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Mechanics of Biological Cells Studied with Atomic Force Microscopy

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The importance of the study of living cells as fundamental "bricks" of life is hard to overestimate. Under regular functioning inside organisms, cells are subject to various mechanical forces. Mechanical properties of cells define a different physical response of cells to these external stresses. Besides just pure fundamental interest, there is a recently emerged large practical need to measure cell mechanics quantitatively. The correlation between elasticity/rigidity of cells and different human diseases or abnormalities has been recently found. It has been implicated in the pathogenesis of many progressive diseases, including vascular diseases, cancer, malaria, kidney disease, cataracts, Alzheimer's Dementia, complications of diabetes, cardiomyopathies, etc.

Atomic force microscopy (AFM) is presumably one of the most versatile approaches of studying cell mechanics [1]. When dealing with indentation of an ideal viscoelastic homogeneous material, the rigidity modulus derived (for example, the Young's modulus) should be independent of the indentation depth. At the same time, virtually all indentation experiments on biological cells have shown the depth-dependent behavior. *Independence of the elastic modulus of indentation depth is the necessary (strong) condition of applicability of any model in which the material is considered (visco)elastic and homogeneous.* Moreover, there is a substantial discrepancy between the modulus values obtained with the conical versus spherical AFM probes (the discrepancy can reach up to an order of magnitude). The above reasons let us question if cells can be described with the elastic modulus (effective Young's modulus) at all.

Here we will present our investigation of the question if cells, being highly heterogeneous objects, can be described with the elastic modulus in a self-consistent way. We analyzed the elastic modulus derived from the indentation data obtained on normal and cancerous human cervical epithelial cells. Both sharp (cone) and dull (2500nm radius) AFM probes were used the indentation experiments [2]. The elastic modulus was calculated in four different models. The cell was approximated as a homogeneous elastic medium which had either a) a smooth hemi-spherical boundary (Hertz/Sneddon models) or b) the boundary covered with a layer of glycocalyx and membrane protrusions ("brush" models) [3, 4]. We investigated the dependence of the elastic modulus on the indentation depth in each of these models.

We will demonstrate that only one model shows consistency with treating cells as homogeneous elastic medium, the bush model when processing the indentation data collected with the dull probe, Fig.1d. The elastic modulus demonstrated strong depth dependence in all other three models: Hertz/Sneddon models (no brush taken into account), Fig.1a,b and when the brush model was applied to the data collected with sharp conical probes, Fig.1c. A sharp conical probe brings strong modulus-indentation dependence for small indentations. This is presumably due to the excessively high stresses/strains produced by this sharp indenter, a phenomenon observed when indenting polymers. While Figure 1 shows representative examples of the dependence of the elastic modulus on the indentation depth, Table 1 shows the result of statistical analysis of more than 300 such curves on dozens of cells per specific case. One can clearly see s that the indentation collected with the dull probe into analyzed with the brush model bring the least indentation dependence.

We conclude [5] that it is possible to describe the elastic properties of the cell body by means of an effective elastic modulus in a self-consistent way when using the brush model to analyze data collected with a dull AFM probe. The brush layer is essentially non-elastic part of cell that is better described as entropic/steric brush. The rest of cell (cell body) can be described with just one value of the elastic modulus (virtually indentation independent if indenting below 10% of the cell height). To amplify, this is rather non-trivial result because cells are highly heterogeneous objects.

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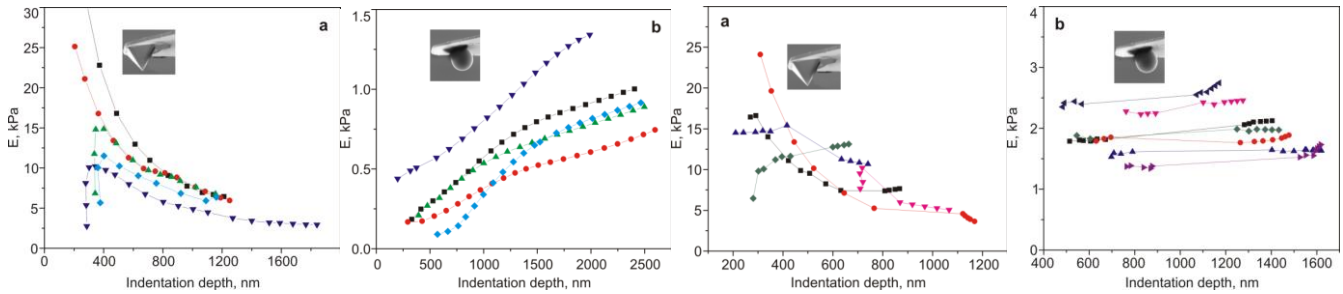

Figure 1. Representative examples of the dependence of the elastic modulus (shown in kPa) on the indentation depth (shown in nanometers) of human cervical epithelial cells when using (a,) conical and (b,d) spherical AFM indenters. Sneddon/Hertz models (a,b) and Brush model (c,d) were used.

Spherical probe			
Normal cells		Cancer cells	
Hertz	Brush	Hertz	Brush
41%	6%	40%	5%
Sharp (conical) probe			
Normal cells	Cancer cells	Normal cells	Cancer cells
Sneddon	Brush	Sneddon	Brush
47%	31%	43%	35%

Table 1. Results of compilation of the elastic modulus dependency on the indentation depth for normal and cancer cells indented with sharp and dull AFM indenters. A relative standard deviation from the average elastic modulus is shown. About 300 values were calculated for each cell type for each indenter.

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