Sensors and Actuators B 171–172 (2012) 155–164

Contents lists available at SciVerse ScienceDirect

Sensors and Actuators B: Chemical

journal homepage: www.elsevier.com/locate/snb

Thin film reference electrodes for aqueous and organic media

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ARTICLE INFO

Article history:
Received 31 October 2011
Received in revised form 21 February 2012
Accepted 22 February 2012
Available online 6 March 2012

Keywords:
Thin-film electrode
Reference electrode
Cyclic voltammetry
Galvanic metal deposition
Biosensor

ABSTRACT

The integration of reference electrodes into electrochemical (sensoric) systems is essential for the stabilization of the potentials and for the determination of reproducible potentials. For many micro-sensoric applications the 'classical' reference electrodes that are based on Ag/AgCl or Hg/Hg_2Cl_2 do not perform. In this work, reference electrodes based on six alternative metals (Pb, Zn, Sn, Fe, Bi, and Cu) were tested alone or in combination with their respective, sparingly soluble salts (forming systems of the second kind). The performance and the stability in aqueous and non-aqueous model systems were investigated. For all systems, at least one suitable reference electrode system could be identified. These systems were implemented in micro-electrode setups; excellent performances were demonstrated with two biochemically relevant redox-systems, vitamin C and vitamin K_3.

1. Introduction

The goal for miniaturization of sensing and actuating devices is driven by the need for an efficient use of energy and material resources. New technologies or new combinations of existing technologies are typically used to build smaller devices that are mobile and cost less in manufacturing and operation. (Bio)chemical sensors, which detect the presence of one or more species and their respective concentrations, are an area of current interest. Among the different principles of chemical sensing, electrochemical sensors have become particularly useful, because the chemical signal is directly converted into an electrical signal, eliminating the need for transduction. Electrochemical sensors typically deliver a current, but if this current is determined as a function of a certain potential, selectivity can be gained without any further selecting principle. This is the case for cyclic voltammetric methods, which can determine the concentration of different redox-active species within the same solution. While the typical laboratory setup uses electrodes that are the size of pens and requires analyte volumes of several tens of milliliters, it is of broad interest to miniaturize this versatile method for integration into mobile applications. Several reports on such miniaturized systems have been published [1–4]. In addition to the obvious advantages of reducing the size, the use of small electrodes also bears other positive aspects because mass transport rates generally increase with a decreasing electrode size [5,6]. In addition, the reduction of charging currents and Ohmic losses result in low signal/noise ratios, permitting the detection of currents as low as $10\,e^-/s$ (equals 10^{-18} A) [6].

Therefore, these systems are very suitable for electroanalytical methods, such as voltammetry [7–18], amperometry [7], potentiometry [19,20], field-effect measurements [21], stripping analysis [22], or impedance spectroscopy [23]. These methods have been used in physiological [24–26] and biological [27–30] research, pH measurements [31–33], kinetics [34], electrophoresis [12,14], and often in combination with microfluidic devices [17–20,35]. In these applications, the microelectrodes are often fabricated from thin metal wires that are embedded in glass or plastic sealing [8–10,31,32,36]. In contrast, the deposition of the electrodes onto surfaces makes them more robust and easy to use, and thin film electrodes are suitable for miniaturization and integration. Many techniques used to fabricate thin-film electrodes have been established including the following: chemical vapor deposition (CVD) [37,38], physical vapor deposition (PVD, in combination with lift-off techniques) [39], sol–gel deposition [40,41], pulsed-laser deposition [42], radio-frequency (RF) sputtering [43], spin coating [44], electrospray deposition [45], ink-jet printing [46], powder pressing [47], and screen-printing [13,16,48–50]. Some techniques require expensive equipment, use complicated procedures, or have inherent difficulties to control the surface properties of the electrodes, hampering their practical application. An easy and inexpensive method for the selective and controlled manufacturing of thin film metal electrodes is galvanic metal deposition. Thickness and appearance of the resulting films can be adjusted by the current and by certain additives.

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doi:10.1016/j.snb.2012.02.071

Table 1
Electrochemical deposition parameters for the films used in this project.

Metal	Bath composition	Metal deposition conditions[a]	Electrooxidation (1 M aq. H_3PO_4)[a]	Electrooxidation (sat. $TBAPF_6$ in DMF)[a]	Electrooxidation (1 M aq. HBF_4)[a]
Zinc	$ZnSO_4 \cdot 7H_2O$ (360 g), $AlCl_3 \cdot 6H_2O$ (20 g), Na_2SO_4 (75 g), H_2O (1 dm^3)	28 °C, −0.4 V, Zn, 60 s	23 °C, +0.5 V, Pt, 2 s	23 °C, +2.0 V, Pt, 60 s	23 °C, +0.1 V, Pt, 2 s
Tin	$Na_2[Sn(OH)_6]$ (8.7 g), NaOH (9.6 g), $NaOAc \cdot 3H_2O$ (14.4 g), gelatin[d] (1.5 g), H_2O (1 dm^3)	60 °C, −2.7 V, Pt, 90 s[b]	23 °C, +2.0 V, Pt, 3 s	23 °C, +2.0 V, Pt, 120 s	23 °C, +0.1 V, Pt, 2 s
Lead	$2PbCO_3 \cdot Pb(OH)_2$ (29.6 g), $B(OH)_3$ (5.5 g), HBF_4 (aq., 50%, 77 cm^3), gelatin[d] (0.22 g), H_2O (1 dm^3)	23 °C, −0.1 V, Pb, 60 s[c]	23 °C, +1.0 V, Pt, 2 s	23 °C, +0.5 V, Pt, 5 s	23 °C, +0.3 V, Pt, 5 s
Iron	$(NH_4)_2Fe(SO_4)_2 \cdot 6H_2O$ (300 g), B_2O_3 (1.0 g), KCl (10 g), SDS (0.3 g), H_2O (1 dm^3)	23 °C, −1.0 V, Fe, 60 s	23 °C, +1.0 V, Pt, 5 s	Not performed	Not performed
Copper	$CuSO_4 \cdot 5H_2O$ (84.4 g), H_2SO_4 (42 cm^3), H_2O (1 dm^3)	40 °C, −2.0 V, Pt, 180 s	23 °C, +1.5 V, Pt, 3 s	23 °C, +2.0 V, Pt, 120 s	23 °C, +1.5 V, Pt, 5 s
Bismuth	$BiCl_3$ (30 g), NaCl (10 g), HCl (aq., 37%, 200 cm^3), gelatin[d] (2.0 g), H_2O (1 dm^3)	23 °C, −0.1 V, Bi, 30 s	23 °C, +2.8 V, Pt, 5 s	23 °C, +2.8 V, Pt, 5 s	23 °C, +0.5 V, Pt, 10 s

[a] The values are deposition temperature, potential, counter electrode, and deposition time.
[b] Vigorous stirring was necessary to obtain glossy layers.
[c] The gold electrode was treated by air plasma at 0.4 mbar before plating.
[d] When gelatin was used, the bath was freshly prepared and heated to 100 °C before use.

A special challenge for the fabrication is the reference electrode, which is needed to warrant a constant potential as a point of reference in the electrochemical system. The use of a pseudo reference electrode, such as one made of the same (noble) metal as the working and counter electrodes, is not an alternative, because the potential is shifting unpredictably (Fig. S1, supporting information). In many setups with miniaturized working and/or counter electrodes, macroscopic reference electrodes are used for simplicity [8–11,14,15,32,36]. Typically, commercially available second-order reference electrodes are based on the Ag/AgCl or the Hg/Hg_2Cl_2 (calomel) system, of which only the Ag/AgCl reference electrode is suitable for miniaturization [51–53] because of the difficulties in handling and dispersing mercury in the micrometer scale. A general characteristic of the second-order electrode system is the presence of the anion of the redox pair in the vicinity of the electrode, which through the solubility product of the poorly soluble salt provides the constant electrochemical parameters, in particular the concentration of the cation [54]. In macroscopic electrodes, the anion solution and the electrode are maintained in a separate compartment where intermixing with the analyte is suppressed by the use of salt bridges. Although this approach also has been tried for miniaturized reference electrodes, the fabrication is quite tedious, and the separation is not complete [33,51]. A typical solution to this dilemma is the addition of the anion to the analyte solution, which is often feasible by using compounds carrying this anion as conducting salts. Unfortunately, chloride, the anion for the reference systems mentioned above (Ag/AgCl, Hg/Hg_2Cl_2), often cannot be used because it is electrochemically active. This limits the accessible potential range and often leads to corrosion of the working and counter electrodes, for example by the formation of $[AuCl_4]^-$ or $[PtCl_4]^{2-}$.

We therefore set out to investigate alternative reference electrode systems and focused on counter anions, which should be electrochemically inert and form insoluble salts at the reference electrode, turning it into a second-order system. The criteria for these reference electrodes include a constant potential from the first cycle (or at least after a short equilibration period in form of a small number of cycles) and the stability over several hundred cycles, which often becomes necessary even for short-term (one-way) applications. It is noted that at this time, the exact chemistry behind the success or failure of each of the electrode systems could not be investigated in detail because the identification of *reliable* systems was the primary goal of this project. Therefore, the fabrication and stability of different macroscopic electrodes in specific reference solutions are presented, followed by the observations upon miniaturization and the application of the optimized systems for the cyclic voltammetry of two biochemically relevant molecules.

2. Materials and methods

2.1. Chemicals

Millipore water with a resistivity greater than 18.0 MΩ cm was used. Since commercial dimethylformamide (DMF) always contains traces of amines that influence the potentials of metal ions by coordination [55], the analytical grade DMF purchased from Sigma Aldrich was distilled in vacuo following the protocol given in Ref. [56] with a significant pre-run (30–40%) being discarded. The purified solvent then was stored under the exclusion of light and humidity. All other chemicals were used as purchased.

2.2. Metal deposition and electro-oxidation

Galvanic metal deposition and electro-oxidation were carried out using a Voltcraft PLUS VSP 1410HE laboratory power supply. The metals were deposited on gold films (80 nm) on glass (with 2 nm of chromium as adhesion promoter) with dimensions of approximately 26 mm × 10 mm. Bath compositions and parameters for electrolysis are given in Table 1 [57]. The substrate-counter electrode distance was 1.5 cm, and the baths were moderately stirred. Deposited metal layers were washed with water and ethanol and dried in a stream of nitrogen. The respective metal salt layers obtained by electro-oxidation of the freshly deposited metal films (Table 1) were dried carefully in a stream of nitrogen.

2.3. Cyclic voltammetry

All cyclic voltammetry experiments were performed with a Compact Stat potentiostat (Ivium Technologies B.V., Eindhoven, Netherlands) in either phosphate buffer (0.1 mol L^{-1}, pH 7) or DMF with the respective conducting salt (0.1 mol L^{-1}). Counter and working electrodes were gold films that were similar to the substrates for the reference-electrode fabrication.

2.4. SEM images

SEM images were taken with a Leo Type 1525 FE microscope. The electron energy used was 5 keV. Samples were fixed onto glass and coated with carbon before the measurements were taken.

2.5. AFM images

AFM measurements were performed with a Solver-Pro from NT-MDT under ambient conditions. The measuring mode was semi-contact from 0.5 to 2 Hz scanning speed using an Au-coated CSG01 cantilever.