Beyond polymer polarity: how the cytoskeleton builds a polarized cell

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Abstract | Cell polarity relies on the asymmetric organization of cellular components and structures. Actin and microtubules are well suited to provide the structural basis for cell polarization because of their inherent structural polarity along the polymer lattices and intrinsic dynamics that allow them to respond rapidly to polarity cues. In general, the actin cytoskeleton drives the symmetry-breaking process that enables the establishment of a polarized distribution of regulatory molecules, whereas microtubules build on this asymmetry and maintain the stability of the polarized organization. Crosstalk coordinates the functions of the two cytoskeletal systems.

Septin family
A conserved family of GTP-binding proteins that were first identified in yeast and later found in a wide range of fungi and animal cells.

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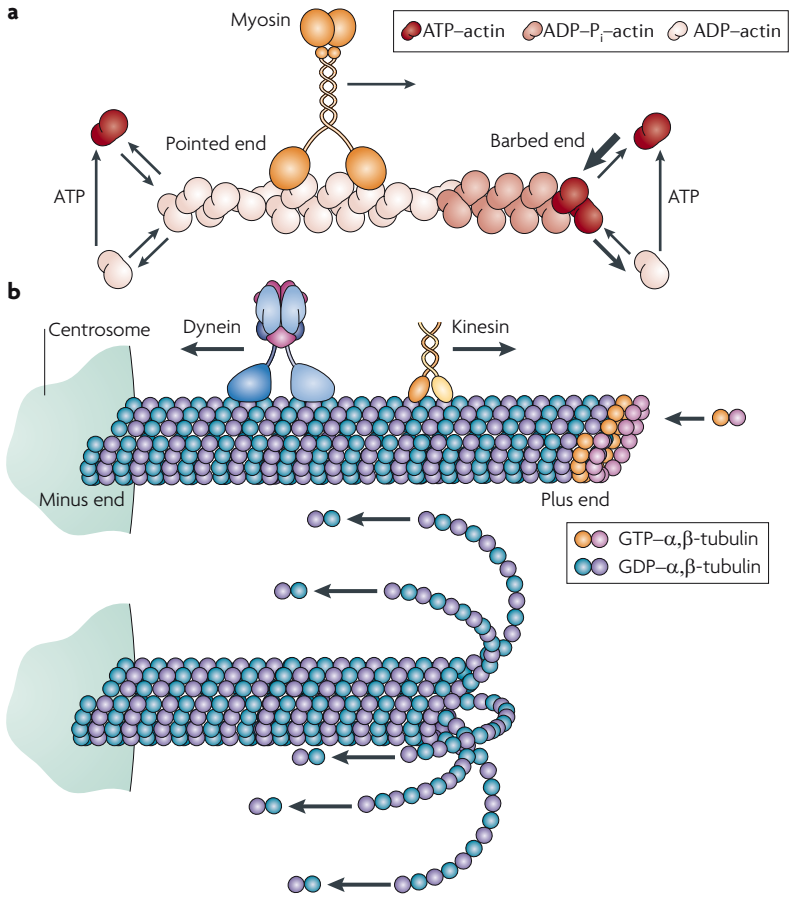
doi:10.1038/nrm2522

Cell polarity results from a vectorial axis that directs the internal organization of a cell, and is observed in most differentiated cell types of metazoans and in unicellular organisms, such as yeast, ciliated protozoa and even prokaryotes. Although observed in different forms in diverse cell types and species, cell polarity is defined by two fundamental properties: the asymmetric accumulation of mobile components (often regulatory molecules) between opposite poles of a cell; and the orientated organization of inherently polar cytoskeletal filaments (particularly actin and microtubules) along the axis of polarity (BOX 1). These properties coexist in most polarized cells, and their interactions are crucial to both the establishment and the maintenance of cell polarity.

Placement of an intrinsically polar object inside a symmetrical entity breaks the host entity's symmetry. Actin filaments (or microfilaments) and microtubules are polar polymers that are composed, respectively, of globular actin (G-actin) subunits that bind and hydrolyse ATP, and α- and β-tubulin heterodimeric subunits that bind and hydrolyse GTP. Polarity results from head-to-tail association of protein subunits, resulting in polymer lattices in which all of the subunits align in the same direction and the two ends differ structurally (BOX 1). Actin and microtubules are also dynamic polymers, of which each end can either polymerize or depolymerize and net growth depends on free subunit concentrations[1,2]. Rate constants that govern growth and shrinkage differ at the opposite ends of the polymers owing to structural differences of the two ends. Such dynamic differences are enhanced through nucleotide hydrolysis by actin and tubulin subunits. The ability to undergo fast turnover and assembly enables actin and microtubules to reorganize rapidly and locally in response to polarity signals. The cell further capitalizes on the intrinsic polarity and dynamics of actin and microtubules through a large number of cytoskeleton-associated proteins, which translate asymmetry in the structure and dynamics of the polymer into polarized functions. Intermediate filaments, however, are nonpolar and are not generally involved in the generation of cell polarity[3]. This generalization might not last, as recent studies have shown that members of the septin family, which form nonpolar polymers that resemble intermediate filaments, are important for cell polarity in a number of cell types[4–6].

A class of cytoskeleton-associated proteins that are particularly important for cell polarity are the motor proteins. These power unidirectional movement along actin filaments or microtubules by irreversibly moving from one tightly bound conformation to another using the energy of ATP hydrolysis (for recent reviews, see REFS 7–10). Myosins are motor proteins for actin, and most members of the myosin superfamily (except myosin-VI) move towards actin barbed ends (BOX 1). Some myosins, such as myosin-V, move processively along actin filaments (that is, they move many consecutive steps before dissociating from the filament) and are thus suitable for transport over long distances. Other myosins, in particular myosin-II, exhibit low processivity but can generate contractile movement through the sliding of actin filaments. Microtubule motors encompass kinesins and dynein. Most kinesins move towards microtubule plus ends with varying degrees of processivity, whereas dynein moves processively towards microtubule minus ends in the presence of the dynactin complex[11] (BOX 1). In principle, cargo molecules or organelles can be trafficked

Box 1 | **Basic properties of actin filaments and microtubules**



Actin filaments are composed of subunits that are orientated in the same direction, which results in a polarized surface lattice and two structurally distinct ends (barbed and pointed; see figure, part **a**). Motor proteins (myosins) move in a unidirectional manner along the filament surface owing to repeated interactions with the subunits in consistent orientations. The barbed and pointed ends of an actin filament exhibit different subunit association and dissociation rates. They also exhibit different critical concentrations for the assembly of ATP–actin as a result of ATP hydrolysis and phosphate release soon after subunit incorporation into the filament. Asymmetry in the state of the nucleotide that is bound along the filament, and structural differences between the two ends enable many actin-binding proteins to differentially regulate their dynamics and stability *in vivo*.

Microtubules are composed of α-tubulin and β-tubulin heterodimeric subunits that are orientated in the same direction, which results in a polarized surface lattice and two structurally distinct ends (plus and minus ends; see figure, part **b**). The minus ends of microtubules are often anchored at the centrosome or other microtubule-organizing centres (MTOCs). Motor proteins (kinesins and dynein) move in a unidirectional manner along the filament surface owing to repeated interactions with the consistently orientated subunits. Tubulin hydrolyses GTP shortly after assembly, resulting in a cap of GTP-containing tubulin subunits on growing plus ends. Because GTP–tubulin and GDP–tubulin have different dissociation rates, microtubules depolymerize rapidly following the loss of the GTP cap. The alternation between growth (top panel) and shrinkage (bottom panel) of microtubules, with infrequent transitions, is known as dynamic instability and contributes to the ability of microtubules to search for polarity factors. Growing and shrinking microtubules have different structures at their ends and this is thought to allow shrinking microtubules to exert force on structures (for example, kinetochores) that may remain associated.

Note that microtubules can assemble in A or B lattices with different lateral contacts between protofilaments. The microtubule depicted is in the B lattice (lateral contacts αα and ββ with a seam of αβ). Most evidence suggests that the B lattice is the predominant form in cells. However, a recent study suggests that microtubule plus end-tracking proteins (+TIPs) might drive microtubules into the A lattice, in which the lateral contacts are αβ (REF. 174).

by motor proteins to specific cellular locations once a cell has established orientated actin or microtubule arrays.

We focus on how polarized arrays of actin and microtubules are generated and how they contribute to the different phases of cell polarity through their dynamic assembly and transport functions. To facilitate the comparison of actin and microtubule functions, we divide the process of cell polarity development into 'symmetry breaking' and 'maintenance of polarity' processes. The term symmetry breaking was chosen over 'establishment of polarity' to highlight the rapid, decisive cellular transition from a symmetric distribution of relevant constituents to an asymmetric distribution along a certain cellular axis. In many well-studied systems, such as yeast and neutrophils, symmetry breaking can occur as a stochastic process that is driven by intrinsic cellular mechanisms. In other systems, however, such as *Caenorhabditis elegans* zygotes, cellular symmetry breaking might depend on an initial external cue (for example, sperm entry). Although our focus in this article is strictly on cytoskeletal-based mechanisms, we also note that there are important contributions by membrane constituents, particularly phosphoinositides (for example, REF. 12), and by the cooperative assembly of signalling complexes (for example, REF. 13) to both symmetry breaking and polarity maintenance.

Forming polarized cytoskeletal arrays

For cytoskeletal polymers to carry out polarized trafficking or localized functions through the dynamics of their ends, actin and microtubules must assemble into organized arrays. The rate-limiting step for spontaneous actin and microtubule polymerization is nucleation — the formation of small oligomers that can rapidly elongate[14,15]. A key mechanism in the assembly of polarized actin arrays is the activation of actin-nucleation factors, such as the actin-related protein-2/3 (Arp2/3) complex and formin-family proteins, at defined locations[16] (FIG. 1a). In cells that undergo crawling motility, Arp2/3 complex-nucleated actin filaments form a branched actin network in the lamellipodium, with filament barbed ends facing the leading-edge membrane[17,18]. This polarized actin organization is determined not only by the intrinsic topology of Arp2/3 complex-mediated actin branching, but also by plasma membrane-associated proteins that control the activation of the Arp2/3 complex and regulate the elongation and turnover of the nucleated filaments. Membrane-bound Rho-family GTPases activate actin-nucleation factors either directly (in the case of formins) or indirectly through nucleation-promoting factors (NPFs)[19]. Through these upstream regulatory factors, Arp2/3 complex- or formin-based actin nucleation occurs maximally near the membrane at sites of Rho GTPase activation.

Whereas nucleation is the principal mechanism that regulates actin during cell polarization, nucleation of microtubules usually occurs near the cell centre at the centrosome (or other microtubule-organizing centres (MTOCs)), which makes it distal to membrane-derived signals that stimulate cell polarity. Consequently, the initial events that localize and orientate microtubules

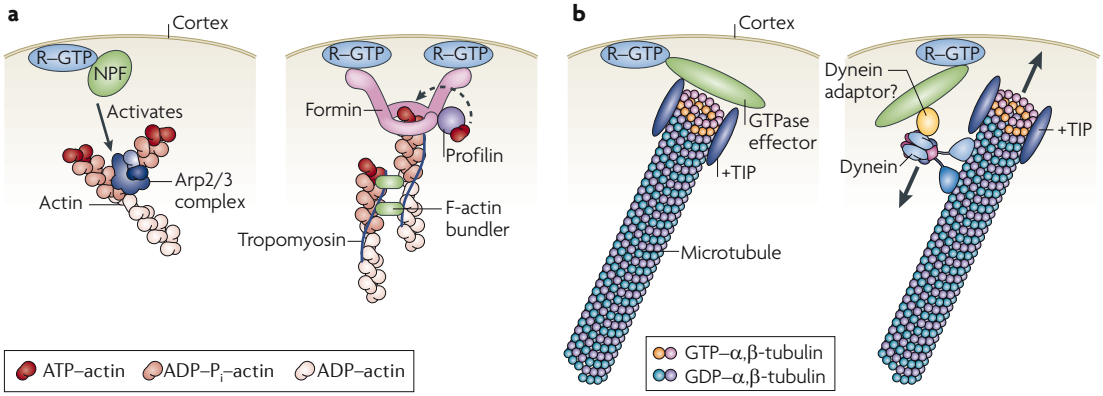


Figure 1 | **Establishment of orientated cytoskeletal arrays. a** | The actin-related protein-2/3 (Arp2/3) complex and formin-family proteins are highly conserved actin-nucleation factors that are important for assembling polarized actin-filament arrays[16]. The Arp2/3 complex is a seven-subunit complex that binds to an existing actin filament and nucleates a new filament at a ~70° angle through its two actin-related proteins, ARP2 and ARP3 (left panel). Formin proteins nucleate actin filaments through their conserved formin homology-2 (FH2) domain, and filament elongation is facilitated by the FH1 domain, which binds to profilin (right panel). Actin nucleation occurs primarily at the cell cortex because nucleation factors are activated directly (for example, some formins) or indirectly (for example, the Arp2/3 complex) through nucleation-promoting factors (NPFs) by the membrane-bound Rho-family GTPases (R–GTP indicates an active Rho-family GTPase). The Arp2/3 complex nucleates the formation of dendritic actin arrays, whereas formins nucleate straight actin filaments that are often decorated with tropomyosin. Depending on the types of crosslinking or motor proteins that are present, these filaments can be organized into parallel bundles (as shown) or contractile networks (see FIG. 2b,c). **b** | Microtubule capture near membranes is mediated by a diverse set of Rho GTPase effectors (TABLE 1) that transmit the activation of Rho GTPases into action on the plus ends of microtubules. Most microtubule capture events stabilize microtubules at least transiently. Proteins that bind to the plus ends of growing microtubules (+TIPs) also contribute to microtubule capture (left panel), and many of these proteins also regulate microtubule dynamic instability by altering assembly rates or transitions between growing and shrinking microtubules. Microtubule capture that involves the motor protein dynein (right panel) can generate pulling forces on microtubules and contribute to centrosome positioning.

during cell polarization mainly involve factors that regulate the dynamic plus ends of microtubules. One frequently used mechanism during cell polarization is microtubule capture by cortical factors that increase the stability of the plus ends and/or generate pulling forces on microtubules[20,21] (FIG. 1b). Other mechanisms include alterations in the assembly properties of microtubules and the bundling of microtubules.

Cortical capture usually involves the interaction of two classes of proteins: proteins that are specifically associated with the plus ends of microtubules (termed +TIPs) and cortical factors that are controlled by Rho GTPases and other membrane-proximal signalling factors (TABLE 1). +TIPs include proteins that are primarily associated with the plus ends of microtubules, for example, end-binding protein-1 (EB1), and other proteins, such as cytoplasmic dynein, that are also found at other sites in the cell. Depending on the factors that are activated, cortical capture results in a range of stabilization from transient increases in microtubule pausing, which last for seconds to minutes, to the long-term capping of microtubules, which persists for hours. Microtubule capture increases the local microtubule density and also provides a means to enhance the delivery of cargoes to specific sites. The best example of direct delivery comes from *Schizosaccharomyces pombe*, in which transiently paused microtubules at the cell poles deliver tip elongation aberrant protein-1 (Tea1; a kelch-repeat protein) and Tea4 (an SH3-domain protein) to regulate actin filaments through the recruitment of the formin For3 (REF. 22). In mammalian cells, long-term (>10–30 minutes) stabilized microtubules become post-translationally modified, and this is known to enhance kinesin-dependent motility in a number of cases (see below).

In microtubule capture that involves dynein, pulling forces that are directed towards cortical sites can influence centrosome or MTOC position and hence microtubule nucleation sites (FIG. 1b). Originally described in the dividing cells of *Saccharomyces cerevisiae* and in asymmetrically dividing *C. elegans* early embryos[23,24], dynein-dependent centrosome positioning also occurs in polarized migrating cells and in T cells that interact with targets (see below). Pulling forces on microtubules also result from captured microtubules that undergo controlled shrinkage, although this mechanism is only known to occur in dividing *S. cerevisiae*[25] and perhaps *C. elegans* one-cell embryos[26].

Cellular symmetry breaking

Historically, the cytoskeleton and its motors were viewed to be under the passive instruction of regulatory molecules that have been localized by pre-existing spatial cues. However, results from many model systems have increasingly strengthened a revised view that the cytoskeleton not only mediates the downstream functions of polarized signals but, more importantly, also drives the symmetry-breaking process by localizing key regulatory molecules to specific cortical sites through motor proteins and the dynamic assembly of mutually exclusive structures.

Lamellipodium
A broad, flat cellular protrusion that contains extensively branched and crosslinked arrays of actin filaments. These are orientated with their barbed ends towards the plasma membrane.

Table 1 | **Microtubule capture factors**

Cortical factor	Regulation	+TIP	Microtubule effect	Refs
Metazoa				
IQGAP1	CDC42 and Rac	CLIP170	Transient stabilization (24–120 seconds)	161
mDia1	Rho	EB1 and APC	Long-term stabilization (hours)	129,131,162
LL5β	PI3K?	CLASP (Rac and GSK3β regulate CLASP)	Transient to long-term stabilization	136,138,163, 164
PAR6	CDC42	Dynein and dynactin	Unknown	122,123,126
β-catenin	Unknown	Dynein	Transient stabilization (>2 minutes)	165,166
Saccharomyces cerevisiae				
Unknown	Cdc42 and Rho	Bim1 and Kar9	Transient stabilization and shrinkage	167–169
Num1	Unknown	Dynein and dynactin	Transient stabilization and pulling	167,170
Schizosaccharomyces pombe				
Mod5	Unknown	Tea1, Tea2, Tea4, Tip1 and Mal3	Transient stabilization (90 seconds)	171–173

APC, adenomatous polyposis coli; CLASP, CLIP-associating protein; CLIP170, cytoplasmic linker protein-170; EB1, end-binding protein-1; GSK3β, glycogen synthase kinase-3β; IQGAP1, IQ motif-containing GTPase-activating protein-1; PAR6, partitioning defective-6; PI3K, phosphoinositide 3-kinase; +TIP, microtubule plus end-tracking protein; Tea, tip elongation aberrant protein.

Symmetry breaking through a processive myosin. The budding yeast *S. cerevisiae* switches from isotropic to polarized growth to form a bud during vegetative proliferation, or during the formation of a mating projection in response to pheromone. Two types of actin structures have been implicated in yeast polarized growth: actin cables, which serve as growth-site-directed transport highways for cargoes, such as secretory vesicles, protein–RNA complexes and various organelles; and cortical actin patches, which are endocytic structures that are concentrated in the general area of the growth site[27]. Actin cables are made up of parallel arrays of actin filaments that are nucleated by two formins, Bni1 and Bnr1, and are assembled into long bundles through the stabilization and bundling effects of tropomyosins and fimbrin, respectively[28–31]. Formation of orientated actin cables relies on the localized activation of Bni1 and/or Bnr1. In turn, this activation relies on active Rho GTPases, as do other diaphanous (Dia)-related formins. Such Rho GTPases include Cdc42 and four of the Rho proteins[32]. Cdc42 is the master regulator of cell polarity in yeast, as in most other eukaryotes[33] (see also the review by Iden and Collard in this issue). Cdc42 binds to the N-terminal Rho-binding domain of Bni1 and is required for the polarized orientation of actin cables[34]. However, genetic analysis suggested that the Rho proteins might be the primary GTPases that control formin activation, whereas Cdc42 has a yet undefined role in localizing this process to the polar cortex[35,36].

Even though the biochemical steps between Cdc42 and the polarized assembly of actin cables are elusive, it is clear that the formation of actin cables is an important target of Cdc42 to control polarized transport. However, recent studies in yeast suggest that actin, in turn, affects Rho GTPase localization through its transport function[37,38]. Such a feedback interaction enables actin to have an active role in the symmetry-breaking process that leads to a polarized distribution of active GTPase. This is best appreciated in an experimental system in which nonpolarized, G1-arrested yeast cells can polarize spontaneously on induction of constitutively active Cdc42 (FIG. 2a). Blocking actin polymerization or myosin-V transport prevents Cdc42–GTP-induced spontaneous polarization, indicating that actin does not passively respond downstream of Cdc42 (REF. 37). A mathematical simulation has shown that a positive-feedback loop that is composed of Cdc42-directed actin-cable formation and actin-cable-mediated Cdc42 transport to the plasma membrane is sufficient, in theory, to turn an initially random distribution of Cdc42–GTP on the cortex into a stable cortical Cdc42 localization, towards which actin cables are orientated. The contribution of this actin-based symmetry-breaking mechanism to physiological polarization was later shown[39]. Along with this, an earlier study[13] also found a parallel cytoskeleton-independent mechanism for symmetry breaking that might involve the cooperative assembly of a signalling complex that regulates Cdc42 activation.

Symmetry breaking through a contractile myosin. Cell polarity is crucial for asymmetric cell divisions, a developmental mechanism that is required for cell fate diversification. In the *C. elegans* zygote, an asymmetric cell division occurs along a plane that is perpendicular to the anterior–posterior (A–P) polarity axis that is established shortly after fertilization[24]. A–P polarity is defined by the localization of distinct sets of PAR proteins (partitioning defective proteins) to opposing cortical hemispheres of the zygote: the highly conserved proteins PAR-3, PAR-6 and atypical protein kinase C (aPKC) localize to the anterior cortex of the zygote, whereas other proteins, such as PAR-1 and PAR-2, localize to the posterior cortex. This asymmetric patterning of cortical determinants is required for the correct orientation and positioning of the first mitotic spindle, which in turn determines the plane of the first cell division.

PAR protein
One of a set of six proteins that were initially identified in *Caenorhabditis elegans*. Inactivation of the PAR proteins results in a partitioning-defective phenotype in early embryos. PAR proteins are now recognized to be widely involved in cell polarity.

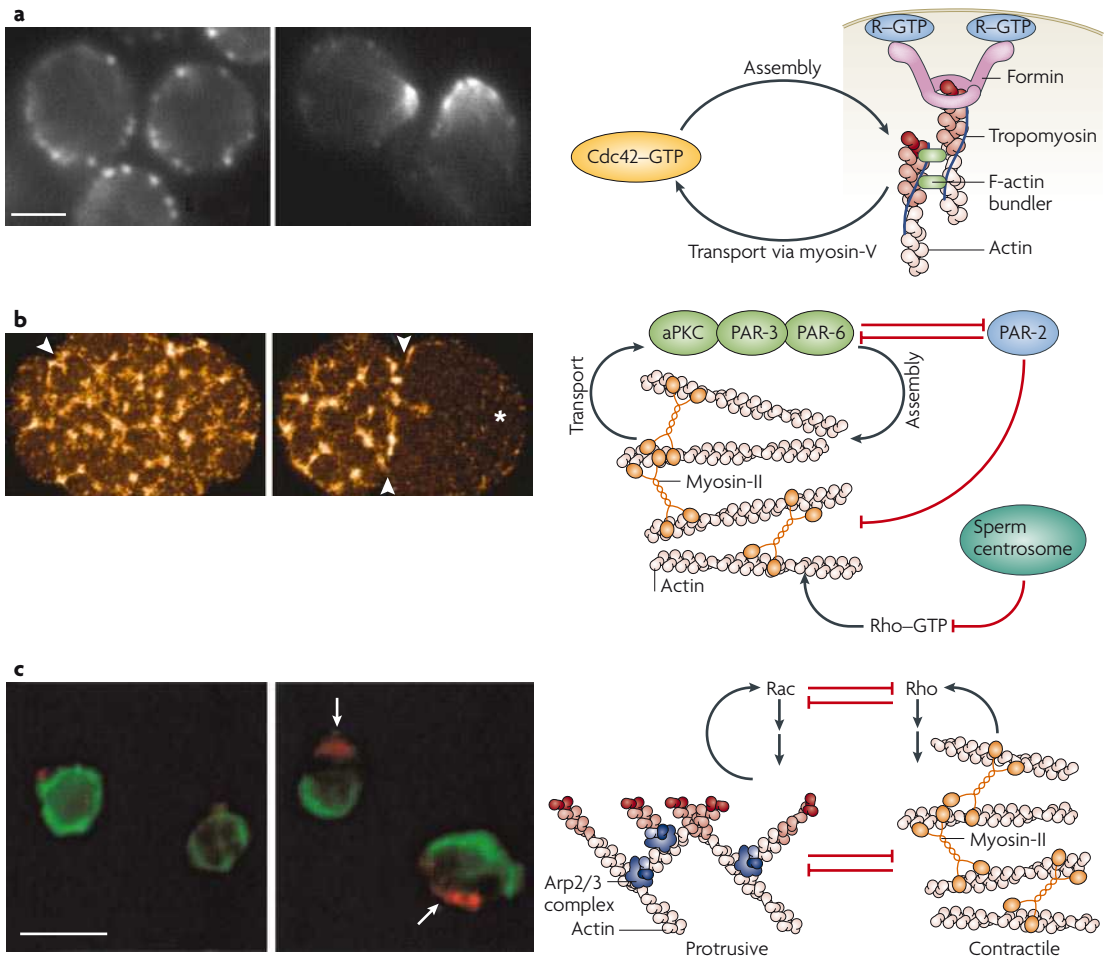


Figure 2 | **Mechanisms of actin-based symmetry breaking in the establishment of cell polarity. a** | Symmetry breaking through an actin and myosin-V-mediated positive-feedback loop. G1-arrested (nonpolar) yeast cells can be induced to polarize spontaneously following expression of constitutively active Cdc42. The images show the distribution of actin in G1 cells that express non-activated Cdc42 (left) or a constitutively active form of Cdc42 (right; left panels). Amplification of stochastic asymmetry in the distribution of Cdc42 through a feedback loop that is composed of Cdc42-stimulated, polarized formation of actin cables and actin cable- and myosin-V-mediated Cdc42 transport (right panel). This positive-feedback loop can lead to symmetry breaking by amplifying stochastic variation in Cdc42 distribution along the membrane. Scale bar, 3 μm. **b** | Symmetry breaking through an actin- and myosin-II-based contractile network in a *Caenorhabditis elegans* one-cell embryo. The images show the cortical network of myosin-II before (left) and after (right) symmetry breaking (left panels). Arrowheads indicate furrows on the egg surface. An asterisk marks the site of sperm entry. Sperm entry locally weakens the contractile network by inhibiting Rho activation, and this initial asymmetry is amplified through feedback interactions between the actomyosin contractile machine and partitioning defective (PAR) and Rho signalling modules, eventually driving stable segregation of anterior and posterior determinants and the establishment of anterior–posterior polarity (right panel). **c** | Establishment of front-to-back polarity in chemotactic neutrophils through mutual inhibition at the level of pathways and cytoskeletal assembly. The fluorescent images show the concentration of Rho (green) and dense protrusive actin (red) to the back and front, respectively, of neutrophils before (left) and after (right) stimulation with the chemoattractant formylated Met-Leu-Phe (fMLP) (left panels). Actin was also present at the back, but the fluorescence signal was too low for it to be observed in these images. Mutual inhibitory interactions between the Rho and Rac signalling pathways and between contractile and protrusive actin structures lead to localization of active Rho and Rac and to assembly of their respective actin structures to opposing cell ends, thus establishing front–back polarity (right panel). Scale bar, 10 μm. aPKC, atypical protein kinase C; Arp2/3, actin-related protein-2/3; R–GTP, an active Rho-family GTPase. Images in part **a** courtesy of R.L. Images in part **b** modified, with permission, from REF. 42 © (2004) Cell Press. Images in part **c** modified, with permission, from REF. 71 © (2003) Cell Press.

Actomyosin
A complex of myosin and actin filaments that is responsible for a range of cellular movements in eukaryotic cells. Myosins can translocate vesicles or other cargo on actin filaments or slide actin filaments to generate contraction.

The localization of anterior determinants is achieved in an actin- and myosin-dependent manner. However, in contrast to the mechanism in yeast, the transport process is powered by myosin-II[40–42]. Sperm entry into the oocytes locally weakens the actomyosin network at the presumptive posterior pole, leading to net contraction of the network in the anterior direction (FIG. 2b). PAR-3 and PAR-6 localize to punctate structures that dynamically associate with cortical actin and move along with the contractile network. The depletion of myosin-II using

RNA interference (RNAi) prevents the concentration of these proteins towards the anterior cortex[42]. However, PAR-3 and PAR-6 proteins are not passive cargoes of the contractile network, as contractile movement is severely diminished in embryos that lack these proteins[42]. The posteriorly localized PAR-2, however, inhibits cortical recruitment of myosin and posterior-directed contractile movement during the maintenance phase. PAR-2 and the anteriorly localized PAR-3–PAR-6–aPKC cohort also antagonize the ability of one another to associate with the cortex[43,44]. This network of interactions (FIG. 2b) might strongly amplify the small initial asymmetry in contractile force that is induced by sperm entry, leading to stable segregation of the PAR proteins and thus well-defined cell polarity.

It is interesting that this symmetry-breaking system seems to require the spatial cue that is provided by the sperm entry site, whereas in many other polarization events spatial cues orientate the polarity axis but might not be required for symmetry breaking *per se*[45]. The sperm cue seems to affect the activation of Rho GTPase[46–48], which exerts dual control over the assembly of the actomyosin network: Rho–GTP stimulates myosin-II activation and filament assembly by promoting the phosphorylation of the myosin-II regulatory light chain (MRLC), and also activates formin proteins to nucleate actin polymerization[49]. It will be interesting to determine, perhaps with the help of mathematical modelling, whether the *C. elegans* zygote system is capable of spontaneous symmetry breaking through the feedback loops in the network that connect PAR proteins and actomyosin contractility.

The involvement of an actomyosin contractile network in cell polarization might not be unique to *C. elegans* zygotes, as suggested by recent findings in epithelial cells. Epithelial cells in their differentiated state exhibit an apical–basal polarity, which defines the microvilli-rich apical domain and a basal–lateral domain that mediates cell–cell and cell–matrix attachment[50]. There is strong evidence for the roles of cell–cell and cell–matrix contacts in the establishment and maintenance of apical–basal polarity. However, a surprising study has found that ectopic activation of the kinase LKB1 in epithelial cells induces spontaneous symmetry breaking and establishes well-defined apical and basolateral domains in the absence of cell–cell contacts[51]. LKB1 is a tumour suppressor protein that is homologous to *C. elegans* PAR-4, which is required for the specification of the posterior cortex[52]. More recently, it was found that in this polarity model, the target of LKB1 is the AMP-activated kinase AMPK, the activation of which also responds to energy deprivation[53,54]. At least one of the substrates of AMPK in the induction of cell polarity is MRLC. Remarkably, expressing a constitutively phosphorylated (active) form of MRLC was sufficient to induce symmetry breaking in otherwise nonpolarized epithelial cells. Although mechanistic details of this process are unclear, it is enticing to speculate that a mechanism that involves the activation of myosin-II-based contractility, which is similar to that described in *C. elegans*, might be key to the establishment of epithelial polarity, given that PAR-3, PAR-6 and aPKC are required for the establishment of epithelial polarity[55].

Symmetry breaking through competitive actin assembly. Polarization is an important first step in cell migration, through which cells establish a protrusive front (the leading edge) and contractile rear. Although both are rich in actin, the filaments of these cellular domains are organized into distinct structures[18]. The leading edge is a protrusive actin-rich structure, in which the filaments are organized in dendritic (in lamellipodia) or parallel (in filopodia) arrays. Lamellipodial actin filaments are likely to be generated through nucleation by the Arp2/3 complex, although one study has suggested that lamellipodial actin might also be assembled in an Arp2/3-independent manner[56]. Other actin-binding proteins, such as ADF or cofilin (hereafter referred to as ADF/cofilin) and capping protein, control the dynamics of the actin at the cell front[57]. Actin at the rear of migrating cells is organized into contractile structures that are rich in myosin-II, a protein that is mostly absent in the protrusive lamellipodia. Contractile actin filaments are often coated with tropomyosins, a family of long coiled-coil polypeptides that form head-to-tail interactions along actin filaments[58], which can influence myosin-II mechanochemistry[59] and might also facilitate actin polymerization through formins[60].

In terms of their assembly, these two types of actin structures are mutually exclusive, both at the structural level and at the regulatory level. *In vitro* biochemistry shows that tropomyosin-decorated filaments cannot be branched by the Arp2/3 complex to form dendritic arrays[61], possibly owing to an overlap between the tropomyosin-binding site and the Arp2/3-binding site on actin[62]. Actin filaments that are bound to certain tropomyosin isoforms are also protected from severing and depolymerization by ADF/cofilin[63,64]. In addition, these two types of actin structures might compete for actin monomers during their assembly. Arp2/3-mediated actin nucleation is stimulated by Rac, whereas Rho promotes the assembly of contractile actomyosin networks[19,49]. In turn, protrusive actin and myosin-II positively influence the activation of their upstream GTPases[65,66]. Active Rho and Rac GTPases also mutually inhibit each other[67–70] (see also the review by Iden and Collard in this issue).

In neutrophils, the mutual exclusivity of the front and rear actin assembly pathways is crucial for polarization and directional motility in response to chemoattractants[71,72] (FIG. 2c). Inhibition of Rho or its downstream targets, such as myosin-II, induces multiple protrusive actin structures and causes the failure to establish a unique axis of cell polarity. Conversely, inhibition of protrusive actin assembly results in the spreading of active RhoA, which is normally restricted to the cell rear. A mathematical model based solely on mutually inhibitory interactions between the front and back pathways has shown that this network is sufficient to drive symmetry breaking and stable segregation of the front and rear cytoskeletal domains[73]. This model might also explain the ability of mechanically induced contraction of nonpolarized cells to generate sustained front–back polarity and to initiate migration[74].

Filopodium
A thin cellular process that contains long, unbranched, parallel bundles of actin filaments.

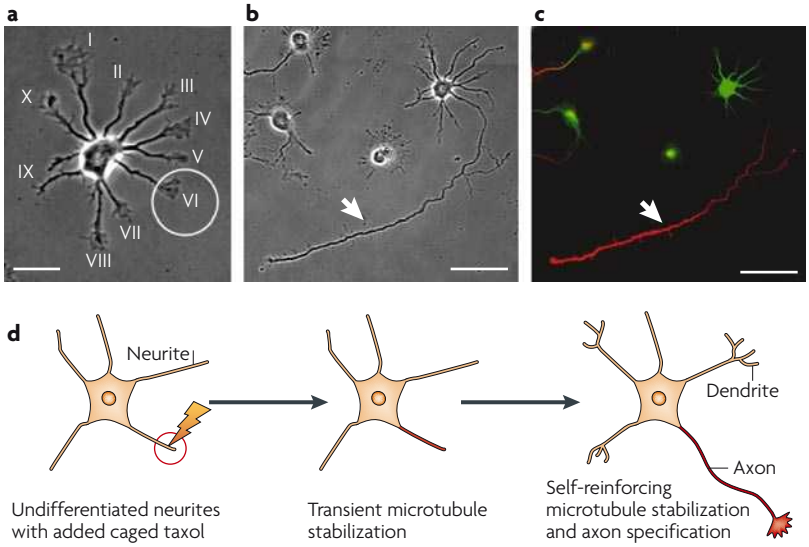


Figure 3 | **Microtubule-based symmetry breaking in neurons.** Images in parts **a**–**c** show that local and transient activation of taxol stimulates microtubule stabilization in a single undifferentiated neurite, which causes axon formation (shown by an arrow). **a** | A neuron just before ultraviolet-mediated photoactivation (for 10–15 minutes) of caged (that is, shielded or masked) taxol at the tip of the circled neurite. Scale bar, 20 μm. **b** | Almost 2 days later, the neurite treated with taxol has become an axon. Scale bar, 50 μm. **c** | An immunofluorescence image of the neuron in part **b** showing that an axonal marker (dephosphorylated tau; red), but not a dendritic marker (microtubule-associated protein-2 (MAP2); green), is present in the axon. Scale bar, 50 μm. **d** | A schematic summary of the result in panels **a**–**c** that shows that transient microtubule stabilization by taxol is sufficient to establish self-reinforcing microtubule stabilization and axon specification. Parts **a**–**c** modified, with permission, from REF. 80 © (2008) Rockefeller University Press.

Symmetry breaking through microtubules. Neurons are highly polarized cells that typically have a single long, thin process (the axon) to transmit information, and multiple shorter and thicker processes (the dendrites) to receive information. Studies of cultured embryonic rodent hippocampal neurons have provided a wealth of information on the factors that are involved in specifying neuronal cell polarity[75,76]. Cultured hippocampal neurons initially extend multiple morphologically indistinguishable, undifferentiated neurites. This symmetry is broken when one of the neurites begins to grow rapidly and acquires axonal markers (such as dephosphorylated tau, a microtubule-associated protein (MAP)), whereas the other neurites remain short. Over time in culture, the short neurites begin to grow and differentiate into dendrites by acquiring dendritic markers (such as MAP2).

It has long been known that microtubules in neuronal processes are more stable than the dynamic microtubules in proliferating cells, as shown by their resistance to microtubule-depolymerizing agents and by their higher levels of post-translationally modified tubulin[77–79]. In a recent study of hippocampal neurons, quantification of the ratio of acetylated tubulin to total tubulin showed an elevated ratio in axons compared with undifferentiated neurites[80]. Even before axon formation, one of the morphologically indistinguishable neurites accumulated acetylated tubulin. Importantly, microtubule stabilization was sufficient to induce axon formation, as local stabilization of microtubules in an undifferentiated neurite by photoactivation of a taxol analogue provided strong bias for the treated neurite to become an axon (FIG. 3).

These recent studies make it important to determine how neurons initially stabilize microtubules in the axon. A large number of factors have been implicated in the regulation of the stability of microtubules in axons, including kinases (for example, glycogen synthase kinase-3 (GSK3), PAR1 or SAD, LKB1 and aPKC), adaptors (for example, collapsin response mediator protein-2 (CRMP2)) and MAPs (tau, adenomatous polyposis coli (APC) and MAP1B)[75,76]. However, it is still unclear which of these factors are involved in the initial stabilization of microtubules in the axon and how they might be restricted to the presumptive axon. One possibility is that the centrosome might enhance the delivery of one or more of these factors to the presumptive axon, although the role of centrosome position in axonal specification is controversial[81,82]. Alternatively, localized actin destabilization, which can stimulate axon outgrowth[83,84], might be involved.

The processes affected by enhanced microtubule stabilization in axons are not clear. There is a precedent for enhanced delivery of membrane cargoes to the axon through kinesin-dependent transport, and even for certain membrane enzymes to contribute to axonal fate[85,86]. A more unexpected cargo is the PAR3 polarity protein, which binds directly to kinesin-3 (KIF3A)[87]. PAR3, PAR6 and aPKC are required for axon specification in hippocampal neurons[88,89] (although not in *Drosophila melanogaster* neurons[90]) and might enhance the formation of stable microtubules through negative regulation of MAP/microtubule affinity-regulating kinase (MARK) or the MARK-related kinase PAR1 (hereafter referred to as MARK/PAR1)[91]. MARK/PAR1 can destabilize microtubules by phosphorylating MAPs[92]. Microtubule stabilization directly affects kinesin-based transport: post-translational modifications, such as detyrosination, polyglutamylation and acetylation, enhance kinesin-1 binding to microtubules and contribute to transport processes in neurons and fibroblasts[93–96]. Intriguingly, the information for the enhanced transport to axons might reside in the kinesin motor domain, because kinesin-1 heads are sufficient to direct kinesin to the axon[97,98]. Taken together, the interactions described above outline a potential positive-feedback loop between microtubule, kinesin-based transport and signalling molecules, such as PAR3, in the initial symmetry breaking that establishes neuronal polarity.

Maintenance of cell polarity

Once established, cell polarity varies greatly in longevity, depending on the cell type. Polarity must be stable in cells that require it to maintain their highly differentiated states and function (for example, neurons and epithelial cells), whereas stability might be less important in neutrophils that chase after tumbling bacteria or in yeast cells that are searching for mating partners. Instead, the ability to respond sensitively to spatial and temporal changes in external stimuli (chemoattractant

Detyrosination
The post-translational removal of the C-terminal Tyr residue of α-tubulin by an as yet unidentified carboxypeptidase.

Polyglutamylation
The post-translational addition of chains of Glu residues to the γ-carboxyl groups of specific Glu residues of α- and β-tubulin.

released by bacteria or mating pheromone, respectively) might be more important. Although much remains to be learnt about the principles that govern the longevity of the polarized state, actin contributes to the maintenance of cell polarity in a dynamic manner through its endocytic function, whereas microtubules are important for long-lasting polarity in large cells.

Maintaining dynamic cell polarity through endocytosis. Maintaining a concentration of molecules at a specific cortical location is no trivial matter if diffusion can occur. In yeast, it is clear that many polarized cortical proteins can diffuse readily along the membrane and/or away from the membrane, even after cells have achieved an apparently stable polarized concentration of these proteins on the cortex[39,99–101]. As such, a polarized distribution of these proteins must be achieved and maintained in a dynamic manner through the continuous retargeting of 'escaped' molecules. Recycling of membrane-anchored proteins can be accomplished through endocytic recycling, which in yeast occurs at cortical actin patches that are assembled by the Arp2/3 complex and several NPFs[102]. Polymerization of the branched actin network, together with the myosin-I motor, drives the invagination and elongation of endocytic membranes and leads to scission of endocytic vesicles. In mammalian cells, actin polymerization also propels the movement of endosomes[102].

The effectiveness of endocytosis in the polarization of membrane proteins was elegantly shown in an experiment in yeast, in which the plasma membrane SNARE Sso1, which is normally uniformly distributed around the cell, became polarized after the introduction of an endocytic signal at its cytoplasmic domain[100]. Concentration of membrane-bound Cdc42 at the site of polarized growth requires actin[39,103], probably through its endocytic function[37]. A mathematical model that describes the maintenance of membrane-bound Cdc42, based on a mechanism of balancing diffusion with endocytosis and transport, predicted a non-monotonic relationship between the rate of endocytosis and the degree of concentration of membrane proteins[104], which suggests that polarization of membrane-anchored proteins can be sensitively regulated through endocytosis.

The requirement for endocytosis in cell polarity has also been noted in other cell types, such as fission yeast[105], migrating cells[106], asymmetrically dividing stem cells[107], epithelial cells[108–110] and *D. melanogaster* oocytes[111,112]. Surprisingly, an RNAi screen for genes that regulate membrane trafficking in *C. elegans* found that conserved cell polarity regulators, such as CDC-42 and PAR proteins, are in turn required for efficient endocytosis[113]. Similarly, in *D. melanogaster* oocytes, the posterior determinant protein Oskar is an endocytic cargo and also stimulates endocytosis, possibly by regulating actin dynamics[111,112]. Such bidirectional interaction between cortical regulators and endocytosis suggests a potential role for endocytosis in the Turing–Gierer–Meinhardt theory, which is often used to explain cell polarity and gradient sensing[114]. This theory assumes the presence of a slow diffusing autocatalytic activator and an elusive global inhibitor, which is the product of the activator and restricts the spreading of the activator. Endocytosis, an actin-based process that internalizes membrane-bound activators (for example, CDC42 and Oskar) and is itself stimulated by them, could be a long-sought-after 'global inhibitor' that is required for cell polarity. Phosphatase and tensin homologue (PTEN), which dephosphorylates phosphatidylinositol-3,4,5-trisphosphate, is another candidate global inhibitor in *Dictyostelium discoideum* cells that are undergoing chemotaxis[115,116].

Microtubules in T-cell polarity. Whereas microtubule-directed membrane trafficking is clearly a contributing factor to the breaking of symmetry in neurons, a more common role for microtubules is to reinforce the initial polarity that has been established by the actin cytoskeleton, especially in large cells or cells that require long-lived polarity. When T cells interact with targets, they form an immunological synapse that contains clustered signalling molecules and actin filaments, and they reorientate their centrosome towards the synapse (FIG. 4). Centrosome reorientation occurs during the interaction of T cells with antigen-presenting cells, and during the interaction of natural killer (NK) cells and cytotoxic T lymphocytes (CTLs) with their targets, and is required for the efficient lysis of targets bound to NK cells and CTLs[117]. Engagement of the T-cell receptor leads to signals that trigger actin-dependent formation of the immunological synapse, followed rapidly by the reorientation of the centrosome to a position near the synapse. Activation of the Rho GTPase CDC42 seems to have a key role in initiating the reorientation of the centrosome[118]. During reorientation, microtubules interact laterally with the synapse, which results in the active movement of the centrosome towards the synapse[119]. Dynein interacts with adhesion- and degranulation-promoting adaptor protein (ADAP), a scaffold protein that links T-cell receptor signalling to integrin clustering, and both proteins are found in the peripheral synapse. This leads to the possibility that synapse-associated dynein generates forces on microtubules to pull the centrosome towards the synapse[120]. The localization of the centrosome near the immunological synapse positions the Golgi apparatus and lytic granules near the synapse, to ensure directed secretion towards the target cell[121].

Microtubules and polarity in migrating cells. In adherent migrating cells there are at least two sources of asymmetry during microtubule organization: centrosome orientation and the selective stabilization of a subset of microtubules[20] (FIG. 4). Centrosome orientation is the localization of the centrosome to a position between the nucleus and the leading edge, which occurs in diverse types of migrating cells, including macrophages, fibroblasts, endothelial cells, astrocytes and neurons. A common set of signalling factors, including CDC42, PAR6, aPKC and dynein, regulates centrosome orientation[122–124]. However, diverse CDC42 effectors have been implicated in centrosome orientation, including IQ motif-containing GTPase-activating protein-1 (IQGAP1)[125], myotonic dystrophy kinase-related CDC42-binding

Endocytic recycling
A process of internalization of plasma membrane proteins, which are subsequently sorted in endosomes and either directed to lysosomes for destruction or recycled back to specific locations on the plasma membrane.

SNARE
(Soluble *N*-ethyl-maleimide-sensitive fusion protein-attachment-protein receptor). A family of membrane-tethered, coiled-coil proteins that regulate fusion reactions and target specificity in exocytosis and other membrane trafficking events.

kinase (MRCK)[126] and mammalian formin diaphanous-1 (mDia1)[127], suggesting that the precise mechanism might depend on the cell type or the use of distinct downstream pathways. This diversity of effectors in centrosome orientation might also reflect the coordinated requirement for both microtubules and actin filaments, as shown by studies in fibroblasts[126]. In these cells, the centrosome does not move to its position in front of the nucleus, but instead the nucleus moves away from the leading edge in an actin- and myosin-dependent fashion. The centrosome is maintained at the centre by dynein, which is thought to act on microtubules at peripheral sites[126].

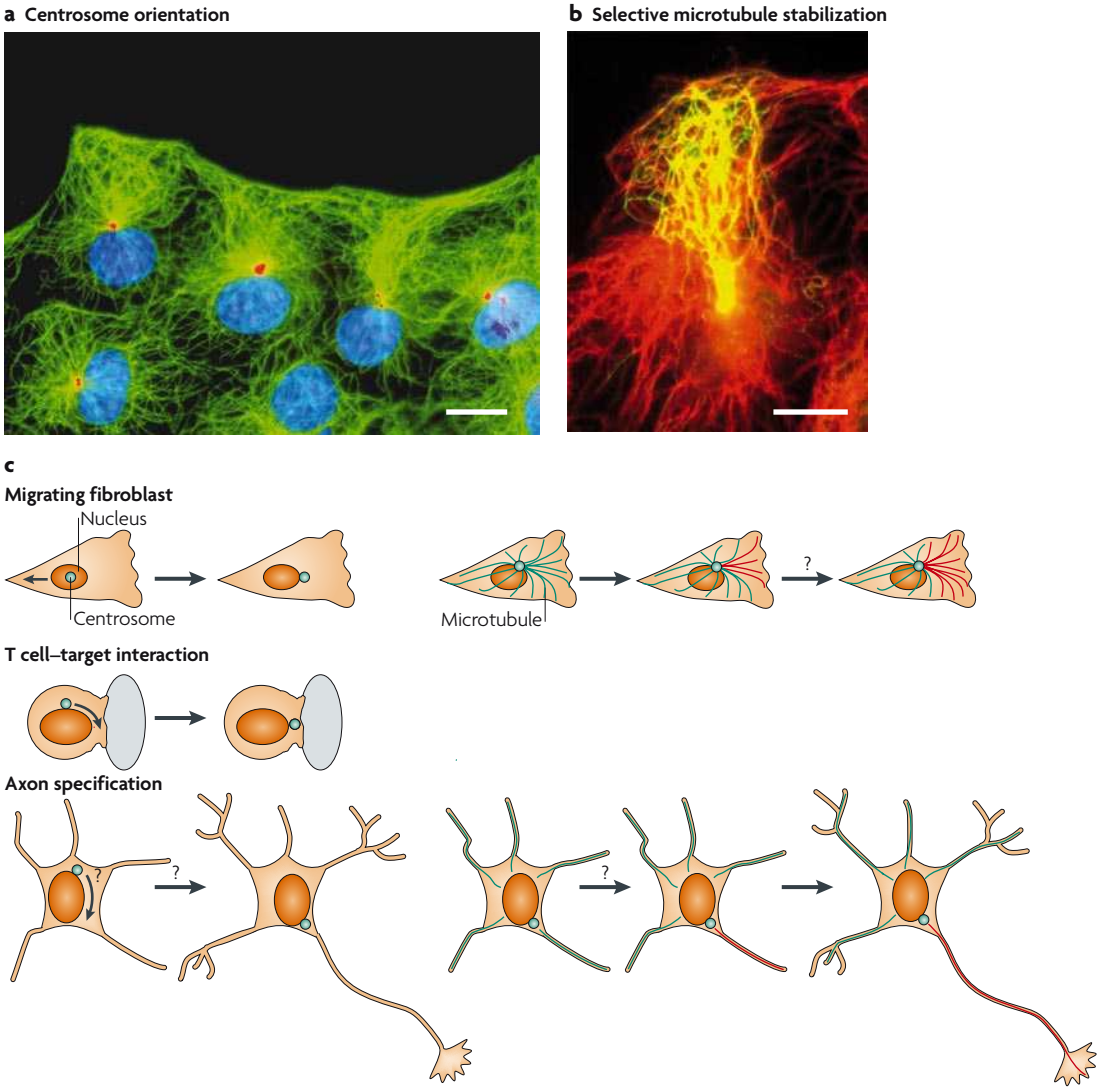


Figure 4 | **Polarization of microtubule arrays in migrating fibroblasts, T cells and differentiating neurons.** **a** | An immunofluorescence image showing centrosome orientation in NIH3T3 fibroblasts that are migrating into an *in vitro* wound. The centrosomal marker pericentrin is shown in red, microtubules are shown in green and 4′,6-diamidino-2-phenylindole (DAPI)-stained nuclei are shown in blue. **b** | Immunofluorescence image showing the polarized formation of stabilized microtubules in an NIH3T3 fibroblast that is migrating into an *in vitro* wound. Tyrosinated tubulin, a marker of bulk and dynamic microtubules, is shown in red, and detyrosinated tubulin, a post-translationally modified form of tubulin that accumulates in stable microtubules, is shown in yellow. Note that only a subset of the microtubule array is stabilized and that these stable microtubules are preferentially directed towards the leading edge. **c** | The diagrams show that centrosomes orientate towards the leading edge in migrating cells, towards sites of target interaction in T cells and possibly towards the presumptive axon in neurons. Note that the nucleus moves to orientate the centrosome in fibroblasts, whereas the centrosome moves to become orientated in T cells. It is not known whether the centrosome moves towards the presumptive site of the axon in neurons. The diagrams also show the selective formation of stabilized microtubules in the leading edge of migrating cells and in the presumptive axon of neurons (stable microtubules are shown in red and dynamic microtubules are shown in green). In fibroblasts, stable microtubules initially form *de novo* in the leading edge, whereas it is unclear how stabilization arises in one of the undifferentiated neurites in neurons. Neurons are known to possess a self-reinforcing mechanism to continually generate new stable microtubules as the axon grows. It is not known if fibroblasts share such a self-reinforcing mechanism. Scale bars, 10 μm. Images in parts **a** and **b** courtesy of A. F. Palazzo, Harvard University, Massachusetts, USA, and G.G.G.

Microtubule arrays in migrating cells are also polarized by the formation of a subset of persistent stable microtubules near the leading edge that accumulate post-translational modifications of tubulin[128]. Centrosome polarization and the selective stabilization of microtubules occur concurrently in fibroblasts, although each process is independently regulated[123]. In fibroblasts, embryonic endodermal cells and glial cells, the formation of the stabilized microtubules is regulated by Rho GTPase and its effector mDia1 (REFS 127,129–131). The Rho and mDia1 pathway generates stabilized microtubules in fibroblasts by capping microtubule plus ends, as shown by the lack of tubulin subunit addition or loss from the ends of the stabilized microtubules[131,132]. mDia1 is likely to be a key player in the capping of microtubules, as the related protein, mDia2, directly stabilizes microtubule ends *in vitro*[133]. The restricted formation of stable microtubules near the leading edge is due to integrin engagement and focal adhesion kinase signalling, which limit the ability of active Rho to activate mDia1 only near the leading edge[134]. A number of +TIPs have been implicated in Rho–mDia1-mediated microtubule stabilization, including EB1 and APC in fibroblasts and actin crosslinking family protein-7 (ACF7; also known as microtubule–actin crosslinking factor (MACF)) in endodermal cells[130,135]. Another type of +TIP, the cytoplasmic linker protein (CLIP)-associating proteins (CLASPs), also contributes to the formation of stabilized microtubules in fibroblasts[136]. CLASPs interact with the membrane protein LL5β, and this contributes to microtubule capture at cortical sites[137]. CLASPs seem to be regulated by Rac, GSK3β and phosphoinositide 3-kinase (PI3K), rather than by Rho[136,138]. Additional studies are needed to determine whether the diversity of +TIPs involved in microtubule stabilization reflects a single, but complex mechanism, or diverse mechanisms, each of which can be activated to achieve microtubule stabilization.

Centrosome polarization and microtubule stabilization are thought to contribute to polarized delivery of membrane precursors from biosynthetic and endocytic recycling pathways. The Golgi apparatus and endocytic recycling compartment are localized near the centrosome and become polarized towards the leading edge as a result of centrosome polarization. The polarized orientations of these organelles might direct the delivery of membrane vesicles to the leading edge through kinesin-dependent transport. As described above, certain kinesins might preferentially use stable, post-translationally modified microtubules, and this would also direct delivery to the leading edge. Model biosynthetic cargoes, such as the viral VSVG protein, preferentially undergo exocytosis near the leading edge and initially accumulate at the leading-edge membrane before rapidly diffusing[139,140]. Biased exocytosis and accumulation near the leading edge are randomized by treatments that break down microtubules.

Microtubules in the maintenance of epithelial polarity. Following initiation of epithelial polarity, microtubules undergo a dramatic rearrangement from a radial centrosomal array to a non-centrosomal array. In columnar epithelia, non-centrosomal microtubules become aligned along the apical–basal axis, predominantly with minus ends at the apical pole and plus ends at the basal pole[141]. There are also arrays of short microtubules of mixed polarity at both apical and basal surfaces. Formation of epithelial (E)-cadherin adherens junctions triggers the formation of non-centrosomal microtubules and an overall increase in epithelial microtubule stability[142]. A recent study in *D. melanogaster* embryos showed that aPKC is involved in generating the non-centrosomal arrays in epithelia, by allowing the release of microtubules from the centrosome[143]. EB1 and APC contribute to the basal microtubule network, but little else is known about how these arrays are formed and maintained[144].

The basolateral and apical membrane domains of epithelial cells exhibit a distinct composition of proteins, and studies with viral membrane proteins have clearly shown that proteins destined for these membranes take distinct routes during intracellular targeting[145]. There is strong evidence that proteins bound for the apical domain are trafficked by microtubules. Nocodazole breakdown of microtubules leads to nonpolarized accumulation of apical proteins in both apical and basolateral domains. This might reflect the redistribution of the normally apical SNARE protein syntaxin-4 to basolateral sites in the absence of microtubules[146]. In addition, several kinesins have been shown to be specifically involved in the trafficking of apical proteins from the Golgi to the apical membrane[147,148].

In contrast to apical proteins, microtubules do not seem to be required for delivery of basolateral proteins. This result is surprising given the highly polarized array of microtubules that extends from the apical region in which the Golgi is localized, towards the basal membrane of epithelial cells. It has been suggested that the normal basolateral route might be overridden by nocodazole treatment, which disperses the apically situated Golgi to sites near the basolateral membrane[145]. If kinesins can be implicated in basolateral trafficking, it will reinvigorate the idea that there is a microtubule-dependent route to the basolateral membrane.

Microtubule–actin filament crosstalk

For microtubules to build and stabilize the polarity that is initiated by actin, the two cytoskeletal systems must be coordinated through crosstalk. This crosstalk occurs in both directions, with actin contributing to the initial polarization of microtubule arrays and with the polarized microtubule arrays reinforcing the initial asymmetry in the actin cytoskeleton. As noted above, a molecular mechanism for the microtubule-dependent delivery of actin regulatory factors has been established in *S. pombe*. Below we discuss studies in migrating cells in which there is abundant bidirectional crosstalk between actin and microtubules.

Early studies on fibroblasts showed that breakdown of microtubules with microtubule antagonists collapsed the front–back polarity of migrating cells[149,150], and more recent studies have confirmed this result for rapidly moving neutrophils undergoing chemotaxis[151]. Two mechanisms have been invoked to explain the contribution of microtubules to front–back polarity. In neutrophils,

Focal adhesion
A plaque-like cellular structure that links the extracellular matrix on the outside of the cell to the actin cytoskeleton inside the cell through integrin receptors and associated proteins.

Adherens junction
An adhesive structure that connects adjacent cells through cadherins and other membrane proteins and is associated with cortical actin filaments.

microtubule disruption causes the Rho-mediated contractility in the tail to spread, abrogating the ability of the cell to establish protrusive activity at its leading edge[151]. The molecular basis for this might involve the ability of microtubules to regulate Rho GTPases, which control the protrusive actin in the front and the contractile actin in the back (see above). Microtubule disruption by nocodazole or colchicine activates Rho globally, with a concomitant increase in contractility[152,153]. Microtubules bind to one of the Rho guanine nucleotide-exchange factors (GEFs), RhoGEF H1, that are involved in activating Rho. Following breakdown of microtubules, the exchange activity of RhoGEF H1 is increased, which leads to increased levels of active Rho[154]. A greater understanding is required of the endogenous factors that release RhoGEF H1 from microtubules and allow its activation.

Microtubules can also affect actin in migrating cells by acting on focal adhesions. Focal adhesions are sites of clustered integrins and associated molecules that are linked to actin stress fibres and provide traction for cell migration. Imaging studies have shown that dynamic microtubules contact focal adhesions and, in doing so, trigger the disassembly of focal adhesions and their associated actin stress fibres[155,156]. The disassembly process that is triggered by microtubules involves endocytosis of integrins. The canonical endocytic protein dynamin is clustered at disassembling focal adhesions and is required for microtubule-induced focal adhesion disassembly[157]. This result suggests that microtubules might deliver factors that trigger the endocytosis of focal adhesion components. As the disassembly of mature focal adhesions occurs primarily in the middle and tail of migrating cells, this contributes to a polarization of focal adhesions with assembly predominant in the front and disassembly in the rear. The targeting of focal adhesions is an example of the contribution of dynamic microtubules to cell polarization and might explain the observation that low concentrations of microtubule antagonists that dampen microtubule dynamics reduce the speed of migrating cells[158].

The actin cytoskeleton contributes to the assembly of polarized microtubule arrays in migrating cells in a number of ways. As mentioned above, polarization of the centrosome in fibroblasts involves the collaboration of actin-dependent movement of the nucleus and microtubule-dependent centring of the centrosome. Another example is the targeting of focal adhesions by microtubules, although, unlike centrosome polarization, the effects of this are local rather than global. Although microtubules usually grow straight owing to constraints imposed by the microtubule lattice, imaging studies have shown that microtubules bend towards focal adhesions when in their vicinity[156]. Stress fibres are likely to guide the microtubule to the focal adhesion, although it is not clear whether this involves a directed molecular mechanism or a physical one. A type V myosin (myosin-Va) can interact with the +TIP EB1 through the adaptor protein melanophilin, and this might guide microtubules to the barbed ends of actin filaments that are associated with focal adhesion components[159]. Indeed, earlier work in budding yeast showed that a similar mechanism, which involves the EB1 orthologue (Bim1) and myosin-V (Myo2), directed microtubules towards the bud to move the nucleus and orientate the spindle along the mother–bud axis[160]. As actin arrays are generated at the membrane during many symmetry-breaking processes, this might be a widespread crosstalk mechanism that enhances microtubule capture during cell polarization.

Concluding remarks

The examples discussed above include cells of various sizes and polarized cell states that last from minutes (for example, yeast and neutrophils) to days or even years (for example, epithelial cells and neurons). These different cell types rely on actin and microtubules to polarize to different degrees and in different ways, but some general themes have begun to emerge. Actin seems to have a key role in the initial symmetry-breaking process and enables a rapid response to stimuli in cells of all sizes, with the possible exception of neurons, in which symmetry breaking is driven by a microtubule-based mechanism. This role might stem from the versatility in the assembly and organization of actin filaments, especially at the cell cortex, and from the many ways by which actin mediates the transport of cytosolic and cortical components. Microtubules, however, build on and stabilize the initial asymmetry that is created by actin-based forces. This function requires transport along microtubules that can be orientated by positioning the centrosome and/or by stabilizing and adding post-translational modifications to a subset of microtubules. The contributions of microtubules to cell polarization seem to increase with cell size and with the longevity of the polarized state. Some of these generalizations might not last, however, as the field has just begun to 'break' the secretive shell of cell polarity.

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Acknowledgements

This work is supported by National Institutes of Health grants to R.L. and G.G.G. The authors apologize for not being able to discuss all relevant and important data in this review owing to space limitations.

DATABASES

UniProtKB: http://www.uniprot.org
APC | aPKC | Bni1 | Bnr1 | Cdc42 | cofilin | For3 | myosin-II | myosin-VI | PAR-3 | PAR-6 | RhoA | tau | Tea1 | Tea4 |

FURTHER INFORMATION

Rong Li's homepage: http://www.stowers-institute.org/labs/RongLiLab.asp

Gregg Gundersen's homepage: http://www.cumc.columbia.edu/dept/gsas/anatomy/Faculty/Gundersen

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