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Detection of Staphylococcus Aureus by Amoxicillin Modified Natural Phosphate Electrode: Analytical Application Potato Juice

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Abstract- The electrochemical detection of staphylococcus aureus bacteria by the amoxicillin modified natural phosphate (AMX-Np) is decied. The AMX-NP electrodes were used for the detection of low optical densities of staphylococcus aureus by using the cyclic voltammetry (cv) and the square waves voltammetry (swv). Some electrochemical properties, in particular the influence of the pH, the optical density of the bacterium were studied. The elaborate electrode was the subject then of an analytical application for purposes of the detection of staphylococcus aureus in the potato juice.

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I. INTRODUCTION

The gilded staphylococcus(staphylococcus aureus) is the stock of staphylococcus most frequently met in pathology human and veterinary. It shares with the bacterium Escherichia coli the unhappy privilege to be in the forefront of the germs responsible for infection nosocomial (infection contracted at the hospital) [1]. The staphylococcus aureus is pathogenic opportunist which can cause various diseases at the human ones, energy of the affections which evolve spontaneously to the cure with pathologies mortals [2].The food poisoning by the staphylococcus is characterized by a brutal appearance of nauseas, vomiting, abdominal pains, cramps and of diarrhea [2,3].

The food which facilitates the growth of the staphylococcus is mainly pastry makings with the cream, dairy ice creams, the food treated such as hams, the pies and rilette, and tuna and poultry, the potato salads. Cooked products contaminated after cookings (chopped meats, fish, sections of pork-butcher). Products with water content reduced (saltings, fish dried and smoked, dried milk). Cheeses, following an insufficient acidification of curd. Ovoproducts, mayonnaise, dairy products (e.g. condensed milk), creams, ices. The contaminated food has the same

aspect (appearance, odor, taste) that the healthy food [4].The placement of detector of this bacterium is essential in order to prevent the risks of contamination.

A preliminary work was published on the AMX-NP characteristics as for the detection of the staphylococcus aureus [5].The objective of this work is to make an analytical application of this electrode in potato juice by using the method of the square waves voltammetry.

II. EXPERIMENTAL

a) Reagent

Provisions were made for oxygen removal by bubbling the solution with azotes gas for about 5 min then the solution was blanketed with azotes gas while the experiment was in progress. For reproducible results, a fresh solution was made for each experiment.

b) Instrument

Voltammetric experiments were performed using a voltalabpotentiostat (modelPGSTAT100, EcoChemie B.V., Utrecht, The Netherlands) driven by the general purpose electrochemical systems data processing software (voltalab master 4 software) run under windows 2007. The three electrode system consisted of a chemically modified carbon paste electrode as the working electrode a saturated calomel electrode (SCE) serving as reference electrode, and platinum as an auxiliary electrode

c) Electrodes

The working natural phosphate paste electrode was prepared by mixing appropriate weight of natural phosphate powder with paraffin oil. The whole cell modified natural phosphate paste was subsequently packed firmly into the electrode cavity (0.1256 cm²) and polished to a smooth shiny finish by gently rubbing over an ordinary weighing paper. Electrical contact was established with a bar of carbon. Amoxil-modified natural phosphatepaste electrodes (AMX-NP) were prepared by immobilizing the Amoxil system by soaking the preformed natural phosphate paste electrode in a solution containing the Amoxil solution.

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d) Analytical procedure

The modified natural phosphate paste electrode was immersed in a cell containing bacteria sample to get a chemical accumulation. Meanwhile, the solution was rotated about 600 rpm at open circuit. After a desired contact time, the electrode was removed from the preconcentration cell, rinsed with DW and placed in the measurement cell containing the supporting electrolyte ($1.0 \text{ mol L}^{-1} \text{ NaCl}$). The solution was deaerated with nitrogen for 10 min. The voltammetric curve was recorded. The same procedure was carried out in sample analysis and all electrochemical experiments were carried out at room temperature. The square wave voltammograms were recorded in different bacteria concentrations using 5 mV of the pulse amplitude, step potential 50 mV and the duration time is 2 s at scan rate 1 mVs^{-1} .

III. RESULT AND DISCUSSION

In order to study the effect of the optical density of the bacterium on the electrode, we made the electrochemical characterization of this one by using the methods of voltammetry square waves. The electrode with natural phosphate paste modified by the amoxicillin with a concentration of 30g/L was characterized in the presence of bacteria with different density optical. In the presence of the bacteria, the electrode displays a significant increase in the density of current. This electrochemical behavior of the electrode is confirmed by the voltammetry square waves. The capacity of detection of the electrode thus increases with the increase in the bacterial load. (figure 1).

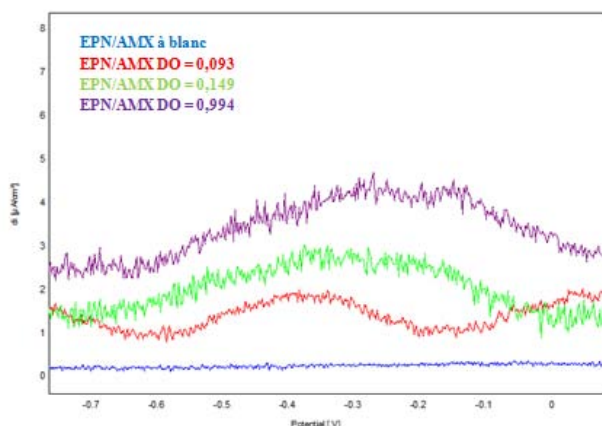


Figure 1: Superposition of the voltammograms with square waves of EPN/AMX ads. to white and EPN/AMX ads. (30g/l) in the presence of the bacteria with various optical densities in NaCl to 0,1 M; $v = 100 \text{ mV/s}$, of -2V with 2V; $\text{pH} = 7,42$

This sensor was the subject of an electrochemical characterization by the cyclic voltammetry for identifying the duration of detection of the bacterium. Within sight of the results, it arises that

after 50 cycles, corresponding to one hour duration six minutes (1.06 min), in the presence of the bacterium, the electrode displays an increase in the electroactivity (figure 2).

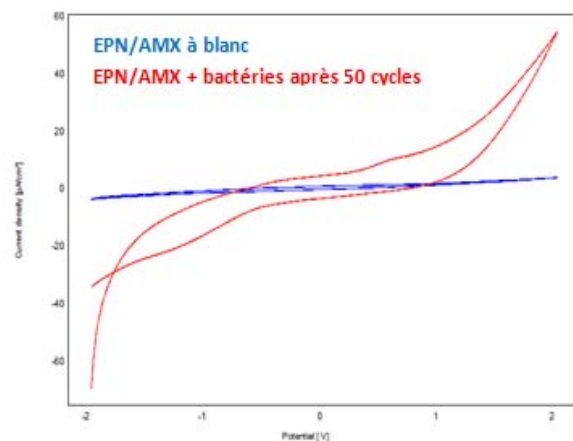


Figure 2: Comparison of the voltammograms cyclic of AMX-NP ads. with white and the bacteria after 50 cycles in NaCl to 0,1 M; $v = 100 \text{ mV/s}$, of -2V with 2V; $\text{pH} = 7,42$

A study of the influence of the pH on the electrochemical sensor in the presence of the bacteria was made. The electrode showed a good electroactivity in the presence of the bacteria in the acid media, neutral and basic. We thus compared the aforementioned voltammograms in order to identify the medium which is most favorable for him. The results are illustrated by (figure 3).

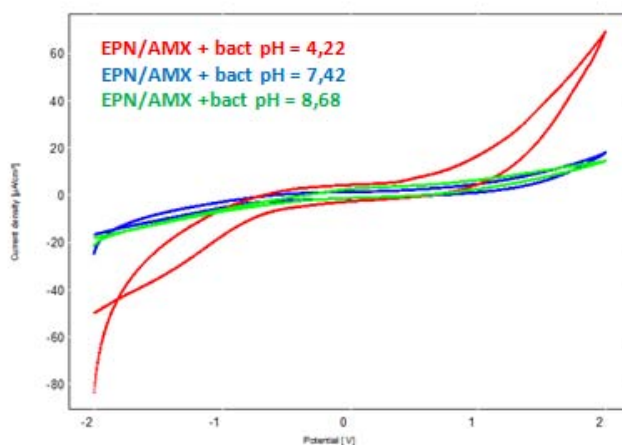


Figure 3: Superposition of the cyclic voltammograms of EPN/AMX ads. to different pH in the presence of the bacteria in NaCl to 0,1 M; $v = 100 \text{ mV/s}$, of -2V with 2V

Taking into consideration these result, we can say that our electrode presents a better electro activity in acid medium, $\text{pH} = 4, 22$.

a) Analytical application in the potato juice

Under the optimized conditions, the sensor (AMX-Np) was used for the detection of *staphylococcus aureus* in sample of potato juices which were the subject of no preliminary treatment. The analytical application consisted in adding various quantities of

physiological water containing the bacterium in the potato juice in order to vary the optical density and we have each time made an electrochemical characterization by using the voltammetry with square waves. The seresults are presented in table 1.

Table 1: Density of current according to the optical density

DO	0	0,410	0,520	0,661	0,733	0,818	0,912	0,991
di ($\mu\text{A}/\text{cm}^2$)	1,45	1,679	1,995	1,980	2,105	2,063	2,155	2,351

Figure 4 shows the variation of the density of current according to the optical density.

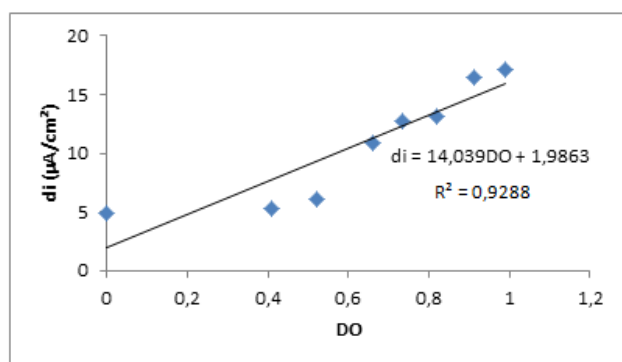


Figure 4: Density of current according to the optical density

The density of current increases with the evolution of the optical density of the bacteria, measured using a spectrophotometer. Figure 4 shows a typical linear answer, which can be expressed according to the following equation: $di = 14,039DO + 1,9863$.

b) Comparison of the characteristics of the electrochemical sensor (SD, LD et LQ)

Table 2: Sensibilities of detection (SD), Limitate detection (LD) and limits of quantification (LQ) of the bacterium according to analytical mediums'

Sensor	Characteristics	Analytical mediums	
		Electrolyte support	Potato juice
AMX-NP(adsorption)	SD ($\mu\text{A}/\text{cm}^2$)	$5,800.10^{-7}$	$6,350.10^{-9}$
	LD	$4,900.10^{-7}$	$2,218.10^{-8}$
	LQ	$1,635.10^{-6}$	$7,390.10^{-8}$

Taking into consideration these result, it arises that the electrochemical sensor being studied, shows a better SD, LD and LQ in the potato juice. This result

comes to confirm the effectiveness of this sensor, considering the potato juice is a hostile environment with the bacteria. Indeed the potato juice contains certain active molecules anti-bacterial in the fight against the bacteria (*the helicobacter pylori*) responsible for the ulcer of the stomach [5].

IV. CONCLUSION

The electrochemical sensor (AMX-Np) is extremely sensitive to the bacterium. The pH has an influence on the electroactivity of this electrode and the acid medium seems more favorable. Also, its duration of detection is satisfactory. The analytical study in a potato juice sample showed good results.

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