Sensors and Biosensors group

Giovanna Marrazza

Maria Minunni

Ilaria Palchetti

Post-docs

Francesca Bettazzi

Simona Scarano

PhD students

Andrea Ravalli

Diego Voccia

Stefano Mariani

Visiting professors

A.P.F. Turner

Marco Mascini

Erasmus link

Aristotele University of Thessaloniki (Greece)
Bucharest University (Romania)
Cranfield University (UK)
Brno University (Czech Republic)
Cluj-Napoca University (Romania)
Group’s expertise

1. Development and evaluation of sensors and biosensors based on the coupling of electrochemical, optical and piezoelectric sensors with enzymes, antibodies, bacteria, whole tissues and nucleic acids.

2. Solving analytical problems in clinical chemistry, experimental medicine, food chemistry and environmental analysis.

3. Development of new immobilization chemistries of biomolecules (enzymes, proteins, nucleic acids, etc.) and analytical procedures suitable for use with biosensor devices, i.e. flow injection analysis, flow systems, and microdialysis.
Electrochemical instrumentations

Autolab Potentiostat/Galvanostat (Metrohm, NL)

Palm Sens electrochemical sensor interfaces (NL)

Home made screen-printed electrodes
Electrochemical & Piezoelectric instrumentations

Scanning electrochemical microscope (SECM)

Universal sensor

- 9.5 MHz AT-Cut quartz crystal (14mm); Gold evaporated (42.6mm² area) on both sides

Seiko quartz microbalance

Microfluidic system

Immuspeed

GRAVI - CHIP

Elbatech QC magic (4 channels)
Optical instrumentations

Fully automated Biacore X™ (GE Healthcare)

Spreeta ™ (Texas Instruments Inc. USA)

SPR-imaging (Genoptics- France)
Recent applications and main research topics

**Artificial Aptamers**

**Nucleic Acid and Peptide Aptamers: Fundamentals and Bioanalytical Aspects**

Marco Mascini,* Ilaria Pulchetti, and Sara Tombelli

**Angewandte Reviews**

DOI: 10.1002/anie.200808630

**Quasi-monodimensional polyaniline nanostructures for enhanced molecularly imprinted polymer-based sensing**

Francesca Bert,* Silvia Todros,* Dhana Lakshmi,* Michael J. Whitcombe,* Iva Chianella,* Matteo Ferroni,* Sergey A. Piletsky,* Anthony P.F. Turner,* and Giovanna Marrazza*+

1 Department of Chemistry, University of Florence, via della Lastrina 2, 50125 Firenze, Florence, Italy
2 CNR-INSTM SBRC laboratory, Department of Chemistry and Chemical Engineering, University of Perugia, via Villoresi 6, 06123 Perugia, Italy
3 School of Chemistry, University of Cambridge, Cambridge, CB3 0HE, UK

**Surface Plasmon Resonance Imaging: What Next?**

Giuseppe Spoto* and Maria Mininni*†

1 Dipartimento di Scienze Chimiche, Università di Catania, Viale Andrea Doria 6, 95123 Catania, Italy
2 Istituto Nazionale di Biotecnologie e Biotecnologie, Catania, Italy
3 Dipartimento di Chimica e CSGI, Università di Roma, Via della Lastrina, 3 00198 Sesto Fiore (FI), Italy

**Angewandte Chemie**

**The Journal of Physical Chemistry Letters**

**Abstract:** This Perspective discusses recent advances in the field of surface plasmon resonance imaging (SPRI) for the label-free, multiplex, and sensitive study of biomolecular systems. Large efforts have been made during the past decade with the aim of developing even more sensitive and specific SPRI-based platforms. Metal nanostructures have been used to enhance SPRI sensitivity and to build specific SPRI-active surfaces, while special effects such as long-range SPRI have been investigated to develop more effective SPRI platforms. Here, we review some of the significant work performed with SPRI for the ultrasensitive detection of biomolecular systems and provide a perspective on the challenges that need to be overcome to enable the wider use of SPRI in emerging key areas such as health diagnostics and nanotagging controls.
This Workshop on Bioinspired nanotechnology for Biosensing is an exciting event, to be held on 16 May 2013, and will be organized by the COST Action TD 1003 and supported by Elsevier. Keynote talks will be invited from leaders in the field and there will be ample opportunity for participants to contribute oral or poster presentations and discussion topics. The Workshop will be held immediately after the 3rd International Conference on Bio-sensing Technology, 13-15 May 2013, which will take place in the same location.

Workshop Chair:
Prof Maria Minunni, University of Florence, Italy

Organizing Committee:
Prof Arben Herkoçi, Catalan Institute of Nanotechnology, Spain
Prof Luigi D. Blasi, Université Lyon 1, France
Prof Fred Lisdat, University of Applied Sciences Wildau, Germany
Prof Giuseppe Spoto, University of Catania, Italy
Prof Anthony P.F. Turner, Linköping University, Sweden
Prof Roy Quinnan, Durham University

Following the perspective of the COST action, innovative approaches to elaborate new concepts in biosensing inspired by the combination of biology and nanotechnology we will discussed.

The co-location of the meeting with an international conference will facilitate discussion and exchange with specialist in biosensing from across the world who will be attending the conference, generating opportunities for new collaborations and novel ideas for collaborative projects.

No fee to attend the workshop is required and delegates may register for the Workshop without registering for the main conference, if they wish.

For administrative convenience, however, registration for the Workshop will be via Elsevier's Conference Registration System http://www.bioinsensingconference.com/conference-register.html

Topics include:
- Natural & synthetic receptors
- Nucleic acid and protein based strategies for biosensing
- Nanostructuring of surfaces
- Nanobiosensors, nanomaterials & nanoanalytical systems
- Theranosics & nanotheranostics

COST Action TD 1003 "BioChem: Chemical and Molecular Sciences and Technologies for Sensing"
<table>
<thead>
<tr>
<th>Project Title</th>
<th>Date and Duration</th>
<th>Funding organisation</th>
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<tbody>
<tr>
<td>Detection of Hepcidin As A New Biomarker of Erythropoiesis Stimulators Abuse: A Pilot Study Optical sensing, P.I. M. Minunni</td>
<td>2010-2011</td>
<td>world anti-doping agency (WADA)</td>
</tr>
<tr>
<td>An integrated approach with Affinity-Based Biosensing (ABBs) for gene doping detection: a pilot study Optical sensing, Coordinator M. Minunni</td>
<td>2009-2010</td>
<td>world anti-doping agency (WADA)</td>
</tr>
<tr>
<td>GAPOGROWTH, Growth of Large GaPO4 Single Crystals and their use for Special Sensor Applications Piezoelectric sensing, P.I. M. Minunni</td>
<td>2002-2005</td>
<td>EU (FP5)</td>
</tr>
<tr>
<td>CARE-MAN- Health CARE by biosensor Measurements and Networking Optical sensing, P.I. M. Mascini</td>
<td>2004-2010</td>
<td>EU (FP6)</td>
</tr>
<tr>
<td>MICS Innovative functional materials and associated technologies Electrochemical, P.I. M. Mascini</td>
<td>2001-2004</td>
<td>EU (FP5)</td>
</tr>
<tr>
<td>Evaluation/validation of novel biosensors in real environmental and food samples (VALIDATION OF BIOSENSORS) Electrochemical, P.I. M. Mascini</td>
<td>2000-2003</td>
<td>EU (FP5)</td>
</tr>
<tr>
<td>Sensors for monitoring water pollution from contaminated land, landfills and sediments Electrochemical, P.I. M. Mascini</td>
<td>1998-2002</td>
<td>EU (FP5)</td>
</tr>
<tr>
<td>National projects</td>
<td>Date of duration</td>
<td>Funding Institution</td>
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<tr>
<td>Development of Biosensor for point mutation detection Optical sensing; P.I. M. Minunni</td>
<td>2010-2013</td>
<td>Fondazione Arpa</td>
</tr>
<tr>
<td>Minimally invasive mycosystem for glucose monitoring in diabetic patients, Electrochemical sensing, P.I. M. Minunni</td>
<td>2010-2012</td>
<td>Regione Toscana, Programma per la ricerca Regionale in materiali di Salute 2009</td>
</tr>
<tr>
<td>Bioanalytical method based on biosensors for the new frontiers of doping: detection of exogenous proteins and genes&quot; Coordinator: M. Minunni</td>
<td>2006-2008</td>
<td>Ministry of Health, Programma di Ricerca 2005 sui farmaci, sulle sostanze e pratiche mediche utilizzabili ai fini di doping nelle attività sportive</td>
</tr>
<tr>
<td>Cicloxygenase-2 inhibitors (COX2) for the colon rectal cancer treatment&quot; Piezolectric sensing P.I. M. Minunni</td>
<td>2003-2005</td>
<td>Ministry of Health, progetti finalizzati</td>
</tr>
</tbody>
</table>
Research:
• Développement d’un biocapteur à base d’un nouvel **aptamère** pour la diagnostique dans la **pathologie d’Alzheimer**
• Sviluppo di **biosensori di affinità** a base di un nuovo recettore aptamerico per la diagnostica molecolare della **malattia di Alzheimer**

Parternership 'Università di Grenoble I, Joseph Fourier (Prof. Eric Peyrin).

**Investigator: M. MINUNNI**
Case studies:

**Drug discovery:** Studies of amyloid fibrills aggregation and its inhibition by drugs (Alzheimer treatment?)

- **DNA-based sensing** for target sequence or mismatch detection, i.e. Single nucleotide polymorphism (SNPs);
  Molecular diagnostic, theranostic, tailored therapy
  Ermini, M.L., S. Mariani, S. Scarano, D. Campa, R. Barale, M. Minunni*, *Analytical Bioanalytical Chemistry*, 2013, 405, 985-993

- **Aptasensor:** Aptamer –ligand interaction detection of MMP9; Molecular diagnostic

- **Immunosensors:** Ag-Ab interaction; Molecular diagnostic

- **Improving** analytical performances by **nanostructuring**
Analytical Methods

Cite this: Anal. Methods, 2012, 4, 2228
www.rsc.org/methods

Label-free methods for probing the interaction of clioquinol with amyloid-β†

Xin Ran Cheng, Vinci Wing Sze Hung, Simona Scarano, Marco Mascini, Maria Minunni* and Kagan Kerman*

Received 4th February 2012, Accepted 5th May 2012
DOI: 10.1039/c2ay25123j

The presence of amyloid-β (Aβ) fibrils is characteristic of Alzheimer’s disease (AD), and the aggregation of these amyloidogenic proteins is a nucleation-dependent process. In this report, label-free methods based on surface plasmon resonance (SPR) and thickness shear mode acoustic wave sensors (TSM-AWS) were used to detect monomer elongation in real-time. The modulation of Aβ aggregation using a well-described flavonoid, clioquinol (CQ) was also observed. Established methods like fluorescence and electrochemistry were also employed to confirm the interaction of CQ with Aβ. Good correlation between the designed label-free methods creates a promising platform for the screening of novel amyloid inhibitors.

Fig. 1 Fibril elongation process on the surface of the seed-immobilized TSM-AWS; (a) baseline signal in buffer, before Aβ1-42 monomer addition on the seed-immobilized surface; (b) deposition and elongation of Aβ1-42 fibrils; (c) surface wash to remove the non-specifically bound monomers. The analytical signal was calculated as the difference (c-a) in frequency (Hz).

Fig. 3 Serial injections of 2 μM Aβ1-42 monomers with 10 μM CQ. Subsequent injections of 2 μM Aβ1-42 monomers initiated the elongation process.

Fig. 4 Sensorgram showing the inhibitory effect of 10 μM CQ on aggregation, when introduced in the presence and absence of 2 μM Aβ1-42 on the seed-immobilized surface.
Single nucleotide polymorphisms high sensitive detection on human DNA – with surface plasmon resonance Imaging

S. Mariani\textsuperscript{1}, S. Scarano\textsuperscript{1}, R. Barale\textsuperscript{2}, M. Minunni\textsuperscript{1}

\textsuperscript{1}Dipartimento di Chimica “Ugo Schiff”, Università degli Studi di Firenze (Italiy)
\textsuperscript{2}Dipartimento di Biologia, Università degli Studi di Pisa (Italiy)

Thematic COST meeting: “Nano-scales arrangements of proteins, aptamers and other nucleic acid structures - and their potential applications”
Surface Plasmon Resonance (SPR)

SPR imaging (SPRi)


Biochips and Array Preparation

Biochip design

Resonance couplers:
Glass prisms or slides coated with gold

Microsyringe

Array of wells
PDMS layer

Gold layer
Glass prism

Biochip view by CCD before interaction

Probe 1
Probe 2
Probe 3
Negative controls
Surface Plasmon Resonance imaging DNA sensor

Principle and instrument

- Interaction light\electron → Plasmon excitation → SPR
- DNA Probes immobilization
- Affinity interaction → DNA Probe \ DNA Analyte → Signal (Sensorgram + Digital Image)
• SNP discrimination on non-amplified hDNA

Case study

• Gene ABCB1 → ATP-binding cassette transporter
• SNP rs 1045642 → Protein expression → Opioids drug absorption

Pharmacogenomic interest

Patient’s SNP → Tailored drug therapy with opioids (*Pain therapy*)

Mondiali di ciclismo a Firenze