REVIEW

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Dynamic molecular processes mediate cellular mechanotransduction

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Cellular responses to mechanical forces are crucial in embryonic development and adult physiology, and are involved in numerous diseases, including atherosclerosis, hypertension, osteoporosis, muscular dystrophy, myopathies and cancer. These responses are mediated by load-bearing subcellular structures, such as the plasma membrane, cell–adhesion complexes and the cytoskeleton. Recent work has demonstrated that these structures are dynamic, undergoing assembly, disassembly and movement, even when ostensibly stable. An emerging insight is that transduction of forces into biochemical signals occurs within the context of these processes. This framework helps to explain how forces of varying strengths or dynamic characteristics regulate distinct signalling pathways.

There is growing recognition that mechanical factors, such as applied forces or the rigidity of the extracellular matrix (ECM), crucially influence the form and function of cells and organisms[1–3]. Biological regulation has classically been understood through the concepts of solution chemistry, in which enzyme activities, reaction rates and affinities govern cellular processes. However, mechanotransduction, the conversion of mechanical forces into biochemically relevant information, contributes to numerous developmental, physiological and pathological processes and is a rapidly advancing area of current research[1].

In the vasculature, blood flow exerts fluid shear stresses on the endothelial cells lining the vessels, whereas blood pressure stretches the vessel wall[4]. Shear stress is crucial for remodelling the primitive vascular plexus into a hierarchical vascular tree[5] and for patterning the cardiac outflow tract in developing mouse embryos[6]. Hypertension causes thickening of the arterial walls and is a major risk factor for cardiovascular diseases[4]. Atherosclerosis, the chronic cholesterol-dependent inflammation of artery walls, occurs preferentially at regions of disturbed flow such as branch points and areas of high curvature, where both the magnitude and temporal characteristics of the flow are disturbed. The development and pathobiology of bone and muscle also strongly depend on mechanical forces from weight and muscle contraction, whereas lung physiology and pathology are strongly influenced by forces from inflation[1].

Tissue rigidity or stiffness affects many biological processes[7]. Tumours have long been identified by palpation, owing to local increases in tissue stiffness. Recently, these changes in the mechanical environment have been shown to be causal for tumour progression[8]. Fibrotic lung disease begins with a small change in tissue stiffness, which is sensed by cells, inducing more severe, irreversible remodelling[9]. Furthermore, the rigidity of the extracellular environment potently controls the differentiation of mesenchymal stem cells[10] and the self-renewal of haematopoietic stem cells[11]. Developing scaffolds with tunable mechanical properties to control cellular behaviour has become a major effort in tissue engineering[12].

Although applying forces to cells and altering the rigidity of their environment are clearly distinct processes, the underlying mechanisms of mechanotransduction seem to be similar[2]. A key event in rigidity sensing is the modulation of cellular contractility. Cells on soft materials exert lower forces than cells on stiff materials, decreasing tension on force-bearing elements. These elements are the same whether forces are generated internally or externally[7]; thus, many of the cellular responses to distinct mechanical stimuli are similar. Another unifying principle is that the structures that generate and bear cellular forces are involved in sensing forces[1]. Therefore, cytoskeletal proteins such as actin and tubulin are crucial for mediating mechanical effects in nearly all systems[2,13] (Fig. 1a, b). Cellular adhesions, both to the ECM and to other cells, are also important, as they mechanically connect cells to their surroundings. Correspondingly, many of the candidate genes associated with diseases that can be considered 'mechanotransduction disorders' — such as aortic aneurism, heart failure, hypertension and muscular dystrophy — encode proteins involved in adhesion complexes, the cytoskeleton and the ECM[14–17]. There are often drastic changes in the protein composition, dynamics and mechanics of these structures during metastatic progression[18] and stem-cell differentiation[7].

Although much progress has been made towards understanding mechanotransduction, a complete picture is lacking. Mechanotransduction is typically depicted as a series of rapid switch-like events, activated in response to step-like applications of force, which eventually lead to cellular responses. This level of detail, however, is insufficient to explain the cellular responses to dynamic mechanical stimuli often found in physiological settings.

In this Review, we first outline the basic features of the switch-like model of mechanotransduction. We divide this process into mechanotransmission, mechanosensing and mechanoresponse, and then highlight the limitations of the model. We also describe recent advances in our understanding of the dynamic processes regulating load-bearing subcellular structures and the behaviour of single molecules in response to applied forces. With guidance from mathematical models of adhesion assembly, these examples are used to develop a more complete model of mechanotransduction based on the concept that forces alter the rates of key subcellular processes to affect cell function. This perspective allows us to understand how cells respond to time-varying mechanical stimuli. We end by suggesting that the cell may function mechanically as a multiband pass filter in which stimuli with different temporal characteristics activate distinct signalling pathways that affect cell state and disease progression.

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Switch-like models of mechanotransduction

Descriptions of mechanotransduction typically begin with the forces acting on cellular elements and end with the integrated response from the cell or tissue. These can be divided into three steps.

Mechanotransmission

A force must be transmitted to mechanosensitive elements before it can be sensed. For example, the adhesion receptors that mediate cell–cell and cell–ECM contacts are strongly implicated in mechanotransduction[19]. Equally important are the cytoskeletal structures that adhesion receptors universally connect to, which allow adhesions to resist deformation from applied forces. The cytoskeleton is composed of filaments, such as F-actin, intermediate filaments and microtubules, that are relatively stiff on the micrometre-dimensional scale and are stable on minute-to-hour timescales[20]. This mechanical continuity allows forces to propagate relatively long distances along filaments in the cell, a process known as mechanotransmission. Fluid shear stress, for instance, is exerted on the apical domain of endothelial cells; yet the displacements in vimentin filaments are largest at select areas, often near cell–cell and cell–ECM adhesions[21]. This result correlates with data implicating junctional proteins in responses to shear[22]. Twisting of magnetic beads bound to integrins also showed long-distance effects, which were primarily transmitted by F-actin[23], although some studies have proposed a role for microtubules[24] and intermediate filaments[25]. Using a laser trap to apply forces as small as 5.5 pN to actin stress fibres triggered an influx of calcium ions, presumably owing to the activation of mechanosensitive ion channels in the plasma membrane[26]. Cellular responses to force can also be extremely fast, of the order of hundreds of milliseconds[24,27], consistent with direct mechanical effects.

Mechanosensing

Transmitted forces ultimately impinge on mechanosensitive macromolecules to alter their conformation and hence their function. Although the biological consequences of such events are specific to each system, the underlying physical response is similar; forces promote changes in protein conformation that accommodate the applied force. The best studied examples at the structural level are bacterial mechanically gated ion channels[28], which open in response to increased lateral tension in the plasma membrane during osmotic swelling. Similar ion channels are present in all organisms and are essential for survival under changing osmotic conditions (Fig. 1c)[29].

There is also evidence that the unfolding of protein domains under tension mediates responses to applied forces. The first reported instance was fibronectin, which self-assembles into fibrils in the ECM. The formation of fibronectin fibrils requires cell-generated force[30]. Conversely, purified fibronectin undergoes self-association when stretched *in vitro*[31,32]; fibril assembly is mediated by the unfolding of domains revealing cryptic-binding sites. Another well-studied example is talin-1, which connects integrins to F-actin, thereby transmitting forces between actomyosin filaments and the ECM[33]. Talin-1 binds to vinculin, which also links to F-actin and is recruited to adhesions in response to applied forces. Curiously, many of the vinculin-binding sites on talin reside within the interior of bundles composed of four or five α-helices and are therefore inaccessible[33]. Both biochemical and cellular studies provide evidence that tension unfolds these bundles to expose vinculin-binding sites, thereby allowing vinculin recruitment[34,35] (Fig. 1c). Another protein in the cytoplasmic region of integrin-mediated adhesions is the adaptor protein $p130^{Cas}$ (also known as BCAR1). When phosphorylated on tyrosine residues by Src family kinases, $p130^{Cas}$ binds several guanine-nucleotide exchange factors (GEFs) that activate small GTPases[36]. Stretching cells enhances the phosphorylation of these tyrosines, leading to GEF binding and activation of Ras-related protein 1, widely known as Rap1 (refs 37–40). Studies with purified proteins have shown that stretching increases the susceptibility of $p130^{Cas}$ to phosphorylation, without changing the intrinsic activity of Src family kinases[40]. Although it is unclear how forces might be transmitted across

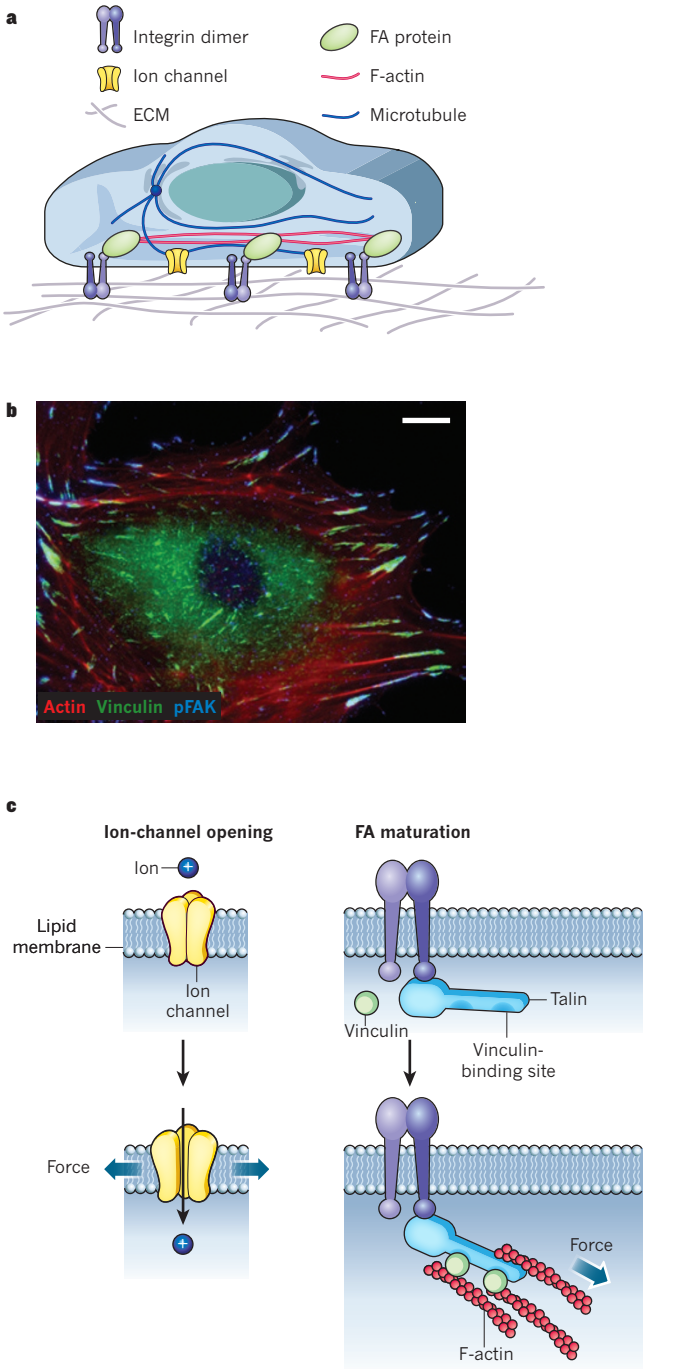


Figure 1 | Switch-like models of mechanotransduction. **a**, Cells are mechanically integrated structures, in which the ECM and actin cytoskeleton are connected by integrins and focal adhesion (FA) proteins. Microtubules and many ion channels are also integrated with this network. Forces can be applied directly through the ECM or transmitted through the cytoskeleton to mechanosensitive components, such as FAs, to mediate cellular response to forces. **b**, Immunostaining of a vascular smooth muscle cell. F-actin filaments (red) link to variably sized, punctate FAs, as shown by vinculin (green) and phosphorylated focal adhesion kinase (pFAK; blue) staining. The variable amount of pFAK staining in the FAs is indicative of different local signalling environments that are probably linked to distinct mechanical signals. Scale bar, 10 μm. **c**, A common mechanism of mechanotransduction is force-induced conformational change. For example, membrane tension can cause ion-channel opening. Also, talin connects the integrin cytoplasmic tail to F-actin; tension on talin exposes cryptic vinculin-binding sites, and the subsequent binding of vinculin (green) reinforces the linkage.

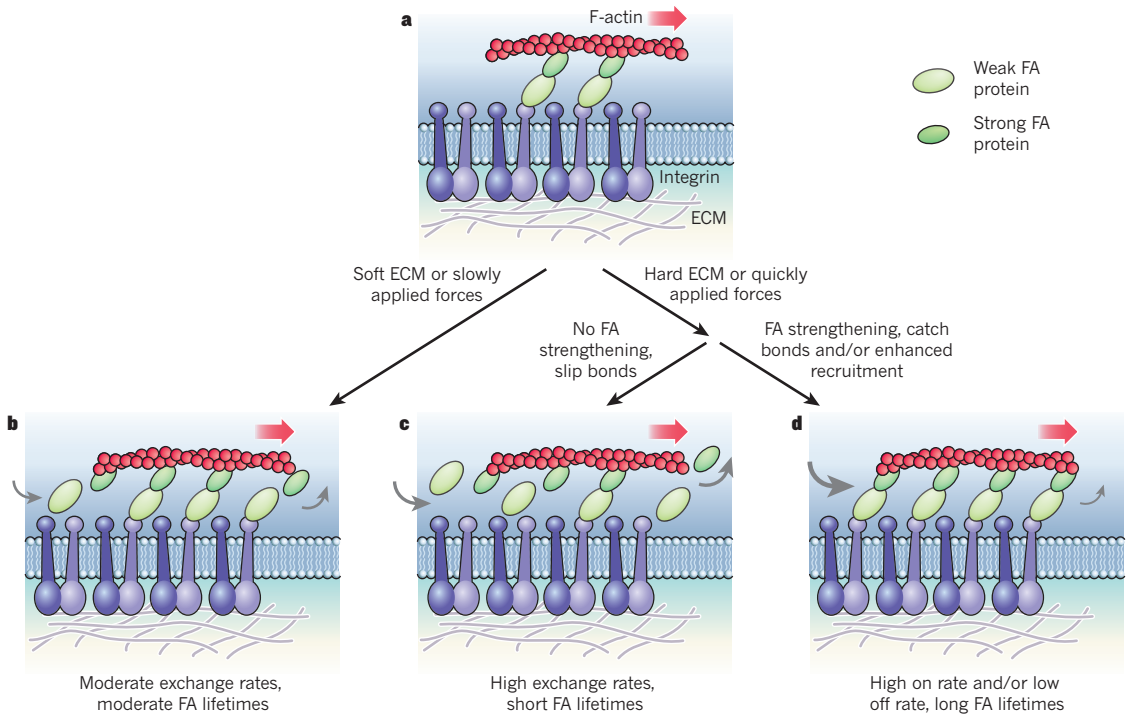


Figure 2 | The focal-adhesion clutch. **a**, Owing to forces from actin polymerization and myosin-dependent contractility, actin filaments flow backwards over FAs towards the nucleus. Through FA proteins that link actin to integrins, force is applied to the ECM. Force-dependent changes in FA exchange rates (arrows; see Box 1 for details) alter the dynamics and size of FAs. **b**, On soft surfaces or when external force is applied slowly, on rates and off rates are moderately high, and FAs have moderate lifetimes. **c**, **d**, By contrast, on stiff surfaces or when external forces are applied quickly, distinct behaviours can be observed. In the absence of FA strengthening (**c**), molecular-linker dissociation rates increase (slip-bond behaviour). These proteins are sometimes replaced through rebinding, resulting in large exchange rates and short-lived FAs. FA strengthening is, however, associated with catch bonds, which slow FA–protein dissociation under force (**d**). This exposes cryptic binding sites and recruits proteins that reinforce the adhesion, and causes conformational changes in FA proteins that activate signalling pathways to recruit other molecular linkers, resulting in large, long-lived FAs.

p130^{Cas} *in vivo*, these studies illustrate that forces can affect substrate availability through effects on protein conformation.

For physiologically significant mechanosensing, these initial conformational changes must be followed by a second step in which the new conformation triggers downstream events. This step can be fairly direct, as for the mechanosensitive ion channels discussed earlier. In other instances, changes in protein conformation, especially the opening of domains that contain cryptic sites, lead to the binding of proteins that mediate downstream events. This can result in reinforcement of the linkage, as in the case of talin and vinculin[34], or the recruitment and activation of signalling proteins, as in the case of p130^{Cas} (ref. 40). This general model is applicable to a wide range of mechanotransduction events in many systems[1]. Understanding in detail how protein domains change conformation under force and how subsequent events transpire has been the major direction in this field.

Mechanoresponse

Ultimately, sensed mechanical signals influence information processing through complex cellular signalling and transcriptional networks that are not specifically force dependent. In many cases, these responses feed back to alter the mechanosensitive structures that initiated the responses. Both integrin-mediated and cadherin-mediated adhesions enlarge and strengthen in response to tension[19]. Distinct from the very rapid, direct recruitment described earlier, signalling pathways that are activated over minutes (such as the small GTPase RhoA, which stimulates the formation of actin stress fibres[41]) and gene-expression pathways that operate over hours or days (such as the induction of vinculin through serum response factor[42,43]) change the composition and structure of adhesions and the cytoskeleton.

Similar principles apply at the tissue level. High blood pressure, for instance, results in the thickening of artery walls to bear the increased tension[4] and in hypertrophy of the left ventricle of the heart to allow stronger pumping against high back pressure[44]. Analogously, bone deposition increases under weight-bearing exercise[1]. These integrated responses depend on the intensity and time course of stimulation in ways that differ from the initial responses. For example, a single, brief interval of high blood pressure during exercise will stretch vessel walls and tax the heart but does not trigger compensatory arterial and cardiac remodelling, unlike sustained hypertension[44].

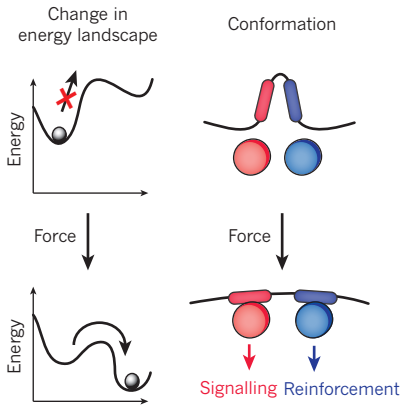
Limitations of switch-like models

In switch-like models of mechanotransduction, applied forces are instantaneously transmitted to load-bearing subcellular structures and induce conformational changes in mechanosensitive proteins. Different forces are sensed largely by conformational changes in protein domains that are stronger or weaker, and thus respond to forces of different magnitudes[45]. This view, however, seems to be incomplete. For example, the frequency of applied cyclic stretch or compression can have major effects. Steady stretch and cyclic stretch of equivalent magnitude induce distinct genes in endothelial cells[46] and induce differential phosphorylation of sites on focal adhesion kinase in rabbit aortas stretched *ex vivo*[47]. The frequency of applied cyclic stretch also determines endothelial alignment[48]. In aortic smooth muscle cells, stimulation of integrin activation and subsequent cellular alignment by cyclic stretch depends strongly on stretch duration and frequency[49].

These observations are particularly relevant in the vascular system, in which shear stress and circumferential stretch in arteries undergo strong, time-dependent variations during the cardiac cycle. Furthermore, these variations are different in various parts of the vasculature[50] and correlate with the location of atherosclerotic lesion formation[4]. Notably, recent evidence demonstrates that particular

BOX 1

Physics of force–activated bond dissociation



The non-covalent bonds that mediate protein–protein interactions have finite lifetimes, ranging from milliseconds to days[63]. Applied force typically shortens the lifetime of a bond (like applying force to remove tape). In a molecular context, these are referred to as slip bonds[63,64]. There are also molecules in which the bond lifetimes increase, although not infinitely so, in response to applied forces[64]. These are called catch bonds and, although relatively rare, are often found in cytoskeletal and cellular adhesion structures.

Protein conformational changes can be understood as a type of slip bond in which the dissociation is internal, owing to non-covalent bonds between amino acids in a single protein instead of between two proteins[63,65]. Moreover, both processes can be represented in terms of an energy landscape in which two energy minima are separated by a high energy state that slows the rate of transition. Applied force acts catalytically to accelerate rates by lowering the energy requirement for the transition and by changing the free energy of the states, typically stabilizing the more open conformation or unbound conformations. Thus, the likelihood of a conformational change depends on both the magnitude of the force and its duration[63]. These conformational changes mediate subsequent mechanotransduction events by exposing binding sites for signalling and cytoskeletal proteins (see Figure; boxes and circles denote binding sites and proteins, respectively; red denotes signalling proteins, and blue represents cytoskeletal proteins).

Furthermore, the bonds mediating protein conformations and protein–protein interactions tend to have similar affinities and force sensitivities. As protein dissociation will terminate the tension that induces conformational changes, these processes will in effect compete. This competition was recently studied using a single-molecule system that contained both a dissociable bond and a protein that could undergo force-dependent conformational opening[66]. Protein dissociation and conformational changes were both observed, but, interestingly, the frequency of conformational changes was enhanced at higher loading rates.

frequencies of mechanical stimulation preferentially activate the inflammatory pathways implicated in atherosclerosis, even when the total power applied to the cell is conserved (R. E. Feaver, B. D. Gelfand and B. R. Blackman, manuscript submitted). The characteristic time between the peaks in wall stretch and fluid shear stress may also crucially regulate endothelial activation[51]. These characteristics of mechanotransduction are not readily captured by switch-like models.

Subcellular structures are dynamic

These time-dependent aspects of mechanotransduction can be attributed to the highly dynamic characteristics of the cellular components that bear and respond to force. Cell adhesions to the ECM go through a complicated, force-sensitive maturation process[41]. Nascent cell–ECM adhesions are very small structures (<1 μm in diameter) at the edges of lamellipodia and usually disassemble within tens of seconds, or else mature into slightly larger focal complexes that persist for only a few minutes[41]. A fraction of these focal complexes mature into larger focal adhesions (FAs) that persist for tens of minutes. However, even within stable FAs, proteins are constantly exchanged, with lifetimes from tens of seconds to at most a few minutes[52]. Thus, even stationary FAs undergo rapid internal dynamics.

Detailed analyses from high-resolution techniques such as fluorescence speckle and correlation microscopy[53,54] have shown complex interactions between cytoskeletal and FA dynamics. Actin filaments constantly polymerize at the leading edge of the cell and flow backwards over the FAs, with the speed of this flow influenced by the nature of the linkages between the cytoskeleton, the integrins and the ECM. Actin flow is faster over areas with few FAs or in which the FAs undergo treadmilling towards the centre of the cell ('sliding FAs'), and slower in areas with stable FAs[53]. These and other results[55] led to the notion of a 'clutch' that controls force transmission between the flowing actin and the integrins (Fig. 2). Furthermore, in areas with stable FAs, integrins are immobile, and actin flows at 0.1–0.2 μm min^{-1}; however, different FA proteins have different velocities between these limits[53]. These results indicate the presence of many proteins that act as clutches, or force-sensitive linkages, within FAs.

Cadherin-dependent adhesions have not been studied in as much detail, but available evidence indicates similarities to FAs[2]. Several studies have shown that cell–cell contacts bear considerable forces[56,57] and, like FAs, they undergo dynamic, myosin-dependent elongation[58]. Applied forces[56] and stiffer substrata[59] enhance cell–cell contact assembly, indicating that these adhesions also undergo force-dependent adhesion strengthening. There is also evidence for actin flow along cell–cell contacts[60]. Although the other molecular players are different, vinculin is recruited to both structures in a myosin-dependent manner, thereby contributing to adhesion strengthening[61,62]. These data indicate that the dynamic properties of cell–cell contacts are regulated by processes physically similar to FA regulation.

Single-molecule responses to dynamic forces

Studies of single molecules or pairs of molecules under applied force have rarely shown simple, switch-like behaviours. Of particular relevance, recent work on the effects of force application on the rates of bond dissociation shows that bonds mediating protein–protein interactions can either decrease or increase their average lifetime in response to applied force, referred to as slip or catch bonds, respectively[63,64] (Box 1). Notably, both the strength of the force and the rate of application affect the rates of protein conformational changes[63,65]. The development of assays involving several molecules (such as a crosslinking protein adhered to F-actin) has shown competition between force-activated unbinding and conformational changes[66]. A current challenge is determining how these molecular processes are integrated to mediate complex phenomena such as mechanotransduction.

Models of dynamic FAs

FAs offer a convenient system for understanding the relationship between dynamics and mechanotransduction that may be more generally applicable. From a mechanical perspective, FAs are dynamic, deformable links between an elastic ECM and the force-generating actin cytoskeleton[13,41]. Myosin-generated forces are transmitted through the actin cytoskeleton to FA proteins. The applied forces affect the dissociation rates of the FA proteins from integrin receptors, from each

BOX 2

Mechanical properties of the cytoskeleton

Materials such as rubber or polyacrylamide gels are elastic solids, meaning that they deform rapidly in response to applied stress but spring back to their original shape when the stress ends. By contrast, liquids such as water or honey flow in response to applied stress such that deformation increases irreversibly and linearly with time. Many materials, including cells[77] and *in vitro* mixtures of cytoskeletal components[20], show behaviours between these two limits and are referred to as viscoelastic. The behaviour of many such materials more closely resembles elastic materials on short timescales and viscous liquids on long timescales. A common example is silly putty, which flows like liquid when slowly squeezed but bounces like an elastic ball when thrown against the floor. On a molecular level, viscoelasticity is due to the presence of stress relaxation, usually through bond dissociation. At times shorter than the dissociation time, stress cannot be relaxed and the materials act like elastic solids, whereas on longer timescales, the bonds dissociate and the materials flow. The details of viscoelasticity in cytoskeletal networks are still controversial, but it is likely that the dissociation of, or conformational changes in, cytoskeletal crosslinking proteins is involved[77].

other and from F-actin. Forces are then transmitted through bound molecules to deform the ECM (Fig. 2a).

Several models that address how cells respond to the mechanical properties of the ECM propose that rigidity determines how quickly forces act on the integrin–actin linkages[13,67–69]. The models can be classified on the basis of whether rapidly applied forces increase or decrease adhesion turnover by promoting adhesion breakage or strengthening, respectively. Although the detailed predictions are distinct, in all cases FA kinetics are determined by the balance of protein association and dissociation. On soft substrates or in response to slowly applied forces, the on rates and off rates of molecules into and out of the adhesions, and the lifetimes of the whole adhesions, are moderate[67,68] (Fig. 2b). In models without FA strengthening, exposing cells to large, rapidly applied forces or plating cells on rigid substrata increases the dissociation of linker molecules, such as vinculin or talin, from the integrin or the actin (slip-bond behaviour). However, with large numbers of unoccupied sites, there is also rapid rebinding. This model leads to FAs with faster exchange of linker molecules and shorter whole adhesion lifetimes (Fig. 2c). In models with FA strengthening, applied force results in decreased protein dissociation (catch-bond behaviour) and/or a conformational change that induces protein recruitment[67,69]. Either way, force leads to large, reinforced FAs with slower exchange rates for linker molecules and longer lifetimes (Fig. 2d).

Notably, FA dynamics consistent with both classes of model have been observed[53,68,70]. In some cases, the difference is cell-type dependent. There is also evidence for spatial specificity within single cells, such that FA strengthening is restricted to the front of migrating cells[71]. This result makes intuitive sense, because if adhesions always strengthened under force, cells could not migrate. A polarized mechanism that strengthens adhesions at the front while allowing those at the rear to break under tension will produce forward movement when the cell contracts.

The polarized signalling pathways that determine whether adhesions strengthen or weaken under force are unknown. A recent study[72] using a biosensor that reports the tension across the FA protein vinculin has helped to shed some light on the mechanism. It showed that vinculin is under high tension in FAs that assemble, whereas it is under low tension in FAs that disassemble under cellular contractile force in migrating cells. Furthermore, vinculin is required for adhesion strengthening under force. These results indicate that the pathways that determine adhesion strengthening versus weakening under force regulate whether the force is transmitted across vinculin or other linkages.

A dynamic model of mechanotransduction

The examples listed above suggest that a dynamic treatment of mechanotransduction is necessary. A key concept is that applied forces can regulate the rates of biochemically detectable processes, such as protein unbinding and protein conformational changes. Although switch-like models emphasize the serial nature of the steps of mechanotransduction, a dynamic model shows a more integrated picture in which mechanotransmission, mechanotransduction and mechanoresponse are intimately related and can affect each other.

Dynamic mechanotransmission

Broken linkages cannot transmit forces. Thus, the stability of the load-bearing subcellular structures dictates paths of force transmission and their duration. On short timescales (subsecond to tens of seconds), mechanotransmission is governed by the physics of force-activated bond dissociation[63,64]. Whereas some cellular structures may simply be strong enough to bear the relevant forces, others will not. For example, bonds between actin and its crosslinking protein α-actinin are slip bonds, and other calponin-homology-domain actin-binding proteins are likely to behave similarly[73]. Other crucial linkages in mechanotransduction, such as fibronectin–integrin ($\alpha_5\beta_1$ integrin)[74,75] and actin–myosin[76] bonds, show catch-bond behaviour. These considerations suggest that only certain dynamic, subcellular structures may be stabilized in response to applied force to allow force transmission to mechanosensitive areas or molecules.

On larger length scales, the dynamic nature of cytoskeletal protein–protein bonds directly leads to viscoelasticity[77] (Box 2). Applied forces can result in either reinforcement[23] or fluidization[78] of the cytoskeleton. The exact mechanisms are still debated, but reinforcement is associated with the maintenance of physical linkages, stiffening of the actin network and increased cell contractility[23,79]. Fluidization involves disruption of the cytoskeleton, from either breakage of mechanical linkages[80,81] or force-induced, biochemically controlled disassembly[82]. In terms of mechanotransmission, these properties are extremely important as forces will be propagated along reinforced, elastic filaments, but quickly dissipate in a fluidized, viscous environment. Furthermore, viscoelastic effects can allow certain frequencies of mechanical stimulus to be selectively transmitted over greater distance in cells[83]. The efficiency of transmission for different frequencies is determined by the rates of bond dissociation that cause cellular viscoelasticity. These effects have been observed in force-induced movements of FAs[84] and mitochondria[85]. Mechanical stimuli with frequencies that are transmitted efficiently are likely to promote greater mechanoresponses.

Dynamic mechanosensing

The basis of mechanosensing is thought to be force-sensitive changes in the rates of conversion between different protein conformations. These transitions depend on the strength and duration of force application (Box 1). For instance, when forces are applied to talin, 50 pN induces conformational changes within 25 ms, whereas at 20 pN, the same changes require 200 ms[34]. For successful mechanosensing, forces must be transmitted for sufficient time to induce conformational changes and subsequent biochemical detection. But force can also accelerate slip-bond breakage, which will terminate the force transmission. Thus, there is competition between conformational change and bond breakage. Transmission pathways with catch bonds will therefore be more sensitive.

The rate at which forces are applied influences force transmission and subsequent signalling. The rate of force application through F-actin to the actin-crosslinking proteins α-actinin or filamin has been shown to determine the relative frequency of dissociation of the actin-linker bonds versus conformational changes[66]. Conformational changes

are more likely to occur at higher rates of force application. Other experiments have shown that fibronectin-coated beads are less likely to dissociate from integrins when forces are applied quickly[86]. The crucial effect of the rate of force application underscores the importance of these dynamic aspects of mechanotransduction.

Dynamic mechanoresponse

The downstream mechanoresponse pathways are not innately force sensitive but often regulate cytoskeletal and adhesion structures that therefore feed back to influence mechanotransduction. The cytoskeletal protein zyxin, for instance, specifically localizes to areas of strain-induced stress-fibre thinning, and recruits α-actinin and vasodilator-stimulated phosphoprotein, which promote actin polymerization and stress-fibre repair[87,88]. The myocardin-related transcription factor (also known as MAL) pathway is activated by actin polymerization in response to force or other stimuli, and regulates the expression of numerous cytoskeletal genes, including those encoding vinculin, filamin and actin[42,43]. Cyclic strain also enhances the expression of ECM proteins and induces the assembly of ECM structures[89]. Thus, on timescales of the order of minutes to days, cells use signalling or transcriptional programs to alter or maintain force-transmission pathways.

Cell alignment in response to applied force is a form of adaptation that involves local regulation of dynamic cytoskeletal elements, largely through the regulation of Rho family GTPases[90]. In two-dimensional cultures, uniaxial static stretch ('stretch and hold') induces actin stress fibre and FA alignment parallel to the applied force, consistent with the general notion of adhesion strengthening[90]. By contrast, cyclic stretch induces alignment perpendicular to the applied force[91] in a frequency-dependent manner[49].

A mathematical model recently proposed that forces applied faster than the characteristic rates of remodelling in load-bearing subcellular structures induce cell alignment perpendicular to the direction of strain to minimize stretching of these elements[92]. By contrast, when forces are applied slower than the remodelling rate, cells can internally remodel and align parallel to the applied stress. This concept may also explain a fascinating effect in which inhibiting Rho kinase or the Rho effector protein mammalian diaphanous has been found to shift the direction that cells align under cyclic stretch from perpendicular to parallel[93]. Inhibiting Rho signalling is also known to deplete cells of stable FAs and stress fibres, resulting in more dynamic subcellular structures[41]. The switch in direction of alignment may be explained if the higher cytoskeletal-remodelling rate now exceeds the characteristic rate of the cyclic stretch, which, according to the mathematical model, would yield alignment in the direction of strain. This model provides insight into how the internal dynamics of the cytoskeleton can determine responses to dynamic mechanical stimuli.

Adhesion strengthening and rigidity sensing

The principles enumerated above can provide at least a first explanation for how cells sense the mechanical properties of their substrata (Fig. 2). A key point is that forces on ECM–integrin–cytoskeletal linkages

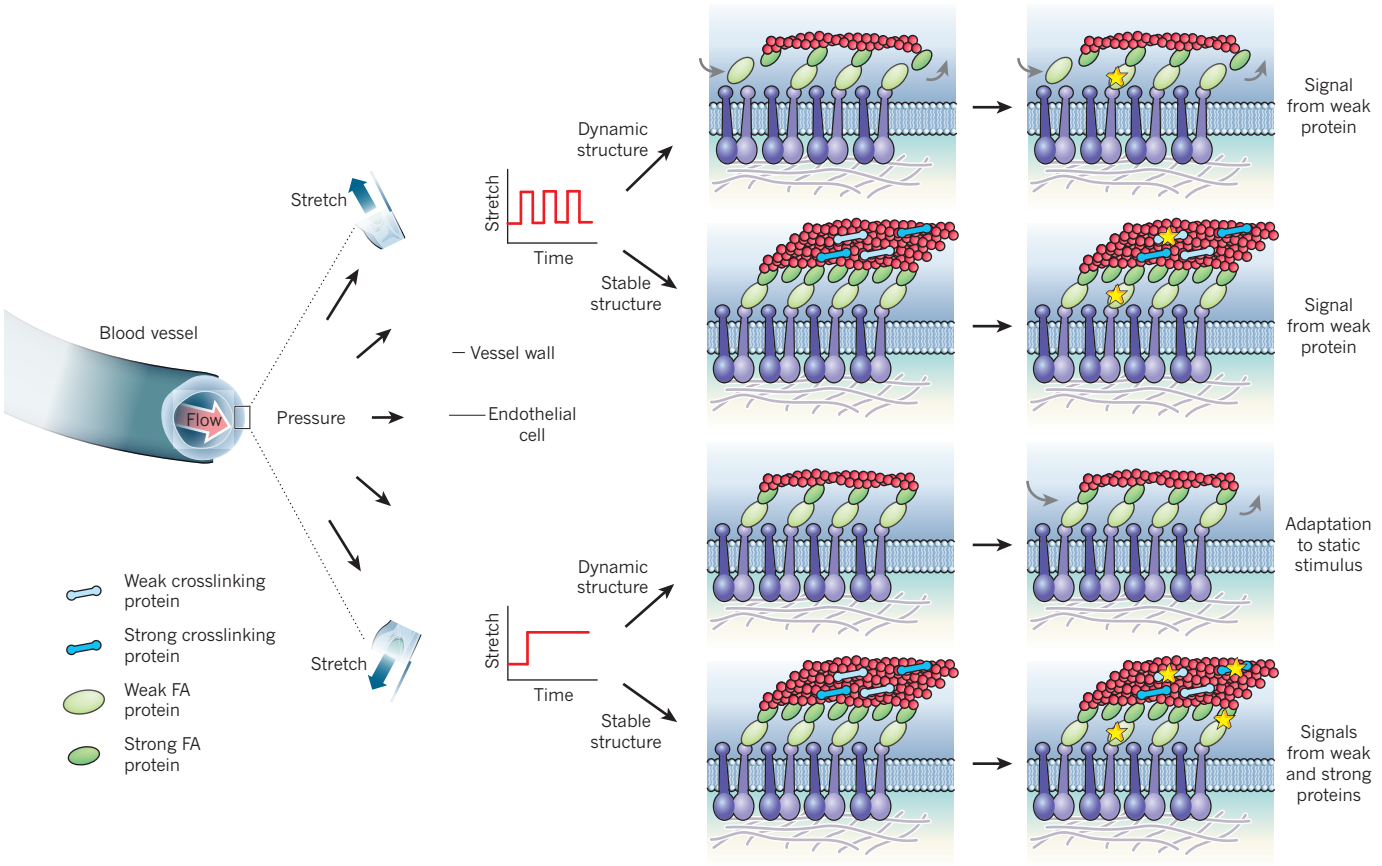


Figure 3 | Dynamic aspects of mechanotransduction. The endothelial cells that line blood-vessel walls are subject to both cyclic stretch (upper) and static stretch (lower). Even when the signal strengths are matched, dynamically distinct mechanical stimuli can activate common or unique signalling pathways based on the strength of the proteins — specifically the resistance to conformational changes — and the dynamic nature of the structures bearing loads. In dynamic structures, such as nascent adhesions, and stable structures, such as mature adhesions and stress fibres, cyclic stretch does not apply forces for sufficient time to induce conformational changes in strong proteins, but weak proteins will signal (yellow star). In response to static stretch, dynamic structures can readily adapt, and there is no long-term signalling. In stable structures, the long force application causes conformational changes in both weak and strong proteins. Thus, signalling pathways that are preferentially activated by cyclic stretch are probably induced by weak proteins that localize exclusively to dynamic structures. Pathways selectively activated by static stretch are likely to contain mechanosensors that are strong proteins in stable structures. Pathways activated by both types of signal probably involve weak proteins that localize to both dynamic and stable structures.

build up more rapidly in cells on rigid surfaces than on compliant surfaces[67,68]. These more rapidly applied forces are better at triggering conformational changes in cytoskeletal proteins than at causing bond dissociation[66]. Furthermore, if these linkages contain catch bonds — whose conversion to a high-affinity state is predicted to be enhanced at high loading rates[94] — they will be further stabilized. As a result, mechanotransmission will be more efficient and longer lived. As a consequence of domain unfolding under force, additional proteins are recruited to support crucial linkages. Adhesion strengthening will then occur through the mechanisms we describe above.

Sensing dynamic applied forces

One set of physiologically important mechanotransduction events involves the stretching of artery walls by blood pressure. Hypertension increases static stretch, whereas cyclic pumping during the cardiac cycle causes time-varying stretch[4], both of which lead to the transmission of forces to vascular cells and activation of many signalling pathways[95]. However, cyclic stretch and static stretch of the same amplitude (and probably similar force-application rates) induce both common and unique mechanoresponses[46]. For example, a 10% cyclic stretch of endothelial cells increases the expression of vascular endothelial growth factor receptor-2 (VEGFR-2) and the angiopoietin receptor TIE-2, but not VEGFR-1 expression. By contrast, a 10% static stretch increases the expression of VEGFR-2 and VEGFR-1, but not TIE-2 (ref. 46). We propose that these various responses can be explained by the dynamic processes intrinsic to the FAs and the cytoskeleton that sense the applied force (Fig. 3). Responses selectively activated by static stretch are probably mediated by protein conformational changes in strong proteins in relatively stable structures, requiring long applications of force to unfold. Responses selectively activated by cyclic stretch probably involve weaker proteins in dynamic structures that adapt to the statically applied forces but are constantly stimulated by dynamic signals. Responses that are activated by both are probably mediated by weak proteins in relatively stable structures.

Future perspectives

The ability of mechanical perturbations to influence cellular signalling in a frequency-dependent manner can be conceptualized as mechanotransducers that function as bandpass filters, which selectively transmit specific frequencies. The ability of the cytoskeleton to transmit certain frequencies of mechanical stimuli to subcellular structures selectively provides one such mechanism[83–85]. Mechanosensitive elements and mechanoresponse pathways are also rate sensitive and frequency sensitive, owing to their own intrinsic timescales. In this regard, the timescale of the applied force must match the crucial timescale of a given signalling process to affect it. Stimuli that change too quickly are simply averaged, whereas stimuli that vary too slowly are not detected at all. Knowledge of the dynamics of cellular mechanotransducers could therefore enhance our understanding of frequency-dependent cell and tissue responses. As cells contain several mechanically sensitive biochemical signalling pathways with wide variations in important timescales, they may act as multiband pass filters, which pass several ranges of frequency. These systems would allow cells to distinguish multiple stimuli based on their frequencies or timescales.

Our understanding of mechanical signalling is still slim compared with our understanding of signalling by hormones and growth factors. But the more we learn, the more it seems that mechanical forces can have subtle and precise roles in governing morphogenesis, physiology and disease. We propose that just as conventional signals from soluble regulators act together in regulatory networks in which complex temporal and spatial characteristics determine outputs, so mechanical stresses may also convey large amounts of information through precise time-dependent and force-dependent modulation. For periodic stimuli, this will take the form of frequency and amplitude features that determine cellular outputs. Elucidating the dynamics of cellular mechanotransduction systems holds the key to understanding these mechanisms. ■

1. Orr, A. W., Helmke, B. P., Blackman, B. R. & Schwartz, M. A. Mechanisms of mechanotransduction. *Dev. Cell* **10,** 11–20 (2006).
2. Chen, C. S. Mechanotransduction — a field pulling together? *J. Cell Sci.* **121,** 3285–3292 (2008).
3. Geiger, B., Spatz, J. P. & Bershadsky, A. D. Environmental sensing through focal adhesions. *Nature Rev. Mol. Cell Biol.* **10,** 21–33 (2009).
4. Hahn, C. & Schwartz, M. A. Mechanotransduction in vascular physiology and atherogenesis. *Nature Rev. Mol. Cell Biol.* **10,** 53–62 (2009).
5. Lucitti, J. L. *et al.* Vascular remodeling of the mouse yolk sac requires hemodynamic force. *Development* **134,** 3317–3326 (2007).
6. Yashiro, K., Shiratori, H. & Hamada, H. Haemodynamics determined by a genetic programme govern asymmetric development of the aortic arch. *Nature* **450,** 285–288 (2007).
7. Discher, D. E., Janmey, P. & Wang, Y. L. Tissue cells feel and respond to the stiffness of their substrate. *Science* **310,** 1139–1143 (2005).
8. Levental, K. R. *et al.* Matrix crosslinking forces tumor progression by enhancing integrin signaling. *Cell* **139,** 891–906 (2009).
9. Liu, F. *et al.* Feedback amplification of fibrosis through matrix stiffening and COX-2 suppression. *J. Cell Biol.* **190,** 693–706 (2010).
10. Engler, A. J., Sen, S., Sweeney, H. L. & Discher, D. E. Matrix elasticity directs stem cell lineage specification. *Cell* **126,** 677–689 (2006).
11. Gilbert, P. M. *et al.* Substrate elasticity regulates skeletal muscle stem cell self-renewal in culture. *Science* **329,** 1078–1081 (2010).
12. Dado, D. & Levenberg, S. Cell–scaffold mechanical interplay within engineered tissue. *Semin. Cell Dev. Biol.* **20,** 656–664 (2009).
13. De, R., Zemel, A. & Safran, S. A. Theoretical concepts and models of cellular mechanosensing. *Methods Cell Biol.* **98,** 143–175 (2010).
14. Saratzis, A. *et al.* Abdominal aortic aneurysm: a review of the genetic basis. *Angiology* **62,** 18–32 (2011).
15. Hershberger, R. E., Morales, A. & Siegfried, J. D. Clinical and genetic issues in dilated cardiomyopathy: a review for genetics professionals. *Genet. Med.* **12,** 655–667 (2010).
16. Chandrasekharan, K. & Martin, P. T. Genetic defects in muscular dystrophy. *Methods Enzymol.* **479,** 291–322 (2010).
17. Laurent, S., Boutouyrie, P. & Lacolley, P. Structural and genetic bases of arterial stiffness. *Hypertension* **45,** 1050–1055 (2005).
18. Yu, H., Mouw, J. K. & Weaver, V. M. Forcing form and function: biomechanical regulation of tumor evolution. *Trends Cell Biol.* **21,** 47–56 (2011).
19. Schwartz, M. A. & DeSimone, D. W. Cell adhesion receptors in mechanotransduction. *Curr. Opin. Cell Biol.* **20,** 551–556 (2008).
20. Gardel, M. L., Kasza, K. E., Brangwynne, C. P., Liu, J. & Weitz, D. A. Chapter 19 Mechanical response of cytoskeletal networks. *Methods Cell Biol.* **89,** 487–519 (2008).
21. Helmke, B. P., Rosen, A. B. & Davies, P. F. Mapping mechanical strain of an endogenous cytoskeletal network in living endothelial cells. *Biophys. J.* **84,** 2691–2699 (2003).
22. Tzima, E. *et al.* A mechanosensory complex that mediates the endothelial cell response to fluid shear stress. *Nature* **437,** 426–431 (2005).
This paper identifies a crucial complex, consisting of PECAM1, VE-cadherin and VEGFR-2, in the pathway leading to integrin activation by shear flow.
23. Matthews, B. D., Overby, D. R., Mannix, R. & Ingber, D. E. Cellular adaptation to mechanical stress: role of integrins, Rho, cytoskeletal tension and mechanosensitive ion channels. *J. Cell Sci.* **119,** 508–518 (2006).
24. Na, S. *et al.* Rapid signal transduction in living cells is a unique feature of mechanotransduction. *Proc. Natl Acad. Sci. USA* **105,** 6626–6631 (2008).
25. Wang, N. & Stamenovic, D. Contribution of intermediate filaments to cell stiffness, stiffening, and growth. *Am. J. Physiol. Cell Physiol.* **279,** C188–C194 (2000).
26. Hayakawa, K., Tatsumi, H. & Sokabe, M. Actin stress fibers transmit and focus force to activate mechanosensitive channels. *J. Cell Sci.* **121,** 496–503 (2008).
27. Poh, Y. C. *et al.* Rapid activation of Rac GTPase in living cells by force is independent of Src. *PLoS ONE* **4,** e7886 (2009).
28. Sukharev, S., Betanzos, M., Chiang, C. S. & Guy, H. R. The gating mechanism of the large mechanosensitive channel MscL. *Nature* **409,** 720–724 (2001).
29. Árnadóttir, J. & Chalfie, M. Eukaryotic mechanosensitive channels. *Annu. Rev. Biophys.* **39,** 111–137 (2010).
30. Zhong, C. *et al.* Rho-mediated contractility exposes a cryptic site in fibronectin and induces fibronectin matrix assembly. *J. Cell Biol.* **141,** 539–551 (1998).
31. Oberhauser, A. F., Badilla-Fernandez, C., Carrion-Vazquez, M. & Fernandez, J. M. The mechanical hierarchies of fibronectin observed with single-molecule AFM. *J. Mol. Biol.* **319,** 433–447 (2002).
32. Smith, M. L. *et al.* Force-induced unfolding of fibronectin in the extracellular matrix of living cells. *PLoS Biol.* **5,** e268 (2007).
33. Ziegler, W. H., Gingras, A. R., Critchley, D. R. & Emsley, J. Integrin connections to the cytoskeleton through talin and vinculin. *Biochem. Soc. Trans.* **36,** 235–239 (2008).
34. del Rio, A. *et al.* Stretching single talin rod molecules activates vinculin binding. *Science* **323,** 638–641 (2009).
This study demonstrates force-induced binding of vinculin to cryptic sites in talin at the single molecule level.
35. Zhang, X. *et al.* Talin depletion reveals independence of initial cell spreading from integrin activation and traction. *Nature Cell Biol.* **10,** 1062–1068 (2008).
36. Defilippi, P., Di Stefano, P. & Cabodi, S. p130Cas: a versatile scaffold in signaling networks. *Trends Cell Biol.* **16,** 257–263 (2006).
37. Tamada, M., Sheetz, M. P. & Sawada, Y. Activation of a signaling cascade by cytoskeleton stretch. *Dev. Cell* **7,** 709–718 (2004).

38. Sawada, Y. *et al.* Rap1 is involved in cell stretching modulation of p38 but not ERK or JNK MAP kinase. *J. Cell Sci.* **114,** 1221–1227 (2001).
39. Sawada, Y. & Sheetz, M. P. Force transduction by Triton cytoskeletons. *J. Cell Biol.* **156,** 609–615 (2002).
40. Sawada, Y. *et al.* Force sensing by mechanical extension of the Src family kinase substrate p130Cas. *Cell* **127,** 1015–1026 (2006).
This study shows that stretching of $p130^{Cas}$ leads to the exposure of tyrosine residues, which can be phosphorylated to affect signalling pathways.
41. Parsons, J. T., Horwitz, A. R. & Schwartz, M. A. Cell adhesion: integrating cytoskeletal dynamics and cellular tension. *Nature Rev. Mol. Cell Biol.* **11,** 633–643 (2010).
42. Asparuhova, M. B., Gelman, L. & Chiquet, M. Role of the actin cytoskeleton in tuning cellular responses to external mechanical stress. *Scand. J. Med. Sci. Sports* **19,** 490–499 (2009).
43. Olson, E. N. & Nordheim, A. Linking actin dynamics and gene transcription to drive cellular motile functions. *Nature Rev. Mol. Cell Biol.* **11,** 353–365 (2010).
44. Mallion, J. M., Baguet, J. P., Siche, J. P., Tremel, F. & De Gaudemaris, R. Left ventricular hypertrophy and arterial hypertrophy. *Adv. Exp. Med. Biol.* **432,** 123–133 (1997).
45. Vogel, V. Mechanotransduction involving multimodular proteins: converting force into biochemical signals. *Annu. Rev. Biophys. Biomol. Struct.* **35**, 459–488 (2006).
46. Zheng, W., Christensen, L. P. & Tomanek, R. J. Differential effects of cyclic and static stretch on coronary microvascular endothelial cell receptors and vasculogenic/angiogenic responses. *Am. J. Physiol. Heart Circ. Physiol.* **295,** H794–H800 (2008).
This study provides evidence that statically and dynamically applied stretches lead to the activation of distinct pathways in stretched endothelial cells.
47. Lehoux, S., Esposito, B., Merval, R. & Tedgui, A. Differential regulation of vascular focal adhesion kinase by steady stretch and pulsatility. *Circulation* **111,** 643–649 (2005).
48. Hsu, H. J., Lee, C. F. & Kaunas, R. A dynamic stochastic model of frequency-dependent stress fiber alignment induced by cyclic stretch. *PLoS ONE* **4,** e4853 (2009).
49. Liu, B. *et al.* Role of cyclic strain frequency in regulating the alignment of vascular smooth muscle cells *in vitro*. *Biophys J.* **94,** 1497–1507 (2008).
50. Gelfand, B. D., Epstein, F. H. & Blackman, B. R. Spatial and spectral heterogeneity of time-varying shear stress profiles in the carotid bifurcation by phase-contrast MRI. *J. Magn. Reson. Imaging* **24,** 1386–1392 (2006).
51. Dancu, M. B. & Tarbell, J. M. Large negative stress phase angle (SPA) attenuates nitric oxide production in bovine aortic endothelial cells. *J. Biomech. Eng.* **128,** 329–334 (2006).
52. Wehrle-Haller, B. Analysis of integrin dynamics by fluorescence recovery after photobleaching. *Methods Mol. Biol.* **370,** 173–201 (2007).
53. Hu, K., Ji, L., Applegate, K. T., Danuser, G. & Waterman-Storer, C. M. Differential transmission of actin motion within focal adhesions. *Science* **315,** 111–115 (2007).
54. Brown, C. M. *et al.* Probing the integrin–actin linkage using high-resolution protein velocity mapping. *J. Cell Sci.* **119,** 5204–5214 (2006).
55. Maruthamuthu, V., Aratyn-Schaus, Y. & Gardel, M. L. Conserved F-actin dynamics and force transmission at cell adhesions. *Curr. Opin. Cell Biol.* **22,** 583–588 (2010).
56. Liu, Z. et al. Mechanical tugging force regulates the size of cell–cell junctions. *Proc. Natl Acad. Sci. USA* **107,** 9944–9949 (2010).
57. Maruthamuthu, V., Sabass, B., Schwarz, U. S. & Gardel, M. L. Cell–ECM traction force modulates endogenous tension at cell–cell contacts. *Proc. Natl Acad. Sci. USA* **108,** 4708–4713 (2011).
58. Mège, R. M., Gavard, J. & Lambert, M. Regulation of cell–cell junctions by the cytoskeleton. *Curr. Opin. Cell Biol.* **18,** 541–548 (2006).
59. Ladoux, B. *et al*. Strength dependence of cadherin-mediated adhesions. *Biophys. J.* **98,** 534–542 (2010).
60. Kametani, Y. & Takeichi, M. Basal-to-apical cadherin flow at cell junctions. *Nature Cell Biol.* **9,** 92–98 (2007).
61. Riveline, D. *et al.* Focal contacts as mechanosensors: externally applied local mechanical force induces growth of focal contacts by an mDia1-dependent and ROCK-independent mechanism. *J. Cell Biol.* **153,** 1175–1186 (2001).
62. le Duc, Q. *et al.* Vinculin potentiates E-cadherin mechanosensing and is recruited to actin-anchored sites within adherens junctions in a myosin II-dependent manner. *J. Cell Biol.* **189,** 1107–1115 (2010).
63. Evans, E. A. & Calderwood, D. A. Forces and bond dynamics in cell adhesion. *Science* **316,** 1148–1153 (2007).
64. Thomas, W. E., Vogel, V. & Sokurenko, E. Biophysics of catch bonds. *Annu. Rev. Biophys.* **37,** 399–416 (2008).
65. Bustamante, C., Chemla, Y. R., Forde, N. R. & Izhaky, D. Mechanical processes in biochemistry. *Annu. Rev. Biochem.* **73,** 705–748 (2004).
66. Ferrer, J. M. *et al.* Measuring molecular rupture forces between single actin filaments and actin-binding proteins. *Proc. Natl Acad. Sci. USA* **105,** 9221–9226 (2008).
67. Bruinsma, R. Theory of force regulation by nascent adhesion sites. *Biophys. J.* **89,** 87–94 (2005).
68. Chan, C. E. & Odde, D. J. Traction dynamics of filopodia on compliant substrates. *Science* **322,** 1687–1691 (2008).
This study proposes and validates a model describing rigidity sensitive FA dynamics in terms of force-activated protein dissociation.
69. Li, Y., Bhimalapuram, P. & Dinner, A. R. Model for how retrograde actin flow regulates adhesion traction stresses. *J. Phys. Condens. Matter* **22,** 194113 (2010).
70. Gardel, M. L. *et al.* Traction stress in focal adhesions correlates biphasically with actin retrograde flow speed. *J. Cell Biol.* **183,** 999–1005 (2008).
71. Schmidt, C. E., Horwitz, A. F., Lauffenburger, D. A. & Sheetz, M. P. Integrin–cytoskeletal interactions in migrating fibroblasts are dynamic, asymmetric, and regulated. *J. Cell Biol.* **123,** 977–991 (1993).
72. Grashoff, C. *et al.* Measuring mechanical tension across vinculin reveals regulation of focal adhesion dynamics. *Nature* **466,** 263–266 (2010).
This paper reports a biosensor that measures forces across specific proteins in dynamic FAs and shows that molecular tension across vinculin correlates with FA strengthening.
73. Miyata, H., Yasuda, R. & Kinosita, K. Jr. Strength and lifetime of the bond between actin and skeletal muscle α-actinin studied with an optical trapping technique. *Biochim. Biophys. Acta* **1290,** 83–88 (1996).
74. Kong, F., Garcia, A. J., Mould, A. P., Humphries, M. J. & Zhu, C. Demonstration of catch bonds between an integrin and its ligand. *J. Cell Biol.* **185,** 1275–1284 (2009).
This study shows that the linkage between $\alpha_5\beta_1$ integrin and fibronectin acts like a catch bond at the single molecule level.
75. Friedland, J. C., Lee, M. H. & Boettiger, D. Mechanically activated integrin switch controls $\alpha_5\beta_1$ function. *Science* **323**, 642–644 (2009).
This study shows that force and increased extracellular rigidity switch $\alpha_5\beta_1$ integrin between a relaxed and a tensioned state that is required for mechanically induced focal adhesion kinase signalling.
76. Guo, B. & Guilford, W. H. Mechanics of actomyosin bonds in different nucleotide states are tuned to muscle contraction. *Proc. Natl Acad. Sci. USA* **103,** 9844–9849 (2006).
77. Hoffman, B. D. & Crocker, J. C. Cell mechanics: dissecting the physical responses of cells to force. *Annu. Rev. Biomed. Eng.* **11,** 259–288 (2009).
78. Trepat, X. *et al.* Universal physical responses to stretch in the living cell. *Nature* **447,** 592–595 (2007).
79. Gardel, M. L. *et al.* Prestressed F-actin networks cross-linked by hinged filamins replicate mechanical properties of cells. *Proc. Natl Acad. Sci. USA* **103,** 1762–1767 (2006).
80. Lee, H., Ferrer, J. M., Lang, M. J. & Kamm, R. D. Molecular origin of strain softening in cross-linked F-actin networks. *Phys. Rev. E* **82,** 011919 (2010).
81. Chaudhuri, O., Parekh, S. H. & Fletcher, D. A. Reversible stress softening of actin networks. *Nature* **445,** 295–298 (2007).
82. Chen, C. *et al.* Fluidization and resolidification of the human bladder smooth muscle cell in response to transient stretch. *PLoS ONE* **5,** e12035 (2010).
83. Shafrir, Y. & Forgacs, G. Mechanotransduction through the cytoskeleton. *Am. J. Physiol. Cell Physiol.* **282,** C479–C486 (2002).
84. Mack, P. J., Kaazempur-Mofrad, M. R., Karcher, H., Lee, R. T. & Kamm, R. D. Force-induced focal adhesion translocation: effects of force amplitude and frequency. *Am. J. Physiol. Cell Physiol.* **287,** C954–C962 (2004).
85. Hu, S. & Wang, N. Control of stress propagation in the cytoplasm by prestress and loading frequency. *Mol. Cell. Biomech.* **3,** 49–60 (2006).
86. Jiang, G., Huang, A. H., Cai, Y., Tanase, M. & Sheetz, M. P. Rigidity sensing at the leading edge through $\alpha_v\beta_3$ integrins and RPTPa. *Biophys. J.* **90,** 1804–1809 (2006).
87. Smith, M. A. *et al.* A zyxin-mediated mechanism for actin stress fiber maintenance and repair. *Dev. Cell* **19,** 365–376 (2010).
88. Wojtowicz, A. *et al.* Zyxin mediation of stretch-induced gene expression in human endothelial cells. *Circ. Res.* **107,** 898–902 (2010).
89. Chiquet, M., Gelman, L., Lutz, R. & Maier, S. From mechanotransduction to extracellular matrix gene expression in fibroblasts. *Biochim. Biophys. Acta* **1793,** 911–920 (2009).
90. Katsumi, A. *et al.* Effects of cell tension on the small GTPase Rac. *J. Cell Biol.* **158,** 153–164 (2002).
91. Kanda, K. & Matsuda, T. Behavior of arterial-wall cells cultured on periodically stretched substrates. *Cell Transplant.* **2,** 475–484 (1993).
92. De, R., Zemel, A. & Safran, S. A. Dynamics of cell orientation. *Nature Phys.* **3,** 655–659 (2007).
This theory-based study suggests how cytoskeletal dynamics affect the ability of cells to align to dynamically applied stretches.
93. Kaunas, R., Nguyen, P., Usami, S. & Chien, S. Cooperative effects of Rho and mechanical stretch on stress fiber organization. *Proc. Natl Acad. Sci. USA* **102,** 15895–15900 (2005).
94. Prezhdo, O. V. & Pereverzev, Y. V. Theoretical aspects of the biological catch bond. *Acc. Chem. Res.* **42,** 693–703 (2009).
95. Haga, J. H., Li, Y. S. & Chien, S. Molecular basis of the effects of mechanical stretch on vascular smooth muscle cells. *J. Biomech.* **40,** 947–960 (2007).

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