Force Generation, Transmission, and Integration during Cell and Tissue Morphogenesis

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Abstract
Cell shape changes underlie a huge set of biological processes ranging from cell division to cell motility. Stereotyped patterns of cell shape changes also determine tissue remodeling events such as extension or invagination. In vitro and cell culture systems have been essential to understanding the fundamental physical principles of subcellular mechanics. These are now complemented by studies in developing organisms that emphasize how cell and tissue morphogenesis emerge from the interplay between force-generating machines, such as actomyosin networks, and adhesive clusters that transmit tensile forces at the cell cortex and stabilize cell-cell and cell-substrate interfaces. Both force production and transmission are self-organizing phenomena whose adaptive features are essential during tissue morphogenesis. A new era is opening that emphasizes the similarities of and allows comparisons between distant dynamic biological phenomena because they rely on core machineries that control universal features of cytomechanics.
INTRODUCTION

During development, cells assemble into tissues that are extensively remodeled to yield the characteristic shapes of embryos and organs. How local cell shape changes and regulation of cell-cell contacts drive tissue-level deformations such as invagination or elongation is a fundamental question that has fascinated scientists for decades. We also have learned much about how individual cell shape changes stem from interactions between cytoskeletal elements, motors, and adhesion molecules. Now that the fundamental and evolutionarily conserved molecular constituents driving cell motility and deformation have been identified, important challenges are to explain how subcellular mechanics emerge from the interaction between these molecules and how they drive cell and tissue morphogenesis.

Historically, studies of tissue morphogenesis focused on describing the cellular basis of tissue shape changes as well as the genes and pathways that control them in space and time. Recent advances in live microscopy have opened the door to more quantitative investigation of subcellular dynamics. As a result, it becomes possible to integrate different scales of morphogenetic description into a single coherent framework. This review attempts to synthesize insights from physical and biochemical studies of cytoskeletal dynamics in cultured cells or cell-free systems with those from cell biological and genetic studies of morphogenesis in whole organisms. We look beyond earlier physical descriptions of tissue morphogenesis based on energy minimization principles (reviewed in Lecuit & Lenne 2007) to focus instead on the dynamics of subcellular and tissue shape changes. Rather than focusing on the conservation of specific proteins, we emphasize organizational principles underlying force generation, force transmission, and tissue-level integration by drawing parallels between different molecular assemblies.

ACTIN-BASED FORCE PRODUCTION AS A SELF-ORGANIZING PHENOMENON

Embryonic cells assemble, use, and disassemble two kinds of actin-based machines as they divide, move, and change shape. Local actin filament assembly powers cellular protrusions; interactions between nonmuscle myosin II and arrays of actin filaments, drives local contractility. Cells regulate force production by controlling the assembly and architecture of actin filament arrays and by regulating myosin II’s motor activity and assembly state. Local protrusive and contractile forces in turn lead to...
deformation and flows that redistribute or reorganize actin filaments, motors, and their various regulators. Thus, force production in embryonic cells involves locally tuned, self-organized machines that evolve minute to minute through the continuous interplay between biochemical regulation and cytomechanics.

**Structure and Regulation of Nonmuscle Myosin II**

Nonmuscle myosin II is a hexamer composed of two heavy chains, two essential light chains, and two myosin regulatory light chains (MRLCs) (Sellers 2000). The heavy chain contains an N-terminal head domain that couples ATP hydrolysis to filament binding, conformational change (the “power stroke”), and filament release; an intermediate neck domain that binds the light chains; and an extended C-terminal α-helical tail domain that mediates coiled-coil dimerization to form a myosin II homodimer. The tail domain mediates self-assembly of homodimers into bipolar minifilaments containing a few dozen heads (Mahajan & Pardee 1996, Niederman & Pollard 1975, Verkhovsky et al. 1995), which are fundamental units of contractile force generation in nonmuscle cells (Figure 1). Minifilament assembly is essential for force production by myosin II because it converts a unipolar and nonprocessive hexamer into a highly processive bipolar machine.

Although F-actin and ATPase-dependent force generation and minifilament assembly are characteristic of all myosin IIs, the details vary across different subtypes and species. Indeed, comparative analyses of the three vertebrate isoforms (NM2A–C) reveal variations in mechanochemistry (Kovacs et al. 2003, 2007; Wang et al. 2003) and minifilament organization (Ronen & Ravid 2009) that accompany significant differences in localization and function (Maupin et al. 1994, Rolo et al. 2009, Skoglund et al. 2008, Smutyń et al. 2010, Vicente-Manzanares et al. 2007).

A primary mode of nonmuscle myosin II regulation is phosphorylation of the RLC on two highly conserved residues (serine 19 and threonine 18 in vertebrate myosin IIs). Numerous kinases operate in diverse regulatory pathways to target these residues, including the myosin light chain kinase (MLCK; activated by Ca\(^{2+}\)/calmodulin), Rho kinase and citron kinase (activated by RhoA), myotonia dystrophy-related Cdc-42-binding kinase (MRCK; activated by Cdc-42) and others (Leung et al. 1998, Matsumura 2005). In vitro studies showed that when the MRLC is unphosphorylated, the myosin II homodimer adopts a folded conformation in which head-head associations inhibit F-actin binding and ATPase activity and head-tail association prevents minifilament assembly (Jung et al. 2008). MRLC phosphorylation promotes unfolding of the homodimer to relieve this inhibition (Figure 1; Craig et al. 2008).
Contractility: the ability of a material to contract by producing active internal stress (e.g., through local actomyosin interactions)

1983). The same transition likely occurs in vivo, although this remains to be demonstrated.

Another mode of regulation involves phosphorylation of the heavy chain, which destabilizes existing minifilaments and/or prevents their de novo assembly (Figure 1). This was first demonstrated in Dictyostelium (Egelhoff et al. 1993, Yumura et al. 2005). However, more recent studies of vertebrate nonmuscle myosin IIs have identified numerous phosphorylation sites within the C-terminal rod and nonhelical tailpiece that a variety of kinases target in different physiological contexts (reviewed in Vicente-Manzanares et al. 2009). Loss of this function leads to overaccumulation and/or mislocalization of myosin II, which suggests that myosin II turnover is essential for its redistribution as cells move or transit between different contractile activities (Breckenridge et al. 2009, Clark et al. 2008, Even-Faitelson et al. 2005), but the underlying mechanisms are still poorly understood.

Finally, F-actin can directly and efficiently promote minifilament assembly in vitro (Mahajan & Pardee 1996), presumably by concentrating encounters among myosin II homodimers. On purely kinetic grounds, minifilament assembly should increase the affinity of myosin II for F-actin and other cortical binding partners and thus bias minifilament accumulation to regions where these binding sites are denser. Indeed, although F-actin is not strictly required for equatorially biased myosin II recruitment during cytokinesis, most cells require it for normal levels and persistence (Dean et al. 2005, Foe & von Dassow 2008, Kamijo et al. 2006, Zang & Spudich 1998). In Dictyostelium, cortical recruitment of myosin II (but not minifilament assembly) in response to chemoattractant is abolished by depolymerizing F-actin or by removing the globular F-actin-binding head domain (Levi et al. 2002). In fly embryos, actin depolymerization causes a similar loss of cortical myosin II (Bertet et al. 2009), whereas experimentally induced increases in F-actin lead to increased accumulation of myosin II (Bertet et al. 2009, Homem & Peifer 2008). Whether these observations reflect purely kinetic effects on myosin II recruitment or something less direct remains to be seen. Understanding how local assembly/disassembly, recruitment to specific binding sites, and active redistribution (see below) shape myosin II distributions within embryonic cells is an essential goal for future studies.

Modes of Actin Filament Assembly

Cells use different actin filament arrays for different modes of contractile and protrusive force generation (Figure 2). Most cells maintain a pool of actin monomers that are primed for rapid addition to the fast-growing (barbed) ends of existing actin filaments. However, capping protein binds to most barbed ends to restrict monomer addition and filament growth (Pollard 2007). Cells overcome this restriction through nucleation or severing factors that create new barbed ends, or elongation factors that compete with capping protein to allow monomer addition to existing ends. Controlling the activities of these factors in space and time is the first step in defining the architecture of filament arrays.

ARP2/3

The most common and best-understood regulators of filament nucleation are the ARP2/3 complex and formins (reviewed in Chesarone & Goode 2009, Pollard 2007). Several other nucleators have been identified, but these do not appear to operate in most embryonic tissues (for recent reviews, see Chesarone & Goode 2009, Renault et al. 2008). The ARP2/3 complex nucleates new side branches on existing filaments. It remains tightly bound to both the mother filament and the pointed end of the new daughter filament, which leaves the barbed ends free to grow for a short time before they are capped. Combined with the actions of proteins such as ADF/cofilin, Aip1, and coronin that promote debranching and filament disassembly, ARP2/3 function promotes the formation of densely branched, rapidly treadmilling, filament arrays in which barbed-end growth rectifies local
fluctuations in the plasma membrane, the filament array, or intracellular obstacles (e.g., vesicles or pathogens) to generate protrusive forces (Mogilner & Oster 2003, Peskin et al. 1993, Shaevitz & Fletcher 2007; Figure 2a).

ARP2/3 activation requires members of the WASP and WAVE/SCAR families, which bind ARP2/3 and actin monomers to stabilize an essential intermediate step in initiation of a new side branch (Pollard 2007). WASP and WAVE are in turn activated locally—often synergistically—by binding small Rho family GTPases and phosphoinositides, and by phosphorylation (Stradal & Scita 2006). Additional control may come via spatial localization of ARP2/3 itself, e.g., via binding to cadherins (Kovacs et al. 2002). This requirement for multiple inputs to WASP or WAVE plus the simultaneous presence of WASP/WAVE, ARP2/3, and F-actin filaments allows cells to achieve tight spatial and temporal control over actin assembly. Confining activation of ARP2/3 to the plasma membrane ensures that protrusive force generation follows the membrane during protrusive extension or the propulsion of intracellular vesicles. For nonextending membranes, e.g., at domains of contact between adjacent cells, this confinement promotes the formation of subsurface layers or patches of branched actin that could serve as scaffolds to facilitate recruitment/clustering of signaling molecules, adhesion proteins, or myosin II, or that could stabilize points of cell-cell or cell-substratum contact.

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**Figure 2**

Modes of actin filament assembly and formation of protrusive and contractile arrays. (a) Activation of ARP2/3 at membranes promotes branched actin assembly to drive lamellar protrusion. Activation of formins promotes nucleation and elongation of unbranched filaments that are organized by fascin into stiff protrusive bundles (b) or organized by cross-linkers and myosin II into contractile networks (c) or bundles (d). Debranching of networks assembled by ARP2/3 could also contribute to formation of contractile networks. Abbreviations: FH1, formin homology; NPF, nucleation-promoting factor; PIP2, phosphatidylinositol 4,5-biphosphate.

**Formins**

The formins are a family of related proteins that share highly conserved formin homology (FH1 and FH2) domains (Liu et al. 2008).
Formins nucleate new filaments and remain associated with their growing ends, where they cooperate with profilin to promote monomer addition and rapid elongation of long, unbranched filaments (Figure 2b, Kovar et al. 2006), which form the raw material for higher-order protrusive and contractile structures including filopodia (Yang et al. 2007), the cytokinesis contractile ring (Watanabe et al. 2008), stress fibers (Watanabe et al. 1999), circumapical filament bundles (Homem & Peifer 2008), and more loosely organized filament networks that mediate contractility and flow in oocytes and early embryos (Schuh & Ellenberg 2008; Figure 2b–d).

Most of what we know about formin regulation comes from studies of the Diaphanous-related formins (DRFs). In isolation, DRFs adopt an inactive folded configuration mediated by the intramolecular association of the C- and N-terminal domains. Active (GTP-bound) Rho partially relieves this inhibition by binding to a conserved N-terminal GTP-binding domain to promote unfolding and activation of the catalytic C terminus (Li & Higgs 2003). This dual ability to promote both local myosin II activation and formin-dependent filament assembly makes RhoA a potent activator of contractile force generation (Watanabe et al. 1999 and see below). However, additional inputs are required for full activation in many cases (Higashi et al. 2010, Watanabe et al. 2010; see Chesarone & Goode 2009 for a more comprehensive review).

Similar to ARP2/3, formin activities are controlled through their localization to specific sites of actin assembly. For example, fission yeast localize three distinct formins to control contractile ring assembly (Cdc12p; Yonetani et al. 2008), orientation and growth of cytoplasmic cables that direct protein traffic and polarized growth (For3p; Martin et al. 2007), and formation and extension of mating structures (Fus1p; Paterson et al. 2008). Examples more directly relevant to morphogenesis include the recruitment of formins to the leading edge of motile cells (Brandt et al. 2007, Gupton et al. 2007) or to sites of cadherin-based adhesion (Homem & Peifer 2008, Kobielak et al. 2004), where they promote filament assembly and maturation of adherens junctions (discussed below).

As for ARP2/3, the combination of synergistic activation and local recruitment yields tight spatial and temporal control over filament nucleation. Cross-linkers such as fascin, α-actinin, or filamin can then organize filaments into higher-order structures (e.g., orthogonal networks, parallel versus antiparallel bundles) that are specialized for different functions (Figure 2c,d). By localizing and/or activating formins at specific sites, cells can both orient contractile forces and define the anchor points against which those forces pull. Such a mechanism is thought in fission yeast to drive actomyosin-dependent contractile ring assembly from dozens of distributed formin-containing nodes (Wu et al. 2006). Anchoring barbed ends at nascent focal adhesions or cadherin spot junctions could provide analogous mechanisms to capture and recruit myosin to antiparallel filaments and thus to drive self-assembly of stress fibers (Hotulainen & Lappalainen 2006) or circumapical filament bundles (Homem & Peifer 2008).

Branched and unbranched modes of filament assembly can also synergize in different ways. For example, formin-mediated filament assembly may help to supply raw material for ARP2/3-dependent branching and lamellipod extension (Yang et al. 2007), whereas branched actin produced by ARP2/3 may supply the starting material for filopodial extension (Lee et al. 2010). Similarly, contractile filament bundles can form through myosin II–dependent realignment and/or merger of preformed elements at the leading edge (Anderson et al. 2008b, Hotulainen & Lappalainen 2006, Nemethova et al. 2008). Although these types of transitions have been documented mainly in vitro or at the leading edge of motile cells where filament architecture and dynamics are easily observed, similar synergies likely contribute to the formation and dynamic organization of actin arrays in other contexts, e.g., in epithelial cells during morphogenesis.
Mechanics of Actin Networks

The dynamics of cell- and tissue-scale movement and shape change depend critically on how actin networks resist or deform in response to the forces they produce or experience. To push, actin networks must be stiff enough to resist compression. To contract, actin networks require a minimal degree of network connectivity (i.e., cross-linking), but too much connectivity makes a network too stiff to deform or reorganize (Bendix et al. 2008).

As semiflexible polymers, actin filaments exhibit both entropic (owing to changes in degrees of freedom as filaments deform) and enthalpic (owing to resistance to structural deformation) elasticity (MacKintosh et al. 1995). Enthalpic elasticity dominates when filaments are short. Entropic elasticity dominates when they are longer. Networks of unbranched filaments are too soft to explain cellular stiffness, even at high densities, and thus much attention has focused on how branching or cross-linking shapes the mechanical properties of these networks.

Branched actin networks assembled by ARP2/3 are stiff enough to sustain forces produced during leading edge extension of motile cells or actin-based propulsion of beads in vitro (Marcy et al. 2004, Chaudhuri et al. 2007). Interestingly, these networks can rupture or tear when stretched beyond a certain limit, a property that some cells (and intracellular pathogens) may exploit to break symmetry (Paluch et al. 2006). The same networks exhibit reversible softening in response to compressive forces, probably owing to buckling of individual filaments (Chaudhuri et al. 2007), which may allow cells to avoid rupture when they encounter large loads. Cross-linking unbranched filaments into rigid bundles may allow unbranched filament elongation to drive extension of filopodia (Vignjevic et al. 2006; Figure 2b) or to propel intracellular bacteria (Briecher et al. 2004).

The viscoelasticity of rigidly and irreversibly cross-linked networks can be explained in terms of filament and cross-link density and the elastic responses of single semiflexible filaments (Gardel et al. 2004, MacKintosh et al. 1995), for physiological cross-linkers, however, in vitro studies reveal a more complex dependence of network viscoelasticity on the elasticity of both filaments and cross-linkers, on their relative densities, and on their turnover kinetics. A key property of cross-linked actin networks that has emerged from these studies is their ability to stiffen over orders of magnitude when strained by external or internal (e.g., protrusive and contractile) forces (Gardel et al. 2006, Koenderink et al. 2009, Mizuno et al. 2007). Interestingly, myosin II activity can stiffen networks under some conditions, and it can soften or fluidize networks under others by promoting filament sliding and rearrangement (Humphrey et al. 2002, Le Goff et al. 2002) or filament disassembly (Haviv et al. 2008). Myosin II can also exhibit load-dependent kinetics, which causes it to behave more like a cross-linker under reverse tension (against its direction of motion) and accelerates cycling under forward tension (Kovacs et al. 2007). This ability to tune compliance on demand by stiffening or softening in response to different levels of tensile or compressive force provides cells with intrinsic mechanisms to balance loads across heterogeneous structures or to maintain global shape in the face of fluctuating forces.

Another key feature of actin network mechanics that is particularly relevant to morphogenesis is the time dependency of viscoelasticity. On timescales that are fast relative to typical turnover times for cross-links (a few seconds; Wachsstock et al. 1994) or filaments (a few tens of seconds; Theriot & Mitchison 1991, Watanabe & Mitchison 2002), actin networks behave more like viscoelastic solids that deform reversibly in response to an applied force, returning to their original shape after the force is released. On longer timescales (seconds to tens of minutes), however, the same networks behave more like fluids that creep or flow in response to applied force with an effective viscosity whose magnitude depends on the stiffness of individual elements as well as their density, connectivity, and lifetimes.

A striking consequence of this property is that gradients of contractile tension will

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cause actin networks to flow on these longer timescales. Bray & White (1988) proposed this idea more than 20 years ago, but modern imaging studies have now begun to reveal the importance of actomyosin-based flows in organizing morphogenetic dynamics in a variety of contexts, for example, during symmetry breaking in *Caenorhabditis elegans* zygotes (Hird & White 1993, Mayer et al. 2010, Munro et al. 2004) and motile cells (Yam et al. 2007), during wound healing in *Xenopus* oocytes (Mandato & Bement 2003), and during epithelial tissue remodeling (Rauzi et al. 2010). We discuss cortical flows and related phenomena (pulses) below.

**ADHESION STRUCTURES AS FORCE TRANSMISSION ELEMENTS**

Contractile forces must be anchored at the cell cortex to produce cell shape changes. In epithelial cells, adhesion structures are dynamic modules that transmit the cell-generated forces through the plasma membrane to other cells or to the extracellular matrix (ECM). The major transmembrane proteins mediating cell-cell and cell-ECM adhesions are cadherins and integrins, respectively. Despite their structural and functional differences, cadherin- and integrin-based adhesion structures share striking similarities. Both are capable of binding to actin through adaptor proteins and signaling cytoskeleton remodeling. Both form clusters whose properties are most often dependent on actomyosin contractility. To date, cell-ECM adhesion is much better characterized than cell-cell adhesion, mostly because cell-ECM adhesions are amenable to more direct imaging and experimental tools such as biochemically patterned substrates and mechanical probes. We believe that lessons gained from the former can be instrumental in understanding the latter.

**Clusters Are the Elementary Units of Adhesion Sites**

First identified in electron micrographs of migrating fibroblasts in culture, cell-ECM adhesions were initially described as dense plaques anchoring bundles of actin filaments (stress fibers) (Abercrombie et al. 1971). A few years later, adhesion plaques were shown to be the regions where the plasma membrane comes closest to the ECM, with a separation distance of 10 to 30 nm (Abercrombie & Dunn 1975, Lazard & Lochner 1976). Meanwhile, immunofluorescence microscopy was used to image cytoskeletal components such as α-actinin and vinculin, which appeared to form patches localized along or at the ends of stress fibers (Geiger 1979, Lazarides & Burridge 1975), suggesting that they could be involved in the assembly of actin filaments at cell-ECM contact. These pioneering observations paved the way for many studies, which have now identified many components of cell-ECM adhesions.

Today, cell-ECM adhesions are classified by composition and organization into several types: focal complexes, focal adhesions, fibrillar adhesions, podosomes, and invadopodia. We discuss some aspects of force transmission by focal complexes and focal adhesions but refer to several excellent recent reviews for details of cell-ECM adhesions (Albiges-Rizo et al. 2009, Dubash et al. 2009, Geiger et al. 2009, Puklin-Faucher & Sheetz 2009). Focal complexes are nascent adhesive foci that localize along the lamellipodia of migrating cells soon after cell-ECM contact. These circular domains, 0.5–1 μm in diameter, contain transmembrane proteins of the integrin family and a few cytoplasmic partners such as talin (Zaidel-Bar et al. 2003; Figure 3). These structures are not connected to stress fibers but are linked to the actin network. Focal complexes mature into focal adhesions, which are elongated structures 3 to 10 μm long associated with stress fibers.

The molecular complexity of focal adhesions is daunting, with up to 90 resident components and 66 temporary players that interact with the resident molecules and regulate their activity (Zaidel-Bar et al. 2007). Despite this complexity, a small set of components capture the fundamental principles of focal adhesion assembly and dynamics (Geiger et al. 2009). α-Integrin/β-integrin dimers constitute...
the core of the receptor module, which binds ECM components. The cytoplasmic domain of the β-integrin subunit binds to talin, which connects integrin with actin. Talin binding can trigger conformational changes of the α-integrin/β-integrin dimers to an active state with strongly increased affinity to ECM ligands (Tadokoro et al. 2003, Wegener et al. 2007; Figure 3b, d, f).

Maturation of focal complexes into focal adhesions relies on the binding of other cytoplasmic partners such as vinculin (Chen et al. 2005, Humphries et al. 2007, Izard et al. 2004), which promote clustering of nascent complexes and reinforce the integrin links to actin (Figure 3b, d, f). The signaling module of cell-ECM adhesions consists of several components including kinases and phosphatases that are able to initiate a cascade of events. This results in local changes in cytoskeleton dynamics and the generation of mechanical force, which in turn modify adhesion (see below).

The organization of adhesion molecules in finitely sized clusters is also a striking feature of cell-cell adhesion. Similar to integrins, cadherins form dense protein clusters connected to the actin network. E-cadherin clusters have been observed in cultured epithelial mammalian cells and in early epithelia of nonvertebrates. In migrating cells undergoing mesenchymal to epithelial transitions, nascent adhesions of E-cadherin organize in puncta (Angres et al. 1996, Kametani & Takeichi 2007). These puncta are thought to represent

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**Figure 3**
Cell-cell and cell-extracellular matrix (ECM) adhesion clusters and their interaction with actomyosin networks. Cell-cell and cell-ECM adhesion initiate by homophilic binding of E-cadherin (a) and by binding of integrin to ECM (b), respectively. Actin-dependent protrusions are important in these processes. Next, E-cadherin binds actin filaments through adaptor proteins such as β-catenin, α-catenin, and vinculin (c). Similarly, integrin binds the actin cytoskeleton through proteins such as talin and vinculin (d). Actomyosin contractility produces pulling forces on adhesion complexes. These forces can induce conformational changes in α-catenin and talin, thereby exposing buried vinculin-binding sites. In turn, vinculin binding promotes further binding of actin filaments to adhesion clusters (e, f). This feedback mechanism enhances the mechanical coupling between actomyosin networks and adhesion clusters.
clusters of homophilic dimers in trans association (Kametani & Takeichi 2007). In Drosophila embryos, a similar organization has been reported (Harris & Peifer 2004, Müller & Wieschaus 1996). E-cadherin has slower dynamics in these clusters than outside (Cavey et al. 2008), and these structures likely are equivalent to spot adherens junction structures observed by electron microscopy (Tepass & Hartenstein 1994).

The mechanisms of cadherin clustering remain debated, and structural, biochemical, and in vivo studies are essential to illuminate this question. Cadherins are Ca$^{2+}$-dependent transmembrane proteins (Nose et al. 1990) whose extracellular domains are composed of multiple repeat domains. The outermost extracellular repeat is thought to mediate trans-cadherin binding (Chen et al. 2005), but force measurements between tethered extracellular domains suggest that additional extracellular domains may be involved in trans binding (Zhu et al. 2003). Increased extracellular domain density enhances trans-binding affinity, an observation suggestive of collective effects, which could strengthen clustering (Zhang et al. 2009). Selectivity of binding between different cadherin subtypes is thought to operate by trans homophilic binding of extracellular domains, as suggested by in vitro aggregation experiments on cells expressing different types of cadherins (Nose et al. 1990) and in vivo observations (Patel et al. 2006). However, this does not correlate with the significant differences in homotypic and heterotypic trans interactions between extracellular domains (Prakasam et al. 2006), which suggests that other factors such as kinetic effects or cis interactions may be involved.

At cell junctions, E-cadherin interacts with adaptor proteins, which can modulate cluster formation (Figure 3a,c,e). E-cadherin binds constitutively the cytoplasmic protein β-catenin, which is required for its export from the endoplasmic reticulum to the plasma membrane. α-Catenin, which is essential for epithelial integrity (Cavey et al. 2008, Kofron et al. 1997, Torres et al. 1997, Vasioukhin et al. 2000), binds β-catenin and mediates interaction with the actin cytoskeleton (Abe & Takeichi 2008, Cavey et al. 2008), but it does not do so directly, contrary to previous expectations. Biochemical and protein dynamics analyses indicate that the link between E-cadherin/β-catenin and actin through α-catenin is more dynamic than anticipated (Drees et al. 2005, Yamada et al. 2005). However, a recent study showed that actomyosin tension at adherens junctions unmasks a vinculin binding site in α-catenin (e.g., by protein unfolding as for talin) and requires direct actin association with α-catenin at the cell surface during epithelial wound healing (Yonemura et al. 2010, Figure 3a,c,e). Regardless of the nature of the link, several studies demonstrate the mechanical coupling between actin and E-cadherin clusters as well as the essential role of α-catenin in this association (Cavey et al. 2008, Yonemura et al. 2010). The intrinsic dynamics of interactions between E-cadherin/β-catenin complexes and F-actin is important, as it is expected to reflect frictions between tensile networks in epithelial cells and the cortex as well as to affect the rate of tension transmission at cell junctions. Cell-ECM adhesion involves a large repertoire of direct and indirect interactions that change dynamically the links between actin and integrin. This is likely also true for links between actin and cadherins, and further studies will unravel the intermediate missing links, such as EPLIN (Abe & Takeichi 2008) and vinculin (Yonemura et al. 2010). Biochemical studies cannot rule out that α-catenin may bind F-actin through E-cadherin/β-catenin directly but with very low affinity. According to this alternative, individual E-cadherin molecules would diffuse without an apparent link to actin, whereas clusters (comprising a few tens to thousands of molecules) would be tethered by actin, as the number of E-cadherin molecules in the clusters would increase the resultant affinity (Cavey et al. 2008).

**Adhesion Structures Under Forces**

Under forces, adhesion structures reorganize spatially as well as in size and shape. In cell
cultures, cadherin clusters are found either at the tips of protrusions, connected to an actin ring via radially oriented actin bundles (reviewed in Vasioukhin & Fuchs 2001), or tangentially organized along contacting cell edges and in association with circumferential actin bundles. Maintenance of the spatial organization of cadherin clusters depends critically on membrane tension (Ayari et al. 2004). For instance, inhibition of myosin II is sufficient to trigger transition from one type of organization to another (Krendel et al. 1999). In the Drosophila embryo, E-cadherin clusters are immobilized along a cortical actomyosin belt. Changes in myosin II contractility modulate the lateral mobility of cadherin clusters, an observation that is suggestive of an actively controlled tethering mechanism (Cavey et al. 2008).

Efficient transmission of forces requires strong links between actomyosin and adhesions. This property critically depends on the composition and size of the adhesions: a single fibronectin trimer-cytoskeleton link can resist a 1–2 pN force (talin dependent) (Jiang et al. 2003), whereas focal adhesions few micrometers long can bear forces up to 100 nN.

Remarkably, adhesion adapts to mechanical stress by modifying the size and shape of individual adhesion clusters. Protrusive forces provided by ARP2/3-induced actin polymerization are important for the maturation of cadherin clusters in elongated structures parallel to cell-cell interfaces (Verma et al. 2004). Changes in membrane tension were also shown to promote cadherin clustering in in vitro assays (Delanoe-Ayari et al. 2004). Membrane tension may favor cadherin trans binding and clustering by rectifying the fluctuations of apposed membranes.

The assembly of cell-ECM adhesions is also dependent on forces (Balaban et al. 2001, Riveline et al. 2001). Cell-ECM adhesions respond to forces by strengthening the connections between the liganded integrins and the force-generating cytoskeleton in a process called reinforcement (Choquet et al. 1997). Reinforcement of adhesion structures under force is puzzling, as increasing forces reduce the lifetime of single ligand-receptor bonds (Merkel et al. 1999). Which molecular mechanisms could underlie this behavior? First, forces can regulate the activity of integrins. Molecular dynamics simulations indicate that forces can provoke an allosteric change in integrins that results in increased binding strength with the ECM (catch bond). Second, talin, which provides the first mechanical resistance, recruits vinculin to initial adhesions, thereby increasing the mechanical stability of focal adhesions (Galbraith et al. 2002, Grashoff et al. 2010, Humphries et al. 2007, Figure 3b,d,f).

Force measurements have shown that stretching of single talin molecules induces unfolding that exposes cryptic binding sites for vinculin (del Río et al. 2009). Because talin contains multiple vinculin-binding sites, biochemical amplification could occur upon application of force. This may be a general mechanism for force transduction, and cell-cell adhesion structures likely also incorporate components that have the ability to unfold under forces, as recently proposed (Yonemura et al. 2010).

Importantly, the mechanical stress that an adhesion structure experiences depends not only on internally generated forces but also on the mechanical constrains of the surroundings, which can be other cells (possibly exerting forces) or ECM. Forces generated by the cytoskeleton on a focal adhesion give rise to an opposite balancing force in the ECM. Thus, the focal adhesion is under mechanical stress. When the ECM is not rigid, stresses do not develop properly at adhesions, and focal adhesions fail to mature (Galbraith et al. 2002). Strikingly, traction forces exerted on focal adhesions are directly proportional to the rigidity of the ECM (Saez et al. 2005). Conversely, forces applied externally through ECM components give rise to mechanical stress at focal adhesions if a resistive force builds up in the cytoskeleton (which can be passive or active) (Choquet et al. 1997). This strongly indicates that cell adhesions are adaptive coupling elements.

Therefore, changes in cell contractility are able to trigger, through adhesion structures, changes in the physical properties of the ECM or of other cells. Fibrillar adhesions are an...
illustration of this coupling: ECM components, such as fibronectins, can be reorganized through cell-ECM adhesions in parallel arrays in the direction of pulling stress fibers. This coupling is dynamic by nature, as the different components of adhesions are not covalently bound and are renewed with force-dependent rates (Hu et al. 2007).

Remarkably, in several epithelial cell lines, vinculin is also recruited at E-cadherin adhesion sites placed under tension by the actomyosin cytoskeleton (Yonemura et al. 2010; Figure 3a,c,e). Cell delamination triggers wound healing and formation of a junctional actomyosin ring whose contraction recruits vinculin at adherens junctions via direct association with α-catenin at the cell surface. Vinculin recruitment further enhances mechanical coupling by increasing stabilization of α-catenin at adhesion sites as well as actin recruitment. Such mechanical feedback suggests that intercellular adhesion sites also may be capable of tension-dependent organization. The role of vinculin in tension-dependent regulation of both integrin- and E-cadherin-based adhesion points to similar mechanical properties.

Adhesion Structures Regulate Actomyosin Assembly

Forces modify adhesion. Conversely, adhesion is able to change forces by actomyosin assembly. A growing body of evidence indicates that cell adhesions nucleate actin assembly and activate myosin II, thereby modifying the actomyosin organization and dynamics. During the formation of cell-cell junctions, actin bundles stabilize the cadherin clusters at the tips of filopodia in contact with cells (Vasioukhin et al. 2000). In turn, cadherin clusters control actin assembly by modulating the concentration of at least two types of actin nucleators: the ARP2/3 complex, which promotes assembly of branched actin (Helwani et al. 2004, Kovacs et al. 2002, Verma et al. 2004), and formins, which enhance the linear growth of F-actin. Formin-1 binds directly to α-catenin, whereas ARP2/3 binds β-catenin in competition with α-catenin (Drees et al. 2005). As adherens junctions mature, a local increase in α-catenin inhibits ARP2/3 (reviewed in Perez-Moreno & Fuchs 2006, Pokutta & Weis 2007) and promotes interaction with bundled actin via the formin Dia (Kobielak et al. 2004).

At cell-ECM adhesions, formins promote actin nucleation and growth (Butler et al. 2006) even though direct evidence for the specific association of formins to focal adhesions is still lacking. A mechanism for F-actin linear growth promotion by formins is consistent with the fact the stress fibers associated with focal adhesions grow by incorporation of new components at the focal adhesions (Hotulainen & Lappalainen 2006).

Adhesions also activate Rho GTPases that regulate cytoskeletal assembly by different mechanisms. Activation of RhoA at nascent cell-ECM adhesions leads to stress fibers and focal adhesions through a cascade of events. Activation of the Rho-kinase ROCK causes an increase in MRLC phosphorylation and thus myosin II activation (Figure 1). This results in an increase in contractility and actin bundling into stress fibers (Pellegrin & Mellor 2007).

Because integrin- and cadherin-mediated adhesions share common signaling components and both regulate actomyosin assembly, biochemical and mechanical cross talk exists between them (de Rooij et al. 2005, Genda et al. 2000, Martinez-Rico et al. 2010, Yano et al. 2004). Cross talk can either enhance or suppress adhesion depending on the environmental and cellular contexts. This has important implications in morphogenesis as well as in tumor invasion and metastasis. For example, integrins affect cadherin-mediated adhesion during convergent extension movements in Xenopus (Marsden & DeSimone 2003). In vitro, high traction forces exerted at cell-ECM adhesion sites can disrupt cell-cell adhesion, which suggests that actomyosin tension at cell-cell junctions is integrin dependent (de Rooij et al. 2005).

Our view of the adhesions at cell-cell and cell-ECM interfaces has moved from a static to a dynamic picture in which a small set
of modules connect cadherin and integrin clusters physically and biochemically to the actin network. In both cases, adhesion foci are force dependent but also signaling elements for actomyosin assembly. Despite the molecular complexity of the different modules, simple principles of organization emerge (Figure 3) that could account for the adaptive and differential transmission of the forces allowing cell movement, cell shape changes, and tissue morphogenesis as described below.

**Tissue-Level Integration of Tension and Adhesion**

Intercellular adhesion in epithelial cells underlies tissue cohesion and enables extensive remodeling (reviewed in Lecuit & Lenne 2007, Montell 2008). Recent studies have shown that generation of forces by contractile actomyosin networks and their transmission at the cell cortex are responsible for junction remodeling. We now review how tissue dynamics and cohesion emerge from the spatial regulation and integration of force generation (Figures 1 and 2) and force transmission (Figure 3) at the cell cortex.

**Spatial Regulation of Tension**

We review below three examples of morphogenetic processes illustrating how changes in tissue shape emerge from the polarized regulation of force generation and transmission at adhesive sites.

Cells often segregate into compartments, groups of cells that maintain a strict segregation at their boundary throughout tissue growth. Compartment boundaries are smooth because cells are not miscible. In *Drosophila* embryos each segment is composed of two compartments, anterior and posterior. In the zebrafish central nervous system, rhombomeres also form compartments (Mellitzer et al. 1999, Xu et al. 1999). The differential adhesion hypothesis proposed by Steinberg (1970) considers different groups of cells to behave as fluids (Steinberg & Poole 1982), which minimizes the contact surface depending on the relative strengths of the adhesion between cells of different groups. The adhesion molecules underlying differential adhesion at compartment boundaries have, however, remained elusive (Dahmann & Basler 2000, Milan et al. 2001). A different model suggests that increased interfacial tension at compartment boundaries controlled by the concentration of myosin II and F-actin maintains the compartment boundaries and prevents cell mixing in the *Drosophila* wing (Major & Irvine 2005, 2006). This model has been further validated in other instances using a combination of chromophore-assisted light inactivation of myosin II (Monier et al. 2010) and laser ablation to measure differences in cell bond tension (Landsberg et al. 2009). In the embryo, myosin II–based tension rectifies the boundaries that cell divisions distort. A simple upregulation of myosin II–dependent interfacial tension can thus explain the shape of tissue boundaries.

Experiments on cell sorting mechanisms in the gastrulating zebrafish embryo also showed that these cannot be explained by differential adhesive properties, but can by interfacial tension controlled by cortical myosin II contractility as measured with atomic force microscopy (Krieg et al. 2008, Schotz et al. 2008). The role of adhesive clusters in transmitting tension in compartment segregation remains unknown.

The anisotropic regulation of myosin II–dependent contractility also plays a key role in controlling convergence-extension movements via cell intercalation (Figure 4a). Intercalation in adhesive mesenchymal cells (reviewed in Keller 2006) relies on a striking polarization of cell motility. Polarized motility emerges from the interaction between protrusive activity (Jessen et al. 2002, Marlow et al. 2002, Shih & Keller 1992, Wallingford et al. 2000) and contraction powered by myosin IIB on a meshwork of dynamic actin foci (Rolo et al. 2009, Skoglund et al. 2008). The polarity of contractility is due to the polarized transmission of contractile forces at adhesive foci at protrusive ends. The interplay between contraction and protrusive activity is thought to drive cell sliding and intercalation. In epithelia,
Planar polarity: a characteristic of a cell that is oriented in the plane of the tissue of which it is a part. Cell intercalation is apparently quite different, but some parallels in the underlying mechanics can be drawn. For example, in the notochord of ascidian embryos, protrusive activity along the lateral surface was reported (Munro & Odell 2002). However, the most significant advance comes from studies in Drosophila that have emphasized the central role of anisotropic contractility at cell junctions. During elongation of the Drosophila embryonic epithelium known as the germ band, medial lateral cell intercalation produces elongation along the anteroposterior axis (Irvine & Wieschaus 1994). This is driven by the planar polarized remodeling of cell junctions, also called T1 processes, which comprises two steps (Bertet et al. 2004; Figure 4a). First, contacts oriented along the dorsal-ventral (or vertical) axis shrink, bringing four or more cells together. Subsequently, new junctions grow on a perpendicular, along the anteroposterior axis. The remodeling of cell junctions requires the specific enrichment of myosin II in vertical contacts (Bertet et al. 2004). Rosettes form when myosin II accumulation in vertical junctions spans several junctions in supracellular cables (Blankenship et al. 2006, Fernandez-Gonzalez et al. 2009, Rauzi et al. 2008). Laser ablation experiments that severed the actomyosin network at the cortex showed that cortical tension is anisotropic, reflecting differences in the local concentration of myosin II (Rauzi et al. 2008). Interestingly, E-cadherin complexes (Blankenship et al. 2006) and the polarity protein Par-3 (Zallen & Wieschaus 2004), which binds β-catenin (Wei et al. 2005), are present at lower levels in shrinking junctions. This could affect adhesion and/or the distribution of tension-anchoring points at cell junctions, as recently reported (Rauzi et al. 2010). Planar polarized remodeling of cell contacts has been suggested to control morphogenesis at early stages of cell invagination in the Drosophila tracheal placode (Nishimura et al. 2007). Elongation and bending of the vertebrate neuroepithelium, in particular in chicken embryos, is associated with the planar polarized enrichment of MRLC and of its activator ROCK at cell junctions (Nishimura & Takeichi 2008). Live imaging of cell remodeling is still lacking, so it is too early to establish further comparisons between vertebrate neural tube morphogenesis and germ band extension. In the chick embryonic epiblast, epithelial intercalation is important for formation of the primitive streak, but...
the mechanisms driving this process remain unknown (Voiculescu et al. 2007).

Tissue invagination is a widespread process that requires apical cell constriction (reviewed in Sawyer et al. 2009b; Figure 4b). As postulated from physical principles (Odell et al. 1981), this process depends on isotropic upregulation of contractility by myosin II. In vertebrates, such as in the mouse or Xenopus neural tube, bending of the neuroepithelium is associated with recruitment and activation of myosin II at apical cell junctions (Haigo et al. 2003, Hildebrand 2005, Hildebrand & Soriano 1999). This is induced by the protein Shroom, an actin-binding protein necessary and sufficient for the junctional recruitment of myosin II and apical constriction. A striking manifestation of myosin II–based junctional tension increase is the straightening of cell contacts by Shroom (Hildebrand 2005). The GTPase Rap1 but not Rho1 is required for this process (Haigo et al. 2003).

In other cases, apical constriction is associated with an enrichment of myosin II in the medial apical region of the cell rather than at cell junctions, for example in the invaginating C. elegans (Lee & Goldstein 2003, Lee et al. 2006) and ascidian endoderm (Sherrard et al. 2010; Figure 4b) and the Drosophila mesoderm (Barrett et al. 1997, Dawes-Hoang et al. 2005, Kolsch et al. 2007, Martin et al. 2009). In Drosophila, this medial mesh is anchored at cell junctions by E-cadherin/β-catennin/α-catennin complexes (Dawes-Hoang et al. 2005, Martin et al. 2009) but also via the actin-binding protein Canoe/afadin at vertices (Sawyer et al. 2009a). It is still unclear what localizes Canoe/afadin because absence of E-cadherin and/or of the Ig domain adhesion protein Echinoid does not affect Canoe/afadin localization (Sawyer et al. 2009a). Activation of myosin II in the medial region requires activation of the Rho1GTPase by RhoGEF2 (Barrett et al. 1997) and the combined activity of the ligand Fog (Costa et al. 1994, Morize et al. 1998), the small Gx protein Concertina (Gαq12q13) (Parks & Wieschaus 1991), a still unknown G protein–coupled receptor, and the transmembrane protein T48, which binds the PDZ-binding domain of RhoGEF2 (Kolsch et al. 2007). Finite element modeling of mesoderm invagination delineates the key contribution of increased apical cell tension (Munoz et al. 2006). Interestingly, this suggests that apicobasal shortening is also required, which was recently shown in ascidian endoderm invagination through the differential regulation of myosin II phosphorylation at the apical and basal lateral surfaces (Sherrard et al. 2010; Figure 4b). In the C. elegans endoderm, apical recruitment of myosin II depends on a Wnt-dependent signal (Lee et al. 2006), apical activation of Cdc42, and recruitment of the polarity proteins Par3 and Par6 (Nance et al. 2003). Polarized apical activation of Cdc42 is due to the recruitment of the RhoGAP PAC-1 at cell contacts (Anderson et al. 2008a). As discussed earlier, Cdc42 and MRCK play important roles in the activation of the MRLC.

**Actomyosin Contractile Dynamics: Pulses and Flows**

A simple framework thus emerges in which subcellular regulation of myosin II distribution dictates local subcellular forces and cell shape changes. A simple physical interpretation assumes that (a) actomyosin contractility and adhesion define the energy of the cell and (b) tissue dynamics follows a pathway of local energy minimization (Farhadifar et al. 2007, Hilgenfeldt et al. 2008, Landsberg et al. 2009, Rauzi et al. 2008). However, thermodynamic approximation is inadequate if one seeks to understand how tissue-level dynamics emerges from subcellular mechanics, and to answer the following questions: How do forces emerge from the interaction among many actin filaments and myosin motors? How are these forces coupled to adhesive complexes to produce cell shape changes? How do the inherent viscosity within actomyosin networks and frictions with adhesion complexes and the membrane resist force-dependent cell deformations? Answering these key questions is essential if one wants to understand the speed...
at which cell shape changes occur as well as to reveal the existence of possible rate-limiting steps, cross talk, and feedback among the force generation and force transmission systems within and between cells as they attempt to coordinate or isolate their individual mechanics. Subcellular mechanics must be described as resulting from the local biochemistry/exchange kinetics between actin filaments, myosin motors, and adhesion complexes.

Several recent observations concerning the contractile dynamics in epithelial cells emphasize the need for such models, in particular pulsed contractility and actomyosin flow patterns (Figure 5).

Actomyosin contractility is sometimes stationary, in that intensity levels of myosin II are kept relatively constant, as occurs during cytokinesis and related processes such as Drosophila cellularization (Lecuit et al. 2002). In other situations, however, actomyosin networks are pulsatile: Intensity levels of myosin II show strong fluctuations over a few tens of seconds. This was first described in the C. elegans zygote, where pulsatile contractions accompany cortical flow during polarization (Munro et al. 2004; Figure 5g,b). Subsequent work identified similar dynamics during apical constriction in the Drosophila mesoderm (Figure 5a–c). A rapid increase in myosin II intensity in the medial apical region correlated with constriction phases of the apical cell surface followed by phases of stabilization (Martin et al. 2009). A similar behavior occurs during Drosophila cell intercalation when medial myosin II contraction contributes small steps of junction shrinkage, whereas junctional myosin II stabilizes cell shape (Rauzi et al. 2010; Figure 5d–f). Apical constriction in the Drosophila amnioserosa cells is also pulsatile (Solon et al. 2009, Blanchard et al., David et al. 2010). The resulting amnioserosa constriction drives dorsal closure of the abutting lateral ectoderm. The realization that pulses of myosin II contraction are widespread raises important questions. What properties of the actin network and of myosin II activation are required for the formation of pulses? Whether pulsed actomyosin contractility per se serves an essential function during morphogenesis is unclear. The amplitude of pulses may define the range of cell deformations, and their frequency could determine the speed of the process. Pulsed contractility may allow separate regulation of amplitude and speed during morphogenesis. However, pulsed contractility may be a by-product of some other property of actomyosin networks. Another important question is: What underlies deforming or stabilizing properties of actomyosin networks?

Actomyosin networks display other striking dynamics, namely the tendency to flow (Figure 5d–b). This was first described in the C. elegans cortex (Hird & White 1993, Mayer et al. 2010, Munro et al. 2004; Figure 5g,b) and at the leading edge of migrating cells,
but has been reported recently in the medial region of *Drosophila* intercalating cells, which suggests striking parallels between different morphogenetic processes (Rauzi et al. 2010). In the latter case, medial pulses do not accumulate in the center (as is the case in apical constriction) but instead flow toward vertical junctions and shrink them. The directionality of the flow determines the spatial orientation of cell deformation and is dependent on the anisotropic distribution of E-cadherin/β-catenin/α-catenin anchoring points at cell junctions (Levayer et al. 2011). In the *C. elegans* cortex, where the physical properties could be probed in detail, long-range flow may stem from the dominance of the actin network intrinsic viscosity over actomyosin network friction with the plasma membrane (Mayer et al. 2010).

A new framework for tissue morphogenesis thus emerges centered on two fundamental features of actomyosin networks, namely pulsation, which sets the speed of deformation, and flow, which determines spatial orientation. Centripetal flow drives apical constriction and anisotropic flow drives cell intercalation.

### Coordinating Contractility Between Cells and Tissues

Tissue morphogenesis often requires the coordination of multiple cell or tissue behaviors. Several recent reports suggest the possibility that intercellular coordination involves mechanical coupling of cell contractility between adjacent cells. Cell intercalation in the *Drosophila* germ band involves formation of multicellular rosettes in addition to simple T1 events. Rosettes arise when myosin II enrichment in vertical junctions spans multiple junctions in so-called myosin II cables (Blankenship et al. 2006, Fernandez-Gonzalez et al. 2009, Rauzi et al. 2008). Cable formation could result from the feedback amplification of cortical tension by isolated T1 processes (Fernandez-Gonzalez et al. 2009). Consistent with this, tension in cables is greater than that in isolated T1 events. Moreover, application of local stress (e.g., by membrane aspiration) increases myosin II cortical recruitment. Although artificial, this situation suggests a plausible scenario for the formation of cables.

During mesoderm invagination, a phase of randomly distributed apical constriction is followed by another phase in which constriction is more concerted within the presumptive mesoderm (Leptin & Grunewald 1990, Sweeton et al. 1991). Recent data suggest that this too could result from mechanical coordination between adjacent cells, as local contractility amplifies the contractility of neighboring cells (Martin et al. 2010).

It will be important to test this hypothesis further to see the extent to which and how local contractility affects the mechanics of neighboring cells. Mechanotransduction is expected to require the tension transmission machineries described above. In addition, it should require specific properties of actomyosin networks, in particular a high viscosity (i.e., internal friction among actin filaments and high coupling with the tension transmission apparatus) to produce mechanical coordination between neighboring cells.

Mechanical coordination also exists between different tissues, such as during *Drosophila* dorsal closure, when the lateral ectoderm migrates toward the dorsal midline. Leading edge cells accumulate high levels of actin and myosin II at their fronts to form a supracellular actomyosin cable. Laser ablation experiments (Franke et al. 2005, Hutson et al. 2003) identified four sets of force-generating processes required for closure: (a) amnioserosa contraction, (b) leading edge contraction by the actin cable to produce purse string tension, (c) zipping forces from the edges of the closing ectoderm, and (d) protrusive activity for completion of closure. Myosin II concentration at the leading edge of the lateral ectoderm and in amnioserosa cells is essential for closure (Franke et al. 2005).

Recent studies indicated that contraction of amnioserosa cells is coordinated with contraction of the supracellular actin cable (Solon et al. 2009). Amnioserosa cells display pulses of contraction responsible for transient dorsal
displacements of the ectodermal sheet. Solon et al. (2009) have proposed that actomyosin contraction in the supracellular cables stabilizes the ectoderm, although this view was later challenged (Gorfinkiel et al. 2009).

The mechanisms of this coordination are unclear but involve E-cadherin (Gorfinkiel & Arias 2007) and integrins (Gorfinkiel et al. 2009, Hutson et al. 2003), most likely because these ensure tension transmission within and between the lateral ectoderm and the amnioserosa. Detailed kinematic analysis of dorsal closure indicates that adhesion to the ectoderm may resist the contraction of the amnioserosa itself (Gorfinkiel et al. 2009). In integrin mutants, amnioserosa contraction is enhanced, which suggests a high degree of coupling and coordination between different tissues during dorsal closure (Gorfinkiel et al. 2009).

The existence of such feedback between elements of contractile systems and transmission at the cortex makes it hard to predict their collective dynamic behavior in single cells and even more between cells if mechanical coupling is important. In this context, computational modeling that describes local molecular mechanics (e.g., viscosity, elasticity, and active stress imposed by motor-driven contractility) and local biochemistry along with mutual interactions should open new windows for the study of morphogenesis, especially to connect scales of organization from molecules to tissues.

**SUMMARY POINTS**

1. Force production in embryonic cells involves locally tuned, self-organized protrusive and contractile machines that evolve through the continuous interplay between biochemical regulation and cytomechanics.

2. Cells regulate force production in part through local control over actin filament assembly, cross-linking, and disassembly, and in part by regulating myosin II motor activity and assembly state.

3. The dynamics of actin-based movement and shape change depends critically on the viscoelastic properties of actin networks, i.e., how they resist or deform in response to the forces they produce or experience. On short timescales actin networks behave like viscoelastic solids that deform reversibly in response to an applied force. However, on the longer timescales that characterize cell shape change during morphogenesis, the same networks behave more like fluids that creep or flow.

4. Contractile forces must be anchored at the cell cortex via cadherins or integrins to produce cell shape changes. Both integrins and cadherins are organized in supramolecular clusters that comprise adhesion surface molecules and adaptors that mediate coupling to actomyosin networks.

5. Under tension, integrin and cadherin adhesion clusters can be reinforced and strengthen mechanical coupling to actomyosin networks. These features could account for the adaptive and differential transmission of forces that allows cell movement, cell shape changes, and tissue morphogenesis.

6. Contacting epithelial cells in a tissue are tightly adherent but also extensively remodeled during morphogenesis. Regulation of force generation by contractile actomyosin networks and their transmission at the cell cortex by adhesion molecules controls cell and tissue shape changes.
7. Actomyosin pulsed contractility and flow patterns determine the speed and spatial orientation of cell deformations in a tissue.

8. Complex tissue morphogenetic processes require coordination and/or integration of cortical tension among cells or tissues.

**DISCLOSURE STATEMENT**

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**LITERATURE CITED**


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