

**Title: Development of a multiplexed amperometric immunosensor for detection of antibiotics in milk**

**Student:** Alejandro Gracia Gil

**Date:** JUNE 2019

**Supervisor/s:** Dra. Mónica Martínez López

*Departament de Ciència de Materials i Química Física*

Dr. J. Pablo Salvador, Nanobiotechnology for Diagnostics group (Nb4D),  
CIBER-BBN, Nb4D, IQAC-CSIC

Involuntary human exposure to antibiotics can cause bacterial resistance to them, making treatment of common and simple diseases a harder issue and strengthening bacteria which cause more serious illnesses. One of the main potential sources of those antibiotics into the human body is milk. Therefore, the EU has set limits to the presence of antibiotics in this hugely consumed food.

Quantitative analysis of these molecules has been possible for a while now, but most of them imply chromatographic techniques. Even though these methods reach very good limits of detection, they require high money, time and personnel investment. These factors make them unable to be a part of quotidian analysis that may be performed in hospitals, first aid centers or even at home by civilians.

Because of that, a multiplexed amperometric immunosensor based on immunoassays was developed. Polyclonal specific antibodies working under competitive indirect format make the chemical base of this assay and the detection protocol. The transduction of the chemical signal came through an amperometric detection technique using gold screen-printed electrodes previously functionalized.

The results reached are going to be presented as the  $IC_{50}$  values for the calibration curves in milk/PBST dilution 1:10, as well as their standard deviations. Tylosin B detectability value does not correspond to the requirements, so the determination of this antibiotic in milk may need a change in the procedure.

Table I: Mean and SD for the  $IC_{50}$  in milk dilution 1:10 in PBST

$IC_{50}$ ( $\mu\text{g/L}$ )	<b>TYL B</b>	<b>SPYR</b>	<b>CPX</b>	<b>CAP</b>
<b>MEAN</b>	1430	4.100	9.01	6.30
<b>SD</b>	293.4	0.2687	1.103	0.2404

**Keywords:** Antibiotic, antibody, immunosensor, amperometric, ELISA, milk