

IMPEDANCE BASED BIO-CHIP TO MONITOR INTERACTIONS BETWEEN CELLS

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ABSTRACT— An impedance method was developed to quantify the interaction between immune system cells (hemocyte) with intruder cells (parasites). When the hemocyte cells interact with the parasites, they cause a defensive reaction and the parasites start to aggregate in clusters. The level of aggregation is a measure of how strong the host-parasite interaction is, and provides information about the efficiency of the immune system response. The cell aggregation is monitored using a set of microelectrodes. The impedance spectrum is measured between each individual microelectrode and a large reference electrode. As the cells start to aggregate and settle down towards the microelectrode array, the impedance of the system is changed. It is shown that the system impedance is very sensitive to the level of cell aggregation and can be used to monitor in real time the interaction between hemocyte cells and parasites.

INTRODUCTION— Cell impedance sensors have been reported in the literature to measure the interaction between cells and their surroundings, for cell identification, adhesion, coverage, and even cell motility [1-3]. This work expands on the use of micro-electrical impedance spectroscopy for the characterization of the interaction between different cells.

The motivation for this work comes a bivalve of economic interest in the Atlantic coast, the clam *Ruditapes decussatus* which is often infected by a parasite, the *Perkinsus atlanticus*. It was found that some of their species seem to resist better than others to the infection, suggesting differences in their immune system (also known to occur in other animals as well as in humans). The interesting point is that the parasites show a peculiar behavior; they aggregate in clusters when they feel threatened by the hemocytes. The level of this aggregation may provide a measure of the immune system efficiencies. Furthermore, both cells can survive at room temperature in normal atmospheric conditions, and are therefore easy to handle. These characteristics make the bivalve/parasite system an ideal prototype to study interaction of immune systems with intruders. These studies can be extended to other types of cells and used as biosensing techniques to monitor in real time how the immune system responds to infections. Figure 1 shows a schematic view of the measuring system. A glass slide with a microelectrode array consisting of 14 gold electrodes (20 μ m x 3000 μ m) formed by photolithography methods is placed in a small reservoir, built using a thin plastic ring (2 mm height). The reservoir is filled with the medium containing the cells. The AC impedance between 100 Hz and 1 MHz was measured between each individual gold microelectrode and a large top electrode made of a transparent conducting glass. The impedance was measured using a Fluke PM6306 impedance analyzer.

RESULTS— We observe that different types of cells show a distinct temporal evolution of their impedance spectrum. Figure 2a, shows the behavior when only hemocytes are present in the medium. The main relaxation frequency remains almost unchanged with time, while the capacitance at low frequencies decreases. When parasites are added to the medium containing hemocytes, we observe a dramatic change in the behavior, the main relaxation frequency shifts now rapidly to lower frequencies as shown in Figure 2b. This movement stops after about 30 minutes. We believe that this pronounced shift to lower frequencies is caused by the aggregation of parasites in clusters followed by their sedimentation on top of the microelectrodes (see

Fig.1b). This sedimentation of the clusters causes an increase in the contact resistance of the electrode/medium interface.

The aggregation of parasites is a defense mechanism against the attack of the hemocytes. Therefore, the kinetics movement of the loss peak provides information about the efficiency of the immune system cells. Current studies are focused in the optimization of this technique, so it can be used in a biosensor to screen hosts with good immune systems.

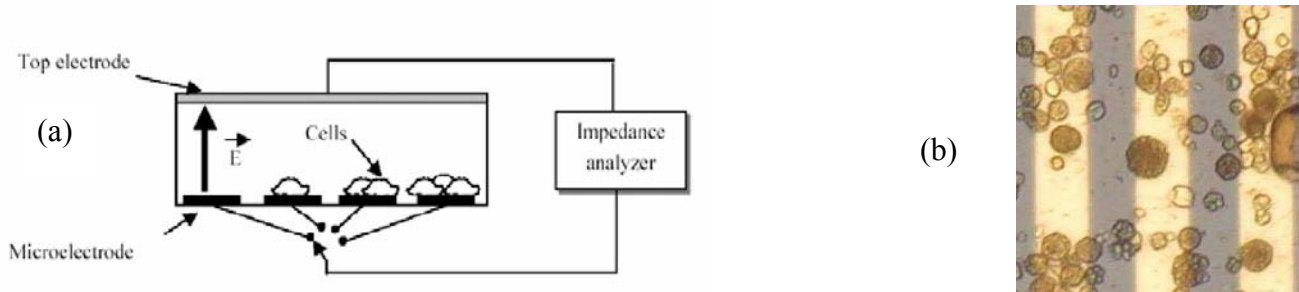


Figure 1 (a) Schematic view of the measuring system. (b) photograph of parasites cells aggregated in clusters and deposited on top of gold microelectrodes. The distance between the gold electrodes is 20 μm .

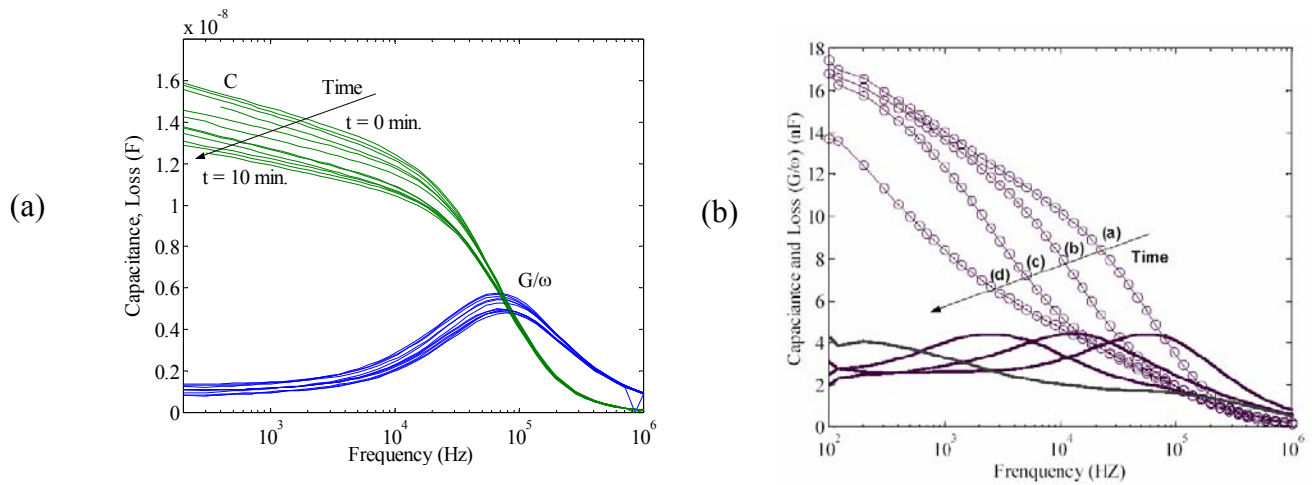


Figure 2. (a) Time dependence of the impedance spectra of a medium with hemocytes cells. (b) Temporal evolution of the impedance spectra of a medium with mixture of hemocytes and parasites cells. (O) Capacitance and (-) Loss (Conductance/ ω). (a) Soon after insertion, (b) $t=18$ minutes, (c) $t=25$ minutes, (c) $t=32$ minutes, and (d) $t=50$ minutes.

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