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On the Mechanism of Cleavage Furrow Ingression in *Dictyostelium*

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ABSTRACT. **The ability of *Dictyostelium* cells to divide without myosin II in a cell cycle-coupled manner has opened two questions about the mechanism of cleavage furrow ingression. First, are there other possible functions for myosin II in this process except for generating contraction of the furrow by a sliding filament mechanism? Second, what could be an alternative mechanical basis for the furrowing? Using aberrant changes of the cell shape and anomalous localization of the actin-binding protein cortexillin I during asymmetric cytokinesis in myosin II-deficient cells as clues, it is proposed that myosin II filaments act as a mechanical lens in cytokinesis. The mechanical lens serves to focus the forces that induce the furrowing to the center of the midzone, a cortical region where cortexillins are enriched in dividing cells. Additionally, continual disassembly of a filamentous actin meshwork at the midzone is a prerequisite for normal ingression of the cleavage furrow and a successful cytokinesis. If this process is interrupted, as it occurs in cells that lack cortexillins, an overassembly of filamentous actin at the midzone obstructs the normal cleavage. Disassembly of the crosslinked actin network can generate entropic contractile forces in the cortex, and may be considered as an alternative mechanism for driving ingression of the cleavage furrow. Instead of invoking different types of cytokinesis that operate under attached and unattached conditions in *Dictyostelium*, it is anticipated that these cells use a universal multifaceted mechanism to divide, which is only moderately sensitive to elimination of its constituent mechanical processes.**

Key words: cytokinesis/cell division/mechanical lens/actin disassembly/bending stiffness/cortexillin

Since the discovery of myosin II-independent mitotic cell division in *Dictyostelium discoideum* (Neujahr *et al*., 1997a), several attempts have been undertaken to clarify its mechanism and relate it to the classical model of cytokinesis, which is based on the concept of contractile ring (Zang *et al*., 1997; Uyeda *et al*., 2000). Basic assumption in these propositions is that cytokinesis in wild-type *Dictyostelium* cells is driven by contraction of a contractile ring, an annular structure that assembles at the central zone of a dividing cell and consists of actin and myosin II filaments. Thus, cells that express myosin II are assumed to divide primarily by the standard sliding-filament mechanism, and this mode of cell division was termed cytokinesis A. It was argued that myosin II-null cells use a different mechanism to divide by a process termed cytokinesis B, apparently not used by wild-type cells. In this view, cytokinesis B depends on expansion of the polar regions of a dividing cell, which induces a passive ingression at the cleavage furrow by drawing the cytoplasmic material out of the midzone (Uyeda *et al*., 2000). Traction forces between the ventral cell surface and a substratum are viewed as necessary in order to support the polar expansion. A sufficient magnitude of the traction forces and their adequate orientation are dependent on a sufficiently strong attachment of a dividing cell to a substratum, which is known to be an essential condition for myosin II-independent cytokinesis.

In the present article, an alternative view at the mechanism of cytokinesis in *Dictyostelium* will be presented. It will be proposed that these cells, instead of activating distinct mechanisms to divide under different conditions, use a universal multifaceted mechanism of cytokinesis. When one of the components of this compound mechanism is eliminated by means of genetic, pharmacological or mechanical manipulation, the remaining components may or may not suffice for a successful cell division. In the first part of the article, major molecular constituents present in the cortical zone

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Abbreviations: CMC, cortical (cortexillin-containing) midzonal complex; MSP, major sperm protein; FRAP, fluorescence recovery after photobleaching; GFP, green fluorescent protein; PIP_2, phosphatidylinositol (4,5)-bisphosphate.

where the cleavage furrow ingression is initiated will be described. Next, clues from experimental observation of cytokinesis in mutant cells and from pharmacological and mechanical manipulations will be used to identify hypothetical processes that contribute to the furrowing, and to tentatively assign roles of individual molecules in these processes. Finally, an integrative model of compound cytokinesis in *Dictyostelium* will be proposed, and its relevance for cell division in general will be discussed.

Major players in the center: myosin II and a cortical midzonal complex

In dividing *Dictyostelium* cells a relatively broad zone is formed in the central cortical region at anaphase, which occupies about a third of the full cell length. This region is enriched in myosin II and a heterodimer formed by actin-binding proteins cortexillins I and II. The term midzone will be used to designate this central zone, and regions on both sides of the midzone will be called polar regions. Polar regions are enriched in filamentous actin, but myosin II and cortexillins are largely absent. This arrangement of cortical zones in dividing *Dictyostelium* cells is different from the situation in, for example, yeast cells and echinoderm oocytes, where the so-called contractile ring enriched in F-actin and myosin II occupies a rather narrow band at the cleavage site.

It is well documented that myosin II accumulates in the central cortical zone of dividing *Dictyostelium* cells, and that this localization is dependent on a domain in the coiled-coil tail of the myosin heavy chain, which is also necessary for formation of myosin II filaments (Shu *et al*., 1999; Nock *et al*., 2000). Although myosin II filaments accumulate at the location of the cleavage furrow formation, there is no evidence that their activity directly constricts the furrow by moving antiparallel actin filaments against each other, as it occurs in animal muscle cells. Myosin II filaments could also be acting solely to link actin filaments into bundles and more complex networks, or to provide isometric tension to the cortex. Furthermore, independence of the cortical myosin II localization from its actin-binding activity suggests that myosin II directly interacts with a factor bound to the cell plasma membrane, suggesting its possible role in coupling between the cortical actin filaments and the membrane.

Cortexillins are actin-bundling proteins that organize actin filaments preferentially into anti-parallel bundles and associate them into three-dimensional networks (Faix *et al*., 1996). Cortexillins are enriched in the cortex of interphase cells and translocate to the equatorial region of dividing cells during anaphase (Weber *et al*., 1999a). Mutant cells lacking both cortexillin I and II are extremely flat and severely impaired in cytokinesis (Weber *et al*., 1999b). The N-terminal halves of each cortexillin subunit encompass a conserved actin-binding calponin-homology domain (Faix *et al*., 1996). A central coiled-coil domain is responsible for the assembly of the cortexillins into parallel dimers (Steinmetz *et al*., 1998). Following this rod domain is a C-terminal region that proved to be pivotal for the targeting and biological activity of the cortexillin I molecule. It harbors the strongest actin-bundling activity and a phosphatidylinositol (4,5)-bisphosphate (PIP_2) binding site (Stock *et al*., 1999). Studies with green fluorescent protein (GFP) fusions have shown that the C-terminal fragment of cortexillin I is sufficient for cortical localization, recruitment to the cleavage furrow and for the complete rescue of cytokinesis in cortexillin I-null mutants (Weber *et al*., 1999a).

Accumulation of the cortexillin heterodimer (in further text referred to as cortexillin) in the cortical midzone during cytokinesis is mediated by the IQGAP-related and Rac1-binding protein DGAP1, which specifically interacts with the C-terminal, actin-bundling domain of cortexillin I (Faix *et al*., 2001). Like cortexillin, DGAP1 is enriched in the cortex of interphase cells and translocates to the cleavage furrow during cytokinesis. The activated form of the small GTPase Rac1A recruits DGAP1 into a quaternary complex with cortexillin I and II, and formation of the complex is of key importance for normal cytokinesis in *Dictyostelium*. Since this complex contains cortexillin and assembles at the cortical midzone in cytokinesis, it will be designated as the cortical cortexillin-containing midzonal complex (CMC). In DGAP1-null mutants, a second IQGAP-related protein, GAPA, mediates CMC formation. The simultaneous elimination of DGAP1 and GAPA, however, prevents CMC formation and leads to a severe defect in cytokinesis, due to an inappropriate localization of cortexillin and inability of double-mutant cells to form a proper cleavage furrow (Faix *et al*., 2001).

Failure of cytokinesis in myosin II-deficient cells

Inability of myosin II-null cells to divide without support of a substratum is used by the proponents of the sliding-filament mechanism as the crucial argument for their scheme of two separate modes of cytokinesis. It should be recognized, however, that mitotic divisions of attached and unattached myosin II-null cells have a number of features in common (Fig. 1). Under both conditions, CMC assembles at the midzone of a dividing cell in anaphase. At approximately the same time point, the cell poles start to expand and display a ruffling activity, which is also typically seen at leading lamellipods of moving cells. Simultaneously, change of the shape that is typical for a dividing cell commences in which the cell contour flattens at the midzone and elongates slightly. After this point in cytokinesis of myosin II-null cells, an asymmetry at the midzone starts to develop when one side of the cylindrical midzone starts to constrict while the other side opens up (Fig. 1). This lateral asymmetry occurs in majority of attached and in all unattached myosin II-null cell. In attached cells, an asymmetric formation of the cleavage

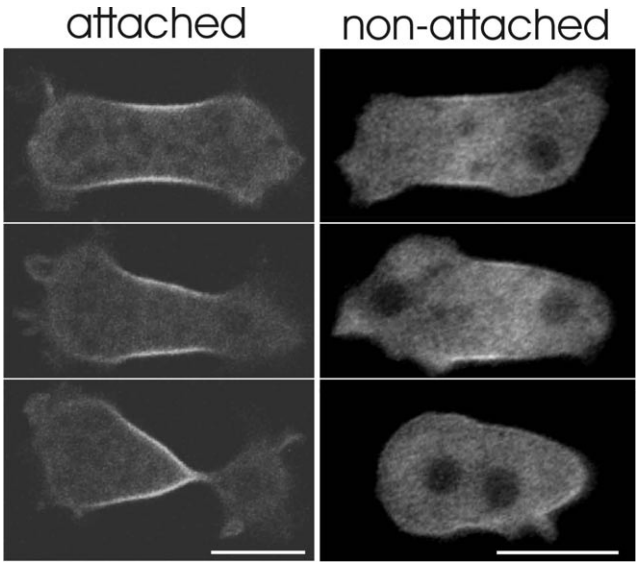

Fig. 1. Asymmetric cytokinesis in myosin II-deficient *Dictyostelium* cells. A cell attached to the glass substratum succeeded to divide, albeit in a strongly asymmetric fashion (left column). Cytokinesis of a cell cultivated in melted agar failed (right column). The cells express a fusion protein between GFP and cortexillin I, a marker of the midzone in mitotic *Dictyostelium* cells. Note that in the first two frames dividing cells have similar shapes under both attached and non-attached conditions. Quality of images in the right column is not optimal due to absorption and scattering of light in the agar. Relative accumulation of cortexillin I in the midzone should not be judged on the basis of these images (Weber *et al*., 2000). Bars, 10μm.

furrow regularly leads to an asymmetric cytokinesis, and in about 20% of these cases the cytokinesis fails (Weber *et al*., 2000). In the case of unattached cells, cytokinesis invariably fails.

Modes of failure of asymmetric cytokinesis in attached and unattached myosin II-null cells are basically equivalent. After an initial imbalance between the two edges of the midzone, the tapered end continues to narrow, whereas the widened end continues to open. The funnel-shaped CMC-containing region appears to move relative to the cell contour in the direction of its tapered end, and the endoplasmic material flows in the opposite direction. After the onset of this process in unattached cells, the CMC zone slides rapidly towards one cell pole and coalesces there into a continuous area shaped as a hollow cone. As one end of the dividing cell becomes covered by CMC, the protrusive activity at the leading edge of that part of the cell gradually terminates and the former polar region eventually retracts. In attached cells, this process is counteracted by an enduring motile activity at the shrinking pole. In this way, a cell is pinched in two unequally sized fragments at a border of the CMC-containing zone (Fig. 1).

It appears therefore that two autonomous activities, ingression at the midzone and expansion at the poles, govern the cell shape changes during cytokinesis. Bisection of the CMC zone and its constant position in the central region of a dividing cell depend on myosin II. Consequently, a stable relative position of the polar and the CMC-containing regions is not warranted in myosin II-null cells, in contrast to wild-type cells. In the absence of myosin II, one of the polar regions shrinks at the expense of the opposite polar region, but the width of the CMC zone remains constant. Most attached myosin II-null cells still can divide because the cell-substratum adhesion at the shrinking polar region seems to provide resistance to fast retraction of that pole, enables its prolonged motile activity, and thus makes completion of furrowing possible. In unattached cells, on the contrary, movement of the CMC zone towards the pole and the resulting suppression of the polar expansion occurs faster than the furrowing. It is likely that relative paces of the two processes, furrowing at an edge of the CMC zone and its sliding towards a polar region, which is counteracted by the polar expansion, determine whether a cytokinesis will be brought to a completion. In this scenario, the main function of myosin II is to prevent the CMC sliding and not to generate furrowing.

Two questions follow from this view on the furrowing process in *Dictyostelium*. First, how does myosin II act to prevent sliding of the CMC zone relative to the central region of a dividing cell? Second, which mechanism provides force for ingression of the cleavage furrow in myosin II-deficient cells?

Myosin II filaments as a mechanical lens

It is known that recruitment of myosin II into the central cortical layer of a dividing cell is independent of its binding to actin (Yumura and Uyeda, 1997; Zang and Spudich, 1998). Consequently, it can be postulated that a population of binding sites for myosin II exists in this region, probably anchored to the cell plasma membrane. Fluorescence recovery after photobleaching (FRAP) studies indicate that binding of myosin II to these sites is highly dynamic (Yumura, 2001). GFP-labeled myosin II is recruited to the cortical binding sites directly from a cytoplasmic pool and remains bound there for an average period of about seven seconds. Cortical recruitment of myosin II depends on its ability to form filaments, and its release depends on ability of these filaments to rapidly disassemble. These two processes are regulated by dephosphorylation and phosphorylation of myosin heavy chain tail domains in the molecule, respectively. It has been shown that mutants of myosin II in which three threonine subunits were replaced by aspartate residues cannot form filaments in the cell, and consequently do not show cortical localization (Sabry *et al*., 1997; Shu *et al*., 1999; Nock *et al*., 2000). On the other hand, replacement of threonines by alanines results in an overassembly of myosin II filaments in the cortex and inhibits their rapid reshuffling into a cytoplasmic pool (Yumura, 2001).

Taking these findings into account, it is plausible to pro-

pose that cortical myosin II mini-filaments act as linkages between actin filaments and the cell plasma membrane. Actin-binding activity of myosin II is known to be essential for its full functionality, since truncated molecules that lack the actin-binding activity cannot rescue the cytokinesis defect in myosin II-null cells (Yumura and Uyeda, 1997; Zang and Spudich, 1998). Also, dynamic shuffling of myosin II between cortical and cytoplasmic populations seems to be of crucial importance for its function, since mutant cells that express the triple-alanine construct have a strong cytokinesis defect (Egelhoff *et al*., 1993). Recent micromechanical experiments support the notion that myosin II activity plays an important role in dynamic couplings within the actin cortex and between the cortex and the plasma membrane. When *Dictyostelium* cells are locally subjected to suction pressures in the range between several tens to several thousands Pa from a micropipette, local aspiration into the pipette occurs at a certain threshold value (Merkel *et al*., 2000). It has been shown that, at the threshold point, cell plasma membrane locally detaches from the actin cortex. It has been inferred that this phenomenon is brought about by a local fracture of bonds between the cell envelope, which is aspirated into the pipette, and the cell body.

Interestingly, the threshold pressure is larger approximately by a factor of 5 for myosin II-deficient cells as compared to wild-type cells. This big difference indicates that myosin II plays an important role in coupling between the lipid bilayer-based plasma membrane and the actin-based cortical layer. In mutant cells, this role of myosin II is probably substituted by other coupling mechanisms, which form stronger and less dynamic cortex-membrane links. Consistent results have been obtained in an independent study where a microrheology technique has been applied to measure local viscoelastic properties of *Dictyostelium* cells (Feneberg *et al*., 2001). Again, local cytoplasmic domains of myosin II-null cells exhibited larger resistance to movement of colloidal beads than equivalent regions of wild-type cells, indicating that the former have an increased viscosity. Myosin II, therefore, appears to act as a dynamic linker, which contributes to softening of the cell envelope, defined as a composite stratified shell comprising the lipid/protein bilayer and the associated actin-based cortex.

Cell division involves a global mechanical deformation of the cell envelope. Deformation of the cell envelope depends, among other factors, on the coupling strength between the cortical cytoskeleton and the plasma membrane, and on the cortical bending elasticity. It can be inferred from the experimental findings described above, and the predominant mode of cytokinesis failure in mutant cells, that myosin II decisively influences these two parameters. First, myosin II stabilizes the CMC-containing zone at the center of a dividing cell, perhaps by linking it to a specific domain at the midzone of the plasma membrane. In the absence of myosin II, CMC usually slides relative to the central membrane region because their coupling is mediated by a less specific mechanism. Also, an elevated isometric tension in the midzone provided by the myosin motor activity may play a role in resisting the sliding. Second, myosin II apparently reduces the bending elasticity modulus of the cell envelope, and thus facilitates its bending deformation that takes place during ingression of the cleavage furrow. In its absence, the CMC-containing zone rarely undergoes a concave deformation, which indicates that it has become rigidified. Since these two activities amount to focusing the cleavage furrow to the center of a dividing cell, it is proposed that in cytokinesis myosin II plays a role of a mechanical lens.

A continuous actin turnover during cytokinesis

During cell locomotion, actin assembles at the leading front of a moving cell and drives a local expansion of the plasma membrane through an elastic polymerization ratchet mechanism (Mogilner and Oster, 1996a). Actin filaments, interconnected by actin-crosslinking proteins that are abundant at the leading edge, are transported towards the rear region of the moving cell, where the actin meshwork is eventually disassembled. The actin cycle, polymerization at a membrane-apposed region of the cell, transport away from the membrane, and depolymerization in a distal region, is a generic process essential for many cellular activities that depend on actin cytoskeleton, such as directed cell movement, cell spreading, endocytosis, and also for cytokinesis (Gerisch and Weber, 2000). In some cell types filamentous actin moves backwards also relative to the substratum, in others it remains stationary. Whether one or the other takes place depends on a number of factors, primarily on a balance between polymerization and depolymerization activities and on the strength of coupling between the actin meshwork and the substratum (Heidemann and Buxbaum, 1998). The term retrograde actin flow is used to describe rearward transport of actin relative to the substratum. In fast moving cells like keratinocytes there is no significant actin flow, because translocation speed of the cell body matches the rate of the actin turnover. In slower, more strongly anchored cells like fibroblasts, F-actin has to be transported away from the front because its buildup outpaces the rate of the leading edge protrusion. Whether the retrograde actin flow is actively driven by a molecular motor or it develops as a consequence of a resistance of the anterior membrane to the thrust of actin polymerization at the leading edge is still a matter of debate, but in *Dictyostelium* the flow is for the most part independent of myosin II activity (Fukui *et al*., 1999; Aguado-Velasco and Bretscher, 1997).

In cytokinesis, the two polar regions of a dividing cell can be regarded as equivalent to immobilized leading edges of locomoting cells. Indeed, many actin-binding proteins involved in regulation of actin polymerization, cross-linking and depolymerization are localized to both zones in *Dictyostelium* cells, e.g. Arp 2/3 complex (Insall *et al*.,

2001), DAip1 (Konzok *et al*., 1999), coronin (Gerisch *et al*., 1995), p34 (Weber *et al*., 1999a), cofilin (Aizawa *et al*., 1995), talin A (Niewöhner *et al*., 1997), and myosin VII (Tuxworth *et al*., 2001). Since a dividing cell is immobilized, actin polymerized at the front has to be depolymerized to prevent accumulation of F-actin in the middle of the cell. The greater part of this depolymerization process occurs at the rear edge of the leading lammelipod, since the zone of a high F-actin concentration does not extend far into cytoplasm, except for a narrow layer of cortical F-actin that extends throughout the cell. Filamentous actin can in principle be recruited to the midzonal cortex via three routes: by polymerization *in situ*, through binding of the preformed actin filaments from the cytoplasm, and by means of a cortical actin flow. The question of relative contributions of the three routes is still not resolved in *Dictyostelium*, but there is evidence from this and other cell types that cortical flow plays a significant role in the process (Fukui *et al*., 1999; Fishkind and Wang, 1995).

In dividing *Dictyostelium* wild-type cells, there is no gross accumulation of filamentous actin in the midzone and the cleavage furrow (Neujahr *et al*., 1997b). Consequently, a steady supply of F-actin to the midzone must be counterbalanced by depolymerization at the same site. Indeed, when actin disassembly is inhibited by the use of agents that stabilize actin filaments, phalloidin and jasplakinolide, actin overassembles at the cleavage furrow and interferes with its constriction (Lee *et al*., 1998; Fukui *et al*., 1999). Likewise, overexpression of a C-terminal talin A fragment which tightly binds to actin filaments in *Dictyostelium* cells leads to a massive accumulation of F-actin in the cleavage furrow and a strong delay of cytokinesis (Niewöhner and Weber, unpublished).

Independent indication that actin disassembly at the midzone facilitates cytokinesis was provided by local application of cytochalasin D, which causes disruption of actin filaments, at the cleavage furrow site of dividing normal rat kidney epithelial cells (O'Connell *et al*., 2001). Application of cytochalasin D to one side of the midzone triggered a faster progression of furrowing on that side compared to the opposite side. Genetic evidence was provided that a protein directly involved in F-actin disassembly is essential for normal cell division. Mutations in *twinstar*, a *Drosophila* gene encoding a cofilin/ADF homologue, result in defects in centrosome migration and cytokinesis (Gunsalus *et al*., 1995). Interestingly, the mutations lead to abnormal accumulation of actin in the cleavage furrow and at the centrosomes. Large and persistent F-actin aggregates are formed at these sites, which interfere with the proper execution of cytokinesis. Also, redistribution of phosphatidylethanolamine from the inner to the outer leaflet of the plasma membrane in CHO fibroblasts results in a blockade of F-actin disassembly at the cleavage furrow and precludes completion of cytokinesis (Emoto and Umeda, 2000).

An active role for F-actin disassembly in constriction of the cleavage furrow

The presented body of evidence, obtained by diverse experimental approaches, points to the importance of an appropriate actin turnover and, in particular, of an unimpeded actin disassembly for cytokinesis. Thus, a delay or failure in disassembly of F-actin at the midzone of a dividing cell leads to its accumulation and creates an obstruction for ingression of the cleavage furrow. Is this simply a passive effect, or can actin disassembly also be functional in inducing the ingression? It has been shown theoretically that disruption of an actin network can generate a contractile stress (Mogilner and Oster, 1996b). Newly polymerized actin filaments at the leading edge are relatively flexible, that is they have a short persistence length and undergo thermal undulations. As these filaments are crosslinked into higher-order aggregates, their thermal writhing is suppressed and they assume a straighten configuration, which is accompanied with a decrease of entropy. In this process, actin network stores the entropic elastic energy of entangled filaments, which will provide the contractile stress upon breakage of filament interconnections. Interestingly, the contractile force generated by rupture of a single linkage between actin filaments has been estimated to correspond to the force generated by one myosin II molecule.

Experimental evidence that disassembly of a crosslinked network of a filamentous protein can generate contraction forces stems from ameboid sperm cells of *Ascaris suum*. Motility of nematode sperm cells is based on a cytoskeleton in which the role usually played by actin is taken over by the 14-kD major sperm protein (MSP). It has been shown that localized depolymerization of the MSP correlates with the forward movement of the cell body, when this movement is uncoupled from protrusion of the lamellipodium (Italiano *et al*., 1999). Based on these data, a push-pull mechanism for amoeboid cell motility has been postulated (Roberts and Stewart, 2000). In this model, lammelipodial protrusion is driven by localized assembly of the MSP cytoskeleton at the leading edge (the pushing mechanism). The bulk of the cell is transported by retraction forces induced by disassembly of the MSP fibres at the transition zone between the rear of the lammelipodium and the cell body (the pulling mechanism). No motors associated with MSP filaments have been identified, and since the MSP filaments, in contrast to actin filaments, do not have an intrinsic polarity, a directed movement of motor molecules along such filaments would not be feasible. Motility of *Ascaris* sperm cells has recently been successfully modeled theoretically (Bottino *et al*, 2002). In the model, it has been explicitly assumed that the cell body is pulled forward by contraction brought about by release of the entropic stress, which occurs through disassembly of a pre-stressed MSP cytoskeleton.

Interestingly, an overassembly of actin filaments at the

midzone is observed in dividing *Dictyostelium* mutant cells devoid of cortexillin (Neujahr and Weber, unpublished). So, it seems that formation of CMC plays a role in regulation of F-actin disassembly. It is not clear how a complex that contains an actin-bundling protein could induce disruption of actin filaments. A cortexillin heterodimer harbours multiple actin-binding sites and at least one lipid-binding site. Such molecular architecture would enable cortexillin to bind actin filaments to the lipid bilayer in a specific orientation and thus facilitate disassembly. It is conceivable that a fraction of actin filaments polymerized at a polar region gets linked to the plasma membrane at the rear side of lammelipodium and thus evade the depolymerization process at that site. These filaments could be crosslinked by cortexillin at the margins of the midzone and transported towards the center of the midzone by a centripetal actin flow. Inhibition of cortexillin actin-binding activity at the midzone, possibly by PIP_2 or through inactivation of CMC, would release interconnections between the filaments, set off its disassembly, and thus generate constriction of the cleavage furrow by the aforementioned entropic mechanism.

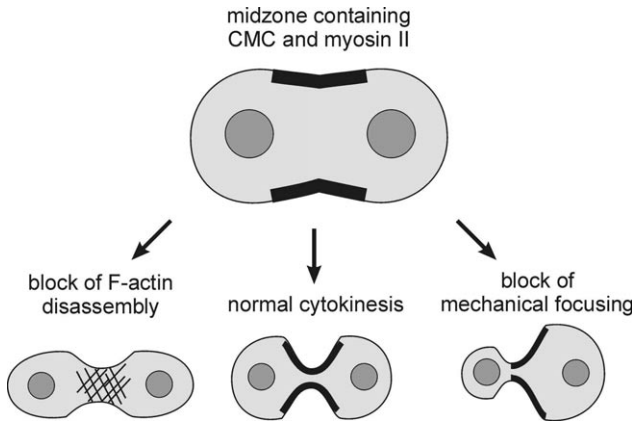

Fig. 2. Defective ingression of the cleavage furrow in *Dictyostelium* mutant cells. Block of filamentous actin disassembly corresponds to situation in cortexillin-deficient mutants. Block of symmetrical dissection of the midzone, or mechanical focusing, corresponds to inactivation of myosin II.

Towards an unified model of cytokinesis

In this article, an alternative view at the mechanism of cleavage furrow ingression in dividing *Dictyostelium* cells was presented. It was hypothesized that an entropic contractile stress generated by disassembly of entangled actin filaments initiates constriction at the midzone, and that myosin II stabilizes and focuses the furrow. Although the emphasis was put on processes taking place at the midzone of a dividing cell, expansion at the polar regions brought about by actin polymerization probably also contributes to cytokinesis. Molecules which control equilibrium between actin monomers and polymers, and which regulate global cortical tension, are also known to play an important role in cytokinesis of *Dictyostelium* cells (Haugwitz *et al*., 1994; Gerald *et al*., 1998; Robinson and Spudich, 2000). If myosin II is absent, a reliable focusing of the furrow to the midzone is not warranted, but traction forces at the poles still make possible a cleavage at the interface between the midzone and a polar region.

It is proposed that *Dictyostelium* cells use a universal, multifaceted mechanism to divide, irrespective of external conditions. From this perspective, different apparent types of cytokinesis result from disturbances of its constituent elemental processes. Erroneous variants of cytokinesis do not rely on separate mechanisms, but are the "stripped off" versions of the integral process, defective in some of its aspects (Fig. 2). Intervening into structural integrity or the spatio-temporal dynamics of elemental activities, by means of genetic, pharmacological or mechanical manipulation, will have more or less severe consequences for the entire process.

A related integral view on cytokinesis has been elaborated in a theoretical study of the sea urchin egg cleavage division, which was based on a comprehensive hydrodynamical model (He and Dembo, 1997). Although molecular mechanisms of elemental processes were assumed to be different from those proposed in the present article, it was emphasized that polar expansion and midzone contraction cooperate to produce the cleavage. Necessity for cycling of cytoskeletal material between its polymerized and depolymerized states was also implied. Stability of numerical simulation of cytokinesis depended critically on a parameter describing the rate of actin depolymerization. In particular, too low values of this parameter led to an accumulation of the material in the cortical furrow region and to a stall of cytokinesis.

In conclusion, I would like to propose that the suggested multifaceted model of cytokinesis in general, and the mechanism of cleavage furrow ingression in particular, are of some universal significance and not restricted only to amoeboid *Dictyostelium* cells. It is possible that, in the course of evolution, molecular basis of one or another elemental process that contribute to cytokinesis in *Dictyostelium* was superseded or reinforced by more efficient and reliable molecular mechanisms. Nevertheless, mechanical principles of cytokinesis that are becoming apparent from studies in *Dictyostelium* can provide a new perspective for analyzing mechanism of cell division in other organisms.

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