Supplementary material

for

AFM study shows prominent physical changes in elasticity and pericellular layer in human acute leukemic cells due to inadequate cell-cell communication

Nataliia V. Guz[1], *Sapan J. Patel*[1,2], *Maxim E. Dokukin*[3], *Bayard Clarkson*[2], *Igor Sokolov*[3,4,5*]

[1] Department of Chemistry, Clarkson University, 8 Clarkson Avenue, Potsdam, New York 13699-5820, USA

[2]Memorial Sloan Kettering Cancer Center, Sloan Kettering Institute, Molecular Pharmacology and Chemistry Program, 415 East 68th Street, New York, NY, 10065

[3] Department of Mechanical Engineering, [4]Department of Biomedical Engineering, [5] Department of Physics and Astronomy, Tufts University, 200 Boston Ave, Medford, MA, USA;

Corresponding authors: Igor Sokolov, Department of Mechanical Engineering, Tufts University, 200 Boston Ave, Medford, MA, USA; phone 1-617-627-2548; email: igor.sokolov@tufts.edu.; Bayard Clarkson, Memorial Sloan Kettering Cancer Center, 415 East 68th Street, New York, NY, 10065, USA; phone 1-646-888-2080: email: clarksob@mskcc.org

Measurement of the cell radius using the AFM data collected in the force-volume mode

The radius of the cell R_{cell} used in this work has been measured from the AFM topographical image of the cell obtained in the force-volume mode, and corrected by the cell deformation *i* (the latter was calculated using eq. (1) of the main text). The below is an example describing how to extract this from the force volume data collected with AFM.

Figure S1a shows a representative height image collected in the force volume mode. Because we don't want to disturb cells for excessively long time, only 16 x 16 pixels maps are collected. However, as one can see, it is quite enough to obtain the cell radius. It is worth noting that the same information has to be used to identify the force curves which will be processed for the data analysis. It should be noted that because we use the Hertz model, we can only use the pixels around the top (see the main text for more details).

To calculate the radius of the cell, we have to correct the height data for their deformation. This is important because the cell is soft and deformation can be substantial. This can easily be done by increasing the height at each pixel by the amount of deformation calculated with the help of equation 1 of the main text. The result of such correction is shown in figure S1b. Figure S1c shows the cross-section of the undeformed cell of figure S1b. The radius of the cell was calculated by parabolic fitting (done with the help of SPIP software by Image Metrology Inc.). It is 5.8 μm in this specific example.

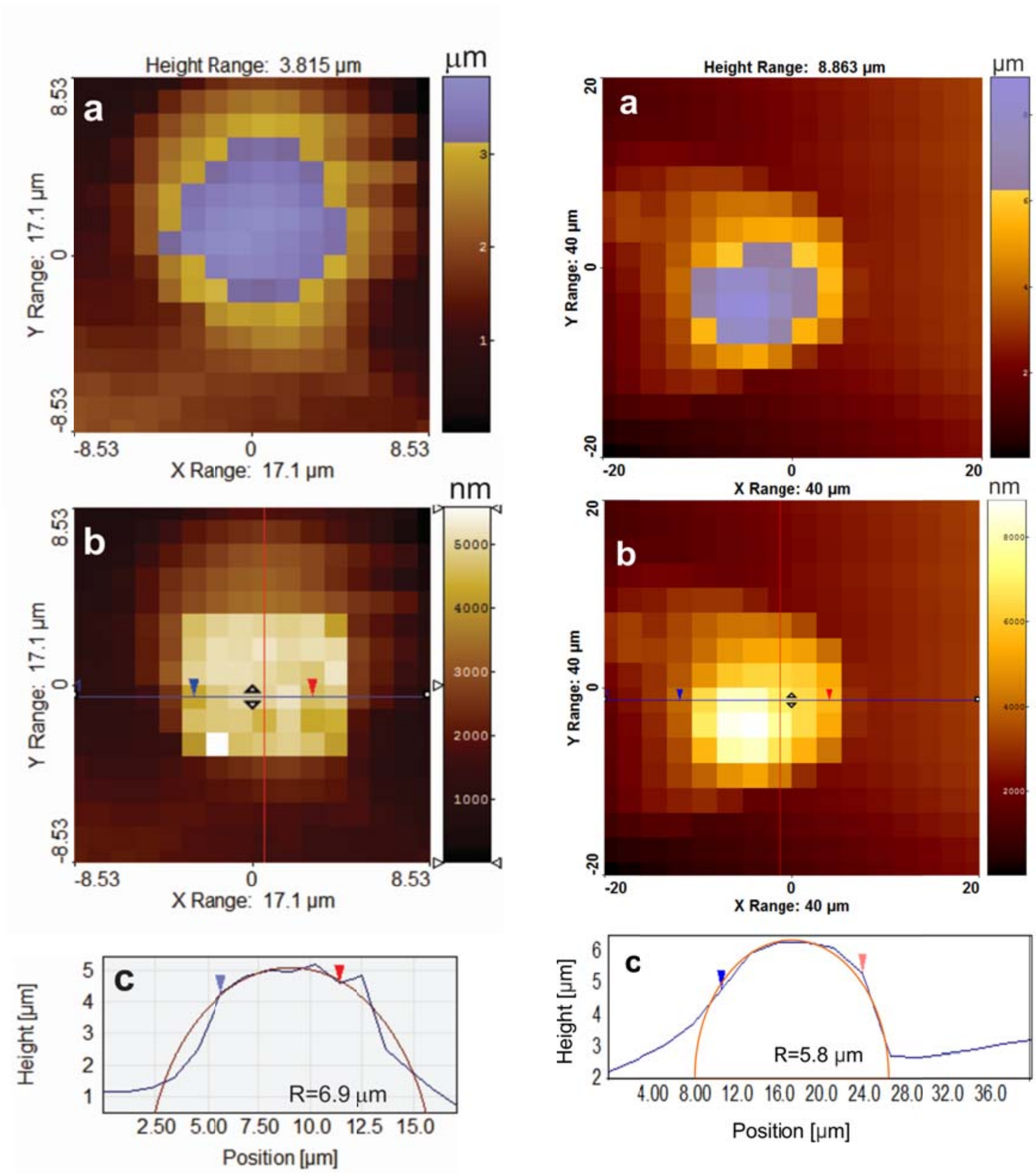

FIGURE S1. A representative (a) Height image of a deformed ALL3 cell. Area in the center of the cell where force curves were extracted is highlighted by blue color. (b) Undeformed cell topography restored near the cell center. (c) Radius of an undeformed cell calculated from the cross-section. Radius used for the final calculation was derived as a geometrical average from radiuses taken from vertical and horizontal cross-sections.

Measurement of biophysical parameters of cells around its top area

The model described in this work was developed for two spheres deforming along the line connecting their centers. Therefore, we consider only the force curves from the top area of cells. Specifically, we take the force curves in the surface points around the top when the incline of the surface is <10-15 degrees). To identify such curves, the cell height image was used, and the radius of the cell was derived as described in the previous section.

A particular example of data processing of cell #7 of low density ALL3 cells done in the way described above is shown in figure S2. The highlighted pixels shown in the height image (the left top panel) are the ones processed through the algorithms described in this work. The values of the elastic modulus (sometimes called cell stiffness), the brush length and its grafting density are shown in corresponding panels. One can see quite large heterogeneity of the derived parameters. Such heterogeneity is quite typical for cancerous cells. This emphasizes the need for taking into account more than just one measurement per cell, which sometimes is done in some reports.

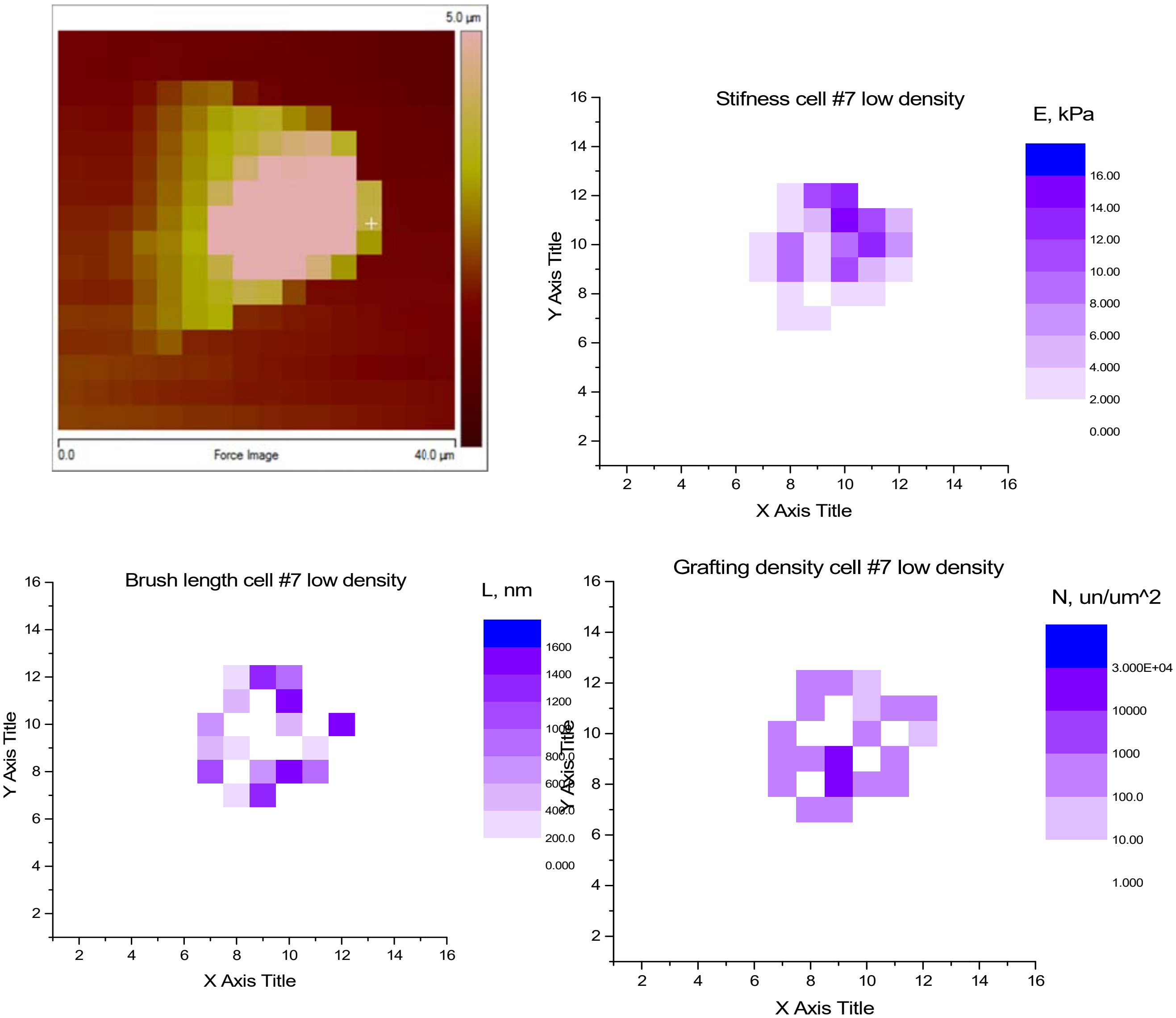

FIGURE S2. An example of data processing for each point over the cell top. The highlighted pixels in the height image (the left top panel) are the used ones for the data processing. The elastic modulus (aka known as the cell "stiffness"), the brush length and grafting density are shown for each pixel/cell surface point.

Self- consistency of the model used: Independence of the elastic modulus of the indentation depth

Independence of the elastic modulus of the indentation depth is the necessary condition of applicability of any model in which the material is considered elastic and homogeneous/isotropic. This includes the model described in this work. Therefore, it is important to verify such independence. As was mentioned in the main text, such independence has been demonstrated that virtually all cells of study. A typical dependence of the elastic modulus on the indentation depth (the mean value of the indentation calculated for the fitting interval) is shown in figure S3.

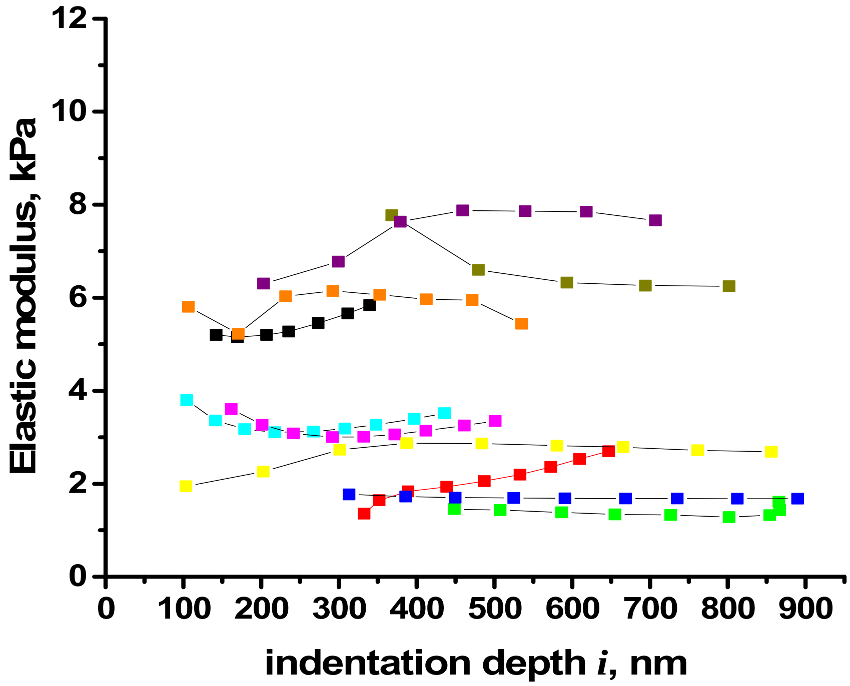

FIGURE S3. An example of a typical dependence of the elastic modulus on the indentation depth (the mean value of the indentation calculated for the fitting interval).