ELASTIC INTERACTIONS OF BIOLOGICAL CELLS

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Abstract   We review recent theoretical work that analyzes experimental measurements of elastic interactions of biological cells with their environment. Recent experiments have shown that adhering cells exert polarized forces on substrates. The interactions of such "force dipoles" in either bulk gels or on surfaces can be used to predict the nature of self-assembly of cell aggregates and may be important in the formation of artificial tissues. Cell adhesion strongly depends on the forces exerted on the adhesion sites by the tension of the cytoskeleton. The size and shape of the adhesion regions is strongly modified as the tension is varied and we present an elastic model that relates this tension to deformations that induce the recruitment of new molecules to the adhesion region.

Keywords: Cell elasticity, cytoskeleton, adhesion

1.   Introduction

In this review, we summarize recent progress in our understanding of the physics of the interaction of biological cells on elastic substrates and in bulk gels. Cell adhesion is quite different from adhesion of fluid-filled vesicles because the interior of the cell is an elastic medium (that

is continuously and actively reorganized) due to the presence of the cytoskeleton. In most cells, the cytoskeleton [1] is composed of several components, including actin, microtubules and intermediate filaments, each of which have different elastic properties [2] that can be used by the cell in a variety of circumstances. Force generation that leads to tension in the actin network arises from the action of myosin bundles that are activated by ATP to change conformation and exert forces on the actin filaments.

Recent experiments [3, 4] show that, in contrast to artificial vesicles that exert only normal forces when they adhere to a substrate, adhering cells show both normal and lateral forces. The normal forces arise from the action of either specific adhesion molecules or non-specific interactions (*e.g.* van der Waals interactions) while the lateral forces arise from elastic deformations of the adhesion region by cytoskeletal forces. These lateral forces regulate the size and shape of the adhesion regions (often called focal adhesions (FA)) [4] and allow a cell to probe and to adjust the strength of its adhesion to its physical environment. The physical origin of the mechanosensor in the cell and the conversion of elastic information to a biochemical response that recruits additional proteins to the adhesion region is an important problem in cell biology today.

These forces that arise from the tension in the actin cytoskeleton tend to polarize the actin filaments (also called stress fibers). Thus, one can sum over all the local focal adhesions and in a coarse-grained picture, model such an adhering cell as a pair of nearly equal and oppositely directed contraction forces (termed an elastic force dipole) with typical forces of 100 nN over a scale of tens of microns. An interesting physics problem [5, 6] concerns the interactions and self-assembly of many such force dipoles; this corresponds to the interactions of many cells, each of which adheres to an elastic medium such as a substrate or in a three-dimensional gel. These dipoles interact via the elastic deformations of the medium, and can form chains or other self-assembled structures. Because these interactions are long-range, the details depend on the boundary conditions and sample shape. In addition, the interaction strength depends on the elasticity of the substrate as well as on its deformations and cells have been observed to migrate towards stiffer substrates and to rotate on elastically strained media [7]. The physics of cell adhesion and the interactions of cells in elastic media are important for the understanding of tissue formation and engineering [8] as well as wound-healing and metastasis.

2. Elastic Effects in Cell Adhesion

Adhesion of live cells to external surfaces [9] plays an important role in many cellular processes, such as cell growth, differentiation, motility and apoptosis (programmed cell death) [10]. Cell adhesion is not a passive process, restricted to the formation of bonds between membrane receptors and extracellular ligands. Adhering cells actively probe the physical properties of the extracellular matrix; their cellular contractile machinery participates in the formation of the adhesive junctions. It has been shown that rigid surfaces give rise to large and stable adhesions, termed focal adhesions (FA), that are associated with the termini of actin stress fibers and trigger signaling activity that affects gene expression, cell proliferation, and cell survival. On the other hand, soft surfaces mainly support the development of relatively small, transient dot-like or fibrillar adhesions that are involved respectively in cell motility and matrix reorganization. In addition, observation of the early phase in the assembly of FA shows small, primordial adhesions, termed focal complexes (FX) as precursors of FA. FX are formed close to the edge of the advancing membrane protrusions of cells and can grow in some cases into FA when subjected to mechanical stress due to either cell contractility or external perturbations [11]. Indeed, the adhesion process has been shown to be mechanosensitive; cells can probe the physical properties of their environment and respond by modulating their adhesions or migratory activity [6, 12, 13].
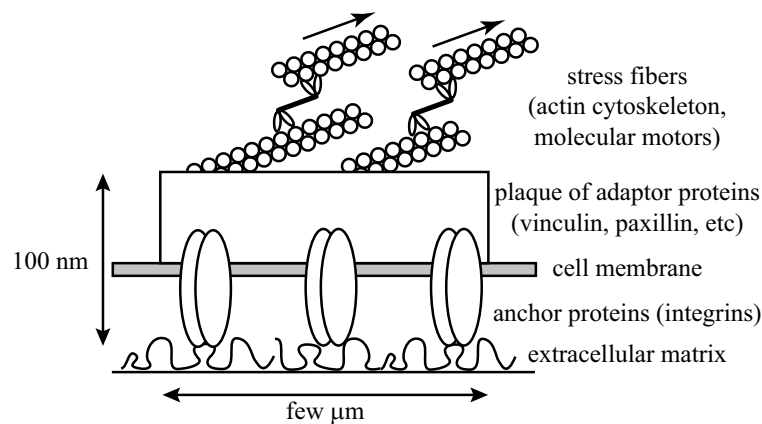
Figure 1. Schematic representation of a focal adhesion. The bottom of the contact is anchored to the extracellular matrix and the top surface is acted upon by the cytoskeletal tension. (Reprinted with permission from [27], Copyright (2004), American Physical Society).

Live cells exert directional, lateral forces on adhesive junctions; these forces originate from the organization of actin filaments and their associated molecular motors into stress fibers (Fig. 1). Adhesions respond dynamically to the local stresses: increased contractility leads locally to larger adhesions, whereas FA are disrupted when myosin II is inhibited [14]. The physics challenge in the understanding of these effects lies in (i) quantifying on a coarse-grained scale, the forces that cells exert on surfaces or in the bulk (ii) understanding the implication of these forces for adhesion of a single cell, the interactions among many cells in an elastic medium and the consequent self-assembled structures that may form (iii) predicting the microscopic origin of these forces and why adhesion growth is sensitive to the magnitude and direction of the internal or external cytoskeletal stresses.

Lateral Forces at Adhesion Sites

Biological cells can exert strong physical forces on their surroundings. One example are fibroblasts, that are mechanically active cells found in connective tissue. The main technique to measure cellular forces is the elastic substrate method [15] which was introduced by Harris and coworkers in the early 1980s [16]. Quantitative analysis of elastic substrate data was pioneered by Dembo and coworkers [17].

Recently, a novel elastic substrate technique to measure cellular forces at the level of single FA [4] was developed using a micropatterned, thick polymer film. From the deformations of the grid on the film surface, the forces exerted by FA were estimated. Correlation of these forces with the lateral size of the FA showed that there exists a linear relationship between force, F, and area, A, of a single FA. This finding translates into a force of several pN per receptor, which is consistent with recent experiments on strength of single molecular bonds at slow loading [18].

Since the adhering cells are rather flat, forces exerted on the substrate can be considered to be tangential to the plane of the substrate surface. Also, because surface displacements are much smaller than the film thickness, one can use linear elasticity theory for an elastic isotropic halfspace. For incompressible elastic substrates, the resulting displacement of the substrate surface remains within the plane of the substrate surface [19]. Therefore, the whole elastic problem becomes two-dimensional. Since lateral deformations of the substrate are not observed in the absence of FA, we assume that forces are exerted mainly at the sites of FA. For a single, discrete force density (force per unit volume) $\vec{f}$ at position $\vec{r}$, the displacement field $\vec{u}$ at position $\vec{r}$ is:

$$u_i(\vec{r}) = \int d\vec{r}' \; G_{ij}(\vec{r} - \vec{r}') f_j(\vec{r}'), \tag{1}$$

where the subscripts refer to the vector components and where G_{ij} is the Green function of the elastic isotropic halfspace, that is, a tensor of rank two:

$$G_{ij}(\vec{r}) = \frac{3}{4\pi E r}\left(\delta_{ij} + \frac{r_i r_j}{r^2}\right). \tag{2}$$

We use the convention of summation over repeated indices, δ_{ij} denotes Kronecker Delta and E is the Young modulus. The Poisson ratio has been set to 0.5 and the indices i, j take the values 1,2. Note that in finite systems, the Green function $G_{ij}(\vec{r} - \vec{r}')$ will be an explicit function of both $\vec{r}$ and $\vec{r}'$. This is because the elastic forces can be long range and thus depend on the boundary conditions. In the case of the half-space, the displacements are mostly in the plane; there are negligible normal displacements and the translational symmetry is approximately restored.

Since linear elasticity theory allows for superposition, one can generalize the above to treat a set of discrete forces that act at the sites of the observed focal adhesions. One must solve the inverse problem of predicting the forces, given the observed diplacement. The resulting force pattern turns out to be very sensitive to small changes in the displacement data. To regularize the problem [20], a reasonable constraint is that the forces should not be exceedingly large.

The combination of the experimental and computational method described above allowed, for the first time, a measurement of the cellular force at the level of FA and its correlation with aggregation characteristics [4]. One finds a linear relationship between magnitude of force and the area of the FA. The direction of force usually coincided with the major axis of the elliptically shaped FA. The observed force is proportional to the area of the FA, with a stress constant of $5.5\,\mathrm{nN}/\mu\mathrm{m}^2$. Since several thousand integrin adhesion molecules and myosin II molecular motors correspond to one FA, the pN scale of single molecules is amplified to the nN-range at FA.

Elastic Interactions of Cells: Force Dipoles

Anchorage-dependent cells constantly assemble and disassemble focal adhesions, thereby probing the mechanical properties of their environment. The protein aggregates in FA are connected to the actin cytoskeleton. This generates a contractile force pattern, that is actively produced by myosin II molecular motors interacting with the actin cytoskeleton. The minimal configuration of this machinery is a set of two focal adhesions connected by one bundle of actin filaments (*stress fiber*), that

leads to a pinch-like force pattern. In condensed matter physics, such an object is known as an *anisotropic force contraction dipole* [21, 22].

By analogy with electric dipoles, a force dipole, P_{ij} is a second rank tensor defined as the second moment of a spatially distributed force density (force per unit volume), $\vec{f}(\vec{r})$

$$P_{ij} = \int d\vec{s} \; s_i f_j(\vec{s}), \tag{3}$$

where the subscripts refer to the vector components. The elastic deformation field, $\vec{u}(\vec{r})$, is related to the force distribution and to the Green function as given in Eqs. (1,2) above. If the force distributions are described in terms of force dipoles, the displacement field at point $\vec{r}$ due to a dipole at point $\vec{r}'$ can be written [6]

$$u_i(\vec{r}) = G_{il,k'}(\vec{r}, \vec{r}') \; P'_{kl}, \tag{4}$$

where the extra index after the comma, denotes a derivative of the Green function with respect to the variable $\vec{r}$; a prime on this index indicates that the derivative is with respect to the variable $\vec{r}'$. As before, there is an implied summation over repeated indices. The prime on P indicates that the dipole is located at position $\vec{r}'$. The interaction of two dipoles proceeds via the deformation of the medium; each dipole feels the deformations induced by the other and this creates an effective interaction between them. For dipoles located at $\vec{r}$ and $\vec{r}'$ one can derive the effective interaction due to the elastic deformation energy of the medium. This energy is proportional to the elastic constant tensor and the product of the gradients of the displacements, $\vec{u}(\vec{r})$ that are related to the forces by Eqs. (1,2). One finds [6] that the elastic deformation of the medium results in a contribution to the elastic energy, W:

$$W = P_{li} \; u_{i,l} = P_{li} \; G_{ij,lk'}(\vec{r}, \vec{r}') \; P'_{kj}, \tag{5}$$

where P_{li} and P'_{kj} are the force dipoles located at $\vec{r}$ and $\vec{r}'$ respectively, $u_{i,l}$ is the derivative of the displacement component u_i with respect to component l of the position vector $\vec{r}$; the last two subscripts in the Green function represent derivatives with respect to $\vec{r}$ and $\vec{r}'$ respectively. For translationally invariant systems, such as an elastic half space with planar displacements,

$$G_{ij,lk'}(\vec{r}, \vec{r}') = G_{ij,lk'}(\vec{r} - \vec{r}') = -G_{ij,lk}(\vec{r} - \vec{r}'). \tag{6}$$

This interaction energy scales as $1/|\vec{R}|^3$ where $\vec{R}$ is the distance between dipoles and is proportional to P^2/E where P is the dipole strength and E is the Young's modulus of the medium. Equation (5) is not the only

relevant energy to be considered since in the case of inert matter one must add to the deformation energy of the medium, an energy proportional to the product of the local force and the local displacement; this accounts for the direct interaction of the dipole with the environment. The elastic theory can also describe [6] the interaction of a single dipole with an external strain imposed on the elastic medium or with the boundary of a finite-sized sample.

Recently, we have extended the concept of force dipoles to model the mechanical activity of cells [5]. Cells in an isotropic environment often show isotropic (that is round or stellate) morphologies. However, since the focal adhesion dynamics is local, even in this case there is an anisotropic probing process, that can be modeled by anisotropic force *contraction* dipoles. The anisotropy of focal adhesion dynamics becomes apparent when stress fibers begin to orient in one preferential direction, either spontaneously during a period of large mechanical activity, or in response to some external anisotropy, or during cell locomotion. In this case, cellular dipoles have been measured to be of the order of $P \approx -10^{-11} J$ (this corresponds to two forces of $200 \, nN$ each, separated by a distance of $60 \, \mu m$) [4, 23]. In Fig. 2 we show schematic representations of the physical and cellular cases discussed here.

For inert, physical dipoles, the equilibrium configuration follows by minimizing the sum of the elastic energy of the strained medium and the direct interaction energy between the force dipole and elastic environment. The first term represents a restoring force and raises the energy (*i.e.*, its sign is always positive), while the second term is a driving force that reduces the total energy (*i.e.*, its contribution will always be negative). The total interaction between two dipoles has the form of Eq. (5) but with a negative sign due to the effect of the local force-displacement interaction.

The equilibrium configuration corresponds to the minimum of the total energy as a function of position and orientation of the force dipoles, which results in an effective, so-called *elastic interaction* between the force dipole and other dipoles, sample boundaries or external strain fields. We have predicted that the competition between direct and image interactions should lead to hierarchical structure formation, with the direct interaction leading to structure formation on a length scale set by the elastic constants and similar to that of electric quadrupoles [5]. We suggested that such a behaviour should be expected for artificial or inert cells, that is for physical particles with a static force contraction dipole, but without any internal dynamic or regulatory response.

In contrast to this physical case, the effective behavior of active cells usually follows from dynamic and tightly regulated non-equilibrium pro-

(a)   (b)   (c)
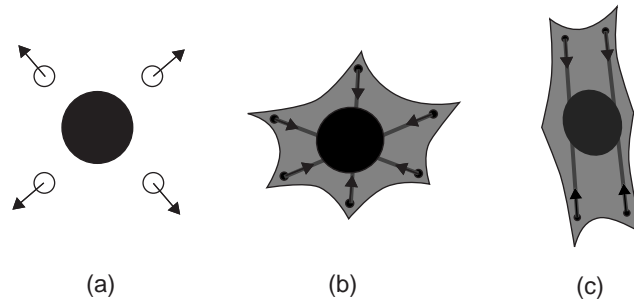
Figure 2.   Schematic representation of physical and cellular force dipoles. (a) Physical case: an intercalated defect deforms the simple cubic host lattice, thus acting as an isotropic force expansion dipole. (b) Cellular case: anchorage-dependent cells probe the mechanical properties of the soft environment through their contractile machinery. Actin stress fibers (lines) are contracted by myosin II molecular motors and are connected to the environment through focal adhesions (dots). Even if cell morphology is round or stellate, different stress fibers probe different directions of space and compete with each other for stabilization of the corresponding focal adhesions. Therefore the probing process can be modeled as an anisotropic force contraction dipole. (c) Cell morphology becomes elongated in response to anisotropic external stimuli, during locomotion or spontaneously during times of strong mechanical activity. Then most stress fibers run in parallel and the whole cell appears as an anisotropic force contraction dipole. (Reprinted with permission from [6], Copyright (2004), American Physical Society).

cesses inside the cell. More recently, we have shown that despite this severe complication, it is still possible to describe the active response of mechanosensing cells in an elastic material in the same framework as the physical case [6]. Motivated mainly by recent experiments with elastic substrates [24], we have suggested that effective cellular behavior can be described as simple preference for large effective stiffness in the environment (including both rigidity and tensile prestrain). Moreover, we have shown that this principle is equivalent to minimization of the energy cost of straining the environment. (But this does not include the energy due to the direct force-displacement interaction.) This results in an interaction energy between two force dipoles with a positive sign as given in Eq. (5). The direct elastic interaction between cells has been predicted to be similar to that of electric dipoles, leading to strings of cells [6]. This is not the case for inert dipoles (due to the difference in the sign of their interaction), where more compact structures are predicted [6].

The different energies and opposite sign of the interactions for the case of inert force dipoles and active cell force dipoles leads to interesting differences in their behavior. For example, active cells are attracted and repeled by clamped [25] and free sample boundaries, respectively. In the

case of inert dipoles, this behaviour is inverted. The predictions for the elastic response cells explain several experimental findings reported in the literature [6] and can be used for rational design of tissue equivalents.

3.   Theoretical Model for Mechanosensitivity of Adhesion

Cell Adhesion Mechanosensitivity

The coupling of the FA to the internal stresses that are regulated by the cell (*e.g.*, via actin-myosin activity or actin polymerization) means that the cell can respond to the elasticity of its adhesive environment. Recent experiments have shown that external forces can also cause the anisotropic growth of focal adhesions. Independent of the origin of the stress (e.g. internal contractility [4], shear flow [26], micropipette-induced shear stress [14]), small adhesions grow into focal adhesions (FA), that elongate in the direction of the force. The FA grows via a biochemical process that is very sensitive to the force applied to the adhesion region by the cytoskeleton and the origin of this mechanosensitivity is an important problem in cell biology.

Our model (see Fig. 1) assumes that the biochemical response of adhesions to cytoskeletal stresses originates from the stress-induced elastic deformation of the adhesion site. These force-induced deformations modify the local density of the proteins found in and around the adhesive junction, and thus their interactions. This, in turn, can trigger a conformational change or a molecular reorganization that initiates the biochemical cascade responsible for the aggregation of new proteins and the directional, anisotropic growth of the adhesion. For example, one could identify the mechanosensor with the layer of transmembrane, integrin proteins [11], since they are connected to the extracellular matrix and are elastically deformed by cytoskeletal forces; this deformation can activate conformational or organizational changes that enhance their binding with plaque proteins [10].

In addition to the fact that adhesion to an anchoring surface makes a junction sensitive to shear [27] (even if the latter is composed of fluid-like assemblies of molecules), several observations support the view we propose. First is the observation that independent of the origin of the stress, small adhesions grow into focal adhesions (FA), that elongate in the direction of the force. This anisotropy is predicted by our theory since stress-induced deformations are correlated with the applied force and exert different effects on the proximal (front) and the distal (back) aspects of the adhesive junctions. In our model, where we relate the biochemical response to deformations, this anisotropy also leads to an

anisotropic addition (or loss) of proteins. Moreover, the size and the strength of the adhesive junctions depend on the elastic properties of the extracellular matrix [28, 29]. Our model predicts that a local force applied to an adhesive junction induces a pure translation of the junction (and thus no deformation) if the extracellular matrix is very soft, whereas strongly grafted adhesions on a rigid matrix are elastically deformed by lateral, local stresses.

Description of the Model

The total force transmitted by FA has been shown to be proportional to their surface [4], suggesting that the region over which stress fibers act grows with the junction. As a model, we consider the adhesion and the membrane-associated integrins as an elastic, thin film grafted at its bottom surface and stressed by a local lateral force on its top surface; "local" means here that the force only acts on a finite region of the adhesive junction, which, as explained below, needs only to be somewhat smaller than the total area of the adhesion by a few thicknesses of the FA (typically, a few tens of nanometers).

The localization of the force is a crucial aspect of our model, since a homogeneously applied stress would give rise to a uniform shear of the adhesive junction and a translational motion of the surrounding lipid membrane. A *local*, tangential stress induces a compression at the front edge and an expansion at the back of the stressed region because the unstressed part is also grafted and cannot slide. In this case, a lateral force induces anisotropic density changes in proteins located inside or close to the stressed region. The resulting stress-induced deformation profile can be calculated using continuum elasticity theory [27]. Our model assumes that this change in density triggers the biochemical response responsible for the growth or dissolution of FA. Anisotropic variations of density thus give rise to different molecular dynamics in the front and back regions near the edge and can result in either overall growth or shrinking of the FA as shown below.

We first consider a quasi-two-dimensional model of FA in which the elastic thin film is represented by particles that interact via springs of stiffness k_0 (see Fig. 3). The particles are bound to the anchoring surface by springs of stiffness k_b, that measure the resistance to normal shear. Each particle in our model is a protein complex consisting of one integrin molecule and the associated intracellular proteins that connect it to the actin cytoskeleton. The elastic properties of the extracellular matrix and its connection to the anchoring surface are included by fixing the anchors to the surface via a sinusoidal potential which, in the limit of
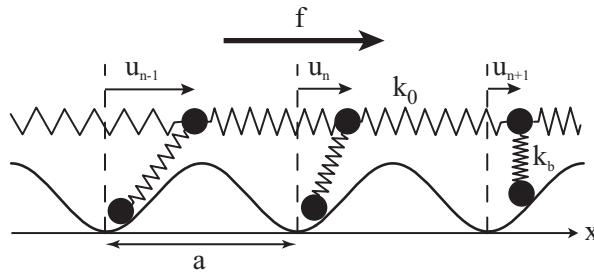
Figure 3.   One dimensional chain of interacting particles bound to anchors, as a model for focal adhesions grafted to an extracellular matrix. The potential $V(x)$ accounts for the grafting properties of the extracellular matrix. The distance a between the anchors is the integrin-integrin spacing and u_n is the displacement of the nth particle from its position in absence of force. (From Ref. [30], Copyright (2004), National Academy of Sciences, USA)

small displacements, reduces to a spring-like interaction where k_m is the stiffness of the anchorage close to the equilibrium position. The case of a grafted layer is found by taking the limit $k_m \to \infty$.

Predictions of the Model

In a one dimensional version of this approach (Fig. 3, which can easily be generalized to a two-dimensional layer), the mechanical equilibrium for a such a system of particles pulled by a force $F(x)$ is, in the continuum limit:

$$k_0 a^2 \frac{d^2 u}{dx^2} - k_b u + F(x) = 0, \qquad (7)$$

where k_0 is the spring constant connecting the particles and k_b accounts for the restoring force due to the connections to the fixed and rigid extracellular matrix; the integrin-integrin spacing is denoted by a. We solve Eq. (7) for a gate-shaped field of force of amplitude, f, per unit length and width L; that is, the force is only non-zero for $-L/2 < x < L/2$. This force profile corresponds to the local pulling force due to stress fibers, and induces the following spatial variation of the local, average density, $\delta\Phi(x)$, relative to the undeformed density Φ:

$$
\begin{aligned}
\frac{\delta\Phi}{\Phi}(x) &= -\frac{du}{dx} \\
&= \frac{f}{\sqrt{k_b \, k_0}} \begin{cases} \sinh \frac{x}{\ell} \, e^{-L/2\ell} & \text{if} \quad |x| < L/2 \\ \sinh \frac{L}{2\ell} \, e^{-|x|/\ell} \, \text{sign(x)} & \text{if} \quad |x| \geq L/2. \end{cases} \quad (8)
\end{aligned}
$$

Equation (8) is derived in the case of a stiff matrix ($k_m \rightarrow \infty$) and shows that the deformation decreases exponentially with a range $\ell = a\sqrt{\frac{k_0}{k_b}}$. Remembering that an actual adhesion has a finite thickness fixes the order of magnitude of ℓ: force-induced deformations must vanish at the bottom surface due to the grafting boundary condition appropriate to the case of a immobilized extracellular matrix. The range ℓ is thus of order of the thickness of the junction (~ 100 nm). The representation of the adhesion as an infinite elastic medium pulled by a force acting on a region of size L is therefore valid once the actual size of the adhesion is larger than $L + 2\ell$, where $\ell \sim 100$ nm $\ll L$.

The three dimensional case is treated more rigorously in [27], using a full elastic treatment of a thin film subject to a surface force, F, on its top surface with zero displacement boundary conditions at the bottom surface where the film is grafted to the substrate. The density, $\delta\Phi$ at the top surface is anisotropic and varies like,

$$\delta\Phi \sim \frac{F}{E}\frac{h}{\ell}\frac{e^{-r/\ell}}{\sqrt{r/\ell}}\cos\phi, \qquad (9)$$

where ϕ is the azimuthal angle, E is the Young's modulus of the film, h is the film thickness, r is the distance from the applied force, and as above, ℓ is proportional to the thickness and is a function of the elastic constants of the film [27]. This predicts a highly anisotropic response to the force in two dimensions and is consistent with the observation that the FA only grow in the direction of the applied force. The variation of density due to a local stress is only significant close (within a distance ℓ) to the edges of the stressed area. The predicted anisotropy means that changes in density are therefore much smaller on the sides of the stressed area perpendicular to the force, in accord with observations [14].

Growth of Focal Adhesions

The main hypothesis of our model is that force-induced deformations trigger the biochemical response of FA, via conformational (e.g. activation of integrins) and/or organizational changes (e.g. binding of plaque proteins) of molecules embedded in a narrow region of size ℓ where the density varies. This local activation increases the rate of association of free proteins within this area. The competition of the elastic and biochemical energies determine whether additional proteins will join the FA. Within our model, the addition of a protein complex of cross-sectional area a^2 to the FA results in a deformation with an elastic energy cost, ΔE_{el} proportional to $\frac{f^2 a^2}{2k}$ where k (which replaces k_b defined above) is an effective elastic constant that includes the finite stiffness of the extra-

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cellular matrix. We now suppose that the association of a free protein complex to the front edge of the adhesion releases an energy, ΔE_a, that depends on the change in density at the front edge of the stressed region. In the absence of density variations, the probability of attaching a new particle is unchanged, and $\Delta E_a = 0$. For small variations of the density, ΔE_a can be taken to vary linearly with the change in density at the edge. This energy also scales with the free energy of reaction of one protein complex, e. Adding a protein complex to an adhesive junction thus involves an overall variation of free energy:

$$\Delta E = \frac{f^2\, a^2}{2k} - e\,\frac{f}{\sqrt{k\, k_0}}. \qquad (10)$$

When $e < 0$, aggregation of new particles to an existing adhesion is thermodynamically unfavorable ($\Delta E > 0$), and an additional input of energy is required to stabilize the protein cluster. For this thermodynamically unfavorable growth process, increasing the force per unit length, f, leads to smaller adhesions. FA can only grow when the overall free energy of Eq. (10) is negative, corresponding to $e > 0$ – *i.e.* favorable aggregation energy and an exothermic, local chemical interaction. However immobilization of the extracellular matrix ($k \to 0$) or very large forces can lead to positive value for ΔE, and, if large enough, may arrest the growth process, even if $e > 0$. This analysis can be generalized to predict the kinetics of growth of FA [30].

Acknowledgments

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