Interdigitated biosensor for multiparametric monitoring of bacterial biofilm development

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Abstract

Adhesion and development of bacterial biofilms onto any kind of surfaces causes numerous problems in a wide variety of sectors, and more particularly, in those related with the medical environment or the industry. Specifically, medical implants are frequently infected due to bacterial adhesion threatening the health of the patients and involving additional hospital costs [1].

Biofilms are formed when bacteria adhere to surfaces and excrete a sticky and viscous substance that forms a complex and heterogeneous polymeric structure. This extracellular matrix increases the antimicrobial resistance of the adhered microorganisms by blocking the access of antibiotics through it [2]. Thus, treatment and disposal of these kinds of infections is often hampered by the surrounding structure and by the total lack of symptoms in the early stages of the infectious process. Therefore, it is necessary to find new methods for the early detection of biofilm development and growth since the first steps.

One of the most widely used detection methods for microorganism detection is the electrical impedance spectroscopy [3]. The working principle is based on the impedance changes induced after cell adhesion onto the measured electrodes [4]. Sensors based in interdigitated micro and nanoelectrode arrays are very commonly used for this purpose because they offer notable features such as a large sensitive area in a limited space. In this work, chromium-gold interdigitated electrodes (**Figure 1 (a)**) have been designed and fabricated on silicon wafers using conventional lithographic techniques. **Figure 1 (b)** shows the experimental setup developed ad hoc for electrical impedance spectroscopy. Measurements have been supported by a multiplexer system [5] that allows an automated sampling. **Figure 2** shows the results obtained for the impedimetric monitoring of bacterial biofilm adhesion and development. The presence of microorganisms has been detected since the first steps of bacterial adhesion.

On the other hand, so as to enhance both the specificity and the sensitivity of the detection method, electrochemical monitoring through cyclic voltammetry has been conducted. Measurements have been taken using the redox cycling technique that allows signal amplification by the use of two working electrodes: collector and generator. By exciting one electrode at a potential that allows the reduction of the species and the other electrode at the oxidation potential, species are continuously being transformed at the generator electrode and are converted back to their original form at the collector. Moreover, the measurement of the temperature onto the surface where biofilm deposits can give additional information about its metabolic activity. Real-time surface temperature measurements can also be used to correct the thermal drifts of the monitoring curves.

For these purposes, a multiparametric sensor has been designed and fabricated on thermally oxidized silicon wafers and on layers of Cyclic Olefin Polymer (COP). COP increases the adhesion of bacteria and its malleability allows its use in 3D applications. For electrochemical experiments, the choice of biosensors based on interdigitated electrodes has been kept because of their good performance [6]. As the gap between electrodes is very small, molecules reach the opposite electrode immediately and the resulting current is increased. Two interdigitated working electrodes of titanium-gold, a titanium-silver reference electrode and a titanium-platinum counter electrode have been deposited for the electrochemical sensing part (**Figure 3 (a)** and **Figure 3 (b)**). Moreover, a titanium-platinum temperature sensor has been added onto the chip (**Figure 3 (c)**). **Figure 3 (d**) shows the developed setup for multiparametric monitoring. The measurements and characterizations carried out proved the linearity of the temperature sensor that has a TCR value of 1950 ppm/ºC. Moreover, the first measurements carried out in order to compare impedimetric and electrochemical techniques have proved that the specificity and the sensibility have been improved.

References

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Figures

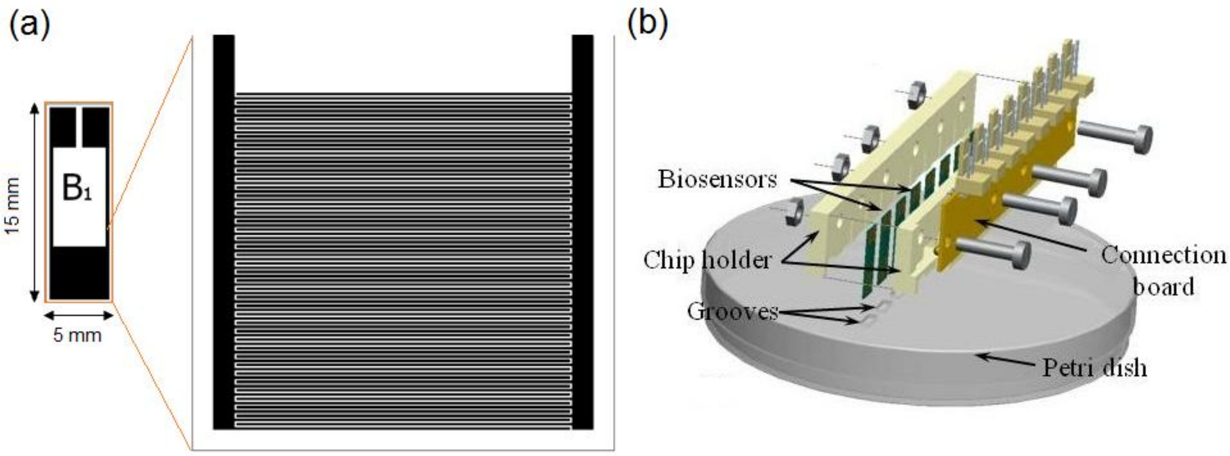

Figure 1: (a) Interdigitated microelectrodes array based sensor and (b) experimental setup for electrical impedance measurements.

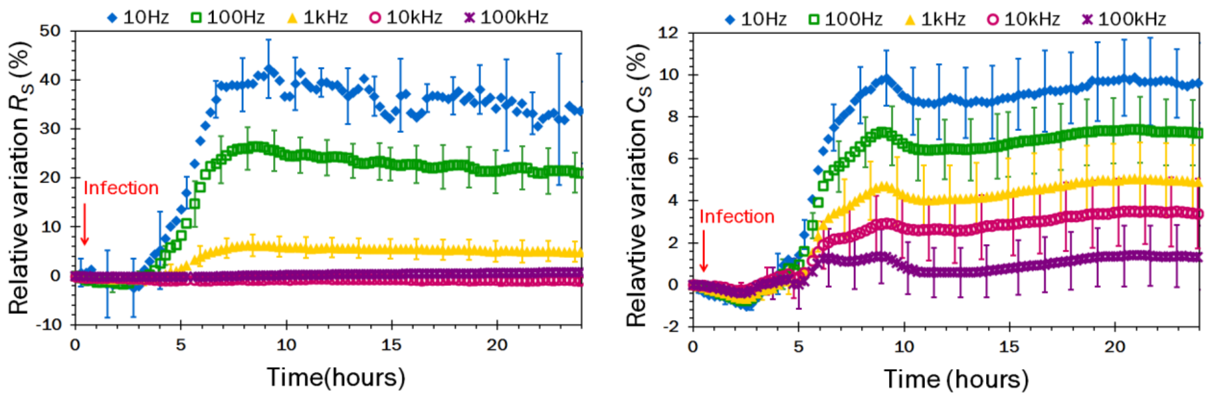

Figure 2: Impedance measurement during the development of a microbiological culture of *S. epidermidis*.

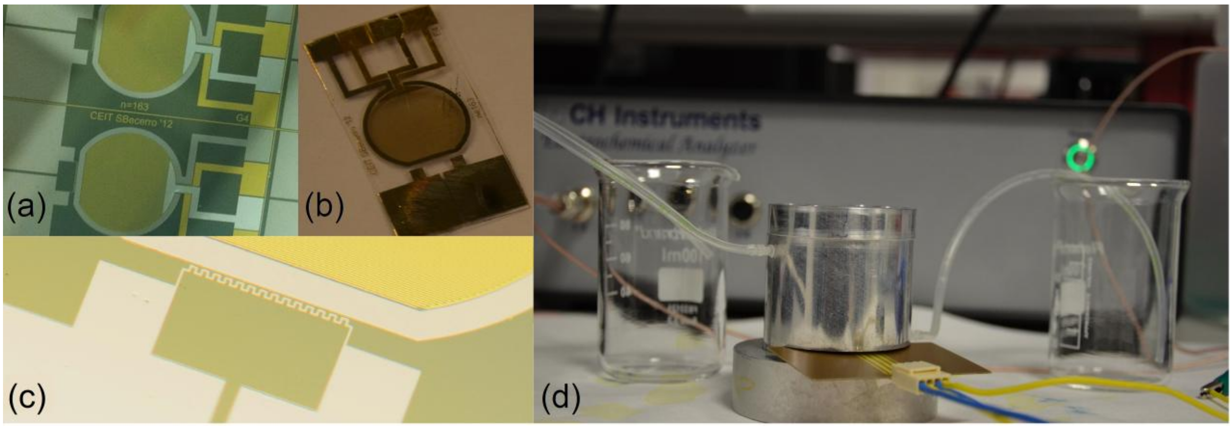

Figure 3: Interdigitated microelectrode array based sensor (a) on silicon substrate and (b) on COP polymer; (c) integrated temperature sensor and (d) experimental setup for multiparametric measurements.