

CATHETER BASED BIOSENSOR: ASSEMBLING AND CHARACTERIZATION

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ABSTRACT

The development of a catheter based biosensor for real time and *in situ* monitoring of biological signals is described. Our main application is the detection of paracetamol, a well-known chemical for pain and fever relieve. The biosensor is based on the electrochemical reaction of this radical at the surface of the detection electrode made of gold, using silver chloride (Ag/AgCl) as reference electrode and a platinum auxiliary electrode. All electrodes were made by wires ($\Phi=200\ \mu\text{m}$) and assembled on to catheter ($\Phi_c=1.34\ \text{mm}$). Amperometric technique was used for paracetamol detection and cyclic voltammetry for gold electrode characterization, mainly its area, which is important to determine the sensibility and to assert the reproducibility of the sensor. Both techniques were performed by using a CV-50W Bionalytical System (BAS) electrochemical workstation. The sensor was able to detect concentration as low as $0.17\ \text{mmol.L}^{-1}$ of paracetamol which is the usual value found in the blood. We applied this sensor to obtain the amount of active principle in tablets of Tylenol[®].

1. INTRODUCTION

Electrochemical sensors have had a tremendous development in many different applications^[1,2] from pH determination, gas sensing, and analysis of complex biological molecules. Catheter based electrochemical sensors present many advantages such as^[3,4]: small size therefore less invasive, possibility of *in situ* and real-time measurements of many important biochemical compounds such as paracetamol, a well-known chemical for pain and fever relieve and active principle in tablets of Tylenol[®] found at any drugstore shelf. The fabrication and characterization of the sensor is described as well as its calibration curve for paracetamol.

2. ELECTRODES IN CATHETER

The standard electrochemical detection is based on three electrodes system: 1) the working or detection electrode (gold), where the chemical reactions are performed, 2) reference electrode, a stable and reference potential inside of the sensor's environment (modified silver to obtain silver chloride film) and finally 3) auxiliary or counter electrode, which closes the electric circuit path in solution (platinum). All electrodes were made by wires ($\Phi=200\ \mu\text{m}$, 99% pure) and assembled on to catheter ($\Phi_c=1.34\ \text{mm}$). It was used a P2 audio cable (figure 1), to connect the sensor to a CV-50W Bionalytical System (BAS) electrochemical workstation.

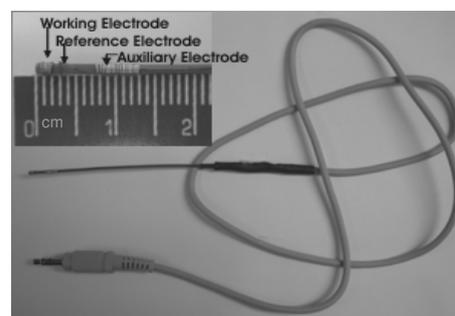


Fig. 1: Complete scheme of biosensor and electrodes in detail.

3. ELECTROCHEMICAL CLEANING PROCESS

After the standard cleaning, we performed an electrochemical cleaning process that provides an extra increasing of the area of the working electrode^[5], which is proportional to the sensor's response in amperometric detection. Using cyclic voltammetry technique, we swept ten consecutive cycles of the potential applied to the electrode from $-900\ \text{mV}$ to $+900\ \text{mV}$ at $300\ \text{mV/s}$ scanning rate in standard solution of $0.1\ \text{mmol.L}^{-1}$ potassium ferricyanide - $\text{K}_3[\text{Fe}(\text{CN})_6]$, $0.5\ \text{mmol.L}^{-1}$ KCl. we observed in the seventh cycle that the signal became stable with an increase of 30.8% in relation to the initially m

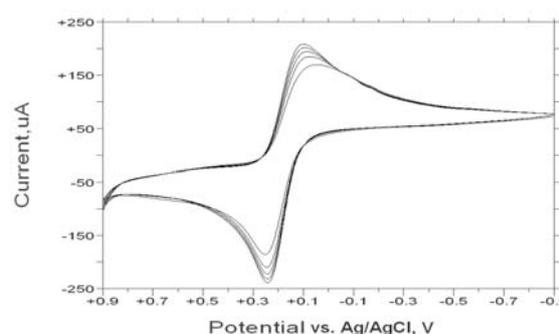


Fig. 2: Standard response in electrochemical cleaning process.

4. ELECTRODES CHARACTERIZATION

Measuring the effective exposed area of the working electrode permits to determine the sensibility and to assure the biosensor reproducibility. Also, in electrochemical sensors, it is essential to characterize the reversibility of the reference electrode to guarantee trustful measurements^[6]. It was chosen the Ag/AgCl reference electrode obtained by chlorine electrodeposition over a silver surface in $0.1\ \text{mmol.L}^{-1}$ HCl solution – to obtain silver chloride^[7]. The potential difference between a commercial reference electrode and the fabricated one was $4\ \text{mV}$, in $3\ \text{mmol.L}^{-1}$ NaCl solution.

Reversibility tests with the produced reference electrode in relation of the commercial reference one, presented the expected linear^[8] behavior.

5. BIOMEDICAL APPLICATION

Through the amperometric analysis technique (Single Potential Time Base - TB) with constant potential equal to 700 mV it was obtained a calibration curve by adding consecutively ten injections of 50 mmol.L⁻¹ paracetamol solution, 17 µl each, into 5 mL of physiological solution (that is isotonic with blood plasma) up to a concentration of 0.17 mmol.L⁻¹ (figure 4).

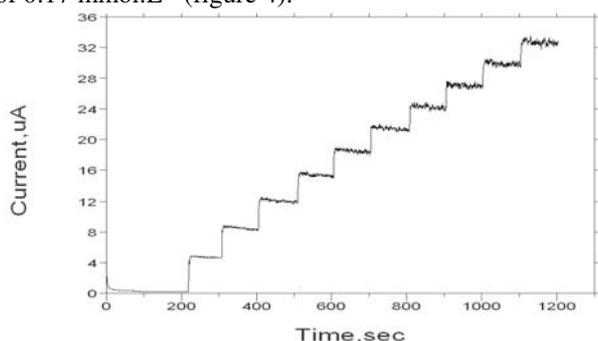


Fig. 4: Sensor's response for 0.17 mmol.L⁻¹ of paracetamol injections.

In the calibration curve (figure 5) one can observe a slight deviation from the straight line for low concentrations.

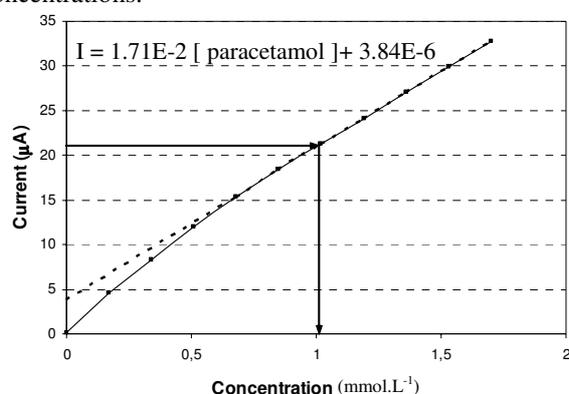


Fig. 5: Calibration curve for paracetamol.

6. RESULTS AND DISCUSSION

We applied the fabricated sensor to determine paracetamol in pharmaceuticals formulations. A tablet of Tylenol[®] 750 mg, which presented a net weight of 841.8 mg, was fragmented in 1/10 of the total mass (84.18 mg) and dissolved in 10 mL of physiological solution. An injection of 100 µL of this solution in 5 mL of physiological serum (dilution 1:50) produced a sensors response that is shown in figure 6. The observed steady current of 21.3 µA corresponds to a paracetamol concentration of 1.02 mmol.L⁻¹ according to the calibration curve (dotted line in figure 5), i.e., 772 mg in a tablet (considering MW = 151.2 g.mol⁻¹), which means an error of 2.9%. The observed error is comparable to others techniques such as Raman Spectroscopy^[9] and Spectrophotometric determination^[10].

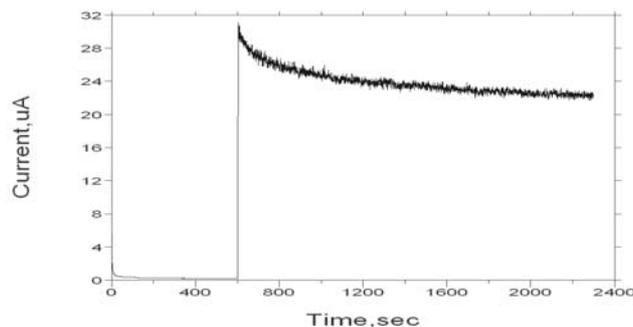


Fig. 6: 100 µL injection of paracetamol for measure of mass.

7. CONCLUSION

The assembling, characterization and paracetamol detection of a catheter based microwired electrochemical biosensor was presented. The cleaning procedure allowed increasing the sensors sensitivity by 30.8%, enabling to measure a concentration of paracetamol in a physiological solution of 0.17 mmol.L⁻¹. The cleaning procedure allowed increasing the sensors sensitivity by 30.8%, enabling to measure a concentration of paracetamol in a physiological solution of 0.17 mmol.L⁻¹. The amount of paracetamol in a tablet of Tylenol[®] 750 mg was determined to be 772 mg, which correspond to an error of 2.9%, that is comparable to others analysis techniques.

The presented results showed the efficiency of the developed biosensor to detect paracetamol electrochemically and we are looking forward to apply it in other bioenvironmental applications such as detection of heavy metals, ascorbic acid, uric acid, nitrate and glucose.

8. REFERENCE

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