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About us

According to the Agreement between UNESCO and Romanian Government, the International Centre of Biodynamics-ICB has been established through the decision No. 1378/2000 of the Romanian Government, under the aegis of UNESCO, as a non-profit organisation of general interest and public utility to pursue graduate training, research and development initiatives in the field of Biodynamics.

ICB focuses on noninvasive, real time assays and (non) linear data analysis for appraisal of biosystems or/and of their dynamics. In a multidisciplinary approach, it integrates precise measurements into detailed models (of the dynamics) of living systems and biointerfaces, taking into account the complexity of the interaction between biological structures and the measuring systems.

The potential applications of these methodologies have far reaching implications for Ecology, Food industry, Bio-Medicine and Pharmaceutical industry. Some examples are: fast detection of analytes, e.g. contaminants or/toxic compounds in liquid media, appraisal the efficacy and cytotoxicity of chemical compounds, biomass monitoring etc.
Our Mission

The International Centre of Biodynamics (ICB) is a flexible, highly dynamic structure active on the national and international research areas. It initiates and coordinates research related to noninvasive, sensitive and cost effective methods to analyze and control biosystems. The applicative domains span BioMedicine, Food Industry and Ecology. ICB aims to maintain a balance between fundamental and applied research.

ICB conducts and is involved in a number of collaborative national and international research projects having the following headlines:

- Characterization techniques for cellular systems, with emphasis on noninvasive multi-parameter real-time monitoring
- Detection of contaminants and adulterates in food products and water (microorganisms, heavy metals, antibiotics & toxins)
- Monitoring cell cycle progression & Biomass assessment
- Biomedical applications - pre-clinical, in vitro assays to assess the interaction mechanisms between selected drugs and cellular structures as well as related cytotoxic effects
- Monitoring & Nonlinear Analysis of the external stimuli (e.g. drugs, toxins, pathogens, EM radiation, pollutants) effects on: (1) evolution of cellular systems and (2) fish behavior
- ICB aims to provide a competitive research environment supporting the (re) integration of valuable researchers and formation of new ones.
Important Dates in the Institute’s History

November 1996: The proposal to create the International Centre of Biodynamics, ICB, was discussed at the UNESCO Centre of Membrane Science and Technology, UNSW, Sydney, Australia

April 1998: The first International Symposium on Biodynamics

October 1999: The Agreement between Romania and UNESCO to establish the ICB

December 2000: Romanian Government issued the Decision No. 1378/2000 to establish ICB

January 2001: The ICB commences its independent activities; starts AFRAMILK, the first European Research Project (under FP5) in ICB’s portfolio

March 2001: Romanian Parliament passes the law 110/2001 to ratify the Agreement between Romania and UNESCO to establish the ICB

December 2001: In Agreement with UNESCO the first Management Board of the ICB is appointed by Romanian Ministers of Education and Research

April 2004: Appointment of the first International Advisory Board of ICB

April 2005: Starts ROBIOS, the first European Project (under FP6) coordinated by ICB

April 2006: ICB activities start in the new lodging

May 2006: The International Conference on Biosensing and Biodynamics: From Basics to Applications co-financed by UNESCO

April-May 2008: The II
  nd European Young Investigator Awardees Symposium EURVIAS 2008- Self-organization and Selection in Evolution of Matter, Molecules and Life

July 2012: Starts EXTRACELL – the first ERC- like project “Monitoring the extracellular space with catalytic self-propelled nanomotors”, hosted by ICB

July 2012: Starts BIOSCOPE – the first Complex project “Electro-Plasmonics for the analysis of the dynamics of cellular processes and biomolecular interactions”, coordinated by ICB

Director's Message

This Annual Report highlights the major ICB accomplishments during 2016 with emphasis on both the progress of ICB research and the capabilities of ICB staff and laboratories for continuous development.
I acknowledge with pride the landmarks of 2016:

Two international projects started in 2016:

✓ FLAG ERA Graphene Flagship - GRAPHTIVITY: Graphene-based optoelectrochemical sensor for the simultaneous monitoring of the electrical and chemical activity of single cells, Project Director: Szilveszter Gaspar

✓ NATO-SPS 985042: Cell Biosensors for Detection of Chemical and Biological Threats. Project Director: Eugen Gheorghiu

Complemented by Manunet II project SENS4WINE which has been proposed and recommended for funding in 2016: New automated system based on biosensors for winemaking monitoring and assessment of allergen risk along the wine production chain

❖ The projects supporting ICB activity:
  o BIOSCOPE, Electro-Plasmonics for the analysis of the dynamics of cellular processes and biomolecular interactions, Contract No. 11/2012, ID: PN II-ID-PCCE-2011-2-0075, coordinated by ICB; Project Director: Eugen Gheorghiu;
  o ESPRIm - Single-molecule detection of DNA hybridization, based on electrochemical surface plasmon resonance microscopy and magnetic tweezers, Contract No. 29/2015, ID: PN-II-RA-TE-2014-4-2363, Project Director: C. Polonschi;
  o ANTIOXWIN-Exploiting the antioxidant capacity of black grapes for producing wines with high authentic quality – Contract No. 101/2012, ID: PN-II-PT-PCCA-2011-3.1-1809; Project Director: A. Vasilescu;

And 5 applicative projects proposed and approved in 2016:

• FIND PATHOGEN - Advancement of a Portable System for Fast and Sensitive Detection of Pathogenic Cells, Contract No. 2PED/2017, ID: PN-III-P2-2.1-PED-2016-1041, Project Director: E. Gheorghiu

• GRAPHTOOL - Graphene-based, miniaturized, electrochemical tool for the investigation of tumor cell pH regulation (GrapHtool), Contract No. 110PED/2017, ID: PN-III-P2-2.1-PED-2016-1106, Project Director: S. Gaspar


- **COLDSENSOR** - Novel aldehyde dehydrogenase from Antarctic bacterium as highly efficient catalyst for low temperature biosensing and biotechnologies, Contract No. 1PED/2017 PN-III-P2-2.1-PED-2016-0116, Coordinator: The Institute of Biology of the Romanian Academy, Partner institution: International Center of Biodynamics Partner responsible: A. Vasilescu

⚙️ New scientific developments

**International Patents:**

**Papers:**

**Published:**

**Accepted:**
serum on Micrococcus lysodeikticus-modified graphene oxide surfaces, accepted, *Biosensors and Bioelectronics*

Submitted:

**Book chapters:**

These accomplishments could not have been possible without the constant support, motivation and efforts of ICB staff and collaborators as well as of ICB Management and International Advisory Boards that fostered the continuous increase of Centre’s RTD capabilities and visibility during 2016.

*Thank you!*

*Eugen Gheorghiu*
Organizational Chart

**ICB Functional Structure**

- Cell Cultures Laboratory
- Electrochemistry Laboratory
- Applied Electrochemistry Laboratory
- Microfabrication & Prototyping Laboratory
- Bioanalysis Laboratory (including a mobile laboratory)
- Electrical Measurements Laboratory
- Impedimetric and Plasmonic Biosensors
- Optical Microscopy Laboratory
- Modeling and Data Analysis Laboratory
- Pilot Laboratory on Monitoring Fish Behavior
- Atomic Force Microscopy Laboratory
The Managing and Advisory Boards

The International Advisory Board, the Managing Board and the Director coordinate the activity of the International Centre of Biodynamics.

According to ICB statute, the current Managing Board was appointed in 2013 by the International Advisory Board.

Managing Board members (in alphabetical order):

- Mircea Dumitru - Rector, University of Bucharest, Corresponding Member of Romanian Academy
- Alexandru Morega - University Politehnica of Bucharest, Corresponding Member of Romanian Academy
- Romeo Resiga - Polytechnic University of Timisoara
- Octavian Popescu – Babes-Bolyai University of Cluj-Napoca, Member of Romanian Academy
- Nicolae Zamfir- Director General, Institute for Nuclear Physics and Engineering “Horia Hulubei”, Member of Romanian Academy

The members and observers of the International Advisory Board (in alphabetical order):

- Prof. Dr. Koji ASAMI
  Japan
- Prof. Dr. Anton ANTON
  Representative of Romanian University Research Council
- Prof. Dr. Jean-Michel KAUFFMANN
  Universite Libre de Bruxelles, Belgium
- Prof. Dr. Maciej NALECZ
  Director, Division of Basic and Engineering Sciences UNESCO –Paris, France
  Director, UNESCO – CEPES
  Minister of Education and Research

Representative of the European Commission, to be nominated
... and us

Eugen Gheorghiu – 2001*
ICB Director
Research Professor
PhD, Physicist, theoretical physics
EIS&SPR assays
Modeling & Data analysis

Dumitru Bratu – 2001*
Engineer Electronics
Development of Electrochemical devices

Szilveszter Gáspár – 2006*
Head of Electrochemistry Laboratory
PhD, Chemist
Electrochemical biosensors

Mihaela Gheorghiu – 2001*
Head of (Bio) Sensors, (Bio) Surface
EIS&SPR biosensing
Analyses and Cell culturing Department
Research Professor, Physicist
Electro - Optical Analysis of Biointerfaces

Mihai Sorin David – 2003*
PhD, Biophysics/Biodynamics
EIS&SPR biosensing
Bioprocess Impedance Measurements, Biosensor
Development and
Nonlinear Analysis

Cristina Polonschii – 2006*
PhD, Biophysics/Biodynamics
EIS&SPR biosensing
Development of analytical methods to
interrogate bio interfaces

Alina Vasilescu – 2011*
PhD, Analytical Chemistry
EIS&SPR biosensing
Practical applications of Biosensors

Luciana Stanica – 2012*
PhD student
Biologist
Electrophysiological analysis

Mihnea Rosu Hamzescu – 2012*
PhD student
EIS&SPR biosensing

Florica Moranescu – 2011*
Cell culture technician

Cezar Giubalca – 2001*
Head of the Administrative Office
Economist

Valeria Nane – 2001*
Chief Accountant
Economist

* Date of employment at ICB
Cell Cultures Laboratory

Team: Mihaela Gheorghiu
Luciana Stanica
Florica Moranescu

Facilities

- Cell cultures facilities that enable cultivation of different epithelial tissues: cell collection which contains renal epithelial cells (A6, MDCK-I, MDCK-II, MDCK parental, LLC-PK1), intestinal epithelial cells (CaCo-2), epithelial cells from a cervical carcinoma (HeLa), cells derived from an adrenal medulla (PC12); Neuroblast from human neural tissue SH SY-5Y, optogenetically engineered HEK 293 cells containing bacterial channel rhodopsin; HT29 cancer cells expressing Carbonic anhydrase IX under hypoxic conditions; CO₂ incubators (from Sanyo Biomedical Division–U.K), cryogenic storage facilities and deep freezer, Two microelectrodes Voltage Clamp (TEVC) system, Osmometer (from Gonotec Germany), Transepithelial, impedance and noise system (TINS), inverted microscope; microinjection setup and support peripherals (pumps, pipette puller, Faraday cages, dedicated measurement chambers and antivibration table); hypoxic chamber, computer controlled LED illuminators.
- other lab utilities (purified air, microbiological hood class II, ultra pure water)

Current Research

In the Cell Cultures Laboratory we have developed a battery of methods to study the effects of external factors (pharmaceutical compounds or bacteria) assessing several useful parameters. The methods consider:

A. Analysis of transport and cellular permeability;
B. Toxicity studies at cellular level (changes in transport properties and cellular viability);
C. Changes in cell surface attachment and cell-to-cell communication upon interaction with stressors;
D. Cell behavior under hypoxic conditions;

This expertise has been advanced with optogenetically engineered HEK 293, hypoxic conditions and multisensory approaches towards continuous opto-electrical evaluation and integration of cells in biosensing platforms.

Accomplishments

- Impact of Controlled Illumination on stable Cell Lines Containing Light Activated Channels (Channel Rhodopsin)
• A novel biosensing method based on an exogenous driven rhythm induced using optogenetic tools and assessment of the dynamics of cell response and recovery as reporter processes of bioactive analytes’ presence

• Cell behavior under hypoxic conditions and carbonic anhydrase inhibition.

2 articles submitted:

• L. Stanica*, M. Rosu-Hamzescu, M. Gheorghiu, M. Stan, L. Antonescu, C. Polonschii, E. Gheorghiu Bioanalytical electro-optical platform for evaluation of bioeffects; Case study – cancer hypoxia


3 project applications submitted (1 PED projects, one PCE project, one PCE exploratory)

2 oral and 1 poster presentations at international conferences


Oral L. Stanica, M. Gheorghiu, E. Gheorghiu Electro-optical evaluation of cellular dynamics of ChR2 expressing HEK293 cells induced by light stimulation The EMBO Meeting 2016, 10–13 September 2016 Mannheim, Germany

Poster L. Stanica, M. Gheorghiu, E. Gheorghiu Characterization of light responsive, genetically modified cells as cellular sensors, 10th FENS Forum of Neuroscience, July 2–6 2016, Copenhagen, Denmark
Electrochemistry Laboratory

Team: Szilveszter Gáspár

Facilities
The resources of the laboratory include the following equipment: VSP modular potentiostat/ galvanostat (from Bio-Logic S.A., France), CellTest multi-channel potentiostat/ galvanostat (from Solartron Analytical, UK), Nanoband Explorer II anodic stripping voltammetry-based heavy metal analyzer (from TraceDetect, USA), 797 VA Comprtrace PC controlled system for voltammetry (from Metrohm AG, Switzerland), Scanning Electrochemical Microscopy (SECM, from Sensolytics, Germany), trinocular stereo microscope (from World Precision Instruments, USA) equipped with a Coolpix 995 digital camera (from Nikon, Japan), EG-40 micropipette beveller (from Narishige, Japan), and 3D micropositioner (HS6 from World Precision Instruments, USA).

Current Research and associated projects
The Electrochemistry Laboratory was mainly focused on Graphtivity a project developing electrochemical sensors which i.) allow monitoring small molecules (e.g. hydrogen peroxide, dopamine, glucose, etc.) at cellular level, ii.) are also compatible with optical microscopy methods (e.g. Surface Plasmon Resonance microscopy), and iii.) build on the advantageous electrical and optical properties of graphene. The laboratory has also made contributions to projects on protein aggregation and on electro-plasmonics. The following papers were published during 2016 with the involvement of the Electrochemistry Laboratory:

Facilities

Main resources are represented by an 8-channel potentiostat from Dropsens, Spain and a portable SPR system (SPREETA, Nomadics). The group shares with the Electrochemistry group the VSP modular potentiostat/ galvanostat (from Bio-Logic S.A., France).

Current Research and associated projects

For the Applied Electrochemistry group, 2016 marked the end of the Partnership Project ANTIOXWIN. In 2016, the focus of the group was on publishing the research results and strengthening collaborative ties with other groups from Romania (Institute of Biology of the Romanian Academy, National Institute for R&D in Biological Sciences, Bucharest, Faculty of Chemistry-University of Bucharest, National Institute for Laser, Plasma and Radiation Physics (INFLPR), Magurele), France (Prof. Jean-Louis Marty's group at BAE, University of Perpignan and Prof. Sabine Szunerits group at Institute of Electronics, Microelectronics and Nanotechnology-IEMN, University of Lille 1, France) and the US (dr. Serban Peteu at Michigan State University and Prof. Silvana Andreescu at Clarkson University). This was reflected in papers published or accepted, done in collaboration with the above groups. An important achievement was the CSII title obtained by Alina Vasilescu.

Accomplishments:

- CSII title accorded to Alina Vasilescu
- 7 papers published, 1 accepted and 1 submitted for publication;
- 5 book chapters—three published, one accepted, one submitted for publication;
- 2 oral and 1 poster presentations at national/international conferences in Romania
- 1 national research project finished in 2016 (ANTIOXWIN, PN-II-PT-PCCA-2011-3.1-1809).
- 1 research project accepted for funding (Horizon 2020, Manunet II SENS4WINE);
- 4 project applications submitted (2 PED projects, one PCE project, one Bilateral Romania-France Brancusi);
- Workshop “Modern approaches for producing high quality wines rich in antioxidants” organized in the frame of ANTIOXWIN project on July 05, 2016 at the International Centre of Biodynamics.
Articles:
Published:
Accepted:
Submitted:

Book chapters:


Conferences:
Oral presentations:


Posters:
Microfabrication & Prototyping Laboratory

Team: Mihai Sorin David
     Dumitru Bratu

Facilities

CNC Shape Cutting Machine-2000 series 8 direction mill (from Sherline Products, USA), P6700 Spin coater (from Specialty Coating Systems, USA), lathe and rectifier lathe. Physical Vacuum Deposition PVD 75 (Kurt J Lesker, USA). Modular design configured to suit a variety of thin film deposition applications, typically for research and development or small batch production.

Physical Vapor Deposition-PVD 75

PVD 75 thermal evaporator from KJ Lesker is currently used for manufacture of different configuration of electrodes and for different applications requiring precise deposition of several layers of materials. For example: SPR – we manufacture SPR chips by depositing 2 nm of Chromium/Titanium and 50 nm of Gold; Magnetic SPR – we prepare metallic “sandwiches” comprising of Chromium, Cobalt and Gold; Electrode passivation – we deposit thin layer of insulating materials (e.g. SiO).

Additional facilities: Spin coating machine within glove box, laminar flow hood for specific surface functionalization, ultrasonic processors and UV ozone cleaning system. The spin coater is used for controlled deposition of thin polymer layers on solid substrates for specific applications including electrode preparation for impedance measurements, investigation of cell adhesion to various materials, controlled local electrical insulation for electrode design, customized PCB boards, optical coupling for SPR measurements.

Current Applications

Developing user defined parts and accessories from holders and test fixtures electro-optically compatible measurement cells (static and flow through) and tailored plasmonic surfaces, with sizes down to tens of microns.
Bioanalysis Laboratory
(including a mobile laboratory)

**Team:**
- Mihaela Gheorghiu
- Szilveszter Gáspár
- Mihai Sorin David
- Cristina Polonschii

**Facilities**
Anodic Stripping Voltameter Nanoband Explorer II (from Tracedetect U.S.A) Dual system EIS/SPR (ICB patent), Glomax Luminometer 20/20 (from Promega U.S.A), pH-meter and conductometer (from WTW Germany), portable UV-VIS Spectrophotometer Nova 60A (from Merck, Germany), Mobile autolaboratory equipped with working areas, separate access and electrical connections.

**Current Status**
The Mobile and Fixed Bioanalysis Laboratory was accredited in 2010 by RENAR as a third party testing laboratory, in compliance to the ISO/IEC 17025:2005 clauses and to the provisions of European directives and national legislation on water quality. Following the reduction of the commercial activities within the accredited laboratories during 2011, the activity of this laboratory was suspended in 2012, at our request.
Electrical Measurements Laboratory

Team:
Eugen Gheorghiu
Mihai Sorin David
Cristina Polonschii
Mihnea Rosu Hamzescu
Dumitru Bratu

Facilities
Impedance analyzer Agilent 4294A (from Agilent Technologies, USA), Impedance / gain phase Analyzer Solartron; 1470 E Cell Test System (both from Solatron Analytical, U.K), Digital Oscilloscope TDS 3052 (from Tektronix SA-France), Signal generators, Phasemeter, Spectral Analyzer HP 3585; 8 channel low frequency impedance spectrometer (designed and produced in ICB);

Dedicated working lab and current utilities (purified air, ultra pure water, Faraday and thermostated chambers, 4 point electrode configurations and wide range of surface electrodes – circular and/or interdigitated, compatible with complementary optical assays).

Current Research and related accomplished projects

- DYNANO (FP7), BioScope
  - Investigation of lipid film formation in view of lipid sensors development - mimics for actual biological membranes.
  - Investigation of the dynamics of electrical parameters of cellular platforms in relation to engineered interfaces (with attractive/repulsive chemical and morphological features),
  - Investigation of the dynamics of electrical parameters of cellular platforms in relation to chemical and biological stressors (heavy metals, pathogen cells).
  - Monitoring cell cycle progression on non/synchronized cell suspensions – an on line system for electro-optical evaluation of yeast cell suspensions has been developed.
  - Evaluation of functionalization protocols (deposition of thin polymeric layers, biorecognition compounds or ligand – e.g. thiol layers).
  - Development of a multi channel dual SPR impedance set-up.
Impedimetric and Plasmonic Bio Sensors

Team: Mihaela Gheorghiu
      Mihai Sorin David
      Cristina Polonschii
      Dumitru Bratu
      Luciana Stanica

Facilities
- Biacore 3000 system (from Biacore AG Sweden)
- Dual spectrometer EIS/SPR (ICB patent),
- 3 channel Spreeta Modules, Contact Angle Meter CAM 100 (from KSV, Finland).
- Dedicated working lab and current utilities (purified air / N₂, chemical hood and ultra pure water, thermostated chambers)
- Advanced set-up for monitoring the dynamics of aquatic species and the quality of growth environment.
- Advanced set-up for high speed impedance assay with synchronization with light pulses.
- Custom design generators for multifrequency assays.

Additional facilities: Evolution 600 UV-VIS Spectrophotometer (Thermo Scientific USA) with variable angle specular module, AFM module Nano Wizzard II (JPK, Germany) Multimode readers Promega / Turner.

Current Research and associated projects
The design of novel (bio) sensing interfaces for selectively recognition of the analyte of interest, from low molecular compounds to cells, and for providing a concentration-dependent phenomenon that is easy to translate into useful analytical signal) towards a new generation of analytical tools. These analytical tools will extend our ability of detecting compounds of interest:

- in very low concentrations,
- in complex samples without prior separation,
- in small sample volumes (microliters) or exotic environments (such as cellular sub compartments), and
- in a timely manner traditional analytical approaches are very often lacking such abilities.
The integration of complementary analytical tools (SPR, electrochemistry) with microtechnologies (fluidics, electronics) for the development of portable, sensitive solutions for real time detection of target analytes.

**Accomplishments**


**Supported by:**

 kazhki

- 2 national research projects:
  - BIOSCOPE, Electro-Plasmonics for the analysis of the dynamics of cellular processes and biomolecular interactions, Contract No. 11/2012, ID: PN II-ID-PCCE-2011-2-0075, coordinated by ICB; Project Director: Eugen Gheorghiu;
  - Sensitive quantitation of target microorganisms using dual electro plasmonic analysis and magnetic actuation, Contract No. 82–30.04.2013, ID: PN-II-RU-PD-2012-3-0467, Project Director: Mihai Sorin David

- 1 international research project:
  - Tumoranalyser- Response of in vitro hypoxic tumor models to potentially therapeutic compounds as revealed by an advanced analytical platform, Romanian-Swiss Research Programme IZERZ0_142236, 2012, Promoter from Switzerland, École Polytechnique Fédérale de Lausanne; Hubert Girault and from ICB: Eugen Gheorghiu;

Participation in the Cost Action Proposal “European Perspectives Towards Bioelectronics”
Optical Microscopy Laboratory

Team: Mihaela Gheorghiu  
Mihai Sorin David  
Luciana Stanica

Facilities
Zeiss Total Internal Reflection Microscope (TIRFM), Zeiss AxioObserver Z1, fully motorized with xyz nm resolution, equipped with EMCCD ANDOR IXon DU-885K camera, with facilities for epifluorescence, laser (3 line Ar) and white light TIRF, and various contrast methods (Ph, DIC) and for cell cultivation (OkoLab controlled environmental cage); Extended image processing facilities (AxioVision, Andor IQ and Andor IQ Tracker); dedicated chambers for combined (optical and electric) assays; Additional filter sets 02, 09, 14. NIKON Eclipse 400 microscope with epiflourescence system and CCD camera for data acquisition.

Additional facilities:
- Spectrophotometer Evolution 600 (Thermo Scientific, USA), with variable angle specular reflectance accessory and thermostated carrousel.
- GloMax 20/20 Multimode reader (Promega), with 2 injectors and UV and Blue fluorescence modules.
- Compatible AFM module fully integrable with advanced optical assessment.

Current Research and associated projects
- Dual electro-optical measurements based on dedicated flow cells with optimized electrode configurations.
- Time lapse assessment of the dynamics of cells (morphology, attachment and metabolism) and cell organelles (pH, ions, morphology) in response to external stimuli and engineered bio-interfaces cues; membrane processes in normal and pathologic conditions; manipulation and characterization of natural and synthetic lipid environments (including cholesterol). ATP & Cytotoxicity assays; Intracellular Ca^{2+};

The push - pull microfluidic probe developed by the Swiss Partners within TUMORANALYZER Project has been integrated within the AxioObserver Z1 set-up and
experiments enabled 1. Precise control of probe positioning with optical feedback; 2. Fluidic and electric cell stimulation 3. Experimental validation of modelling studies.

Accomplishments

2 manuscripts on cell actuation and imaging to be submitted

Participation in the 14th EMBO Practical Course in Advanced Optical Microscopy, 6-16 April 2016, Plymouth England (L. Stanica, fellowship)
Modeling and Data Analysis Laboratory

Team: Eugen Gheorghiu  
Cristina Polonschii  
Sorin David

Current research and associated projects

We are currently developing fast, efficient methods to assess target analytes using periodic actuation.

**Dielectric modeling and nonlinear time series analysis**

Strong emphasis is placed on both experimental and theoretical aspects regarding:

- Development of new Plasmonic-EIS instrumentation;
- Development of microscopic models of dielectric behavior with emphasis on:
  1. Membrane Potential (discussion of GHK approach) and the role effect of ionic channels
  2. non-spherical cells (focusing on yeasts, red blood cells, and gap junction connected cells).

A quantitative approach providing shape evolution of budding yeasts during the cell cycle, consistent with experimental findings, is available.

- Time series analysis of dielectric data yielding quantitative measures of the system dynamics (revealing changes in the tissue structure and function and the invariants of the cell cycle

3 project applications submitted (2 PED projects, one PC Exploratory)

An international patent application was granted: U.S. Patent 9,315,855/2016, Systems and Methods for Detection and Quantitation of Analytes Using an Oscillating Stimulus, Authors: E. Gheorghiu, S David, C. Polonschii, D Bratu

Participation to international conferences


Pilot Laboratory on monitoring fish behavior for quality assessment of aquatic environments

Team: Eugen Cheorghiu
Cristina Polonschii
Dumitru Bratu

Facilities

- A platform consisting in a 3D array of ultrasound transducers and of supporting analytic modules to monitor fish positions and assess fish dynamics (ICB patent);
- Dedicated water tank with adequate infrastructure for controlling water temperature, composition, including dissolved Oxygen (DO);
- A module for controlled dispensing of nutrients;
- Facilities for complementary video observations of fish behavior;
- A set of sensors for continuous monitoring of water parameters: pH, DO, conductivity and temperature.

Current Research

Extending the capabilities of the platform to remotely monitor and analyze the behavior/dynamics of various fish species (including sturgeons) both in tanks and cages immersed in aquatic media (lakes, rivers). The monitoring system fosters detection of behavioral changes of aquatic species triggered by harmful compounds that might incidentally occur in water (even in very small concentrations, well below the lethal threshold of those species).

Accomplishments

A patent application was granted:

Patent Ro 128065/2015: “Platform and method to monitor the quality of an aquatic environment based on analysis of the behavior of a fish population”, Authors: E. Cheorghiu, C. Polonschii, D. Bratu
Atomic Force Microscopy Laboratory

Teams: Szilveszter Gáspár
       Mihaela Gheorghiu
       Sorin David

Facilities
The major piece of equipment of the laboratory is a NanoWizard® II Atomic Force Microscope JPK AG (Berlin, Germany) combined with optical microscope Zeiss AxioObserver D1 fully equipped with phase contrast, fluorescence and dark field for complementary optical observations.

Current Research and associated projects
Our AFM instrument has the necessary features to work with biological samples (i.e. soft samples in liquid), and thus is often used to observe small cellular structures (e.g. tight junctions, microvilli, etc.) or surface absorbed / immobilized proteins and bacteria (see Figure 1A). In addition to the study of biological samples, it is also used to investigate the quality and thickness of thin metal films deposited by PVD or metal sputtering and the structure of graphene-modified surfaces (see Figure 1B).
Acomplishments:
1 published paper

1 Accepted:
International projects

ONGOING

- Cell biosensors for detection of chemical and biological threats, Contract: NATO SPS 985042, Project director: Eugen Gheorghiu, 2016-2019
- GRAPHTIVITY - Graphene based optoelectrochemical sensor for the simultaneous monitoring of the electrical and chemical activity of single cells, Contract: Nr. 40 din 2016, ID: FLAG-ERA-Graftivity, Project director: S. Gaspar, 2016-2018

ACCOMPLISHED

- CAPTALLERGENES- Aptamer based sensors for allergen detection, Cooperation Romania-France, PN-II-CT-RO-FR-2012-1-0012, Project Director: Alina Vasilescu;
- Tumoranalyser-Response of in vitro hypoxic tumor models to potentially therapeutic compounds as revealed by an advanced analytical platform, Romanian-Swiss Research Programme IZERZ0_142236, 2012, Promoter from Switzerland, École Polytechnique Fédérale de Lausanne: Hubert Girault and from ICB: Eugen Gheorghiu;
- “DYNANO” Dynamic Interactive Nanosystems, FP7-PEOPLE-2011-ITN N°289033 7th FP (2011-2015);
- “Non-invasive electro-optical methods to assess the effect of nano/micro substratetopography on living cell dynamics”, Academic Cooperation between Romania and Russian Federation, Project Director: Eugen Gheorghiu
- “NANOMAGMA” - NANOstructured active MAGnetoplasmonic MAterials”, Funded under 7thFWP (Seventh Framework Programme), Contract no.: 214107 /2008, 2008-2011;
- „ROBIOS“- Strengthening ROnanian Research and Training Capacities in BIOSensing and Related Areas”, 2005-2008;
- Role of membrane organization and dynamics on cell behavior and response to external stimuli; Romania -Flanders Bilateral Agreement, 2005-2006;
- Novel Impedimetric Affinity Biosensors (IAB) for toxicological applications exploiting E.Coli membrane protein LamB - Romania-Flanders bilateral agreement, 2003-2005
- Development of an Impedance Assay for Immunocapture - Brancusi Romania - France bilateral agreement, 2003-2005
- Towards the development of a rapid portable immunoassay device for the detection of Microorganisms and Toxins in Food industry & Ecology - COBASE Grant Program 2001
• Impedance Spectroscopy of biological membranes: modeling the epithelial tissues and the dielectric dispersion of the plasma membrane; Romania-Flanders bilateral agreement 2001-2003;

• Biomonitoring quantitative evaluation of biological systems, Romanian-German Bilateral Agreement reference number RUM 003-98;

• AFRAMILK -Antifraud impedimetric and ultrasonic control system to detect fraud (adulteration) of milk GRD1-2000-25801, within the 5th Framework Program of the European Commission 2001-2004;

• Fluid Rheology – Application to biology and medicine, financed by the World Bank Coordinator: Bucharest Politehnic University (UPB), Faculty of Energetics;
# National projects

## ONGOING

<table>
<thead>
<tr>
<th>Financing period</th>
<th>Project title and managing/partnership status</th>
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<tr>
<td>2012-2016</td>
<td>BIOSCOPE - Electro-Plasmonics for the analysis of the dynamics of cellular processes and biomolecular interactions, Contract No. 11/2012, ID: PN II-ID-PCCE-2011-2-0075, coordinated by ICB; Project Director: E. Gheorghiu</td>
</tr>
<tr>
<td>2012-2016</td>
<td>ANTIOXWIN - Exploiting the antioxidant capacity of black grapes for producing wines with high authentic quality – Contract No. 101/2012, ID: PN-II-PT-PCCA-2011-3.1-1809; Project Director: A. Vasilescu;</td>
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## ACCOMPLISHED*

<table>
<thead>
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<tbody>
<tr>
<td>2013-2015</td>
<td>Sensitive quantitation of target microorganisms using dual electroplasmonic analysis and magnetic actuation, Contract No. 82-30.04.2013, ID: PN-II-RU-PD-2012-3-0467, Project Director: S. David</td>
</tr>
<tr>
<td>2011 – 2014</td>
<td>Tracing proteins through food processing with biosensors (PN II-RU-TE-3-0302), Project supporting development of Young Teams, Principal Coordinator ICB - director A. Vasilescu</td>
</tr>
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<td>2011-2014</td>
<td>SENSMOTION Sensing using the electrochemically-triggered motion of catalytic nanomotors - Contract No. 16/05.10.2011, ID: PNII-RU-TE-2011-3-0237, Project Director: Szilveszter Gaspar</td>
</tr>
<tr>
<td>2011-2013</td>
<td>A new strategy for effective assessment of DNA hybridization, based on periodical magnetic actuation, Post Doctoral Grant - director C. Polonschii</td>
</tr>
<tr>
<td>2012-2014</td>
<td>EXTRACELL - Monitoring the extracellular space with catalytic self-propelled nanomotors, ERC-like project Contract No. 9/02.07.2012, Project Director: Szilveszter Gaspar</td>
</tr>
</tbody>
</table>
2010-2011 Improvement of Management and Marketing Abilities within the International Centre of Biodynamics (MANMAR) – Structural Funds, Economic Competitiveness Improvement, Axis 2.2.4.

2007-2010 Dielectric modeling of biological cells and heterostructures with fast and efficient algorithms for boundary integral method; Principal Project Coordinator ICB

2007-2010 DEMENTJUNCTION - Expression and function of the tight junction proteins – a study in experimental models and dementia diagnosed patients Partnership with “Victor Babes” National R&D Institute

2007-2010 BIOSADN – Development of biosensors based on nucleic acids for the evaluation and monitoring of some toxic agents with applications in bioterrorism; Partnership with Faculty of Chemistry, University of Bucharest

2007-2010 PROPETHAD – Advanced Research towards medical applications of nuclear technologies; Partnership with “Horia Hulubei” National Institute of Physics and Nuclear Engineering

* Full list is given on the institution website.
Visibility actions

Oral presentations of ICB representatives


Posters

2. Stănică L, Gheorghiu M, Gheorghiu E, Poster „New twist for optogenetics - characterization of light responsive, genetically modified cells as cellular sensors”10th FENS Forum of Neuroscience 02-7July 2016 Copenhagen, Denmark.
Emergent Applications fostered by ICB

1. Sensors and biosensors

Developing of rapid, sensitive, cost effective detection platforms to determine low concentration of target compounds. In this context, surface plasmon resonance (SPR) based biosensors represent attractive solutions in environmental monitoring and for quality-control, due to their sensitivity, miniaturization amenability and wide range of detectable compounds.

Since 2006, the SPR technology is operational within ICB through several instrument including: Biacore 3000 (Biacore AB, Sweden), SPREETA based devices (complemented by EIS and Magneto-Optic SPR) and a recently developed bench system allowing for flexible illumination at various wavelengths.

The analysis methods already implemented allow:

- multianalytes detection (mixture of compounds e.g. with toxicogen potential)

SPR-based techniques enable real time, label-free assessment of the interaction between an analyte (e.g. chemicals, DNA, RNA, proteins) and a covalently immobilized specific ligand, simultaneously with a high sensitivity in determination of concentrations and kinetic dates, and also the possible integration with other analysis and fluidic techniques (FIA) or with approaches like Lab-On-a Chip.

In this respect, ICB has developed cellular platforms for assessment of food quality and cytotoxicity of water samples. Including specific electrochemical instrumentation (impedance spectrometer, potentiostat) and a detection unit based on electrochemical sensors, electrical components and fluidic elements to maintain the temperature at 37°C, and applying a previously validated measurement method, multiple cell parameters can be potentially determined.

The ICB proprietary technology based on periodic actuation supports major sensing advantages of ICB platform versus the current state of the art

1.) Portability in a lab on a chip format;
2.) High sensitivity & specificity
3.) Reduced analysis time;
The analytical platform is versatile, so it can provide relevant information not only for water monitoring activities, but also in chemical or pharmaceutical industry, efficiently responding to the Romanian and European socio-economic needs regarding the environmental quality control and/or biosensing applications in other specified domains.

2. Measurement and control systems for optimization of aquaculture activities

An issue of real interest facing both our country and other European countries is represented by the strong demand for automated systems that will lead to optimization of aquaculture activity through increasing productivity and reducing costs. Systems already on the market do not optimize feeding process as a function of fish behavior, which is one of the causes that generate many losses both in terms of quantity and quality of food (about 30% of the costs of aquaculture operations).

Taking into consideration these trends and the promising results obtained by ICB in research and experimental studies undertaken in this direction, some of the most important applications developed in this field are as follows:

- support system of automation and optimization of feeding
- pilot system for non-invasive, multichannel assessment of behavior (dynamics) of aquatic species
- integrated, remote monitoring system for water (aquatic environment) quality control able to activate a specific system based on biosensors whenever the behavior of aquatic species deviates from a normal pattern;
- (non) linear - analysis method which provides a set of quantitative measures, characteristic of the behavior of species with high economic value (e.g., sturgeons, carps) in relation to actual environmental conditions (including feeding conditions).

3. Implementation of light driven dynamic processes for cell based sensing

Cell-based biosensors traditionally exploit living cells capability to alter physiologic processes or cellular rhythms emphasized by cell cycle or exhibited by excitable cells in response to extracellular stimuli. They are often slow responding, nonspecific and irreproducible. We highlight for the first time the analytic potential of fast, noninvasive electrical assessment of the discrete changes of characteristic cell response to an externally induced stimulus as modulated by bioactive (e.g., pharmacologic or toxic) compounds. The sensing concept highlights in 30 minutes, signature evolutions of impedance data characteristic for cellular status and the nature of extracellular stimulus.
4. Quantitative assessment of specific carbonic anhydrase inhibitors effect on hypoxic cells using electrical impedance assays

Carbonic anhydrase IX (CAIX) is an important orchestrator of hypoxic tumour environment, associated with tumour progression, high incidence of metastasis and poor response to therapy. Due to its tumour specificity and involvement in associated pathological processes: tumorigenicity, angiogenesis, inhibiting CA IX enzymatic activity has become a valid therapeutic option. Dynamic cell-based biosensing platforms can complement cell-free and end-point analyses and support the process of design and selection of potent and selective inhibitors. The effectiveness of recently emerged CA IX inhibitors (sulfonamides and sulfocoumarins) and their antitumour potential is assessed using electrical impedance spectroscopy (EIS) biosensing platform. The analysis allows discriminating between the inhibitory capacities of the compounds and their inhibition mechanisms.
Relevant publications


- Polonschii C., David S., Gáspár S., Gheorghiu M., Rosu-Hamzescu M., Gheorghiu E., “Complementarity of EIS and SPR to Reveal Specific and Nonspecific Binding When Interrogating a Model Bioaffinity Sensor; Perspective Offered by Plasmonic Based EIS”, Anal. Chem.(2014), 86 (17), 8553–8562


- Gaspar, S. Enzymatically Induced Motion at Nano- and Microscale. Nanoscale 2014, 6, 7757–7763.


The International Centre of Biodynamics

- Polonschii C., Bratu D., Gheorghiu E., Appraisal of fish behaviour based on time series of fish positions issued by a 3D array of ultrasound transducers, Aquacultural Eng (2013), 55, 37–45


- E. Gheorghiu, M. Gheorghiu, S. David, C. Polonschii "Biodynamics: sensing through dynamics of hybrid affinity / cellular platforms; towards appraisal of Environmental and...