Support information

Single-cell correlative nanoelectromechanosensing approach to detect cancerous transformation: Monitoring the function of F-actin microfilaments in the modulation of ion channel activity

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S1. Tungsten needle formation by electrochemical etching

To prepare tungsten (W) needle we must clean the tungsten wire by the solution of $C_6H_8O_7$, CH_3COOH and HNA. Then we sharp the tip of the wire with electrochemical etching method. In this manner, an electrochemical sharpening system (schematic presented in below) has been designed and made. We used from KOH (3.5 M) electrolyte and a steel electrode. The W wire was entered into the electrolyte (with the length of 1 mm). Electrochemical etching of the wire was started by applying a voltage between wire and electrode (~ 3.5 v) and carried on until the tip of the wire became so sharpened in which the current of the system suddenly would be decreased. Such decrement in the current would be monitored by an intermediate circuit based on current differentiation and the voltage would be turned off immediately after decrement to avoid further etching of the wire (which would lead to blunting the tip). An optical image of the tip was presented below.

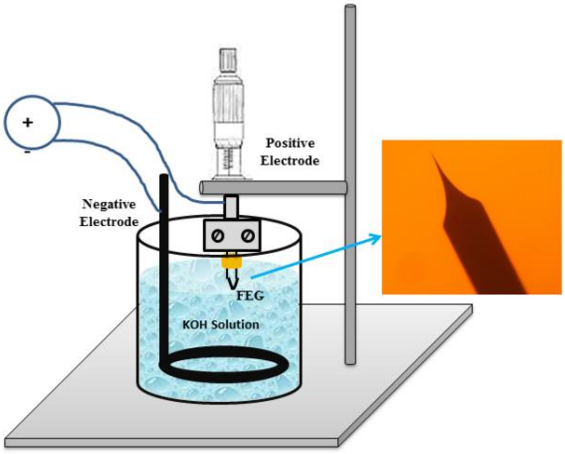


Figure S1. Schematic from the setup of tungsten (W) electrochemical etching. Optical image: sharpened W needle

S2. Lifetime measurement of doped SiNT

An enhancement in the magnitude of the charge carrier lifetime is found depending on the incorporation of impurities after the growth. The results of lifetime measurement from non doped SiNT (figure S2-A) in comparison with doped one (figure S2-B) (measured by lifetime measurement system: Micro PED. WT-2000 PVN. SEMILAB Co. Germany) indicate this increment. Our doping process has increased the life time of charge carriers in SiNT probe to 47 µsec.

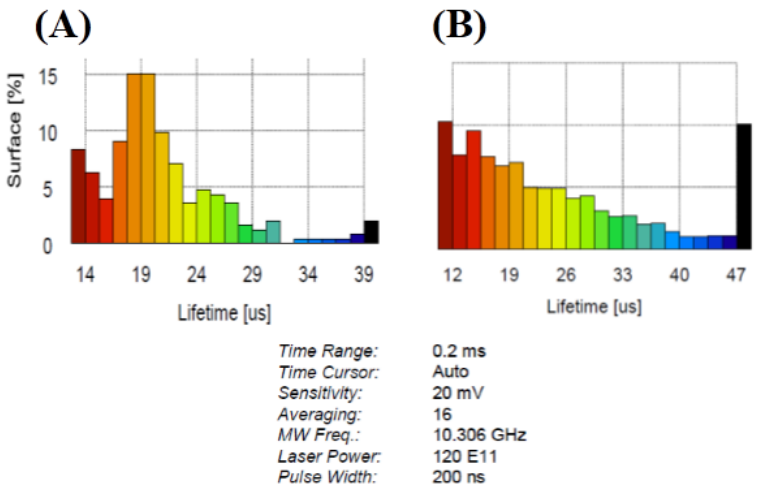


Figure S2. Lifetime measurement profile from A) non doped and B) doped SiNT probes.

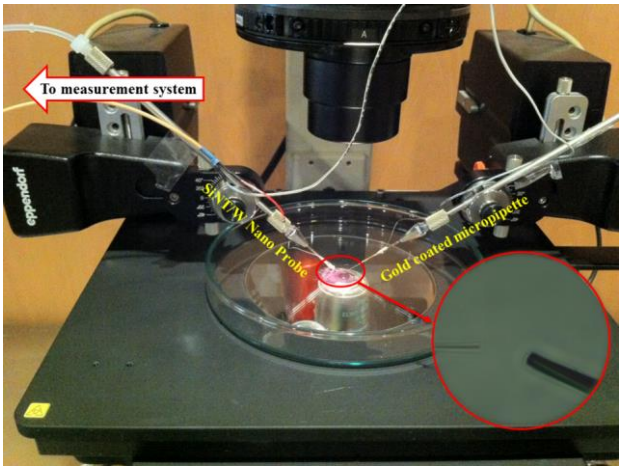


Figure S3. Optical image from the testing system. Both SiNT nanoprobe and electrically activated glass micropipette were assembled on microinjection microscope system (Narishige & Nikon Co. Japan) and connected to readout system.

S3. Response comparison between dry, solution and cell connected situations between the SiNT/W nanoprobe and Au/ glass µpipette

The basic electrical sensitivity of the device was characterized by entering the Sensing system in ionic solution (Figures S4-A and D) followed by comparing with the sensitivity after connecting the nanoprobe and micropipette to the cell (Figure S4-B and D) and finally was scaled with dry ambient (Figure S4-C and E). The magnitude of the impedance (device sensitivity) decreased when the ambient has been replaced from air to cellular media solution in a fixed distance between SiNT and micropipette (~7 µm).

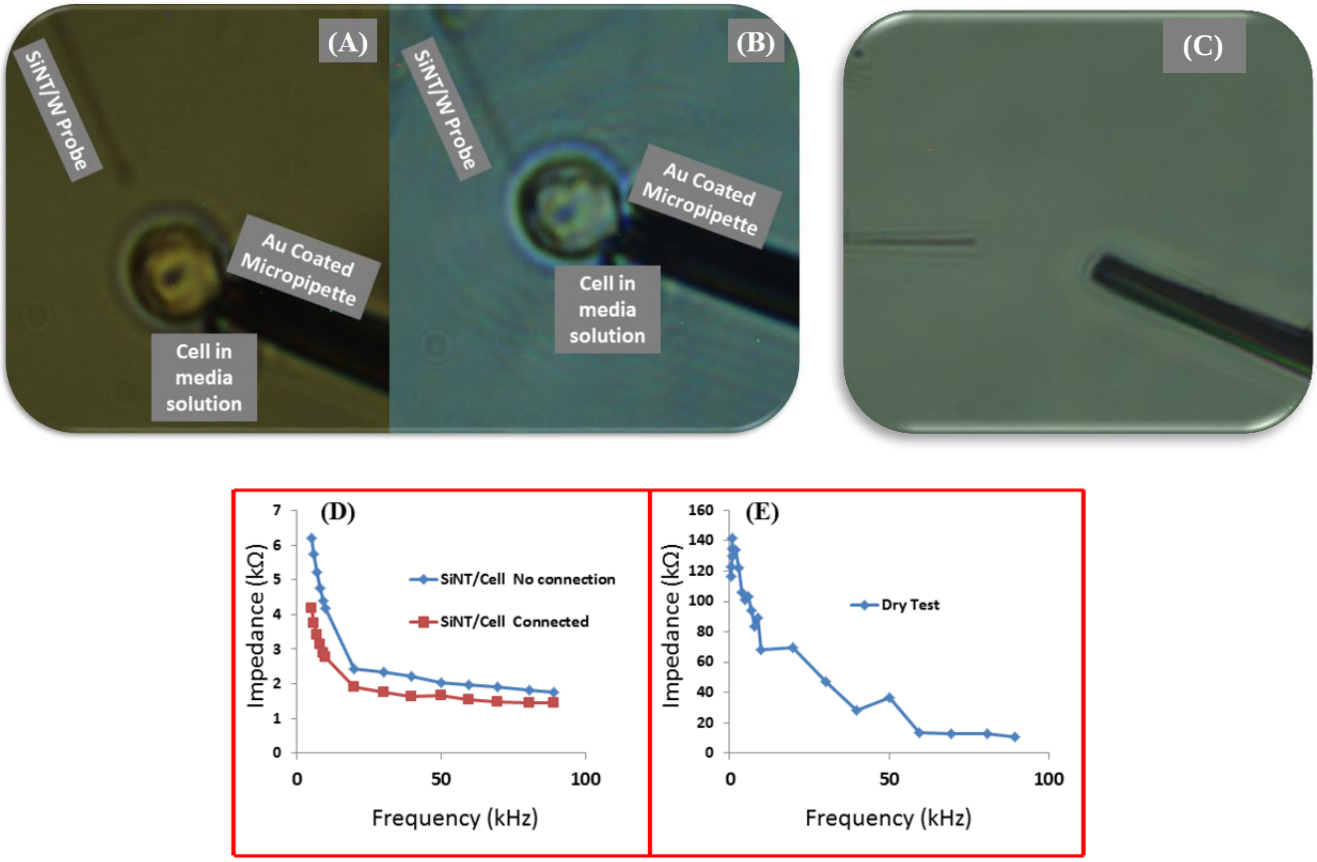


Figure S4. Correlative nanoelectromechanosensing system in which A) cell has been aspirated to pipette and A) hasn't been concede to SiNT nanoprobe. B) has been connected to SiNT nanoprobe. C) no cell was aspirated to the pipette. Impedance response of the system in D) media solution and E) air ambient.

S4. Cell culture

All cell lines were obtained from the National Cell Bank of Iran, Pasteur Institute. HT-29 and SW-48 cells had been isolated from grade I and IV human colorectal tumors. QU-DB and MRC-5 had been derived from human lung carcinoma and normal lung tissue respectively. Cells were maintained at CO_2 incubator (37 °C, 5% CO_2) in RPMI-1640 medium (Sigma) supplemented with 5% fetal bovine serum, and 1% penicillin/ streptomycin (Gibco). The fresh medium was replaced every other day. Prior to each experiment, cells were trypsinized to be detached from

the substrate and suspended in the culture medium. To minimize the effect of trypsinization[1], the procedure was taken less than 4 minutes at room temperature around 20-22^{o}C.

S5. Measurement of young modulus and aspiration pressure of the suctioned cells.

Mechanical properties of a cell can be measured by micropipette aspiration quantitatively. Considering appropriate mechanical models and subsequent theories, micropipette aspiration technique has been broadly utilized for estimation of whole body mechanical properties of cells such as cancerous[2] [3], Chondrocytes[4], Fibroblast[5] and mesenchymal stem cells[6]. In the present study, Micropipette Aspiration (MA) technique was implemented to evaluate mechanical properties of HT29, SW48, QU-DB, and MRC-5 cells. The procedure was performed according to previous work in which a generalized Maxwell model including a spring (k_1) in parallel with series of a damper (μ) and a spring (k_2) were used to extract the mechanical properties of the aspirated cell[3] [7] [8] [9] [10].

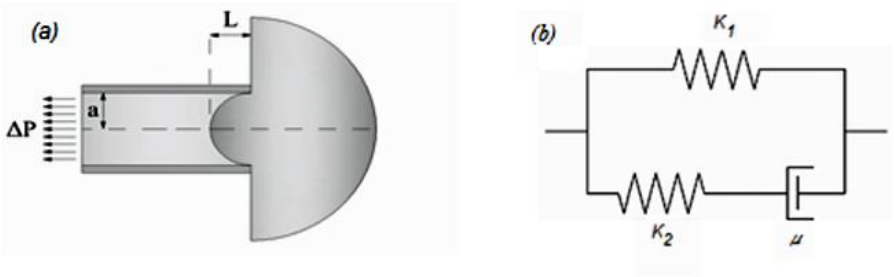


Figure S5. (a) The theoretical representation of the MA experiments. The cell is modeled as an axisymmetric elastic half-space. A negative pressure, ΔP, is applied through the micropipette. (b) A three-parameter viscoelastic model was utilized to represent the material behavior of the cell.

The relation between elastic and viscous parameters is defined by Equ. 1 :

$$\frac{\mathrm{Lp(t)}}{\mathrm{a}} = \frac{2\Delta p}{k_2 \pi}\left(1 + \left(\frac{k_1}{k_1 + k_2} - 1\right)e^{-\frac{t}{\tau}}\right)h(t) \qquad (1)$$

Where ΔP, $L_p(t)$, $h(t)$, and a are the applied pressure, the aspirated length, the unit step function, and the inner radius of the micropipette respectively. Viscoelastic parameters of cells are obtained by curve fitting of experimental data (L/a) with time using least square method in Matlab.

The equilibrium Young's modulus (E_∞) is calculated according to Equ. 2

$$E_\infty = \frac{2}{3} k_1 \tag{2}$$

In steady state . The analysis of the cell aspirated into a micropipette based on cells Young's modulus gives the following result:

$$\Delta P = 1.4\pi E \frac{L_p}{R_p} \tag{3}$$

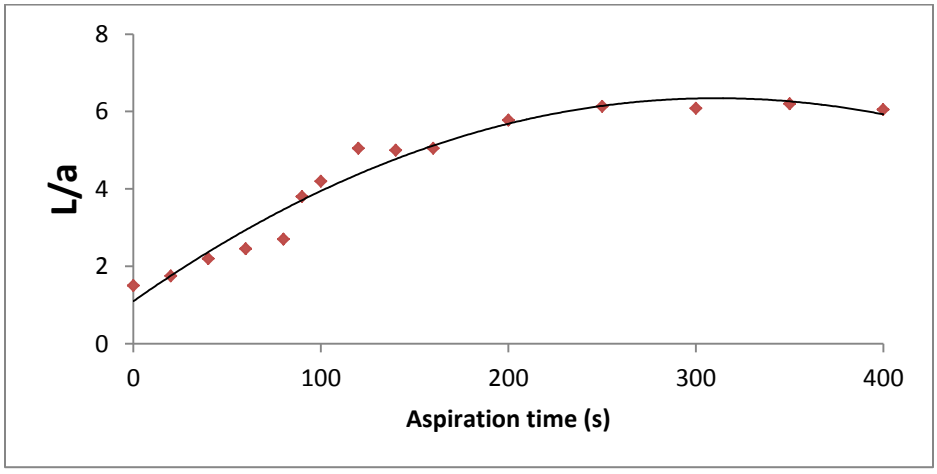


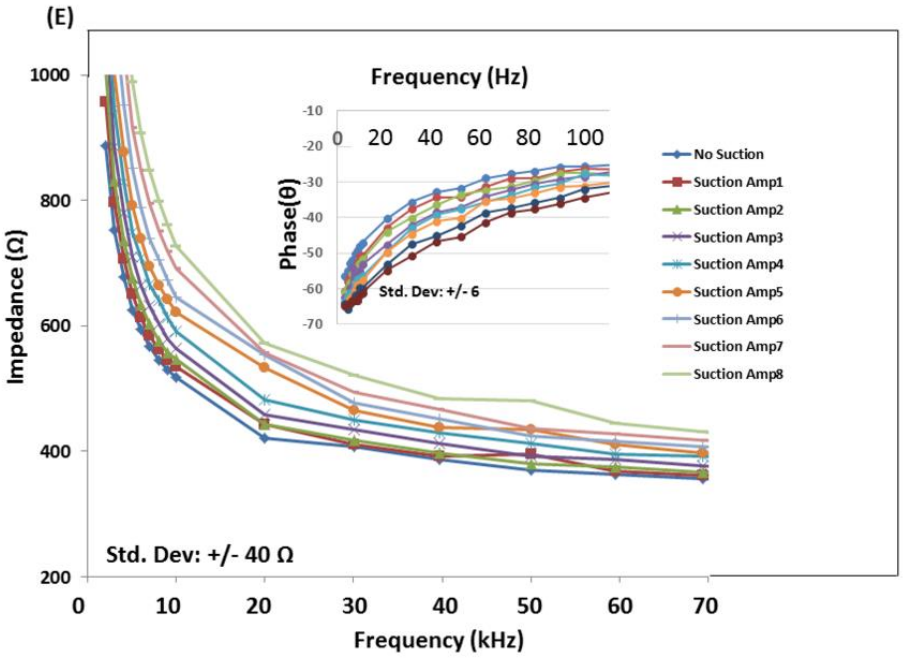
Figure S6. Creep response of grade IV colon cancer cells line (SW-48), the data was fitted with nonlinear regression,

S6. Effect of pipette size on impedance

Because of the deformability of cancer cells they will flow into the micropipettes with bigger nozzle radius (more than 5 um) during their aspiration with larger suction forces. So we couldn't

applied various suction force during their aspiration to such pipettes. Hence we investigated the effect of pipette radius on the aspiration and electromechanical properties of lung healthy (MRC-5) cell.

The diagram of cell impedance as well as phase variations during its aspiration into 10um micropipette with various suctions forces have been plotted in figure S. as a well confirmation to our reported data in the paper, the impedance of the cell has increased observably by increasing the suction force.



FigureS8. Impedance and phase responses of MRC-5 single cell aspirated by micropipette with the inner diameter of 10 µm.

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