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COMMENTARY

The mechanical regulation of integrin–cadherin crosstalk organizes cells, signaling and forces

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ABSTRACT

Cadherins and integrins are intrinsically linked through the actin cytoskeleton and share common signaling molecules. Although mechanosensing by the integrin–actin axis has long been appreciated, a growing body of literature now demonstrates that cadherins also transduce and respond to mechanical forces. Mounting evidence shows that mechanically driven crosstalk between integrins and cadherins regulates the spatial distribution of these receptors, their signaling intermediates, the actin cytoskeleton and intracellular forces. This interplay between integrins and cadherins can control fibronectin matrix assembly and signaling, and a fine balance between traction forces at focal adhesions and intercellular tension at adherens junctions is crucial for directional collective cell migration. In this Commentary, we discuss two central ideas: (1) how the dynamic interplay between integrins and cadherins regulates the spatial organization of intracellular signals and the extracellular matrix, and (2) the emerging consensus that intracellular force is a central mechanism that dictates cell behavior, guides tissue development and ultimately drives physiology.

KEY WORDS: Mechanotransduction, Focal adhesion, Adherens junction, Actin cytoskeleton

Introduction

Amid the complex extracellular milieu of chemical and mechanical signals, cells rely on adhesion receptors to probe and make sense of their microenvironment. Integrins and cadherins are two of the best-studied classes of adhesion receptors. Integrins mediate adhesion between the cell and its extracellular matrix (ECM), and cadherins mediate homotypic adhesion between cells. Clustered integrins associate with focal adhesions, which are multi-protein complexes that link these receptors to the actin cytoskeleton (Sastry and Burridge, 2000) (Fig. 1). Cadherin-based adherens junctions, which consist of α- catenin, β-catenin and p120-catenin (also known as CTNND1) as the core molecular components, serve a similar role in cadherin–actin interactions (Gumbiner, 2005) (Fig. 1). At the cellular level, engagement of both integrins and cadherins stimulates the Rho family of GTPases (Rho, Rac and Cdc42) to remodel the architecture of the actin cytoskeleton adaptively in response to adhesion (Etienne-Manneville and Hall, 2002; Parsons et al., 2010; Watanabe et al., 2009). In addition to its structural role, the actin–myosin network forms the internal contractile machinery of the cell (Fig. 1) and generates intracellular tension in response to different forces transduced by focal adhesions and adherens junctions (see Box 1 for definitions of relevant mechanobiology terms).

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The integration of adhesion, biochemical signaling and cytoskeletal network remodeling dynamically tunes the generation of intracellular force and regulates signal transduction, as well as transcriptional events that control fundamental biological processes, including proliferation, differentiation and migration. At the tissue level, the coordinated interaction between integrins and cadherins transmits local information globally, mechanically connecting the actin cytoskeleton to neighboring cells and the matrix in order to regulate multicellular processes, such as collective migration and tissue patterning during morphogenesis. The development of different techniques that are now commonly employed to manipulate physical and adhesive environments of the cell (Box 2) has allowed for a deeper exploration and understanding of the mechano-chemical interplay between integrins and cadherins.

Integrins and cadherins have been reviewed extensively, as has the idea that these receptors interact (Schwartz and DeSimone, 2008; Weber et al., 2011), but a number of recent reports have revealed exciting new mechanisms of crosstalk, especially in different mechanical contexts. In this Commentary, we focus on mechanotransduction as a fundamental driver of integrin–cadherin crosstalk and the importance of these interactions in regulating cellular functions. Specifically, we discuss new evidence demonstrating that focal adhesion proteins play new roles at adherens junctions, that cadherins regulate integrin activation and matrix assembly, and that integrin–cadherin crosstalk spatially organizes signaling components and forces within cells and coordinates cell movement.

New roles for focal adhesion proteins in mechanotransduction, cadherin expression and junctional stability

Focal adhesions and adherens junctions have been historically regarded as functionally and spatially distinct adhesion structures with different molecular compositions. However, as both focal adhesions and adherens junctions are intracellularly linked to the actin cytoskeleton, they recruit and activate a common set of signaling proteins and actin regulators, such as Rho family GTPases. Interestingly, several canonical focal adhesion proteins have now been detected in adherens junctions, indicating that these structures might not be as distinct as once thought.

Vinculin is among the best-studied components of focal adhesions that also appears in adherens junctions. Unassociated vinculin assumes an auto-inhibited conformation in which the vinculin head domain binds to its tail domain. In focal adhesions, the vinculin head and tail domains bind to talin and actin, respectively (Galbraith et al., 2002). These interactions control the ability of bound vinculin to bear force, which in turn determines whether focal adhesions assemble or disassemble under tension (Grashoff et al., 2010). Similarly, within cadherin-dependent junctions, α-catenin exposes a cryptic vinculin-binding site under

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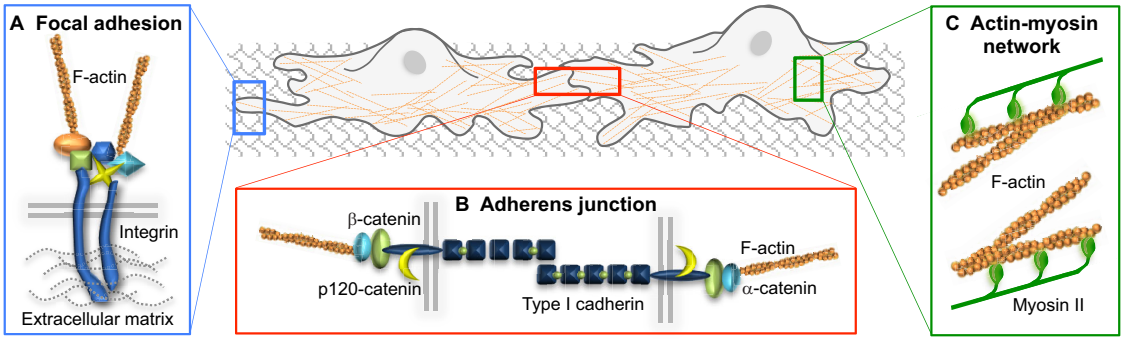


Fig. 1. Integrins and cadherins modulate the mechanical landscape of the cell. Integrin-based focal adhesions (A) and cadherin-dependent adherens junctions (B) relay mechanical signals through a contractile actin–myosin network (C) to actively modulate the mechanical landscape of the cell. Focal adhesions and adherens junctions form the linkages of the cell to the ECM and to neighboring cells, respectively. Integrins and cadherins are linked to the intracellular actin–myosin network and are thus intrinsically linked to each other. Forces are relayed between integrins and cadherins through the tensional changes in the actin–myosin network to shape the mechanical landscape of the cell.

high intracellular tension and recruits vinculin to reinforce adherens junction linkages to F-actin (Yonemura et al., 2010).

Distinct site-specific tyrosine phosphorylation of vinculin distinguishes its mechanical role in adherens junctions and focal adhesions. DeMali and colleagues have demonstrated that applied force on E-cadherin increases the Abl-mediated phosphorylation of vinculin on tyrosine 822 (Y822) and allows vinculin to integrate into E-cadherin-based intercellular junctions (Bays et al., 2014). The degree of Y822-phosphorylated vinculin within adherens junctions then determines the extent to which cadherins transduce force. In contrast, vinculin phosphorylation at Y822 is not required for force transduction by integrins. Consistent with these results, Goldmann and colleagues have shown that Src-dependent phosphorylation of vinculin at Y100 and Y1065 regulates force transmission from the ECM to focal adhesions and the actin cytoskeleton (Auernheimer et al., 2015). Thus, spatial regulation of vinculin phosphorylation is an important regulatory modality that determines its mechanical effects at integrin- versus cadherin-dependent adhesions.

Remarkably, even focal adhesion kinase (FAK, also known as PTK2), the canonical focal-adhesion-associated protein, might localize to and modulate cell–cell adhesions. Schlaepfer and colleagues have reported that FAK binds to VE-cadherin and phosphorylates β-catenin^{Y142} in vascular endothelial growth factor (VEGF)-stimulated human umbilical vein endothelial cells (Chen et al., 2012). Here, FAK-dependent β-catenin phosphorylation facilitates its dissociation from VE-cadherin and is associated with reduced junctional stability and increased cell permeability. Although focal adhesion proteins are typically activated by tension, this interaction between FAK and VE-cadherin occurs in a tension-independent manner, as the FAK–VE-cadherin interaction persists in the presence of blebbistatin, an inhibitor of myosin-generated contractility (Chen et al., 2012).

Box 1. Glossary

Selected terms in mechanobiology

Actomyosin contractility: contraction of actin fibers by myosin motors, which leads to the generation of intracellular tension.

Mechanotransduction: conversion of mechanical signals into biochemical signals.

Stress: force divided by the area to which the force is applied.

Tensional homeostasis: equilibrium of tensional forces within a cell.

Cells tune their intracellular tension proportionally to ECM stiffness as evidenced by increased phosphorylation of focal adhesion proteins, such as FAK, p130Cas (also known as BCAR1), vinculin and paxillin, in cells cultured on relatively stiff polyacrylamide hydrogels. Functionally, phosphorylation of FAK and p130Cas (but not vinculin or paxillin) are required for stiffness-induced entry into S-phase in mouse embryonic fibroblasts (MEFs) (Bae et al., 2014). In this system, FAK and p130Cas are core components of a focal adhesion signaling module that activates Rac. Interestingly, this integrin-initiated module also triggers crosstalk with cell–cell adhesion pathways. For instance, we have found that

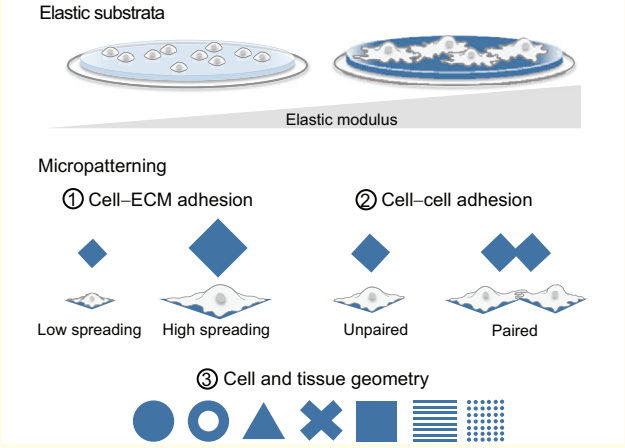
Box 2. Selected techniques for manipulating the physical and adhesive environment of the cell

Elastic substrata

The use of elastic substrata such as hydrogels (polyacrylamide, hyaluronan, polyethylene glycol or collagen) or silicone gels allows cells to experience physiologically relevant substrate stiffness. Cells cultured on relatively stiffer substrata typically are more spread and exhibit more organized actin structures than their counterparts on soft substrata.

Micropatterning

Micropatterning provides a means to control cell adhesion and cell shape by limiting matrix protein deposition to defined shapes and areas. (1) Cell spreading on ECM can be controlled by varying the area of the micropatterned matrix protein. (2) Cell–cell adhesion can be manipulated by changing the shape of the micropatterned matrix protein. (3) Different shapes and patterns modulate the geometry of single cells and tissues.



stiffness-dependent activation of the FAK–p130Cas–Rac pathway leads to increased expression of N-cadherin (also known as CDH2) mRNA and protein in MEFs and mouse vascular smooth muscle cells (VSMCs) (Mui et al., 2015).

Furthermore, we have assessed the functional importance of this effect on cell proliferation by measuring S-phase entry in VSMCs cultured on micropatterned islands. We utilized micropatterning (see Box 1) as a means to systematically control cell–cell contact (paired versus unpaired) and the area of cell spreading on matrix, and thereby examine cell proliferation under different adhesive contexts. Our results indicate that FAK–p130Cas–Rac-stimulated induction of N-cadherin allow cells to enter S phase when spreading is constrained, a condition that typically precludes cell proliferation (Mui et al., 2015). This *in vitro* finding was corroborated *in vivo*: studies using conditional knockout mice bearing floxed alleles of FAK or N-cadherin demonstrate a FAK-dependent upregulation of N-cadherin that correlates with VSMC proliferation after vascular injury, a model of arterial stiffening (Klein et al., 2009). Thus, activation of FAK–p130Cas–Rac signaling might determine the degree to which ECM-dependent cell spreading is required for proliferation. These results and those mentioned above (Chen et al., 2012) lead to the idea that FAK can participate in both tension-independent and tension-dependent events to regulate crosstalk between cell–ECM and cell–cell adhesions.

Although paxillin does not have a role in N-cadherin-mediated proliferation downstream of FAK (Bae et al., 2014), paxillin regulates N-cadherin in a different functional context. An early study has demonstrated that paxillin works in concert with FAK to stimulate assembly of N-cadherin-dependent junctions and inhibit cell migration (Yano et al., 2004). By analyzing experiments where different focal adhesion proteins had been knocked down by using small interfering RNA (siRNA), the authors concluded that paxillin is required for the recruitment of FAK to focal adhesions at the cell periphery of motile HeLa cells, where it downregulates Rac activity to promote the formation of N-cadherin-based cell–cell adhesions. Thus, different focal adhesion proteins (p130Cas and paxillin) can differentially regulate N-cadherin function depending on cellular context or cell type.

In addition to the stimulatory effect of Rac on N-cadherin gene expression as discussed above, Rac regulates the assembly and maintenance of cadherin-based adhesions. A recent study by Nelson and colleagues demonstrates that the activation of Rac at adherens junctions is under the control of Elmo and DOCK proteins (Toret et al., 2014). Elmo proteins act as scaffolding proteins that bind to DOCK proteins, which are Rac guanine exchange factors (GEFs). These authors showed that a DOCK–Elmo complex (consisting of DOCK1 and Elmo2) transiently localizes to nascent cell–cell contacts in Madin–Darby canine kidney cells, where it colocalizes with areas of Rac activity and results in the recruitment of E-cadherin and the local reorganization of actin (Toret et al., 2014). Although DOCK–Elmo complexes have established roles in regulating focal adhesion dynamics during cell migration and spreading on ECM, the biological roles of these proteins can now be expanded beyond focal adhesions to include important effects in the maturation of adherens junctions.

Overall, the new work summarized here reveals the many layers by which several focal adhesion proteins (vinculin, FAK, p130Cas, Rac, paxillin and Elmo–DOCK complexes) regulate adherens junctions and cadherin biology. This body of work also makes clear that adherens junctions are more complex than generally acknowledged. As more evidence emerges for shared regulatory components between integrin- and cadherin-based adhesions, it becomes increasingly apparent that these adhesion systems are interconnected through shared components that extend well beyond actin itself (Fig. 2).

Regulation of mechanosignaling and forces by crosstalk between integrins and cadherins

Because forces are experienced simultaneously across integrin-based focal adhesions and cadherin-dependent adherens junctions,

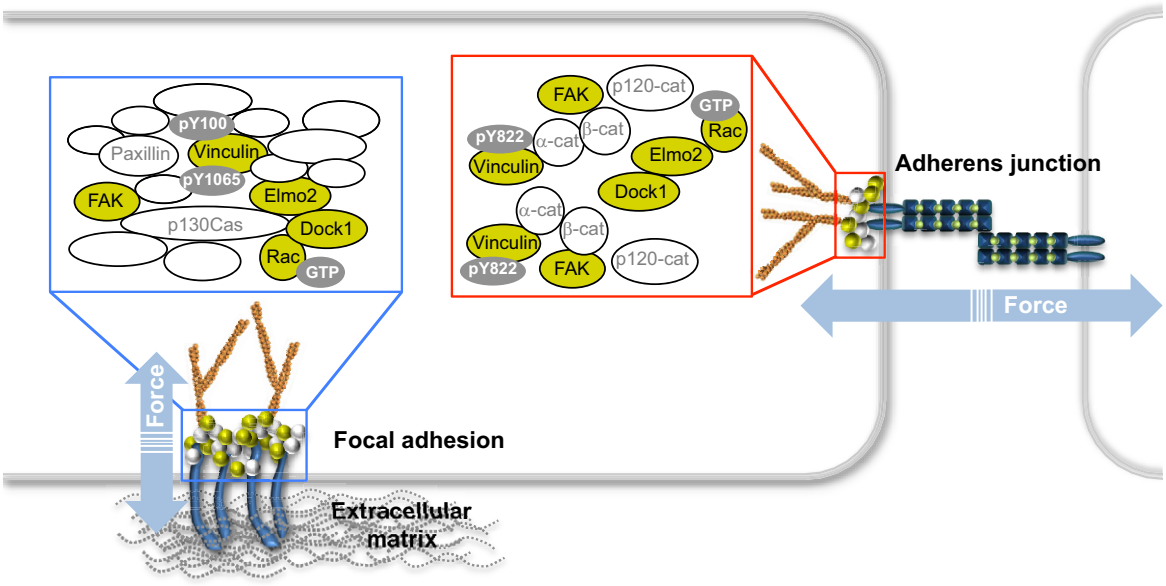


Fig. 2. Focal adhesions and adherens junctions signal through common molecular components. Focal adhesions and adherens junctions are each comprised of unique constituents (white), but they also share many common signaling components (yellow). For instance, several molecules typically associated with focal adhesions, such as FAK, vinculin, Rac, and DOCK and Elmo proteins, also localize to adherens junctions and regulate cadherin dynamics. Mechanical forces transduced across integrins and cadherins activate many of these signaling molecules. (Proteins are not drawn to scale and not all reported protein interactions are depicted.) pY1065, phosphorylation of Y1065 in vinculin.

variations in adhesion strength across the cell can generate shifts in the balance of tension across both adhesion systems. This might be a means to achieve tensional homeostasis, which is important for organizing and localizing adhesions within cells and the application of force across cells. Several reports have demonstrated that strengthening of one type of adhesion opposes the formation of the other. For instance, early studies have shown that FAK activation downstream of strong ECM–integrin engagement leads to a loss in VE-cadherin- (Wang et al., 2006) and N-cadherin-mediated intercellular contacts (Yano et al., 2004). Conversely, cells on soft polyacrylamide hydrogels have weakened adhesions to the underlying ECM, and this stimulates cell aggregation and compaction (Guo et al., 2006). However, the view that cell–cell and cell–matrix adhesions are in a constant tug-of-war is too simplistic. Interactions between cell–cell and cell–matrix adhesions are not necessarily antagonistic, but can also be cooperative and interdependent to allow the cell to achieve tensional homeostasis.

Using an inducible, endothelial-specific integrin β1 knockout mouse model, a recent study has demonstrated that β1 integrin is required for the localization of VE-cadherin and p120-catenin to intercellular junctions, the internalization and trafficking of VE-cadherin, preservation of junctional integrity and endothelial cell sprouting (Yamamoto et al., 2015). Conversely, any misregulation of these processes culminates in the formation of leaky, unstable blood vessels owing to defects in endothelial cell junctions.

Other recent studies have also examined how cadherins and integrins regulate their mutual distribution within cells and establish polarized signaling schemes by segregating distinct molecular components. For instance, Ouyang et al. cultured MEFs on micropatterned fibronectin strips to investigate polarized phosphoinositide 3-kinase (PI3K) and Rac signaling at the free end of a cell compared with the end that contacts an adjacent cell (Ouyang et al., 2013). PI3K and Rac activities were stimulated by integrin signaling at the free end, whereas N-cadherin–p120-catenin complexes excluded α5β1 integrin from intercellular junctions to suppress local PI3K and Rac activity. Myosin II light chain and actin filaments localized to cell–cell junctions in this system, and based on data from ectopic expression of mutant forms of N-cadherin and catenin, the authors concluded that this polarized localization was selectively regulated by N-cadherin–β-catenin complexes. The results of this study suggest that we need further understanding of the role of actomyosin contractility in adhesion-polarized signaling and raise the question of how the organization of cells and the resulting positioning of cell–cell and cell–ECM adhesions might affect tension between integrin- and cadherin-mediated adhesions and drive differential signaling within the same cell.

The interaction between integrins and cadherins can also direct the localization of forces within cell aggregates. For example, Mertz et al. have performed experiments where they cultured keratinocytes on two-dimensional (2D) fibronectin-coated, silicone gels and incubated the cells in low or high Ca^{2+} to either preclude or support cadherin-mediated adhesion (Mertz et al., 2013). Keratinocytes in high Ca^{2+} aggregated into colonies, with coordinated actin fiber organization across multiple cells and spatially organized intracellular traction stresses that were most prominent at the periphery and aligned radially inwards. By contrast, colonies in low Ca^{2+} lost multicellular coordination of the actin cytoskeleton, and the traction stresses were much more evenly distributed throughout the colony. Using function-blocking antibodies, and knockout or knockdown cells, the authors were able to show that the effects of high Ca^{2+} were mediated by E-cadherin (Mertz et al., 2013). Thus, the degree of E-cadherin engagement controls the localization of F-actin and traction forces observed in response to fibronectin-mediated adhesion. However, how cadherin-driven organized patterns of stress within groups of cells translates into functionally relevant outcomes remains to be determined.

A similar scenario unfolds in colonies of pluripotent stem cells. In a recent study, human embryonic stem cells (hESCs) were micropatterned on Matrigel islands of different geometries to manipulate the localization and the relative magnitudes of integrin- and E-cadherin-based adhesions at the periphery and interior of the cell colony, respectively (Toh et al., 2015). hESCs on Matrigel islands with a greater geometric anisotropy and a higher perimeter-to-area ratio exhibited a polarized distribution of integrins and E-cadherin. Under these conditions, competition between the adhesion receptors led to a heterogeneous distribution of phosphorylated myosin light chain and thus actomyosin tension, which correlated directly with the spatially restricted differentiation of the colony into mesoendoderm. The association of activated myosin II with integrins and regions of high stress supported mesoendoderm differentiation, whereas activated myosin II associated with cell–cell junctions and regions of lower stress leads to the maintenance of pluripotency (Toh et al., 2015). Thus, spatial polarization of integrins and cadherins creates a mechanical landscape that drives heterogeneity during stem cell differentiation. Using computational modeling and *in vitro* experimental approaches, Danuser and colleagues have recently quantified force transmission within multicellular clusters (Ng et al., 2014) and have demonstrated that the distribution of forces through E-cadherin cell–cell junctions is dynamic and fluctuates with local variations in cell–ECM adhesion and actomyosin contractility. Taken together, these studies demonstrate that a dialog between cadherins and integrins, which occurs through shifts in actomyosin contractility, determines the organization of molecular and mechanical signals at both the cell and tissue level.

Cadherin-dependent regulation of integrin activation and fibronectin matrix assembly

As discussed above, integrins and focal adhesion proteins can act as upstream regulators of cadherin dynamics, but there are also reports that cadherin itself functions as an upstream regulator of integrin activation and localization. Perhaps the clearest example of this is work by the Schwartz group on the response of endothelial cells to flow. Initial work in this system defined an intercellular mechanosensory complex, involving PECAM1, VE-cadherin and VEGF receptor (VEGFR), that transmits force, activates integrins and leads to alignment of endothelial cells in response to fluid shear stress (Tzima et al., 2005). In this model, mechanical forces exerted on endothelial cells by shear stress are directly transduced through PECAM1, VE-cadherin serves as an essential adaptor between PECAM1 and VEGFR, and VEGFR, in turn, activates PI3K and results in PI3K-mediated activation of integrins to regulate cell alignment in the direction of the shear stress. This crosstalk between VE-cadherin and integrins is coordinated in part by the Shc adaptor protein (Liu et al., 2008).

Using tension sensors for VE-cadherin and PECAM1, the same authors have subsequently demonstrated that shear stress elicits a tensional decrease in VE-cadherin, while simultaneously stimulating an increase in tension across junctional PECAM1 (Conway et al., 2013). More recently, the same group generated a series of VE-cadherin–N-cadherin chimaeras to identify the crucial domain(s) of VE-cadherin that are needed for its adaptor function. Both VEGFR2 and VEGFR3 bind specifically to the transmembrane

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domain of VE-cadherin and this binding facilitates the mechanical responses to fluid shear flow (Coon et al., 2015).

Another recent study has suggested an additional role for VE-cadherin in mechanotransduction (Barry et al., 2015). Using magnetic twisting cytometry to mechanically stimulate VE-cadherin adhesions in endothelial cells, these authors demonstrated that mechanical force on VE-cadherin triggers local recruitment of F-actin and vinculin to VE-cadherin-containing adherens junctions, as well as cell stiffening. This mechanosensitive response depends on Rho-associated protein kinase 1 (ROCK1) and PI3K signaling, and propagates global changes in cellular traction forces. Interestingly, both means of mechanical stimulation on VE-cadherin trigger downstream activation of the PI3K pathway, which in turn stimulates integrin activity. The different effects downstream of shear stress compared with the application of a local twisting force on VE-cadherin suggest that cells have evolved elaborate mechanisms to discriminate between different types of forces. However, how cells are able to transduce different mechanical stimuli through cadherins to integrins remains to be uncovered.

Cadherins can also regulate integrin function by organizing the ligands to which integrins bind. For example, cell–cell adhesion mediated by C-cadherin (also known as EP-cadherin), the major cadherin in *Xenopus* oocytes, increases mechanical tension to promote assembly of a fibronectin fibrillar matrix during *Xenopus* morphogenesis (Dzamba et al., 2009). In a recent study, Jülich and co-authors used fluorescence crosscorrelation spectroscopy (FCCS) to identify protein–protein interactions during zebrafish development. They found that α5 integrins (presumably α5β1) physically associated with each other on adjacent cells when the integrins were in an inactive conformation. There, N-cadherin stabilized the complex of inactive α5 integrins and inhibited fibronectin fibrillogenesis (Jülich et al., 2015). This interaction between N-cadherin and inactive α5 integrins biased the assembly of fibronectin matrix towards tissue surfaces that lack cell–cell adhesions. The author also showed that downregulation of N-cadherin was associated with α5 integrin activation and fibronectin matrix assembly and, ultimately, guided the ECM patterning necessary for body elongation and segmentation during zebrafish development. Whereas C-cadherin-generated tension is crucial for fibronectin remodeling during *Xenopus* development (Dzamba et al., 2009), N-cadherin blocks fibronectin fibrillogenesis in the developing zebrafish (Jülich et al., 2015). Thus, context-dependent molecular interactions and differential adhesion strength might be responsible for the different roles of cadherins in fibronectin matrix remodeling.

Interplay between focal adhesions and adherens junctions controls the polarization of forces and directional motility

Cell migration occurs through a complex array of mechanochemical signaling events that involve precise spatiotemporal coordination of adherens junctions, focal adhesions and intracellular tension. A fine balance between substrate traction and intercellular adhesion dictates collective migration within a tissue. This mechanical communication between cells was described by Liu et al., who have shown that increased endogenous stresses between cells leads to a proportional increase in the size of their adherens junctions (Liu et al., 2010). However, increased traction force from the ECM also affects cell–cell adhesions and results in a proportional increase in endogenous tension at cell–cell contacts (Maruthamuthu et al., 2011).

Desai and colleagues asked how cadherin-dependent adhesion might regulate cell polarity required for directed migration. Using micropatterned substrates to facilitate cell–cell contact and constrain cell protrusion, they demonstrated that E-cadherin is necessary for directing nuclear positioning, centrosome orientation and lamellipodial ruffling away from cell–cell junctions and towards the free boundaries of cells. Moreover, they showed that polarization is dependent on an intact actin cytoskeleton and Cdc42 activity (Desai et al., 2009).

Polarized cadherin adhesion strength and actomyosin contractility are also detectable during collective migration of border cells within the *Drosophila* ovary, an effect that is dependent on E-cadherin (Cai et al., 2014). Use of an E-cadherin tension sensor revealed that E-cadherin is under higher tension at the front of migrating border cells. Furthermore, the polarized tension on E-cadherin helps to localize activated Rac at the front of the border cells, thus forming a feed-forward loop that amplifies tension on E-cadherin-mediated adhesions. All these effects are crucial for directionally persistent migration (Cai et al., 2014). As Rac activity is also a major target of cell–ECM adhesion (see above), it is likely that cadherins and integrins within migrating cells are integrating Rac signals to direct motility.

During morphogenesis, cadherin-dependent adhesions are crucial for determining directionality in migrating cell populations within the embryo (Breau and Schneider-Maunoury, 2015). Transient N-cadherin-dependent adhesions formed between two cell types (embryonic neural crest cells and placode cells) in co-culture produce an asymmetric distribution of traction forces in placode cells and a significant reduction in focal adhesions at their shared intercellular junctions (Theveneau et al., 2013). This asymmetry in traction forces and focal adhesion distribution is dependent on N-cadherin, as reducing N-cadherin levels by treatment with morpholinos reversed the asymmetry. Thus, N-cadherin-mediated contact locally inhibits cell protrusions in placode cells and controls the direction in which they migrate (Theveneau et al., 2013).

Furthermore, signals that emanate from focal adhesions guide the formation of cadherin-based adhesions during gastrulation in *Xenopus* (Bjerke et al., 2014). There, deletion of FAK with morpholinos disrupts the actin cytoskeleton, alters the spatial distribution of keratin and reduces the binding of plakoglobin (also known as JUP and γ-catenin) to C-cadherin in mesendoderm explants. These changes are associated with impaired cell spreading and traction force generation and ultimately delay collective cell migration as well as disrupting mesendoderm tissue polarity (Bjerke et al., 2014).

Thus, these different studies provide compelling evidence that cadherin-specific reorganization alters the mechanical state of cells to guide collective cell movement. The degree of substrate traction and cell cohesion is finely tuned at focal adhesions and adherens junctions, respectively, and an intricate interplay between integrins and cadherins controls spatial stresses within the cell that guide migration.

Integrin–cadherin crosstalk as a new mechanism for spatial control of signaling and ECM deposition

Collectively, the studies reviewed here raise the intriguing idea that the interplay between forces, cadherins and integrins controls spatial signaling inside the cell and ECM remodeling outside the cell (Fig. 3). For example, vinculin associated with adherens junctions is phosphorylated at Y822 (Bays et al., 2014), whereas vinculin in focal adhesions is phosphorylated at Y100 and Y1065 (Auernheimer et al., 2015). To date, this differential phosphorylation has been associated with selective binding to β-catenin and actin, respectively, and viewed in the context of controlling the open versus closed

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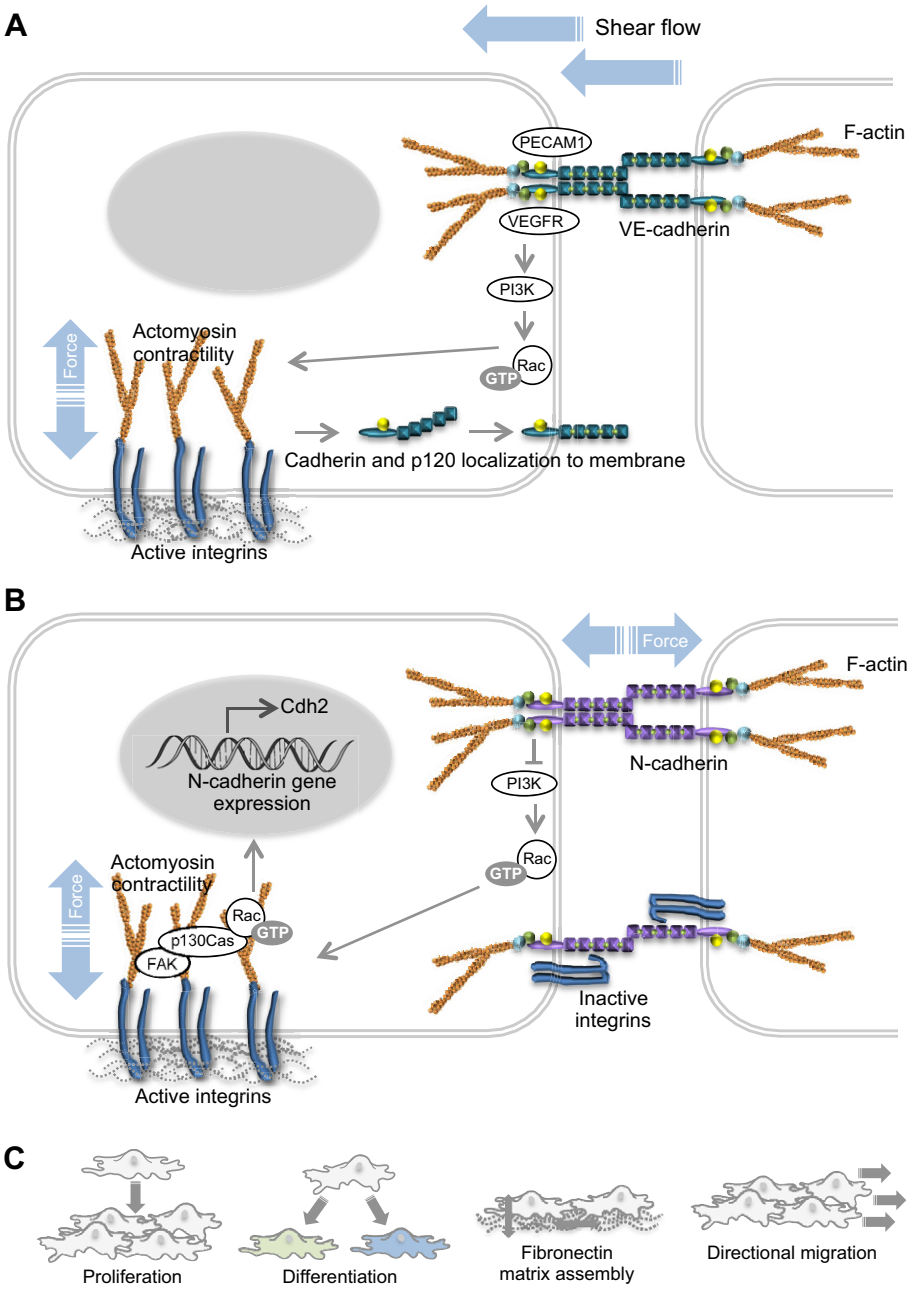

Fig. 3. Cross-regulatory pathways between focal adhesions and cadherins. Reciprocal regulation between focal adhesion proteins and cadherins occurs through different mechanochemical signals and is crucial to establish tensional homeostasis within cells. (A) A mechanosensory complex comprised of VE-cadherin, PECAM1 and VEGFR signals upstream of integrins to activate PI3K and Rac in response to shear flow. Conversely, actomyosin contractility drives integrin-dependent localization of VE-cadherin and p120-catenin to cell–cell junctions. Focal adhesion proteins linking integrins to F-actin are not shown. (B) Activation of the FAK–p130Cas–Rac signaling pathway at focal adhesions in response to substrate stiffness stimulates N-cadherin gene expression. Within adherens junctions, N-cadherin inhibits local PI3K and Rac activity. At adherens junctions, N-cadherin maintains α5 integrins in an inactive conformation to direct fibronectin matrix away from the junction. Focal adhesion proteins linking integrins to F-actin are not shown. (C) The coordinated effects described above in A and B regulate the spatial distribution of forces and signals to drive processes such as cell cycling, stem cell differentiation, fibronectin matrix assembly and collective cell migration.

vinculin configuration. However, we speculate that these site-specific phosphorylations of vinculin at adherens junctions and focal adhesions might also allow for the site-specific recruitment of distinct SH2-containing proteins, which could then initiate spatially distinct signaling pathways. Indeed, the recruitment of distinct DOCK–ELMO complexes to focal adhesions and adherens junctions (Toret et al., 2014) provides a conceptual framework for understanding how cadherins and integrins might spatially or kinetically direct Rac activity.

Subtle differences in the composition of N-cadherin junctions can contribute to changes in the spatial distribution of PI3K and myosin II. N-cadherin–p120-catenin complexes suppress local PI3K signaling to Rac, whereas N-cadherin–β-catenin complexes enrich adherens junctions with myosin II (Ouyang et al., 2013). Polarized Rac and myosin II signaling within cells might then generate spatially distinct intracellular regions of high and low tension within the cell. Adherens junctions containing N-cadherin can also confer spatial remodeling to the ECM by excluding α5β1 integrin from adherens junctions and by inhibiting the activation of α5 integrins within adherens junctions (Jülich et al., 2015). Forces on cadherins can even direct the spatial localization of macromolecular structures, such as nuclei, centrosomes and lamellipodia (Desai et al., 2009).

Context or cadherin-specificity might play a role in the effects of cadherin on spatial organization: whereas N-cadherin engagement inhibits fibronectin fibrillogenesis in zebrafish (Jülich et al., 2015), cell–cell adhesion mediated by C-cadherin promotes assembly of a fibronectin fibrillar matrix in *Xenopus* (Dzamba et al., 2009). Nevertheless, we envision that mechanically directed crosstalk between integrins and cadherins will cooperate with the more classically established biochemical effects of amino acid motifs (localization sequences, pleckstrin

homology domains, poly-proline sequences, etc.) and chemical modifications (phosphorylations, prenylation, myristylation, etc.) to localize subcellular signaling and ECM remodeling within cells and tissues. These interactions will ultimately guide cell fate and fundamental cell behavior (Fig. 3C).

Conclusions and perspectives

In this Commentary, we discuss how the mechanical landscape of the cell is continually modulated through multiple layers of crosstalk between integrins and cadherins. This crosstalk allows cells to actively adapt to changes in their physical and chemical environments. It is becoming increasingly clear that focal adhesions and adherens junctions share signaling molecules, yet some execute site-specific mechanical roles depending on their localization and (at least for vinculin) discrete post-translational modification. Coordinated interplay between integrins at focal adhesions and cadherins at adherens junctions also leads to a spatial organization of molecular signals and forces, and these guide diverse processes from cell fate determination and ECM patterning to directed migration.

The mechanical forces that are relayed between focal adhesions and adherens junctions ultimately regulate the transcriptional programs that control cell and tissue function. Two of the most-studied mechanosensitive transcriptional regulators are MRTF and the YAP or TAZ proteins (Janmey et al., 2013). MRTF-dependent transcription is modulated by actin cytoskeleton dynamics, whereas YAP and TAZ proteins can be regulated by cell–cell and cell–substratum adhesion, as well as by ECM stiffness, actin polymerization and mechanical forces (Imajo et al., 2012; Kim and Gumbiner, 2015; Kim et al., 2011; Schlegelmilch et al., 2011; Silvis et al., 2011; Wada et al., 2011). Elucidating the mechanistic links between adhesion receptor crosstalk, cytoskeletal tension and mechano-sensitive transcription will be crucial to fully understanding how integrins, cadherins, the actin cytoskeleton and intracellular forces control proliferation, migration and differentiation. Cytoskeletal tension is transduced by linker of nucleoskeleton and cytoskeleton (LINC) complexes in the nucleus. Nuclear mechanics is a rapidly growing field of study, and the chemical and mechanical pathways by which forces are transduced from adhesion receptors at the plasma membrane to LINC complexes at the nuclear membrane stands out as a fertile area of investigation.

Competing interests

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