



Contents lists available at ScienceDirect

Biomaterials

journal homepage: www.elsevier.com/locate/biomaterials

Relationships among cell morphology, intrinsic cell stiffness and cell–substrate interactions

Martin Y.M. Chiang ^{a,*,1}, Yanzi Yangben ^{b,1}, Nancy J. Lin ^{a,1}, Julia L. Zhong ^{b,1}, Li Yang ^{b,*,1}

^a Biosystems and Biomaterials Division, National Institute of Standards and Technology (NIST), Gaithersburg, MD 20899, USA²

^b College of Bioengineering, Chongqing University, Chongqing, China

ARTICLE INFO

Article history:

Received 8 August 2013

Accepted 4 September 2013

Available online xxx

Keywords:

Cell modulus

Cell morphology

Substrate rigidity

Roundness

Cell–material interaction

Free energy

ABSTRACT

Cell modulus (stiffness) is a critical cell property that is important in normal cell functions and increasingly associated with disease states, yet most methods to characterize modulus may skew results. Here we show strong evidence indicating that the fundamental nature of free energies associated with cell/substrate interactions regulates adherent cell morphology and can be used to deduce cell modulus. These results are based on a mathematical model of biophysics and confirmed by the measured morphology of normal and cancerous liver cells adhered on a substrate. Cells select their final morphology by minimizing the total free energy in the cell/substrate system. The key mechanism by which substrate stiffness influences cell morphology is the energy tradeoff between the stabilizing influence of the cell–substrate interfacial adhesive energy and the destabilizing influence of the total elastic energies in the system. Using these findings, we establish a noninvasive methodology to determine the intrinsic modulus of cells by observing global changes in cell morphology in response to substrate stiffness. We also highlight the importance of selecting a relevant morphological index, cell roundness, that reflects the interchange between forms of energy governing cell morphology. Thus, cell–substrate interactions can be rationalized by the underlying biophysics, and cell modulus is easily measured.

Published by Elsevier Ltd.

1. Introduction

The impact of cell modulus (the deformability of cells or resistance to morphological change) extends beyond knowledge of a mechanical property to include cellular processes important in developmental biology, pathology, molecular biology, etc., as well as cell–material interactions in tissue engineering and regenerative medicine. For instance, cell modulus also affects many cell functions [1–3], correlates with disease states [4,5], has potential as a biomarker to distinguish normal and cancerous cells, and corresponds with embryonic stem cell fate [6,7]. Smooth muscle cell stiffness has been recognized recently to contribute to aortic stiffening, which relates to a host of aging processes and vascular diseases, including hypertension, atherosclerosis, and aortic

aneurysms [8]. Glial cell stiffness has also been suggested as both a mechanism responsible for rigid glial scars that impair neuro-regeneration after spinal cord injury as well as a target for regeneration therapies [9]. Likewise, tumor cell modulus corresponds with metastatic potential, and biochemical means to alter the modulus can change that metastatic potential [10], leading to the proposal of cell modulus as a “mechanical signature” of cancer cells and potentially a new biomarker of malignancy to aid in diagnosis and treatment of cancer [11]. In addition, controlling cell modulus is one potential approach to direct stem cells into the desired lineage for regenerative medicine applications, where a key challenge is optimizing mechanical and structural properties of scaffolds (substrates) to promote the desired tissue regeneration [12].

Our objective is to provide a noninvasive measurement of intrinsic cell modulus based on fundamental free energy concepts in cell–substrate interactions. Currently available methods to measure cell modulus typically involve either physical perturbation of cells adherent to two-dimensional substrates [11,13,14], where mechanical contributions from the substrate can be difficult to delineate from cell properties (e.g., atomic force

* Corresponding authors.

E-mail addresses: martin.chiang@nist.gov (M.Y.M. Chiang), yangli@cqu.edu.cn (L. Yang).

¹ Authors contributed equally in this work.

² Official contribution of the National Institute of Standards and Technology; not subject to copyright in the United States.

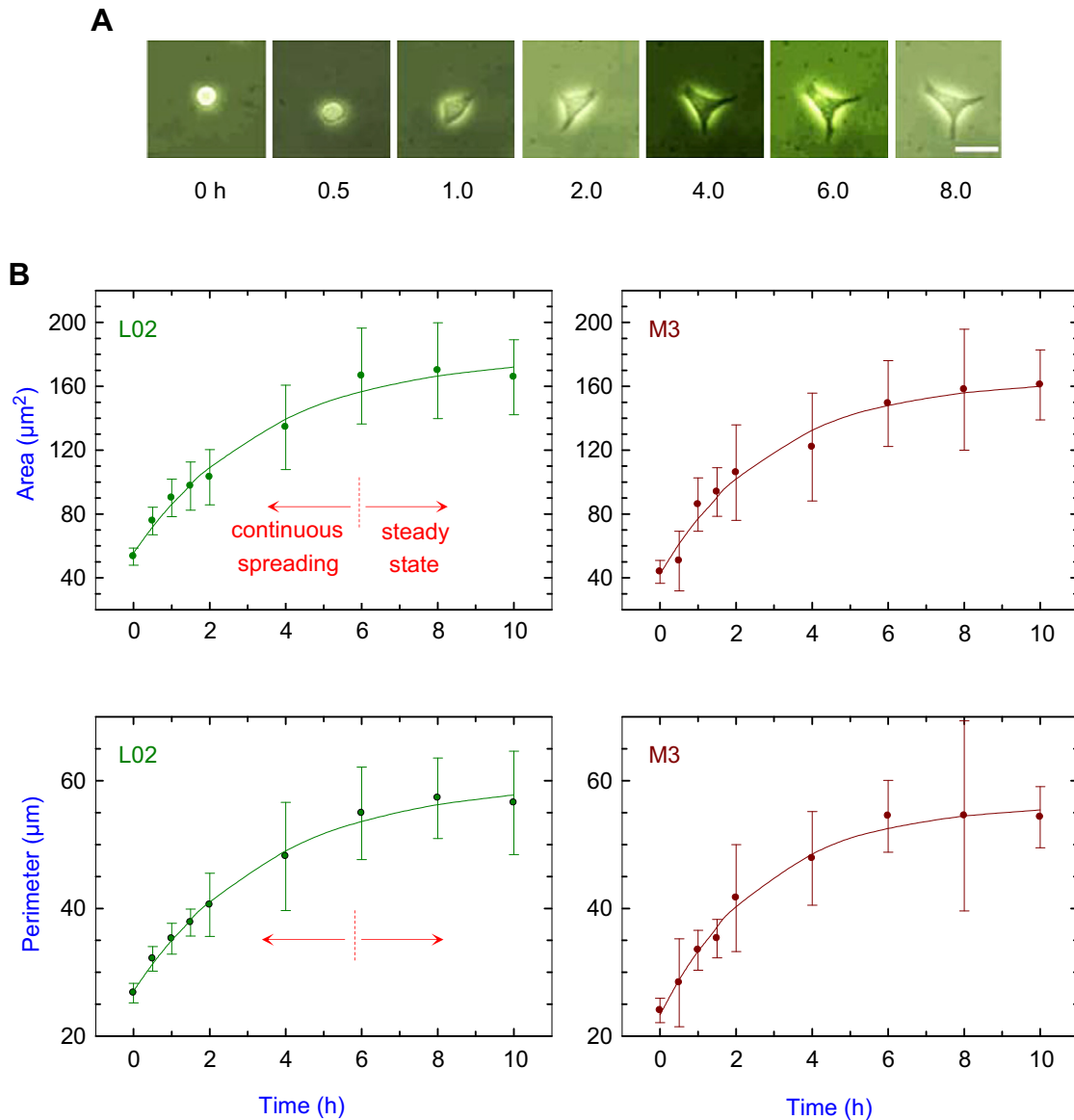


Fig. 1. The process of cell spreading. A, Typical microphotographs of M3 cell spreading obtained at different time points up to 10 h. Scale = 20 μm . B, Cell area and perimeter obtained as a function of time for normal (L02) and cancerous (M3) liver cells attached to glass substrates coated with fibronectin (2.5 $\mu\text{g}/\text{mL}$). In general, cell spreading exhibits three regimes (three-phases) starting with an initial basal growth regime for the nucleation of adhesion sites (not recorded for the plots). This basal phase is followed by a phase of fast continuous spreading, a non-steady process where cells constantly move and modulate their shapes. Finally, cell spreading slows down and effectively reaches a steady state. Cells were imaged for 10 h using inverted phase contrast microscopy. Each data point represents the mean value from at least 30 cells, and each error bar is one standard deviation and represents the standard uncertainty. Lines are drawn to aid the reader's eyes.

microscopy (AFM), poking, magnetic twisting cytometry, micro-pipette aspiration), or qualitative evaluation of deformability via filtration processes [15]. These measurements provide a relative or apparent cell modulus that depends on the measurement technique rather than an intrinsic modulus, which is a fundamental property of the cell that is independent of cell adhesion and assembled cytoskeletal networks of adherent cells. Our interest here is not to compare the relative merits of existing methods but to establish a method to quantify intrinsic cell modulus based on cell-substrate interactions. The effect of deformable substrates on cells is an active research area in cell mechanics [16–20]. *In vitro* studies of cells on substrates with different moduli indicate that cells sense substrate modulus and respond by changing their cell morphology (shape and size)

[14,21]. This morphological change is accomplished through assembly/disassembly of focal adhesions and used by cells to regulate physiological processes [22–25]. In this investigation, we show that the underlying physics of free energy in the cell/substrate system can describe how substrate modulus is translated into cell morphology, and that morphological stability of cells is dictated by the minimum total free energy in the system provided that the surrounding chemical environment remains constant. The combination of mathematical modeling and cell measurements reveals a new method to obtain intrinsic cell modulus using the variation of cell morphology in response to substrate stiffness. Moduli of normal and cancerous cells are quantified via our morphology analysis and compared with conventional AFM measurements.

Download English Version:

<https://daneshyari.com/en/article/10228087>

Download Persian Version:

<https://daneshyari.com/article/10228087>

[Daneshyari.com](https://daneshyari.com)