POSTER PRESENTATIONS

PP001
LACTATE DEHYDROGENASE BIOSENSOR BASED ON AN HYBRID CNT-CONDUCTING POLYMER ELECTRODE

LOURDES AGÜÍ, MARCOS EGUÍLAZ, PALOMA YÁÑEZ-SEDEÑO*, and JOSE M. PINGARRÓN

Department of Analytical Chemistry, Faculty of Chemistry Complutense University of Madrid, 28040-Madrid, Spain
yseo@quim.ucm.es

Biosensors based on nanostructured electrochemical transducers have a great interest because of their particular advantages. Carbon nanotubes (CNTs) and conducting polymers have been used successfully for the preparation of hybrid electrodes which exhibit special properties due to the synergic effect from both components. However, poly(3--methylthiophene) P3MT has been scarcely applied for the fabrication of composite materials. In a previous report, we synthesized a P3MT/ CNTs hybrid composite onto a glassy carbon electrode (GCE)[1]. Recent investigations in our lab have revealed that this type of hybrid electrodes possess the ability to decrease significantly, the overpotential for electrochemical oxidation of H_2O_2 and NADH.

In this communication, a lactate biosensor in which the enzyme lactate dehydrogenase (LDH) was immobilized onto a CNTs-P3MT-GCE has been developed. The resulting design is very attractive because of its simplicity, high sensitivity and low potential without using redox mediators.

The influence of experimental variables that could affect the performance of the biosensor: enzyme immobilization procedure, CNTs loading, applied potential, pH and NAD^+ concentration, were investigated in order to optimize the electroanalytical characteristics of the amperometric detection.

A good electroanalytical behaviour of the hybrid electrode was observed. The NADH analytical signal obtained by CV in 0.1 M PBS at pH 7.4 was approximately, three times higher at CNTs-P3MT-GCE than those at CNTs-GCE or P3MT-GCE. Furthermore, amperometric measurements at the LDH biosensor in stirred 0.1 M PBS pH 8.0 in the presence of 2.5 mM NAD^+, using 300 mV as the potential value, allowed the achievement of a quantification limit of 1 μM lactate. A good electrode-to-electrode reproducibility (RSD: 7.4 %, n=5) was also obtained. The influence of other species: ethanol, citric, malic, tartaric, uric (UA) and ascorbic (AA) acids was also investigated. Only UA and AA interferred, but this effect can be drastically reduced by casting a Nafion film over the working electrode.

REFERENCE

1. Agüí L., Peña-Farfal C., Yáñez-Sedeño P., Pingarrón J. M.: Electrochim. Acta *52*, 7946 (2007).

PP002
STUDY OF METAL COMPLEXATION BY THIOL-RICH PEPTIDES USING BISMUTH FILM ELECTRODES

ARÍSTIDES ALBERICH *, NÚRIA SERRANO, CRISTINA ARIÑO, JOSÉ MANUEL DÍAZ-CRUZ, and MIQUEL ESTEBAN

Departament de Química Analítica, Facultat de Química, Universitat de Barcelona, Martí i Franquès 1-11, E – 08028 – Barcelona, Spain
aristidesah@hotmail.com

Living beings have developed different strategies to face up toxicity of heavy metals. Particularly, plants response with immobilisation, exclusion, chelation and compartmentalisation of the metal ions, cleaning up and restoring the damage ecosystems, main objective of the phytoremediation technologies.

A number of metal-binding ligands have nowadays been recognised in plants. Among them, phytochelatins (PC) play a key role since they are capable to bind metal ions -mainly via thiolate coordination-, to complex and then to place them into vacuoles where they are no longer toxic. PC's are polypeptides synthesized enzymatically by plants and constituted basically by three amino acids (Glu, Cys and Gly) with the general structure $(\gamma\text{-Glu-Cys})_n\text{-Gly}$, where *n* can be as high as 11, but it usually ranges from 2 to 6.

To understand the role of phytochelatins in the detoxification of metals, mechanisms of formation of PC-metal complexes must be examined. The combined use of voltammetric techniques and Multivariate Curve Resolution by Alternating Least Squares (MCR-ALS) has proved to be useful to obtain the possible stoichiometries and some suggestions about the relative stability of the complexes, especially with Cd(II) and Zn(II). However, for the study of Pb(II)-PC systems this approach presents the problem that thiol-rich peptides facilitate the oxidation of mercury electrodes, producing many anodic signals (of free ligands and some complexes) which appear in the reduction region of Pb(II)[1]. These anodic signals strongly overlap with the electrodic signals to be studied and, moreover, they undergo potential shifts and over-distort baselines, complicating in all cases a complete resolution of the complexation mechanisms.

A suitable solution could be the use of electrodes based in other material than mercury, expecting the non-formation of these signals and therefore the simplification of electroanalytical data. Bismuth film electrode (BiFE)[2] is an electrochemically attractive and environmentally friendly electrode that has proved to be a good alternative to the prevalent use of mercury electrodes for the determination of trace metals and organic compounds[3].

The present work tries to examine the possibilities of the BiFE for the study of the complexation of phytochelatins and related compounds by lead, checking what species of the

complexation process provide signals in this electrode, its dependence on metal speciation, and the possible application of MCR-ALS.

As regards to the latest point, the applicability of this chemometric tool needs a good linearity of the data, but a peak splitting effect has been observed for some heavy metals in stripping techniques using bismuth film electrode[4]. Thus, a rief study of this possible behavior in differential pulse voltammetry, the more widespread technique used in complexation analysis, is carried out.

The authors acknowledge support of the Spanish Ministry of Education and Science (Project CTQ2006-14385-C02-01/ BQU) and from the Generalitat of Catalonia (project 2005SGR00186).

REFERENCES

1. Alberich A., Ariño C. Díaz-Cruz J. M., Esteban M.: Talanta *71*, 344 (2007).
2. Wang J., Lu J., Hocevar S. B, Farias A. M., Ogorevc B.: Anal. Chem. *72*, 3218 (2000).
3. Economou A.: Trends Anal. Chem. *24*, 334 (2005).
4. Serrano N., Alberich A., Díaz-Cruz J. M., Ariño C., Esteban M.: Electrochim. Acta, submitted.

PP003

CYP250 2B4 COVALENTLY BOUNDED TO CARBON AND GOLD SCREEN PRINTED ELECTRODES BY DIAZONIUM AND THIOLS MONOLAYERS

M. ASUNCION ALONSO-LOMILLO[a]*, CEREN YARDIMCI[b], OLGA DOMINGUEZ RENEDO[a], and M. JULIA ARCOS MARTINEZ[a]

[a] Analytical Chemistry Department, Faculty of Sciences, University of Burgos, Pza. Misael Bañuelos s/n, 09001 Burgos, Spain, [b] Analytical Chemistry Department, Faculty of Pharmacy, Hacettepe University, 06100 Sihhiye, Ankara, Turkey malomillo@ubu.es

Modification of carbon and gold screen printed electrodes (SPEs) surfaces has been performed in order to ensure the covalent immobilization of biological molecules. SPE units based on three-electrode configurations have been homemade printed by using a DEK 248 (Fig. 1). Carbon counter, Ag/AgCl reference and gold or carbon working electrodes have been used.

Fig. 1. **Screen printed carbon electrodes units developed**

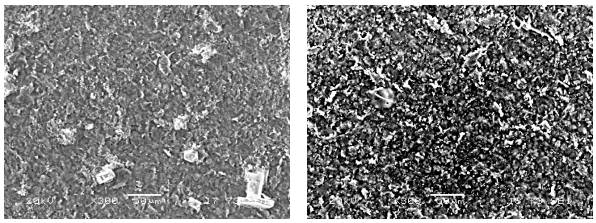

Fig. 2. **Carbon working electrode modified by gold nanoparticles (left) and by gold nanoparticles-MPA (right)**

A first step consisted of the electrochemical reduction of diazonium salts onto the carbon SPEs, which leads to covalent attachment of aryl radicals. This chemical modification of the composite may improve its properties[1–4].

In the case of gold SPEs, 3-Mercaptopropionic acid (MPA) was self assembled[5]. The performance of this working electrode in the building of biosensors has been improved by electrochemical deposition of gold nanoparticles before the self-assembled monolayer (SAM) modification.

The latter modification has also been attempted onto the carbon working electrodes (Fig. 2, ref.[6]), showing an improvement of conductivity defined by the analysis of the cyclic voltammetric peaks of ferricyanide system.

Then, these four different modified SPEs have been covalently bonded to Cytochrome P450 2B4 (CYP450) for the selective determination of Phenobarbital.

Experimental variables in the enzyme immobilization and in the chronoamperometric determination of PB have been optimized by means of the experimental design methodology.

Reproducibility, repeatability and limit of detection of the CYP450 biosensor have been analyzed. In order to check the performance of the proposed methods, the sensors have been applied to the PB determination in commercial pharmaceutical drugs.

Authors would like to acknowledge funding via the Spanish Ministry of Education and Science (MAT2005-01767) and Dr. J.M. Trejo from the Hospital General Yagúe in Burgos for his kind collaboration. M.A. Alonso-Lomillo is funded by a Ramón y Cajal fellowship from the Spanish Ministry of Education and Science.

REFERENCES

1. Allongue P., Delamar M., Desbat B., Fagebaume O., Hitmi R., Pinson J., Saveant J. M.: J. Am. Chem. Soc. *119*, 201 (1997).
2. Pinson J., Podvorica F.: Chem. Soc. Rev. *34*, 429 (2005).
3. Corgier B. P., Marquette C. A., Blum L. J.: J. Am. Chem. Soc. *127*, 18328 (2005).
4. Alonso-Lomillo M. A., Rudiger O., Maroto-Valiente A., Velez M., Rodriguez-Ramos I., Munoz F. J., Fernandez V. M., De Lacey A. L.: Nano Lett. *7*, 1603 (2007).
5. Susmel S., Guilbault G. G., O'Sullivan C. K.: Biosens. Bioelectron. *18*, 881 (2003).
6. Renedo O. D., Martinez M. J. A.: Anal. Chim. Acta *589*, 255 (2007).

PP004
VOLTAMMETRIC LEAD DETECTION USING LTCC BASED MICROFLOW SYSTEM

EVA ARASA and JULIAN ALONSO*

Grup de Sensors i Biosensors, Departament de Química Analítica, Facultat de Ciències, Edifici Cn-Nord, Universitat Autònoma de Barcelona, 08193, Bellaterra
julian.alonso@uab.es

Electrochemical detectors are an attractive alternative in microfluidics because small sample volumes are needed and provide low detection limits.

The aim of the present work is the construction of miniaturized voltammetric system for the determination of heavy metals., in this case lead ion. The microanalytical system was constructed using the LTCC (Low Temperature Co-Fired Ceramics) technology. Monolithic integration of fluidic and detection system was done using the same technology and substrate. The voltammetric system includes two platinum sheets (5 × 5 mm) acting as working and counter electrode and a miniaturized Ag/AgCl reference electrode fabricated using embebed Ag conductive pads screen-printed over the auxiliary microfluidic channel. The dimensions of the whole microsystem was 55 × 25 × 2 mm (see Fig. 1).

Results obtained demonstrate the posibility to determine lead ions. However, the detection limit obtained was high for its aplication in drinking water. Moreover, the device has a limited life time due to a irreversible passivation of working electrode under the working conditions. Better results had been obtained in preliminary experiments when platinium was replaced for gold microchip as working electrode.

In order to improve the analytical features and increase the device life span, the performance of other metals like gold and silver as working electrodes will be tested.

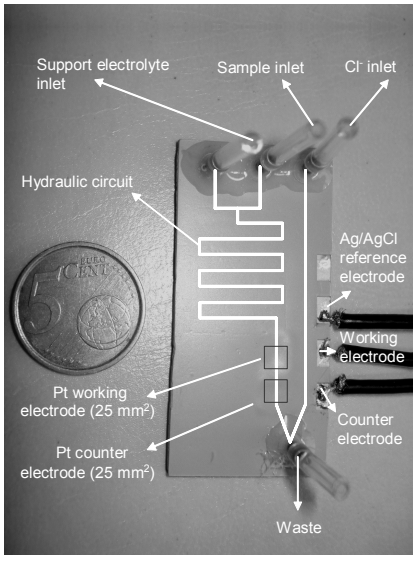

Fig. 1. **LTCC device picture**

PP005
DETERMINATION OF As(III) USING SILVER NANOPARTICLE-MODIFIED SCREEN-PRINTED ELECTRODES

M. JULIA ARCOS-MARTÍNEZ, OLGA DOMÍNGUEZ-RENEDO, and SILVIA SANLLORENTE-MÉNDEZ*

Department of Chemistry, University of Burgos, Plaza Misael Bañuelos, E-09001, Burgos, Spain
jarcos@ubu.es, olgado@ubu.es, silsan@ubu.es

Arsenic is one of the most toxic elements found in nature, and it constitutes one of the main concerns in relation to human health. Arsenic is second only to lead as the main inorganic contaminant in the original US Environmental Protection Agency's (EPA) National Priority List (NPL) of Superfund sites. Arsenic can be found in drinking water, in the air as volatile arsines, and in soil, where it can concentrate if absorbed on soil components. Consumption of water with high concentrations of this non metal over an extended period of time causes serious diseases, including development of cardiovascular and peripheral vascular disease anomalies, neurological and neurobehavioral disorders, diabetes, hearing loss, portal fibrosis of the liver, lung fibrosis, hematological disorders, and carcinoma, especially in rural and semi-urban areas where water irrigates food or drinking water is often used without treatment[1].

Several analytical techniques have been used for arsenic determination at trace levels, such as atomic absorption spectrometry[2], atomic fluorescence spectrometry[3], and high-performance liquid chromatograph-inductively coupled plasma mass spectrometry (HPLC-ICPMS)[4]. The most reliable techniques are more suitable for laboratory conditions only and are, in addition, time consuming. In contrast, electrochemical methods provide accurate measurements of low concentrations of metal ions at the ppb levels with rapid analysis times and low cost instrumentation.

Screen-printed electrodes are planar devices with plastic substrates that are coated with layers of electroconductive and insulating inks at a controlled thickness. The advent of screen-printed (thick-film) technology has made it possible to mass-produce inexpensive disposable electrodes for use with electrochemical instruments[5].

The design of new nanoscale materials has acquired ever-greater importance in recent years due to their wide-ranging applications in various fields. Among these materials, metallic nanoparticles are of great interest due to their important properties and their numerous possible applications. Moreover, the advent of screen-printed (thick-film) technology has made it possible to mass-produce inexpensive disposable electrodes for use with electrochemical instruments.

The aim of this work is to determine As(III) by differential pulse voltammetry (DPV) using screen-printed electrodes modified with silver nanoparticles and, to the best of the author's knowledge, presents the first ever electrochemical detection of As(III) with this type of electrodes.

The financial support made available by the Junta de Castilla y León (BU022A07) and the Ministerio de Educación y Ciencia (MAT2005-01767) is gratefully acknowledged.

REFERENCES
1. Mandal B.K., Suzuki K.T.: Talanta *58*, 201 (2002).
2. Niedzielski P.: Anal. Chim. Acta *551*, 199 (2005).
3. Ma G., Xie W. B., Liu J., Li X. Y., Jin Y. W.: Spectrosc. Spectr. Anal. *27*, 807 (2007).
4. Brennan R. G., Murdock S. A. E. O., Farmand M., Kahen K., Samii S., Gray J. M., Montaser A.: J. Anal. At. Spectrom. *22*, 1199 (2007).
5. Wang J., Lu J., Tian B., Yarnitzky C.: J. Electroanal. Chem. *361*, 77 (1993).

PP006
ANALYSIS OF MACROCYCLIC LACTONE MYCOTOXINS IN MAIZE FLOUR SAMPLES BY CAPILLARY ELECTROPHORESIS WITH AMPEROMETRIC DETECTION

ALBERTO SÁNCHEZ ARRIBAS[a], MÓNICA MORENO[a], ESPERANZA BERMEJO[a], MANUEL CHICHARRO[a], ANTONIO ZAPARDIEL[b], HELENA TÉLLEZ[c], MOHAMMED ZOUGAGH[c], and ÁNGEL RÍOS[c]

[a] Dpto. de Química Analítica y Análisis Instrumental, Facultad de Ciencias, Universidad Autónoma de Madrid, 28049 Madrid, Spain, [b] Dpto. de Ciencias Analíticas. Facultad de Ciencias, Universidad Nacional de Educación a Distancia (UNED), 28040 Madrid, Spain, [c] Dpto. de Química Analítica y Tecnología de los Alimentos, Facultad de Química, Universidad de Castilla-La Mancha, 13004 Ciudad Real, Spain
alberto.sanchez@uam.es

Mycotoxins are secondary metabolites produced by fungal species, growing on agricultural products during cultivation, harvest, transport and storage. Their occurrence in food has been recognized as potential human hazard either caused direct contamination of grains and fruits and their products or by "carry over" of mycotoxins and their metabolites in animal tissues. Zearalenone (ZON) and its metabolites α-zearalenol (α-ZOL) and β-zearalenol (β-ZOL) are produced by *Fusarium* species, which colonize several grains and high amounts of ZON can most frequently be found on maize, wheat, oats and barley. Maximum levels of 200 and 100 ppb have been fixed for ZON in unprocessed corn and unprocessed cereals other than corn, respectively.

In this communication we report on a new analytical scheme for the screening and quantification of ZON, α-ZOL and β-ZOL by capillary electrophoresis with amperometric detection in extracts collected by supercritical fluid extraction (SFE) from maize flour samples. The sample screening was carried out by capillary zone electrophoresis (CZE) using 25 mM borate running buffer at pH 9.2 and 25 kV as separation voltage and following the amperometric signal at +700 mV of a carbon paste electrode (CPE, 0.5 mm diameter). In this way, total mycotoxins containing is determined and samples could be processed in 4 minutes with a detection limit of 20 ppb, enough to discriminate between positive (more than 200 ppb total mycotoxins, reference value stablished for ZON by directive 2005/38/EC)) and negative samples (less than 200 ppb total mycotoxins). Positive samples were then subjected to CZE separation and quantification of each analyte with 50 mM borate running buffer modified with 30% methanol at pH 9.7 and 17.5 kV as separation voltage. Under these conditions, separation is achieved in 15 minutes with detection limits of 50 ppb for each analyte.

Ministerio de Educación y Ciencia of Spain (Grants CTQ2004-06334-C02-01-02 and CTQ2007-61830) and JJCC Castilla-La Mancha (Grant PCC08-015-0722) are gratefully acknowledged for funding this work.

PP007
MELDOLA'S BLUE/SINGLE-WALLED CARBON NANOTUBES/SOL-GEL COMPOSITE ELECTRODE FOR ELECTROCHEMICAL ANALYSIS OF NADH

ADINA ARVINTE[a], ADAMA SESAY[b], VESA VIRTANEN[b], and CAMELIA BALA[a,*]

[a] LaborQ, Department of Analytical Chemistry, University of Bucharest,4-12 Regina Elisabeta Blvd. 030018 Buchares-3, [b] Kajaani University Consortium, Biotechnology Laboratory, Salmelantie 43, 88600 Sotkamo, Finland
cbala@chem.unibuc.ro

An electrochemical sensitive electrode for NADH determination was developed based on the integration of the redox mediator (Meldola's blue, MB) and single-walled carbon nanotubes (SWNT) with polymeric matrix (silica sol-gel). The resulting composite material was used for coating the surface of screen-printed electrodes and investigated and characterized by electrochemical methods. Analytical parameters of the sensors with and without SWNT in the hybrid film were compared, and the results showed that analytical performance of the sensor could be improved greatly after introduction of the SWNT (sensitivity 3 times higher and a wider linear range). Experimental parameters of the NADH sensor, such as applied potential, amount of SWNT, drying time for sol-gel matrix were studied and optimized. The applied working potential (–0.050 V vs. Ag/AgCl) is low, consequently avoiding the interference from electroactive compounds and thus increasing the selectivity of the sensor.

This nanomaterials-based composite may be used as electrochemical transducers and have potential application for designing a variety of NAD+-dependent electrochemical biosensors.

We acknowledge financial support from MECT contract no. 1637/2007, 51-006/2007, 11-055/2007 and Bioprof.

REFERENCES
1. Song Z., Huang J.-D., Wu B.-Y., Shi H.-B., Anzai J.-I., Chen Q.: Sens. Actuators, B *115*, 626 (2006).
2. Salimi A., Compton R. G., Hallaj R. : Anal. Biochem. *333*, 49 (2004).
3. Du P., Liu S., Wu P., Cai C. : Electrochim. Acta *53*, 1811 (2007).
4. Sampath S., Lev O.: J. Electroanal. Chem. *446*, 57 (1998).

PP008

ELECTROCHEMICAL ANALYSIS OF CORROSION BEHAVIOUR OF DENTAL AMALGAMS

BAJSMAN ANITA[a]*, MALIC MIRJANA[a], KAHROVIC EMIRA[b], SABINA BEGIC[b], KONJHODZIC - PRCIC ALMA[a], TURKUSIC EMIR[a], and KALCHER KURT[c]

[a] Dental Faculty, University of Sarajevo, Bosnia and Herzegovina, [b] Faculty of Science, University of Sarajevo, Bosnia and Herzegovina, [c] Department for Analytical Chemistry, Institute for Chemistry, Karl-Franzens University Graz, Austria

Dental Amalgam represents restorative material that has been used in dentistry since it's beginings. Throughout history of its use there have been certain dilemmas within dental profession that are focused on dental amalgam safety, regarding harmfulness of mercury from amalgam. Attitudes in this scientific area were, and still are, so divided, that we speak about three "amalgam wars". Mercury is one of basic components of dental amalgam. From toxicological point of view it is one of the most toxic heavy metals, considering all possible risks, both for dentists and patients. Antiamalgamists based their based their opposing on dental amalgam use mostly on the fact that there is a corrosion of amalgam within oral cavity, under different physical and chemical influences. Contrary, proamalgamists state that there is no proof that could support attitude that mercury from dental amalgam makes influence on general health and well-being of patient.

According to that, goals of this investigation were:

1. To examine quantity of corrosion of 5 different dental amalgams *in vitro,*
2. To make quantitative and qualitative analysis of corrosion products of analyzed amalgams *in vitro*,
3. To establish influence of polishing of dental amalgams on degree of their corrosion.

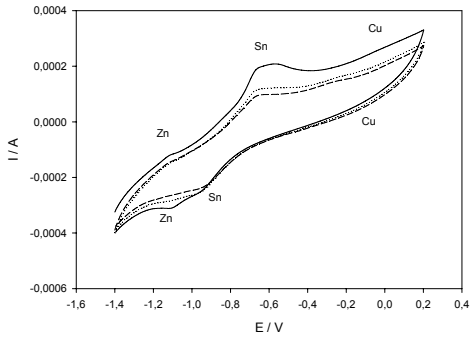

Fig. 1. **"NG70 Septalloy"in artificial saliva** (Quezada Duffo – Castillo), unpolished

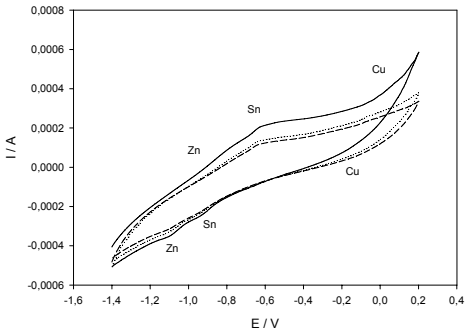

Fig. 2. **"NG70 Septalloy"in artificial saliva** (Quezada Duffo – Castillo), polished

Experimental material is represented by 5 different dental amalgams. Electrodes were made from them, respecting manner that is usual for electrochemical investigations. Those electrodes represented artificial teeth. Electrodes were tested using cyclic voltammetry (unpolished and polished) and inductively coupled plasma-mass spectrometry (ICP-MS) – polished. Electrochemical behavior of electrodes was tested in artificial saliva (Quezada Duffo – Castillo).

Cyclic voltammetry proved that corrosion of dental amalgams within defined potential range always happens. Its intensity depends on polishing of amalgam surface. They were determined as isotopes. According to this *in vitro* investigation, quantities of liberated mercury and other metals can not be consider harmful for patient, according to standards of World Health Organization (WHO). Polishing of dental amalgams has positive influence on their corrosion behavior because of reduced corrosion surface.

The authors wish to acknowledge support for this work by the Ministry of education and science Sarajevo Canton and Federal ministry of education and science.

REFERENCES

1. Wang J.: *Study of Electrode Reactions*. In: (Wang J., ed.). *Analytical electrochemistry*, 2nd ed., p. 28−59. Wiley-VCH, New York 2001.
2. Horasawa N., Nakajima H. J. L., Takahaschi S., Okabe T.: Dent. Mater *12*, 154 (1996).
3. Leung V. W.-H., Darvell B. W.: J. Dent. *6*, 475 (1997).
4. Duffo G. S., Quezada Castillo E.: Corrosion *60*, 594 (2004).
5. Karov J., Hinberg I.: J. Oral. Rehab. *28*, 212 (2001).
6. Yap A.V. J., Ng B. L., Blackwood D. J.: J. Oral. Rehab. *31*, 595 (2004).

PP009
FABRICATION AND APPLICATIONS OF INJECTION-MOULDED POLYMERIC CELL-ON-A CHIP DEVICES WITH INTEGRATED CONDUCTING ELECTRODES FOR ELECTROANALYTICAL MEASUREMENTS

S. J. BALDOCK[a], N. G.NASERI[b], Z. M. REDHA[a], P. R. FIELDEN[a], N. J. GODDARD[a], A. ECONOMOU[c]*, and C. KOKKINOS[c]

[a] School of Chemical Engineering and Analytical Science, University of Manchester, PO BOX 88, Manchester, M60 1QD, [b] Petroleum University of Technology, PO BOX 63431, Kut Abdualah, Ahwaz-Abadan Rd., Ahwaz, Iran, [c] Department of Chemistry, University of Athens, Panepistimiopolis, Athens 157 71, Greece

The use of microfabrication technologies enables the mass fabrication of small, reproducible, low-cost, portable and disposable sensors[1]. In particular, the integration of electrochemical analysis techniques into microfluidic devices is an area of increasing interest[2,3]. Injection-moulding offers some advantages over alternative micro-fabrication approaches: in addition to providing planar structures (such as channels) quickly and at a reasonable cost, it enables the creation of three-dimensional structures and the incorporation of preformed elements (such as electrodes) into the plastic during the moulding process.

In this work, cell-on-a-chip devices for electrochemical analysis were injection-moulded from polymeric materials to form plastic micro-devices. The electrodes were moulded from polystyrene loaded with carbon-fibres (40 %) By using a mould insert and an injection over-moulding procedure, the polymer electrodes were integrated into the device substrate. The moulded electrodes can be used unmodified or can be pre-coated either with metal layers (such as Au, Ag or Bi by e-beam/thermal evaporation and eletroplating) or with Ag/Cl paste, thereby forming working, reference and counter electrodes as required.

Furthermore, the moulded electrodes were be incorporated into micro-flow-cells. The flow channels in these devices were produced in a clear grade of polystyrene. Ultrasonic welding was used to bond the substrate containing the electrode to the substrate containing the microchannel. Figure 1(i) illustrates the two parts forming a microfluidic device while Figure 1(ii) shows the assembled micro-flow-cell.

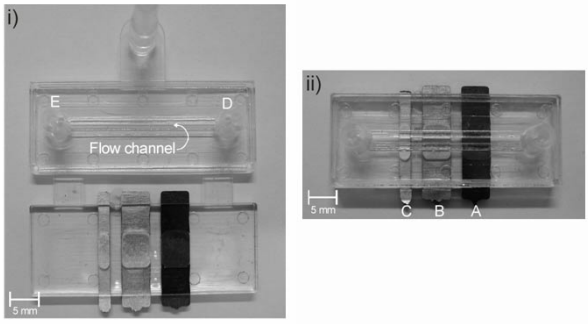

Fig. 1. **A example of an injection-moulded microfabricated device**: i) the two separate parts forming the flow cel, and; ii) the assembed flow-cell (A is the carbon CE, B is the gold WE, C is the Ag RE, D is the solution outlet and E is the solution inlet)

Electrochemical techniques such as cyclic voltammetry (CV), anodic stripping voltammetry (ASV), electrochemiluminescence (ECL) and catalytic adsorptive stripping voltammetry (CAdSV) have been tested on the micro-fabricated devices. In addition electrochemical procedures for *in situ* activation of the working electrodes, long term drift studies on the reference electrodes and device-to-device reproducibility were evaluated.

The results obtained demonstrate "proof-of-principle" of these cell-on-a-chip devices and suggest that they can be employed as disposable sensors in electronalysis.

REFERENCES

1. Becker H., Locascio L. E.: Talanta *56*, 267 (2002).
2. Suzuki H.: Electroanalysis *12*, 703 (2000).
3. Bakker E., Telting-Diaz M.: Anal. Chem. *74*, 2781 (2002).

PP010
UTILIZING OF BRDICKA REACTION FOR ANALYSIS OF LOW MOLECULAR MASS THIOLS

JIRI BALOUN[a,b], VIOLETTA SHESTIVSKA[a,b], VACLAV DIOPAN[a,b], VOJTECH ADAM[a,c], LADISLAV HAVEL[a], JOSEF ZEHNALEK[b], MIROSLAVA BEKLOVA[d], RENE KIZEK[b]*

[a] Department of Plant Biology, [b] Department of Chemistry and Biochemistry, and [c] Department of Animal Nutrition and Forage Production, Faculty of Agronomy, Mendel University of Agriculture and Forestry, Zemedelska 1, CZ-613 00 Brno, Czech Republic, [d] Department of Veterinary Ecology and Environmental Protection, Faculty of Veterinary Hygiene and Ecology, University of Veterinary and Pharmaceutical Sciences, Palackeho 1-3, CZ-612 42 Brno, Czech Republic kizek@sci.muni.cz

Low molecular mass thiols represents group of compounds rich in –SH moieties. Due to reactivity of this moiety thiols play number of crucial roles in organisms e.g. control of gene expression and receptor signalling, signal transduction and heavy metal detoxification, and many others. Glutathione (GSH) belongs to the most important thiols. As a ubiquitous tripeptide thiol it is a vital intra- and extra-cellular protective antioxidant. Glutathione is found almost exclusively in its reduced form; since the enzyme, which reverts it from its oxidized form (GSSG) called glutathione reductase, is constitutively active and inducible upon oxidative stress. The sulfhydryl group of glutathione is highly reactive and is often found conjugated to other molecules such as nitric oxide (NO) via its sulfhydryl moiety. Particularly, nitrosation of the glutathione may serve as a signal event and/or as a deposition of NO to S nitrosoglutathione (GSNO). The aim of this work was to utilize Brdicka reaction to study GSH, GSSG and GSNO.

Primarily we optimized the experimental conditions to detect the thiols of interest. Under the optimal conditions we measured the dependence of Cat2 peak height on concentration of the thiols of interest. The dependence was linear from 5 to 100 μg ml^{-1} for all thiols with R^2 higher than 0.99. The detection limits were lower than 1 μg ml^{-1} for all thiols. Moreover the differential pulse voltammograms differed markedly according to thiols determined.

We gratefully acknowledge the GACR 522/07/0692 for financial support to this work.

REFERENCES

1. Vitecek J., Petrlova J., Petrek J., Adam V., Potesil D., Havel L., Mikelova R., Trnkova L., Kizek R.: Electrochim. Acta *51*, 5087 (2006).
2. Zitka O., Huska D., Krizkova S., Adam V., Chavis G. J., Trnkova L., Horna A., Hubalek J., Kizek R.: Sensors *7*, 1256 (2007).

PP011
STRATEGIES TO DEVELOP AMPEROMETRIC BIOELECTRODES FOR INFECTION DIAGNOSIS

MARÍA S. BELLUZO[a], MARÍA E. RIBONE[a], EMMA STELLA[b], AGUSTINA DE LA IGLESIA[b], IVÁN S. MARCIPAR[c], HÉCTOR R. MORBIDONI[b], and CLAUDIA M. LAGIER[a]*

[a] *Analytical Chem. Dep., Biochemistry Faculty,*
[b] *Microbiology Dep., Medical Sciences Faculty. Universidad Nacional de Rosario. Suipacha 531 (2000) Argentina,*
[c] *INTEBIO, UNL clagier@fbioyf.unr.edu.ar*

Most of infection diagnostic tests rely on detecting either the infecting agent (whole microorganism or selected molecules) or specific antibodies raised by the host[1]. We developed amperometric systems to detect both of these infection markers. In one approach, we detected antibodies to *Trypanosoma cruzi* parasite, by using a classical indirect immunoassay with electrochemical detection. We used recombinant proteins specifically designed to achieve three goals: *i*) to avoid cross-reactions with antibodies generated by the host during other diseases, *ii*) to link these molecules to the electrode in an oriented manner so as to enhance sensitivity and *iii*) to attach them covalently, so that the device could be reused after rigorous treatments. This strategy allowed us not only to efficiently and specifically detect the infection marker but also to regenerate the device to be reused in consecutive sample analysis, envisaging automatization[2]. The results obtained showed 100 % specificity for the entire positive and negative samples assayed. The sensitivity was in the same order as that obtained using a commercial ELISA kit following an indirect immunoassay format with spectrophotometric detection. The biosensors could be regenerated and then reused to analyze 10 different samples consecutively before showing a signal loss of 50 % ($P < 0.05$), with a cut-off value corresponding to a 20 % of the original signal measured. In another approach, to verify the presence of pathogenic bacteria, we detected by amperometry the consumption of catechol, a model substrate of bacterial enzymes. Considering that several bacteria have enzymatic systems that can degrade catechol, we enhanced the assay specificity by biologically releasing exclusively the enzymatic content of the target bacteria. Amperometries were performed in a classical three-electrode cell where a carbon paste electrode was the working electrode, using PBS as bathing electrolyte. The current difference obtained between the background and the tested sample was used to monitor the presence of catechol-degrading enzymatic system. Results obtained with samples of intact bacteria and specifically lysated showed significant differences ($P < 0.05$).

ANPCyT & CONICET are acknowledged for grants PICTR2002-00057 and PIP5303, respectively.

REFERENCES

1. Belluzo M. S., Ribone M. E., Lagier C. M.: Sensors *8*, 1366 (2008).
2. Lagier C. M., Marcipar I. S., Ribone M. E., Belluzo M. S., Camussone C. : INPI, Inscription N° P060103687.

PP012
SELECTIVE DETECTION OF MOLECULES WITH DIFFERENT SIZE BY GOLD ELECTRODE MODIFIED WITH DI-*n*-OCTADECYL-DISULPHIDE. APPLICATION TO VITAMIN B_2 DETERMINATION

ELÍAS BLANCO, PEDRO HERNÁNDEZ, JOSÉ VICENTE, and LUCAS HERNÁNDEZ*

Department of Analical Chemistry and Instrumental Analisys, Faculty of Sciences, Universidad Autónoma de Madrid,28049 Madrid, Spain
lucas.hernandez@uam.es

The objective of our work is modifying a gold electrode to allow selective arrival of molecules with different size at the surface of electrode through formed channels. It is got with the control of surface recover by di-*n*-octadecyl-disulphide. This control is obtained by change the concentra-

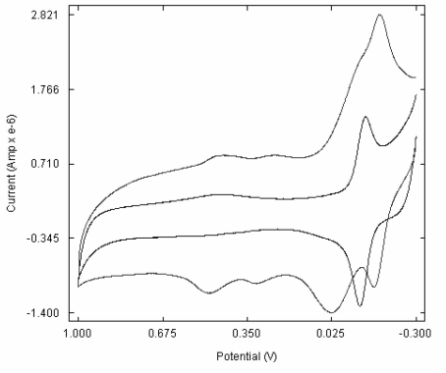

Fig. 1. **Cyclic voltammograms of vitamin B_2 in H_2SO_4 0.1 M at 100 mV/s**. In red, without modification; in blue, with modification

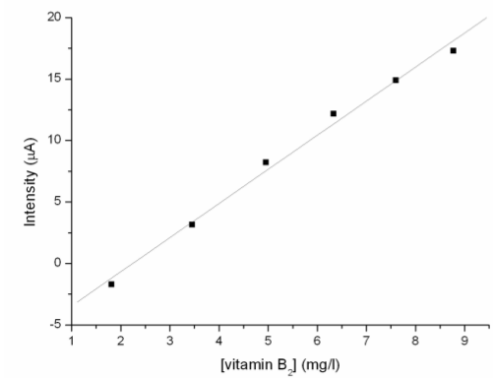

Fig 2. **Calibration curve of vitamin B_2 using SWV**.
I = 2,7748 C – 6,21106, r^2 = 0,996

tion of di-*n*-octadecyl-disulphide (solved in tetrahydrofuran) and stay time of the electrode in solution of the modifier. Ferrocen, hydroquinone and vitamin B_2 are the studied molecules. All work is being done in acid media (sulphuric 0,1 M) what is the most adequate media for its. The used electrochemistry technique is cyclic voltammetry what allows us to study oxidized and reduced molecules of ours compounds in the electrode surface. Each molecule and the mixture are studied with modifier and without modifier using different times and concentrations. In Fig. 1 is represented the response of the electrode with modifier and without modifier for vitamin B_2 and in Fig. 2 a calibration curve using the electrochemistry technique SWV. We are now working in the analysis of a drug formulation with vitamin B_2.We can deduce in these conditions if possible cause di-*n*-octadecyl-disulphide to make a physical barrier for molecules and to do selective detection by molecular size.

We are grateful to MEC of Spain for financial support (CTQ2004-041/BQU).

PP013
MINIATURE, MAINTENANCE AND INTERNAL SOLUTION FREE MULTI-ELECTRODE GALVANIC CELLS FOR POTENTIOMETRIC MEASUREMENTS

TERESA BLAZ, JAN MIGDALSKI, BOGUSLAW BAS, BARBARA ZRALKA, and ANDRZEJ LEWENSTAM

University of Science and Technology, Faculty of Material Science and Ceramics, al. Mickiewicza 30, 30-059 Cracow, Poland
teblaz@uci.agh.edu.pl

Construction of the miniaturized, maintenance and internal solution free galvanic cells are an important challenge, because of the envisaged medical and biochemical applications. The use of conducting polymers in designing of such a cell was found to be especially beneficial.

It has been shown that electropolymerization is a convenient method to obtain conducting polymer films (CP films) doped with bulky dopants, e.g. metal complexing or biological buffer ligands, which retain their specific chemical properties inside CP-films[1–5]. In this way it is possible to obtain conducting polymer based reference or indicator membranes as well as mediating layers for solid-contact ion-selective or reference electrodes, consequently to obtain miniaturized, maintenance and internal solution-free multi-electrode galvanic cells. In this communication recent advances describing technology, electrochemical properties and selected applications are presented.

Financial support from the State Committee for Scientific Research (KBN project 3T08E 085 30) is gratefully acknowledged.

REFERENCES

1. Migdalski J., Błaż T., Lewenstam A.: Anal. Chim. Acta *322*, 141 (1996).
2. Migdalski J., Błaż T., Lewenstam A.: Chem. Anal. (Warsaw) *47*, 371 (2002).
3. Błaż T., Migdalski J., Lewenstam A.: Talanta *52*, 319 (2000).
4. Migdalski J., Błaż T., Żrałka B., Lewenstam A.: Anal. Chim. Acta *594*, 204 (2007).
5. Błaż T., Migdalski J., Lewenstam A.: Analyst *130*, 637 (2005).

PP014
THE SILVER AMALGAM FILM ELECTRODE IN ADSORPTIVE STRIPPING VOLTAMMETRIC DETERMINATION OF PALLADIUM(II) AS ITS DIMETHYDIOXIME COMPLEX

ANDRZEJ BOBROWSKI[a]*, PAWEŁ KAPTURSKI[a], VALENTIN MIRCESKI[b], FILIP SPASOVSKI[b], and JERZY ZARĘBSKI[a]

[a] Department of Building Materials Technology, Faculty of Materials Science and Ceramics, AGH - University of Science and Technology, Al. Mickiewicza 30, 30-059 Kraków, Poland,
[b] Institute of Chemistry, Faculty of Natural Sciences and Mathematics, University Cyril & Methodius, 91000 Skopje, Republic of Macedonia
gcbobrow@cyr-kr.edu.pl

A new, cylindrical silver-based amalgam film electrode (Hg(Ag)FE) of prolonged analytical application has recently been introduced for voltammetric measurements and has been applied for the determination of Pb, Zn, Cd, Cu, Cr, Ni and Co by means of stripping voltammetry[1,2,4] and stripping chronopotentiometry[3]. The electrode design enables easy and quick regeneration of the liquid layer before each measurement cycle and, consequently, good reproducibility of results. The paper features a description of a sensitive adsorptive stripping voltammetric protocol for palladium(II) determination in the presence of dimethylglyoxime (DMG) at an amalgam film electrode (Hg(Ag)FE) of prolonged analytical applicability. The procedure is based on the adsorptive preconcentration of the Pd(II)-dimethylglyoxime complex onto the (Hg(Ag)FE) at –0.55 V, followed by a negatively-going

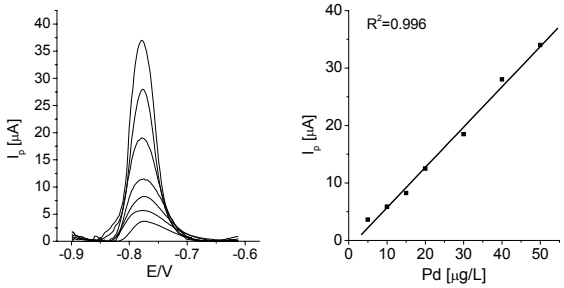

Fig. 1. **The SW-AdSV curves of Pd for successively increased concentrations of palladium in the range from 5 to 50 μg L^{-1}** (obtained after subtraction of the background curve). Inset: calibration plot. Supporting electrolyte: 0.1 M acetate buffer (pH 4.4), 0.2 mM DMG. E_{dep} = – 0.55 V, t_{dep} = 60 s

square-wave voltammetric scan. Factors affecting the stripping performance, such as the composition of the supporting electrolyte, including different ligands, DMG concentration, pH, potential and time of preconcentration, type of voltammetric mode, SW pulse amplitude and electrode surface have been investigated and optimized. The optimized procedure yields favorable and highly stable stripping responses with good precision (RSD = 3.0 % for a Pd concentration of 2.0 μg L^{-1}), a low detection limit (0.13 μg L^{-1} for a Pd concentration of 0.3 μg L^{-1} Pd(II)), and good linearity (from 0.5 μg L^{-1} up to 100 μg L^{-1}, R^2 = 0.996, Fig. 1) for a deposition time of 60 s. Possible interferences from coexisting ions and surface active substances were also studied.

Financial support from the Ministry of Science and Higher Education (project N507 063 32/1767) is gratefully acknowledged. F. Spasovski would also like to thank the CEEPUS programme for support (project No.Cz-212-07/08).

REFERENCES

1. Baś B., Kowalski Z.: Electroanalysis *14*, 1067 (2002).
2. Baś B.: Anal.Chim. Acta *570*, 195 (2006).
3. Kapturski P., Bobrowski A.: Electroanalysis *19*, 1863 (2007).
4. Kapturski P., Bobrowski A.: J. Electroanal. Chem., doi:10.1016/j.jelechem.2008.01.007 (2008).

PP015
A CATALYTIC ADSORPTIVE STRIPPING VOLTAMMETRIC PROCEDURE FOR HEXAVALENT CHROMIUM DETERMINATION IN PORE AND OVERLYING WATER

ANDRZEJ BOBROWSKI[a]*, PAWEŁ KAPTURSKI[a], JERZY ZARĘBSKI[a], JANUSZ DOMINIK[b], and DAVIDE VIGNATI[b]

[a] *Department of Building Materials Technology, Faculty of Materials Science and Ceramics, AGH - University of Science and Technology, Al. Mickiewicza 30, 30-059 Kraków, Poland,* [b] *Institute F.-A. Forel, University of Geneva, 1290 Versoix, Switzerland*
gcbobrow@cyr-kr.edu.pl

Catalytic Adsorptive Stripping Voltammetry (CAdSV) has proved to be useful in the determination of Cr(VI) with a very low detection limit in different water samples including rain[1], sea[2], river[2,3], lake[2,3] water and landfill leachate[4]. The method is based on the accumulation of the Cr(III)-DTPA complex formed instantaneously from the very active Cr(III) ions generated by the reduction of Cr(VI) at the electrode surface, and the utilization of the catalytic reaction in the presence of nitrate ions[2]. One of the main reasons why the application of electrochemical methods to the analysis of real samples has not always been successful is the inevitable presence of surface active substances (SAS). Depending on their concentration and type, SAS can partially or completely suppress the observed chromium voltammetric signal. In particular, overlying and pore water samples from natural aquatic systems often contain high concentrations of organic matter (up to 160 mg L^{-1}) coupled with low concentrations of chromium (less than 1 μg L^{-1} Cr(VI)), thus Cr(VI) determination is exceptionally difficult. This paper features a description of a new protocol for Cr(VI) determination in pore and overlying water samples.

In this work, well-known voltammetric procedures for the determination of chromium with DTPA and nitrate[1–4] have been adopted for the Cr(VI) quantification in the presence of an excess of SAS. This task was achieved by means of a selective separation of organic matter on Amberlite XAD-7 (ref.[5]) resin. The SAS were separated directly in the voltammetric cell by adding Amberlite XAD-7 (ref.[6]) and stirring the solution for 5 min before determination. The difference in CAdSV curves with and without separation is illustrated in Fig. 1. The procedure was applied in hexavalent chromium analysis in pore and overlying water samples collected in the upper Dunajec river and the Czorsztyn reservoir (Poland). The results obtained using the elaborated method were confirmed by those achieved by the CAdSV procedure based on matrix exchange in the flow system[7].

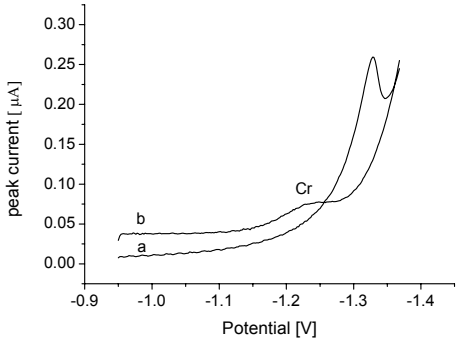

Fig. 1. **CAdS voltammograms obtained for overlying water sample**. Supporting electrolyte: 0.1 M acetate buffer, 0.25 M KNO_3, 0.01 M DTPA; pH 6.2 (a); Curve (b) – with addition of 0.6 g XAD–7 (5 min adsorption with stirring). E_{acc}= –0.95 V; t_{dep}= 15 s

Financial support from the Ministry of Science and Higher Education (project N507 063 32/1767) and from the Swiss National Science Foundation (grant 200020-109608) is gratefully acknowledged.

REFERENCES
1. Golimowski J., Valenta P., Nuernberg H. W: Fresenius' Z. Anal. Chem. *322*, 315 (1985).
2. Boussemart M., van den Berg C. M. G., Ghaddaf M.: Anal.Chim. Acta *262*, 103 (1992).
3. Bobrowski A., Dominik J., Niewiara E., Szalińska E., Vignati D., Zarębski J: Talanta *63*, 1003 (2004).
4. Li Y., Xue H.: Anal. Chim. Acta *448*, 121 (2001).
5. Petting M., Capri S.: Anal. Chim. Acta *540*, 239 (2005).
6. Grabarczyk M.: Anal. Bioanal. Chem. *390*, 979 (2008).
7. Korolczuk M., Grabarczyk M.: Anal. Chim. Acta *387*, 97 (1999).

PP016
ELECTROCHEMICAL AND MICROSCOPIC CHARACTERISTICS OF A LEAD-MODIFIED CARBON PASTE ELECTRODE

ANDRZEJ BOBROWSKI*, AGNIESZKA KRÓLICKA, and EWA ŁYCZKOWSKA

Faculty of Materials Science and Ceramics, AGH University of Science and Technology, Cracow, Poland
gcbobrow@cyf-kr.edu.pl

Carbon electrodes plated with metal films have been the subject of investigation for many years. Recently, the electrochemically plated lead layer was examined as the prospective electrodes' modifier[1,2]. The interest in this material was inspired by the possibility of plating the lead film on the support surface *in situ*, in fairly alkaline media. *In-situ* plating indisputably simplifies and shortens the experimental procedure for adsorptive stripping voltammetry, where work in such media is routine. The aim of this work is to show that a lead film plated *in situ* at a carbon paste support could work as the electrode for the adsorptive stripping voltammetric determination of cobalt traces in an ammonia buffer solution.

To show the applicability of the new electrode, a catalytic adsorptive Co system in a supporting electrolyte containing 0.1 M ammonia buffer, 5×10^{-4} M nioxime and 0.25 M nitrite was selected and investigated. Pb and Co ions were simultaneously accumulated *in situ* on the electrode surface. The lead layer was deposited electrochemically after applying the potential of −1.3 V ('nucleation potential') and then at −0.75 V, at which potential also the Co(II)-nioximate complex was pre-concentrated via adsorption. Instrumental parameters, such as the time of nucleation and formation of Pb film deposits, the time of accumulation of the Co-nioxime complex at the PbF/CPE, and the procedures of electrode regeneration, were optimized to obtain good reproducibility and sensitivity of the Co response. The optimized procedure yields favorable and highly stable stripping responses with good precision (*RSD* = 3 % for a 5×10^{-8} M Co) and good linearity (up to 5×10^{-7} M, coefficient of determination, R = 0.996). The detection limit was 4×10^{-10} M Co (0.023 μg L^{-1}) for an accumulation time of 120 s. The described method enables Co determination in the presence of a high excess of Ni or Zn. Finally, the voltammetric data were correlated with structural characterization by means of scanning electron microscopy (SEM) and X-ray fluorescence spectroscopy (XRF).

Financial support from the Ministry of Science and Higher Education (project N507 063 32/1767) is gratefully acknowledged.

REFERENCES
1. Korolczuk M., Tyszczuk K., Grabarczyk M.: Electrochem. Commun. *7*, 1185 (2005).
2. Grabarczyk M., Korolczuk M.: Electroanalysis *18*, 70 (2006).

PP017
DEVELOPMENT OF AMPEROMETRIC PUTRESCINE BIOSENSOR FOR FOOD ANALYSIS

BEATA BOKA[a] NORA ADANYI[b], JENŐ SZAMOS[b], ATTILA KISS[a], and CSABA CSUTORAS[a]

[a] Department of Biochemistry, College of Károly Eszterházy, Eger, Leányka u. 6, 3300, Hungary, [b] Unit of analytics, Central Food Research Institute, Budapest, Herman Ottó u. 15, 1022, Hungary
bokab@ektf.hu

Biogenic amines are organic bases with low molecular weight and biological importance. Although they are essential for all living cells, consumption of food containing high amounts of them may have toxicological effects. The analysis of biogenic amine content of foodstuff gives information about the potential health risk and quality, since high level in non-fermented foods indicates undesired microbial activity during food processing and storage[1].

The aim of our work is to develop an enzyme based-amperometric biosensor applicable as food quality marker. This biosensor selectively measures putrescine which can be considered as an indicator of microbial spoilage. Putrescine oxidase (EC 1.4.3.10) was isolated from *Kocurea rosea* (*Micrococcus Rubens*) by an improved and simplified purification procedure[2] in our laboratory. Cells were grown on brain heart infusion medium supplemented with putrescine. Cell-free extract was prepared in pH 8.0 Tris buffer by Beadbeater and purified with three-phase partitioning (TPP), DEAE-cellulose chromatography and ultrafiltration steps.

Our putrescine biosensor works in flow injection analysis system (FIA) using a potentiostat (QuadStat 164, eDAQ, Australia). Evaluation of measured data was done with eDAQ e-corder A/D converter and eDAQ Chart software.

The enzyme was immobilized on the surface of a spectroscopic graphite electrode in redox hydrogel with horseradish peroxidase, Os mediator and poly(ethylene glycol) (400) diglycidyl ether (PEGDGE) as crosslinking agent[3,4]. This modified working electrode was used in wall-jet type amperometric cell together with the Ag/AgCl (0.1 M KCl) reference electrode and a platinum wire as counter electrode. The

effect of hydrogel composition, pH and potential dependence were studied, the range of linear response was determined and real food samples were analyzed.

This study was supported by EgerFood Regional Knowledge Center application 9/2005.

REFERENCES

1. Silla Santos M. H.: Int. J. Food Microbiol., 29213-231, (1996).
2. DeSa R. J.: J. Biol. Chem. *247*, 5527 (1972).
3. Bott A. W.: Cur. Separ. *21*, 3 (2004).
4. Ruzgas T.: Anal. Chim. Acta *1996*, 123.

PP018
PORTABLE ELECTROCHEMICAL URIC ACID MONITORING SYSTEM BASED ON SCREEN-PRINTED ELECTRODES

PABLO FANJUL BOLADO, DAVID HERNÁNDEZ SANTOS, and AGUSTÍN COSTA-GARCÍA*

Departamento de Química Física y Analítica, Universidad de Oviedo, Julián Clavería, 8, 33006, Oviedo, Asturias, Spain costa@fq.uniovi.es

Uric acid(UA, 7,9-dihydro-1*H*-purine-2,6,8(3*H*)-trione) is the major end product of purine metabolism. Purines from catabolism of dietary nucleic acid are converted to uric acid directly. However, the most of purines excreted as uric acid arises from degradation of endogenous nucleic acids. Its determination serves as a marker for the detection of disorders associated with purine metabolism such as gout arthritis, Lesch-Nyhan syndrome and renal retention. Also, hyperuricemia may indicate other diseases mainly related with cell degradation such as chemotherapy of malignant tumors as leukemias or lymphomas. Diagnosis of these clinical disorders are carried out by monitoring UA blood or urinary levels. The normal UA serum levels range from 3.5−7.2 mg dL^{-1} in men and from 2.6−6 mg dL^{-1} for women.

Methods in current use for measuring uric acid fall into two groups: phosphotungstic acid methods and uricase methods.

Phosphotungstic acid methods rely on the reduction of this acid by urate in alkaline medium to give a blue color in solution (tungsten blue) that is read spectrophotometrically at wavelengths of 650 to 700 nm.

Uricase methods are inherently more specific than colormetric methods. In this case urate oxidation is catalyzed by the enzyme uricase to allantoin and hydrogen peroxide. These methods became feasible and popular as a result of availability of high quality, low-cost preparations of the bacterial enzyme.

HPLC methods using reversed-phase columns have also been used to measure uric acid.

Some electrochemical methods based on enzymatic and non enzymatic approaches have been already reported for UA analysis[1,2]. Electrochemical activated surfaces, carbon nanotube and redox mediator modified electrodes can be used for the enhancement of the oxidation UA analytical signal.

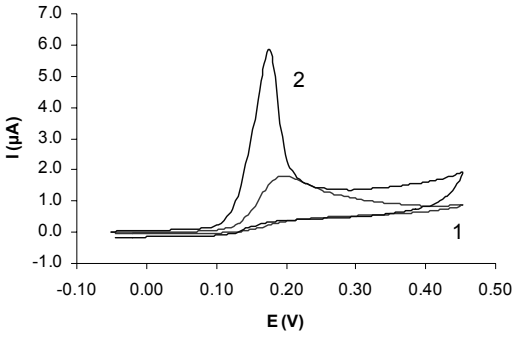

Fig. 1. **Cyclic voltammograms of uric acid 5×10^{-5} M in 0.1 M AcO- buffer solution pH 5,0; v=50 mV s^{-1}**. (1) Red line: screen-printed carbon electrode, (2) blue line: multi walled carbon nanotube modified screen printed electrode

A trend in the development of electrochemical sensors for decentralized analysis is the miniaturization of the analysis device and the instrumentation. Therefore in this work screen printed electrodes are evaluated for the detection of uric acid in combination with a hand held potentiostat.

Different carbon screen-printed electrodes are evaluated (Fig. 1) and biological fluids as urine and blood are analysed through voltammetric techniques. Furthermore, the proposed analysis system is compared with clinical serum analysis carried out by enzymatic method.

REFERENCES

1. Zen J. M., Jou J.-J., Hangovan G.: Analyst *123*, 1345 (1998).
2. Bravo R., Hsuch C., Jaramillo A., Brajter-Toth A.: Analyst *123*, 1625 (1998).

PP019
DOUBLE-TAGGING PCR AND GOLD NANOPARTICLES USED IN IMPEDIMETRIC GENOSENSING

ALESSANDRA BONANNI, MARIA I. PIVIDORI, SALVADOR ALEGRET, and MANEL DEL VALLE*

Sensors and Biosensors Group, Department of Chemistry, Universitat Autonoma de Barcelona, 08193 Bellaterra, Spain manel.delvalle@uab.cat

Real sample DNA analysis is proposed employing an electrochemical impedimetric genosensor, previously developed in our laboratories[1], applied to double stranded DNA sequences produced by the polymerase chain reaction (PCR) amplification.

Electrochemical Impedance Spectroscopy is a rapid developing technique for the transduction of biosensing events at the surface of an electrode[2]. Transduction principle exploits changes in interfacial resistance of charge transfer after modification of the genosensing transducer with DNA[3].

In the present work, an avidin bulk-modified graphite-

epoxy biocomposite (Av-GEB) was employed to immobilize –onto the electrode surface– the double-tagged DNA, modified in each end with biotin and digoxigenin, respectively. Impedance spectra were recorded to detect the change in interfacial charge transfer resistance (R_{ct}) as observed from the ferri/ferrocyanide redox marker reaction. A further step in the genosensing procedure was the amplification of impedimetric signal by the use of gold nanoparticles modified with Anti-Mouse IgG (whole molecule)–Gold antibody. The latter were immobilized to the digoxigenin-modified end of the amplicon by a monoclonal IgG1kappa anti-Digoxigenin antibody from mouse.

Results obtained by the comparison of Rct values after each further modification of the electrode surface show a significant difference in the impedimetric signal variation between experiments and negative controls. Moreover, this difference results thoroughly amplified thanks to the procedure used for signal enhancement.

The sensitivity of the technique and the good reproducibility of results confirm the validity of this method based on a universal affinity biocomposite platform coupled with the impedimetric technique. The described strategy was used for the rapid and sensitive detection of PCR amplified samples of *Salmonella spp*, the most important pathogen affecting food safety.

REFERENCES

1. Bonanni A., Pividori M. I., del Valle M.: Anal. Bioanal. Chem. *389*, 851 (2007).
2. Berggren C., Stålhandske P., Brundell J., Johansson G.: Electroanalysis *11*, 156 (1999).
3. Katz E., Willner I.: Electroanalysis *15*, 927 (2003).

PP020
ELECTROCHEMICAL METHOD FOR EVALUATION OF OXIDANT/ANTIOXIDANT ACTIVITY OF BIOLOGICAL FLUIDS

BRAININA KH. Z.[a,b], GERASIMOVA E. L.[a,b], and MOROZOVA O. B.[a]

[a] Institute of Medical Cell Technologies, Ekaterinburg, [b] Ural State University of Economy, Ekaterinburg

Free-radical oxidation, on the one hand, ensures the normal progress of metabolism processes in the organism, but, on the other hand, the disturbance of the oxidant/antioxidant balance leads to the oxidation stress, which is a disbalance between the generation of active forms of oxygen and the antioxidant protection of the organism. Surplus active forms of oxygen can destroy lipids of cells, proteins or DNA, inhibiting the normal functioning of the organism.

The total antioxidant activity of a sample should be considered as an integral parameter. We have proposed a method involving the use of a mediator system and measurements of the potential across a screen-printed platinum sensor as the source of information about the antioxidant/oxidant activity of test materials.

The proposed potentiometric method for measurement of the antioxidant activity is a promising alternative choice against the available methods. The subjects of study were seminal and follicular fluids.

The optimal concentrations of the oxidized and reduced forms of the mediator system were determined for each of the aforementioned test fluids. The sampling and sample handling methods were selected. It was shown that the devised potentiometric method is highly informative and provides reliable results.

The proposed method has the advantages of simplicity and a low cost. The obtained data suggest that the aforementioned parameter of the antioxidant activity can be used as a criterion for evaluation of the general antioxidant activity of the seminal and follicular fluids and serve as an index pointing to the condition of the reproductive function in humans.

The authors are very grateful to ISTC and RFBR for their financial support (ISTC project 2132; RFBR project 07-03-96071-p_ural_a).

PP021
AMPEROMETRIC GLUCOSE BIOSENSOR BASED ON ELECTROPOLYMERIZED CARBON NANOTUBE/ POLYPYRROLE COMPOSITE FILM

VIOREL BRÂNZOI[a]*, LUISA PILAN[a], FLORINA BRÂNZOI[b], and FLORENTINA GOLGOVICI[a]

[a] University Politehnica of Bucharest, 132 Calea Griviţei, district 1, 010737,Bucharest, Romania, [b] Institute of Physical Chemistry, 202 Splaiul Independenţei, 060021, district 6, Bucharest, Romania
iv_branzoi@chim.upb.ro

An important work in the design and fabrication of biosensors is to find an effective immobilization method, an appropriate support material and a fast and simple procedure. Thus, we report a one-step preparation route of amperometric enzyme electrodes based on incorporating a carbon-nanotube (CNT) dopant and the enzyme within an electropolymerized polypyrrole (PPY) film.

Because electropolymerization represents an attractive wellcontrolled one-step procedure for preparing amperometric enzyme electrodes[1,2], the nanocomposite films were electrochemically synthesized by electrooxidation of pyrrole in an aqueous solution containing appropriate amounts of GOD and CNTs. The main advantages of this immobilization way are the simple one-step preparation, exclusion of electroactive interferences, control of the film thickness, and localization of enzymes onto electrode surfaces[3–5]. The electropolymerization was carried out at a pH above the isoelectic point of the protein in order to provide a negative charge. The relevant parameters of the film preparation were examined and optimized. The amperometric detection of glucose was assayed by potentiostating the enzyme electrode at a potential to reduce the enzymatically produced H_2O_2 with minimal interference from the coexisting electroactive compounds. The results indicate that the PPY-CNTs-GOD nanobiocomposite film is highly sensitive and suitable for glucose biosensor function.

Financial support from CEEX ET 67/2006 contract

(Romanian Ministry of Education and Research) is gratefully acknowledged.

REFERENCES
1. Emr S. A., Yacynych A. M.: Electroanalysis *7*, 913 (1995).
2. Trojanowicz M., Krawczynski T.: Mikrochim. Acta *121*, 167 (1995).
3. Zhang S., Wright G., Yang Y.: Biosens. Bioelectron. *15*, 273 (2000).
4. Wu X. J., Chio M. M. F.: Anal. Chem. *76*, 4279 (2004).
5. Puig-Lleixa C., Jimenez C., Bartroli J.: Sens. Actuators, B *72*, 56 (2001).

PP022
MODIFIED ELECTRODES OBTAINED BY ELECTRO-DEPOSITION OF POLYMERIC AND COMPOSITE FILMS ON WOLFRAM SURFACE ELECTRODE

VIOREL BRANZOI[a], FLORENTINA GOLGOVICI[a]*, FLORINA BRANZOI[b], and LUISA PILAN[a]

[a] University Politehnica of Bucharest, Calea Grivitei, 132, 010737, Bucharest, Romania, [b] nstitute of Physical Chemistry, Spl. Independentei, 202, 77208, Bucharest, Romania
floregov@yahoo.com

Polypyrrole, polyaniline films and polypyrrole-polyaniline composite films were obtained by electrochemical oxidation of the monomers: pyrrole, aniline and pyrrole-aniline mixture of different compositions in aqueous solutions containing different dopant anions as supporting electrolyte.

Cyclic voltammetry and electrochemical impedance spectroscopy (EIS) were used to investigate the electrochemical properties of modified electrodes. The results indicate that the surfactant anions favour redox processes which are faster and more reversible than those associated to the usual polymeric electrodes. In fact, while in polymeric films doped by conventional, small anions (e.g. Cl^-), the charge compensation is assured by uptake of the anions during oxidation and the release of the same anions during reduction, in the polymeric films prepared in the presence of the large–anions salts (e.g. SDS, AOT) the charge compensation appears to involve incorporation of cations since the large anions are not easily released from the polymer matrix.

Multilayered coatings, consisting of combinations of the conducting polymers polyaniline (PANI) and polypyrrole (PPY) were deposited onto wolfram by different electrochemical techniques, such as potentiodynamic, galvanostatic and potentiostatic modes. The overall system has a bilayered structure, indicating that the electrochemical interface during polymers electrodeposition might be situated at the polypyrrole-solution interface in case of PANI/PPY wolfram electrode, respective the polyaniline-solution one in the case of PPY/PANI wolfram electrode. The presence of the surfactant (SDS or AOT) makes easier the pyrrole and aniline electropolymerization. Indeed, the presence of micelles decreases the monomer oxidation potential and induces an acceleration of the polymerization. In the used electrosinthesys solution pyrrole and aniline have approximative the same monomer oxidation potentials. Therefore, an electropolymerization mixture of these monomers was carried out with different ratios in order to prepare copolymers and the resulting products were characterized by electrochemistry, IR and IV-visible spectroscopy and SEM.

REFERENCES
1. Akundy G. S., Rajagopalan R., Iroh J. O.: J. Appl. Polym. Sci. *83*, 1970 (2002).
2. Branzoi V., Pilan L., Golgovici F., Branzoi F.: Mol. Cryst. Liq. Cryst. *446*, 305 (2006).

PP023
NANOCOMPOSITE FILMS OBTAINED BY ELECTROCHEMICAL CO-DEPSITION OF CONDUCTING POLYMERS AND CARBON NANOTUBES

VIOREL BRÂNZOI[a], LUISA PILAN[a]*, and FLORINA BRÂNZOI[b]

[a] University Politehnica of Bucharest, 132 Calea Griviței, district 1, 010737,Bucharest, Romania, [b] Institute of Physical Chemistry, 202 Splaiul Independenței, 060021, district 6, Bucharest, Romania
luisa_pilan@yahoo.com

The electrochemical growth of carbon nanotubes (CNTs) – conducting polymer composites offers the ability to produce tree-dimensional nanostructured films that combine the redox pseudo-capacitive charge storage mechanism of conducting polymers with the high surface area and conductivity of CNTs[1–6]. Nanocomposite films of CNTs and either polyaniline (PANI) or polypyrrole (PPY) were prepared using electrochemical polymerization technique in which nanotubes and conducting polymer were deposited simultaneously from solutions containing acid treated CNTs and the corresponding monomer. The electropolymerization process was carried out by galvanostatic method. The concentration and dispersion of CNTs in polymerization electrolyte, in addition to any supporting electrolyte used, were found to have a significant effect on thickness of the composite film and CNTs loading in the produced films. Electrochemical impedance spectroscopy (EIS), cyclic voltammetry and square wave voltammetry were used to investigate the electrochemical properties of the composite films. The electrochemical response, both in cyclic voltammetry and square wave voltammetry is significantly increased for the composite films in comparison with the respective polymers alone. The EIS on CNTs/PPY and CNTs/PANI composite electrodes were carried out in the supporting electrolyte in dependence of the following parameters: film thickness, supporting electrolyte concentration, temperature and polarisation potential.

REFERENCES
1. Hughes M., Chen G. Z., Shaffer M. S. P., Fray D. J., Windle A. H.: Chem. Mater. *14*, 1610 (2002).
2. Hughes M., Shaffer M. S. P., Renouf A. C., Singh C., Chen G. Z., Fray J.: Adv. Mater. *14*, 382 (2002).
3. Hughes M., Chen G. Z., Shaffer M. S. P., Fray D. J., Windle A. H.: Comp. Sci. Techn. *64*, 2325 (2004).

4. Chen G. Z., Shaffer M. S. P., Coleby D., Dixon G., Zhou W., Fray D. J., Windle A. H.: Adv. Mater. *12*, 522, (2000).
5. Wu M., Snook G. A., Gupta V., Shaffer M., Fray D. J., Chen G. Z.: J. Mater. Chem. *15*, 2297 (2005).
6. Peng C., Jin J., Chen G. Z.: Electrochim. Acta *53*, 525 (2007).

PP024
DETERMINATION OF THEOPHYLLINE AT A GLASSY CARBON ELECTRODE MODIFIED BY ELECTRODEPOSITED CYSTEIC ACID

BARBARA BRUNETTI* and ELIO DESIMONI

DiSTAM, University of Milan, Via Celoria 2, I-20133 Milano, Italy
barbara.brunetti@unimi.it

It was recently shown that glassy carbon electrodes (GCE) modified with cysteic acid can be successfully used in the determination of nimesulide[1] and other analytes of pharmaceutical interest[2] thanks to their electrostatic accumulation onto the modified electrode surface. Cysteic acid was deposited onto the GCE surface by electrochemical oxidation of cysteine by mean of cyclic voltammetry.

In this work, such modified GCE are used for determining methylxanthines, with particular attention to theophylline. According to several Authors, common GCE don't allow acquiring sufficiently reproducible results in the determination of these analytes. This because their oxidation occurs at very positive potentials, overlapping with the discharge of the background medium[3]. Several solutions were proposed, including the use of electrodes with a wider anodic potential range[3] or modified electrodes[4], or unusual combination of solvent/supporting electrolyte[5]. Cysteic acid modified electrodes appear to be a straightforward solution to the problem, thanks to their affinity to the above mentioned analytes, the low cost, the simplicity of the modification procedure and the reproducibility of the electrode surface.

Preliminary cyclic voltammetric experiments were made to evaluate the influence of the supporting electrolyte and of the electrodeposition parameters (cysteine concentration, scan rate, potentials, etc).

Differential pulse voltammetry experiments allowed to estimate the linear range, the calibration function and the limit of detection. The results confirmed the potentiality of the modified electrode, characterized by higher sensitivity, selectivity and reproducibility in comparison to unmodified GCE.

The possible interferences when analyzing real samples were also considered. The results are discussed in the light of the most recent literature information.

REFERENCES

1. Wang C. Y., Shao X., Qingxiu L., Qu Q., Yang G., Hu X.: J. Pharm. Biomed. Anal. *42*, 237 (2006).
2. Wang C. Y., Wang Z. X., Guan J., Hu X. Y.: Sensors *6*, 1139 (2006).
3. Spataru N., Sarada B. V., Tryk D. A., Fujishima A.: Electroanalysis *14*, 721 (2002).
4. Zen J.-M., Ting Y.-S., Shih Y.: Analyst *123*, 1145 (1998).
5. Lau O.-W., Luk S.-F., Cheung Y.-M.: Analyst *114*, 1047 (1989).

PP025
IMPROVEMENT OF STORAGE STABILITY FOR AMPEROMETRIC BIOSENSORS BASED ON ALCOHOL OXIDASE IMMOBILIZED BY ENTRAPMENT IN A PHOTOPOLYMERISABLE MATRIX

BOGDAN BUCUR[a]*, MADALINA PETRUTA BUCUR[b], and GABRIEL LUCIAN RADU[a,b]

[a] National Institute of Biological Sciences, Centre of Bioanalysis, 296, Splaiul Independentei, Bucharest, 060031, Romania, [b] Politehnica University of Bucharest, Faculty of Applied Chemistry and Materials Science, 1-7, Polizu, Bucharest, 011061, Romania
bucurica@yahoo.com

One important impediment for successful commercial application of enzymatic biosensors is their limited storage stability caused to the instability of the used biocomponent As model enzyme was used alcohol oxidase (AOX), a model enzyme with a poor storage stability. The detection was made by cronoamperometry in a drop of solution and the transducers were screen-printed electrodes modified with mediator Co-phthalocyanine. The enzyme was immobilized onto screen-printed electrodes by entrapment in a photopolymerisable PVA matrix, a method that already provides a stabilizing effect.

The storage stability of immobilized AOX was improved by the use of three different stabilizers: sorbitol, Tween 20 and PEG 6000 and compared with the enzyme immobilized without stabilizers.. All stabilizers have a positive effect on the enzyme stability. The increase of storage time in comparison with biosensors without stabilizers was by 25 % for Tween 20 while the sorbitol allow a double storage time of the biosensors. The most appropriate stabilizer proved to be PEG 6000 that allowed the increase of the storage time of the biosensors by more than 3 times.

This work was supported by Romanian Ministry of Education and Research through Grant CEEX-156/2006 (BAME).

REFERENCES

1. Bucur B., Radu G. L., Toader C. N.: Eur. Food Res. Technol. in press.
2. Bucur B., Mallat E., Gurban A. M., Gocheva Y., Velasco C., Marty J. L., Noguer T.: Biosens. Bioelectron. *21*, 2290 (2006).
3. Bucur B., Bucur M. P., Radu G. L.: Rev. Chim *58*, 1145 (2007).