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Mechanical behavior characterization of glioblastoma cell using Scanning Ion Conductance Microscope (SICM)

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1. Introduction

Understanding the mechanical properties of biological materials and tissues, especially at the single cell level, is crucial for understanding various physiological processes and diseases such as cancer. Many methods have been developed to study the mechanical properties of single living cells, but most involve direct mechanical contact with the cell, which can lead to contamination or damage. The Scanning Ion Conductance Microscope (SICM) provides a non-contact alternative for mapping stiffness and imaging topography on living cells, avoiding physical contact with the sample and making it less invasive than other methods. This study proposes to use the SICM to measure the stiffness of glioblastoma cancer cells using the method proposed by (Rheinlaender, J., & Schäffer, T. E. 2013).

2. Methods

2.1 SICM experiment

The study was performed on human glioblastoma cells from the LN229 lineage in Tyrode medium. The scanning ion conductance microscope (SICM) was employed to assess cellular mechanical properties. SICM operates by detecting variation in ionic current between a sensor located within a probe and another in the surrounding cell medium. As the probe approaches the cell surface, a decrease in current is observed, enabling non-contact determination of the relative distance between the probe

and the cell surface. By applying a constant pressure through the probe and lowering it towards the cell, we can measure the decrease in current as a function of the position of the probe (Figure 1). The cell's local Young's modulus can be determined by comparing the current slope to a rigid (plastic) substrate (Pellegrino.M, et al. 2012).

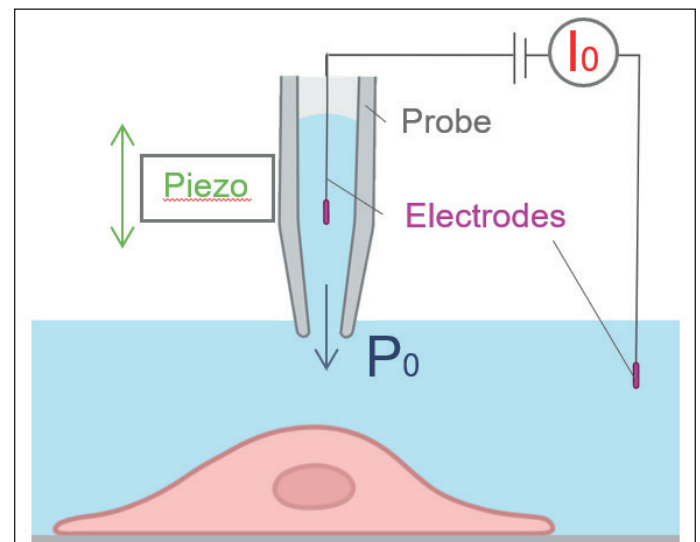


Figure 1. Schematic of the experimental SICM setup.

2.2 Numerical simulation

The probe dimensions used in SICM are obtained from scanning electron microscope (SEM) images. A

digital representation of the borosilicate probe can be recreated and meshed. The SICM can also generate a topography of the cell surface, allowing the shape of the cell to be extracted and meshed. The stiffness reliability of the SICM experiment can be numerically reproduced using a combination of fluid and solid mechanics through the finite element method.

3. Results and discussion

The SICM allows for the measurement of ion current as a function of the probe height during its descent towards a cell (Figure 2). A decrease in current is observed as the probe approaches the surface. Additional measurements are performed on the bottom of a petri dish, which is considered to be highly rigid. By considering the slopes of the curves obtained from cell and petri dish measurements, as well as the geometric parameters of the pipette, it is possible to calculate the Young's modulus associated with the cell (Kalinin.S, et al. 2011). The local modulus of elasticity associated with the slopes shown in Figure 2 is 1.4kPa.

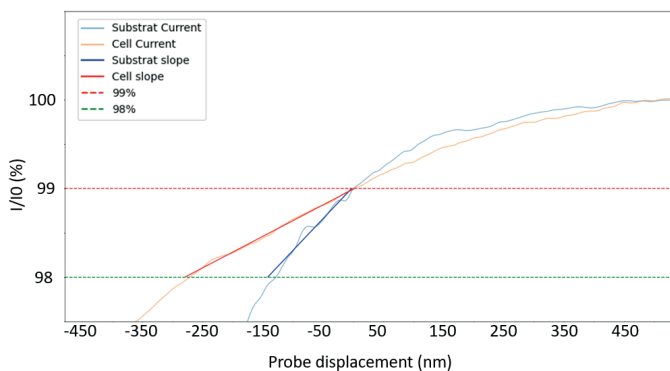


Figure 2. Decrease in current as a function of the relative vertical position of the probe on a cell and the plastic.

To improve results obtained by SICM, a numerical simulation of the experiment was conducted. Flow simulation within the probe was performed to evaluate pressure drop, between the inlet and outlet. It was observed that there was a 95% pressure drop at the outlet with 8 kPa is applied at the inlet, which was attributed to the geometry of the pipette. This pressure drops directly impacted observation of stiffness of the cell obtained experimentally and had to be considered to correct the cell's stiffness. Additionally, the significance of the opening angle and outlet radius of the pipette on pressure drop

was noted. Moreover, geometric parameters such as the opening angle and the outlet radius of the probe influence pressure drop. It seems crucial to measure experimentally these parameters.

The results obtained by SICM are consistent with those in the literature for other cell types (Rheinlaender, J., & Schäffer, T. E. 2013, Pellegrino.M, et al. 2012, Rheinlaender,J, et al. 2020). The studies assume that the cell exhibits linear mechanical properties, but the proposed methodology could be used with other behaviours (hyperelastic, viscoelastic, etc.).

4. Conclusions

The capability of using SICM, employing the method proposed by (Rheinlaender,J, et al. 2013), to characterize the mechanical behaviour of cancerous cells was demonstrated in this study. Numerical simulation of the experimentation highlighted pressure losses associated with pipette geometry and aids in improving experimental results. The coupling of these two techniques provides a reliable non-contact method that does not damage the cell to identify its stiffness. Characterizing the mechanical behaviour of a cell is a crucial step towards developing a cellular model and understanding physiological processes and diseases.

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Conflict of Interest Statement

The authors declare that they have no conflicts of interest.

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