstracts of Plenary, Keynote, Oral and Poster Presentations

B E S
The Bioelectrochemical Society

Award Lecture

From nanobiosensors to bioelectrocatalysis and photobioelectrocatalysis

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Measurement of physiological relevant molecules in and at individual cells with minimal disturbance of the cellular function is a key. Coupling of biocatalytic re-dox reactions with electrode surfaces is on the one hand the basis for the design of targeted biosensors and on the other hand the basis for harvesting energy using biomolecules as recognition elements and catalysts. This implies the choice of a suitable long-term stable active enzyme and, in particular, a stable immobiliza-tion/fixation of the biorecognition element on the electrochemical transducer. In this context, redox polymers present a well-suited immobilization matrix that en-sures a highly solvated environment for the biorecognition element.

The presentation is hence focusing on recent developments on:

1. Reproducible fabrication of carbon nanoelectrodes and design of nanometer-sized amperometric biosensors.
2. Double-barrel carbon nanoelectrodes for spearhead field effect transistors.
3. Self-powered biosensors and instrument-free substrate determination.
4. Biocatalyst protection using redox-polymer based protection shields.
5. Biophotovoltaic cells

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Introducing Robustness into Fragile Bioelectrochemical Systems

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The preconception on enzyme fragility often limits the ambition for the industrial applicability of bioelectrochemical systems. However, isolated enzymes are commonly used in white biotechnology. So why not aim for large scale bioelectrochemical systems such as biophotovoltaic cells, H2/O2 biofuel cells, biocapacitors, etc.? Evidently, it is unreasonable to imagine water splitting at industrial scale with isolated photosystem 2 or it was until recently not thinkable that highly O2 sensitive hydrogenases can be operated in the presence of O2. Nevertheless, the recent breakthrough with redox-active polymer films serving as protective matrices for preventing oxidative deactivation of hydrogenases (1, 2) may provide motivation to also use comparatively fragile enzymes and to develop protection strategies to exploit their potential for applications.

The protection mechanism for hydrogenases relies on the diversion of electrons from H2 oxidation within the matrix toward reduction of O2 at the electrolyte-film boundary (3). This promotes anaerobic conditions within the matrix to the extent that even thin films (4) can provide O2-immunity while achieving highly efficient catalyst utilization. Our empirical data were explained by modeling demonstrating that resistance to O2 inactivation can be obtained for non-limiting periods of time even when using the most fragile hydrogenases. We show that different protection mechanisms operate depending on matrix dimensions and intrinsic catalyst properties, and can be integrated together synergistically to achieve large and stable H2 oxidation currents in the presence of O2, potentially enabling a plethora of practical applications for biological catalysts in harsh oxidative conditions.

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Protein pores as nanoreactors for single-molecule covalent chemistry

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Covalent chemistry can be observed at the single-molecule level by using engineered protein pores as "nanoreactors". By recording the ionic current driven through single engineered alpha-hemolysin pores in a transmembrane potential, individual bond-making and bond-breaking steps are monitored with sub-millisecond time-resolution. Covalent chemistry observed by this approach has included multiple turnovers of reversible reactions, polymer chain elongation and complex reaction networks. The scope of the approach can be expanded by using unnatural amino acid mutagenesis to place a variety of reactive side chains within the lumen of the pore. Besides the intrinsic interest of single-molecule chemistry, the technique allows the detection of a variety of reactive analytes. Most recently, we have focused on using the nanoreactor approach to monitor small moving molecules, walkers and hoppers, in real time. This chemistry can be applied to the stepwise translocation of biopolymers and selective transmembrane signaling.

2D and 3D Bioelectrochemical Lab-on-a-Chip Systems For Environmental and Life Science Applications

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In the past few years we have focused on developing 3D lab-on-a-chip systems relevant for monitoring environmental and liver toxicity as well as systems relevant for mimicking neurodegenerative disorders such as Parkinson's disease (PD). The main focus has been on PD, which is a disease characterized by having insufficient dopamine in the brain, a neurotransmitter involved in the motor function of PD patients. One of the future treatments are believed to be cell replacement therapy, using stem cells. We have previously reported that pyrolysed 3D carbon micropillars promotes differentiation of human neural stem cells (hNSCs) into dopaminergic neurons and that these pillars simultaneously can function as sensors for detecting dopamine release from maturing neurons attached to them [1]. We hereby will demonstrate how 3D printing [2,3] and/or photolithography combined with pyrolysis into pyrolytic carbon [1] can be used to develop scaffolds, equipped with different properties for support and culturing of cells, that inherently have: (i) structured perfusable channel network, enabling delivery of necessary nutrients and oxygen to the interior of the scaffolds, (ii) secondary more arbitrary random porous network that can enclose a hydrogel phase with a "nearby" source of important cell factors, supporting the growth and differentiation of cells, (iii) ability to conduct or sense electrical currents, and/or finally (iv) having both optical and electrochemically properties, which are our newest results, involving development of what we call optoelectrical scaffolds potentially applicable as brain implants for treatment and alleviation of symptoms of PD patients.

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How Low Can We Go? Ultrasensitive Electrochemical Detection of Biomolecular Analytes

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To put disease-related biomarkers to work in the clinic, new high-performance technologies are needed to enable rapid and sensitive analysis of clinical specimens. Electrochemical methods providing low cost and direct biomarker readout have attracted a great deal of attention for this application. We exploit controlled nanostructuring of electrode surfaces to enhance biomolecular capture rates and efficiencies to solve this long-standing problem,[1] and showed that the nanoscale morphologies of electrode surfaces control their sensitivities. An electrocatalytic reporter system that leverages a pair of redox reagents is used to readout the presence of specific nucleic acids and other analytes bound to electrode surfaces. Recently, we have developed assays that are able to detect nucleic acids, proteins and small molecules,[2] with universally high sensitivity levels. This presentation will highlight how electrodeposited metals can be used to create high-performance sensors that can be applied to a broad collection of clinically-relevant analytes,[3-6] and will discuss how surface morphology, surface area and molecular diffusion can affect the detection limits obtained.

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Transport Through Electrically Stressed Cell Membranes — Introduction to the Electropermeome

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The "standard model" of electroporation describes material transport into cells through pores formed when the transmembrane voltage exceeds a critical value. Current implementations of the model evolved from the transient, stochastic pore model proposed over 30 years ago [1–5]. Despite improvements, serious shortcomings in the model remain, including the inability to predict key features of electropermeabilized systems: pore lifetimes; dynamics of membrane potential, electrical conductivity, and permeability to small molecules; multiple-pulse protocol outcomes; dependencies on cell size and type and medium conductivity.

A model that is predictive beyond the initiation of permeabilization must include *both* the immediate physics of lipid electropore formation *and* the subsequent physical and biological responses of the cell to the stress of membrane barrier disruption. Long-lived permeabilizing structures (electropermeasomes) must be identified from molecular models and experiments. Transport-related stress and damage responses must be represented: restoration of ion concentration gradients (Ca^{2+}, K^{+}, Na^{+}) and transmembrane potential, osmotic and volume regulation, membrane repair, and metabolic adjustments. Transport contributions from electromodulated membrane proteins and from quasi-stable lipid and lipid-protein associations resulting from perturbations of membrane structure must also be incorporated. All these electro-transport-relevant structures and processes taken together comprise the regime of the new model, the electropermeome.

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Genetically Encoding for Improved "Wiring" in Enzymes and in Microorganisms

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One of the research questions that we have found most fascinating in bioelectrochemistry is, what makes electrogenic bacteria, electrogenic? In order to tackle this question, we have commenced our studies by attempting to turn non-electrogenic bacteria to electrogenic bacteria. Using the tools of synthetic biology, synthetic chemistry and standard as well as new genetic engineering tools, we have replaced the pili of *pseudomonas aeruginosa* with pili of electrogenic bacteria using CRISPR/Cas 9 technology and studied the new bacteria properties. We have genetically expanded the genetic code of *pseudomonas aeruginosa* and modified its flagellin peptides using an unnatural amino acid (UAA) in order to site-specifically attach to it electro transferring enabling moieties (such as gold nanoparticles). We have expanded the genetic code of the photoautotrophic bacteria (cyanobacteria *Synechococcus sp.* PCC7942) in order to site-specifically wire one of its cytochromes to electrodes, and we engineered and incorporated UAAs into various redox active enzymes and proteins in order to improve and direct their electron transferring capabilities in an efficient way to electrodes. Using these novel technologies, we could successfully confirm some of the suggested mechanisms of electron transfer in such electron transferring microorganisms, furthermore we could successfully demonstrate that if specific molecular components are correctly identified as electron transferring components, transplantation of these proteins into non-electron transferring microorganisms, greatly enhances their electron transferring properties.

Electron transfer out of *Geobacter*: most models are wrong, are some useful?

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Geobacter representatives are the most commonly enriched bacteria on anodes in microbial electrochemical systems. These organisms possess a molecular mechanism capable of linking complete oxidation of substrates in their cytoplasm to electron transfer across an inner membrane, periplasm, outer membrane, and conductive space outside the cell. Early studies hypothesized that this process was due to a single pathway, consisting of a few key redox proteins or novel conductive structures lacking any redox cofactors. However, recent work shows that most early models were overly simplistic. Three different multiheme cytochrome quinone oxioreductases, operating at different redox potentials, are essential for electron transfer by *G. sulfurreducens*, while at least five periplasmic triheme *c*-type cytochromes appear to contribute to electron transfer across the periplasm. Five different 'porin-cytochrome' complexes are needed for electron transfer across the outer membrane, depending on the electron acceptor being used, with some of these complexes containing Fe-S clusters instead of cytochromes. Finally, at least four different multiheme cytochromes are essential for electron transfer beyond the cell to electrodes or metals, depending on conditions. Further altering our view of electron transfer in *Geobacter* is a surprising new structure for conductive filaments that does not involve pili. Together, these new data suggest a more complex model for extracellular electron transfer that changes depending on the redox potential, substrate, and environmental conditions experienced by the organism.

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Portable bioelectroanalysis systems for real-time patient and human tissue monitoring

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The possibility of real-time biochemical monitoring of both patients and vital tissue, such as transplant kidneys, is currently of great interest clinically (1). The chemical and biochemical signals obtained can provide the means to detect the onset of pathology in acutely ill patients and guide treatment in a personalised way, or determine the functional capacity of potential transplant organs ex vivo before use. To address these problems we have developed a portable, potentially wearable microfluidic based bioelectroanalysis systems designed to be used with the tissue sampling technique of microdialysis.

A sterile perfusable microdialysis probe (MD) is implanted into the tissue of interest. Slow pumping (0.5-1.0 μl min^{-1}) of a sterile ionic liquid through the MD probe extracts biomolecules from the tissue for ex vivo analysis. To make robust, portable devices robust we use 3D printed microfluidic systems with custom high performance electronics (2). We have analysed biomolecules related to tissue energy metabolism (glucose, lactate, pyruvate) tissue activation (glutamate, ionic changes) and tissue function (creatinine for kidneys). In each case the analysis is performed using a miniature needle-based integrated electrochemical cell. For biosensors working electrodes (50 μm) are coated with an electropolymerised selectivity layer, a hydrogel bioreconition enzyme layer and if required an outer layer to tune mass-transport into the biosensor. Increasingly we use microfluidic networks to allow homogeneous enzyme reaction with the dialysate stream as this can provide greater stability and sensitivity.

The presentation will describe the system design and give example from recent clinical monitoring.

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How is Electromagnetic Field Generated in and Interacts with Organisms at Molecular level?

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We share our current molecular level understanding of how electromagnetic field interacts with organisms and how organisms generate electromagnetic field. In the first part, we show that intense nanosecond pulsed electric field affects protein structure and function on the example of cytoskeletal protein tubulin both from molecular dynamics simulation and experimental data. We also demonstrate that molecular dynamics simulations enable prediction and interpretation of dielectric properties of biomolecular systems to rationalize design of high-frequency biochips. [1,2]

In the second part, we explain biophysical foundations of electromagnetic activity of cells[3] in two frequency bands: microwave and visible light range. Electromechanical fluctuations of cytoskeletal structures microtubules were proposed to be the underlying mechanism in microwave band[4], although it lacks convincing experimental evidence so far. In the spectral range of visible light, all organisms undergoing oxidative metabolism generate very weak chemiluminescence.[5] We show selected details of the molecular mechanisms leading to this weak, yet ubiquitous biological light, and how it can be harnessed for non-invasive and label-free monitoring of oxidative processes and stress in organisms.

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Operando, in situ and on-line techniques for real-time measurements in Bioelectrochemistry

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In this keynote lecture, several techniques for real-time measurements under bioelectrochemical control will be discussed. We shall show experimental setups that use differential electrochemical mass spectrometry (DEMS), electron paramagnetic resonance (EPR), Micro-FTIR (mid region), far infrared spectroscopy (FIR, below 500 cm^{-1}) and X-ray absorption spectroscopy (XAS), in order to provide on-line, in situ and *operando* information.[1-3] Real-time monitoring of the conversion of substrates using redox enzymes will be discussed for dehydrogenases and oxidases. On-line measurements were able to answer some important question related to the pathway of redox reactions, such as the calculation of kinetics parameters from metabolites generated in DEMS. In addition, redox chemistry of drugs as well as its dynamics inside living cells (e.g. lung carcinomas) could be studied by using Micro-FTIR. Other examples of application will be presented and discussed in detail.

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Ag^+ as electron scavenger: bioinorganic aspects

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Silver, in particular silver ions, are well known and studied for their antimicrobial properties.[1] Yet, the question arises whether bacteria can become resistant against silver, or at least tolerate higher silver concentrations. Indeed, two ways are known by which this seems possible.
Some Gram-negative bacteria possess a silver ion specific efflux pump in which a unique protein, SilE, can act as a silver ion sponge and transporter. Studying this protein and its interactions with silver ions gives insight on how silver ions bind to proteins reversibly without electron transfer occurring.[2]
On the other hand, anaerobic bacteria like *Geobacter sulfurreducens*, are able to cope with high levels of silver ions by reducing them to silver nanoparticles (AgNPs). AgNPs are less soluble and toxic, hence this might be a way to survive and inactivate antimicrobial silver ions. We are interested in how proteins can coordinate to silver ions, under which conditions silver ions can be reduced when bound to proteins and finally, how anaerobic *Geobacter sulfurreducens* is able to reduce silver ions to AgNPs.[3]
Hence, the binding of silver ions to proteins like SilE, to model peptides and to cytochromes will be presented. From the results, we learn about the role of some key amino acid residues in proteins that allow or not for electron transfer and hence silver ion reduction.

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Keynotes

Ag^+ as electron scavenger: EET rates in *Geobacter sulfurreducens*

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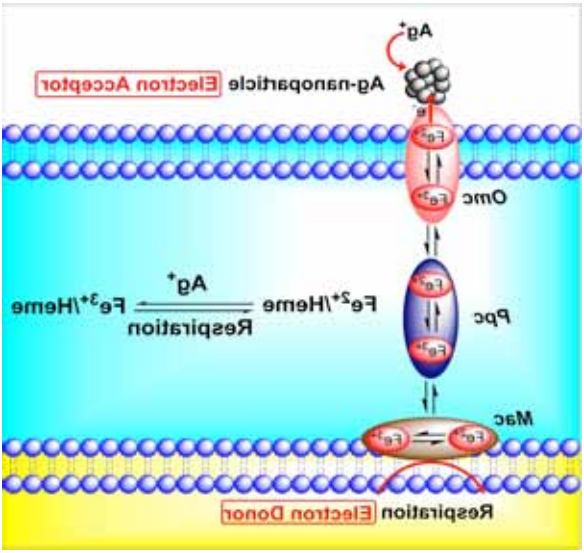
We have developed an assay, which allows the measurement of rates and activation parameters for extracellular electron transfer (EET) through *Geobacter sulfurreducens.* Silver nanoparticles (AgNPs), bound to the outer membrane cytochromes, establish the metal/bacteria contact. Outside Ag^+ ions are the external oxidants, and the iron-hemes of cytochromes act as electron shuttles.



ET rates were determined by the time dependence of the concentration changes, analyzed by ICP (decrease of Ag^+ ion concentration), light scattering (AgNP growth), and Q-bands (redox changes of the iron-hemes). TEM experiments quantified the metal/bacteria contacts, which indicate the number of the reaction pathways. Activation parameters were measured by the temperature effect on the EET rate. It will be discussed why the low EET rates and activation parameters via this cytochrome pathway requires the production of pili in living *Geobacter sulfurreducens.*

Electrochemical tunneling spectroscopy to study charge transport in redox proteins

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Electron Transfer (ET) plays essential roles in biological processes such as cell respiration and photosynthesis. It takes place between redox proteins and in protein complexes that display an outstanding efficiency and environmental adaptability. Although the biochemistry of ET processes is well characterised, nanoscale experimental methods are needed to understand electronic pathways in these redox protein structures, both for fundamental and for technological purposes. Electrochemical Scanning Tunneling Microscopy (ECSTM) is an excellent tool to study electronic materials and redox molecules including proteins. It offers single molecule resolution and allows working in aqueous solution, in nearly physiological conditions in the case of proteins, and under full electrochemical control. ECSTM also allows performing conductance measurements by current-potential and current-distance tunneling spectroscopy. An overview of these methods and current projects in the laboratory will be presented.

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Keynotes

Selective potentiometric sensing of ions and polyions by solid-state nanopore-based ion channels

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Apart from the overwhelming majority of nanopore sensors that are based on measuring the modulation of a transpore ionic current by the detected species, here we present nanopore-based sensing schemes that use potentiometric readout. Permselective, charged nanopore membranes were shown to generate potential response either for cations or anions depending on the charge sign of the nanopore surface.[1] This response was found to be described by the Nernst-Planck/Poisson model for water permeable charged pores, which approaches ideal Nernstian behavior as the pore diameter is reduced to the range of the Debye layer thickness.[2] While such permselective pores exhibit no selectivity among ions of the same charge sign we have shown that additional chemical modifications of the permselective nanopores can turn them into highly selective ion sensors. Two different potentiometric sensing strategies will be presented that are based on complex chemical modifications of the sensing zones of gold nanopores. The first approach inspired by the selectivity filter of biological ion channels involves the functionalization of gold nanopores of a few nm diameter with three different functional thiol and dithiolane derivatives, most importantly with an ionophore to induce selective complexation of the targeted ion (e.g., Ag^+, Cu^{2+}).[3,4] By simply monitoring the membrane potential of these functionalized nanoporous membranes highly selective ion sensors with ideal Nernstian response were obtained. The second approach is based on potentiometric detection of the charge inversion that occurs in the sensing zone of conically shaped gold nanopores upon binding of negatively charged microRNA strands to positively charged peptide-nucleic acid modified nanopores.[5] A quantitative theoretical treatment of the real-time potentiometric response and hybridization kinetics will be also provided.

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Biophotovoltaics: exoelectrogenic activity of photosynthetic microorganisms

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Photosynthetic microorganisms (both prokaryotes, such as cyanobacteria, and eukaryotes) typically show exoelectrogenic activity, that is to say they generate small amounts of extracellular current, which is stimulated by illumination. However, very little is known about what determines the magnitude of this exoelectrogenic activity. I shall discuss our present understanding of some of these factors, such as genotype and physiological status of the cells. Bioelectrochemical devices generating power in this way are referred to as 'biophotovoltaic' systems. I shall also discuss the feasibility of using biophotovoltaic systems to power small electrical devices.

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Keynotes

Direct Electron Transfer Performance of Protein Engineered D-Fructose Dehydrogenase

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D-Fructose dehydrogenase (FDH) from *Gluconobacter japonicus* NBRC3260 is a heterotrimeric membrane protein consisting of subunit I (67 kDa), II (50 kDa), and III (20 kDa). Subunit I contains a flavin adenine dinucleotide (FAD) as the catalytic center, while subunit II contains three heme *c* moieties (heme 1*c*, 2*c*, and 3*c* from N-terminus of subunit II). Subunit III plays a significant role in the expression of FDH. FDH catalyzes the oxidization of D-fructose and shows high DET-type bioelectrocatalytic activity. Subunit complex I/III does not show any DET-type catalytic activity, though it shows a catalytic activity in solution Our group was pointed out that the electron is transferred from the substrate-reduced FAD, to heme 3*c*, heme 2*c*, and an electrode, and that heme 1*c* was not involved in the catalytic electron transfer judging from the relation between the formal potentials of the hemes and the location of the catalytic wave and the effect of replacing the sixth axial methionine ligand (M301, M450, or M578) of one of the hemes the waves with glutamine.

An FDH variant which lacks 143 amino acid residues including the heme 1*c* binding site (Δ1*c*FDH) showed an increase in the catalytic current density. This was presumably due to the downsizing of the enzyme, which resulted in an increase of the surface concentration of effective Δ1*c*FDH.

An FDH variant (M450QΔ1*c*FDH) in which M450 in Δ1*c*FDH was replaced with glutamine to negatively shift the formal potential. The variant-adsorbed electrode produced a clear sigmoidal steady-state DET-type catalytic wave. The limiting current density was 1.4 times larger than that of FDH. The half-wave potential of the wave shifted by 0.2 V to the negative direction. M450QΔ1*c*FDH adsorbed rather homogeneously in orientations suitable for DET-type bioelectrocatalysis.

An FDH variant (Δ1c2cFDH) which lacks 199 amino acid residues including two heme 1*c* and 2*c* moieties showed a clear DET-type catalytic current. Detailed analysis of the steady-state catalytic current indicated that Δ1c2cFDH transports the electrons to the electrode via heme 3c at a more negative potential and at more improved kinetics than FDH.

Molecular sensing through autocatalytic deprotection of a chemosensitive probe into a redox cycler and its promising use in bioelectroanalysis

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Among the wide variety of molecular amplification schemes, those based on non-linear signal-gain are undeniably the most powerful in terms of sensitivity and dynamic range. An emblematic example is the exponential molecular amplification of nucleic acids by PCR, a technique that was proven decisive in achieving the ultimate specific quantification of a few target nucleic acid sequences in a few tens of microliters. Generally speaking, exponential amplifications originate from autocatalytic reaction schemes in which one of the products catalyzes its own formation, a process that is facilitated with nucleic acids thanks to the rich panoply of biological reagents (*e.g.,* polymerases, nucleases, etc.) available to selectively replicate DNA or RNA. The autocatalytic concept of target replication is however not transposable to non-nucleic acid such as proteins or small organic molecules, and so alternative strategies must be developed if one want to see the amplification power of an autocatalytic reaction to be exploited for improving the analytical detection of such targets.[1-3] In this talk, I will present our ongoing research work on a new molecular autocatalytic reaction scheme that leads to the sensitive detection of either H_2O_2 or redox cyclers through a H_2O_2-mediated autocatalytic deprotection of a chemosensitive probe (pro-RCC) into a redox cycling compound (RCC), generating thus an exponential optical or electrochemical signal gain in the presence of O_2 and a reducing agent (or enzyme) (see the Figure below).

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Keynotes

Phage Display Based Biosensing and Nanomedicine

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Specific probes against targets play siginificant role in biosensing and nanomedicine. Phage display enables to express peptides on the coat protein, which demonstrate many excellent features including multivalency, stability, and high structural homogeneity. In this talk, we present our recent advances in selecting specific phage clones binding with different targets for rapid, selective and sensitive biosensing plotform for antigen, cellulase, tumor cell and ubiquitous pathogens, etc. Additionally, phage fusion protein assembled bio-nanocomposite was applied for optical imaging and targeted photothermal therapy of cancer cells. Taken together, phage display can provide attractive biomolecular probes and should become efficient and cost-effective in bioanalysis and healthcare.

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Controlling Cell Migration and Differentiation using Extracellular Electric Fields

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Extracellular electric fields (EFs) exist during development and injury repair; a natural consequence of directional ion transport across epithelia (McCaig et al., 2005; McCaig et al., 2009). The resulting EFs, which are many-fold smaller than those used for electroporation, guide cell migration, direct the axis of cell division and influence cell differentiation. The roles of such physiological EFs will be discussed in the contexts of mammalian wound healing and in stem cell differentiation during axial regeneration in a planaria flatworm model.

In mammalian wounds the endogenous EFs influence a variety of cells important for the wound resolution, including directional migration of immune T cells as well as keratinocytes, endothelial cells and nerve outgrowth. In addition to directional migration phagocytic uptake is enhanced in human macrophages (Hoare et al., 2016; Cunha et al., 2019; Rajnicek et al., 2018).

The planaria flatworm model is attractive for study of stem cell differentiation because ~30% of the worm's cells are stem cells, called neoblasts. After injury neoblasts undergo selective programming to replace spatially appropriate tissues, but the signals that drive this specificity are unclear. We have measured a head to tail voltage gradient inside planaria and have shown that reversal of the natural gradient can induce spatially inappropriate tissues (eg, head structures at the regenerating 'tail'). With better understanding of the underlying mechanisms the ability to control wound healing and stem cell differentiation electrically may be exploited in clinical therapies.

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Keynotes

Protein Biomarker Diagnostics for Cancer and Metastatic Cancer using Low Cost 3D-printed microfluidic arrays

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Reliable early detection is currently the best hope for successful cancer outcomes in the treatment of primary cancers and metastatic disease. To facilitate translation of early detection multiprotein assay technology into hospitals and clinics, it need to be cheap, fast, accurate, sensitive, and multiplexed. Desktop 3D printing can be used to design and fabricate low cost microfluidic devices that fit these requirements very well. In this paper, we will describe a family of 3D printed, automated immunoarrays that detect multiple proteins with very low detection limits. These low cost, miniature immunoarrays employ electrochemiluminescent (ECL) or chemiluminescent (CL) detection measured with a CCD camera and employ microprocessor control for automated use. Arrays feature prefilled reservoirs to deliver sample and reagents to microwell detection chips and complete sandwich immunoassays. The detection chips achieve high sensitivity by using nanoparticle-antibody or hydrogel-antibody conjugates in the microwells, and ultrasensitive multilabel detection strategies. The total cost arrays is <1 Euro. An 8-serum protein assay can be done in duplicate for $0.14 per protein with LODs as low as 78 fg mL^{-1} in dilute serum. A CL-based array has been designed for single cell detection of oral cancer metastasis in lymph nodes, with LODs of 0.1 fg/mL. A simplified CL assay was been designed that can be implemented in a 3D printed pipette tip. These arrays are well suited to future clinical diagnostic testing, and could be adapted to resource-limited environments.

Flexible Ionophore-Based Sensors for Continuous Monitoring of Electrolytes

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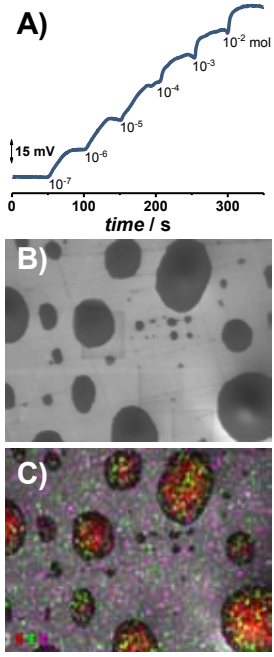
Continuous monitoring of physiological processes allows early detection/diagnosis of possible medical conditions. Information on physiological processes can be extracted by analyzing electrolytes concentration in sweat. These parameters play an important role in diagnostics and attention is paid to new devices that allow their continuous monitoring.



The objective of this research was to design flexible ionophore-based sensors for quantification of targeted electrolytes such as Ca^{2+}, H^+, NH_4^+ and Cl^-. The first and the most important step in the sensors development process was the immobilization of the ionophore at the electrode surface. Different immobilization matrixes were investigated: liposomes of saturated and unsaturated lipids, supported bilayer lipid membranes and nafion. Experiments were performed on $Au/SiO_2/Si$ as model electrodes and finally on Au/PMMA/PET flexible electrodes obtained by electro-spinning process. The interface was characterized by Fourier-transformed infrared spectroscopy (FTIR), scanning electron microscopy (SEM) with energy-dispersive X-ray spectroscopy (EDX), cyclic voltammetry (CV) and electrochemical impedance spectroscopy (EIS). Potentiometry was used to determine the response of the ionophore to the target ion. Sensors parameters like sensibility and limit of detection were determined.

Fig. 1. **(A)** Potentiometric response; **(B)** SEM image and **(C)** EDX map of the Ca^{2+} ionophore sensor.

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Incorporation of Phytantriol Cubosomes into Gold (111) Supported DMPC Biomimetic Bilayer: a Spectroelectrochemical Study

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Cubosomes are lipid liquid-crystalline nanoparticles that can be used as drug delivery systems transporting drugs across the biological membranes. Due to huge internal lipid area ($400 m^2/g$) cubosomes can host large amounts of drugs not exposed to the external solution. They are especially important as carriers hosting toxic, unstable or ill-soluble drugs used in the treatment of cancer.[1]

In this report we use a phospholipid bilayer prepared by the Langmuir-Blodgett method as a simple model of biological membrane Electrochemical impedance spectroscopy (EIS) and photon polarization modulation infrared reflection absorption spectroscopy (PM-IRRAS) were employed to investigate how the properties of DMPC bilayers supported on gold (111) change upon contact with the phytantriol cubosomes present in the solution. The EIS data exhibited the interaction of cubosomes with the lipid bilayers leading to changes of the membrane resistivity. PM-IRRAS measurements provided information on changes in the orientation of the acyl chains to less tilted in the supported lipid membrane exposed to the cubosomes – in result, the lipid layers became more ordered and densely packed.

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Electroanalysis of Enzymatic Activities at a Single Biomimetic Microreactor

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Enzymatic reactions are ubiquitously involved in physiological and pathological processes in living organisms. These reactions are based on protons and electrons transfers and can lead to the production of by-products. Among them, reactive oxygen and nitrogen species (ROS and RNS) are of great interest as they play a dual role: ROS and RNS provide the organism chemical means to react to a stress by the activation of signaling redox pathways, and cause oxidative damages to tissues ensuing dysfunctions in the organism. The high reactivity of such species induce their short lifetimes (ns-min) and leads to uncertainties when searching for deciphering the enzymatic reaction mechanisms in bulk conditions.
This work aims at developing a biomimetic microreactor [1] for the study of enzymatic activities producing ROS/RNS. Indeed, by confining a reaction within a cell-sized compartment (20-100 µm diameter), the generated species (H_2O_2, $NO^{\bullet}$, NO_2^-) could be analyzed in situ with a quantitative and kinetic resolution. Giant unilamellar vesicles (GUVs) are formed in physiological conditions [2] and are used as microreactors [3] for the monitoring of enzymatic activities of glucose oxidase (GOX) and NO-Synthases. Fluorescence microscopy allows individual vesicle observation and the monitoring of reactions triggered by microinjection. Then, released species are detected in real-time by electrochemistry in order to decipher the diverse enzymatic pathways of NO-Synthases [4]. Platinized ultramicroelectrodes (UME) are used to monitor low concentration variations (nM-µM) of ROS and RNS. A single UME is placed at the vicinity of the microreactor membrane and species diffusing through lipidic bilayer, including H_2O_2 and $NO^{\bullet}$ are measured by chronoamperometry [5]. True fluxes of species generated by GOX and NOS under controlled activation/inhibition can then be quantified with this micrometric methodological approach.

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DNA Biosensors as a Tool to Determine the Mode of Action and Nuclease Activity of Novel Bioinorganic Compounds

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Some bioinorganic compounds, on interaction with DNA, can cause significant changes in the DNA structure such as unwinding, oxidation of DNA bases and/or single- and double-stranded breakage in the DNA backbone [1]. Changes in the DNA structure, caused by interaction with the bioinorganic compounds, can inhibit DNA function and, consequently, may result in cell death. Thus, some bioinorganic compounds have the potential to be used as anticancer drugs as they can efficiently kill cancer cells.

The aim of this research project is to use electrochemical DNA biosensors to investigate the interactions between DNA and bioinorganic compounds, in particular, focusing on the nuclease activity of bioinorganic compounds.

The DNA biosensor was fabricated through chemisorption of thiol-modified double-stranded DNA oligonucleotides to create a self-assembled monolayer (SAM) on the gold electrode surface.

The DNA biosensor was used to determine the mode of action and nuclease activity of novel bioinorganic copper complexes. The redox wave of bioinorganic compounds can be observed at the DNA sensor as oxidation and reduction of the compound metal centre is facilitated by DNA *via* a long-range electron transfer mechanism – stacked DNA base pairs behave as the electron conduit [2]. Comparison of the bioinorganic compound redox waves in the presence and absence of DNA can reveal the type of binding mechanism.

The nuclease activity of the complexes was investigated through the measurement of changes in the DNA electrode surface coverage before and after interaction with the bioinorganic compounds. DNA nuclease efficacy can be recorded as a percentage of the amount of DNA cleaved from the DNA-SAM, thus, quantifying the nuclease efficacy of the bioinorganic compounds.

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Enzymatic Biopower: Approaches for optimisation of enzyme electrode performance

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Applications of biomedical devices as implantable and semi-implantable systems such as sensors, valves and pumps are of increased importance. These devices rely heavily on battery power which are reliant on re-charging or sufficient reagents contained within. An alternative strategy is fuel cell technology, using readily available in-vivo substrates (glucose and oxygen) as fuel and oxidant. This is difficult to achieve using chemical catalysts as they are non-selective and operate under harsh conditions (pH and temperature). Enzymes as catalysts offer an alternative route towards powering of such devices. Enzymatic active sites are selective and operate under mild conditions thus offering a potential solution for semi-implantable devices.Here we report on co-immobilisation of osmium based redox polymers with specific enzymes on electrode surfaces. These enzymes are capable of substrate catalysis (glucose oxidation and oxygen reduction). Tailoring of the osmium redox potential to the enzyme active site improves electron transfer to electrode surface. Combining enzyme and redox centre with nano supports such as carbon nanotubes achieves higher current densities and greater power outputs. This is achieved through refinement of the immobilisation procedures as well as optimisation of the enzyme electrode components. Our research uses a design of experiment (DoE) approach for optimisation of electrode surface chemistry to improve current density. The DoE approach improved current density by >50% over traditional one factor at a time (OFAT) approaches, with current densities of 1.2 mA cm^{-2} in biologically relevant glucose concentrations (5 mM) [1]. Fuel cell testing in artificial plasma produced 49% less power than those tested in PBS. Current response of electrodes in the presence of each component of artificial plasma as well as operational stability were recorded in an attempt to understand the reduced performance and identify the component(s) responsible for the change. Electrodes tested in the presence of uric acid produced the lowest current densities and had the lowest operational stability [2]. Electrodes were prepared with different glucose oxidising enzymes to identify if the uric acid affects the enzyme or another component of the biofilm. Electrodes prepared using glucose oxidase were the most stable in the presence of uric acid. This is an important finding for continuous glucose monitoring devices as well as inclusion in fuel cell assemblies.

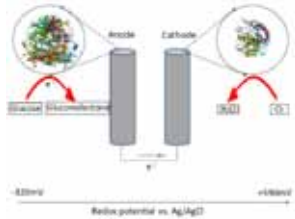

Figure 1: Schematic representation showing the principle of an enzymatic fuel cell with the immobilisation of a glucose oxidising enzyme at the anode and an oxygen reducing enzyme at the cathode.

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Electrochemical Biosensors Based on Microdot Arrays

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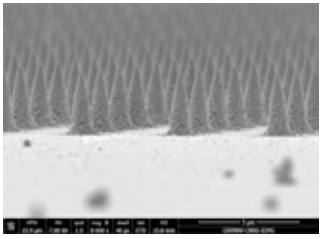
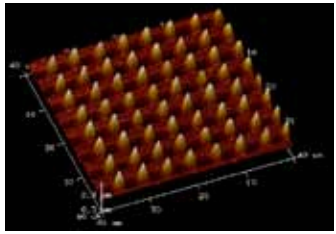
Biosensors based on electrochemical micro and nano-structured transducers have drawn considerable attention over the past few decades[1,2]. Control of their structural surface dimensions is a key factor to improve the biosensor performances (e.g., limit of detection (LOD), sensitivity, linear range, maximum current density, etc…). This study aims to provide valuable insights into controlling the structural features by exploring platinum electrode surfaces modified with microdot arrays (figure a, b) using electrochemical techniques like amperometry. Developing such structures by electron-beam lithography allows adaptation of their size and their composition to provide optimal performance for multiple purposes.

(a)  (b) 

On the basis of electrodes modified with microdot arrays, glucose biosensors[3] were fabricated as a model to study the sensitivity and the LOD. The glucose biosensors were constructed by immobilization of glucose oxidase via covalent interactions with electrogenerated polypyrrole-N-hydroxysuccinimide as already used in recent immunosensor[4]. One key benefit of microstructured surfaces for biosensing is that the sensitivity can be drastically increased (by a factor 12) to provide enhanced performance for just relatively small increases in surface area (by a factor 2). This result demonstrates better electrochemical detection of the generated H_2O_2 and the interest of using microstructures surfaces.

This work benefited from the support of the project ImmuNanoCaps (ANR - 2016 CES09) of the French National Research Agency.

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Electrogenic Transport of Na^+/K^+-ATPase Incorporated in Lipidic Cubic Phase Films as Biomimetic Membranes

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The transmembrane pump Na^+/K^+-ATPase was reconstituted in a biological membrane-mimetic system of monoolein liquid-crystalline cubic phase (LCP) either in form of thin layer or nanoparticles - cubosomes. LCP consists of curved lipid bilayer forming the walls of a network of aqueous channels. It provides the lipid environment for the hydrophobic part of the Na^+/K^+-ATPase, and the aqueous channels enable protein contact with the aqueous environment. We demonstrate that Na^+/K^+-ATPase reconstituted in the cubic phase film preserves native properties and mediates transport of sodium ions through the lipid matrix. To investigate the electrogenic properties of the Na^+/K^+-ATPase chronoamperometry (CA) was employed and increasing potential pulses of positive and negative values were applied to the electrodes covered by LCP or LCP with Na^+/K^+-ATPase films. The results of the ion transport studies using CA were compared with the spectroscopic [1] and square-wave voltammetry (OSWV) assay of the protein activity based on phosphates released from ATP. The net transients generated by Na^+/K^+-ATPase demonstrate the protein mediated ion transport in the cubic phase. Results of the SAXS measurements proved that the structures of cubic phases with incorporated Na^+/K^+-ATPase before and after chronoamperometry experiments are the same, thus confirming that the changes in the net currents reflecting ion-transport efficiency through the film are connected with the activity of Na^+/K^+-ATPase in the cubic phase. In the absence of Na^+, the current related to the pump functions is not observed since Na^+ ions are required in the mechanism involving the switch of the enzyme from E1 to its P-E2 conformation. The differences in ion transport through the empty and Na^+/K^+-ATPase doped LCPs decrease when oubain, the Na^+/K^+-ATPase inhibitor was added to the solution. The same behaviour was observed in ATP-hydrolyzing assay using spectrophotometry or OSWV. It confirmed that oubain effectively inhibits the Na^+/K^+-ATPase when the protein is embedded in the cubic phase. The cubic phase film proves to be a suitable environment for the studies of the protein activation and inhibition.

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Long-term stable glucose determination using redox polymer/enzyme-based amperometric biosensors

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Since the initial concept of enzymatic glucose biosensors was introduced, tremendous progress has been made resulting in the successful commercialization of skin patched or subcutaneously implanted sensors for continuous glucose monitoring (CGM). By the virtue of their accuracy and substrate specificity, combined with their general biocompatibility, enzyme-based electrochemical sensors are seen as key components for such devices. Amperometric CGM systems still suffer a limited long-term and signal stability which is mainly caused by the inevitable foreign body response after sensor implantation, but also by the design of the sensor itself.[1] This implies the choice of a suitable long-term stable active enzyme and, in particular, a stable immobilization/fixation of the biorecognition element on the electrochemical transducer. In this context, redox polymers present a well-suited immobilization matrix that ensures a highly solvated environment for the biorecognition element. Moreover, potential tuning of the polymer bound redox species allows interference free detection of the analyte, *e.g.* the co-oxidation of ascorbic acid or uric acid. In addition, anchoring sites for productive crosslinking of the polymer enzyme films are introduced to ensure stable film formation.

Here, we report the synthesis of Os-complex modified co-polymers that are equipped with suitable anchoring and crosslinking groups (*i.e.* epoxy functions) allowing the attachment of the redox mediator as well as bifunctional crosslinkers. Moreover, styrene sulfonate groups were introduced to control the hydrophilic properties of the polymer matrix. The polymer was combined with either glucose oxidase or PQQ-GDH that were wired via the polymer bound redox species.[2,3]

Long-term amperometric detection was evaluated by using a flow-injection analysis system to minimize background effects and to allow continuous measurements. The effect of polymer composition, type of crosslinkers as well as pre-modification of the electrode surface on film stability and current response will be discussed.

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The Interaction of Heat Shock Proteins with Lipid Membranes: a Novel Diagnostic Target

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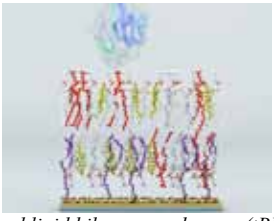
Many members of the underlying heat shock proteins (HSP) are expressed at low levels under physiological conditions and act as chaperones, while others emerge only during stressful conditions to assure cell protection by reassembling protein homeostasis and interfering with apoptotic pathways [1]. As an intracellular polypeptide HSP70 and HSP90 can be exposed on the plasma membrane and/or released into the circulation. However, the role of HSP70 and HSP90 in various nondisease and disease conditions remains unknown.

Our research experiments were designed and performed with heat shock proteins (HSP90 and HSP70) that play key role in the cell membrane damage mechanisms identified in Acute Pancreatitis (AP) and cancer and etc., cases [2], which later can be used as biomarkers for AP and cancer detections.

For the study of biomolecular interactions with membranes, we have developed tethered bilayer lipid membranes (tBLMs) as a long-term stable and versatile experimental model for protein reconstitution and for lipid-protein interaction studies [3]. In this work, different isoforms of HSP's were used to investigate their interaction with tBLM. These HSP's proteins exhibited the membrane damaging properties as probed by the electrochemical impedance spectroscopy (EIS). Membrane composition was found to be one of the important factors affecting the interaction of the HSP proteins to phospholipid membranes.



1pav. Representative image of tethered lipid bilayer membranes (tBLM) interaction with HSP70.

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A Kinetic Model for Redox-Active Film Based Photoelectrodes

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Redox-active films are advantageous matrices for the immobilization of photosynthetic proteins, due to their ability to mediate electron transfer as well as to achieve high catalyst loading on an electrode for efficient generation of electricity or solar fuels. A general challenge arises from various charge recombination pathways along the light-induced electron transfer chain from electrode to the charge carriers or to the solar fuels. Experimental methods based on current measurement or product quantification are often unable to discern between the contributions from the photocatalytic process and the detrimental effect of the short-circuiting reactions. Here we report on a general electrochemical model of the reaction-diffusion processes to identify and quantify the "bottlenecks" present in the fuel or current generation. The model is able to predict photocurrent-time curves including deconvolution of the recombination contributions, and to visualize the corresponding time dependent concentration profiles of the product. Dimensionless groups are developed for straightforward identification of the limiting processes. The importance of the model for quantitative understanding of biophotoelectrochemical processes is highlighted with an example of simulation results predicting the effect of the diffusion coefficient of the charge carrier on photocurrent generation for different charge recombination kinetics.

Redox behavior of azathioprine and it's interaction with DNA

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Azathioprine (AZA), **Figure 1**, a purine analogue discovered in 1957, is used as an immunosuppressive drug and is administrated in a various condition such as autoimmune diseases or in transplantation for prevention of rejection. According to World Health Organization's List of Essential Medicines, AZA is one of the most effective and safe medicines needed in a health system. However, since 2009 AZA is listed as a human carcinogen by the U.S. Food and Drug Administration.
In this context, the electrochemical behavior of AZA and it's interaction with DNA was investigated using the voltammetric techniques and a glassy carbon electrode.

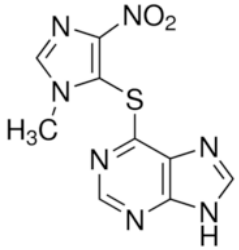

Figure 1 - Chemical structure of azathioprine

The voltammetric results obtained for AZA, in different supporting electrolytes, showed that the redox mechanism takes place in a cascade mechanism. First step was represented by the reduction of the nitro moiety and it was followed by different oxidation\reduction processes. Nevertheless, the interaction of AZA with different DNA structures was investigated.

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Ultra-fast glutamate biosensor recordings in brain slices reveal complex single exocytosis transients

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Glutamate, the main excitatory neurotransmitter in the brain, released through single vesicle compartments during neuronal communication is involved in a majority of brain functions. Sensor technology for detection of glutamate have suffered from detection schemes not providing temporal resolution that is fast enough for monitoring neuronal activity on the time scale for resolving single vesicle exocytosis release events. We here present development of an ultra-fast enzyme-based nanoparticle modified measuring bursts of hundreds to thousands of spontaneous bursts of single vesicle glutamate release when implemented in the nucleus accumbens in mouse and rat brain slice. The ultra-fast sensor temporally resolve a diversity of amperometric current spike shape characteristics, where the quantal release indicate different modes of exocytosis modes in this brain region. Hence this new sensor technology is important for offering new insights in the regulatory aspects of glutamate release and critical to understanding of brain glutamate function and in dysfunction.

Porous electrodes for miniaturized biofuel cells

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The use of enzymes in fuel cells has been extensively investigated.[1-3] Advantages in the use of enzymatic fuel cells such as ease of miniaturization and ability to operate at mild conditions made them a perfect alternative to metal catalyst fuel cells. However, common disadvantages such as low power output and low cell voltage are still not fully overcome. To achieve efficient performances of biofuel cells, enzymes can be immobilized within porous electrodes. Highly ordered macroporous materials generated by gold electrodeposition through a colloidal crystal template are used as bioanode and biocathode of the biofuel cell. The fabrication procedure of macroporous gold electrodes allows a perfect control of the electrode thickness that is tuned precisely at the desired level.[4] Despite the small size of the electrodes, the increase in active surface area due to a high surface/volume ratio of the porous material allows a significant increase of current, a cell voltage of ~0.6 V and an improved long term stability of the enzymes. The miniaturized biofuel cell is also tested for *in vivo* implantation in rats.

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Alteration of tubulin binding and microtubules formation by nanosecond pulsed electric field

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Nanosecond pulsed electric field (nsPEF) treatments of mammalian cells have been shown to permeabilize both plasma and organelle membranes, disturb intracellular vesicles, induce cellular blebbing, swelling, alter intracellular Ca2+ homeostasis, damage cytoskeleton and other effects [1]. Cytoskeleton filaments, especially microtubules (MT), are of a great importance for cellular trafficking and cell division. MT are formed from tubulin heterodimer subunits in a dynamic polymerization process [2]. The control of this process is of a great interest for medical applications. To contribute to the mechanistic understanding of PEF action on MT, we analyze how nsPEF modulates tubulin's binding and microtubules formation.
ZetaPALS from the Brookhaven Instruments Corporation (USA) was used to determine the isoelectric point of tubulin and the Zeta potential of our samples before nsPEF treatment and after nsPEF treatment. Later on, the control and the treated samples with nsPEF were subjected to the polymerization process. The kinetics of tubulin assembly to MT was monitored by standard method of absorbance at 370 nm using microplate reader (Spark, TECAN, Austria). Based on the isoelectric point and zeta potential measurements, the results clearly show that pulsed electric field modifies the surface charge of tubulin. Additionally, the polymerization kinetics of PEF treated tubulin was different from in control tubulin sample as it was assessed by absorbance.

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Use of Alginate Hydrogel Coating to Improve Buckypaper-based Bioanode Stability

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Enzymatic biofuel cells (EBFCs) using glucose and oxygen are attractive power sources for wearable and implantable electronic devices, which consist of a glucose oxidizing bioanode and an oxygen reducing biocathode. Buckypapers (BPs) are emerging materials for bioelectrode fabrication due to their exceptional electrical conductivity, mechanical stability and electrochemical properties [1-2]. We have demonstrated a high-power glucose-based EBFCs using 1,10-Phenanthroline-5,6-dione (PLQ) as mediator for FAD-dependent glucose dehydrogenase FADGDH [3]. However, the instability of bioanode has impeded the further application of this type of bioelectronic devices, which can be attributed to factors such as the deactivation and leaching of both the mediator and the enzyme immobilized on the electrode surface.

Here, we study the effect of hydrogel coating on long-term stability of bioanode. Sodium alginate was chosen as the hydrogel coating material, which is a naturally occurring polysaccharide and can be cross-linked in the presence of calcium ions. A 'sandwich' electrode structure was developed by deposition of a hydrogel layer directly on the nanostructured carbon electrode functionalized with mediator and enzyme. A homogenous gel layer can be visually seen on the electrode surface. Cyclic voltammograms reveal that both operational and storage stability of the hydrogel coated electrode has improved compared to the uncoated one.

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Photobioelectrodes with Anisotropic Electron Flow by Oriented Immobilization of Photosystem 1 Monolayers

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The large abundance, robustness, and high quantum efficiency of certain protein complexes participating in the photosynthetic process make them particularly interesting building blocks for the fabrication of efficient and cost-effective biophotovoltaic devices for solar energy conversion. In particular, photosystem 1 (PS1) is a natural photodiode capable of generating a charge separated state upon the absorption of visible light, revealing a potential difference of about 1 V. However, the considerably large voltage difference achieved between the two opposite redox sites at PS1 also translates into a substantial driving force for short-circuiting processes cancelling out part of the generated photocurrent and making the design of PS1-based devices particularly challenging. Therefore, recovery of the high-energy photoexcited electrons exiting the PS1 protein complex requires immobilization strategies that allow the fabrication of stable and well-defined structures capable of providing an optimized electron transfer with, ideally, a unidirectional electron flow.

The effective modification of electrode surfaces with PS1 protein complexes under a preferred orientation has been achieved by taking advantage of the amphiphilic nature of the isolated protein complex. After spreading of PS1 onto the air-water interface, the hydrophilic ends at PS1 bearing the terminal redox sites adopt a specific orientation either facing air or being solubilized in water. Moreover, stacking of the hydrophobic regions enables the formation of a stable monolayer. After transfer of the film to a gold surface serving as electrode substrate by means of the Langmuir-Blodgett technique, the achieved PS1-based photobioelectrodes revealed an anisotropic electron transfer, enabling the effective extraction of photoelectrons.

Fabrication and development of microelectrodes for label-free biosensing

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With the electroanalytical advantages of reduced *iR* drop, improved signal to noise, relative insensitivity to convection and steady state currents, microelectrodes represent an exciting candidate technology for the development of biosensors. One particular attraction is the ability to fabricate arrays of microelectrodes for the simultaneous detection of multiple analytes. This is particularly advantageous for diagnosis and monitoring of complex medical conditions such as sepsis, drug resistant infections and oncogenic mutations. Problems with the development of microelectrodes for biosensing, particularly those utilising electrochemical impedance (EIS) spectroscopy have included: interpretation of the more complex EIS response and developing reproducible surface functionalisation chemistries to give stable baseline measurements. This talk will initially focus on fabrication of the multi-microelectrode arrays we use for biosensing applications. Emphasis will then shift to measurements of DNA-DNA hybridisation for MRSA using microelectrodes[1] (identifying the necessary SAM formation conditions[2] and electrode choice to successfully record the impedimetric response upon DNA target binding). The talk will conclude by showing a newly fabricated array of microelectrode sensors on a needle shaped substrate which have been shown capable of detecting a common inflammatory biomarker of sepsis (Interleukin-6) at clinically relevant levels in near real time[3].

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Redox Glyconanoparticles and Buckypapers for the Design of Enzymatic Fuel Cells

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The need for clean methods of producing electricity have stimulated the emergence of a new generation of a subcategory of fuel cells: enzymatic fuel cells. These biodevices that convert chemical energy into electrical energy by electro-enzymatic reactions, have attracted considerable attention over the last decade [1]. Recent advances in the design of bioelectrodes based on electrically wired enzymes onto carbon nanotube coatings will be reported. In particular, a new generation of flexible buckypaper electrodes was produced by using linear polynorbornene polymers containing multiple pyrene groups as crosslinker and applied to the elaboration of enzymatic and hybrid fuel cells [2,3]. In addition, the use of compression for the staking of several buckypapers with the entrapment of enzymes will be described.

An innovative approach based on the electrical wiring of enzymes in solution by redox glyconanoparticles resulting from the self-assembly of bio-sourced block copolymers will be presented. We demonstrate the self-assembly, characterization and bioelectrocatalysis of redox-active cyclodextrin-coated nanoparticles. The nanoparticles with host-guest functionality are easy to assemble and permit entrapment of hydrophobic redox molecules in aqueous solution. These nanoparticles (diameter: 195 nm) were used as electron shuttles between electrode and bilirubin oxidase providing enhanced and stabilized current densities for enzymatic O2 reduction. [4]. Finally, we will demonstrate the innovative elaboration of solubilized enzymatic fuel cell that can be easily restored or replenished [5].

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Scanning Ion Conductance Microscopy (SICM) for Determining the Local Charge Environment of Gram-positive and Gram-negative Bacteria

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Scanning Ion Conductance Microscopy (SICM) has well-known capabilities in non-invasive probing of the local environment of biological samples, extending now to live bacteria held in physiological conditions. In addition to compatibility with optical microscopy techniques,[1] SICM versatility and resolving power allows pointwise measurements across a sample, producing a detailed sample topography map, along with other analytical measurements including delivery of metabolites,[2] local pH measurements,[3] activation of ion channels,[4] and surface charge[5]

Previous surface charge has commonly been measured by proxy, through non-localised Zeta potential measurements.[6] Herein, the charge distribution across the surface of Gram-positive *Bacillus subtilis*, and Gram-negative *Escherichia coli* bacteria was investigated using the ionic current rectification at a 150 nm SICM nanopipette tip, as induced by the bacterium surface. Results indicate a negatively charged region directly over both bacteria types, with a heterogeneous charge distribution over the surface. We observe with notable charge differences was found between the two different bacteria strainsspecies. These observations provide a good starting point to elucidate how cell charge in microbes relates to other physiological parameters and processes, such as metabolism, membrane potential, and growth.

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Detection of Interleukin 6 from Human Serum by Label Free Electrochemical Sensors

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Cytokines, soluble proteins secreted by the immune and non-immune cells, are involved in the modulation of many immune reactions including chemically-induced tissue damage, control of cell replication, apoptosis and cancer progression and development [1]. Interleukin 6 (IL6), an inflammation biomarker, is a circulating protein that can be found in plasma of healthy individuals at low levels and in increased concentrations in inflammations, cardiovascular diseases, metabolic syndrome and diabetes, as well as in mood disorders. Due to its implication in several pathologies, its detection from serum is of paramount importance from biomedical point of view. Two approaches using label free immuno and aptasensors were designed and will be compared in terms of analytical performances and ability to selectively detect IL6 from human serum samples.

The covalent immobilization of IL6 antibody through amidic bound formed with the carboxyl functionalities found at the surface of protein G-functionalized magnetic microparticles immobilized on graphite based screen printed electrodes, provided an immunoassay with electrochemical label free detection, with a limit of quantification of 1 pg mL^{-1} and a limit of detection of 0.3 pg mL^{-1} [2]. In the second approach, the glassy carbon electrode was modified with *p*-aminobenzoic acid, *p*-aminothiophenol and gold nanoparticles and then a thiol-terminated aptamer for IL6 was immobilized *via* the formation of gold-sulphur bonds. This approach showed a good linear response from 10 pg mL^{-1} to 100 ng mL^{-1}, with a detection limit of 3.3 pg mL^{-1}.

A critical comparison between antibodies and aptamers as well as the influence of the immobilization platform will be discussed in terms of detection limit, selectivity and tests in blood serum collected from patients.

Acknowledgements: This work was supported by the Romanian National Authority for Scientific Research and Innovation, CNCS/CCCDIUEFISCDI, project no. PN-III-P1-1.2-PCCDI-2017-0221 IMPROVE.

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Bioelectrochemical Sensor Commercialisation

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This presentation gives an overview of the operation and detection mechanisms of four types of bioelectrochemical sensor::

- ISFET pH sensor
- Continuous Glucose Monitoring
- Nanopore nucleic-acid base identification and sequencing
- Capacitive DNA detection

The commercialization history of each (or lack thereof) is discussed, as well as commercialization timelines – decades in some cases. The prospects for more rapid commercialization of emerging bioelectrochemical sensor technologies are discussed, in the context of emerging trends of eHealth, Internet-of-Things, and The Trillion Sensor Initiative.

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Combining Membrane-Bound Enzymes on Electrodes for Electroenzymatic ATP Regeneration or Detection

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Modification of electrode surfaces with supported phospholipid bilayers as biomimetic membranes allows arranging membrane-bound enzymes on a controlled way for electrochemical applications. In this way, the catalytic activity of the enzymes modulated by the associated transport processes across the biomimetic membrane is combined with the driving force given by the electrode and the sensitivity of electrochemical detection.

The combined immobilization of the membrane-bound NiFeSe hydrogenase from *Desulfovibrio vulgaris* with *E. coli* ATPsynthase F1-FO (ATPase) allows conversion of H_2 fuel into the biochemical energy vector ATP. The system consists in a membrane-modified electrode where a lipid bilayer serves as enzyme support and simultaneously provides a confined aqueous phase close to the electrode; which allows a local acidification that triggers ATPase's ATP production [1]. This electroenzymatic assembly can be used as ATP regeneration system of biocatalytic reactions that require a high consumption of ATP, such as phosphorylation reactions catalyzed by kinases.

We also show that ATP-synthase reconstitution on a membrane-modified gold electrode can be used for specific ATP detection using the SAM on the gold surface as redox probe.

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In situ Fluorescence Confocal Microscopy for the Study of Electro-enzymatic Reactivity

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Thermodynamic and kinetic data on relevant redox enzymes have been obtained through electrochemical techniques during the last decades. However, in the case of immobilized enzymes, several key-features remain unknown, especially spatial information since the electrochemical measurement is averaged over the entire surface of the electrode. No data about surface heterogeneities are therefore provided. In this context, there is a major interest in coupling electrochemical techniques to microscopy methods. Such coupling enables collecting simultaneously spatial information [1] and affords a cartography of the entire surface homogeneity with respect to electrochemical reactivity. We show here that *in situ* coupling between fluorescence confocal microscopy and electrochemistry [1, 2, 3] can be implemented to characterize electro-enzymatic catalysis. For example, enzymatic O_2 reduction or H_2 oxidation involves proton transfers, which can be evidenced via fluorescence change from a pH-dependent fluorophore such as fluorescein. We demonstrate that local pH changes in the electrode plane can be visualized both during O_2 reduction catalyzed by an immobilized bilirubin oxidase and during H_2 oxidation catalyzed by an immobilized hydrogenase [4]. Moreover, proton gradients generated during the enzymatic electrode reaction can be readily observed since recording fluorescence intensity in the volume adjacent to the electrode under potential control enables rebuilding the diffusion layer.

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Bio-inspired laser-induced electrochemical sensing strategies: antigen/nanobody based magnetosensing

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In my talk a sensing strategy inspired by the advantages of enzymes and photoelectrochemical sensing, namely the integration of aerobic photocatalysis and electrochemical analysis, will be presented. The photosensitizer, a bioinspired perfluorinated Zn phthalocyanine, generates singlet-oxygen from air under visible light illumination and oxidizes analytes, yielding electrochemically-detectable products while resisting the oxidizing species it produces. Compared with enzymatic detection methods, the proposed strategy uses air instead of internally added reactive reagents, features intrinsic baseline correction via on/off light switching and shows C-F bonds-type enhanced stability. It also affords selectivity imparted by the catalytic process and nano-level detection, such as 20nM amoxicillin in ml sample volumes. [1]

Additionally, this strategy can be applied to improve the sensitivity of assays by using magnetic beads in combination with an amperometric readout in a configuration so-called a electrochemical magnetosensor. The main difference between of a magnetosensor from a biosensor is that a biorecognition element is linked to magnetic beads and can be temporally dispersed in a sample for improving binding kinetics and, then, accumulated on an electrode by a magnet to register an analytical response. In comparison with conventional optical read-out systems, electrochemical sensors can register an analytical response almost immediately and without the need of developing the reaction in bulk volume, because the response of electrochemical sensors is determined by a thin layer of measuring solution near the electrode surface where the beads with immunoreagents are placed. This approach may improve sensitivity and decrease analysis time in comparison to conventional ELISA.

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Scanning electrochemical microscopy imaging of poly (3,4-ethylendioxythiophene)/thionine electrodes for lactate detection via NADH electrocatalysis

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Herein we report the use of scanning electrochemical microscopy (SECM) together with electrochemical and spectroscopic techniques to develop and characterise a stable and uniformly reactive chemically modified platinum electrode for NADH electrocatalysis. In order to achieve this, a range of different approaches for thionine entrapment within an electropolymerised poly(3,4-ethylendioxythiophene) (PEDOT) film were evaluated using SECM imaging in the presence of NADH, demonstrating the uniformity of the reactive layer towards NADH oxidation. The effect of electrolyte type and time scale employed during PEDOT electropolymerisation was examined with respect to thionine loading and the resulting charge transport diffusion coefficient (D_{CT}) estimated via chronoamperometry. These studies indicated a decrease in D_{CT} as thionine loading increased within the PEDOT film, suggesting that charge transport was diffusion limited within the film. Additionally, thionine functionalised nanotubes were formed, providing a stable support for lactate dehydrogenase, lowering the rate of thionine leaching via SECM imaging and enabling lactate determination at E_{app} = 0.0 V vs Ag/AgCl over the range 0.25-5 mM in the presence of 1 mM NAD^+.

Towards Low-cost, Simple and Biodegradable Microbial Fuel Cells for Energy Harvesting and Sensing Applications

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Microbial Fuel Cell (MFC) is an attractive technology for energy harvesting, bioremediation and self-powered environmental monitoring applications. Key features of the technology include no emission of harmful gases, use of renewable feedstocks, including waste, and, depending on the materials and design implemented, cost effectiveness.

We focus on the use of low-cost materials and extremely simple device designs, which facilitate large scale applications and implementation in poor areas. We use several feedstocks, in either liquid, slurry or semi-solid phase, such as digestate from food waste, urine, algae, soil. We also explore the use of low-cost membrane as well as membrane-less designs, which include a fully biodegradable paper-based MFC device. Both energy harvesting and sensing applications are explored, with also some preliminary results on soil bioremediation.

Our results demonstrate the feasibility of the technology as low-power energy supply in remote areas, as well as the ability of providing on site, real time detection of toxicants in water, for shock sensing purposes.

Magnetic Electrodes and Nanoparticles for Direct Immobilization of Biomolecules through Magnetic Forces

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A new approach to simplify electrode architectures based on the magnetic immobilization of biomolecules is proposed. The procedure involves the preparation and characterization of metallic electrodes with magnetic properties, at which magnetic metallic nanoparticles (MeNP) are directly attracted, without externally applied magnetic field. The electrodes can play a dual role: as immobilization support for biological molecules and as transducers of the biological reactions. Additionally, magnetic MeNP can act as carriers for the biomolecules toward the immobilization support.
Both magnetic electrodes and MeNP were obtained by simple and cost-effective methods: Ni electrodes by electroplating and NiNP by the chemical reduction of a Ni salt. Also, Pd was employed for doping Ni electrodes (Pd@Ni electrodes), so as to benefit from both the catalytic effect of the Pd and the magnetic properties of Ni. The magnetic properties of these electrodes and of the NiNP were investigated by Magneto-optic Kerr effect (MOKE) measurements. The electrode surface and the NiNP were characterized by X-ray diffraction (XRD), scanning electron microscopy (SEM) with energy dispersive X-ray spectroscopy (EDX) while their chemical composition was determined by X-ray photoelectron spectroscopy (XPS). Electrochemical methods, such as cyclic voltammetry and electrochemical impedance spectroscopy, allowed to determine the nature of the electrochemical process at Ni and Pd@Ni electrodes. Both electrodes were applied as electrochemical sensors for: i) the detection of H_2O_2 and ii) immobilization of the model enzyme glucose oxidase (GOx) functionalized with NiNP solely through magnetic interactions. The performance of the biosensor for glucose determination was investigated by fixed potential amperometry. The biosensor was able to operate at +0.05 V (vs. Ag/AgCl) with no interferences.

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Connecting photosystem I with electrodes by fullerens

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One of the most attractive natural tools for photo-to-current conversion represents photosystems I (PSI). Many approaches for the construction of PSI-based photobioelectrodes have already been reported in the literature[1-3]. However, the defined immobilization of this protein supercomplex on electrodes represents still a demanding issue[4]. In this study a new assembly of PSI on the electrode has been employed and a novel approach for electric wiring of PSI with the electrode has been introduced. Results indicate a preferred orientation of PSI on a thiol-modified gold surface, which has subsequently been modified with ruthenium hexamine. Compared to the direct electron transfer of PSI at this electrode a 10-fold higher photocurrent is found when PSI is connected via fullerenes. The turnover number (Te = 230 e s^{-1}) and the internal quantum efficiency (IQE = 10 %) illustrate the rather high efficiency of this approach.

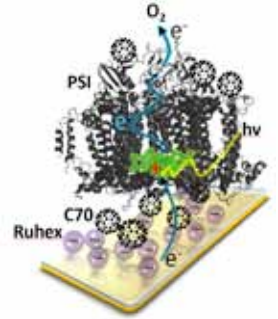

Figure 1. Schematic representation of PSI-C70 electrode

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Thiolated chitosan layers adsorbed on gold surface as a system for deposition of lipid bilayer

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Chitosan is a polysaccharide biopolymer composed of D-glucosamine and N-acetylated glucosamine units linked by glyosidic bonds. Naturally occurring chitosan is one of the components of the fungal cell wall; therefore, chitosan shows biocompatibility properties.[1] Moreover, the presence of the polar moieties provides the ability to absorb a large amounts of water. Therefore molecular films of chitosan derivatives can be considered as a cushion for deposition of lipid bilayer.

In this work, we have synthesized 3 thiol derivatives of chitosan: chitosan modified by lipoic acid, chitosan oligosaccharide lactate modified by lipoic acid, chitosan oligosaccharide lactate modified by 3-(tritylthio)propionic acid. In order to characterize the self-assembled films of chitosan derivatives, we used electrochemical methods such as a cyclic voltammetry, electrochemical spectra impedance and alternating current voltammetry. In the next step, atomic force microscopy (AFM), was applied to visualize the adsorbed layer. By applying potential to the electrode, we were able to observe the structural changes of the film at different potential values. The highest capacitance value was observed for chitosan oligosaccharide lactate modified by lipoic acid and this system was further examined by electrochemical quartz crystal microbalance technique with dissipation monitoring (EQCM-D)

The ACV results showed that the thiolated chitosans remain stable at the gold surface over a wide potential range. All three systems can absorb water, however, its content varies with the potential.

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Size dependent catalysis of phosphine-modified gold nanoparticles

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Gold nanoparticles (AuNPs) possess distinct physical and chemical features that make them useful as scaffold components for the fabrication of novel electrocatalytic and bioelectrocatalytic systems[1]. Gold nanoparticles of 2 nm diameter differ from larger nanoparticles in terms of optical, electrical and chemical properties. Small particles can behave more like molecules than as particles and exhibit enhanced catalytic activities and interesting charging properties[2,3]. In this work we focus on the size of AuNPs and on the behavior of the smallest gold nanoparticles – called clusters, with diameters ranging from 2 nm to subnanometer size. We present the synthesis of gold nanoparticles modified with phosphines[4] and their utilization in nanostructuring of electrodes using particles of different sizes. In contrast to nanoparticles stabilized with thiols, stabilization with phosphines enables further modification and in addition - the particles are characterized by a higher monodispersity index. The size of the gold nanostructures was controlled by the ratio of the stabilizing ligand to the gold precursor used in the synthesis. The performance of the electrode in the catalytic processes of oxygen reduction depends on the size of the nanoparticles modifying the electrode surface.

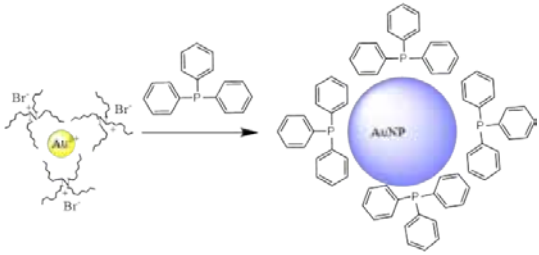

Figure 1: Scheme of the synthesis of gold nanoparticles modified with phosphines.

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Enzymatic Histamine Biosensor Based On Prussian Blue-Modified 3D Pyrolytic Carbon Microelectrodes

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In allergic reactions, mast cells and basophils release histamine[1]. Here we develop a novel electrochemical enzymatic biosensor for the detection of histamine in biorelevant media. A three-electrode chip consisting of pyrolytic carbon working electrode (WE) with micropillars, and platinum counter (CE) and reference (RE) electrodes is fabricated by photolithography with SU-8 photoresist followed by pyrolysis. CE and RE are patterned by e-beam metal deposition through a shadow mask[2]. The 3D WE is modified with Prussian Blue (PB) film by electrodeposition[3]. PB film on 3D pyrolytic carbon microelectrodes displays enhanced deposition, uniformity and stability compared to flat electrodes. Next diamine oxidase (DAO) in a solution containing BSA and glutaraldehyde is cross-linked on the WE. The modified chip is placed in a magnetic clamp holder fabricated with PMMA by micromilling. The holder has an inlet above the electrodes to provide access for manual sample introduction. A miniaturized PCB interfaces the chip with the potentiostat. The mechanism of histamine biosensing consists of several steps. PB on WE is electrochemically reduced to form Prussian White (PW) and the open circuit potential (OCP) measurement is started. When the electrode is exposed to the solution containing histamine, imidazole acetaldehyde and hydrogen peroxide (H_2O_2) are generated at the electrode by DAO. H_2O_2 subsequently oxidizes PW producing PB imposing a shift in OCP. The amount of the produced PB is then assessed by recording chronoamperometric current by applying a step of reducing potential. The histamine assay is done in a stagnant sample volume (150µl). The above-described two-step electrochemical method- i) open circuit potentiometry followed by ii) chronoamperometry- is expected to provide practically sufficient sensitivity. During OCP measurements, histamine molecules have sufficient time to diffuse towards the WE and, due to catalytic chain reactions, result in charge accumulation in the PW/PB layer. From the preliminary results, the detection limit of 500 nM histamine in buffer was obtained. Currently, we are working towards detecting histamine in biorelevant media at concentrations below 100 nM, which is the target level of histamine in tissue released from mast cells.

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Adsorption and Electroreduction of Cytosine Oligonucleotide Blocks at the Mercury Electrode

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Behavior of nucleic acids (NA) at electrodes is influenced by their nucleobase composition, sequence and secondary structure. Despite recent data demonstrating reduction of all NA bases at the pyrolytic graphite electrode, mercury electrodes are still most frequently used tool for the studies of electrochemical reduction of natural nucleobases. Among them, cytosine and adenine are irreversibly electroreduced while reduction of guanine (possibly through a mechanism involving chemical reduction by hydrogen evolved at the electrode surface) gives a product that can be electrooxidized back to guanine, giving a well-developed anodic signal. Moreover, signals of polyanionic NAs interacting with the negatively charged atomically smooth mercury electrode surface are strongly influenced by the NA structure, allowing sensitive detection of helix-coil transitions, DNA damage, interactions of DNA with small molecules as well as G-quadruplex formation.

Our recent data indicate that interactions of NA chains with/at the electrode surface are remarkably influenced by the presence of homonucleotide blocks. Homopyrimidine oligonucleotides exhibit 2D condensation at negatively charged electrode surface and stronger adsorption than homopurine ones. In this work we focus on cytosine blocks which, among all homonucleotide blocks, were most tightly adsorbed, as indicated by the dominance of C-E curves characteristic for homocytosine stretches obtained in competition experiments. Moreover, long homocytosine stretches gave well developed reduction signals at unusually high pH values (above pH 8) at which random (but containing cytosine at levels corresponding to at least ¼ of all nucleobases) yielded no reduction peaks. Notably, maximum pH values at which cytosine reduction was observed were dependent on the length of homocytosine blocks separated by other nucleotide sequences and followed similar trends as the propensity of the same nucleotides to form cytosine tetraplexes - i-motifs, (as assessed by CD spectra measurements).

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Wet Oxidation Liquor Treatment using Microbial Fuel Cells

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Treating wastewater using cheaper and more sustainable methodologies is becoming a greater concern throughout the world for water efficiency and resource reusability. This study focuses on assessing the use of Microbial Fuel Cells (MFCs) to treat wastewater liquor samples generated by sub-critical wet oxidation (WO) of simulated human excreta at a variety of temperature and pressure levels. These waste products were provided by Scion.

Semi-permeable membrane of the MFCs was built out of cylindrical terracotta ceramic with carbon veil as the anode and activated carbon/PTFE mixture pressed over a carbon veil cathodes [1]. This work further analyses the electrochemical output and the organic/nutrients degradation of a cascade of four MFCs running in continuous flow with a flow rate of 8.5ml/h^{-1}. MFC performance was measured in terms of power density, chemical oxygen demand (COD) reduction as well as pH, conductivity and ammonia levels.

MFCs fed with WO liquor reduced the COD content by 24.4% over a time of 24 h generating levels of power of <0.4 Wm^{-3}. When WO liquor was mixed with hydrolyzed urine at a 1 to 1 ratio, power density increased tenfold to 4Wm^{-3} and COD reduction increased to 29% after 24 h, whilst ammonia concentration increased.

The study concludes with the use of multiple MFCs connected in a cascade, intensifying the liquor treatment and achieving a COD reduction of 45.85% and a power density of 0.8 to 2 Wm^{-3} with neat WO, compared to WO + urine, which achieved a 37.10% COD reduction and produced a power density of 7 to 10 Wm^{-3}.

Thanks should be given to the Bill and Melinda Gates Foundation for the financial support of this work.

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Microbial electrochemistry enables nutrient recovery from urine while delivering benefits to urban wastewater networks

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Establishing the circular economy of the future requires the development of innovative concepts that will allow for the continuous removal and recovery of valuable resources – including nutrients from wastewater – so that they can be reused continuously. Source-separated urine contains 79% of the nitrogen (N), 71% of the potassium (K) and 47% of the phosphorous (P) released in human bodily wastes, yet constitutes ≤ 1% of the total volume of wastewater generated in sewered urban environments. Separated urine is therefore an ideal target for macro- and micro-nutrient removal and recovery, and for this reason we have developed the bio-electroconcentration (BEC) concept through a partnership between the Advanced Water Management Centre and Queensland Urban Utilities. The concept, better known as UGOLD, can be described as a hybrid between electrodialysis and microbial fuel cells, whereby nutrients are concentrated across ion-exchange membranes by an electric field generated by the action of electrochemically active bacteria. Nutrients are recovered in a liquid concentrate with low-to-nil energy input, while bacterial catalysis also helps to partially treat the urine (*e.g.* organics and sulfate removal) before it is discharged to sewer, delivering additional benefits to the sewer network (*e.g.* less sulfur = less corrosion by sulfide) and end-of-pipe treatment plants (less N to remove = less energy/chemicals used). In 2017, our team demonstrated the BEC concept in the laboratory with 3-compartment microbial electrochemical cells of 600 cm^3 working volume, attaining current densities of up to 29 A m^{-2} for a specific energy consumption of 2.4 kWh kgN^{-1} to produce a liquid fertiliser containing 2.6% N. In 2018, a demonstration was completed for a scaled-up system of approximately 6 litres, capable of treating the urine from ~40 users while generating up to 2 litres/day of concentrated liquid fertiliser. Substantial economic benefits and added flexibility for urban wastewater networks are demonstrated.

Towards the understanding how silver species influence electroactive *Geobacter sulfurreducens* biofilms

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Recovery of heavy metals from waste streams of mining and industry can be considered as an important contribution to a "green" world. The dissimilatory-metal reducing microorganism *Geobacter sulfurreducens* (*Gs*) is established in microbial fuel cells for electricity production coupled to the oxidation of acetate. Less known is its ability to produce nanoparticles from a variety of partially surprising metal ions like silver [1] and uranium [2] in the presence of a suitable electron donor. This ability is important for the immobilization and, therefore, filtration of heavy metals from waste streams and indicates a resistance mechanism towards such metal ions. The recent investigations on the mechanism by which planktonic *Gs* forms silver nanoparticles support the hypothesis that cytochromes are involved and give indications for two mechanisms depending on the silver species (i.e. silver ions and silver chloride) [3]. While this study was done using planktonic *Gs* cells, the influence of silver species on *Gs* biofilms grown on electrodes for the electrode-assisted bioremediation is still in its infancy.
Here we present results obtained with *Gs* biofilms on carbon electrodes which were either operated as anodes, where silver reduction is a competing reaction to electricity generation, or as cathode, where electrons needed for the reduction process are provided by the electrode.

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Long term bio-power of Microbial Fuel Cells in individual and stacked configurations

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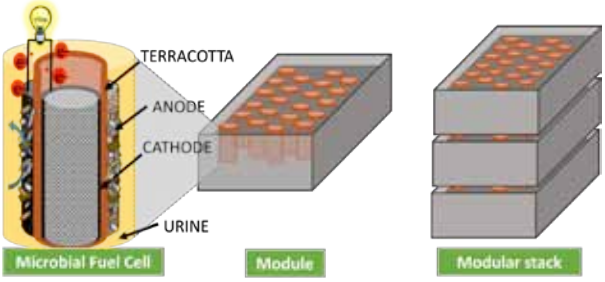
Microbial Fuel Cells (MFCs) utilise organic feedstocks such as urine as fuel for direct electricity production, by employing anode respiring microbes that convert organic matter into electrons while treating waste. In order to scale-up the MFC technology towards real-world applications and reach usable power levels, the MFCs units can be operated in collectives (stacking) of small-scale MFC units [1] using affordable and durable materials. Ceramic materials have shown to be suitable and cost-effective separators for MFCs [2] therefore the aim of this work is to assess the MFC performance and long-term operation of MFCs, as individual units, as well as a plurality of units connected together as modules (stacks) and modular stack cascades (Fig.1).

MFCs were constructed using carbon veil anodes on the outer side of the terracotta cylinders and activated carbon cathodes on the inner side of the ceramic (Fig.1).

MFCs were tested as (i) individual units as well as (ii) multiplied in a module (22 MFC units) forming a stack where MFC units were electrically connected in parallel and (iii) in a cascade configuration using 3 modules (66 MFCs).

Power output showed that the individual MFCs produced ca. 1.6 mW (3 replicates), while the module of 22 MFCs produced >22 mW and the modular stack cascade produced >70 mW. Long term operation was assessed over 1 year of continuous feeding with neat human urine.



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Interfacial Electron Transfer of Cytochrome *c* at an Electrified Liquid-Liquid Interface

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To date metallic electrodes have dominated the study of protein electrochemistry. However, proteins are highly liable to undergo denaturation when immobilized on a variety of functionalized electrode surfaces due to undesirable protein-electrode interactions.[1] Thus, a major focus of bioelectrochemistry is the development of different platforms to preserve the native state of immobilized proteins.

An interesting approach is protein electrochemistry at a "soft" electrified liquid-liquid interface, known as *i*nterface between *t*wo *i*mmiscible *e*lectrolyte *s*olutions (ITIES). The ITIES has long been considered a biomimetic model of half a biological cell membrane.[2] Most studies at the ITIES have been performed in highly acidic aqueous solutions displaying a characteristic electrochemical signal inherent to facilitated ion transfer.[3] However, studies under more gentle conditions imitating real biological environments are essential to provide key insights into biological electron transfer systems and processes.

Herein, we studied the electrocatalytic behavior of Cyt *c* at the ITIES at different pH values (2, 7 and 9). Cyt *c* was chosen as a model biomolecule due to its well-studied role in cell death, known as apoptosis.[4] Electrochemical and spectro-electrochemical experiments involving interfacial electron transfer between aqueous Cyt *c* and decamethylferrocene (on the oil side) demonstrate that Cyt *c* acts as an interfacial catalyst towards the oxygen reduction reaction (ORR). These experimental findings are supported by molecular dynamic simulations of Cyt *c*'s interaction with the liquid-liquid interface.

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Development of electrochemical and optical biosensors for the detection of mosquito borne diseases

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Mosquito borne diseases are endemic in more than 100 countries, causing about 1 million deaths every year. The early and facile diagnosis of vector borne infectious diseases using electrochemical and optical methods has attracted considerable interest in the field of biosensors. The challenges associated with conventional diagnostic methods like polymerase chain reaction (PCR), enzyme linked immunosorbent assay (ELISA) can be addressed using these methods. The goal of the present work is to develop electrochemical and optical fiber biosensors, which can easily detect the presence of dengue and chikungunya. The electrochemical detection method makes use of electrochemical impedance spectroscopy, cyclic voltammetry and differential pulse voltammetry. The capture of the chikungunya and dengue virus by corresponding immobilized antibodies was evaluated using these methods. A clear change in electrochemical responses was observed for virus exposed electrodes (figure 1). The preliminary studies to analyze the performance of localized surface plasmon resonance based silica optical fiber as a biosensor were performed with GaHIgG (antibodies from goat) and HIgG ((Human IgG) as model samples. The real time absorbance for antibody immobilization showed a significant increase in absorbance response which suggests that the optical fiber can be further explored for the detection of analytes of interests.

Fig. 1 Nyquist plots of impedance spectra showing each stage of surface modifications during biosensor development and detection of ChikV at 10µg/ml

Wiring of Bacterial Cells to Electrodes through Redox Polymers

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We have throughout the years since 2004 pioneered "wiring" both Gram-positive and Gram-negative bacteria [1-3] (and thylakoid membranes [4]) to electrodes using osmium redox polymers (Os RPs) that will strongly facilitate extracellular electron transfer (EET). Because of the cationic nature of the Os RPs they will electrostatically form very strong complexes with the cells and form hydrogels that will precipitate on the electrode surface and will allow substrates and products to freely diffuse in and out of the hydrogel. We have investigated the influence of E°'-value and structure of the Os-complexes of the Os RPs on the rate of the EET. However, only recently we have obtained a much more clear picture on how the RPs and the cells interact and how the interaction changes with time. In these recent investigations we have "wired" wild type and some mutants of *Enterococcus faecalis* with both 4 different Os RPs as well as with a quinone RP [5-8]. These recent results will be shown and discussed.

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New electrogenic bacteria Paenibacillus dendritiformis MA-72

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In this study, the electrogenic properties of bacterial strain *Paenibacillus dendritiformis* MA-72, isolated from the anodic surface of long-term operated sediment microbial fuel cell (SMFC), were examined. Initially, the optimal conditions for biofilm formation were explored by cultivation of bacteria in three-electrode cell at different applied potentials. The SEM observations reveal that *P. dendritiformis* forms dense biofilms on carbon felt only under polarization conditions and moreover, the orientation of bacteria in the obtained biofilms diverges at different poised potentials. The electrochemical activity of the mature bacterial biofilms was further investigated by means of cyclic voltammetry, differential pulse voltammetry and electrochemical impedance spectroscopy. The results obtained show that the bacteria *P.dendritiformis* MA-72 are capable of exchanging electrons with the electrode via direct electron transfer mechanism. Under strict anaerobic conditions, the highest current generation was achieved at poised potential of +0.220 V (vs. SHE). Carbon felt electrodes covered by *P.dendritiformis* biofilm were also explored as bioanodes in a single-chamber MFC with two air-cathodes. The maximal power density achieved with this MFC-configuration was 18 mW/m^2. The results from the complex analyses carried out undoubtedly show that *P.dendritiformis* MA-72 is an electrogenic strain, contributing to the current generation of the SMFC, from which it was isolated.

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Fundamental insights into the electrooxidation of halocytosines at carbon based electrodes: mechanism, electroanalytical and computational studies

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Epigenetic modifications in DNA are associated with a plethora of pathophysiological and metabolic dysfunctions. One of the major epigenetic modifications is the methylation of DNA. Although DNA methylation takes place naturally as a mechanism of gene expression, hypermethylation (non-natural) is associated with the aforementioned disorders. On the other side, radical species can give rise to the formation of halocytosines such as 5-chloro (ClC) and 5-bromocytosine (BrC) which can mimic the function of methylation, causing a fraudulent effect in the transcription modulation. In this regard, improved analytical methods for the determination of these epigenetic modifications in DNA as biomarkers may allow the detection of certain diseases in the early stages of development. This work reports for the first time the exploration of the electrochemical behaviour of ClC on different carbon electrodes using cyclic voltammetry and square wave voltammetry. When comparing for example both glassy carbon (GC) and boron doped diamond (BDD) materials, the former turned out to be the appropriate carbon material in terms of a more well-defined anodic wave and higher sensitivity for the electrooxidation of ClC. The electrochemical oxidation potential of ClC was found to be linearly pH-dependent with a maximum current intensity in acetic acid buffer solution at pH 5.0. A linear response between peak current intensity and ClC concentration was obtained within the range of 200 and 1,000 μM with a limit of detection of 200 μM. The main oxidation products after a preparative electrolysis were detected by HPLC-Mass Spectroscopy. Simultaneous detection of ClC in the presence of the unmodified cytosine and mixtures containing other nucleic bases such as guanine, adenine and thymine was also addressed. Finally, the effect of the halogen atom (X=F, Cl, Br), on the electrooxidation process was examined by theoretical calculations.

Design and characterization of marine-based plasma membrane vesicles

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Screening of natural products found in the marine environment is a highly competitive field, where pharmaceutical companies have strong interests. Systematic understanding of isolated bio-interfaces like plasma membrane vesicles, where other complex, intracellular biochemical processes are excluded, may be a prerequisite for future discoveries and innovations. Unique to the photosynthetic marine microalga *Dunaliella* is the absence of a rigid cell wall. *Dunaliella* possesses a glycocalyx type of cell envelope, and its softness and deformability facilitate the manipulation of its plasma membrane by osmotic stress. Isolated plasma membrane vesicles mimic biological membranes because they are formed from the complex assembly of proteins, sugars, and lipids that make up the cell membrane, which distinguishes them from commonly used model systems like phospholipid vesicles. Several imaging techniques have been employed for structural characterization of these *Dunaliella*-derived vesicles and to image the efficiency of intracellular release. We carried out qualitative comparisons of molecular transport of fluorescent dye through membrane systems of different levels of complexity (plasma membrane vs. model lipid membrane). The observed differences in transport are ascribed to differences in membrane structural features and physicochemical properties.

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Phenotype-based Nanoamperometric Signalling of Breast Cancer Drug

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Tamoxifen (TAM) and toremifene (TOR), which are selective estrogen receptor modulator (SERM) drugs, are among the most prescribed breast cancer treatment drugs. Variation in the inter-individual response patterns of patients to the SERM drugs pose real problems for the adequate prescription of the drugs. Drug toxicity due to overdose and non-response to treatment due to under-dose, are some of the consequences of improper prescription. Clinicians and biochemical analysts have been involved in active research work aimed at developing protocols for reliably determining the metabolic classification (as slow, regular or rapid metabolisers) of breast cancer patients for TAM and TOR. Assays based on cytochrome P450 (CYP) enzymes that metabolise TAM and TOR or the CYP genes (since they exhibit genetic polymorphism) are being explored. Commercially available genotype-based tests (genosensors) for determining the drug-response patterns (i.e. whether a patient exhibits impaired, extensive or rapid metabolism of the SERM drugs) of breast cancer patients are very costly and do not incorporate the effect of environmental factors. Electro-phenotype biosensing (with *b-cancer phenosensor*) of SERM drugs have been developed as a possibly, more efficient drug-response pattern test than the gene-based tests. The *b-cancer phenosensor* consists of a biocompatible telluric chalcogenide quantum dot-CYP enzyme composite sensing film, that is chemically printed on gold chip electrode, which responds to SERM drugs by amperometric signalling.

Lactoglobulin as a platform for designing biologically active carriers

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β-lactoglobulin (LGB) can serve as a potential transport and depot protein, particularly for hydrophobic molecules, through the binding of potential ligands at active sites. The primary binding site of LGB is located inside the β-barrel (~2mm deep) and the second one on the protein surface. This study reports the effectiveness of LGB as a potential drug carrier for the anesthetic tetracaine (TCA) under varying environmental conditions (pH, ionic strength, concentration, LGB-TCA complex molar ratio). TCA is a linear, hydrophobic molecule of length ~1.5mm, similar to the natural ligands (fatty acids) of LGB. In this work, spectroscopic UV-vis and Laser Doppler Velocimetry (LDV) were utilised to determine the physicochemical properties of LGB and LGB-TCA complex in a sodium chloride solution. Electrophoretic mobility measurements showed that in comparison to a pure LGB solution, the addition of TCA deceased the zeta potential as well as shifting the isoelectric point to a higher pH. This suggests the formation of LGB-TCA complexes. UV-vis spectra for LGB, TCA and its complex were recorded. The binding constant (K_{UV}) of the complex was calculated. The adsorption capacity of LGB on the gold surface was studied using quartz crystal microbalance with dissipation energy monitoring technique (QCM-D). From this technique, the optimal conditions (pH, ionic strength, concentration) to form the LGB-TCA complex were determined. Molecular docking was implemented to estimate the binding position of the ligand. The correlation between the adsorption capacity of LGB-TCA and pH or ionic strength of the solution was evaluated, and the optimised conditions for LGB-ligand binding were investigated. The experiments revealed that the characterisation of the layers formed on the QCM-D sensor might play an important role in studies of future protein-hydrophobic ligand interactions.

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Building Better Biosensors: Controlling the Surface Density of Redox Probes on a Boron-Doped Diamond Electrode

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Boron-doped diamond electrodes have significant potential for utilization in DNA biosensors due to their bio-compatibility, robustness and non-fouling properties.[1] However, unlike more commonly used gold and glassy-carbon substrates, the chemistry required to attach DNA and other biomolecules with controllable surface density to a boron-doped diamond electrode is relatively unexplored. In this contribution, we demonstrate that grafting of N-Boc terminated diazonium salts to a diamond substrate can be performed reproducibly, with subsequent attachment of a redox-active anthraquinone label or DNA probe. The surface density of the probe can be controlled through reduction of a mixed diazonium salt solution in which one of the components acts as an inert 'spacer molecule' in an approach similar to that demonstrated by others previously at glassy-carbon and gold substrates.[2] Surface densities for redox probes on diamond can be controlled in the region of $\sim 10^{11} - 10^{12}$ molecules/cm^2, comparable to that utilized for DNA immobilized on gold electrodes.[3]

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Enzyme Inhibition in Continuous Flow Measurements in a Microfluidic System

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Bilirubin oxidase (BOD) is an enzyme that is frequently used in biocathodes because of its ability to reduce O_2 directly to water. It is known that many enzymes of the blue multicopper oxidase family are inhibited by the presence of halogen ions such as chloride. Although BOD is less sensitive to Cl^- inhibition than e.g. laccase, there are still many open questions about the inhibition mechanism and function.

Here we investigate inhibition of BOD in a flow-injection microfluidic system. This setup allows us to distinguish between temporary and permanent effects of a particular inhibitor by making it not only possible to quickly inject a certain concentration of inhibitor in question, but also to quickly remove the inhibitor. This also enables us to study the time-scale of the recovery of enzyme function after the inhibitor is removed.

In this work we study inhibition of BOD by three compounds often encountered during fuel cell operation; ascorbic acid, hydrogen peroxide, and chloride ions. We show that these three inhibitors act in completely different manner. Ascorbic acid lowers the oxygen reduction current but leaves no lasting effects after it has been removed. After Cl^- exposure the enzyme function is only partially recovered, while after exposure to H_2O_2 the enzyme function is permanently lost. These data are compared with experiments using laccase. We also discuss merits and problems with the present setup and possible improvements.

Electrochemical Catalytic Probes for Amplified Biosensing

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Recently, the investigation on electrochemical biosensing has made great progress. The developed biosensors have extensively been used in different fields, particularly, the sensitive and accurate detection of DNA, protein and metal ions. The designs of specific electrochemical catalytic probes are becoming the main topic of electrochemical biosensing research. This presentation will introduce new electrochemical catalytic probes designed in our group for development of amplified biosensing strategies in last two years. For example, we prepared two electrochemical catalytic probes to design two molecular switches for electrochemical DNA sensing by coupling with porphyrin-encapsulated metal-organic frameworks. Several new signal amplification strategies based on nanotechnology and molecular biology have been presented for DNA detection and immunoassay. By the recognition of nucleic acids to metal ions, some label-free methods have been developed for specific and sensitive detection of heavy metal ions. Some new electrochemiluminescent emitters have been synthesized and used for design of new ECL biosensing mechanisms. A wavelength-resolved ratiometric photoelectrochemical technique has also been developed for sensing applications. These works led to a series of amplified methods for sensitive detection of DNA, microRNA and proteins.

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Development of a DNA based electrochemical biosensor to determine the overall genotoxicity of multi-pesticide soil extract

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Modern agriculture is reliant on the efficient application of pesticides to maximize crop yields, by preventing losses due to pests (Popp et al, 2013). Pesticide genotoxicity in soil is a complex issue which can be influenced by soil characteristics, environment and pesticide stability. Electrochemical DNA biosensors are rapid, sensitive and cost effective compared to other conventional methods currently in use to assess pesticide content. Electrochemical impedance spectroscopy (EIS) is a label free method that measures changes in the charge transfer resistance (Rct) before and after damage to DNA oligo probes attached to the sensor surface. This allows a highly sensitive detection of DNA alterations compared to voltammetric techniques.

In this study, we will utilize screen printed graphite working electrodes (SPE) due to their low cost and ease to fabricate in bulk. Incorporation of gold nanoparticles (AuNPs) onto the working electrode surface increases the electrode conductivity and surface area, while allowing for the stable attachment of the thiolated probe oligonucleotides (Yang et al, 2011, Omidfar et al, 2013, Wang et al, 2014). Therefore, we will electrochemically deposit AuNPs onto the surface of the working electrodes to bind thiolated double stranded DNA probes.

Upon fabrication, the biosensors will then be exposed to different mixtures of pesticides and other oxidising agents, and the genotoxicity will be determined by the change in Rct over time (20-30 minutes) as measured by EIS. The biosensor design will then be validated using pesticide extracts from contaminated soils collected from representative sites around Ireland. Representative sample sites and the composition of the pesticide mixtures that will be used to test the sensor will be determined in late January-early February. Once validated the biosensors will become an important tool as part of a National Soil Screening programme.

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A Novel Approach to 7-dehydrocholesterol Determination in Smith-Lemli-Opitz syndrome Diagnosis

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Smith-Lemli-Opitz syndrome (SLOS) is an autosomal recessive, multiple congenital malformation/mental retardation syndrome. Its clinical characteristics vary greatly in severity and manifestations. SLOS is caused by an inborn mutation of DHCR7 enzyme coding gene. This enzyme facilitates the biotransformation of 7-dehydrocholesterol (7-DHC) to cholesterol. Therefore, a clinical suspicion of SLOS can be confirmed by demonstrating elevated levels of 7-DHC [1], for example in amniotic fluid (prenatal) or plasma (postnatal). There is a lack of routine neonatal methods of screening for SLOS, and thus new methods are highly desirable [2].

In this contribution, a simple, fast, and cheap method of 7-DHC determination is presented. The determination process is based on electrochemical oxidation of 7-DHC which, to our knowledge, has not been described until now [3]. We have shown that 7-DHC can be oxidized at approximately +800 mV (*vs.* $Ag/AgNO_3$) in non-aqueous medium of acetonitrile, or mixed medium acetonitrile-artificial serum on boron-doped diamond electrode. The current response can be employed for 7-DHC determination with LOD of 0.2 μM in both the acetonitrile medium, and acetonitrile-water medium. This LOD value is several orders of magnitude lower than the expected 7-DHC levels in SLOS patients. When perchloric acid is used as the supporting electrolyte, a second oxidation signal appears. We believe this is due to a dehydration reaction analogical to the one we have described for the end-product of cholesterol catabolism, bile acids [4].

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Effect of calcium ions on the cell membrane models

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Calcium electroporation is a new experimental anti-cancer treatment where calcium is internalized into cells by application of short, high voltage pulses. The anti-tumour effectiveness has been demonstrated *in vitro*, *in vivo* and in clinical studies as well as effect on vasculature has been explored [1,2]. However basic physical principles of calcium ions on the cell membrane is not fully elucidated. The aim of the study was to introduce the calcium ions to the simplified and controllable models of cell membrane such as computer Molecular Dynamics simulation (MD), and experimental planar lipid bilayers and liposomes. The results of each model can be analysed and compared. In all of the investigated models $CaCl_2$ were added in a buffer solution on one side of the membrane. Results of molecular dynamics simulating POPC membrane shows binding of calcium ions to the lipid headgroups and decreasing of the area per lipid [3]. Experimentally measured capacitance on planar lipid bilayers increases due to adding $CaCl_2$ into a buffer solution in steps of 0.5 mM from 0 to 5 mM. Experiments on liposomes were performed using gas phase electrophoresis to evaluate the diameter of the prepared vesicles [4]. Diameters of the vesicles were measured in buffer of 40 mM ammonium acetate and the same buffer with added $CaCl_2$. Results shows that diameter of vesicles reduces with added calcium ions. Three simplified models of cell membrane were studied. MD simulation results of capacitance increase are in agreement with planar lipid bilayer experiments. This increase of planar lipid bilayer capacitance results from calcium ion binding on the lipid headgroup. Also, the reduction of diameter of the vesicles experiment are in agreement with the area per lipid value calculated from MD simulations.

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Flexible neural electrodes based on self-supporting carbon nanotube films

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Electrical stimulation as the way to treat neural tissue has been known in medicine for the last decades. Currently, deep brain stimulation (DBS) is a common tool to treat several neurological disorders, including chronic pain, depression, tremors associated with Parkinson's disease and epilepsy. However, the major drawback of the existing DBS technologies is the design of the neural electrodes, which lack in flexibility, biocompatibility and capacitance. In this study, we introduce self-supporting CNT films as robust neural interfaces. This flexible material made entirely from carbon moieties was found to possess a highly capacitive nature, elevated electrically active surface area and superior charge injection behaviour, in theory, allowing for the use of low electrical potentials in neuromodulation approaches without compromising on the therapeutic effects of stimulation. Cytocompatibility studies indicate that CNT films can significantly promote neurite adhesion and facilitate neurite outgrowth, as well as decrease the presence of reactive astrocytes *in vitro*. CNT films have been found to outperform platinum control electrodes as well as other materials commonly used as neural interfaces. Demonstrating minimal loss of performance following repeated deformation, CNT films are shown to be promising materials for the design of flexible and cytocompatible neural interfaces.

Acknowledgements
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Helping enzymes to fight against thermodynamics: bioelectrocatalytic uphill production of hydrogen

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Chemical systems do not allow the coupling of energy from several simple reactions to drive a subsequent reaction, which takes place in the same medium and leads to a product with a higher energy than the one released during the first reaction. In the here presented example, Gibbs energy considerations are not favorable to drive water splitting by the direct bioelectrochemical oxidation of glucose as a model reaction. This presentation will however show that it is nevertheless possible to carry out such an energetically uphill reaction, if the electrons released in the oxidation reaction are temporarily stored in an electromagnetic system, which is then used to raise the electrons' potential energy, so that they can power in a second step, after electrical uncoupling, the electrolysis of water in the same reaction vessel. We thereby demonstrate the general concept that lower energy delivering chemical reactions can be used to enable the formation of higher energy consuming reaction products, even if both electrochemical cells (biofuel cell and electrolysis cell) are combined in a single closed system. The presented device can be therefore considered as a semi-artificial version of a hydrogenase, but with the advantage of operating under ambient aerobic conditions, using a rather robust enzyme and thus leading to a higher stability.

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Molecular Imprinting of Peptide Nucleic Acid (PNA) in an Electropolymerized CG-Rich Artificial Oligomer Analogue for Determination of Genetically Relevant Oligonucleotide

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We devised and fabricated a chemosensor for selective determination of genetically relevant 5'-GCGGCGGC-3' (G-guanine, C-cytosine) oligonucleotide. Toward that, we simultaneously synthesized electrochemically and deposited on a Pt electrode a sequence-defined *octakis*(2,2'-bithien-5-yl) polymerized film of a DNA hybridizing probe.[1] For that purpose, we used both an approach of macromolecular imprinting in a polymer and a peptide nucleic acid (PNA) template of a programmable sequence. For transducing an oligonucleotide recognition event into the analytical signal, we applied electrochemical impedance spectroscopy (EIS) and surface plasmon resonance (SPR) spectroscopy under stagnant-solution and flow-injection analysis (FIA) conditions, respectively. Using EIS, we determined the target oligonucleotide with the 200-pM limit of detection. With the EIS determined apparent impact factor, IF≈4.0, the chemosensor discriminated both two-nucleotide-mismatched oligonucleotides and Dulbecco Modified Eagle Medium sample interferences.

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Bioelectrode Engineering - Control of Catalytic Film Thickness for Enzymatic Fuel Cells

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Redox hydrogel films are commonly applied for bioelectrode fabrication and we recently demonstrated that such matrices can be engineered to efficiently protect the hydrogenases from O_2 damage to enable their integration into H_2/O_2 biofuel cells [1,2]. The formation of these films is highly reproducible for thicknesses between 30 to 300 µm. However, the most relevant thickness range for technological applications is in the low micrometer thicknesses which are more challenging to produce due to film thickness heterogeneities. Here, we propose the in-situ gelation of monodispersed viologen-modified macromolecules to achieve homogeneous films with thicknesses down to 100 nm. The relative standard deviations of the mean film thickness, was found to be only 4.8%. Electroanalytical investigations reveal defect-free, efficient electron transfer pathways over the complete film. We illustrate the advantage of this method and the importance of the film homogeneity for bioelectrocatalytic applications by investigating the behavior of bioanodes built from ferredoxin-$NADP^+$-oxidoreductase or NiFe-hydrogenase for NADPH and H_2 oxidation, respectively. The ability of forming thin films reveal that peak catalytic performances are reached at 10-fold lower catalyst loading compared to conventional approaches owing to the extreme homogeneity in film morphologies. This method will be generally transposable to other related applications, involving catalyst deposition by drop-casting with strict demands for film homogeneity.

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Investigation of Ionic interactions at a lipid cubic phase with Electrochemical Impedance Spectroscopy

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Electrochemical impedance spectroscopy was used as a complementary technique to investigate cationic and anionic interactions in the headgroup region in the aqueous channels of lipid cubic phases, LCP. The experiments were performed at freestanding LCP membranes exposed to different electrolyte solutions at both sides and at various ionic strengths. A two electrode set-up was employed for Electrochemical Impedance Measurements.

Lipid Cubic phases were prepared from pure Monoolein and water: MO/H_2O or the ternary mixture of monoolein/dioleylphosphatidylcholine/water: MO/DOPC/H_2O. The membranes were exposed to mono- and divalent metal cations and three different anions.

The impedance data was fitted to relevant equivalent circuits to estimate membrane resistances and membrane capacitances. These values were discussed in the light of earlier findings in the literature and also with SAXRD measurements on the same systems. The cations could be arranged in a direct Hofmeister series that reflects the intrinsic binding constant with DOPC headgroup.

Swelling and condensing effects to various degrees were observed at cationic exposure. At exposure of tryphtophane very large membrane resistances were observed, that is explained with ion channel blocking and a change in the bilayer ordering in the interfacial region.

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The effect of amiloride an ion channel blocker on pore formation by alamethicin in negatively charged and zwitterionic bilayers - a molecular view.

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The formation of an ion conductive pore by alamethicin (a short chain antibiotic peptide) and blocking of the pore conductivity by amiloride (an ion channel blocker) were investigated with the help of scanning microscopies, polarization modulation infrared reflection absorption spectroscopy (PM-IRRAS) and electrochemical impedance spectroscopy (EIS).[1,2] Molecular resolution images of alamethicin channels in phospholipid monolayer and bilayer were obtained. The IR spectra showed that the insertion of alamethicin and the ion conductive pore formation is controlled by the applied electrostatic potential. The changes of the membrane conductivity due to ion channel formation were measured by EIS. Excellent correlations between the changes of membrane conductivity and AFM images of the pore formed by alamethicin molecules and orientation of the inserted peptides (determined from PM IRRAS experiments) were observed. These techniques also were applied to investigate blocking of the alamethicin pores by amiloride. The addition of amiloride to the solution bathing the bilayer with alamethicin caused an order of magnitude decrease of the membrane conductivity. Significant differences between the mechanisms of interactions of amiloride with a zwitterionic and negatively charged membrane were observed. In the case of zwitterionic membranes amiloride binds to the 3_{10}-helix of the alamethicin molecule preventing its insertion into the membrane. In the case of an anionic membrane, amiloride is inserted into the channel increasing the pore dimension. These differences were explained in terms of electrostatic interactions between the positively charged amiloride molecule and negatively charged bilayer.

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Light-triggered sugar detection using semiconductor materials

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Light can be used as a valuable tool for a multiplexed analysis of biosensor surfaces provided a spatially resolved immobilization of different biological recognition elements is feasible. The presentation will demonstrate that such a multiplexed approach can be realized without any crosstalking using two different oxidases on a quantum dot modified gold electrode. Here signal generation is based on the oxygen sensitivity of the QD electrode allowing the parallel detection of glucose and sarcosine on one sensor surface.
Furthermore it can be shown that semiconductor nanowires - which can be prepared by a bottom up method - can be used for improved signal to noise ratio in the detection process [1].
3D electrodes provide the general advantage to harbor larger amounts of recognition elements. It can be shown that they can also be combined with a light-triggered read-out by sensitization of a TiO_2 material with a sulfonated polyaniline and coupling to the PQQ-GDH enzyme [2]. Even higher current densities can be achieved by a sensitization with in situ prepared quantum dots and combination with a redox polymer coupled enzyme [3].
The last two systems also demonstrate another attractive feature of these light-triggered sensors – the rather low potential at which electrons can be collected from the biocatalytic sugar oxidation. This may illustrate that several advantages can be combined in photoelectrochemical sensing schemes.

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Interplay between Orientation at Electrodes and Copper Activation of *Thermus thermophilus* Laccase for O_2 Reduction

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Blue multicopper oxidases have received considerable attention because of their ability to oxidize a large variety of organic substrates while reducing O_2 into water with efficiency comparable to platinum catalyst. Bilirubin oxidases (BOD), laccases (Lac) and copper efflux oxidases (CueO) belong to this family. They share different copper sites: one type 1 copper, the entry site of electrons where the substrate binds, one trinuclear active site where O_2 reduction takes place and which is composed of one type 2 copper and a binuclear type 3 copper (1). In addition to these four Cu sites, CueO is able to weakly bind additional Cu atoms at a methionine-rich helix positioned as a lid over the binding site cavity. Hence, depending on the environmental condition, CueO is involved either in organic substrate oxidation (phenol oxidase activity for example) or in Cu^+ oxidation (2).
As a thermostable enzyme, Lac from *Thermus thermophilus* HB27 (*Tt* Lac) may be coupled with thermophile hydrogenases in H_2/O_2 enzymatic fuel cells. Enzymatic activity using classical Lac substrates is however very low, only enhanced by exogenous Cu^{2+}. As *Tt* Lac possesses a Met-rich hairpin, Cu binding to this domain was proposed to be involved in the catalysis. Little is known however about electrocatalysis using *Tt* Lac (3), and the effect of copper salt addition has never been studied at electrochemical interfaces.
In this work, we discuss the electrochemical behavior and enzymatic properties of *Tt* Lac immobilized on different electrodes. Based on a careful examination of the enzyme structure, we demonstrate that the direct wiring of the enzyme is driven by strong repulsive electrostatic interactions between the Met-rich hairpin domain and the electrode. We then highlight an additional electroenzymatic process in the presence of exogenous Cu^{2+}, and we prove it is related to the Met-rich hairpin. Finally, mutant studies allow to give new insight in the electron transfer pathways.

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Basics to Utilise Direct Electron Transfer Enzymes in Carbohydrate Biosensors

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Enzymes capable of direct electron transfer have been intensively studied for the last three decades, especially as electrode biocatalysts for third generation biosensors. Promising biosensors for various analytes have been build, investigated and published. The first commercialised third generation biosensor – produced by DirectSens GmbH for the detection of lactose in lactose reduced milk and milk products – is now on the market for three years. What are the problems and difficulties encountered during the development of a third generation biosensor?
Based on the structure and natural function [1, 2] of the employed multi-cofactor enzyme cellobiose dehydrogenase, the most important factors for efficient catalysis, intermolecular electron transfer (IET) and direct electron transfer (DET) will be discussed, which are: (i) substrate specificity, (ii) pH [3], (iii) ions, (iv) electrode surface chemistry and (v) the orientation of the enzyme on the electrode [4,5]. The molecular basis for these potentially performance limiting factors will be explained and strategies to overcome them presented. The specifications of the produced lactose and glucose biosensors [6] show their strengths, but also point out weaknesses and the direction of further necessary research to improve their performance.

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Operando X-Ray Absorption Spectroscopy Applied to Bioelectrochemistry

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Although metallic cofactors within enzymes are usually associated to their biocatalytic activity, the mechanisms involving these cofactors are often under debate. One example is bilirubin oxidase (BOD), whose structure contains 4 copper atoms and is an well-known biocatalyst for the oxygen reduction reaction (ORR).[1] We propose an *operando* X-Ray Absorption Spectroscopy (XAS) setup to study the behavior of a BOD-modified electrode under ORR conditions. We observe that the Cu ions within BOD do not reduce themselves in order to provide an energetic environment favorable for the ORR, since no spectroscopic feature attributed to Cu(I) is observed under mild potential conditions. Instead, their reduction to Cu(I) only occurs in a much lower potential, indicating that during the ORR the Cu ions work as an electronic bridge for the electron transfer towards the O_2 molecules. These observations were possible by using a novel *operando* XAS approach that allows the spectroscopic measurement under electrochemical control of the enzyme.

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In Vivo Brain Monitoring Using Minimally-invasive microelectrode biosensors Based on Platinized Carbon Fibers

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Monitoring the chemical composition of the brain interstitial fluid can be achieved with microelectrode biosensors Typically, oxidase enzymes are immobilized on a platinum microelectrode that oxidizes hydrogen peroxide (H_2O_2) produced by enzymatic activity. Non-electroactive molecules like glucose and lactate can be detected by these devices, their size is quite large and could potentially damage brain tissue, modifying the local composition of the interstitial fluid. Here, microelectrodes based on platinized carbon fibers were fabricated to obtain biosensors with less than 15 µm external diameter. Platinization was achieved by evaporation of a 10 nm Ti adhesion layer followed by 100 nm of Pt. Platinized carbon fibers could provide very thin microelectrodes for in vivo brain oxygen detection, or be functionalized with oxidase enzymes to monitor glucose or lactate interstitial concentrations. After implantation in the rat parietal cortex, such biosensors detected smaller basal oxygen and lactate concentrations and a slower diffusion of glucose through the blood brain barrier compared to more conventional biosensors with 90-100 µm external diameter. Interestingly, spreading depolarizations (SD) produced a smaller increase in lactate and a larger decrease in glucose at platinized carbon fiber microelectrode biosensors compared to larger sensors. Therefore, developing smaller, less invasive probes for brain monitoring is key to preserve the blood brain barrier at the site of implantation, and provide more physiological measurements from the brain interstitial fluid.

Implantable Hybrid Biofuel Cell with Abiotic Biocathode and Enzymatic Bioanode

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The lithium batteries are currently the most widely used source to supply power implantable medical devices. However, lithium reach their limitations in terms of the power density required for modern implantable devices that require more electrical power. In this context, glucose/oxygen enzymatic biofuel cell is a promising alternative, since it is able to convert the chemical energy of glucose and oxygen present in physiological fluids into electrical energy[1,2]. Nevertheless, the enzymatic catalytic system has to overcome the three major issues of *(i)* the short lifetime of the enzyme, *(ii)* the biocompatibility of the compounds, and *(iii)* the low energy yield due to the low oxygen concentration present in the body. Here we investigate the biocompatibility and electrochemical performances of an abiotic biocathode based on graphene doped iron and an organic binder. Electrochemical performances show that the chitosan-based biocathode exhibits an electrochemical stability for more than one year (figure 1). Biocompatibility studies shows that the several compound of the biocathode are cytocompatible without any toxicity (figure 2). Moreover, an hybrid biofuel cell based on this abiotic cathode and an enzymatic bioanode exhibits a long-term stability under physiological condition for several months.

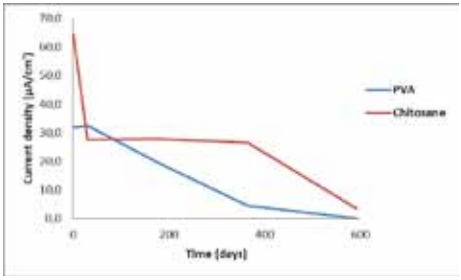

Figure 2: Current density in time of biocathodes with graphene, phosphate buffer saline solution, pH 7,4 200mV vs ECS

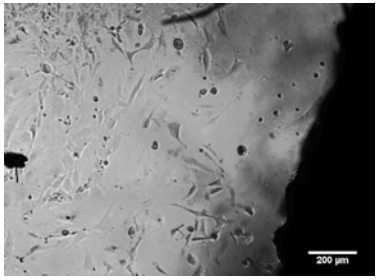

Figure 1: Cytocompatibility biocathode with graphene after 4 days, 37°C, 5% CO_2, DMEM

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Exploration of the Biodiversity of CO Dehydrogenases using Protein Film Electrochemistry

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Ni-containing CO dehydrogenases (CODHs) are very efficient metalloenzymes that catalyse the fast conversion between CO_2 and CO. They are a source of inspiration for designing CO_2-reduction catalysts and can also find direct use in biotechnology. Until now, only a small number of CODHs have been studied and properly characterised, however those enzymes might not be representative of the entire family of the CODHs in terms of catalytic properties and behaviours.
In this work we study several CODHs from different microorganisms, which have never been purified and characterised before, using mainly protein film electrochemistry. This technique consists in adsorbing the enzyme onto an electrode surface that can be characterised using several electrochemical methods, mainly cyclic voltammetry and chronoampeometry. This allows us to study the mechanism of CODHs, in particular their inhibition by oxygen, which is one of the main issues for a potential application of CODH as CO_2-reduction catalyst. In fact, CODHs are deemed to be extremely sensitive to O_2, but very little is known about this feature. Nevertheless, we determined that distinct CODHs exhibit different behaviours towards oxygen [1, 2].
We currently characterise and compare several CODHs from different origins, in order to have a better understanding of the biodiversity of this enzyme family. For example, we investigate their reaction with O_2, and the dependence of their Michaelis-Menten constant (K_M) for CO on the applied potential. In addition, we study their catalytic bias (CO oxidation *vs.* and CO_2 reduction) and their activation/inactivation by the substrate CO. The results obtained with different CODHs will help us to determine the molecular bases of the biodiversity of this metalloenzymes.

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Monitoring changes in the expression profile of long noncoding RNAs associated with prostatic cancer by using an electrochemical biosensing platform

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Recent progress in RNA sequencing technology has revealed the involvement of non-protein coding part of human transcriptome in multiple and very important biological processes. Long noncoding RNAs (lncRNAs) are a large and heterogeneous subclass of transcripts with lengths above 200 nucleotides that, despite being coded by the genome, are not translated into proteins. Altered expression levels of certain lncRNAs in human body fluids have been recently associated with different cancer conditions that turn them into potential non-invasive biomarkers.[1] Among them, prostate cancer antigen-3, PCA 3, has been the only one approved by FDA in the USA for clinical use as a urinary biomarker for prostate cancer diagnosis to date.[2]

The analysis of PCA 3 using hybridization-based biosensors is particularly challenging as a consequence of its large size and internal secondary structure, as well as their low abundance. To address these issues, the incorporation of short DNA fragments acting as hybridization assistant probes is herein proposed. They hybridize in solution with the long target sequence, thus disrupting its secondary structure and, in turn, facilitating its selective capture onto a sensing platform obtained by covalent binding of amino-terminated capture probes to *p*-mercaptobenzoic acid monolayers onto gold supports.[3] Furthermore, these probes were designed to allow the accommodation of multiple redox enzymes per binding event, making use of the monovalent and high-affinity fluorescein-Fab antifluorescein interaction. This strategy enabled on-surface quantification of long transcripts with an enhanced sensitivity and a wider range of linear response.

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Direct electrochemistry of Cytochrome c with conducting polymers

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Direct electron transfer (*DET*) to redox proteins is a major issue for the development of biotechnological devices as biosensors, biocatalytic systems, biofuel cells, etc. Cytochrome c (*cyt c*), a water-soluble heme protein, is probably one of the most extensively explored compounds in electrochemical applications. *DET* to *cyt c* from conventional electrodes as Pt, Au, or Ag is usually hindered since the peptide chain of this protein strongly adsorbs on metallic surfaces and conformational changes suffered by the protein lead to slow electron-transfer kinetics.

The traditional approach to overcome this difficulty is the electrode modification with thiolate-self assembled monolayers (*SAM*) to promote the appropriate orientation of the protein and thus enhancing its electroactivity. However, *SAM* present several drawbacks when used as electrode modifiers: they are limited to some few substrate materials (Au, Ag, Cu), decrease the surface conductivity and present low chemical and electrochemical stability (especially during oxidation). These drawbacks can be overcome by using conducting polymers, as an alternative to SAM. These polymers constitute a class of organic modifiers that offer high versatility in several biotechnological applications of proteins. These materials have been used as enzyme immobilizers, *DNA* biosensors, electronic transducers in biosensors, or drug delivery systems, among others. Due to its adaptability and ease of preparation, conducting polymers have been also used as promoters of *DET* to several redox proteins and, particularly, to *cyt c*.

The present work explores the modification of electrode with poly(3,4-ethylenedioxythiophene) polystyrene sulfonate (PEDOT-PSS) conducting polymer prepared by electrochemical methods. The voltammetric response of the *cyt c* has been analyzed as a function of the chemical composition of the polymer film. The films synthesized were characterized by several electrochemical and spectroelectrochemical methods in order to evaluate the redox state of the polymer as a function of the potential and, in addition, the observation of intrinsic *cyt c* redox activity upon electron transfer from the conducting polymer. During the oxidation process of this protein, lysine residues placed near the heme crevice interact electrostatically with the electrode surface doped with negatively charged PSS moieties. This interaction favors the orientation of the heme group toward the chains of the PEDOT backbone, which is eventually responsible for the electron transfer to the protein.

Controlling The Drug Release From Mesophase – Modifying Release Rates From Lipidic Cubic Phase Carriers By Modulating Electrostatic Interactions or Phase Transitions

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Functionalized lipidic cubic phases (LCPs) have gained considerable interest as drug delivery systems (DDS) in recent years. Among their advantages, one of the most intriguing features that make them interesting as DDS is the possibility of controlling the release of drugs based on electrostatic interactions or phase transitions. The drug release profiles can be modulated by the electrostatic interactions between the drug molecule and the charged LCP used. Modulation of the release of a positively charged drug, doxorubicin (DOX) was obtained by incorporation of small amounts of designed charged derivatives of the monoolein host lipid as additives to the otherwise non-charged LCP. By design, the charged hydrophilic headgroup of the additive was exposed to the aqueous channels, whereas the hydrophobic tail is incorporated in the lipid bilayer and forms an integral part thereof. The release rates of drugs are strongly dependent on the structure of the lipidic mesophases. The drug release characteristics from H2 and V2 mesophases ensures that the symmetry of liquid crystalline structures may be used to control the drug release rate. Transport properties within hexagonal (H2) phase are more strongly affected by the interaction of DOX with the carrier than it was observed in the cubic (V2) phase. The monoolein derived hexagonal phase and its nanoparticles showed very slow discharge profile from H2 without an initial burst release of the drug, contrary to the one observed for the V2 phase. The release profiles of the DOX-loaded hexosomes showed that most of the drug remained encapsulated within the lipid bilayers of the GMO derived hexosome.

Spectroelectrochemical Investigation of Cholesterol Oxidase from *Streptomyces Lividans* at Different pH

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Cholesterol oxidases (ChOxs, EC 1.1.3.6) are monomeric flavoenzymes.[1-6] After glucose oxidase ChOxs are the second most frequently used enzymes in clinical diagnostics.[1] ChOxs catalyze the oxidation of cholesterol to 5-cholesten-3-one and its subsequent isomerization to 4-cholesten-3-one. There are two different types of ChOx, one with a covalently linked flavin adenine dinucleotide (FAD) cofactor and one with a strongly but non-covalently bound FAD cofactor.[6] ChOx from *Streptomyces lividans* (*Sl*ChOx) with a molar mass of about 58 kDa contains a non-covalently linked FAD-cofactor. For an application in electrochemical biosensors it is important to know details of the redox process and the redox potentials. Therefore *Sl*ChOx was investigated by spectroelectrochemistry between pH 5 and pH 9. For the measurements the enzyme was reduced stepwise in the presence of different one and two electron redox mediators. As the mediator methylene blue contributes not significantly to the spectral changes of the enzyme between 350 nm and 500 nm, for each pH value also one measurement with methylene blue only was performed. In the measurements a stable flavosemiquinone radical anion was detected. The formation of a stable flavosemiquinone radical anion was reported previously for different types of ChOxs. Based on global analysis the first (Fl(quinone)/Fl(semiquinone radical anion)) and second redox potential (Fl(semiquinone radical anion)/Fl(hydroquinone anion)) of *Sl*ChOx were determined between pH 5 and pH 9. Also the pH dependent midpoint potential was detected. The pH dependency of the redox potentials will be discussed and compared with those published previously for other types of ChOx, and further flavoenzymes, which also comprise stable semiquionone radicals.

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Electrochemical Cysteamine Assay as a New Approach for Studying the Reactivity of Bioactive Ligands

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A cysteamine assay was designed for the study of reactivity of a broad spectrum of bioactive compounds with primary amines and thiols, which are found in the side chains of amino acid residues of proteins. Modification of these residues often modulates the function and activity of the proteins under investigation [1].
The detection principle is based on the chronopotentiometric stripping (CPS) analysis of cysteamine (**1**) as the molecular probe. This technique was used to observe the redox transformations of both sulfhydryl and amino groups of **1** in a single scan. The decrease in CPS signals of **1** was utilized to monitor its reactivity towards the following metabolic products and electrophiles: methylglyoxal (**2**), sulforaphane (**3**), 4-hydroxynonenal (**4**) and nitrooleic acid (**5**).

LC/MS was used to verify electrochemical data and identify reaction products. The stability and reactivity of various bioactives under different conditions was investigated by this assay under aqueous conditions for the first time.

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Amyloid beta peptides time-dependent structural modifications: AFM and voltammetric characterization

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Alzheimer's disease (AD) is an age-related disease affecting millions of people worldwide, characterized by progressive cognitive deterioration. The predominant constituents of senile plaques are the amyloid beta (Aβ) peptides $A\beta_{1-40}$ and $A\beta_{1-42}$, which are composed of different regions: the C-terminus domain is hydrophobic and adopts a β-strand structure, while the N-terminus region enables the formation of both α-helical or β-strand configurations. However, in small amount, shorter Aβ peptides were also found both in amyloid plaques and cerebrospinal fluid of AD patients, and, under specific fibrilization conditions *in vitro*, the short $A\beta_{1-28}$, $A\beta_{12-28}$ and $A\beta_{14-28}$ peptides were found to form aggregates and fibrils. In this context, short Aβ peptides may present high analytic interest for understanding the aggregation, reactivity and toxicity of the Aβ peptides, helping to highlight the role played by different amino acid domains in the fibrilization process.

Aiming to understand how the specific amino acid domains $V^{12}HHQ^{15}$, $K^{16}LVFF^{20}$ and $I^{31}IGLMVGGVV^{40}$, with different hydrophobic characteristics, influence the Aβ peptides aggregation, the time-dependent redox behaviour and structural modifications undertaken by seven Aβ peptide sequences $A\beta_{1-40}$, $A\beta_{1-42}$, $A\beta_{1-16}$, $A\beta_{1-28}$, $A\beta_{10-20}$, $A\beta_{12-28}$ and $A\beta_{17-42}$, were investigated at carbon electrodes, using voltammetric methods and atomic force microscopy (AFM).

The presence of an intact $K^{16}LVFF^{20}$ domain, the peptide hydrophobic aggregation core, induced time-dependent aggregation and fibrilization of full length $A\beta_{1-40}$ and $A\beta_{1-42}$ peptides, and short chain $A\beta_{1-28}$, $A\beta_{10-20}$, $A\beta_{12-28}$ peptides. AFM images showed a decrease in the adsorption and/or the occurrence of aggregates protofibrils and fibrils that was correlated with the decrease in the oxidation peak currents, observed by DP voltammetry. The hydrophobic C-terminal domain $I^{31}IGLMVGGVV^{40}$ present in the $A\beta_{1-40}$ and $A\beta_{1-42}$ peptides contributed to accelerate the aggregation rate. A truncated $L^{17}VFF^{20}$ aggregation core, but the presence of an intact hydrophobic $I^{31}IGLMVGGVV^{40}$ domain at the C-terminus in the case of $A\beta_{17-42}$ peptide, led to the formation of aggregates immediately in freshly prepared solution, which did not changed with increasing the incubation time. The lack of the hydrophobic $K^{16}LVFF^{20}$ and $I^{31}IGLMVGGVV^{40}$ domains in the case of $A\beta_{1-16}$ peptide, led to absence of aggregation.

Gold nanoparticle decorated graphene nanoflakes for electrochemical biosensing of clinically significant molecules

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Nowadays, graphene-modified electrodes and their application to electrochemical sensing have attracted enormous interest [1]. The behavior of gold nanoparticle-decorated graphene nanostructures is, herein, investigated towards electrochemical sensing of clinically relevant molecules and towards the development of a DNA-based biosensor [2]. An *in situ* novel colloidal chemistry approach has enabled to achieve a high density and very uniform coverage of reduced graphene oxide (rGO) nanoflakes with the Au NPs. In more details, rGO platforms are surface functionalized with 1-aminopyrene or 1-pyrene carboxylic acid that act as heteronucleation and growing sites of the amine- or thiol-coated Au NPs of different dimensions (from 3 to 20 nm). At first, Cyclic Voltammetry (CV) and Electrochemical Impedance Spectroscopy (EIS) measurements are carried out in order to characterize the different hybrids. Then, the materials are applied as electroanalytical sensors for clinically relevant molecules (dopamine, H_2O_2) with very promising results, comparable or even better than analogous systems reported in literature. The properties of the hybrid nanocomposite, enhanced with respect to those of the single components, are ascribed to charge transfer occurring at the heterojunction from the Au NPs to the rGO, assisted and channelled by the pyrene linker. The novel electrochemical platforms have also been evaluated for the development of a genosensor for the detection of miRNAs, a family of small, non-coding RNAs that are considered important clinical biomarker candidates for many diseases, including cancer [2]. Results on spiked serum samples are reported. A LOD of 0.7 amol in 10 μL, with an average percentage standard deviation (RSD) of 13%, in a range of 1 – 5000 pM, has been achieved.

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Tethered Bilayer Lipid Membranes – a Comprehensive Tool for Electrochemical Studies of Pore-Forming Toxins

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Formation of plasma membrane is considered as a crucial event during evolution and life, as known today, would not be possible without them. To investigate this complex membrane environment of lipids and membrane proteins significant progress has been made to model native membranes. The most common artificial systems include lipid monolayers, lipid vesicles, and supported lipid bilayers (SLBs) [1]. Distinct group of solid supported bilayers, tethered bilayers (tBLMs), are considered as perspective experimental platforms for membrane biosensors [2]. In particular, the modulation of the ionic conductance of tBLMs may be utilized to develop biosensors of the membrane damaging agents such as pore-forming toxins (PFTs) [3].

We have used the tBLM platform for detection, visualization and investigation of the mechanism of action of cholesterol dependent cytolysins (CDCs) – a class of PFT produced by a large number of Gram-positive pathogens and considered as virulence factors promoting bacterial invasion and infection. Most of the experimental data was obtained by particularly applying electrochemical impedance spectroscopy (EIS) and atomic force microscopy (AFM) techniques. tBLM modification in aqueous environment has been performed, using Methyl-β-cyclodextrin (MβCD) and cholesterol complex, to achieve picomolar sensitivity of CDC detection. Moreover, the additional receptor – human CD59, essential for some of the CDCs to reconstitute, was successfully attached to tBLM, enabling a better insight into different mechanisms of how the same toxin can operate.

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Underpotential Photoelectrooxidation of Water by SnS_2-Laccase Cocatalysts on Nanostructured Electrodes with Only Visible Light Irradiation.

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Clean energy vectors are needed towards a fossil fuel-free society, diminishing both greenhouse effect and pollution. Water splitting is a clean route to obtain hydrogen, the cleanest fuel; although even after decades of research it is not yet efficient enough to overcome its high overpotentials.

One of the recent advances has considered the combination of inorganic catalysts with biocatalysts searching for new advantages and alternative approaches to this problem. Recent works showed how to force a multi-copper oxidase, laccase, to oxidize water under relatively high applied potentials [1] and improve the energy applied with assistance of light energy absorbed by an In_2S_3 interface [2]. In the present work we have tested a different semiconductor, SnS_2, characterized by a deeper valence band, a visible light band gap of ~2.2 eV (λ= 550 nm). Preparation of a FTO electrode modified with SnS_2 and laccase allowed water oxidation at much lower applied external potential, taking better advantage of light energy. Additionally ITO nanoparticles were added to increase the SnS_2 area connected to the electrode surface and therefore available for bioelectrocatalytic water oxidation. We studied our nanostructured electrode under the effects of external applied potential to minimize the external energy input and different irradiance power from a solar simulator to increase both the photonic and Faradaic efficiency.

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Resisitive Pulse Sensors for Biosensors and Imaging Nanomaterials

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Resistive Pulse Sensors, RPS, commonly known as nanopore sensors, offer single analyte resolution. The translocation of the analyte through the pore causes a change to the conductivity of the channel. Information on the volume, concentration and charge (zeta potential) of the analyte can be quickly measured.

An objective of our research is to increase the number nanomaterials and biomarkers we can analyse simultaneously. To do this we have been developing new RPS platforms, including a 3D printed tool kit for particle analysis and embedding these sensors into nanoparticle manufacturing lines, or bioreactors for inline quality control. The materials we create and characterise range from nanomaterials, exosomes, bacteria and virus particles. We show some recent work characterising nanoparticle shape, with new methodologies to identify the particle shape, and composition when mixture of rods and spheres are both present in solution. Here we combine resistive pulse sensing with predictive logistic regression models, termed RPS-LRM, to rapidly characterise a nanomaterial's shape and size in solution. We demonstrate an RPS-LRM system capable of characterising individual nanospheres and nanorods over a wide size range and present the first method that can predict the ratios of nanospheres to nanorods within mixtures, with high accuracy, giving proof of concept for RPS-LRM to be developed as an imaging modality.

Fig 1. From left to right. Schematic of the RPS setup, example pulses and the vision of the Multipore. Design files for 3D printed particle sensors and far right, measured versus predicted rod to sphere ratio in solution measured using RPS.

Photon-Modulation Infrared Reflection-Absorption Spectroscopy (PM-IRRAS) Study of Mixed Nucleolipid/Phospholipid Monolayers Supported on Au(111) Electrodes. Molecular Recognition Capabilities.

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Nucleolipid are bioactive molecules that combine a lipid or phospholipid chain and a nucleoside or nucleotide moiety in the polar head. Nucleolipid monolayers formed in the air/water interface have demonstrated molecular recognition capabilities by the interaction with the complementary DNA base present in the subphase[1].

In a previous work, we have transferred monolayers of 1,2-dipalmitoyl-sn-glycero-3- cytidine diphosphate (16:0-CDP-DG) on gold (111) electrodes by the Langmuir-Schaefer technique, in order to characterize them by electrochemical and PM-IRRAS techniques[2]. The quantitative analysis of the PM-IRRAS bands provided the tilt angle of the acyl chains and the tilt and rotation angles of the cytosine moiety of the nucleolipid as a function of the rational potential.

This contribution contains the PM-IRRAS study of mixed monolayers of (16:0-CDP-DG) and 1,2-dipalmitoyl-sn-glycero-3-phosphocholine (DPPC) supported on gold (111) electrodes. The molar fraction used provides a more compact monolayer maintaining the molecular recognition capabilities. The influence of the presence of guanine in the subphase is also investigated.

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Plugging redox reactions into wireless devices: RFID-based biosensor tag

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Massive integration of biosensors into design of Internet-of-Things (IoT) is vital for progress of healthcare. However, the integration of biosensors is challenging due to limited availability of battery-less and wireless biosensor designs. In this work, a combination of nanomaterials for plugging redox reaction based biosensors into a battery-less RFID-biosensor tags for wireless detection is described. The design exploits silver nanoparticles as part of the RFID tag antenna.

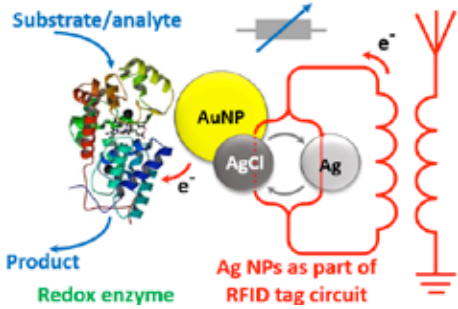

Fig. 1. Conceptual presentation of battery-less, wireless RFID-biosensor tag. AgNPs constitute a part of RFID tag antenna. An enzyme catalyzed redox reaction convert AgNPs to AgCl or AgCl to metallic Ag. This strongly modulates the impedance of the tag antenna. The impedance change is wirelessly monitored as a biosensor response.

We demonstrate that a redox enzymes can oxidize AgNPs to AgCl or reduce AgCl to Ag in the presence of enzyme substrate (analyte). The Ag reactions strongly change the impedance of the RFID tag and thus can be wirelessly monitored. The principle constitute the essence of redox-reaction-to-wireless-signal transduction.

Wireless coupling of several redox reactions have been studied and will be presented, particularly, biosensors for hydrogen peroxide and glucose. In this work, a fast and general route for converting a high number of redox reaction based biosensors into battery-less RFID-biosensor tags will be described.

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Towards detection of steroids: Electrooxidation on bare electrode materials after acid-induced dehydration

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Detection of steroids is a challenging task due to the frequent absence of effective chromophores/fluorophores thus limiting the applicable analytical methods on mass spectrometric or enzyme-based assays. Direct redox activity of steroids with not any or isolated double bonds in the steroid core on bare electrode materials is limited to far positive/negative potentials, if any, mostly in non-aqueous media, as we reviewed in [1].

Herein we present a novel electrochemical approach for detection of steroids in the region of positive potentials in mixed organic-aqueous media, firstly recognized by us for bile acids [2]. For primary bile acids, electrochemical oxidation occurs after acid-induced dehydration within potential window of bare electrode materials (*e.g.*, boron doped diamond, platinum, glassy carbon). This approach can be applied to other steroids. Based on the structure characteristics of the steroid core (*e.g.*, number and position of double bonds and hydroxyl groups, their orientation), the dehydration step and following electrochemical oxidation proceeds at different rate and oxidation potentials. The sterols with Δ5 double bond (*e.g.*, cholesterol (Ch), stigmasterol) provide the oxidation signal at *ca* +1.5 - +1.7 V. On the other hand, the sterols with Δ5,7 conjugated double bonds (*e.g.*, ergosterol, 7-dehydrocholesterol (7-DHCh) are oxidized at lower potentials of *ca* +0.8 -+1.0 V. Thus, simultaneous determination of sterols in mixtures can be performed based on different oxidation potentials. This will be demonstrated for Ch and its precursor 7-DHCh for pulse voltammetric techniques and FIA with pulse amperometric detection. This determination is helpful for diagnosis of Smith-Lemli-Opitz syndrome (SLOS) caused by an inborn mutation of DHCR7 enzyme responsible for biotransformation of 7-dehydrocholesterol (7-DHC) to cholesterol [3].

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Confinement of Bilirubin Oxidase in Biogel Matrix for Bioelectrocatalytic Oxygen Reduction

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The possibility of engineering enzymes confinement in gels opens news possibilities in bioelectrocatalysis, this is because the gel-phase can maintain the integrity of the protein molecular structure and can promote a microenvironment rich in water that allows high performance of the biocatalysts. However, when the substrates are in the gas-phase, as in the case of molecular oxygen, the biocatalysis is limited by the low diffusion capability of the substrate. Here we show a BOD-based gel (biogel) that allow efficient diffusion for oxygen as well as good adherence to carbon electrodes. The biogel is obtained by one-pot reaction, based on cross-linked bilirubin oxidase (BOD) in the gel matrix composed by sodium phosphate electrolyte, glutaraldehyde and Nafion® polymer. Based on FTIR spectromicroscopy (μ-FTIR), it was obtained a collection of chemical images of the of biogel, suggesting the formation of micro-scaled channels with random protein distribution across the biogel. In addition, it was observed that aldehyde groups from glutaraldehyde react with amino groups from lysine resides of BOD, indicating intermolecular crosslinking of the enzyme molecules. The immobilization of BOD in this matrix enabled an efficient direct electron transfer reaction from Cu-groups from enzyme to the carbon electrode. Furthermore, the biogel allows the diffusion of molecular oxygen towards BOD, resulting in an efficient and stable gas-diffusion biocathode. Thus, our results show the incorporation of BOD in biogel matrix provides a suitable and stable microenvironment for the enzymatic activity. We glimpse that these results may open a wide application range in three-phase system (electrolyte-electrode-gas) for bioelectrocatalysis.

Improving the Performance of Lactate/Oxygen Biofuel using MgO-templated Carbon coated Carbon Cloth Electrodes

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A flexible and high-performance lactate biofuel cell has been attractive attention due to it's potential to disposable electrical power source for medical and healthcare applications [1, 2]. In this study, we newly fabricated a high-power thin film-type lactate/oxygen biofuel cell. A carbon cloth was used as electrode material. A MgO-templated carbon (MgOC) particles, which has mesopores of 50 nm diameter, was coated on the carbon cloth surface to be increased the effective surface area of the electrode. In the case of bioanode, lactate oxidase (LOx) was immobilized on the MgOC-coated carbon cloth surface. 1,2-naphthoquinone or 1-methoxy PMS was used as a mediator. Bilirubin oxidase (BOD) and 2,2'-azino-bis(3-ethylbenzothiazoline-6-sulphonic acid) were immobilized on the biocathode surface. Then, a thin film-type lactate biofuel cell was fabricated by combining the bioanode and biocathode, and the output power was evaluated by electrochemical measurement. A catalytic oxidation current of the bioanode attributable to 1-methoxy PMS and LOx was observed around -0.05 V, and a maximum current density was about 50 mA cm^{-2} at 0.4 V vs. Ag/AgCl. A catalytic reduction current of the biocathode attributed to ABTS and BOD was observed around 0.5 V. The maximum current density was -15 mA cm^{-2} at - 0.2 V vs. Ag/AgCl. The maximum output power density of the biofuel cell was about 2.0 mW cm^{-2} and 4.3 mW cm^{-2} under air and oxygen flow conditions. In the present study, we also performed the 3-D impedance spectroscopy analysis of the biofuel cell in detail.

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Triple Hybrid Electric Power Biodevice for Harvesting and Storage of Chemical and Solar Energies

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The very first triple hybrid biosupercapacitor has been developed and characterized. First, the biodevice is hybrid because it can simultaneously convert both solar and chemical energies into electric energy. The biosupercapacitor is double hybride since it is able to store electric energy in the same unit used for conversion. The biodevice is triple hybrid since, in addition to an enzymatic supercapacitive cathode, it also consists of bi-enzyme based supercapacitive anode for conversion of different biofuels.

Indium tin oxide nanoparticles were used to nanostructure flexible indium tin oxide electrodes. Nanostructuration helped to immobilize significant amount of different biocatalysts and to promote the ability to store charges not only electrostatically (due to high double-layer capacitance), but also electrochemically. The electrochemical storage was achieved using poly(3,4-ethylenedioxythiophene) as the conductive polymer, which has a high storage capacity through redox processes and forms a highly porous, highly capacitive amorphous film. An Os-complex based redox polymer was used as the immobilization matrix and redox mediator to wire three anodic biocatalysts to the electrode surface. Lactate and glucose oxidases were co-immobilized as anodic elements to utilize both lactate and glucose present in sweat, whereas thylakoid membranes were immobilized as the anodic photocatalyst to utilize ambient light. The developed solar/chemical bioanode was further combined with a bilirubin oxidase modified cathode to form the triple hybrid biodevice. A portable prototype of biosupercapacitor was designed using a 3D printer and tested, as the first step to show proof-of-concept for triple hybrid wearable electric power biodevices.

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Novel Nanocomposite Based on Poly (Brilliant Cresyl Blue)-Deep Eutectic Solvent/Carbon Nanotubes for Biosensing Applications

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Enzyme-modified electrodes with enzymes incorporated onto nanostructured electroactive polymers and/or carbon nanomaterials have been widely developed using the layer-by-layer strategy and applied as biosensor platforms [1,2]. The preparation of redox polymer films, as redox mediators, in deep eutectic solvents (DES), a new class of green solvent, enhances the polymerization rate leading to the formation of higher molecular weight, more stable polymers than those formed in aqueous electrolytes [3]. The combination of carbon nanotubes (MWCNT) with polymers is beneficial for biosensor applications, their combination leading to improved performance.

The development, characterization and optimization of a novel platform based on poly(brilliant cresyl blue)-DES/MWCNT for glucose oxidase (GOx) and tyrosinase (Tyr) immobilization is described. The use of the DES ethaline (choline chloride: ethylene glycol) contributed significantly to better polymer film growth and the resulting film morphology led to an enhanced voltammetric response and increased biosensor sensitivity compared with the polymer film produced in aqueous solution. The electrochemical performance of the biosensor and the biosensor characteristics including enzyme kinetics, influence of pH, limit of detection and sensitivity was evaluated for glucose and catechol as enzyme substrates.

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A three-electrode µL-cell for green (bio-)electroanalysis with common macro- and microelectrodes

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Electrochemical cell miniaturisation is a green chemistry solution to reduce reagent consumption and waste generation. Recent reports introduced practical solutions with a tapered glass tube with agar gel seal (Skalová *et al*., 2018) or an inverted Ω-shaped Pt-tube as the sample holder (Sripirom *et al*., 2018) as well as a hanging droplet cell for microvolume electroanalysis (Jimenez *et al*., 2015). Though the suggestions worked well, they require adapted electrodes of microscale dimensions, which is against universal applicability. Here, an advanced three-electrode electrochemical mini-cell with suitability for green voltammetry with standard working electrodes (WE) is proposed. A platinum tube (Pt-T, ⌀ 4 mm × 5 mm) functions both as the counter electrode and the electrolyte container when placed on the sensing platform of inverted WEs. A fritted mini Ag|AgCl reference electrode completes the electrochemical mini cell by contacting the electrolyte through the top opening of the Pt-T cell. The Pt-T mini-cell produced typical cyclic voltammograms for 30 μL hexacyanoferrate(III) with all types of WEs tested, i.e. Pt, Au, boron-doped diamond and glassy carbon disk macroelectrodes, a cylindrical pencil lead electrode and Pt disk microelectrodes. The mini-cell was in trials with biologically relevant analytes also successful with drug, hormone, antioxidant and redox label voltammetry, the operation of a glucose biosensor and quantitative differential pulse voltammetric paracetamol analysis in standard addition mode. The significant technical advantages of the mini-cell are its cheapness and simplicity of construction, unlimited reusability, wide analyte adaptability and the ability to incorporate practically all standard macro- and microelectrode designs. Consequently, the mini-cell is not only a valuable option for voltammetry work in general but more importantly it facilitates electrochemical analysis of samples with a mass or volume constraint or when the chemicals involved are costly. Obviously, the μL electrochemical cell described is a 'green' electroanalytical tool that supports eco-friendly practice in research, commercial and teaching laboratories.

High-Performance Gas Breathing H_2/O_2 Biofuel Cells Comprising a Hydrogenase/Polymer Based Bioanode

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Limitations in the use of hydrogenases for energy conversion systems, i.e. H_2 oxidation bioanodes, based on a fast deactivation of such enzymes by O_2 and at high potentials could already be overcome by the incorporation of such biocatalysts into redox polymers exhibiting low potentials and thus being able to reduce O_2 while simultaneously acting as a Nernst buffer for the redox protein.[1] Yet, for H_2/O_2 biofuel cells based on a rather conventional design, the diffusional mass transport of the gaseous substrates is typically the rate limiting step due to their low solubility in aqueous media. Hence, the fabrication of high performance bioelectrochemical devices is still a major challenge. The use of gas diffusion electrodes is a promising approach to overcome this limitation. These electrodes exhibit a three-phase boundary at the electrolyte-catalyst-gas interphase and thus ensure a high local substrate flux at the active sites even under passive conditions. The combination of the benefits of redox polymer/enzyme electrodes, i.e. protection from O_2 and high potential deactivation, with the concept of a gas diffusion electrode (enhanced mass transport) is supposed to ensure high current densities which are desired for the fabrication of high performance biofuel cells.
In this contribution, we present a dual gas-breathing H_2/O_2 biofuel cell equipped with a H_2 oxidizing bioanode modified with redox polymers/hydrogenase layers and a conventional O_2 reducing, bilirubin oxidase-based biocathode operating in a direct electron transfer regime. For the biofuel cell, an open circuit voltage of 1.13 V and an outstanding power output of 3.6 mW cm^{-2} at 0.7 V was obtained, setting a benchmark for redox polymer/hydrogenase based biofuel cells.[2]

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Graphene-Sulfite Oxidase Bioanodes for Enzymatic Biofuel Cells

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Enzymatic biofuel cells (EBFCs) are environmentally friendly electrochemical cells that produce electricity using enzyme catalysts. An EBFC consists of a bioanode in which enzymes catalyze the oxidation of fuels such as sugars, ethanol and sulfite, and/or a biocathode using dioxygen-reducing enzymes. Most EBFCs generate energy from abundant fuels without using noble metals. However, EBFCs are not sufficiently stable and hampered by limited power densities. Graphene possess high conductivity and surface area,[1] and thus the introduction of graphene offers a strategy to promote the performance of bioelectrode.

We have developed a bioanode based on human sulfite oxidase (HSO) electrostatically immobilized on a graphene-modified carbon paper using polyethyleneimine (PEI), Fig. 1a. HSO catalyzes the oxidation of sulfite to sulfate via direct electron exchange with the electrodes.[2] Electrocatalytic performance of the bioanode was further enhanced by electroreduction of graphene oxides in the graphene matrix. A stacked EBFC constructed by combining the HSO bioanode and a commercial platinum cathode is found to deliver an open circuit voltage of 0.64 ± 0.01 V, and a maximum power density of 61 ± 6 μW cm^{-2} which is seven times higher than the earlier reported value,[2] Fig. 1b and c.

Fig. 1 Illustration of (a) the graphene-HSO bioanode and (b) the EBFC. (c) Polarization and power density curves of the EBFC fueled with Tris-acetate buffer (750 mM, pH 8.4) containing 25 mM Na_2SO_3 (2.0 mL min^{-1}) at the bioanode and O_2 (100 mL min^{-1}) at the cathode at 30 °C.

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Enzyme Biosensing of a Urinary Biomarker with Redox Polymer-Based Readout and Allosteric Activation

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p-Hydroxyphenylacetate (*p*-HPA) is a product of human metabolism and as such at discrete levels a component of urine. Abnormal urinary *p*-HPA concentrations indicates intestinal overgrowth of bacteria ('dysbiosis') and may be a sign of e.g. brain disfunction.[1,2] Hence, *p*-HPA became an important candidate as urinary disease biomarker. Chromatographic separation with spectroscopic end-column detection is currently most common for the analysis of urine *p*-HPA. Obviously, this practice is not feasible for personal urine testing at home or in medical offices of general physicians. A novel electroanalytical biosensor assay for *p*-HPA detection that uses the power of the compound to allosterically activate the turnover rate of a bacterial reductase for the conversion of substrate NADH into NAD^+ was recently reported.[3] Here, the advanced redox polymer-based biosensing of *p*-HPA will be introduced and it will be confirmed that the approach indeed allows quantification of the biomarker far below the micromolar level in spiked phosphate buffer solution and even in artificial urine samples. Electrochemical *p*-HPA biosensing has the potential to be integrated into simple handheld glucose monitor-type devices, which ultimately may facilitate health care applications of the strategy in the form of easy and quick portable screens of the compound in real urine samples, autonomous at home or in health centers.

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Enzymatic and Photocatalytic Amplification in Amperometric Immunoassays with Nanobodies

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Detection of protein biomarkers in trace amounts is of crucial importance for medical diagnosis. Particularly, a zoonotic disease human toxocariasis lacks an adequate diagnostic method despite of its broad distribution over the world. The disease is frequently underdiagnosed due to often unclear clinical picture and limitations of the currently used serological analysis. The direct detection of a protein marker that is excreted by the infection in its active stage could greatly improve the diagnosis but stays illusive due to insufficient sensitivity in conventional ELISA.

In our work we used single domain fragments of antibodies (nanobodies) from camelids [1] that were engineered and produced in *E. coli* to tackle specificity and sensitivity in ELISA. To further improve the sensitivity we applied modified magnetic beads and amperometric detection coupled to enzymatic and photocatalytic amplification strategies [2, 3]. In optimized conditions with the double amplification strategy, our method could detect trace concentrations of the specific protein marker of Human toxocariasis with the LOD at least two orders of magnitude lower compared to previously reported assays and conventional ELISA made with the same immunoreagents. Pushing sensitivity limits in detection of biomarkers without need of costly equipment is crucial for development point-of-care diagnostic tools. Nevertheless, multiple factors beside the sensitivity should be addressed to get actual improvement in LODs by such ultrasensitive assays.

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Bioelectrochemical herbicide sensor based on photocurrent inhibition of *Anabaena variabilis*

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Herbicide contamination is a crucial issue for environmental and safety reasons, therefore a continuous monitoring of the water bodies is needed. Thanks to their sensitivity, and immediate response, amperometric biosensors represent a very interesting tool for this purpose. When whole-cells of photosynthetic microorganisms are employed as bioreceptors, this kind of biosensors shows a higher stability and durability compared to the ones employing biomolecules like enzymes, photosystems or chloroplasts. In addition, extraction and purification of such molecules is usually expensive and time-consuming.
When whole-cells sensors have been employed, the biosensor mostly relied on the amperometric measure of photosynthetically generated oxygen. Unfortunately, this parameter is only an indirect measure of the photosynthetic activity, as it can be influenced by environmental factors. Also, the use of gas-diffusion membranes might arise fouling issues.

To tackle the presented issues, we developed a bioelectrochemical herbicide sensor based on the direct inhibition of the photocurrent generated by the cyanobacteria *Anabaena variabilis*. Living cells have been entrapped on a carbon felt electrode using an alginate polymer, and the redox mediator p-benzoquinone has been employed to sustain the electron transfer. The addition of activated carbon to the alginate layer was also tested.

Commonly used herbicides such as Atrazine and Diuron have been employed as model inhibitors, and their influence on the signal has been evaluated. These compounds are able to bind the plastoquinone reaction center of the photosystem II, thus blocking the photosynthetic electron transport chain.

Each time the tested herbicides were injected, an immediate concentration-dependent decrease of the photocurrent was observed, achieving a linear response up to 1 μM, and opening for the on-line monitoring for the inhibition process.

Electrochemistry of Electrophilic Fatty Acids

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Inflammatory as well as metabolic reactions are associated with increased levels of reactive oxygen and nitrogen species, *e.g.* nitrogen oxide and dioxide radicals, hydroxyl radicals, hydrogen peroxide and superoxide [1]. These conditions are associated with the generation of biologically relevant electrophiles, which can accept electrons from electron-rich donor molecules (nucleophiles).
Nitro-fatty acids play an important role in inflammation and cellular stress response, with high application potential in the field of novel electrophilic pharmacophores. In this report, we focused on evaluating the redox behavior of nitro-oleic acid (Fig. 1) and its ability to bind to proteins. The nitro group in nitro-oleic acid undergoes electrochemical reduction at around –0.75 V (*vs.* Ag|AgCl|3M KCl) at pH 7.4 in an aqueous milieu. Based on observations of the NO_2 reduction process, we can quantitatively monitor the stability or reactivity of nitro-oleic acid in comparison to oleic acid, serving as the negative control.

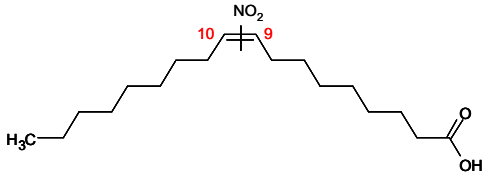

Fig. 1. Structure of nitro-oleic acid.

The data presented here are the first mechanistic view into the reactivity of nitro-oleic acid and could be used in further molecular studies focusing on other electrophiles with high biological relevance.

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Optimization of a biocompatible, implantable bioanode based on glucose dehydrogenase

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Recent decades have witnessed tremendous development in electronics, and wireless technology for Implantable Medical Devices (IMDs). Active IMDs rely on a sustained power supply, and many power sources for IMDs have been investigated. Rechargeable batteries are only appropriate for devices consuming tens of microwatts. Biochemical energy harvesting using *enzyme based biofuel cells to convert chemical bond energy to electrical energy are an important alternative.* Glucose is the most commonly used fuel in biofuel cells, and glucose oxidase (GOx) the most studied. However, the generation of potentially toxic H_2O_2 *in vivo* is unsatisfactory. Alternatively, we studied glucose two glucose dehydrogenases (GDHs): NADP-GDH and FAD-GDH. Two mediators were investigated: Poly(methylene green) and (PMG), Poly(Acrylic) Acid. Both mediators enable cofactor (NADP, FAD) regeneration at the electrode surface. Bioanodes were created by a mechanical compression of GDH, cofactor and CNTs. To achieve biocompatibility, the bioanode surface was covered with a Polyvinyl Alcohol (PVA) layer. Performance showed that that the NADP-GDH based bioanode was capable of three weeks *continuous* generation of a current density of $50\mu A/cm^2$. A first in animal study using tissue implantation in a rat showed good bioanode biocompatibility, with no inflammation or rejection reaction. Furthermore, explanted bioanodes demonstrated activity retention a month after continuous *in vivo* operation.

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Study of electrochemical melting/hybridization of DNA self-assembled monolayers by Fluorescence Microscopy

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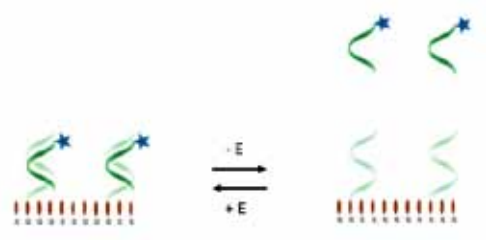
The control of the interfacial behavior of double stranded DNA by means of an electric field is of interest for sensing applications such as the detection of single nucleotide polymorphisms (SNPs). An SNP is the variation of a nucleotide at a single position in a DNA sequence among individuals. The detection of SNPs is of importance as SNPs are associated with various human diseases.

In this work we study the electric-field induced melting/hybridization of DNA by electrochemical in situ fluorescence microscopy. Electrochemistry is used to induce melting/hybridization of DNA, while fluorescence measurements are employed to detect the associated melting/hybridization events. For this purpose, fluorescently labeled DNA sequences are immobilized at gold electrodes as self-assembled monolayers (SAMs), the probe being thiolated and the complementary strand bearing a fluorescent label, in both instances at the 5'-extremity.

We present the results of a comparative study of perfect match and SNP-containing target sequences (e.g. SNP 309T>G of the human *MDM2* gene). The presence of a mismatch in a sequence significantly reduces the stability of the DNA duplex, therefore making it more susceptible to the influence of an electric-field. We studied the electrochemical melting/hybridization of double stranded DNA and present the influence of various parameters, such as the applied potential, time and ionic strength of the measurement buffer.



Surface-Confined Electrochemiluminescence Microscopy of Cell Membranes

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The development of novel single cell microscopy approaches continually opens new perspectives in bio-diagnostics. By labelling membrane proteins with fluorophores, photoluminescence (PL) has become the standard of single cell microscopy techniques. Despite the high sensitivity of PL methods, those techniques still present some inconveniences (e.g. high background due to autofluorescence). In recent years, the employment of chemiluminescence and bioluminescence methods has overcome PL challenges. In particular, electrogenerated chemiluminescence (ECL), thanks to the dual nature of electrochemical and photophysical phenomenon, offers many advantages: a higher selectivity, a very low background, and electrochemical control. The coreactant-based ECL system, $[Ru(bpy)_3]^{2+}$/tripropylamine , operating in buffered aqueous solutions, has permitted the wide diffusion of ECL as a bioanalytical technique.

We report here the development of ECL as a surface-confined detection technique to image single cells and their membrane proteins. Labeling the entire cell membrane allows to demonstrate that, by contrast with fluorescence, ECL emission is only detected from fluorophores located in the immediate vicinity of the electrode surface.[1] Such a result is intrinsically associated to the unique ECL mechanism and is rationalized by considering the limited lifetimes of the electrogenerated coreactant radicals. The resulting ECL microscopy permits the observation of surface details not resolved by classic fluorescence microscopy. Then, to present the potential diagnostic applications of our approach, we selected carbon nanotubes (CNT)-based inkjet-printed disposable electrodes for the direct ECL imaging of a labeled cellular membrane in both reflection and transmission modes.[2] The presented surface-confined ECL microscopy should find promising applications in ultrasensitive cell assays.

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Site Specific Avidity Binding Tags based on Intrachain Histidine Motifs

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The introduction of intrachain affinity tags in solvent exposed secondary structures of proteins is attractive for site-specific labelling and isotropic immobilization. The simple introduction of such sites opens the possibility for site selective bioconjugation toward protein labelling or immobilization on surfaces like electrodes. The affinity binding of the intrachain tags is however, often of insufficient strength which limits its widespread application.
Here we show that the extension from single to double intrachain His-tags strongly enhances the binding interaction due to avidity. When using a double His-tag for protein binding on Ni-NTA monolayers, protein dissociation from the surface becomes irrelevant. In addition, tris-Ni-NTA ligands form strong one-on-one pairs, allowing for site selective bioconjugation. The avidity effect described here results in dissociation constants reaching down to the picomolar range while minimal alterations of the protein structure preserve the full enzymatic activity. This approach presents a simple avidity tag with potential impact in both protein labelling and protein immobilization.

Electroenzymatic NADH Recycling for Selective In-Flow Biocatalytic Reductions

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Biocatalysts such as oxidoreductase enzymes offer exquisite selectivity under mild reaction conditions, but have proved difficult to exploit in electrosynthesis for fine chemical production. Many of these enzymes require stoichiometric hydride transfer from the expensive biological NAD(P)H cofactor, which means that their use is only viable when they are coupled with an efficient cofactor recycling method. Electrochemically-driven regeneration of the NADH cofactor is possible but problematic at unfunctionalized electrodes due to formation of non-active forms of the reduced cofactor.

Here, we present an electrochemically-driven NADH regeneration system with NAD^+-reductase modified carbon electrodes, offering perfect selectivity for the active NADH cofactor. We show that we can immobilize this enzyme onto carbon interfaces and that it is electrochemically active at much more modest potentials than using unmodified electrodes. We demonstrate this recycling system as a modular approach to biocatalytic reductions, coupling NADH-dependent reductases onto the same carbon interface for selective C=X bond reductions (Figure 1). The catalytic activity of this bioelectrocatalyst is enhanced when used within an electrochemical flow cell and is stable over many days of continuous use.

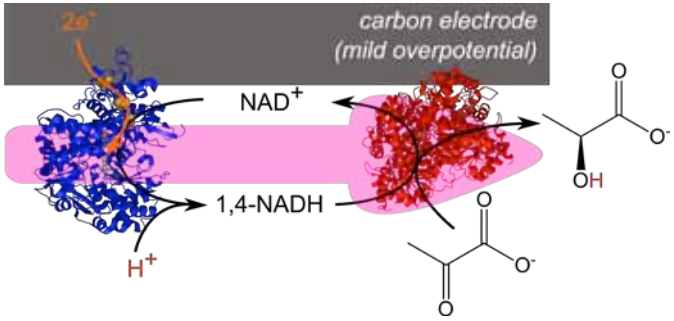

Figure 1 - Example biotransformation with an NADH-dependent reductase (lactate dehydrogenase, red), coupled with the electroenzymatic NADH-regeneration system by feeding electrons directly from an electrode to an NAD+-reductase (blue). The substrate concentration is maximized in high flow conditions (purple).

Biological origins of cyanobacterial exoelectrogenesis

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Certain cyanobacteria exhibit the ability to export electrons outside the cell, called exoelectrogenic activity. There is great potential to harness cyanobacterial exoelectrogenic activity in biophotovoltaic systems to renewably generate electricity, but power outputs remain low. Further, the biological mechanisms of cyanobacterial exoelectrogenic activity are poorly defined. This research aims to increase understanding in order to identify a cyanobacterial mutant with heightened exoelectrogenic activity and the optimal growth conditions to achieve increased peak power output of biophotovoltaic systems. Previous research systematically compared the photoelectrochemistry of photosystem II protein films and biofilms of the model cyanobacteria *Synechocystis* sp. PCC6803, both using inverse opal indium-tin oxide (IO-ITO) working electrodes. These exhibited markedly different photocurrent profiles, and evidence was obtained for the presence of a soluble endogenous electron mediator that is secreted from the cell in response to light. Presented here is a complementary study of the photoelectrochemistry of intermediate layers of *Synechocystis*: whole-cells lacking the surface-layer, spheroplasts, and isolated thylakoid membranes. Films of *Synechocystis* material were formed on IO-ITO electrodes and 3-electrode chronoamperometry experiments were conducted under light and dark cycling regimes. The cytosolic material was additionally analysed by cyclic voltammetry. Comparison of these photoelectrochemical data to those obtained in previous research provides further evidence of a soluble endogenous electron mediator, as well as additional insights into the biological origins of cyanobacterial exoelectrogenesis.

Redox modified gold clusters for ET enhancement

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Monolayer-protected gold nanoclusters, gold crystals consisting of 13 to ca. 200 atoms, modified with corresponding thiol derivatives can have important applications across many fields of biomedical, catalytic and sensing [1]. The size of gold clusters implies interesting behavior where single-electron quantization effects are involved [2, 3]. Below a certain core size, the monolayer-protected clusters exhibit double layer charging as a series of oxidation states. For the smallest clusters (below 1.5nm) a molecule-like behavior is observed as HOMO-LUMO gap [4]. These properties of gold clusters are different from those of bulk metal, gold complexes and common sizes of nanoparticles.

Gold nanoparticles covalently bound to the self-assembled monolayers attached to the electrode can act as rely units for long-range electron transfer. Redox reaction of the probe in the solution or surface-confined probe can be facilitated by the presence of the hybrid junction: electrode-alkanedithiol-gold nanoparticle-redox probe. The role of nanoparticles in this process is to provide electronic energy levels for the tunneling electron that temporarily resides on the nanoparticle so that ultimately the electron can move into the electronic levels of the metallic electrode [5].

In this project we present an investigation of the electron transfer rate using different monolayer protected gold clusters and gold nanoparticles for comparison modified with surface-confined redox probes. We have employed a redox reaction of surface-confined hydroquinone modified with alkanethiol of different length.

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Biosensor for trimethylamine –N-oxide with an chimeric enzyme

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We present electroanalytic studies of TorA and also the development of an electrochemical biosensor system for determination of TMAO. The TMAO reductase TorA from *Escherichia coli* catalyzes specifically the reduction of TMAO to trimethylamine. It is a molybdoenzyme hosted in marine and most enterobacteria. Recently active site variants of this enzyme were generated including chimeric TorA corresponding to the apoform of TorA from *E.coli* reconstituted with the molybdenum cofactor of other enzymes. The sensor employs an active chimeric TorA-FDH, that contains the molybdenum cofactor from *R. capsulatus* formate dehydrogenase (FDH). TorA-FDH was immobilized onto a carbon electrode and protected with a dialysis membrane. The mediated biosensor can be operated at an applied potential of -0.8 V Ag/AgCl (1M KCl) under ambient air conditions because of deoxygenation by GOD and catalase. The biosensor with immobilized TorA-FDH covers the diagnostically relevant measuring range of 2-110 µM TMAO. Furthermore, the sensitivity is almost similar for TMAO in buffer and in spiked serum samples and detection of TMAO in human serum can be performed without interferences from the complex serum matrix. Elevated levels of trimethylamine N-oxide (TMAO) were reported to be related to several disease states. The described enzyme-based biosensor thus represents a promising approach for the future determination of TMAO levels in human blood samples.

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Flexible nanoporous gold film based enzymatic biofuel cells for wearable power sources

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There is growing interest in fabrication of wearable biosensors for personal healthcare. Wearable enzymatic biofuel cells (EBFCs) using sugars in tears or sweat are potential, miniaturized power sources for wearable microelectronics. Those applications require flexible, biocompatible and high-surface-area electrodes. Nanoporous gold (NPG) can be an excellent candidate meeting all the requirements[1]. We fabricated mechanically stable and flexible NPG electrodes by electrochemically dealloying Au/Ag alloy films that had been sputtered on plastic membranes. Enzymes were then immobilized on the NPG electrodes and characterized for EBFC applications.

Flexible NPG based lactate/O_2 EBFCs on commercial contact lenses registered a power density of 1.7 ± 0.1 μW cm^{-2} in air-equilibrated artificial tear solutions, which is slightly lower than that obtained in phosphate buffer solution (PBS) (2.4 ± 0.2 μW cm^{-2})[2]. A quasi-solid-state and flexible EBFC can be worn on skin, consuming the reservoir of fuel stored in a hydrogel electrolyte, eliminating the need for sugar from sweat[3]. Such a device can also function as a self-powered biosupercapacitor, delivering pulses for over 600 cycles, with a power density over 10 times higher than that from the EBFC alone.

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Remediation of Cd Contaminated Soil based on Bioelectrochemical Reduction of Sulfate

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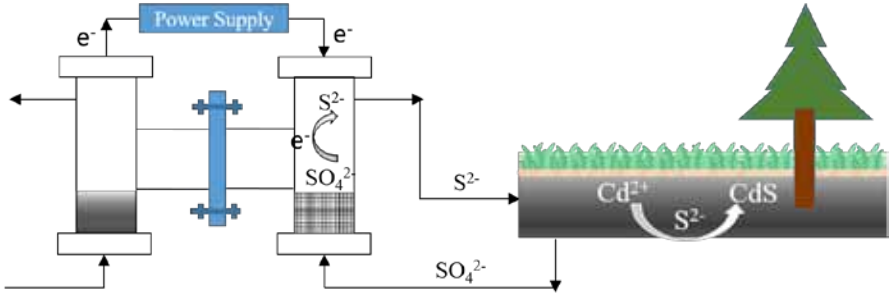
Heavy metal contamination in soil has potential serious harm on human health. Stabilization metal ion with sulfide is a useful strategy to eliminate the migration and transformation of metal in polluted soil. Although most of metal sulfides are chemically stable, sulfide containing stabilization agent is difficult to prepare, storage and transportation. In this work, a bioelectrochemical sulfate reduction reactor was coupled with Cd contaminated soil remediation. In this coupled system, sulfate was bioelectrochemically reduced to sulfide and then stabilized Cd species into CdS. In the bioelectrochemical system, sulfate reducing bacteria using the electrode as an electron donor via hydrogen produced on the electrode or electron transfer. Sulfate containing wastewater was used as the source of sulfur element. When the applied current is 45 mA, sulfate reducing bacteria can reduce 200 mg/L sulfate into 176.81 mg/L sulfide. After reduction, the produced sulfide was used as the stabilization agent and mixed with the Cd contaminated soil. Operational parameters, including mixing time, the concentration of sulfide and soils obtained from different regions, are investigated and optimized. The results shown that in this coupled soil remediation system a high Cd stabilization efficiency can be achieved in 2~3 hours. Cd stabilizing rate is positively related to the total amount of sulfide. The sulfide will combine with other metal iron such as Fe^{2+}, Cu^{2+}, Pb^{2+} in soil first before stabilizing Cd^{2+}. When the total amount of sulfide is 1.2 mg per gram of soil, Cd stabilization efficiency can achieve 50~60%.

In this study, we show a new potential application for bioelectrochemical systems as well as a novel strategy of heavy metal contaminated soil remediation.



Rapid and Sensitive Detection of *Aspergillus niger* Using a Single-Mediator System Combined with Redox Cycling

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Rapid and sensitive mold detection is becoming increasingly important, especially in indoor environments. Common mold detection methods based on double-mediated electron transfer between an electrode and molds are not highly sensitive and reproducible, although they are rapid and simple. Here, we report a sensitive and reproducible detection method specific to *Aspergillus niger* (*A. niger*), based on a single-mediator system combined with electrochemical-chemical (EC) redox cycling. Intracellular NAD(P)H-oxidizing enzymes in molds can convert electro-inactive hydroxy-nitro(so)arenes into electro-active hydroxy-aminoarenes. Since the membrane and wall of *A. niger* is well permeable to both a substrate (4-nitro-1-naphthol) and a reduced product (4-amino-1-naphthol) in tris buffer (pH 7.5) solution, the electrochemical signal is increased in the presence of *A. niger* due to two reactions: (i) enzymatic reduction of the substrate to the reduced product and (ii) electrochemical oxidation of the reduced product to an oxidized product. When a reducing agent (NADH) is present in the solution, the oxidized product is reduced back to the reduced product and then electrochemically reoxidized. This EC redox cycling significantly amplifies the electrochemical signal. Moreover, the background level is low and highly reproducible because the substrate and the reducing agent are electro-inactive at an applied potential of 0.20 V. The calculated detection limit for *A. niger* in a common double-mediator system consisting of $Fe(CN)_6^{3-}$ and menadione is $\sim 2 \cdot 10^4$ colony-forming unit (CFU)/mL, but the detection limit in the single-mediator system combined with EC redox cycling is $\sim 2 \cdot 10^3$ CFU/mL, indicating that the newly developed single-mediator system is more sensitive. Importantly, the detection method requires only an incubation period of 10 min and does not require a washing step, an electrode modification step, or a specific probe.

Improving the long-term Stability of Photosystem 1-based Photocathodes

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Photosystem 1 (PS1), one of the key protein complexes that drives photosynthesis, is able to deliver electrons at a highly negative potential upon light-induced charge separation. In consequence, PS1 has been increasingly explored for the fabrication of biophotoelectrochemical devices for energy conversion. In the development of PS1-based photocathodes, the use of Os-complex modified redox polymers to electrically wire PS1 to the electrode surface has enabled the efficient generation of photocurrents [1]. Conventionally, O_2 has been used as terminal electron acceptor, alone or in combination with MV^{2+} as redox mediator, which effectively scavenges the high-energy electrons from PS1. Although this process enables the efficient generation of photocurrent, an in-depth evaluation of the PS1-based biocathode reveals the formation of partially reduced high-reactive oxygen species under irradiation. These reactive species are responsible for a fast degradation of the photosynthetic protein, thus considerably limiting the lifetime and potential applicability of PS1-based biohybrid devices [2]. To avoid degradation of PS1 during illumination and hence to enhance the long-term stability of PS1-based biohybrid devices, possible strategies are explored to prevent the deleterious effects of generated reactive oxygen species [2, 3].

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Zinc Ion Sensitive Light-addressable Potentiometric Sensor Based on a Self-assembled Monolayer

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The zinc ion (Zn^{2+}) is one of the major metal ions in the human body involved in physiological and pathological processes, and an excess has been associated with diseases such as Alzheimer's disease and Parkinson's disease. The Zn^{2+} accumulation was also shown to be related to aged-related macular degeneration causing blindness [1]. Therefore, the detection of Zn^{2+} in biological systems is important. In this work, a label-free, biocompatible Zn^{2+} sensitive light-addressable potentiometric sensor (LAPS) was developed, which allows imaging of local zinc concentrations with micron resolution and high selectivity. A Zn^{2+} sensitive surface was obtained by modifying p-type silicon-on-sapphire (SOS) with a cyclam terminated monolayer (Fig. 1a). Cyclam is a known chelator for Zn^{2+} [2].

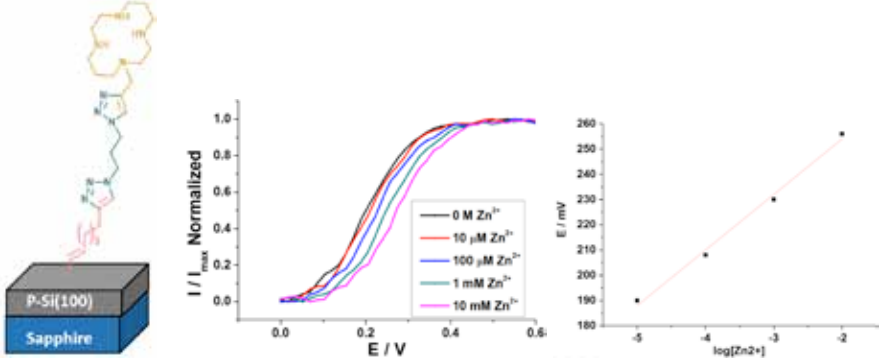

Figure 1 (a) Schematic of cyclam modified SOS; (b) LAPS I-V curves for at different Zn^{2+} concentrations; (c) calibration curve for Zn^{2+}.

The normalised photocurrent-voltage (*I–V*) curves of the Zn^{2+} sensitive LAPS showed a significant shift to positive potentials with increasing Zn^{2+} concentration (Fig. 1b), and the slope of the calibration curve was 22 mV/log[Zn^{2+}] (Fig. 1c). The sensor showed excellent reversibility of the sensor signal. The investigation of the selectivity showed that the sensor surface was insensitive to calcium ions, which makes this sensor a promising tool for biological applications. Investigations into monitoring zinc release from living cell are currently under way.

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Photoelectrochemical imaging system using α-Fe_2O_3 nanorods as substrate

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Light-addressable potentiometric sensors (LAPS) is a photoelectrochemical imaging technique, producing spatiotemporal photocurrent images of the surface potential, ion concentrations and electrical impedance. Recently, an indium tin oxide (ITO) surface without an insulator was reported as a substrate for LAPS imaging [1]. Here, α-Fe_2O_3 nanorods were investigated as a more photo-responsive and robust substrate material, providing photocurrent images of cells with high resolution.

α-Fe_2O_3 has been studied intensively for water splitting, photocatalysts, lithium batteries, and sewage treatment, owing to its earth abundance, non-toxicity, high chemical stability, narrow bandgap (1.9−2.2 eV), low cost and ease of synthesis. By using α-Fe_2O_3, a high AC photocurrent (60 nA) was obtained, which was about 9 times higher than that measured with ITO. Photocurrent imaging of a single living cell was achieved on α-Fe_2O_3 at a relatively low reference bias (0.8 V vs. Ag/AgCl), which shows a great potential to investigate cellular responses.

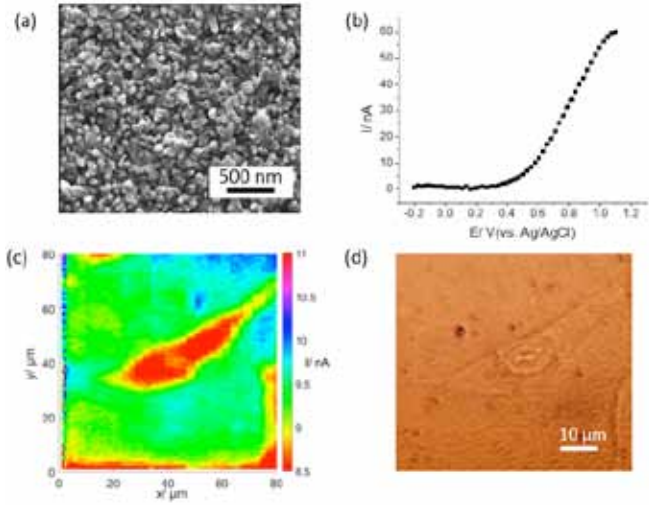

Figure 1(a) SEM image of α-Fe_2O_3 nanorods; (b) I-V curve of α-Fe_2O_3 in pH 7.4 PBS buffer solution; (c) photocurrent image of a mesenchymal stem cell on α-

EIS and SERS Analysis of Anchor Molecules for Tethered Bilayer Lipid Membrane Formation

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The tethered bilayer lipid membranes (tBLMs) are considered as perspective experimental platforms for membrane biosensors and may be suitable for a broad spectrum of biophysical experiments such as peptide/membrane interactions, protein/membrane interactions, lipid phase transitions and others. tBLMs attachment to the silver surface allows the monitoring of biologically relevant events with electrochemical impedance (EIS) and surface enhanced Raman (SERS) spectroscopical techniques.

It is known that structure of the self-assembled monolayers (SAMs) used to anchor phospholipid bilayers to surfaces affects the functional properties of the tethered bilayer lipid membranes.

To evaluate the differences in tBLM formation on flat silver surface two types of anchor molecules were chosen to form SAMs – long strand thiolipid Wilma's compound (WC14) and three different short strand backfillers 3-mercapto-1-propanol (3-M-1-P), 4-mercapto-1-butanol (4-M-1-P) and 6-mercapto-1-hexanol (6-M-1-H). EIS results showed the ability to successfully form functional SAMs and tBLMs on the plain silver surface, which leads to further experiments with this system using the SERS technique on nanostructured silver surface.

For the SERS analysis the same backfillers (3-M-1-P, 4-M-1P and 6-M-1-H) were used in a pair with WC14 compound to form SAMs with different structural properties. Analysis of SERS spectra form anchor molecules before and after tBLM formation showed, that WC14 and all of the tested short strand backfiller molecules are suitable for tBLM immobilization on nanostructured silver surface. Furthermore, by using different length backfiller molecules it is possible to control long strand anchor molecules conformation on the surface.

Poster Presentation

Highly Efficient Flavin–Adenine Dinucleotide Glucose Dehydrogenase Fused to a Minimal Cytochrome C Domain

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As the incidence of Type 1 and Type 2 Diabetes rise around the world, there is a constant need for improvement of blood-glucose monitoring tools. Some glucose biosensing devices utilize Flavin-adenine dinucleotide dependent glucose dehydrogenase (FAD-GDH) as a glucose oxidizing enzyme. FAD-GDH is an oxygen independent glucose oxidizing enzyme, and is thermostable, which renders it a good candidate for enzyme-based glucose biosensors. Electrons from the redox active catalytic site of the enzyme cannot be directly transferred to an electrode. In this study, a natural minimal C-type cytochrome domain was fused to the c-terminus of FAD-GDH, in order to achieve direct electron transfer abilities. Our new fusion enzyme introduced highly efficient direct electron transfer compared to the native enzyme, with ca. seven times higher catalytic current, three times higher k_{cat} and a lower onset potential. The onset potential of the new enzyme is about (-) 0.15 V vs. Ag/AgCl reference electrode. This low potential is beneficial since it diminishes the electrode sensitivity to known interfering molecules such as Vitamin C that is being oxidized at the electrode at higher redox potentials. To conclude, the fusion enzyme can simplify glucose biosensing, it is highly sensitive and selective towards glucose and thus can be utilized in blood-glucose monitoring and possibly with other bodily fluids.

Microwell Array Integrating Ring Nanoelectrodes for the Monitoring of Bioenergetic Responses from Single Mitochondria

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Mitochondria are major cell organelles since they are the main source of ATP owing to the oxidative phosphorylation pathway. They also play an important role into several other metabolic pathways (Krebs cycle, lipid synthesis…) and when defective, they are involved into severe pathologies (myopathies, neurological diseases, cancers...). Consequently, new methodological approaches[1, 2] are required in order to decipher the complex mitochondrial bioenergetics and to provide efficient tools for diagnosis.

In this context, we have developed microsystems, namely ElecWell platforms, which combine electrochemical [1] and optical [2] sensing abilities. These are based on the integration of platinum ring nanoelectrodes (RNE, surface: 10-15 μm^2) into SiO_2-based microwell arrays (well radius: 3 - 4.5 µm, depth: 5 µm, individual volume < 1 pL), as shown on figure 1. Similarly to ultramicroelectrodes, RNE exhibit high current density, fast response time and reduced charging currents. These electrochemical devices were characterized by cyclic voltammetry using single well or array configurations, and optimised according to COMSOL™ simulations.[3] In addition, the glass substrate of the microsystems allows the observation by microscopy of the content and reactivity within each microwell.

Then, a suspension of isolated mitochondria (from yeast cells or rodent cardiac cells) was deposited on the ElecWell array. We monitored by fluorescence the sedimentation of individual mitochondria inside the wells owing to their NADH content intensity [2]. Simultaneously, we monitored by cyclic voltammetry the variations of their oxygen consumption [3] rate in response to activators and inhibitors of respiratory chain (ethanol, ADP, antimycin A). The electroanalytic resolution offered by the ElecWell platform is nearly a few thousands of mitochondria, corresponding to the mitochondrial content of a single cell.

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Poster Presentation

Dopamine Sensing by Boron-Doped Diamond Electrode Fabricated by Microwave Plasma Enhanced Chemical Vapour Deposition Process With Linear Antenna Delivery

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Boron-doped diamond (BDD) has become a well-established electrode material for neurotransmitter sensing due to its outstanding characteristics including biocompatibility, resistance to fouling, and sufficiently wide potential window in the anodic region [1]. Within this study, both as-grown and anodically pre-treated BDD films fabricated by microwave plasma enhance chemical vapour deposition system with linear antenna delivery (MW-LA-PECVD), which enables growth with excellent homogeneity over large areas over a wide temperature range (250 to 750 °C) [2], underwent complex morphological, spectral and electrochemical characterization by scanning electron microscopy, Raman spectroscopy and cyclic voltammetry with different redox probes, along with evaluation of their sensing properties. The electrochemical behaviour of dopamine, an essential biologically active compound of which abnormal levels in physiological fluids are associated with neurological disorders, was thoroughly examined. The obtained results suggest that BDD electrode produced by the MW-LA-PECVD process is a perspective material for dopamine sensing and it may be used advantageously for many applications including fabrication of BDD-based microelectrode array devices.

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Development of Non-enzymatic Electroanalytical Methods for Detection of Cholesterol

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Cholesterol is precursor of steroids species for example bile acids and steroids hormones. Any change in metabolism of steroids in organism is signalized by decreased or increased concentration levels in blood and leads to genesis of various diseases, *e.g.* the Smith-Lemli-Opitz syndrome [1] or cardiovascular diseases. Determination of cholesterol is thus crucial to address health problems. Commonly used methods include HPLC with tandem MS detection [2] or enzymatic methods, electrochemical methods are applied peripherally due to its limited direct redox activity (see review [3]). Modified electrodes have to employed to detect cholesterol within potential window in mixed aqueous-organic media [4].
In this contribution a novel approach of direct voltammetric detection of cholesterol is presented. Strong acids (*e.g.* $HClO_4$, H_2SO_4) are used as dehydration reagents chemically activating the steroid core by dehydration reaction, similarly as in Liebermann-Burchard reaction used for spectrophotometric assay of cholesterol [3]. Two methods were developed for determination of cholesterol in solution containing 0.1 M $HClO_4$ in acetonitrile with water content 0.43% and 5% where oxidation of cholesterol occurs at the potential of +1.5 V (*vs.* $Ag/AgNO_3$ in acetonitrile). The first method is differential pulse voltammetry and the second one is flow injection analysis with amperometric detection (E_{det} +1.95 V) both using boron doped diamond electrode. Limits of detection 4.08 µM and 1.18 µM were achieved. Application aiming at determination of cholesterol in serum and food products are in progress.

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Poster Presentation

Characterisation of a Biosensor for the Real-Time Neurochemical Monitoring of L-Glutamate

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L-Glutamate is the major excitatory neurotransmitter that is released by nerve cells in the mammalian brain and has an *in-vivo* concentration of approximately 4 μM[1]. It is vital for normal brain function, including activities such as development and memory formation. It is an endogenous co-agonist of glutamatergic N-methyl D-aspartate (NMDA) receptors[2] and has been linked to disorders including epilepsy, strokes and many neurodegenerative disorders[3]. The principle objective of this research is the characterisation of a novel biosensor for the *in-vivo* detection of L-glutamate using Long Term In-Vivo Electrochemistry (LIVE). LIVE requires a microelectrochemical sensor to be implanted in a specific brain region allowing changes in the concentration of the targeted analyte to be detected with sub-second time resolution over extended periods. Experiments were performed in a standard three electrode glass electrochemical cell containing 20 mL phosphate buffer saline at pH 7.4, the biosensor, and reference (Saturated Calomel Electrode (SCE)) and auxiliary (stainless steel wire) electrodes. L-Glutamate calibrations (0-10 mM) were performed using constant potential amperometry at +700 mV *vs*. SCE. This biosensor incorporated the enzyme L-glutamate oxidase utilising a unit of activity of 100 U/mL. Several dip-coating procedures were investigated for enzyme immobilisation, including incorporating styrene, glutaraldehyde, bovine serum albumin and polyethylenimine, at various concentrations, in order to develop a simple and reproducible coating method to maximise the sensitivity of the sensor to L-glutamate. The inclusion of a poly-*o*-phenylenediamine underlayer conferred excellent permselective characteristics necessary for *in-vivo* recording. Performance factors – biocompatibility/stability, shelf-life, etc., were investigated utilising the optimum biosensor design.

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Bio-Sensing of Dopamine And its Precursors Using Boron-Doped Diamond Microelectrodes

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The use of boron-doped diamond (BDD) as an electrode substrate in both macroelectrode and microelectrode forms is now well-established due to its excellent properties.

In this work, BDD microelectrodes (BDDμEs) were fabricated by linear antenna microwave plasma enhanced chemical vapour deposition process (MW–LA–PECVD) [1]. BDD layers were deposited onto tungsten wires, which were then embedded in glass to produce a functional electrode. Structural and morphological characterization of BDDμE was performed by scanning electron microscopy and Raman spectroscopy. The layer thickness of samples was evaluated from cross section SEM observations.

The electrochemical performance of BDDμEs was carried out using cyclic voltammetry (CV). The characterization of BDDμEs was performed by recording cyclic voltammograms of 1 mM $[IrCl_6]^{4-/3-}$ redox probe in 1 M KCl in the scan rates range of 5–1000 mV s^{-1}. The evaluation of anodic and cathodic peak potential difference of $[IrCl_6]^{4-/3-}$ and estimation of effective surface area was carried out.

The redox behaviour of dopamine and its precursors, DOPA and tyrosine, on BDDμEs was tested by CV in different supporting electrolytes, such as 1.0 M KCl, phosphate buffer over the pH range of 2.0–7.4, and 20 mM HEPES buffered saline at pH 7.4, which is a biological buffer apt for all cell culture systems with a high proliferation and cell density. The results demonstrate BDDμE's attractiveness as a novel sensor for neurotransmitters and its precursors for bio-sensing.

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Poster Presentation

Electrochemical Impedance Spectroscopy (EIS) Pathogen Detection in a Microfluidic Electronic Lab-on-a-Disc (eLoaD) Platform

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Sepsis is one of the leading causes of deaths in hospitals and remains to be one of the biggest clinical challenges to date. Sepsis affects over 30 million people worldwide, with approximately 6 million people dying from the condition annually [1]. It is a time-dependent disorder and can claim lives in hours [2]. Current gold standards in microbiology labs for pathogen detection may take as long as 72 hours to return results, where 90% of all samples tested are negative [3]. SepTec is a revolutionary eLoaD capable of capturing and detecting the presence of pathogens in real-patient samples, and furthermore, characterising whether they are Gram-positive, Gram-negative and/or Fungal microorganisms. The clever use of EIS techniques allows for label-free detection of the pathogens in a user-friendly device where a sample-to-answer response is delivered in just 15 minutes. Collaborations with St. James' hospital and Beaumont hospital has allowed for a prood of concept device validation of the eLoaDs performance for use with real-samples. The electrochemical antibody-based sensor platform embedded within a centrifugal microfluidic platform requires just 2mLs of blood per test, where each eLoaD has three individual, simultaneously run, testing sites.

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Click Chemistry on Azide Functionalized Graphene Oxide

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Chemical functionalization of graphene oxide represents a challenge in chemical engineering of materials for versatile platforms with a broad range of applications. The development of electrochemical (bio)sensors requires the controlled and rational immobilization of molecules as a key step towards the enhancement of analytical performance. Click chemistry reaction represents an important strategy for the covalent linking of different compounds on the substrate bearing the complementary azide or alkyne groups [1]. The mild reaction conditions allow the preservation of the properties of biomolecules, the orientation towards green chemistry enabling a new range of biomedical applications [2]. The azide group was inserted in the graphene oxide backbone by chemical functionalization and the resulting product was characterized by Fourier-transform infrared spectroscopy, scanning electron microscopy and electrochemical impedance spectroscopy. The successful synthesis of graphene-azide platform was documented by electrochemical methods after clicking ethynylferrocene, an electroactive model molecule. The results highlighted this new approach as a versatile method for the covalent immobilization of biomolecules [3].

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Poster Presentation

Detrended fluctuation analysis of electroporation current signal from cell suspension and tissue

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Despite the success and the intensive use of electroporation in medicine and biotechnology, the understanding of the phenomenon of induced molecular paths by pulsed electric fields is still incomplete. In addition to these induced electro-molecular paths, pulsed electric fields induce at the same phenomenological time scale electrochemical processes. The common denominator of these different microscopic processes is their highly nonlinear dynamics where an on-time distinguishing method of different process is needed. We present experimental evidence for a different scaling behavior within the electroporation current signal fluctuations of cell suspension, tissue and electrochemical process. All three systems were treated in the same conditions used as in electrochemotherapy protocols. Time series data were measured and then analyzed using detrended fluctuation analysis. Detrended fluctuation analysis shows that electroporation current signal contains different scaling features related to each process with different short and long-rang correlated sub-processes. This method based on electric current recorded during electroporation is an easy way for membrane permeabilization assessment that can be implemented for a real-time monitoring tool of the phenomenon.

Spectroelectrochemical study of the adsorption and reactivity of 5-fluorouracil on Au and Ag electrodes.

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Fluorouracil ($C_4H_3FN_2O_2$, 5FU) belongs to the family of uracils and pymiridine analogues. These compounds are found in several biologically active natural products and can have considerable therapeutic potential as they are considered as privileged structures in drug discovery with a wide array of biological activities and synthetic accessibility. This class of compounds and derivatives stand out as bioactive [1], from the antiviral point of view to antibacterial. 5FU has applications in chemotherapy treatment the cancer diseases (colon and breast). The concentration of this drug in the body must be kept constant over time, and this requiring continuous monitoring. However, evidence has been found of the adverse effects of an overdosage of these drugs, which may cause intestinal damage and even death [2]. The current methods of analysis [3]) require much time, use of concentrated solvents, expensive devices and extensive protocols of extraction or separation of the sample (low selectivity for the determination of the analyte due to the presence of interferers).

Spectroelectrochemical methods such as Surface-Enhanced Infrared Spectroscopy-SEIRAS [4] combined with surface electrochemistry, provide one of the few and most powerful analytical tools to study analyte layers of adsorbed species at the nanometer scale, giving information on the adsorption properties and reactivity of molecules on metal surfaces. These techniques have shown high sensitivity, becoming useful in the study species of biological interest and the development of biosensors [5].

This communication presents the spectroelectrochemical behaviour of 5-fluorouracil on nanostructured electrodes of Au and Ag and reports additional information on the reaction mechanisms, intermediates and products of the oxidation-reduction reactions of 5-fluorouracil that could be useful for future applications.

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Poster Presentation

Why citrates shapes both gold and platinum nanoparticles. A fundamental insight for biomedical purposes.

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Citrate is one of the most used agents for nanoparticles synthesis due to its biocompatible characteristics. To name a few, its easy-removal capabilities as well as being non-cytotoxic in human serum originated citrate to be a cornerstone of a wide number of nanoparticles synthesis processes. On the other hands, gold and platinum nanoparticles (AuNPs, PtNPs) have being widely studied showing up the first to be a good candidate for drugs delivery and the latter being able to reduce intra-cellular reactive oxygen species. Due to these facts, it is crucial to understand citrate mechanisms to shape AuNPs and PtNPs, which are still poorly understood. Here, we have focused mainly on how electrochemical reactions are surface-sensitive in order to discern to which crystallographic facet the citrate has preference to be adsorbed, so that we can understand how the citrate works in colloidal AuNPs and PtNPs synthesis in water. To untangle this, we used well-defined surface electrodes to understand how citrate interacts with the different facets. Concerning this, we carried out thermodynamic analysis using a strategic combination of electrochemical experiments (CVs), FTIR spectra (IRRAS spectroscopy) and DFT calculations, on well-defined surfaces. Thank to this, the adsorption behaviour of solvated citrate on the basal planes of gold and platinum ((111), (100) (110)) in water, under different solvation conditions, are for the first time established. With all this, we determine how citrate bonds preferentially on Au(111) and Pt(111) with three dehydrogenated carboxylic groups in bidentated form, which explain why AuNPs and PtNPs grow tetrahedrycal and octahedrycal in water.

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Poster Presentation

SmartInks: Screen-Printing Platforms for the Development of Label-Free DNA/mRNA Assay

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The biosensors market is expected to reach €4bn by 2021, at a CAGR of 6.4% from 2016 attributed to the continuous technological advancements in the biosensors ecosystem. A key barrier to the development of sensors for biomedical, environmental and industrial process monitoring markets is creating rapid, low-cost strategies for the production of high-performance devices. This study shows the development of screen-printing inks that allow sensors with high level of accuracy, sensitivity and selectivity, that is mass-produced at low-cost for label-free DNA/mRNA Immobilization and Hybridization.

A single use microfluidic device based on an ultrasensitive DNA assay that discriminates down to the bacterial subtype was successfully manufactured. Figure 1 shows the amperometric i-t curves for a bare screen-printed carbon electrode (blue line), and (A) C/G (Au) 1 (B) C/G (Au) 2 screen-printed electrodes functionalised with capture, 1µM target and probe DNA labelled with platinum nanoparticles (full sandwich assay, (A) black and green line (B) purple and black line). Potential applied was −0.250 V in 0.1M H_2SO_4. The difference in current before and after the addition of H_2O_2 is displayed.

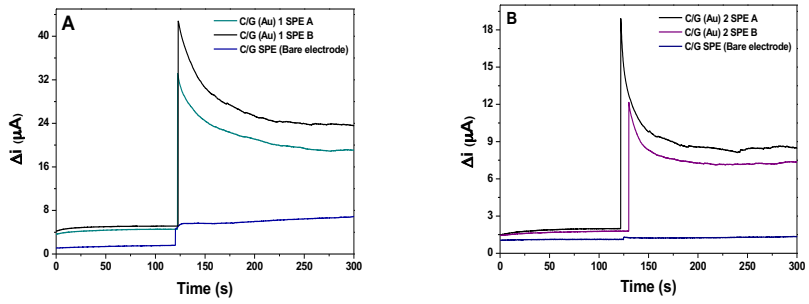

Figure 1. Amperometric i-t curves for a bare screen-printed carbon electrode (blue line), a and **(A)** C/G (Au) 1 **(B)** C/G (Au) 2 screen-printed electrodes functionalised with capture, 1µM target and probe DNA labelled with platinum nanoparticles (full sandwich assay, **(A)** black and green line **(B)** purple and black line).

Poster Presentation

An Electrochemical Study of Urate Hydroperoxide Using Carbon Microelectrodes

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Uric acid (UA) is one of the most important antioxidants present in the blood plasma. This species emerges as a final product of purine metabolism and its oxidation in biological environment produces strong oxidant intermediate species, like urate hydroperoxide (HOOU) and urate free radical. Urate hydroperoxide has a high reactivity and it is generated by peroxidases enzymes in the presence of uric acid and superoxide. The detection in biological samples is difficult due to its high reactivity and, consequently, short lifetime. This work shows an in vitro investigation involving the electrochemical response of HOOU using a carbon microelectrode (Ø = 5 μm) electrochemically activated [1]. The HOOU solution was obtained and purified using the protocol described in [2] and the electrochemical experiments were carried out at 4°C. The Figure 1 shows the linear sweep voltammograms (LSVs) obtained in a fresh 0.5 mmol L^{-1} HOOU in 10 mmol L^{-1} phosphate buffer saline solution (pH = 7.4). The LSVs were recorded after 30 s of pre-concentration at two different applied potentials: -200 mV (Figure 1, red curve) and -800 mV (Figure 1, black curve). As one can see, no response was obtained in the LSV when the applied potential was -200 mV in the pre-concentration step. However, at a more negative potential (-800 mV), a peak around 500 mV in the LSV was observed due to the oxidation of the HOOU reduction product, which was adsorbed onto the carbon surface. A comparable behavior was noticed in an experiment performed with uric acid solution (not shown) and this may suggest that a similar electrochemical mechanism takes place in the UA oxidation.

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Spectroscopic Qualities of Silver Amalgam Particles

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Research and development of novel electrode materials applicable in direct analysis of biopolymers, e.g. nucleic acids and proteins, using (spectro)electrochemical detection systems represents great potential for research, screening or diagnosis of not only cancer and genetic disorders. Silver amalgam particles (AgAP) benefit from the emergence of nanotechnology and nanomaterials. The nanostructuring of the amalgam electrodes holds promise for higher charge transfer efficiencies, larger active surface areas, or even novel catalytic effects. AgAP may be prepared by electrodeposition on various conductive supporting materials or by chemical co-reduction using suitable reducing and stabilizing agents. Electrodeposition of AgAP on ITO and PGE or gold was found to be suitable for voltammetric and spectroelectrochemical applications in analysis of reducible organic compounds [1] and biopolymers, such as proteins or DNA [2,3], respectively. Spectroscopic qualities of AgAP have systematically been studied by spectroscopic elipsometry and electron energy loss spectroscopy (EELS), which revealed plasmonic resonances in the ultraviolet, visible and infrared wavelength range, and reported plasmonic properties at a single-particle level, respectively. [4]

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Poster Presentation

Electrochemical oxidation mechanism of the hair dye reagent *p*-toluenediamine at a glassy carbon electrode

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Permanent hair dyeing is a popular process adopted by millions of people around the world, and involves complex oxidative reactions between precursor agents (aromatic amines) and couplers (phenolic compounds) in oxidizing alkaline medium inside the hair strand.

The *p*-toluenediamine has been increasingly used in permanent hair dye compositions as precursor agent and it is associated with many adverse effects, such as allergies, dermatitis, skin infections, eye irritation, opacity of the corneas, stomach ulcers, aplastic anemia and it is recognized for its high mutagenic properties.

In this way, the investigation of the electrochemical oxidation mechanisms of *p*-toluenediamine is important since this could result in a better understanding of these effects and its physiological mechanisms of action.

The electrochemical behaviour and the electron transfer mechanism of *p*-toluenediamine, by cyclic, differential pulse and square wave voltammetry, in a wide pH range, at glassy carbon electrode, was investigated.

Cyclic voltammograms in *p*-toluenediamine, at glassy carbon electrode, in intermediate pH values, showed one quasi-reversible oxidation process.

The *p*-toluenediamine differential pulse voltammograms presented pH-dependent oxidation peaks, and for very acidic and alkaline pH values a second oxidation peak was observed.

The square wave voltammograms, in *p*-toluenediamine, at glassy carbon electrode, confirmed the reversibility of the electron transfer reactions and the oxidation mechanism was proposed.

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Thiophene-Based Conducting Polymers for Direct Electrochemistry of Cytochrome c

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Due to its adaptability and ease of preparation, conducting polymers have been used in direct electrochemistry of several redox proteins. While polyaniline (PANI) is, probably, the most studied organic conducting material, the lack of electrical conductivity at neutral pH hampers its use in biological systems. Self-doped polyanilines could be adopted for this application, but due to its their low activity, they were mainly used as protein entrappers in combination with carbon materials. Biological applications can be found for other conducting polymers as thiophene-based materials, since they are electroactive at physiological pH. However, in most cases polythiophene films serve merely as the matrix to embed nanostructures (gold nanoparticles, carbon nanotubes, graphene, etc.) that are employed in biosensor applications. In contrast few fundamental studies based exclusively on conducting polymers (in the absence of other promoters) can be found in relation to the direct electron transfer to proteins.

The present work explores the modification of metallic electrodes with different thiophene-based polymer prepared by electrochemical methods from its monomer precursors: 3,4-ethylenedioxythiophene (EDOT), hydroxymethyl-EDOT, 3,4-propylenedioxythiophene (ProDOT) among others. The electrocatalytic performance of these polymers were explored for the direct electron transfer reaction to *cyt c*. The voltammetric response of *cyt c* has been analyzed as a function of the chemical composition of the polymer film. The capabilities of the organic film to transfer charge to the protein have been examined by *in situ FTIR* spectroelectrochemical methods. This method allowed to evaluate the redox state of the polymer as a function of the potential and, in addition, the observation of intrinsic *cyt c* redox activity and its orientation upon electron transfer from the conducting polymer.

Poster Presentation

β-methylumbelliferone Surface Modification and Permeability Investigations at PENTEL™ Graphite Electrodes

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Electrochemical and micro-imaging analysis of a commercial graphite-composite material is presented following electro-oxidation with β-methylumbelliferone. Charge-transfer surface modification was observed for the graphite electrode, presumed to have arisen from adsorbed interfacial umbelliferone moieties. The molecular permeability of the new surface towards a range of similar, yet size-variable ($23Å^3$–$136Å^3$) molecular redox probes is discussed. Red-shift fluorescence in confocal microscopy offers further support for the presence of a surface-bound umbelliferone layer. An SEM-platinum profiling technique was used as an imaging tool to map the umbelliferone surface and size-distribution of electro-active sites.

The *in-vivo* Validation of a D-Serine Biosensor

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D-Serine is a gliotransmitter that modulates neurotransmission at glutamatergic synapses and has an *in-vivo* concentration of ~ 6 μM[1]. It is an endogenous co-agonist of glutamatergic N-methyl D-aspartate (NMDA) receptors modulating both neuronal transmission and synaptic plasticity[2]. D-Serine has been implicated in disorders associated with NMDA dysfunction such as schizophrenia, cerebral ischemia and amytotrophic lateral sclerosis[3]. The principle objective of this research was the development of a novel biosensor for the *in-vivo* detection of D-serine using Long Term In-Vivo Electrochemistry (LIVE). *In-Vitro* experiments were performed in a standard three electrode electrochemical cell containing 20 mL phosphate buffer saline (pH 7.4), utilising constant potential amperometry at +700 mV *vs.* SCE. Several dip-coating procedures were investigated for enzyme immobilisation, including incorporating different immobilisers, cross-linkers and stabilisers at various concentrations, in order to develop a simple and reproducible coating method to maximise the sensitivity of the sensor to D-serine. The inclusion of a poly-*o*-phenylenediamine underlayer conferred excellent permselective characteristics necessary for *in-vivo* recording. The developed biosensor was then characterised *in-vivo* in freely moving animals using pharmacological manipulations. This characterisation involved altering local levels of D-serine, glycine, and potential interferents such as ascorbic acid and O_2, and administration of glutamate receptor agonists and antagonists.

Acknowledgement: We gratefully acknowledge financial support from Science Foundation Ireland (SFI) under Grant Number **15/IA/3176**.

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Poster Presentation

Redox Mechanisms of Proteasome Inhibitors

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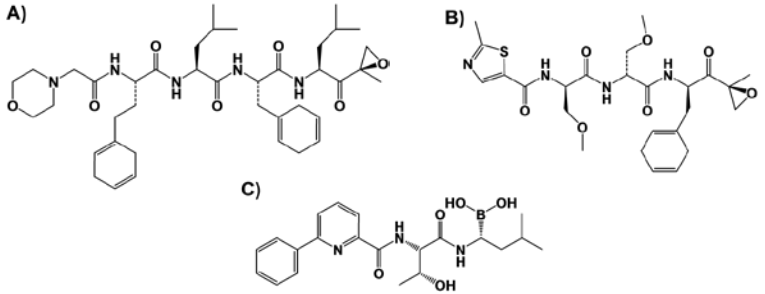
In this work, the redox behavior of proteasome inhibitors carfilzomib, oprozomib and delanzomib **Scheme 1**, was investigated through voltammetric methods using glassy carbon and boron doped diamond electrodes.



Scheme 1 – Chemical structures of A) carfilzomib, B) oprozomib and C) delanzomib

The voltammetric results showed that the oxidation of epoxyketone peptides carfilzomib and oprozomib at glassy carbon electrode surface occurred in one step while at boron doped diamond were observed two consecutive charge transfer reactions due to different adsorption orientation at the electrode surface. The moieties of these peptides, involved in the oxidation process, were morpholine for carfilzomib and thyazole for oprozomib. For boronated peptide delanzomib, at booth GC and BDD electrode surfaces, two irreversible and independent redox processes, oxidation at + 0.8 V and reduction at -1.4 V, were identified at pH = 7.0. The oxidation reaction occurred at the amino group close to the pyridine moiety of delanzomib with the transfer of one electron and one proton whereas the reduction process takes place at pyridine ring in a two-electrons two-protons mechanism.

Acknowledgment: Financial support from the Operational Programme Competitiveness 2014-2020, Project: NANOBIOSURF-SMIS 103528.

Electrochemical Fingerprint of Vancomycin

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Vancomycin is a glycopeptide antibiotic, widely used for the treatment and prophylaxis of severe infections caused by Gram-positive bacteria, but its use can lead to ototoxicity, nephrotoxicity and bacterial resistance to antibiotics, needing a therapeutic drug monitoring of vancomycin that maximizes efficacy, reduces toxicity and avoids the under-dosing, reducing the antibiotic resistance [1,2]. The electrochemical detection of vancomycin can facilitate the monitoring of vancomycin treatments. The purpose of this study was to determine the electrochemical fingerprint of vancomycin.

The electrochemical fingerprint of vancomycin both in oxidation and reduction domain was studied using different electrochemical techniques and several electrode materials: glassy carbon electrode, boron-doped diamond electrode and screen printed electrodes modified with nanomaterials (SPEs). The influence on the vancomycin electrochemical signal of the nature of the electrolyte and of chemical or enzymatic pretreatment of the antibiotic molecule was also investigated.

In order to explain the electrochemical behavior of vancomycin, the mechanism of the anodic oxidation was elucidated by the comparison of the electrochemical signal of vancomycin and other substances, simpler, but with the same functional groups and by investigating the oxidation products by means of small scale electrolysis and HPLC-MS/MS analyses.

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Acknowledgements: This work was supported by a grant of Ministry of Research and Innovation, CNCS - UEFISCDI, project number PN-III-P1-1.1-PD-2016-1132, within PNCDI III.

Poster Presentation

Biological Samples Treatment for Electrochemical Analysis of Vancomycin

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Vancomycin is an antibiotic from the glycopeptides class. It is administered parenterally for the treatment or prophylaxis of severe infections caused by Gram-positive bacteria. Unfortunately, the treatment with vancomycin is associated with a relative high occurrence of nephrotoxicity and ototoxicity. Also, the bacterial resistance to this antibiotic has increased. In order to maximize efficacy and improve the clinical outcome for the patients and to minimize the emergence of antibiotic resistance, there is a need of for the therapeutic drug monitoring of vancomycin [1]. The purpose of this study was the development of a fast and sensitive method for the electrochemical analysis of vancomycin from biological samples (human serum and urine).

The electrochemical analysis of vancomycin from human serum and urine was optimized in terms of electrode material, electrolyte and electrochemical technique in order to eliminate/diminish the electrochemical signal of the interferents from these complex matrices.

To facilitate a fast and simple electrochemical detection of vancomycin, different treatments of the biological samples were tested. The human serum and urine containing vancomycin were treated with different protein precipitation agents, liquid-liquid extraction and different solid phase extraction procedures and the influence of each treatment on the electrochemical signal of vancomycin and of the interferents was tested. Also different electrochemical pretreatments of the working surface and different parameters for the electrochemical techniques were also tested with the goal of further improvement of the selectivity of the method, applied in the aforementioned matrices.

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Acknowledgements: This work was supported by a grant of Ministry of Research and Innovation, CNCS - UEFISCDI, project number PN-III-P1-1.1-PD-2016-1132, within PNCDI III.

Observation of direct electron transfer from glucose dehydrogenase to single sheet graphene electrode

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Continuous glucose monitoring (CGM) is an emerging technology that can provide a more complete picture of the diabetes patient's glucose levels. Amperometric blood glucose tests typically require redox mediators to facilitate charge transfer from the enzyme to the electrode, that are not ideal in CGM settings because of their potential toxicity or long-term stability issues. Direct electron transfer (DET) would eliminate this need and has therefore attracted substantial interest. However, most DETs studies so far have used glucose oxidase leading to controversial results because the oxygen dependency may be misinterpreted as DET. Here, we overcome this challenge by using an oxygen-insensitive glucose dehydrogenase. The biosensor strongly responded to glucose even without a redox mediator, implying direct electron transfer (Fig. 1 A). Control measurements on different surfaces further confirm that the response is enzyme-specific. To enable direct electron transfer, the enzyme was immobilized on the surface of high-quality single-layer graphene electrodes via short pyrene linkers (<1 nm) (Fig. 1 B). The activity of immobilized enzymes was confirmed by glucose measurements with relatively unexplored redox mediator - nitrosoaniline, and the influence of a most potent interferent in blood, ascorbic acid, was assessed (Fig. 1 C). This is the first demonstration of DET from an oxygen insensitive enzyme to single-layer graphene, highlighting the potential of such devices for applications in CGM.

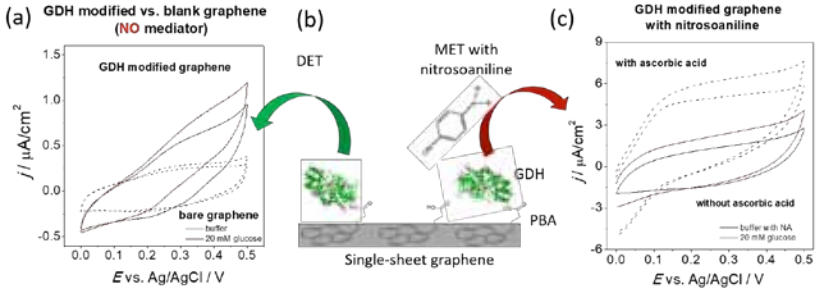

Fig. 1 (a) Direct electron transfer from glucose dehydrogenase (GDH) to graphene electrode (bare graphene – dashed line) (b) Graphene surface modification scheme (c) Mediated electron transfer (MET) using nitrosoaniline derivative (NA) in absence (solid) and presence (dashed line) of ascorbic acid (AA).

Poster Presentation

Rapid Screening of Levo-Thyroxine Using Nanoparticle-Based Sensor

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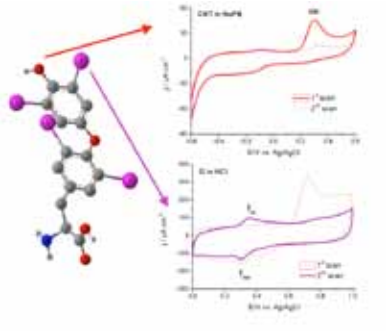
Several medical conditions, such as hypothyroidism, require hormone replacement therapy. In this particular case, administration of levo-thyroxine (LT4) plays an important role. Therefore, monitoring of LT4 levels in the human body lacks a simple, selective detection tool.

Nanoparticles such as graphene, carbon nanotubes and gold nanoparticles are widely used since they are good conducting materials, assure an enhanced surface area and biocompatibility or are able to catalyse reactions. In this work label-free nanoparticle based-sensors were optimised and evaluated for rapid, sensitive and selective detection of LT4 in medication samples as rapid screening tools.

The interfacial changes, which occur due to electrochemical properties of LT4, at different surfaces were studied by electrochemical impedance spectroscopy (EIS) and cyclic voltammetry (CV). The analytical detection was carried out by differential pulse voltammetry (DPV) where a high sensitivity (S = 0.99 μA cm^{-2} μM^{-1}) and detection limit of 0.61 μM for LT4 was achieved.



Acknowledgments: This work was supported by a grant of the Romanian Ministry of Research and Innovation, CCCDI - UEFISCDI, project number PN-III-P1-1.2-PCCDI-2017-0062, contract no. 58, within PNCDI III.

Study of different icosahedral carboranes as a novel electrochemical label

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In our previous studies [1-3] we have investigated several 3, 10, 11, 12 and 18 vertex boron cluster compounds and some of their *endo*- and *exo*- skeletal derivatives. In our present study, we have studied electrochemical behavior of 12 vertex icosahedral carboranes – *orto*-, *meta*- and *para*-carborane with different *exo*-skeletal substituents.

Our studies were done using phosphate buffers in pH range from pH = 2 to 10, in some cases with 10%- 30% dimethylsulfoxide (DMSO) addition (to increase the solubility of some samples). The measurements were done using either polished glassy carbon electrode or home-made screen printed electrodes (they were preferably used for measurements with DMSO addition). We have measured electrochemical responses using differential pulse (DPV) and cyclic voltammogram (CV) techniques.

Compared to our previous results, we have found quite no dependence of electrochemical signal position on cluster substituents. Distinct electrochemical response was observed only for the *orto*-carborane and their *exo*-skeletal substituents. The use of DMSO enable us to observe electrochemical signals for some samples, which were in pure phosphate buffers not detectable.

The study of different *exo*-skeletal substituents on the electrochemical behavior is crucial for the intended use of these compounds as electrochemical labels of biomolecules. Used substituents (-OH, -COOH) are usable for different bioconjugation reactions. The boron cluster compounds family forms a novel and perspective group of electrochemical biolabels.

This work was supported by the Grant No. 19-04630S from Czech Science Foundation.

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Poster Presentation

Relation Between Biological Activity, Structure and Voltammetric Behavior of Novel Antimycobacterial Agents

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Growing number of drug-resistant strains of *Mycobacteria* causing illnesses such as tuberculosis is prompting the development of new promising molecular scaffolds designed for better treatment against those strains. One of these promising novel agents are derivatives of ring-substituted hydroxynaphthalenecarboxanilides [1]. The pilot study of a nitro substituted derivative has confirmed that this compound can be electrochemically oxidized and also reduced at the glassy carbon electrode [2]. In this study electrochemical potentials of oxidation of further 22 derivatives were measured by cyclic voltammetry. All the compounds gave one oxidation signal corresponding to the oxidation of their hydroxyl group. The obtained values of oxidation potentials were correlated with their structure and Hammett substituent constants for *ortho*-, *meta*-, and *para*- derivatives with good fit parameters. The relation of electrochemical potentials and biological activities of novel antimycobacterial agents is discussed.

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This research was carried out within the framework of Specific University Research (SVV 260440). This research was supported by the Czech Science Foundation (project 17-03868S).

Towards a Point-of-Use Nanosensor System for on-Farm Disease Detection

Fernando Garrido Diaz, Alan O' Riordan

Farm diseases in animals and plants have increased because a variety of pathogenic viruses, bacteria, protozoa, and helminths which have developed numerous mechanisms that render them resistant to some antimicrobial agents. Disease outbreaks in high-value crops, can cause a significant economic damage, consequently, a method for the rapid identification of viruses is critical for the protection and prevention of costly outbreaks [3].

Design an immunosensor to detect the presence of ***Potato virus Y*** and process this information with the help of an electrical device generating a response directly to the farmer. Silicon chip electrodes were modified using different reagents to permit covalent attachment of a specific capture antibody to determine the the presence or absence of disease causing (virus) pathogens in plant material. To this end, Cyclic voltametry (CV) and Electrochemical impedance spectroscopy (EIS) were employed. In CV measurements, a decrease in electron transfer from a redox molecule, Ferrocene monocarboxylic acid, to the electrodes was indicative of a binding event, whicle in EIS an increase in impedance arising from the formation of a thicker dielectric layer is expected.

Virus presence was confirmed is suspensions of ground up plant material by a decrease in current intensity (CV) and in increase impedance (EIS), as expected. In case of the negative un-infected control samples (without a target virus), the CV and EIS voltammograms exhibited only a slight change in signal, arising from non-specific adsorption. Time to analysis was 15 minutes

This immunosensor could be a very important tool for food safety from farm-to-fork as the presence virus can be detected in minutes in the field compared to hours or days using existing laboratory methods.

Poster Presentation

Optimization of the fabrication of an abiotic bioelectrode for implantable biofuel cell.

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For more than fifty years lithium-based batteries were used to supply power Implantable medical devices (IMD) [1]. However, in order to respond to the DMI's conditions, the required lithium based battery's size and lifetime fail to reach more than few tens of microwatts. In this context, implantable enzymatic glucose/oxygen biofuel cells (BFCs) are increasingly seen as a potential future technology for replacing lithium batteries [2]. One of the major limitations of enzymatic BFCs is the short lifetime of enzymes especially in the biocathode case. Thus, the use of abiotic catalysts is preferable over enzymes for long-term application of implantable glucose BFC's.Here, we present for the first time a nitrogen/iron doped graphene-based biocathodes able to operate for several months under physiological conditions. These biocathodes were fabricated using two different printing technics: i) a flexible cathode where the catalyst is spread over a larger area fabricated using a doctor blade printing technic ii) a 3D printed cathode with a high surface area fabricated with 3D bioprinter. Electrochemical characterizations under physiological conditions for both cathode types show maximum current densities of 70µA/cm^2. Moreover, long-term stability of 6 months is obtained in physiological conditions. Biocompatibility tests reveal also the biocathodes biocompatibility and the ability to operate inside the animal body without any inflammation reactions.

Figure 1: *a) 3D printed cylindrical electrode 3D b) Flexible electrode.*

Keywords: *Abiotic BFC, Abiotic, Doctor Blading coating, 3D printing.*

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Engineering Functional Enzyme Bioelectrodes with Conductive Polymeric Films for Sensing Applications

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Immobilization of enzymes is a key strategy for the development of point-of care diagnostics such as biosensors and biofuel cells. Conductive polymeric films have been proposed as excellent signal transducers for biomedical applications due to their high electrical conductivity and biocompatibility. In this context, we report a highly innovative, while simple and robust, surface chemistry approach for the covalent immobilization of enzymes onto conductive polymeric nanostructures. In particular, an electrochemical grafting approach is proposed, according to which, polypyrrole (PPy) films on gold surfaces on-chip are modified with β-alanine (Ala) as a linker, bearing free carboxylate groups, for further probe immobilization. As a case study and considering its clinical relevance for glucose monitoring systems, the enzyme Glucose Oxidase (GOx) is used. The fabricated GOx/Ala/PPy/Au electrode exhibited excellent performance for glucose detection, with a linear response within the range of 0.1 mM - 10 mM, and a sensitivity of 3.75 μA mM^{-1} cm^{-2}. The observed low limit of detection is 0.1 mM; a value that lies below the normal glucose concentration in blood and non-conventional physiological fluids, such as interstitial fluid or saliva. The fabricated glucose sensor also demonstrates an excellent reproducibility with a relative standard deviation of 4 %. The methodology proposed paves the way for rapid manufacturing of effective enzyme-based polymeric electrodes, which, contrary to other PPy-based sensors previously suggested, does not involve any chemical pre-treatments of the monomer used.

Poster Presentation

Nitro-fatty acids redox transformations: electrochemical and EPR study

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Over the past several years, nitroalkene derivatives of fatty acids (NO_2-FAs) have gained significant attention among scientists due to their encouraging preclinical and clinical responses. An analysis of synthetic nitroalkene derivatives of oleic and linoleic acid showed that these species possess unique electrophilic reactivities that support metabolic enzyme regulation, the modulation of inflammation and activation of stress responses [1].

Here we focused on the redox behavior and an evaluation of the stability of different nitro fatty acids: nitrooleic acid, nitrolinoleic acid and their derivatives. Cyclic voltammetry and chronopotentiometry were used to study the electrochemical reduction of the R-NO_2 group at mercury and also carbon electrodes. The R-NO_2 reduction process is related to the electrophilic properties of the NO_2-FAs and should be distinguished from adsorption processes, which were investigated by alternating-current voltammetry.

In addition to electrochemistry, an electron paramagnetic resonance (EPR) method was utilized to study NO radical release phenomena and for evaluating the stability of tested NO_2-FAs. Fatty acids without an NO_2-substitution served as the negative control in all experiments.

In our opinion, these electrochemical and EPR results provide new mechanistic data that is potentially useful for further biochemical and pharmacological investigations of NO_2-FAs.

The authors would like to acknowledge *Prof. Bruce Freeman* (University of Pittsburgh, USA) and his group for the preparation of nitro-fatty acids and *Prof. Milos Mojovic* for performing EPR analyses and helping with data interpretation. We also wish to thank Ministry of Education, Youth and Sports (CZ.02.2.69/0.0/0.0/16_027/0008482) for financial support.

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Electrodeposition of Silver Amalgam on Golden Plate Electrodes Using Double Pulse Chronoamperometry

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Metal mercury represents unique electrode material advancing from its ideally smooth surface and high hydrogen overpotential enabling to detect reduction processes ongoing at very negative potential. Thanks to these properties, mercury electrodes found frequent utilization in voltammetric analysis of DNA [1]. Disadvantages of mercury electrodes are their low mechanical stability and toxicity of metal mercury. Silver solid amalgam represents up to now the most suitable alternative electrode material to metallic mercury in electroanalytical chemistry [2]. This work is focused on the preparation of silver amalgam particles by its direct electrodeposition on vapor deposited golden plate electrodes using double pulse chronoamperometry from solution containing soluble Ag^{+} and Hg^{2+} ions. Different parameters of chronoamperometry (pulse and grow potentials and times) and different Ag/Hg ratio in solution were optimized during electrodepositions of silver amalgam nanoparticles. All the prepared amalgam particles were characterized by scanning electron microscope with energy dispersive X-ray spectroscopy. Electroanalytical application of the gold electrodes decorated by silver amalgam particles was confirmed by successful detection of 4-nitrophenol using cyclic voltammetry and of synthetic oligonucleotides labeled with osmium tetroxide complex with 2,2´-bipyridine by DPV, what allows their broad application in analysis of nucleic acids [3].

This work was supported by The Czech Science Foundation (grant 17-23634Y) and part of the work was carried out with the support of CEITEC Nano Research Infrastructure (MEYS CR, 2016–2019).

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Poster Presentation

Aptamer-based Electrochemical Ferrocene-Labeled Biosensor for Tetracyclines Detection

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Tetracyclines (TCs) are one of the most common used antibiotics for the treatment of infectious diseases in veterinary medicine. Therefore, high TCs levels can be found in animal-based food products (*e.g.* meat, eggs, milk) and environmental samples (*e.g.* soil, waste water) due to its high accumulation [1]. Hence, safety and quality in food industry are of crucial importance as foodborne illnesses have become a global menace on public health. Therefore, there is an imperative need to develop high sensitive and selective sensors with easy operation and *in-situ* applications for tetracycline antibiotics.

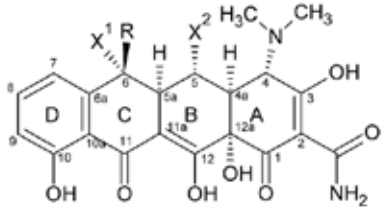

Figure 1. General structure of tetracyclines

Several strategies were addressed for the direct detection and quantification of tetracyclines. Gold was electrodeposited at glassy carbon electrodes and carbon-based screen-printed electrodes from a $HAuCl_4$ solution to obtain different 3D-nanostructures (nanoparticles and nanovoids). Supporting electrolyte and pH choice was a crucial step in obtaining the optimal parameters. A ferrocene-labeled aptasensor for tetracycline detection was developed using the platform with the best analytical performance and possible sensor applications will be presented.

Acknowledgements
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β-Lactoglobulin Allergen Detection based on an Electrochemical DNA-assay

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Cow's milk is one of the most common allergic reaction-induced foodstuffs as being introduced since early childhood. The most frequently allergenic protein which is responsible of cow's milk allergy is *β*-lactoglobulin (*β*-LG) mainly due to its high stability and absence in human milk [1]. As there is no cure for cow's milk allergy yet developed but only effective care and emergency treatments available, the only viable solution is a free milk-based diet. Therefore, risk management and mandatory labelling system of food allergens are critical steps in food industry to guarantee consumers safety. For these reasons, the development of an easy-to-use, fast and accurate device for allergen detection is fundamental to prevent life-threatening reactions in sensitive subjects.

In this work, a DNA-based sensor for *β*-lactoglobulin electrochemical detection was developed. Two oligonucleotide sequences labelled with amino and biotin termination, respectively, were selected using M-fold software in order to obtain a DNA sensor. First, different monomer concentrations and ratios of aniline and anthranilic acid were electropolymerised at graphite screen-printed electrodes by cyclic voltammetry (CV). The obtained platforms were further characterized by Scanning Electron Microscopy and electrochemical techniques (CV and Electrochemical Impedance Spectroscopy) and used as support for the sensor development. The selectivity of the DNA sensor was evaluated by testing unspecific proteins, the results showing promising results for real sample applications.

Acknowledgements
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Poster Presentation

Electrochemical characterization of *Geobacter sulfurreducens* under fumarate-limiting condition, comparison between biofilm and chemical immobilization

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Geobacter sulfurreducens uses sodium acetate as an electron donor; the acetate is completely oxidized to CO_2 using the central metabolism [1,2]. At this point, the resulting electrons can be divided into two routes. The first route can be considered as an "open Krebs Cycle" ending in fumarate as the final electron acceptor. The second route involves a complete Krebs Cycle, and it utilizes IV type pili and the c type cytochromes, this route allows the bacteria the possibility to make direct contact with metallic soluble and insoluble subtracts [3]. In electrochemical studies, it has been found that under fumarate-limiting conditions *Geobacter sulfurreducens* generates more current than the strains grown under standard culture conditions [4]. In this work, the effect of fumarate concentration in the charge transfer process of *Geobacter sulfurreducens* is reported. The purpose of growing the bacterium under limiting (20 and 10 mM) and standard-fumarate conditions (40 mM) was to generate a stress condition in the bacteria such that the flux of electrons derived from acetate metabolism could be directed towards the electrode. A growth curve of Geobacter sulfurreducens was made in different in the previously mentioned concentrations of fumarate finding a modification in the growth kinetics as was reported in the literature [2,3].

Furthermore, a modified carbon electrode was designed to perform the electrochemical studies. This electrode consisted of glassy carbon (GC) electrode as the current collector covered with a nanostructured carbon black film (nCB). The GC/nCB electrode was modified with a gold electrodeposit and chemically functionalized with an organic molecule (4'-mercapto-N-phenylquinone diimine). This electrode allowed both electrochemical analysis, bacterial immobilization (through chemical attachment) and biofilm development where important features about the electrochemical electron transfer mechanism followed by this bacteria were found.

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A glance at the electroanalytical response of carbon materials based films doped with aminocumulene acting as dispersant and N-containing precursor

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Broadly speaking, cumulene is a linear allotrope of carbon without end groups and with all carbons connected by double bonds. Furthermore, cumulene derivatives have applications in the field of molecular wires. To fulfil such applications, linear and rigid molecules made up of cumulated carbons connected at the end with a wide variety of electrochemically functional groups can be synthesised. In this regard, we report herein the synthesis of the technical compound called aminocumulene (AC) from the polycondentation of hexamethylenetetramine in anhydrous acid media.

Accordingly, we are aiming at exploring the spectral, structural and electrochemical properties of the AC molecule in order to evaluate its performance as highly dispersant agent of carbon materials in aqueous solutions. In doing so, carbon materials dispersions of different mixtures based on AC as N-containing precursor and either with distinct oxygen functionalized carbon nanotubes or with p-benzoquinone derivative carbon material were prepared under powerful insonation. Then, carbon based films were prepared by drop casting or painting onto carbon support for further characterization by scanning electron microscopy, X-ray diffraction and X-ray photoelectron spectroscopy. Then, the electrochemical behaviour of the different carbon based films was explored as a function of pH, in the presence of outer-sphere probes and finally inner-sphere probes of interest in electroanalysis.

Poster Presentation

Incorporation of lipid liquid crystalline drug carriers containing doxorubicin into lipid model membranes

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There has been recently a lot of interest in lipid liquid crystalline nanoparticles - cubosomes as potential drug delivery systems [1,2]. We present our studies of the incorporation of cubosomes containing doxorubicin, a potent anticancer drug into Langmuir-Blodgett layers used as simple models of the biological membrane. We demonstrate the differences of the lipid monolayer structure when different lipidic compounds (monoolein or phytantriol) and two different polymers (to stabilize the nanoparticles against aggregation) are used for the preparation of cubosomes. The Langmuir-Blodgett lipid film is transferred from the air –water interface onto the gold electrode surface and using voltammetry we monitor the relocation of the toxic drug from the drug delivery vehicle to the lipid membrane.

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Studies on the affinity-based biosensors for electrochemical detection of HER2 protein

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As cancer diseases are the second main cause of death worldwide, it is necessary to elaborate fast, selective and sensitive early detection tools that would also allow for the application of proper treatment methods.

Currently, early detection methods of cancer refer to biopsy, gene analysis and imaging techniques which are expensive and require sophisticated equipment [1]. Alternatively, the occurrence or threat of a number of cancer diseases can be linked to the elevated concentration of protein biomarkers. An example could be HER2 protein, which overexpression indicates a development of several types of cancer including breast cancer [2].
Herein, we present the studies on the elaboration of affinity-based sensing layers for electrochemical detection of HER2 protein. HER2-specific monoclonal antibodies were immobilized on gold electrodes by the use of cysteamine intermediate layer and applied as capture element in direct, antibody sandwich and antibody-aptamer hybrid assay. Thiolated label-free DNA aptamers or conjugated with methylene blue electroactive specie that were selective towards HER2 were also utilized as capture elements. The binding between HER2 biomarker and sensing layer was analyzed using voltammetric and impedimetric techniques in the presence of redox indicators such as ferri/ferrocyanide. Amperometry technique was applied for HER2 detection in the case of the application of secondary polyclonal antibody combined with HRP enzyme.
It was observed the most promising approach was the utilization of antibody sandwich assay that enabled HER2 detection from 1 to 100 $ng \cdot mL^{-1}$ which covers the range indicating the development of cancer disease (15 – 75 $ng \cdot mL^{-1}$). Though the interaction between interfering proteins and antibody–based layer was evident on the basis of SWV measurements, it led to at least two times smaller current responses than for HER2 protein. Future research will be focused on further limitation of nonspecific adsorption on the transducer surface as well as on the miniaturization of proposed tool.

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Poster Presentation

Neural Interface via Artificial Synapse between Live Neuron and Solid Electrode

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To make robust interaction platform with neural system, there have been numerous studies modifying an electrode with a variety of biocompatible materials such as conducting polymer, hydrogel, and proteins. Previous studies trying to improve on neural interface share the common fundamental strategy, which is to stimulate neurons or read out neural signals through electrical cues. However, neurons communicate each other via synapses where signaling using chemical messengers occurs. Formation of a synapse is triggered by membrane protein binding between neurons. Among out of many synaptic adhesion proteins, neuroligin1 is a most widely studied synaptic protein, which is known to induce the presynaptic terminal without assistance of other proteins.

Here, we genetically engineered the neuroligin1 to make it easy to being immobilized on the artificial substrate, and investigating the feasibility of an induced synapse as a neural interface. The most important features of the neural interface are structural stability, signaling activity, and scalability to bran tissues. Using engineered Nlg1-modified microbeads and primary cultured hippocampal neurons, the durability and scalability of the artificial synapses were examined. Next, we fabricated the functionalized electrode for the electrochemical study. We believe that the strategy using artificial synaptic electrode can open up new vista for neural interface applications.

Mechanism of lipopeptide action on model lipid membranes

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The discovery of antibiotics was a milestone in the human fight against infectious diseases and has led to the substantial increase in quality of human's life. However, rapidly increasing number of multi-drug resistance pathogens resulted in a continuous need for new active compounds with strong activity. Lipopeptides are known to show bactericidal activity and due to their simple structure, ease of design, and low cost of implementation, they are often considered potent replacement for many traditional antibiotics. Another important advantage of lipopeptides is related to the fact that their preferential target is a cell membrane.[1] Lipopeptides are expressed in bacteria e.g. from Bacillus or Pseudonomas groups and exhibit antibacterial, antifungal and surfactant activity.[2]

Here we present results of our studies on the interactions of short synthetic lipopeptides with model lipid membranes. The models of two cell membranes were employed, reflecting the eukaryotic and bacterial cell membrane. The first one was composed of 1-palmitoyl-2-oleoyl-sn-glycero-3-phosphocholine (POPC) and cholesterol. The latter membrane was prepared with lipids extracted from Escherichia Coli cells, i.e. L-α-phosphatidylethanoloamine (PE) and L-α-phosphatidylglycerol (PG). In order to prepare a model lipid membrane, phospholipid monolayers may be transferred onto solid substrate to form a bilayer.[3,4] We employed the combination of Langmuir-Blodgett and Langmuir-Schaefer techniques to immobilized model lipid membranes on solid support. The action of lipopeptides on the membranes was monitored using electrochemical methods combined with atomic force microscopy imaging and quartz crystal microbalance measurements. Since the packing density of the molecules within PE:PG and POPC:Chol bilayers and their elastic properties are similar, we found out that diverse action of the lipopeptide is primarily caused by a different net charge of the membranes.

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Poster Presentation

The Interrelation Between Ion-Exchange Membranes and the Efficiency of Microbial Electrochemical Systems

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In this study, microbial fuel cells (MFCs) – operated with novel cation- and anion-exchange membranes, in particular AN-VPA 60 (CEM) and PSEBS DABCO (AEM) – were assessed comparatively with Nafion proton exchange membrane (PEM).
The process characterization involved versatile electrochemical (polarization, electrochemical impedance spectroscopy – EIS, cyclic voltammetry – CV) and biological (microbial structure analysis) methods in order to reveal the influence of membrane-type during start-up. In fact, the use of AEM led to 2-5 times higher energy yields than CEM and PEM and the lowest MFC internal resistance by the end of start-up.
Regardless of the membrane-type, *Geobacter* was dominantly enriched on all anodes. Besides, CV and EIS measurements implied higher anode surface coverage of redox compounds for MFCs and lower membrane resistance with AEM, respectively. As a result, AEM based on PSEBS DABCO could be found as a promising material to substitute Nafion.

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Electro-responsive nanocomposites as smart antibacterial surfaces

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Being a common phenomenon, the formation of biofilm has a substantial effect on a broad range of industrial areas, including food and biomedical industry. Currently, one of the major challenges is the prevention of the growth of bacteria, since the formation of biofilm may result in the contamination of food products and medical devices. On the other hand, biofilm formation is highly desired in several biotechnological applications, where naturally-immobilized micro-organisms are expected to perform their specific functions, e.g. in bioreactors. Regardless the formation of biofilm is expected or undesirable, there is a pressing need to design new methods allowing the control of this process, either facilitating or preventing the attachment and growth of microorganisms.
The aim of this study was to develop a novel type of an electro-responsive nanocomposite coating than can be used to control the growth of biofilm. The coating was based on a poly(3,4-ethylenedioxythiohene) matrix, which is known to have tunable physical properties and morphology depending on its redox state. Conducting polymer matrix was filled with silver nanowires in order to provide the antibacterial functionality to the nanocomposite. The as-formed electro-responsive nanocomposites were characterized by means of electrochemical (cyclic voltammetry, electrochemical impedance spectroscopy) and microscopic (SEM, AFM) techniques, and were used as platforms for the growth of a model bacterial strain (*E. coli*). The antibacterial effect of the nanocomposite was then correlated with its electrochemical performance and surface morphology to find the optimal design for achieving the superior antibacterial activity.

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Poster Presentation

Cyclodextrin as a Carrier for Anticancer Drug-Temozolomide

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Glioblastomas (GBM) are the most lethal form of human brain tumours with the mean survival time below one year from the time of diagnosis. The current standard treatment for glioblastoma begins with a maximal safe surgical resection, followed by a combined radiotherapy and chemotherapy [1]. Temozolomide (TMZ) is one of the most effective chemotherapeutic agents for glioblastoma multiforme. Various studies have shown that TMZ is an effective alkylating compound. At physiological pH, TMZ is metabolised to its active metabolite 5-(3-methyl-1-triazenyl)-imidazole-4-carboxamide (MTIC) [2]. Due to the short half-life of TMZ, high dose administration of drug is required and this leads to several dose-limiting side effects [3]. To improve the efficacy of TCM and to lower the risk of side effects the use of drug carriers such as cyclodextrin is proposed.
Cyclodextrins (CDs) are cyclic oligosaccharides composed of α-(1,4) linked glycopyranose units. Their spatial structure resembles the shape of a truncated cone with a hydrophobic inner cavity and hydrophilic outer surface. CDs are able to form host-guest inclusion complexes with a hydrophobic molecules as a guest [4].
The aim of this study was to investigate the complex formation between temozolomide and selected cyclodextrin derivatives. Using electrochemical and spectroscopic methods we determine the stability constant of TMZ/CD complex at physiological pH 7.4 and at pH 5.5 - characteristic for cancer cells.

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Enhanced Production of Lipids and High-Value Products from Microalgae by Electrochemical Treatment

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Phototrophic microalgae have been considered as microscopic factories for the production of various biomolecules such as biofuels and value-added chemicals because their physiological characteristics with high content of proteins, carbohydrates, lipids and pigments. Several applications of electricity have been employed in the field of biotechnology such as electroporation, electrophoresis, energy production as well as biosensing area. Especially, electrochemical reactions may affect to the metabolism of microorganisms via direct or indirect pathways, because growth of microorganisms is involved providing and consuming the energy source-the flow of electrons-through chemical reactions. In this study, the effects of electrochemical treatments (*i.e.*, supplementation of anodic and cathodic currents) were investigated to enhance the productivity of microalgal lipids (*i.e.*, polyunsaturated fatty acids, PUFAs) and multifunctional pigments (*i.e.*, astaxanthin) in green microalgae. During the process, accumulation of both PUFAs and astaxanthin was improved within shorten period of time compared with conventional photobioreactor cultivation.

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Poster Presentation

Bioelectrochemical Systems for Selective Conversion of Carbon Dioxide to Value-Added Chemicals

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Electrochemical conversion of CO_2 to fuels using a renewable energy sources is an increasingly important area that addresses global warming and fossil fuel shortages. To apply the concepts in practically, the electrochemical reactions should be proceeded under mild conditions of a moderate temperature and atmospheric pressure. In general, however, a high overpotential exists in the electrochemical reduction of CO_2. For instance, the theoretical potential for reduction CO_2 electrochemically to formic acid under standard conditions is -0.199 V (vs. NHE). To date, extensive efforts to develop electrode materials for the direct and electrochemical reduction of CO_2 have been made also, but so far, they require the application of energetically inefficient potentials or they are nonspecific and produce mixtures of products. In this context, electrochemical reduction of CO_2 has yet to be achieved on appropriately large scale due to the lack of efficient, robust catalysts operating at low operation potential with high selectivity. The biocatalytic reactions involving energy applications are gaining in prominence, especially in the direction of enzymatic and bacterial electrosynthesis of desired fuels under excess electrical energy. In this work, we focus on the electrochemical CO_2 reduction with high selectivity at low bias potential by employing electrochemical system and biocatalysts such as enzymes and microorganisms.

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Nanostructured Samarium Oxide Electrodes for Biosensing Applications

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The development of nanostructured electrochemical biosensors have been a subject of extensive research. Apart from carbon nanomaterials, metal/metal oxide nanoscaled architectures showed several advantages such as increased current densities, high electroactive surface areas, high stability and facile manipulation.
In this work, the development, characterization and applications of samarium oxide electrode is described. Sm_2O_3 was electrochemically deposited on gold electrodes by a single bath method [1]. The construction of the Sm_2O_3/Au/SiO_2/Si electrode was optimized in each step of its construction. Surface characterization was carried out by X-ray diffraction, X-ray photoelectron spectroscopy and scanning electron microscopy.

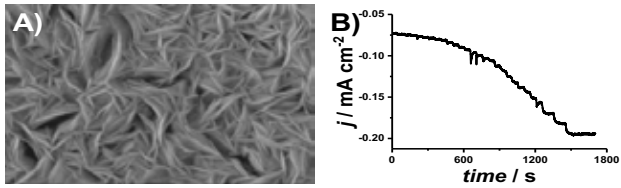

Figure 1. **A)** SEM image and **B)** amperometric response recorded at -0.20 V in pH = 7.0 phosphate buffer at injections of H_2O_2, of the Sm_2O_3/Au/SiO_2/Si electrode.

Cyclic voltammetry and electrochemical impedance spectroscopy were used to understand the surface processes that occur at the electrode/electrolyte interface. The Sm_2O_3/Au/SiO_2/Si electrode was used for the detection of hydrogen peroxide by amperometry at potential values as low as -0.10 V (vs. Ag/AgCl). Finally, glucose oxidase was immobilized [2] at the surface of Sm_2O_3/Au/SiO_2/Si and the biosensor applied for glucose determination by fixed potential amperometry.

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Poster Presentation

Integration of bilirubin oxidase in transparent, inverse opal ATO electrodes for efficient oxygen reduction

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Exploiting the biocatalytic four-electron reduction of oxygen to water has attracted much attention during the last decade for the construction of enzymatic biofuel cells (EBFCs).[1] While in most cases intransparent electrodes hosting oxygen-reducing enzymes such as bilirubin oxidase (BOD) or laccase have been applied, recently the demands for transparent electronics, e.g. for smart electronic contact lenses, has initiated the new field of transparent EBFCs.[2] Previous approaches have relied on modified, nanostructured indium tin oxide (ITO) as transparent electrode material,[2] which are however often limited either by a poor electrode transparency or failing in achieving a proper electrical connection with the enzyme, resulting in low biocatalytic currents and high over potentials.
In this study we have integrated BOD in inverse opal antimony tin oxide (IO-ATO) electrodes for the design of a transparent oxygen-reducing biocathode.[3] Therefore, IO-ATO electrodes with different pore sizes (250 nm and 650 nm) have been constructed by a template approach and evaluated according to their transparency and bioelectrochemical performance. Moreover, the electrode height has been varied between 2 und 8 IO-film layers resulting in a 60-fold surface area amplification (8 layer system) as compared to a flat FTO electrode. Despite the high surface area amplification, the IO-ATO electrodes give rise to high transparencies in the visible wavelength range of about 69% (at 500 nm), 77% (at 600 nm) and 81% (at 700 nm) for 4 IO-film layer systems. Since unmodified IO-ATO electrodes give only rise to a modest biocatalytic response after BOD immobilization, we have applied pyrenecarboxylic acid (PC) as promotor layer between the enzyme and the electrode. The resulting IO-ATO|PC|BOD electrodes show a pronounced oxygen reduction current starting at 0.556 ± 0.006 V vs. Ag/AgCl. Up to -134.5 ± 8.6 µA cm^{-2} and -287 ± 18 µA cm^{-2} can be obtained under air-saturated conditions without and with stirring, respectively.

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Microbial Surface Displaying Enzyme Based Efficient Biofuel Cells

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The power output and stability of biofuel cells (BFCs) greatly depend on the properties of both the biocathode and bioanode. Microbial surface display enables proteins, peptides or antibodies to be efficiently expressed on the surface of microorganisms, which has found potential applications in biocatalysis, environmental governance and bioanalysis. In this talk, we present our recent progress in developing enzyme biofuel cells using glucose dehydrogenase, xylose dehydrogenase displayed bacteria as well as sequential enzymes displayed bacteria as anodic catalysts, which significantly improved the stability of the bioanode in saccharide oxidation. The strategy overcomes the conventional limitations such as the undesirable long-term durability and electron transfer issue in the development of BFCs. Thus, our approach holds great potential to directly convert degradation products of biomass into electrical energy.

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Poster Presentation

In situ evaluation of DNA-antidiabetic drug metformin interaction using a dsDNA-electrochemical biosensor

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Metformin (*N,N*-dimethylbiguanide, MET) is an antihyperglycemic drug administered orally to control type II diabetes. In addition to being widely used to control diabetes, MET is also used to reduce cholesterol and triglycerides, so it is in the first line of choice for treatment especially in people with overweight and fat and normal renal function. Although MET has excellent antidiabetic activity and several potentialities in the treatment of other diseases, MET is one of a group of emerging contaminants, which are compounds that are strong candidates for future regulation due to their potential toxicity.

The evaluation of the antidiabetic drug MET-dsDNA interaction in incubated solutions, by differential pulse voltammetry and ultraviolet-visible spectroscopy, and *in situ* using dsDNA-, poly[G]- and poly[A]-electrochemical biosensors, for different time periods, was investigated.

The effect of the MET-dsDNA interaction was electrochemically followed comparing the changes in the oxidation peaks of guanosine and adenosine residues, in the absence and presence of MET, and monitoring the occurrence of free guanine and free adenine, and the purine biomarkers: 8-oxoguanine, and 2,8-dihydroxyadenine.

The MET-dsDNA interaction occurred in two ways. First, MET interacts with dsDNA causing the condensation/aggregation of DNA double helix, producing a rigid MET-dsDNA complex structure, with a preferential interaction between the adenine hydrogen atoms in the A–T base pair and MET, and subsequently, the intercalation of MET molecules in the dsDNA structure, which led to greater exposure of the dGuo and dAdo residues to the GCE surface and consequently greater ease in oxidizing them, was detected. The peak attributed to the oxidation of 8-oxoGua and/or 2,8-oxoAde was not detected, suggesting that, under the experimental conditions used, MET did not induce oxidative DNA damage.

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Automated Microfluidics for Electrochemical Immunoassays for Prostate Cancer Staging and Grading

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Automated microfluidic platforms are a novel technique to enhance miniaturisation, assure efficient reagent consumption and to produce repeatable results. Inexpensive mass-produced components are used here, such as the programmable Arduino Uno microcontroller, high torque servos, a commercial syringe pump and electronically controlled motors. With a C/C++ programmed microcontroller, repeatable timing for each step in a complex assay protocol including sandwich immunoassays can be achieved. A multiplexed immunoassay for detection of proteins that show potential in prostate cancer clinical decision-making is developed to demonstrate the applicability of the automated microfluidic platform

Prostate cancer (PCa) is one of the leading causes of death from cancer in men worldwide. Detection of elevated prostate-specific antigen (PSA) is an important indication of cancer. However, staging and grading of the disease is problematic. We have selected an 8-protein biomarker panel to test whether diagnostic value is improved for the prognosis of aggressive types of prostate cancer.

Here, we used magnetic particles labelled with horseradish peroxidase and detection antibody on electrodes in the electrochemical assay to capture eight proteins associated with PCa under less than 30 mins [1]. The automated microfluidic platform provides a repeatable and rapid electrochemical immunoassay for PCa detection and it can also assist numerous other immunoassays in detecting different biomarkers in patient samples.

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Poster Presentation

Phospholipid layers as simple models to study the interactions with anticancer and cholesterol-lowering drugs.

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Phospholipid mono- and bilayers were used as simple models of cell membranes to study the influence of selected anthracyclines and cholesterol-lowering statins. Interactions of anthracycline drugs such as doxorubicin (DOx), daunorubicin (DNR) and idarubicin (IDA) with model phospholipid monolayers mimicking healthy (DMPC) and cancer (DMPS) cell membranes were studied first by Langmuir technique. The incorporation of drugs into model membrane leads to the changes in such properties as fluidity of the layer and depends on the type of the anthracycline derivative used, e.g. its hydrophobic/hydrophilic properties and the composition of the model membrane. Since the content of cholesterol in cell membranes may change upon the development of cancer, the effect of cholesterol presence in the model lipid system on the interactions with anticancer drugs was also verified [1]. Due to the presence of electrochemically active quinone-hydroquinone group in the structure of anthracycline drugs, it was possible to employ electrochemistry to confirm the incorporation of the drugs into both DMPC and DMPS supported bilayers and compare the interactions of different anthracyclines with these two types of model systems [2]. The results of Langmuir and electrochemical studies were also compared with the results of measurements performed using spectroscopic (PMIRRAS) and neutron reflectivity methods. Phospholipid layers were also employed as model systems to study the influence of selected cholesterol-lowering statins (pravastatin, fluvastatin, cerivastatin) on the properties of lipid layers mimicking intestinal membranes. Langmuir experiments supplemented with surface potential measurements, Brewster angle microscopy and spectroscopic results showed significant differences in the interactions of selected statins, which are mainly caused again by the differences in the drug hydrophobicity.

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Fluorinated Xerogel-Coated Carbon Fiber Microelectrodes for Brain Nitric Oxide Monitoring

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Nitric oxide (NO) is an important free radical synthetized and released by brain cells. It can modulate synaptic transmission and neuronal network activity but also mediate neuronal injury through oxidative stress. It is believed that NO exerts its physiological functions at low concentrations whereas oxidative stress occurs at much higher levels. However, the quantitative threshold at which NO concentrations become toxic is still poorly defined. Here, we detected endogenous brain NO release using 7 µm diameter carbon fiber microelectrodes first coated with a layer of nickel-porphyrin (Ni-P) and a screening layer composed of trimethoxymethylsilane and Heptadecafluoro-1,1,2,2-tetrahydrodecyl trimethoxy-silane. The fluorinated xerogel improved the selectivity of the sensor compared to Nafion, a fluoropolymer commonly used to block interfering molecules. Nitrite, 5-HT and AA amperometric detection was significantly reduced by the silane layer and NO detection was stable over 7 days of storage and throughout a 3h in vivo experiment. In vivo, these electrodes could quantify brain NO release evoked by a toxic local microinjection of the glutamatergic agonist N-Methyl-D-aspartate at 1.33 [0.49-4.93] µM. The amperometric signal was almost completely blocked by the NO-synthase inhibitor 7-nitroindazole. Fluorinated xerogel-coated carbon fiber microelectrodes therefore provide excellent stability, sensitivity and selectivity to detect brain NO and quantify its concentrations. Toxic NO actions like those evoked by the neurotoxin NMDA may take place in the low micromolar range.
Supported by CNRS, Inserm, University of Lyon., and grant FGC46-2016 from Fondations Gueules Cassées.

Poster Presentation

Hydrogen and/or Formate Facilitate Flavin-Based Electron Bifurcation in *Methanococcus maripaludis*

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Microbial methanogenesis is the main source of the greenhouse gas, methane. In hydrogenotrophic methanogens, hydrogen is oxidized to facilitate the reduction of carbon dioxide to methane. In methanogens without cytochromes, such as in *Methanococcus maripaludis*, capable of using hydrogen or formate as electron donor, the first step of carbon dioxide reduction is driven by low-potential electrons derived from electron-bifurcating heterodisulfide reductase coupling the exergonic reduction of heterodisulfide (CoB-S-S-CoM), which is required for the last step of methanogenesis, with the endergonic reduction of ferredoxin, which is required in the first step of methanogenesis.

Here, we show that formate-grown *M. maripaludis* contains three active heterodisulfide reductase complexes comprised of hydrogenase and/or formate dehydrogenase for electron bifurcation using hydrogen and/or formate, respectively. Overall, electron transfer occurs across 28 iron-sulfur clusters in addition to NiFeSe, Mo and flavin cofactors. We further found that a previously observed stimulation of electron-bifurcating heterodisulfide reductase activity by high concentration of phosphate was due to the promotion of hydrophobic protein-protein interactions between the ferredoxin domain of the heterodisulfide reductase complex and the commonly-used *Clostridium pasteurianum* ferredoxin endergonic electron acceptor. Our findings indicate that multiple complexes of heterodisulfide reductase exist under physiological conditions to utilize formate and/or hydrogen for carbon dioxide reduction.

Photo-induced Phenomena in Sediment Microbial Fuel Cells with Different Architecture

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The presented work summarizes the results obtained with sediment microbial fuel cells (SMFCs) with different constructions, size and electrode materials. In all cases, independently on the SMFC architecture, periodic fluctuations of current generation were recorded, following the natural day-night cycle. The analysis of the data, obtained with differently designed SMFCs, shows that the major factor affecting the light-dependent current oscillations is the anode arrangement in the reactor. In the SMFCs, where the anodes were buried deeply in the sediment layer, the current oscillations strictly followed the intensity of light irradiation. Additional analyses have shown that this response is connected with a formation of electroactive cathodic biofilm, containing photosynthetic microorganisms co-existing with an abundance of other bacteria and eukaryotes. In this connection, we hypothesize that during the photoperiods the in situ produced bio-oxygen on the cathode decreases the mass transport limitations for the oxygen reduction reaction, thus enhancing the kinetics of overall electrochemical process, resp. the current outputs. An inverse current response, observed with flat-SMFCs, is attributed to inhibition of the anodic reaction due to expansion of the oxic zone in the sediment. In this case, the oxygen, produced by existing photosynthetic bacteria, is able to reach the anodic bacteria and compete with the anode as an electron acceptor.

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Poster Presentation

Hydrogen Bonding and Ca^{2+} Affinity of Coenzyme Q_1 and Decylubiquinone

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Benzoquinones and their reduced forms, the hydroquinones, are a broad class of biologically active and physiologically relevant substances involved in essential cellular processes. Due to their central role in the crucial energetic processes in mitochondria, Coenzyme Q_{10} (Q_{10}) and its derivatives are the most prominent members in the family of quinones. The important physiological properties of coenzymes Q (CoQs) involve proton and electron transfers and are controlled by the benzoquinone functional group, while the side chain provides compatibility with hydrophobic membranes.

In contrast to the hydrophobic Q_{10}, Coenzyme Q_1 (Q_1) is much more hydrophilic, because it contains only one instead of 10 isoprenoid groups in its side chain. Another quinone compound, decylubiquinone (DUQ) contains a more lipophilic 10-carbon side chain. Because the length of the side chain does not strongly affect the redox properties of CoQs, Q_1 and DUQ are valuable models to explore the redox properties of Q_{10}.

For optimal voltammetry of their redox transformations, these systems are studied in organic aprotic solvents. Furthermore, aprotic media are to some extent mimicking the lipophilic membrane environment where these species are likely to be located depending on their side chain properties. Hence, in the present study, we investigated the electrochemical behavior of Q_1 and DUQ in acetonitrile.

In acetonitrile, both, Q_1 and DUQ undergo one-electron reduction to the semiquinone radical, which can be further reduced by one electron to form the dianion. The potential separation between first and second electron transfers allows following the effect of different hydrogen-bond donors and calcium ions on the semiquinone and dianion form of Q_1 and DUQ.

The voltammetry is shown to be strongly influenced by addition of hydrogen-bond donors (HB-donors), as the potentials of the first reduction and even more pronounced on the second reduction process are shifting to more positive values with increasing concentration of HB donors. At higher concentrations of HB donors, only one process was detected corresponding to the transfer of two electrons per molecule. Addition of Ca^{2+} ions produces a new peak, which is localized at potentials less negative than that of first reduction wave in the absence of Ca^{2+} ion, indicating interaction of semiquinone radical with Ca^{2+} ions.

Biocompatible Screen-Printed Electrodes: Cell Viability Investigations

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Screen-Printed Electrodes (SPE) are frequently utilised as a platform for the development of (bio)sensors.[1] The formulation of the ink strongly affects the electrode performance and allows its properties to be optimised depending on the application. An important emerging application is in smart-wearables and even implantable bio-sensors. These applications require biocompatible materials.[2]

This contribution describes the formulation of novel SPE inks that use biocompatible binders and avoid toxic solvents. The binder choice is key, as it influences key characteristics that can be tuned for the specific application of the electrode. Moreover, we demonstrate that these formulations allow the hydrophobicity of the printed electrode surface to be optimised, e.g., for the coupling of antibodies to their surface, or to minimise non-specific binding. The further refinement of the ink formulation, e.g., by incorporating metal nanoparticles to optimise conductivity or enhance detection sensitivity, e.g., using electrochemiluminescence, will be presented. Once printed, we have tested the effect of exposing these electrodes to cells in culture. In this protocol, we compared various commercial and in-house compositions of carbon and silver based screen-printed inks. Round disk electrodes were printed using the various ink formulations. HL-1 cardiomyocytes where initially cultured in a T75 flask[3]. Once they reached confluency, they were seeded into wells of a 6 well plate and allowed to establish overnight. Then the media was removed and replaced with 3 ml of supplemented Claycomb media. One electrode per well was laid face down in the media ensuring the ink surface was fully exposed to the media in the well. The cells where left to incubate in an incubator for set time points of 1,2,3,4, and 5 days without media change. Each pair of ink formulation was incubated with a control that did not contain ink to determine the natural decay of the cells without media change for a sustained period. To determine cell viability at the end of each time point, a cell viability assay was conducted. The cells were counted on a haemocytometer using Trypan blue dye to stain dead cells and the living cells were counted and adjusted for dilution to determine the cells per ml of viable cells remaining. These results were compared with the cells per ml of the corresponding control grown for the same period to give a relative determination of the toxicity of the ink during prolonged cell culture. Initial results indicate a significant difference in the relative viability of the cells using our custom ink formulations versus the commercial “biocompatible” ink formulations. The low toxicity of the novel screen-printed electrodes suggests significant potential for the development of biocompatible sensors.

Poster Presentation

Lercanidipine Electrochemical Behaviour at Carbon Black Modified Electrodes

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Lercanidipine, Figure 1, belongs to the 1,4-dihydropyridine group of compounds a new derivative used in the treatment of hypertension, angina pectoris and Raynaud's syndrome. It is characterized by potent, long-lasting, and vascular-selective calcium entry-blocking activity that reversibly blocks voltage-dependent Ca^{2+} influx through L-type channels in cell membranes, promoting peripheral vasodilation and a reduction in blood pressure.

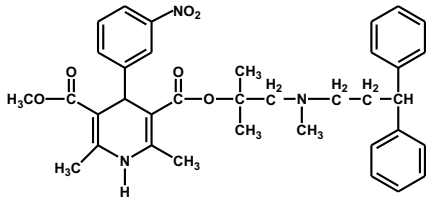

Figure 1: Chemical structure of lercanidipine.

The carbon black (CB) nanomaterial properties include high conductivity, chemical stability, large surface area, and CB is an extremely cheap material. An increase of the electrochemical peak currents and better detection limits was observed using the glassy carbon electrode modified with carbon black (GCE-CB) and a boron doped diamond electrode (BDDE-CB).

The redox mechanisms of lercanidipine at a GCE-CB and a BDDE-CB, using cyclic, square-wave and differential pulse voltammetry, was investigated.

The anodic oxidation is an irreversible process, two consecutive electron transfer reactions, pH-dependent mechanism. The reduction is an irreversible cathodic process. The lercanidipine reduction products are electroactive following a reversible electron transfer reaction.

The quantification of lercanidipine standards using CB-GCE and DP voltammetry, was performed. The detection limit found was LOD = 0.140 μM. The quantification of lercanidipine in ultrapure water, tap water and effluent samples, was determined.

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Poster Presentation

Viologen-modified electrodes for protection of hydrogenases from high potential inactivation

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Hydrogenases, including the O_2-tolerant classes, suffer from reversible inactivation upon applying high potentials, which limits their use in biofuel cell applications. Our previously reported protection strategy based on the integration of the hydrogenase into redox matrices enabled the use of these biocatalysts in biofuel cells even under anode limiting conditions. However, mediated catalysis required application of an overpotential to drive the reaction, and this translates into a power loss in a biofuel cell.[2,3] In the present work, the enzyme is adsorbed on top of a covalently-attached viologen layer which leads to mixed (direct and mediated) electron transfer processes; at low overpotentials, the direct electron transfer process generates a catalytic current, while the mediated electron transfer through the viologens at higher potentials generates a redox buffer that prevents oxidative inactivation of the enzyme. Consequently, the enzyme starts the catalysis at no overpotential with protection from inactivation at high potentials.[1]

Cartoon representing the viologen-modified electrode at electrode potentials lower (A) and higher (B) than $E_{V^{++}/V^{+\cdot}}$

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Poster Presentation

Decreased effectiveness of gene electrotransfer in acidic pH *in vitro*

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Gene electrotransfer (GET) is a method which uses electric pulses to enable intracellular DNA delivery.[1] *In vitro* GET is efficient, however gene expression *in vivo* is often weak, especially in tumor cells.[2] Since acidic microenvironment is one of hallmarks of tumors[3] we tested the effect of acidic medium on GET *in vitro*.

Adherent CHO cells were exposed to two different GET pulse protocols (shorter pulses: 4x200µs, 1Hz, 1.2kV/cm or longer pulses: 4x1ms, 1Hz, 0.8kV/cm) using Electrocell B10 electroporator in presence of pEGFP plasmid in acidic or neutral medium. Cells were incubated for 24h, then three images per well on a distinct area were recorded using a fluorescent microscope and counted by ImageJ program for image analysis.

We observed decreased transfection rates if GET was done in acidic medium. Lower transfection rates in acidic medium were observed with both pulse protocols. In neutral and acidic medium higher transfection rates were achieved with longer pulses. However both pulse protocols led to better cell survival if GET was applied in acidic medium.

In conclusion, we observed decreased transfection rates when GET was done in acidic medium, in spite of better cell survival after GET. Further experiments are needed to clarify if acidic medium decreases GET effectiveness by modulating plasmid DNA characteristics, cellular characteristics or both.

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Bioelectrochemistry of Cytochrome *c* in a Closed Bipolar Electrochemical Cell with Immiscible Aqueous-Organic Electrolyte Solutions

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Bipolar electrochemistry involves the generation of simultaneous asymmetric electrochemical reactions at the opposite ends of isolated conductive objects, known as bipolar electrodes, immersed in ionically conductive electrolyte solutions in the presence of an applied electric field (1). In this study, we investigated the bioelectrochemistry of Cyt *c* using immiscible aqueous-organic electrolyte solutions in a 4-electrode closed bipolar electrochemical cell (see Fig. 1).

In line with previous uses of this technique (2-3), our motivation is to acquire mechanistic insights for studies of interfacial electron transfer events involving Cyt *c* at the interface between two immiscible electrolyte solutions (ITIES). In the natural world, life is sustained by bioenergetic reactions taking place at soft liquid interfaces, *i.e.,* biomembranes. The ITIES is considered a simple model system that mimics biological interfacial processes and is therefore an ideal environment within which to study electron transfer with Cyt *c*, a natural redox shuttle.

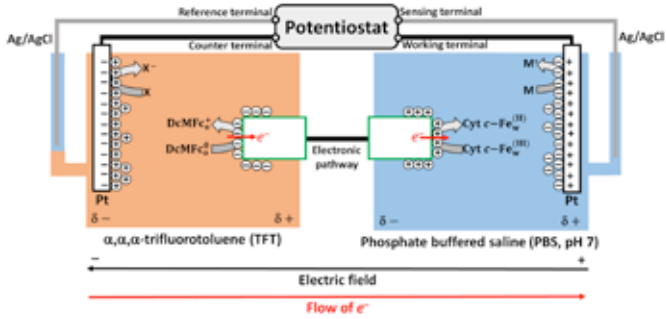

Fig. 1 Schematic of Cyt *c* reduction by lipophilic decamethylferrocene (DcMFc) in a 4-electrode closed bipolar electrochemical cell with immiscible aqueous-organic electrolyte solutions.

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Poster Presentation

Biosensor Development for SECM

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How does a fungus degrade lignocellulose? A lot is known about the extacellularly expressed enzyme machinery that a fungus is using to break down lignocellulosic structures into its molecular building blocks [1,2] but little to nothing is known about the actual *in vivo* ongoings, when a hyphe, growing inside a single wood cell, is releasing its lignocellulolytic enzyme cocktail. The ERC Consolidator Grant funded project OXIDISE (Grant agreement Nr: 726396) aims on the clarification of these ongoings using high resolution techniques such as SECM. This work presents preliminary data of a number of biosensors to detect and locate several reaction educts/products and enzymes involved in lignocellulolytic processes within a single wood cell with spatial respect to a fungal hyphe. Reaction educts/products involve (but shall not be limited to) pH, H_2O_2 as product of oxidases, cellobiose as product of cellobiohydrolases, syringol (2,6-dimethoxyphenol) as model product compound of lignin, cellobiose dehydrogenase and laccase. All sensors were electrochemically tested using gold-, carbon- or platinum electrodes applying chronoamperometry and the sensitivity and limit of detection are presented **(Table 1**). Further development of the presented sensors shall result in ultramicro- to nanoelectrodes with diameters of < 1 µm to resolve the desired measures of a single wood cell.

Table 1. Sensitivity and limit of detection of tested analytes.

Analyte	Sensitivity		LOD	
CDH	1.836	µA cm^{-2} mg^{-1} mL^{-1}	0.007	mg mL^{-1}
Laccase	1.556	µA cm^{-2} $µg^{-1}$ mL^{-1}	0.128	µg mL^{-1}
Cellobiose	0.003	µA cm^{-2} $µM^{-1}$	4.369	µM
Syringol	0.075	µA cm^{-2} $µM^{-1}$	13.714	µM
pH	63.6	mV pH-$unit^{-1}$	0.002	pH-units
H_2O_2	0.043	mA cm^{-2}	4.114	µM

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Poster Presentation

The first commercialized 3rd generation biosensor series LactoSens® for the detection of lactose in low lactose dairy samples

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Biosensors utilising enzymes capable of direct electron transfer are of great interest especially due to their low susceptibility to interfering species and the ease of production using less complex enzyme cocktails.
DirectSens GmbH is developing 3rd generation biosensors for the analysis of glucose and lactose based on the direct electron transfer of cellobiose dehydrogenase. Based on years of academic and industrial research [1, 2] we have commercialised the very first 3rd generation biosensor for the measurement of lactose in low lactose dairy products. The currently sold LactoSens R biosensor was developed with respect to high accuracy and precision, low susceptibility to interferents and high shelf life.
We will present the analytical performance of the sensor in various dairy samples with a broad range of interferents (e. g. vitamins, caffeine, fruit aromas). We will show its capability of detecting lactose as well or better as the currently used gold standard HPLC as validated by external laboratories (NordVal certified). We will highlight important lessons learnt from large scale sensor production responsible for obtaining high sensor shelf life.

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Poster Presentation

The Applicability of Pyrolytic Graphite Electrode Decorated by Silver Amalgam Particles in Bioanalysis

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Metallic mercury represents electrode material with unique electrochemical and mechanical properties. Low mechanical stability liquid mercury restrain its application in development of (bio)sensors. [1,2] Silver solid amalgam, represents up to now the most suitable mechanically stable and non-toxic alternative electrode material to liquid mercury in electrochemical applications. [3]

Pyrolytic graphite electrode (PGE) was used as a conductive surface for electrochemical deposition of silver amalgam particles. The electrodeposition process was optimized, tested to model organic nitro-compound and applied in protein and DNA (oligonucleotide) analysis using adsorptive transfer stripping-Constant current chronopotentiometric stripping analysis and adsorptive transfer stripping-cyclic voltammetry, respectively.

The controlled electrodeposition of silver amalgam particles from Ag^{+} and Hg^{2+} solution on the surface of PGE seems to be convenient method for preparation of new nanostructured electrode materials for electrochemical detection of biomolecules (proteins and DNA) in cathodic range of applied potentials.

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Poster Presentation

Micro- and Nanoelectrodes: Fabrication, Characterization and Application. The Detection of Silver and ROS/RNS in Biological Buffer

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The electrochemical determination of low concentrations of silver, and the short-lived highly reactive oxygen and nitrogen species (ROS/RNS), requires reliable, reproducible measurements with sensitive analytical methods. In this work, gold platinum and platinum black micro and nanoelectrodes were fabricated, and characterisation revealed flat disk-shaped working areas of 20 - 25 µm (micro) and 1 - 10 nm (nano) in diameter, derived from the steady-state limiting currents.[1] Reported studies on silver detection have been carried out under a myriad of conditions but, to date, electrochemical determination of silver in biological buffers and media has not been successful.[2] Ultra-low concentrations of silver (1 nM – 80 nM) were determined, by anodic stripping voltammetry (ASV) with Ag/AgBr as a reference electrode, using micro- and nanoelectrodes of platinum and gold in chloride-free phosphate buffer (PB, pH 7.4). Laser pulled electrodes exhibit highly reproducible stripping voltammetry for the determination of silver and returned calibration curves. A low detection limit in this medium provides the ability to explore silver and silver-based bioinorganic drugs uptake by tumour and microbial cells.

This study also includes the fabrication of Pt/Pt-black nanoelectrodes for detection of ROS/RNS in chloride-free PB (pH 7.4). These data should help with the identification and quantitation of cellular ROS/RNS release. In particular, monitoring oxidative stress, as a cellular response on exposure to bioinorganic drugs, should help to elucidate the interaction pathway of bioinorganic anti-cancer drugs. We envisage that electrochemical speciation studies will help to elucidate the mechanism of action and cellular uptake of bioinorganic silver-based anti-bacterial/fungal and anti-cancer drugs in cell sustainable conditions for *in vitro* and, potentially, *in vivo* single cell analysis.

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Poster Presentation

Single Cell Electrochemistry – Uptake of Bioinorganic Silver-based Drugs

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Heterogeneity in a cell population is a universal phenomenon in all biological systems including whole tissues, and cell cultures.[1] Intra-tumour heterogeneity has been widely reported for decades from morphological perspectives, whereas phenotypic and genotypic heterogeneity has also been detected.[2] DNA platination is widely assumed as the central mode of action for most platinum anti-cancer drugs. This generalisation on the mode of action for conventional chemotherapy might be a hindrance for effective drug development. Clinical approval for most novel anti-cancer drugs were denied, due to uncertainty in optimal usage, and the mode of action is unclear.[3] For a better understanding of the variations, from cell to cell, in response to metal-based drugs, single cell analysis is the key. Single cell experiments should provide critical information about the pathway and disease state, which will guide personalised medicine and therapeutic strategies. The efficacy of tumour-specific metal-based drugs depends primarily on its redox nature. Therefore, electrochemistry is the foremost approach for studying the uptake of metal-based drugs on single cell analysis. This strategy involves positioning a micro or nanoelectrode inside or near the surface of a single cell for the electrochemical monitoring of individual cellular events.[4]

In this work, the aim is to exploit single cell electrochemical analysis, to study the uptake of silver-based bioinorganic drugs and to elucidate the mode of action. Single cell electrochemistry experiments will be carried out using the platinum, gold and platinum-platinum black nanoelectrodes on human lung carcinoma (A549) and human breast adenocarcinoma (MCF-7) cell lines. The speciation of the silver-phenanthroline bioinorganic drugs will be examined using stripping voltammetry for metal detection, and the redox behaviour analysis will be determined by cyclic voltammetry for *in vitro* and *in vivo* single cell analysis.

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Voltammetric Detection of Anthraquinone-2-Sulfonate After its Separation by Means of a Microdialysis Catheter

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The contribution is focused on therapeutic anti-cancer drugs monitoring in blood vessel. Individualized anti-cancer drugs dosages could be optimized based on a feedback of measurement of their blood concentration. It should lead to lowering their toxic effects on the body and other major adverse reactions [1]. For these purposes, a microdialysis catheter combined with electrochemical detection was used. The catheter was connected to the injection pump with flow rate 1 and 5 μL min^{-1} and it was placed in a test tube containing a solution of natrium anthraquinone-2-sulfonate (as a model substance for anticancer drug of Doxo/Daunorubicine type) dissolved in a physiological solution (154 mmol L^{-1} NaCl), which was also used as a carrier solution. The dialysate was collected in a vial and after that it was analyzed by differential pulse voltammetry with three-electrode system: a polished silver solid amalgam working electrode, Ag|AgCl (3 mol L^{-1} KCl) reference electrode, and a platinum wire auxiliary electrode (diameter of 1 mm) and by UV/VIS spectrophotometry as a comparative method. The recovery was ca 54 % and 20 % for flow rate 1 μL min^{-1} and 5 μL min^{-1}, respectively, in the range of concentrations from 1.10^{-3} to 1.10^{-6} mol L^{-1}.

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Poster Presentation

Calcium Affinity of Coenzyme Q_{10} and its Hydroxylated Derivative

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Coenzyme Q_{10} belongs to a family of compounds called ubiquinones. Its chemical structure consists of a 2,3-methoxy-5-methyl benzoquinone ring with a side chain of 10 isoprenoid units at ring position 6. CoQ_{10} is the predominant ubiquinone form synthesized in the human body, present in plasmalemma and endomembranes of the cells. As an essential part of the electron transport chain, located in the inner mitochondrial membrane, CoQ_{10} plays a key role in energy production. Besides the function as a mobile electron and proton carrier, CoQ_{10}, in its fully reduced form, acts as an important cellular antioxidant to reduce the damaging effects of reactive oxygen species (ROS) production. Despite this, the partially reduced semiquinone form can lead to free radical generation (superoxide) and is therefore prooxidant.
Since the redox reactions of CoQ_{10} are very important, they have been extensively studied for many years. Due to the lipophilicity of CoQ_{10}, most of the electrochemical studies are performed in non-aqueous solutions. These conditions do not represent the real environment of CoQ_{10} in living cells. Therefore, we developed a new and simple method for investigating the redox properties of CoQ_{10} in a biomimetic system, using NH_2-functionalized multiwalled carbon nanotubes (NH_2-MWCNTs) modified glassy-carbon electrode (GCE).
Various studies of quinones have been dedicated to investigate their ability to bind biologically relevant metal cations. Metal cations play a crucial role in living processes. They are involved in many enzymatic reactions, regulatory pathways, and electron transfer processes. Considering the important role of calcium in regulating mitochondrial function and the presence of CoQ_{10} in the mitochondrial membranes, studying the calcium affinity of CoQ_{10} is of great importance. Here, we report detailed voltammetric analyses of the calcium affinity of CoQ_{10} in buffered and unbuffered solutions using the Q_{10}@NH_2-MWCNTs modified electrode. In a different set of experiments, the ability of CoQ_{10} for binding calcium was studied in alcoholic solutions.
The hydroxylated derivative of CoQ_{10}, HO-CoQ_{10}, was included in the analyses as well. It is synthesized by exposing CoQ_{10} to high pH, so that one methoxy group is substituted by a hydroxyl group. HO-CoQ_{10} can be also generated in presence of cytochrome P450 and is part of the quinone pool in beef heart mitochondria. We found that both CoQ_{10} and HO-CoQ_{10} have affinity to bind Ca^{2+} in alcohols that may be of great importance for their function in cell membranes.

Abiraterone Conjugated with Gold Nanoparticles for Anticancer Drug Delivery Application and Therapeutic Performance

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Gold nanoparticles (AuNPs) conjugated with drugs are used in diagnostics and therapies [1-3]. For many years research has been carried out to form efficient drug carriers. In our work we have shown a novel method for the successful conjugation of abiraterone (AB) and gold nanoparticles (AuNPs-AB), thus obtaining a unique nanoconjugate with expected anticancer properties and biomedical applications. The direct coupling of AB with gold constitutes an essential feature of the unique AuNPs-AB conjugate that creates a promising platform for future applications in nanomedicine. In creating AuNPs-AB, abiraterone acted as both a reducer and stabilizer. The conjugates were characterized in terms of particle size, surface charge, morphology, drug loading and identity by several techniques, including transmission electron microscopy (TEM), zeta potential, dynamic light scattering (DLS), electrochemistry, UV-Vis, TGA, XRPD, NMR and Raman spectroscopy. We have also synthesized a new analogue of abiraterone - thiolated abiraterone (HS-AB) and designed a gold surface monolayer using HS-AB. As a result of immersing the HS-AB modified gold electrode in an aqueous solution containing $HAuCl_4 \times 3H_2O$, gold nanoparticles were formed, which was confirmed by AFM and electrochemistry. The voltammetric experiments also confirm the formation of gold structures with specific catalytic properties in the process of oxygen reduction.

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Poster Presentation

Fast and Sensitive Detection of Methylglyoxal as an Important Advanced Glycation End Product in Oral Pathology

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The pathology of oral cavity is closely correlated with dietary intake. Advanced glycation end products (AGEs) are associated with the processed food and thus, can be ingested by consuming these types of food items. The majority of AGEs, including methylglyoxal, are considered markers involved in oxidative stress by producing long-term damage to humans [1]. A novel approach for the sensitive electrochemical detection of methylglyoxal has been developed. A lab-made screen-printed platform was developed using a flexible plastic foil as support. An Ag/AgCl conductive ink was employed for the design of the reference electrode and the contacts, whereas carbon ink served for the counter electrode and a carbon ink modified with graphene oxide was used for the working electrode. These disposable sensors were easily produced and tested for direct detection of methylglyoxal using differential pulse voltammetry. The employment of graphene oxide exhibits electron-mediating effect, which leads to a well-defined reduction peak associated to the methylglyoxal. A wide dynamic range and low detection limit were achieved for the target analyte. The developed assay was shown to be specific and sensitive for the analysis of salivary methylglyoxal in healthy volunteers and patients suffering of diet-related diseases.

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Electrochemical Detection of N$^{\epsilon}$-(carboxymethyl)lysine in Raw Human Saliva

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N$^{\epsilon}$-(carboxymethyl)lysine (CML) is a well-characterized advanced glycated end product (AGE), its high levels being correlated with oxidative stress and long-term damage to proteins in ageing, atherosclerotic plaques and diabetes. AGEs are usually detected using ELISA, immunohistochemistry or HPLC, methods that require long time for analysis and implicit delayed diagnostic results. [1, 2]. The present work describes a printed electrochemical sensor for the direct salivary detection of CML. The flexible and bendable sensor was integrated on a customized mouthguard and further placed on a phantom jaw that imitated the structure of the human oral cavity. The disposable sensor was readily attached on the mouthguard before usage and detached after the electrochemical investigation. Thus, it can be replaced whenever required. The sensor was tested by employing a differential pulse voltammetry analytical procedure and exhibited high selectivity and sensitivity for CML (limit of detection of 0.1 µg/mL; dynamic range of 0.5–2500 µg/mL) [2]. The optimized sensor was tested in raw, untreated human saliva collected from healthy volunteers and patients suffering from diet-related diseases and good recoveries were obtained. This non-invasive oral sensor for salivary CML monitoring could potentially provide useful real-time information regarding a wearer's health condition, and thus, it holds considerable promise in improving the management of chronic diseases.

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Poster Presentation

Electrochemical Immitance Spectroscopy and Multivariate Data analysis for Analysis of a Therapeutic Drug - Methotrexate

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Monitoring of therapeutic drugs involves measuring drug concentrations in body fluids such as plasma, serum or blood. This information is then used to individualize dosage based on the pharmacokinetics profile obtained by quantification of the drug concentration after administration. Today, analysis of cytostatic drugs and their metabolites is often based on chromatographic techniques such as high-performance liquid chromatography (HPLC) and gas chromatography (GC), hyphenated with mass spectrometry (MS) [1]. Liquid Chromatography is by far the most extensively used method with drawbacks such as consumption of considerable time and large volume of organic solvents. The hyphenation of LC with MS (LC/MS) also leads to expensive instrumentation and requires a skilled operator to run the analysis.

The current research project concerns development of a biosensor based on electrochemical immitance spectroscopy, for monitoring the level of cytostatic drugs in patients undergoing cancer treatment as an ultimate goal. Methotrexate (MTX) which is one of the most common therapeutic drugs is used as a model drug.

Sensors that are based on electrochemical methods have been proposed as sensitive, cost-effective and simple analytical tools. However, most of the methods which have been used for MTX detection depend on the faradaic electrochemical behaviour of MTX at low pH and application of potential (0.2 to 1.2 V *vs* Ag/AgCl) [2]. Immunosensors based on electrochemical immittance spectroscopy (EIS) are an alternative type of sensors. Recently we reported a non-faradaic method for determination of MTX at physiological pH using electrochemical immittance spectroscopy (EIS) and multivariate data analysis [3,4].

The attachment of antibody on the electrode surface was followed by EIS and confirmed by X-ray photoelectron spectroscopy (XPS). Mutivariate data analysis, using singular value decomposition, was used for data analysis and constraction of a calibration model with good linearity, on a logarithmic scale, in the concentration range of 3×10^{-12} to 3×10^{-4} mol L^{-1} offering a detection limit of 5×10^{-12} mol L^{-1}.

In a parallel project we are investigating the performance of carboxyphenyl layers, covalently electrografted onto gold surface, as a platform for the construction of an immunosensor [5].

Interaction of Selenite with Metallothionein

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Some selenium compounds are potent inhibitors of the cell growth with remarkable tumor specificity and their use in attempts to treat cancer has a relatively long history dating back to at least 1912 when selenite was reportedly used to cure a tongue cancer. The function of selenium relates to its role as an antioxidant. It is as a constituent part of glutathione peroxidase important in the detoxification of peroxides, which leads to a reduction in the level of reactive oxygen species in cells and tissues. A remarkable feature of selenium consists of its ability to oxidize thiols under reducing conditions and one mode of action recently suggested is the oxidation of thiol groups of metallothionein. Metallothionein II (MT) is a cytosolic, ubiquitous, low-molecular-weight protein present in various tissues of mammals and non-mammals. A high content of thiol groups (–SH) of MTs can bind mineral micronutrients and xenobiotic heavy metals. In this study, we investigated the electrode processes of MT at a mercury electrode in the presence of sodium selenite (Na_2SeO_3) by means of the Brdička reaction in the differential pulse voltammetric mode. The interaction between MT and Na_2SeO_3 were analyzed via the hydrogen evolution catalytic signals Cat2. It was found that with the increasing selenite concentration, cobalt in MT is replaced by selenium. When concentration of Na_2SeO_3 increases above the MT binding capacity (MT cannot bind more Se), only selenite ions can be responsible for Cat2 signals. We have answered the questions: (i) how selenite participates in the Brdička reaction, (ii) which competitive behavior of selenium against cobalt should be expected and (iii) what is the sequence of reaction processes in the modified Brdička reaction. A new interpretation leading to complete description of the mechanism is presented. Our results can be helpful in biochemical and clinical studies involving selenium compounds as potential chemotherapeutics.

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Poster Presentation

Amperometric Choline Biosensor with Enzymatic Reactor in Flow Systems: Fabricating, Optimization and Application

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Choline (Ch) plays an essential role in human health as a precursor to neurotransmitter acetylcholine. Moreover, it is a quite important for healthy functioning of liver and in woman's health during the pregnancy. Therefore, there is an ever increasing demand for methods of its detection in biological samples and pharmaceutical products.

This work is focused on the development, fabrication and application of the perspective type of long-time functional amperometric enzymatic biosensor based on choline oxidase (ChOx) which consists of the separated enzymatic (reactor) and detection (working electrode) part. The Ch detection is based on amperometric monitoring of oxygen consumption in the enzymatic reaction, which is directly proportional to the Ch concentration. The measurements were performed in flow-through system with tubular detector of polished silver solid amalgam and in the wall-jet arrangement with the working silver solid amalgam electrode covered by mercury film. Two mesoporous silica powders have been used and compared as the filling materials in the enzymatic reactor because of their inertness, good biocompatibility, and large surface area. The enzyme ChOx has been covalently anchored to the filling material surface, which provides stability and long life-time of the biosensor.

pH of the mobile phase, flow rate, detection potential, injection volume, concentration of the enzyme solution, time required for the recovery of ChOx activity, and volume of the reactor have been optimized. The proposed amperometric biosensor with ChOx-based reactor possesses good repeatability, long-term stability, and sufficient sensitivity. Its applicability has been tested for the determination of Ch in commercial pharmaceutical products.

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Graphene derivatives and their application as platforms for aptasensors

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Increased health care requirements have resulted in a need to develop sensitive, reliable and cost-effective tools for the quantitative detection of biomolecules relevant in medical diagnosis and biomedical research[1]. At the same time, recent advances in nanotechnology have generated a variety of nanomaterials with unique intrinsic properties. Moreover, nanomaterials can be easily synthetically modified with appropriate functional groups to tailor their physico-chemical as well as their structural and dimensional properties. In turn, such nano-engineered platforms offer reproducible immobilization of bioreceptor units, enhanced sensitivity for targets and increased biocompability[2]. In the view of all above mentioned reasons, aptasensors based on different types of nanomaterials along with the combination of electrochemical signal output (i.e. impedance spectroscopy and/or pulse techniques) might offer simple, fast as well as low-cost tool for clinical practice[3].
In this work, we suggested two different graphene derivatives for effective sensing of thrombin and myoglobin. First, hierarchical porous sensing platform based on fluorinated graphene oxide and iron-based metal–organic gel (FGO@Fe–MOG) nanocomposite was used for thrombin detection[4]. Further, myoglobin detection was successful using cyanographene functionalized with gold (GCN-Au). In summary, both proposed label-free aptasensor showed remarkable sensitivity with the picomolar detection level, reproducibility over the time and high selectivity as well as it showed its applicability in real matrix (i.e.blood serum).

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Poster Presentation

Interplay of redox potential and pK_a in aminoferrocene

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Due to the simplicity of the redox reactions of $Fe^{2+/3+}$ ions, ferrocenes are popular for electrochemical measurements. While ferrocene itself is rather nonpolar for direct use in aqueous solvents, it's many derivatives (aminoferrocene, ferroceneboronic acid etc.) are soluble enough to be of use in aqueous solvents.

However, improved solubility is not the only change to the ferrocene properties, due to introduction of protonizable group, pH sensitive behavior is expected. The pH-dependent behavior is not completely straightforward as one could expect, electrochemical behavior drastically differs depending on the presence of buffer in solution (figure). On the other hand, not only pH affects the electrochemical behavior, also the electrochemical processes can affect pH-dependent behavior - the pK_a of the reduced/oxidized molecules.

So, deducing the mechanism behind the effect of buffer hints at very interesting interplay between the pH sensitive group and the redox state $Fe^{2+/3+}$ ions, where one could be controlled by the state of the other, leading to the possibility of pH/redox switches.

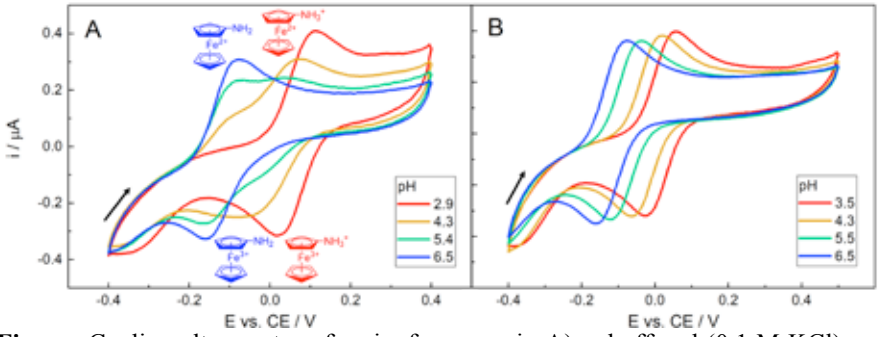

Figure: Cyclic voltammetry of aminoferrocene in A) unbuffered (0.1 M KCl) and B) buffered solution (0.1 M acetate).

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Boron doped diamond microelectrodes: Fabrication, spectral and electrochemical characterization for estimation of coverage quality

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Current technologies enable fabrication of boron doped diamond (BDD) electrodes in many shapes and formats. With micro-sized architecture, they succeeded in *in-vitro* and *in-vivo* sensing of biogenic compounds [1].

In this contribution, BDD microelectrodes (BDDμE) prepared using linear antenna microwave plasma enhanced chemical vapour deposition process (MW–LA–PECVD) [2]. Tungsten wires (d = 20 μm) were used as substrates on which BDD was deposited and then embedded in glass to produce a functional electrode. SEM and Raman spectroscopy revealed inhomogeneous coverage with areas of non-coverage for some wires. Evaluation of the potential windows in different supporting electrolytes (1.0 mol L^{-1} KCl, 1 mol·L^{-1} HNO_3, 0.5 mol·L^{-1} H_2SO_4, and 0.1 mol·L^{-1} H_3PO_4, phosphate buffer pH range pH 2.0–7.4) was performed using cyclic voltammetry. For BDDμEs with non-covered areas, potential windows are shortened in the positive region for solutions of pH > 5. Nevertheless, in acidic media reasonable electrochemical performance was still detected with potential windows ranging from −0.7 V to + 0.7 V, thus enabling estimation of the electrode surface area using $[Ru(NH_3)_6]^{2+/3+}$ redox marker. BDDμE with uniform coverage were characterized using redox systems $[IrCl_6]^{4-/3-}$ and $[Fe(CN)_6]^{4-/3-}$ and exhibited extended potential window to +1.6 V in acidic media. Therefore, these simple electrochemical measurements represent an easy tool to recognize the uniformity of coverage by BDD.

[1] N. J. Yang, et. al., Chem. Soc. Rev. 48 (2019) 157-204.
[2] A. Taylor, et. al., Carbon 128 (2018) 164–171.

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The financial support was provided by Czech Science Foundation (project 17–15319S) and Charles University (SVV 260440, GAUK 1390217).

Poster Presentation

Electrochemical and Spectral Characterization of Flavo[6]helicene and its Application in Bioanalysis and Materials Science

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Helicenes are functional polyaromatic substances with a broad spectrum of applications, especially in the field of optoelectronics, organic semiconductors, biosensing and separation processes, see ref. [1].
Flavo[6]helicene, 15,18-dimethyl-15H-[6]heliceno[2,1-g]pteridin-17,19-dione, was synthesized by reacting 2-nitroso[6]helicene with 3-methyl-6-(methylamino)uracil. The racemic mixture, and subsequently prepared pure P and M enantiomers (Fig. 1), were characterized using nuclear magnetic resonance, UV-vis, fluorescence and Raman spectroscopy, Raman optical activity measurement, circular dichroism spectroscopy, circularly polarized luminescence, and also using electrochemical methods.
In this contribution, we focused on the application of flavo[6]helicene enantiomers in the development of bioanalytical tools and advanced chiral composite materials. The mechanistic study targeting the application of flavo[6]helicene as a molecular DNA structural probe is also highlighted.

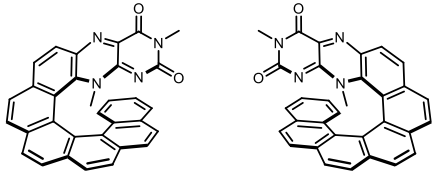

Fig. 1. M and P enantiomers of flavo[6]helicene.

The authors wish to thank the Czech Science Foundation (GACR 19-21237Y) and OP EIC Operational Programme (CZ.01.1.02/0.0/0.0/16_084/0010374).

Reference

[1] J.Vacek et al., *ChemElectroChem* 15, 2080–2088; 2018

Study of H_2O_2 kinetics of an immobilized Lytic Polysaccharide Monooxygenase with FTacV

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Evaluation of the kinetics of immobilized redox enzymes with cyclic voltammetry could give directly the rate of the enzymatic reaction with the substrate and would lead to the extraction of kinetic constants. However, this would require the feasibility of the direct electron transfer between the electrode surface and the redox enzyme as well as the absence of capacitance currents, which is nearly impossible due to relatively low enzyme loadings immobilized on electrode surfaces. In order to deprive of the capacitance currents, one solution could be the use of large amplitude Fourier transform alternate current cyclic voltammetry (FTacV), so as to extract the kinetic constants. In the present work, simulations are run in order to find the expression for the dependence of the kinetic rate of each harmonic, using the finite element method. An experimental methodology is suggested, which is applied for the complete study of the reaction of an immobilized lytic polysaccharide monooxygenase (LPMO) originating from the thermophilic fungus *Thermothelomyces thermophila*. The optimum pH and temperature of the enzyme are calculated from its catalytic efficiency towards H_2O_2, indicating that FTacV is a valuable analytical tool for the study of the activity of redox enzymes.

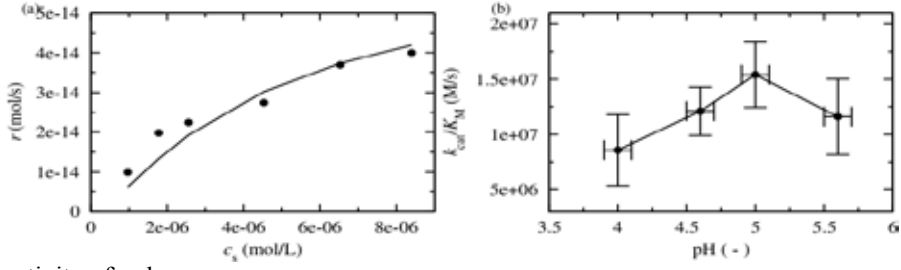

Figure 1: (a) Fitting of experimental data of the reaction rate from the 5th harmonic

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