Cell Adhesion & Migration 7:4, 345–350; July/August 2013; © 2013 Landes Bioscience

α-catenin, vinculin, and F-actin in strengthening E-cadherin cell–cell adhesions and mechanosensing

Sylvie Dufour[1,*], René-Marc Mège[2], and Jean Paul Thiery[3,4]

[1]Institut Curie/CNRS UMR144; Paris, France; [2]Institut Jacques Monod; Université Paris Diderot/CNRS; Paris, France; [3]Department of Biochemistry; Yong Loo Lin School of Medicine; Mechanobiology Institute; National University of Singapore; Singapore; [4]Institute of Molecular and Cell Biology; A*STAR (Agency for Science, Technology and Research); Singapore

Classical cadherins play a crucial role in establishing intercellular adhesion, regulating cortical tension, and maintaining mechanical coupling between cells. The mechanosensitive regulation of intercellular adhesion strengthening depends on the recruitment of adhesion complexes at adhesion sites and their anchoring to the actin cytoskeleton. Thus, the molecular mechanisms coupling cadherin-associated complexes to the actin cytoskeleton are actively being studied, with a particular focus on α-catenin and vinculin. We have recently addressed the role of these proteins by analyzing the consequences of their depletion and the expression of α-catenin mutants in the formation and strengthening of cadherin-mediated adhesions. We have used the dual pipette assay to measure the forces required to separate cell doublets formed in suspension. In this commentary, we briefly summarize the current knowledge on the role of α-catenin and vinculin in cadherin–actin cytoskeletal interactions. These data shed light on the tension-dependent contribution of α-catenin and vinculin in a mechanoresponsive complex that promotes the connection between cadherin and the actin cytoskeleton and their requirement in the development of adhesion strengthening.

Keywords: adhesion, cadherin, catenin, vinculin, mechanosensing, strength, force, cytoskeleton

Abbreviations: F-actin, filamentous actin; VASP, vasodilator-stimulated phosphoprotein; PKC, protein kinase C; VH, vinculin-homology; PIP2, Phosphatidylinositol 4,5-Bisphosphate; ZO-1, zonula occludens-1; Ipa-1, invasion plasmid antigen D of shigella; MDCK, Madin-Darby canine kidney cells; CHO, Chinese hamster ovary; Ncad-FC, N-cadherin-Fc fusion molecule; Ecad-FC, E-cadherin-Fc fusion molecule

Submitted: 04/22/13

Revised: 05/17/13

Accepted: 05/22/13

http://dx.doi.org/10.4161/cam.25139

*Correspondence to: Sylvie Dufour; Email: Sylvie.Dufour@curie.fr

Commentary to: Thomas WA, Boscher C, Chu YS, Cuvelier D, Martinez-Rico C, Seddiki R, et al. α-Catenin and vinculin cooperate to promote high E-cadherin-based adhesion strength. J Biol Chem 2013; 288:4957-69; PMID:23266828; http://dx.doi.org/10.1074/jbc.M112.403774

Forming and Strengthening E-Cadherin Cell–Cell Adhesions

E-cadherin is a calcium-dependent cell adhesion protein that controls interactions between epithelial cells. This protein is the prototypical classical cadherin[1] composed of an extracellular region (ecto-domain) carrying five "cadherin repeats," a transmembrane domain, and a cytoplasmic region. Classical cadherins form one of the six clusters evident in the phylogenetic tree of the superfamily of cadherins. This tree comprises several hundred proteins across 30 species characterized by the presence of at least two cadherin repeats.[2,3] Classical cadherins, herein referred to as "cadherins," play crucial roles during morphogenesis, and are involved in epithelial–mesenchymal transition, epithelial sheet maintenance, cell migration, tissue rearrangement, cell sorting, and synaptogenesis.[4] Their functional deregulation leads to several pathologies, including various cancers, neuropsychiatric disorders, and skin defects.[5-7]

In epithelia, junctional, and extra-junctional, E-cadherin molecules connect neighboring cells, make a bridge between their cortical actin cytoskeleton, and contribute to mechanical coupling within cells and mechanical resistance of epithelia to stretch. The ecto-domain of E-cadherin expressed at the cell–cell surface is responsible for homophilic interactions.[8] Despite interacting in trans, E-cadherin ectodomains are actually thought to cluster by *cis*-interaction.[9] Upon these ecto-domain interactions, the conserved cytoplasmic domain of E-cadherin recruits partners that contribute to its connection to the actin cytoskeleton, leading to the formation of

adhesion plaques that mature in adherens junctions in epithelia. The degree of growth or maturation of these plaques, which is a mechanosensitive process,[10] is thought to be related to the strength of the intercellular contacts.[11-14] In addition, cadherin-mediated adhesion activates signaling cascades that regulate gene expression and cytoskeleton remodeling.[15-20] However, coupling of the cadherin tail to the actin cytoskeleton at the cell surface remains the major factor contributing to junction stabilization (reviewed in ref. 21). The regulation of this coupling is also required for cell–cell adhesion strengthening,[22] collective cell migration,[23] and tissue rearrangement.[24]

The molecular interactions involved in E-cadherin-based adhesion strengthening and mechanosensing are studied mostly in cultured cells but also in vivo. Cadherin–actin coupling is not direct and requires interactions with cytoplasmic partners. The binding of the protein β-catenin at the catenin-binding domain of the cadherin tail is essential but not sufficient for cadherin-based adhesion. β-catenin recruits the vinculin-related protein, α-catenin, a multi-domain F-actin-binding protein with homology to vinculin crucial for strong cell–cell adhesion.[25,26] It was initially proposed that the cadherin/β-catenin complex is already assembled at the reticulum surface and exported at the cell membrane, and then associated to α-catenin, which directly binds the complex to F-actin.[27] Later, biochemical and structural studies argued for a more complex sequence of molecular interactions and the participation of additional partners such as vinculin, to the extent that α-catenin can interact with various partners and recruit them at cell–cell adhesion sites, in addition to its binding to F-actin (see below). However, very recent functional in vivo studies in *Drosophila* have re-established the tripartite cadherin/β-catenin/α-catenin complex as the central unit of cadherin adhesions, leaving a central open question as to the mode of association of this unit to the actin cytoskeleton.[28] In vertebrates, two isoforms of α-catenin coexist, αE- and αN-catenins, encoded by two separate genes.[29]

F-Actin Binding Proteins α-Catenin and Vinculin

Vinculin is a scaffolding protein recruited at integrin-associated complexes[30] that contributes to mechanotransduction at cell–matrix adhesions.[31] It is composed mostly of helical bundles that are organized toward a globular head at its N terminus comprising four vinculin-homology (VH) domains that carry binding sites for talin, α-actinin, IpaA, α-catenin, and β-catenin. A short linker connects the vinculin head to the rod-shaped tail domain (VH5) located at the C terminus, which carries binding sites regulating vinculin binding to actin and cytoskeletal remodeling proteins, including VASP, vinexin, ponsin, Arp2/3, as well for binding of PKC, paxillin, PiP2, and F-actin (reviewed in refs. 32 and 33). Vinculin exists in an auto-inhibitory form where the head and tail domains interact with high affinity. In this closed conformation state, vinculin cannot bind to its partners or to F-actin. The activation of vinculin by talin or its additional partners in a combinatorial activation model disrupts this auto-inhibitory interaction and exposes the head and tail domains for their binding to F-actin.[34-36] This protein is also recruited at cell–cell adhesion sites.[37]

α-catenins are members of the vinculin family of proteins, characterized by three successive VH domains from its N to C terminus.[29,38] The VH1 carries the β-catenin binding site, and a homodimerization site partly overlapping with the β-catenin, ajuba, and spectrin binding sites. The VH2 is subdivided into two parts: VH2a and VH2b. VH2a carries vinculin-, formin-, and α-actinin-binding sites whereas VH2b comprises a modulation domain that overlaps with the afadin-binding site. Finally, the VH3 domain interacts with eplin, ZO-1, and F-actin. Each of these domains is structured in α-helical bundles, as in the vinculin molecule. In vitro, purified αE-catenin is mostly monomeric but can homodimerize.[39] Only the monomeric form can bind to E-cadherin/β-catenin complexes, likely because the dimerization domain and the β-catenin-binding site partly overlap.[40] In contrast, αE-catenin homodimers bind to F-actin with high affinity.[41,42] Moreover, a recent study analyzing the crystal structure of full-length αE-catenin revealed that the αE-catenin dimer is asymmetric and that β-catenin binding to αE-catenin hinders the recognition of F-actin at the C-terminal site of αE-catenin.[42]

Thus, the tripartite cadherin/β-catenin/αE-catenin complex initially thought to directly connect cadherin to actin appears insufficient to tether cadherins to F-actin, even transiently.[43] The interaction of α-catenin with actin-binding proteins such as vinculin and eplin could fulfill the function to connect the cadherin–catenin complex to F-actin.[37,44,45] Alternatively, it was proposed that the transient dissociation of αE-catenin from the tripartite complex induces a local increase in monomers that favors the formation of homodimers in the vicinity of the cadherin-mediated adhesion sites, the latter negatively regulating Arp2/3 complex-dependent actin assembly and promoting actin bundling.[39,46] This assumption on the modulation of αE-catenin monomer vs. dimer concentration near adhesion sites was tested using a mathematical modeling based on experimental data.[47] The findings suggested that the local increase in homodimers near cell–cell adhesion sites depends on the transient homodimerization of αE-catenin at the cadherin/β-catenin/αE-catenin complex.[47] However, this does not provide any molecular cues for the tethering of cadherin-associated complexes to F-actin. Moreover, it has been recently shown that while neither *C. elegans* α-catenin nor vertebrate αN-catenin can assemble into homodimers, both proteins can sustain adherens junction assembly in a similar tripartite cadherin/β-catenin/α-catenin complex.[28,48] These findings suggest that both functions of αE-catenin (i.e., tethering the tripartite adhesion complex and regulating actin dynamics) could be uncoupled.

Vinculin only recently became an important candidate in asserting the link between cadherin complexes and F-actin. Biochemical and crystal structure analyses of the interaction of the VH2a domain of αE-catenin with the VH1 domain of vinculin revealed that an αE-catenin–vinculin complex can form and bind to F-actin by the C-terminal actin-binding domains

of either protein.[36,49,50] The binding of α-catenin on vinculin, playing the role of talin binding, may induce the head-tail opening of the molecule, increasing its affinity for F-actin. These studies further reported the existence of a vinculin–α-catenin heterotetramer, at least in vitro, which may contain a total of four functional binding sites for F-actin. As such, αE-catenin may exist within cells in at least three forms (monomeric, dimeric, and heterotetrameric) that could theoretically be diffusive or bound to cadherin–catenin complexes or F-actin with multiple valences. Whether this tetramer or the αE-catenin–vinculin heterodimers are bound to F-actin, associated to cadherin complexes, or both is unknown so far. A demonstration of the involvement of these F-actin adaptors in cadherin adhesion strengthening in cellulo have been hampered, in part, because α-catenin and vinculin also play biological roles unrelated to cadherin-based adhesions.

α-Catenin and Vinculin in Mechanosensing and Strengthening of Intercellular Adhesions

The formation of cadherin adhesions depends on the environmental stiffness. This has been previously demonstrated by culturing C2 myoblast cells on polyacrylamide gels or on flexible polydimethylsiloxane (PDMS) pillars coated with Ncad-Fc.[10] However, only on rigid surfaces do cells form cadherin adhesions connected to actin cables[51] that transmit myosin II-generated forces.[52] Cadherin adhesion formation following increased environmental stiffness correlates with an increase in cell traction forces at these sites, indicating that cadherins contribute to the distribution of intercellular contractility through a myosin II-dependent mechanism.[10,53] This process shares similarities with the mechanism of mechanosensing described for integrin-based adhesions,[54] which involve acto-myosin cables and mechanosensors located at the interface between adhesion receptors and the cytoskeleton. Borghi and coworkers[55] showed recently that the tension transmitted to cadherin-based adhesion through the actin cytoskeleton requires α-catenin. Independently, α-catenin was proposed to act as an acto-myosin-dependent mechanotransducer at adherens junctions.[56] Structural and biochemical analyses of the αE-catenin VH2 domain suggest that this domain may switch between a closed and open conformation in a force-dependent manner. In a closed conformation, the vinculin-binding domain is masked by the modulation domain, whereas in open conformation, this site is accessible for interaction with vinculin.[38,57,58] Furthermore, vinculin recruitment at adherens junctions is inhibited by blebbistatin, suggesting that the α-catenin–vinculin interaction is increased upon force at adhesion sites.[56,59] Thus, vinculin may contribute to cadherin-based mechanosensing and junctional remodeling.[60-62] However, there has been no direct demonstration of the contribution of the force-dependent conformational switch of α-catenin or of the recruitment of vinculin in mechanosensitive contact reinforcement at intercellular junctions since this point has been mostly investigated in the cases of cell–cell junctions relaxation using myosin II inhibition[56-62] or endothelial junction remodeling using thrombin treatment.[60]

In a recent study, we[63] directly analyzed the effect of the force exerted at cell–cell adhesions and showed that stretching of cell doublets increases vinculin recruitment at intercellular adhesion sites and induces a change of αE-catenin from a closed to an open conformation, as revealed by the recognition by a specific antibody directed against an epitope accessible only in the open conformation. This finding points to a force-driven, α-catenin-dependent mechanism for the recruitment of vinculin at cadherin-based adhesions. Furthermore, we directly addressed the roles of αE-catenin and vinculin in cadherin-based intercellular adhesion strength using the dual pipette assay.[63] In this assay, the force required to separate cell doublets is used as a read-out of adhesion strength.[64] We have shown in earlier studies a time-dependent adhesion strengthening that depends on cadherin expression levels, actin cytoskeleton dynamics, and cadherin–actin coupling.[22,65] Indeed, cytoplasmic domain-deleted E-cadherin does not allow cell–cell adhesion strengthening, whereas a chimeric E-cadherin–αE-catenin can rescue this process as efficiently as wild-type cadherin.[22] In addition, defective adhesions were described in α-catenin-depleted MDCK or CHO cells[66,67] and in DLD-1 cell variant (R2/7) lacking this protein.[68,69] Other studies showed that vinculin depletion produces defective adhesion in F9 and MDCK cells.[59,70] However, no one has tested whether αE-catenin and vinculin, as well as αE-catenin–vinculin interactions, were involved in cadherin adhesion strengthening. Thus, we further investigated the abilities of various αE-catenin mutants and fusion proteins to rescue E-cadherin adhesion strengthening. For this we used first α-catenin-depleted DLD-1-R2/7 cells that express E-cadherin but failed to strengthen their intercellular adhesions over time.[63] We expressed in these cells the wild-type αE-catenin or truncated proteins lacking actin-binding binding domain (aa 1–670 αE-catenin) or both this domain and the modulation domain (aa 1–510 αE-catenin) or an αE-catenin mutated in its vinculin-binding site by inverting the charge of aa residues at 12 positions between aa 326 and 348. We also expressed membrane-targeted αE-catenin such as chimeric E-cadherin fused after its transmembrane domain with wild-type αE-catenin or to a αE-catenin lacking the VH1 domain. The defective adhesion strengthening in these cells was rescued by the expression of soluble or membrane-targeted αE-catenin (E-cadherin αE-catenin chimera), the latter bearing or not the VH1 domain. This indicated that the constitutive recruitment of α-catenin at the cadherin-tail could fulfill the α-catenin function in adhesion strength independent of the α-catenin homodimer regulation of the actin cytoskeleton, as confirmed since then in vivo in *Drosophila* embryo epithelial cells.[28]

We further showed that αE-catenin mutant lacking both the modulation and actin-binding domains (aa 1–510 αE-catenin) rescued E-cadherin-mediated adhesion, the recruitment of vinculin at cadherin adhesions and allowed cells to spread on Ecad-Fc surfaces, while the mutant lacking only the actin-binding domain (aa 1–670 αE-catenin) failed to do so, reinforcing

the requirement of binding of vinculin to an open form of αE-catenin to trigger effective cell contact formation. However, the 1–510 mutant failed to rescue strong adhesion strength; indicating that vinculin recruitment at those sites is not sufficient for mechanosensitivity adhesion strengthening. αE-catenin mutated in the vinculin-binding domain similarly triggered cell–cell contact formation but fails to rescue strong adhesion strength. Furthermore, the vinculin depletion in E-cadherin expression expressing S180 cells that also express α-catenin or the expression of αE-catenin modulation domain (aa 491–660 αEcat) also leads to reduced adhesion strength within cell doublets. Together, these findings demonstrate for the first time that vinculin recruitment at cadherin-based adhesions contributes to intercellular adhesion strengthening and supports the hypothesis that both α-catenin and vinculin are required for and cooperate in intercellular contact maturation and strengthening, although the exact temporal and spatial details of this molecular cooperation is not understand so far and in particular cannot be directly linked to the available structural data.

In conclusion, a large body of very recent work using various cellular models, biophysical approaches, and in vivo studies illustrate that intercellular adhesion strengthening is dependent on α-catenin and vinculin and their cooperation to promote mechanoresponsive cadherin-actin linkage. However, the complexity of the interactome at cadherin-mediated adhesion sites[71] suggests that many other players could also contribute to intercellular adhesion development. Indeed, the presence of the F-actin-binding protein, eplin,[72] regulates mechanosensing at adherens junctions. Additional studies are still required to identify new catenin partners, and characterize their function and dynamics at those sites to help decipher the molecular mechanisms controlling adhesion development and mechanosensing at adherens junctions. Further investigations at the structural, cellular, and functional levels are also needed to understand the exact sequence of molecular interactions and conformational changes operating between the cadherin/β-catenin/α-catenin tripartite complex and vinculin and F-actin during cadherin adhesion formation, strengthening, and tension-dependent remodeling in cell collectives and tissues.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

Acknowledgments

This work was supported by grants from Agence Nationale de la Recherche (ANR) (PCVI program, PCV07-186757; BLANC program, 1515 02), PIC of Institut Curie, and Fondation Pierre Gilles de Gennes, as well as by grants from Association pour la Recherche sur le Cancer (ARC), Association Française contre les Myopathies (AFM), and IMCB A*STAR core funding.

References

1. Nollet F, Kools P, van Roy F. Phylogenetic analysis of the cadherin superfamily allows identification of six major subfamilies besides several solitary members. J Mol Biol 2000; 299:551-72; PMID:10835267; http://dx.doi.org/10.1006/jmbi.2000.3777
2. Hulpiau P, van Roy F. Molecular evolution of the cadherin superfamily. Int J Biochem Cell Biol 2009; 41:349-69; PMID:18848899; http://dx.doi.org/10.1016/j.biocel.2008.09.027
3. Hulpiau P, van Roy F. New insights into the evolution of metazoan cadherins. Mol Biol Evol 2011; 28:647-57; PMID:20817718; http://dx.doi.org/10.1093/molbev/msq233
4. Gumbiner BM. Regulation of cadherin-mediated adhesion in morphogenesis. Nat Rev Mol Cell Biol 2005; 6:622-34; PMID:16025097; http://dx.doi.org/10.1038/nrm1699
5. Redies C, Hertel N, Hübner CA. Cadherins and neuropsychiatric disorders. Brain Res 2012; 1470:130-44; PMID:22765916; http://dx.doi.org/10.1016/j.brainres.2012.06.020
6. Jeanes A, Gottardi CJ, Yap AS. Cadherins and cancer: how does cadherin dysfunction promote tumor progression? Oncogene 2008; 27:6920-9; PMID:19029934; http://dx.doi.org/10.1038/onc.2008.343
7. Amagai M, Stanley JR. Desmoglein as a target in skin disease and beyond. J Invest Dermatol 2012; 132:776-84; PMID:22189787; http://dx.doi.org/10.1038/jid.2011.390
8. Katsamba P, Carroll K, Ahlsen G, Bahna F, Vendome J, Posy S, et al. Linking molecular affinity and cellular specificity in cadherin-mediated adhesion. Proc Natl Acad Sci U S A 2009; 106:11594-9; PMID:19553217; http://dx.doi.org/10.1073/pnas.0905349106
9. Wu Y, Vendome J, Shapiro L, Ben-Shaul A, Honig B. Transforming binding affinities from three dimensions to two with application to cadherin clustering. Nature 2011; 475:510-3; PMID:21796210; http://dx.doi.org/10.1038/nature10183
10. Ladoux B, Anon E, Lambert M, Rabodzey A, Hersen P, Buguin A, et al. Strength dependence of cadherin-mediated adhesions. Biophys J 2010; 98:534-42; PMID:20159149; http://dx.doi.org/10.1016/j.bpj.2009.10.044
11. Adams CL, Chen YT, Smith SJ, Nelson WJ. Mechanisms of epithelial cell-cell adhesion and cell compaction revealed by high-resolution tracking of E-cadherin-green fluorescent protein. J Cell Biol 1998; 142:1105-19; PMID:9722621; http://dx.doi.org/10.1083/jcb.142.4.1105
12. Vasioukhin V, Bauer C, Yin M, Fuchs E. Directed actin polymerization is the driving force for epithelial cell-cell adhesion. Cell 2000; 100:209-19; PMID:10660044; http://dx.doi.org/10.1016/S0092-8674(00)81559-7
13. Lambert M, Padilla F, Mege RM. Immobilized dimers of N-cadherin-fc chimera mimic cadherin-mediated cell contact formation: contribution of both outiside-in and inside-out signals. J Cell Sci 2000; 113:2207-19; PMID:10825293
14. Lambert M, Thoumine O, Brevier J, Choquet D, Riveline D, Mège RM. Nucleation and growth of cadherin adhesions. Exp Cell Res 2007; 313:4025-40; PMID:17765222; http://dx.doi.org/10.1016/j.yexcr.2007.07.035
15. Braga VM, Machesky LM, Hall A, Hotchin NA. The small GTPases Rho and Rac are required for the establishment of cadherin-dependent cell-cell contacts. J Cell Biol 1997; 137:1421-31; PMID:9182672; http://dx.doi.org/10.1083/jcb.137.6.1421
16. Shewan AM, Maddugoda M, Kraemer A, Stehbens SJ, Verma S, Kovacs EM, et al. Myosin 2 is a key Rho kinase target necessary for the local concentration of E-cadherin at cell-cell contacts. Mol Biol Cell 2005; 16:4531-42; PMID:16030252; http://dx.doi.org/10.1091/mbc.E05-04-0330
17. Kovacs EM, Ali RG, McCormack AJ, Yap AS. E-cadherin homophilic ligation directly signals through Rac and phosphatidylinositol 3-kinase to regulate adhesive contacts. J Biol Chem 2002; 277:6708-18; PMID:11744701; http://dx.doi.org/10.1074/jbc.M109640200
18. Kovacs EM, Goodwin M, Ali RG, Paterson AD, Yap AS. Cadherin-directed actin assembly: E-cadherin physically associates with the Arp2/3 complex to direct actin assembly in nascent adhesive contacts. Curr Biol 2002; 12:379-82; PMID:11882288; http://dx.doi.org/10.1016/S0960-9822(02)00661-9
19. Kim NG, Koh E, Chen X, Gumbiner BM. E-cadherin mediates contact inhibition of proliferation through Hippo signaling-pathway components. Proc Natl Acad Sci U S A 2011; 108:11930-5; PMID:21730131; http://dx.doi.org/10.1073/pnas.1103345108
20. Cavallaro U, Christofori G. Cell adhesion and signalling by cadherins and Ig-CAMs in cancer. Nat Rev Cancer 2004; 4:118-32; PMID:14964308; http://dx.doi.org/10.1038/nrc1276
21. Mège RM, Gavard J, Lambert M. Regulation of cell-cell junctions by the cytoskeleton. Curr Opin Cell Biol 2006; 18:541-8; PMID:16905303; http://dx.doi.org/10.1016/j.ceb.2006.08.004
22. Chu YS, Thomas WA, Eder O, Pincet F, Perez E, Thiery JP, et al. Force measurements in E-cadherin-mediated cell doublets reveal rapid adhesion strengthened by actin cytoskeleton remodeling through Rac and Cdc42. J Cell Biol 2004; 167:1183-94; PMID:15596540; http://dx.doi.org/10.1083/jcb.200403043
23. Friedl P, Gilmour D. Collective cell migration in morphogenesis, regeneration and cancer. Nat Rev Mol Cell Biol 2009; 10:445-57; PMID:19546857; http://dx.doi.org/10.1038/nrm2720
24. Cavey M, Rauzi M, Lenne PF, Lecuit T. A two-tiered mechanism for stabilization and immobilization of E-cadherin. Nature 2008; 453:751-6; PMID:18480755; http://dx.doi.org/10.1038/nature06953
25. Ozawa M, Baribault H, Kemler R. The cytoplasmic domain of the cell adhesion molecule uvomorulin associates with three independent proteins structurally related in different species. EMBO J 1989; 8:1711-7; PMID:2788574

26. Imamura Y, Itoh M, Maeno Y, Tsukita S, Nagafuchi A. Functional domains of α-catenin required for the strong state of cadherin-based cell adhesion. J Cell Biol 1999; 144:1311-22; PMID:10087272; http://dx.doi.org/10.1083/jcb.144.6.1311
27. Chen YT, Stewart DB, Nelson WJ. Coupling assembly of the E-cadherin/beta-catenin complex to efficient endoplasmic reticulum exit and basal-lateral membrane targeting of E-cadherin in polarized MDCK cells. J Cell Biol 1999; 144:687-99; PMID:10037790; http://dx.doi.org/10.1083/jcb.144.4.687
28. Desai R, Sarpal R, Ishiyama N, Pellikka M, Ikura M, Tepass U. Monomeric α-catenin links cadherin to the actin cytoskeleton. Nat Cell Biol 2013; 15:261-73; PMID:23417122; http://dx.doi.org/10.1038/ncb2685
29. Kobielak A, Fuchs E. Alpha-catenin: at the junction of intercellular adhesion and actin dynamics. Nat Rev Mol Cell Biol 2004; 5:614-25; PMID:15366705; http://dx.doi.org/10.1038/nrm1433
30. Geiger B, Tokuyasu KT, Dutton AH, Singer SJ. Vinculin, an intracellular protein localized at specialized sites where microfilament bundles terminate at cell membranes. Proc Natl Acad Sci U S A 1980; 77:4127-31; PMID:6776523; http://dx.doi.org/10.1073/pnas.77.7.4127
31. Geiger B, Bershadsky A. Assembly and mechanosensory function of focal contacts. Curr Opin Cell Biol 2001; 13:584-92; PMID:11544027; http://dx.doi.org/10.1016/S0955-0674(00)00255-6
32. Leerberg JM, Yap AS. Vinculin, cadherin mechanotransduction and homeostasis of cell-cell junctions. Protoplasma 2012; 250:817-29; PMID:23274283; http://dx.doi.org/10.1007/s00709-012-0475-6
33. Peng X, Nelson ES, Maiers JL, DeMali KA. New insights into vinculin function and regulation. Int Rev Cell Mol Biol 2011; 287:191-231; PMID:21414589; http://dx.doi.org/10.1016/B978-0-12-386043-9.00005-0
34. Bakolitsa C, Cohen DM, Bankston LA, Bobkov AA, Cadwell GW, Jennings L, et al. Structural basis for vinculin activation at sites of cell adhesion. Nature 2004; 430:583-6; PMID:15195105; http://dx.doi.org/10.1038/nature02610
35. Chen H, Choudhury DM, Craig SW. Coincidence of actin filaments and talin is required to activate vinculin. J Biol Chem 2006; 281:40389-98; PMID:17074767; http://dx.doi.org/10.1074/jbc.M607324200
36. Choi HJ, Pokutta S, Cadwell GW, Bobkov AA, Bankston LA, Liddington RC, et al. αE-catenin is an autoinhibited molecule that coactivates vinculin. Proc Natl Acad Sci U S A 2012; 109:8576-81; PMID:22586082; http://dx.doi.org/10.1073/pnas.1203906109
37. Hazan RB, Kang L, Roe S, Borgen PI, Rimm DL. Vinculin is associated with the E-cadherin adhesion complex. J Biol Chem 1997; 272:32448-53; PMID:9405455; http://dx.doi.org/10.1074/jbc.272.51.32448
38. Sarpal R, Pellikka M, Patel RR, Hui FY, Godt D, Tepass U. Mutational analysis supports a core role for Drosophila α-catenin in adherens junction function. J Cell Sci 2012; 125:233-45; PMID:22266901; http://dx.doi.org/10.1242/jcs.096644
39. Drees F, Pokutta S, Yamada S, Nelson WJ, Weis WI. α-catenin is a molecular switch that binds E-cadherin-β-catenin and regulates actin-filament assembly. Cell 2005; 123:903-15; PMID:16325583; http://dx.doi.org/10.1016/j.cell.2005.09.021
40. Koslov ER, Maupin P, Pradhan D, Morrow JS, Rimm DL. Alpha-catenin can form asymmetric homodimeric complexes and/or heterodimeric complexes with beta-catenin. J Biol Chem 1997; 272:27301-6; PMID:9341178; http://dx.doi.org/10.1074/jbc.272.43.27301
41. Rimm DL, Koslov ER, Kebriaei P, Cianci CD, Morrow JS. Alpha 1(E)-catenin is an actin-binding and -bundling protein mediating the attachment of F-actin to the membrane adhesion complex. Proc Natl Acad Sci U S A 1995; 92:8813-7; PMID:7568023; http://dx.doi.org/10.1073/pnas.92.19.8813
42. Rangarajan ES, Izard T. Dimer asymmetry defines α-catenin interactions. Nat Struct Mol Biol 2013; 20:188-93; PMID:23292143; http://dx.doi.org/10.1038/nsmb.2479
43. Yamada S, Pokutta S, Drees F, Weis WI, Nelson WJ. Deconstructing the cadherin-catenin-actin complex. Cell 2005; 123:889-901; PMID:16325582; http://dx.doi.org/10.1016/j.cell.2005.09.020
44. Abe K, Takeichi M. EPLIN mediates linkage of the cadherin catenin complex to F-actin and stabilizes the circumferential actin belt. Proc Natl Acad Sci U S A 2008; 105:13-9; PMID:18093941; http://dx.doi.org/10.1073/pnas.0710504105
45. Weiss EE, Kroemker M, Rüdiger AH, Jockusch BM, Rüdiger M. Vinculin is part of the cadherin-catenin junctional complex: complex formation between alpha-catenin and vinculin. J Cell Biol 1998; 141:755-64; PMID:9566974; http://dx.doi.org/10.1083/jcb.141.3.755
46. Thomas WA, Boscher C, Chu YS, Cuvelier D, Martinez-Rico C, Seddiki R, et al. α-Catenin and vinculin cooperate to promote high E-cadherin-based adhesion strength. J Biol Chem 2013; 288:4957-69; PMID:23266828; http://dx.doi.org/10.1074/jbc.M112.403774
47. Dawes AT. A mathematical model of α-catenin dimerization at adherens junctions in polarized epithelial cells. J Theor Biol 2009; 257:480-8; PMID:19135061; http://dx.doi.org/10.1016/j.jtbi.2008.11.028
48. Kwiatkowski AV, Maiden SL, Pokutta S, Choi HJ, Benjamin JM, Lynch AM, et al. In vitro and in vivo reconstitution of the cadherin-catenin-actin complex from Caenorhabditis elegans. Proc Natl Acad Sci U S A 2010; 107:14591-6; PMID:20689042; http://dx.doi.org/10.1073/pnas.1007349107
49. Rangarajan ES, Izard T. The cytoskeletal protein α-catenin unfurls upon binding to vinculin. J Biol Chem 2012; 287:18492-9; PMID:22493458; http://dx.doi.org/10.1074/jbc.M112.351023
50. Peng X, Maiers JL, Choudhury D, Craig SW, DeMali KA. α-Catenin uses a novel mechanism to activate vinculin. J Biol Chem 2012; 287:7728-37; PMID:22235119; http://dx.doi.org/10.1074/jbc.M111.297481
51. Gavard J, Lambert M, Grosheva I, Marthiens V, Irinopoulou T, Riou JF, et al. Lamellipodium extension and cadherin adhesion: two cell responses to cadherin activation relying on distinct signalling pathways. J Cell Sci 2004; 117:257-70; PMID:14657280; http://dx.doi.org/10.1242/jcs.00857
52. Ganz A, Lambert M, Saez A, Silberzan P, Buguin A, Mège RM, et al. Traction forces exerted through N-cadherin contacts. Biol Cell 2006; 98:721-30; PMID:16895521; http://dx.doi.org/10.1042/BC20060039
53. Shewan AM, Maddugoda M, Kraemer A, Stehbens SJ, Verma S, Kovacs EM, et al. Myosin 2 is a key Rho kinase target necessary for the local concentration of E-cadherin at cell-cell contacts. Mol Biol Cell 2005; 16:4531-42; PMID:16030252; http://dx.doi.org/10.1091/mbc.E05-04-0330
54. Bershadsky A, Kozlov M, Geiger B. Adhesion-mediated mechanosensitivity: a time to experiment, and a time to theorize. Curr Opin Cell Biol 2006; 18:472-81; PMID:16930976; http://dx.doi.org/10.1016/j.ceb.2006.08.012
55. Borghi N, Sorokina M, Shcherbakova OG, Weis WI, Pruitt BL, Nelson WJ, et al. E-cadherin is under constitutive actomyosin-generated tension that is increased at cell-cell contacts upon externally applied stretch. Proc Natl Acad Sci U S A 2012; 109:12568-73; PMID:22802638; http://dx.doi.org/10.1073/pnas.1204390109
56. Yonemura S, Wada Y, Watanabe T, Nagafuchi A, Shibata M. α-Catenin as a tension transducer that induces adherens junction development. Nat Cell Biol 2010; 12:533-42; PMID:20453849; http://dx.doi.org/10.1038/ncb2055
57. Yang J, Dokurno P, Tonks NK, Barford D. Crystal structure of the M-fragment of alpha-catenin: implications for modulation of cell adhesion. EMBO J 2001; 20:3645-56; PMID:11447106; http://dx.doi.org/10.1093/emboj/20.14.3645
58. Pokutta S, Drees F, Takai Y, Nelson WJ, Weis WI. Biochemical and structural definition of the l-afadin- and actin-binding sites of alpha-catenin. J Biol Chem 2002; 277:18868-74; PMID:11907041; http://dx.doi.org/10.1074/jbc.M201463200
59. Sumida GM, Tomita TM, Shih W, Yamada S. Myosin II activity dependent and independent vinculin recruitment to the sites of E-cadherin-mediated cell-cell adhesion. BMC Cell Biol 2011; 12:48; PMID:22054176; http://dx.doi.org/10.1186/1471-2121-12-48
60. Huveneers S, Oldenburg J, Spanjaard E, van der Krogt G, Grigoriev I, Akhmanova A, et al. Vinculin associates with endothelial VE-cadherin junctions to control force-dependent remodeling. J Cell Biol 2012; 196:641-52; PMID:22391038; http://dx.doi.org/10.1083/jcb.201108120
61. Twiss F, Le Duc Q, Van Der Horst S, Tabdili H, Van Der Krogt G, Wang N, et al. Vinculin-dependent Cadherin mechanosensing regulates efficient epithelial barrier formation. Biol Open 2012; 1:1128-40; PMID:23213393; http://dx.doi.org/10.1242/bio.20122428
62. le Duc Q, Shi Q, Blonk I, Sonnenberg A, Wang N, Leckband D, 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 2010; 189:1107-15; PMID:20584916; http://dx.doi.org/10.1083/jcb.201001149
63. Thomas WA, Boscher C, Chu YS, Cuvelier D, Martinez-Rico C, Seddiki R, et al. α-Catenin and vinculin cooperate to promote high E-cadherin-based adhesion strength. J Biol Chem 2013; 288:4957-69; PMID:23266828; http://dx.doi.org/10.1074/jbc.M112.403774
64. Chu YS, Dufour S, Thiery JP, Perez E, Pincet F. Johnson-Kendall-Roberts theory applied to living cells. Phys Rev Lett 2005; 94:028102; PMID:15698233; http://dx.doi.org/10.1103/PhysRevLett.94.028102
65. Chu YS, Eder O, Thomas WA, Simcha I, Pincet F, Ben-Ze'ev A, et al. Prototypical type I E-cadherin and type II cadherin-7 mediate very distinct adhesiveness through their extracellular domains. J Biol Chem 2006; 281:2901-10; PMID:16253998; http://dx.doi.org/10.1074/jbc.M506185200
66. Capaldo CT, Macara IG. Depletion of E-cadherin disrupts establishment but not maintenance of cell junctions in Madin-Darby canine kidney epithelial cells. Mol Biol Cell 2007; 18:189-200; PMID:17093058; http://dx.doi.org/10.1091/mbc.E06-05-0471
67. Bajpai S, Correia J, Feng Y, Figueiredo J, Sun SX, Longmore GD, et al. alpha-Catenin mediates initial E-cadherin-dependent cell-cell recognition and subsequent bond strengthening. Proc Natl Acad Sci U S A 2008; 105:18331-6; PMID:19017792; http://dx.doi.org/10.1073/pnas.0806783105

68. Vermeulen SJ, Bruyneel EA, Bracke ME, De Bruyne GK, Vennekens KM, Vleminckx KL, et al. Transition from the noninvasive to the invasive phenotype and loss of alpha-catenin in human colon cancer cells. Cancer Res 1995; 55:4722-8; PMID:7553655
69. Shimoyama Y, Nagafuchi A, Fujita S, Gotoh M, Takeichi M, Tsukita S, et al. Cadherin dysfunction in a human cancer cell line: possible involvement of loss of alpha-catenin expression in reduced cell-cell adhesiveness. Cancer Res 1992; 52:5770-4; PMID:1394201
70. Tozeren A, Wu S, Hoxter B, Xu W, Adamson ED, Byers SW. Vinculin and cell-cell adhesion. Cell Adhes Commun 1998; 5:49-59; PMID:9638341; http://dx.doi.org/10.3109/15419069809005598
71. Zaidel-Bar R. Cadherin adhesome at a glance. J Cell Sci 2013; 126:373-8; PMID:23547085; http://dx.doi.org/10.1242/jcs.111559
72. Taguchi K, Ishiuchi T, Takeichi M. Mechanosensitive EPLIN-dependent remodeling of adherens junctions regulates epithelial reshaping. J Cell Biol 2011; 194:643-56; PMID:21844208; http://dx.doi.org/10.1083/jcb.201104124