Generating and Exploiting Polarity in Bacteria

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Bacteria are often highly polarized, exhibiting specialized structures at or near the ends of the cell. Among such structures are actin-organizing centers, which mediate the movement of certain pathogenic bacteria within the cytoplasm of an animal host cell; organized arrays of membrane receptors, which govern chemosensory behavior in swimming bacteria; and asymmetrically positioned septa, which generate specialized progeny in differentiating bacteria. This polarization is orchestrated by complex and dynamic changes in the subcellular localization of signal transduction and cytoskeleton proteins as well as of specific regions of the chromosome. Recent work has provided information on how dynamic subcellular localization occurs and how it is exploited by the bacterial cell.

The main task of a bacterial cell is to survive and duplicate itself. The bacterium must replicate its genetic material and divide at the correct site in the cell and at the correct time in the cell cycle with high precision. Each kind of bacterium also executes its own strategy to find nutrients in its habitat and to cope with conditions of stress from its environment. This involves moving toward food, adapting to environmental extremes, and, in many cases, entering and exploiting a eukaryotic host. These activities often involve processes that take place at or near the poles of the cell. Here we explore some of the schemes bacteria use to orchestrate dynamic changes at their poles and how these polar events execute cellular functions.

In spite of their small size, bacteria have a remarkably complex internal organization and external architecture. Bacterial cells are inherently asymmetric, some more obviously so than others. The most easily recognized asymmetries involve surface structures, e.g., flagella, pili, and stalks that are preferentially assembled at one pole by many bacteria. "New" poles generated at the cell division plane differ from old poles from the previous round of cell division. Even in *Escherichia coli*, which is generally thought to be symmetrical, old poles are more static than new poles with respect to cell wall assembly (*1*), and they differ in the deposition of phospholipid domains (*2*). There are many instances of differential polar functions; among these is the preferential use of old poles when attaching to host cells as in the interaction of *Bradyrhizobium* with plant root hairs (*3*) or the polar pili-mediated attachment of the *Pseudomonas aeruginosa* pathogen to tracheal epithelia (*4*). An unusual polar organelle that mediates directed motility on solid surfaces is found in the nonpathogenic bacterium *Myxococcus xanthus*. The gliding motility of this bacterium is propelled by a nozzle-like structure that squirts a polysaccharide-containing slime from the pole of the cell (*5*). Interestingly, *M. xanthus*, which has nozzles at both poles, can reverse direction by closing one nozzle and opening the other in response to end-to-end interactions between cells.

The Role of Bacterial Cell Polarity in Pathogen-Host Interactions

Unique uses of the cell pole are central to the remarkable tactics that some intracellular pathogens use to co-opt the actin machinery of mammalian host cells. For example, the *Shigella flexneri* IcsA protein and the *Listeria monocytogenes* ActA protein, each located at an old cell pole (*6*, *7*), nucleate host actin filaments to form a comet-like actin tail that propels the invading bacterium forward to penetrate a neighboring cell. Unipolar localization of these bacterial proteins enables unidirectional actin-based motility.

The manner by which these two actin-nucleating proteins reach and are maintained at the bacterial cell pole differs in *Shigella* and *Listeria*. In the case of *Shigella*, IcsA is directly targeted to the membrane of the old pole (*8*). A segment of IcsA within the NH_2-terminal region is believed to make initial contact with a polar target and a short segment near the COOH-terminus maintains this polar interaction. Once IcsA recognizes and attaches to the old pole, it diffuses laterally toward the new pole. To sharpen the IcsA gradient and maintain its polar position, the cell uses an IcsA-specific protease (*9*, *10*). This protease is distributed uniformly around the cell surface and its action creates a polar gradient of IcsA that results in a strong IcsA polar bias (Fig. 1A). Using an apparently different mechanism, the ActA protein initially localizes to both old poles of the dividing *Listeria* cell and then diffuses toward, but not across, the division plane, resulting in newly divided cells that are enriched for ActA at the old poles (*11*).

Dynamically Localized Signal Transduction Proteins and the Maintenance of Asymmetry

Two-component signal transduction proteins are a principal signaling system in bacteria (*12*). In these systems, the sensor domain of the histidine kinase component responds to an internal or external signal by catalyzing phosphotransfer from ATP to an internal histidine residue. This phosphate is transferred, in turn, from the phosphorylated histidine to an aspartate on the second component, a response regulator. Once activated by phosphorylation, the response regulator interacts with a DNA or protein target, or transfers the phosphate to downstream components of a phosphorelay. Many such two-component signaling proteins are dynamically localized to cell poles (*13*, *14*). In *Caulobacter crescentus*, these localized signaling proteins are used to coordinate

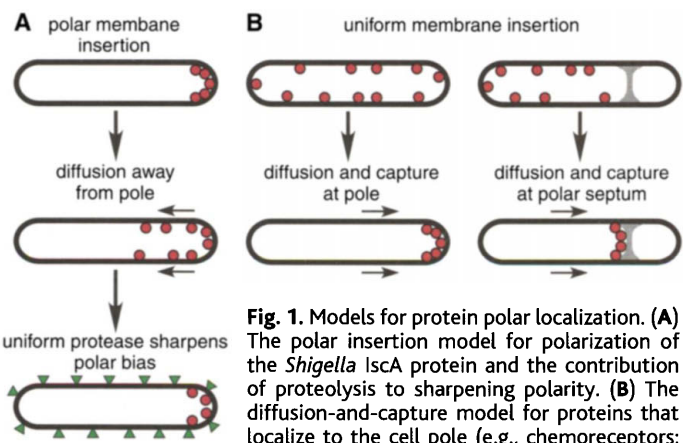

Fig. 1. Models for protein polar localization. (**A**) The polar insertion model for polarization of the *Shigella* IscA protein and the contribution of proteolysis to sharpening polarity. (**B**) The diffusion-and-capture model for proteins that localize to the cell pole (e.g., chemoreceptors; left-hand diagrams) or to the polar septum of sporulating *B. subtilis* (SpoIVFB; right-hand diagrams).

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cell-cycle progression with polar differentiation (*14*).

Caulobacter exploits polar asymmetry to generate dissimilar progeny at each division: a swarmer cell that uses a single polar flagellum and associated chemosensory apparatus to propel the cell toward food and a stalked cell that uses a polar stalk to adhere to a source of food. After a period of motility, the swarmer cell releases the flagellum and grows a stalk at the same pole to continue the cycle. The flagellum, the chemotaxis apparatus, and the pili secretion mechanism are built de novo at one pole of the predivisional cell (Fig. 2A), governed by localized two-component signal transduction proteins (Fig. 2B). For example, the polar localization of the PleC histidine kinase is critical for polar pili biogenesis (*15*).

Asymmetry is maintained in *Caulobacter* by the temporally controlled localization of structural and regulatory proteins to the cell pole and the subsequent proteolysis and release of these polar components in preparation for the next cell cycle. The polar proteins are cleared from the swarmer cell pole as part of its differentiation into a stalked cell, yielding a cell that lacks polar structures opposite the stalk pole (*14*) (Fig. 2A). Removal of these polar proteins involves temporally controlled proteolysis that releases the flagellum, clears the cell of chemoreceptors, and releases pili secretion proteins that then diffuse away from the pole. The polar PleC histidine kinase is required both for its own release and for the removal of the pili secretion proteins from the pole during the swarmer-to-stalked cell transition (*16*). At the same time, another two-component signaling protein, the response regulator PleD, controls flagellar basal body proteolysis (*17*). Thus, different signaling proteins mediate these two critical proteolytic events.

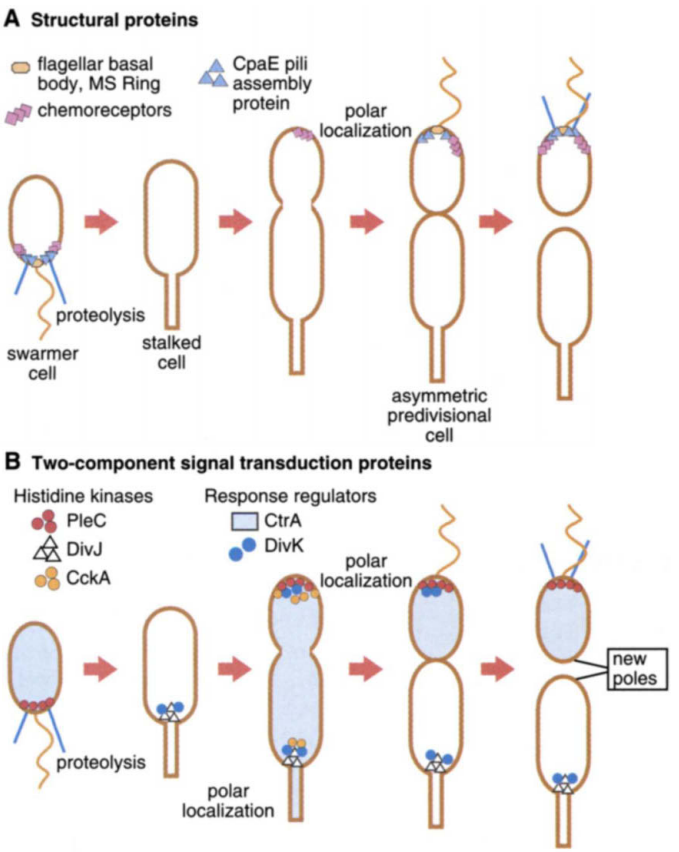

Fig. 2. Dynamic polar localization of two-component signal transduction and structural proteins during the *Caulobacter* cell cycle. (**A**) The asymmetric localization of the flagella basal body MS ring protein, the chemoreceptors, and the CpaE pili assembly protein to the incipient swarmer pole of the predivisional cell and their proteolysis at the subsequent swarmer to stalked cell transition. (**B**) First, the PleC histidine kinase localizes to the incipient swarmer pole, then the CckA histidine kinase transiently localizes to the cell poles coincident with phosphorylation of the CtrA response regulator, and the DivJ histidine kinase takes up the polar position vacated by PleC at the swarmer-to-stalked cell transition where it remains at the base of the stalk. The DivK single-domain response regulator is delocalized in swarmer cells, but localizes to the cell poles at other times in the cell cycle. CtrA is cleared from the stalked cell by temporally and spatially regulated proteolysis to allow the initiation of DNA replication.

Another two-component signaling protein, the histidine kinase DivJ, moves in and occupies the cell pole as it is vacated by PleC when the swarmer cell differentiates into a stalked cell (Fig. 2B). PleC release from the pole is essential for the subsequent occupancy by DivJ. Perhaps a multicomponent polar complex at this pole provides the docking station for signaling proteins sequestered and released at consecutive times in the cell cycle. In support of this idea, the response regulator DivK, which is essential for cell-cycle progression (*18*), is distributed throughout the cytoplasm in swarmer cells, and its subsequent localization at the stalked cell pole depends on the presence of the DivJ histidine kinase at that pole (*19*).

Essential two-component signaling proteins, the histidine kinase CckA and its cognate response regulator CtrA, are critical for progress through the *Caulobacter* cell cycle and are governed by polar positioning (*18*). CtrA, which is present in swarmer cells and predivisional cells, but not stalked cells (Fig. 2B) controls the expression of about 25% of the 550 genes that are temporally controlled during the *Caulobacter* cell cycle (*20*). CtrA~P also transiently binds to and silences the origin of DNA replication to prevent premature reinitiation of replication; initiation of chromosome replication requires clearance of CtrA from stalked cells by targeted proteolysis (*21*). The CckA histidine kinase is transiently localized to the cell pole coincident with the synthesis of CtrA (Fig. 2B). Both autophosphorylation of CckA and activation of CtrA by phosphorylation occur at the time when CckA is localized to the cell poles. Thus, signaling from proteins captured at the cell poles coupled to cell cycle–controlled proteolysis regulates cell-cycle progression and asymmetry in this organism.

Polar Clustering of Chemotaxis Proteins

Chemoreceptor complexes are localized predominantly at the cell poles (*22*, *23*), but the reason for this polar clustering has been mysterious. Studies of the chemotaxis machinery in *E. coli* have shown that chemoreceptors are organized into large membrane-associated, two-dimensional arrays and that formation of these arrays depends on the presence of the cytoplasmic histidine kinase CheA and the coupling protein CheW. Each of the chemoreceptors assembles into a two-dimensional lattice containing hundreds or thousands of receptors, which are held together by bridging connections to CheA and CheW (*24*). The receptors form dimers, and these dimers form trimers, yielding "trimers of dimers" that can be of mixed receptor composition (*24*, *25*). These mixed-receptor trimers bind CheA and CheW to form "signaling teams" that collaborate to activate the shared CheA molecule. Signaling interactions between the receptor teams may play an important role as well (*24*). These mechanisms of communication within and between receptor signaling teams may be the key to the high detection sensitivity of bacterial chemoreceptor networks (*24*).

The mechanism that causes the chemoreceptors to congregate in clusters at the cell poles is not known. In this regard, the pattern of localization of chemoreceptor clusters observed in filamentous cells of *E. coli* is intriguing (*26*). When cell division in *E. coli* is blocked, the cells grow into motile filaments that are as much as 50 times the length of normal cells. Immunofluorescence microscopy shows that the chemoreceptors in the filaments are located in clusters both at the poles and at irregular intervals along the fil-

ament (*26*). The mechanism driving the placement of these clusters along the elongated cells is unknown, but their existence suggests that the polar location in normal cells is not the result of a membrane property unique to the ends of the cell.

Polar Oscillations and Generation of a Medial Division Site

Dividing cells face the fundamental challenges of restricting cytokinesis to a specified position within the cell and initiating division at the appropriate time in the cell cycle. The division site in *E. coli* is determined by the tubulin-like GTPase FtsZ, which polymerizes into a ring-like structure along the inside surface of the membrane across the short axis of the cell (*27*). This "Z ring" is assembled at the cell center between the daughter chromosomes after DNA replication and chromosome segregation have taken place. Early studies showed that when a mechanism that restricts the Z ring to the cell middle is made inoperative, anomalous divisions occur near the cell poles, indicating that the cell poles are capable of supporting cytokinesis unless specifically prevented from doing so (*28*). The mechanism that prevents Z ring formation at the poles involves a complex (MinCD) of the FtsZ polymerization inhibitor MinC and the ATPase MinD, as well as a topological specificity determinant MinE.

The *E. coli* Min proteins exhibit remarkable dynamism (*29*). MinCD forms a membrane-associated polar cap at one end of the cell with MinE then forming a band at the mid-cell (Fig. 3A). As this MinE band moves toward the pole, MinCD is ejected from the membrane. The ejected MinCD proteins regroup at the opposite pole and MinE forms another band at the mid-cell to once again move to the pole (this time the opposite pole) and eject the MinCD cap anew. The period of the entire cycle is a few tens of seconds so that multiple oscillations can occur during each round of cell division. The formation of polar zones requires the ATP-dependent association of MinD with the membrane (Fig. 3B). MinE stimulates the ATPase activity of MinD, thereby dislodging MinD and MinC from the membrane in such a way as to ensure oscillation of MinCD between the halves of the cell (*30*). The net effect of this oscillation is to increase the average concentration of MinE at mid-cell and the average concentration of the membrane-associated and Z ring–inhibiting MinCD complex at all membrane sites other than at mid-cell (*31*).

Although all the reaction mechanisms in the Min system are not fully characterized, dynamic reaction-diffusion models based on the known mechanisms and observed phenomenology can reproduce the behavior of the Min proteins, and they support several general conclusions (*29*, *32*): (i) Active transport of the Min proteins is not required, because diffusion of the free molecules through the cytoplasm is fast enough to support the observed movements. (ii) The oscillations are initiated spontaneously and are robust over a broad range of parameters and cell lengths representative of the normal cell. (iii) Periodic FtsZ bands are predicted for long cells, as observed in filamentous cells. Also, the rod-shaped geometry of the *E. coli* cell appears to be important to the dynamics of system. Indeed, a strictly one-dimensional reaction diffusion model captures the behavior quite well (*32*).

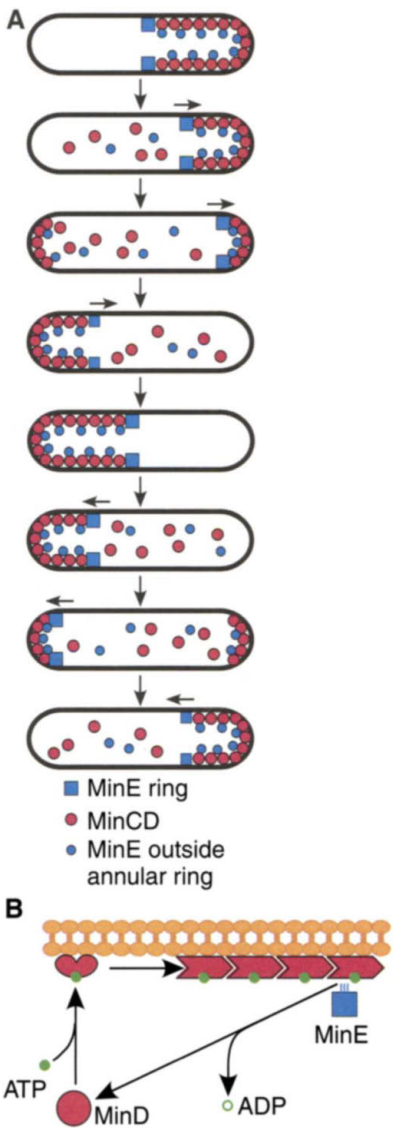

Fig. 3. Polar oscillation of MinCD coupled to dynamic localization of the MinE ring. (**A**) The assembly and disassembly of the MinCD complex around the poles of the cell, bounded by the MinE ring at the rim of the MinCD cap. As the MinCD cap disassembles, the MinE ring moves toward one pole. A new MinCD cap then assembles at the opposite pole, accompanied by the assembly of a new MinE ring at its rim (*29*). (**B**) The dynamic interaction of MinD-ATP with the membrane. When polymerized MinD interacts with MinE, ATP is hydrolyzed and MinD is released from the membrane (*31*).

In *E. coli*, localization of the division plane deviates from mid-plane by only a few percent (*33*). It is remarkable that a yardstick based on concentration gradients can produce this precision. It is important that the yardstick is entirely intrinsic to the properties of the Min proteins and does not seem to depend on any metric cue provided by the cell at its poles or elsewhere.

Not surprisingly, other mechanisms appear to reinforce the mid-cell positioning of the Z ring. During DNA replication, newly duplicated daughter chromosomes separate from each other, forming two large masses known as nucleoids with a DNA-free zone in between. The nucleoid appears to interfere with Z-ring assembly (*34*), perhaps by close contact with the inside surface of the cytoplasmic membrane, where the nucleation of FtsZ polymerization is believed to take place. Polymerization is favored in the cell middle, where chromosome segregation has created a space between the DNA masses. Thus, dynamic protein and nucleic acid determinants work together to ensure that the cell finds its precise mid-point when it undergoes binary fission.

Polar Division in *Bacillus subtilis*

As we have seen, bacteria localize many proteins to an extreme terminus of the cell. Another mode of polarization is asymmetric division, which occurs during spore formation by *B. subtilis*. Growing cells of *B. subtilis* divide by binary fission, but cells that have begun to sporulate produce a septum at a polar position. In this case, the developing cell (the sporangium) divides into dissimilar sized compartments called the forespore (the smaller compartment) and the mother cell.

How does the switch to polar division occur, and what is its biological significance? As in *E. coli*, Z-ring formation in growing cells of *B. subtilis* is restricted to the cell center by the action of the division inhibitor MinCD (*35*). During sporulation, polar division commences with the formation of a medially sited Z ring, but instead of undergoing cytokinesis the medial ring unfurls into a helical structure that grows toward both poles [*36*, (Fig. 4)]. The helix seems to redeploy FtsZ from the center to both ends of the cell, and it is eventually replaced by two polar Z rings (*36*, *37*). This remodeling into a helix and finally into polar Z rings is under developmental control mediated by the FtsZ-interacting protein SpoIIE and a sporulation-specific increase in FtsZ levels (*36*). Both polar Z rings have the potential to form a septum, but cytokinesis is restricted to one pole because the first septum to form sets in motion events that block septum formation at the

distal pole (*38*, *39*). Thus, the formation of a septum at one pole locks in asymmetry by triggering events that prevent a second round of septation.

Once the polar septum is formed, the sporangium has to differentially activate programs of gene transcription in the forespore and the mother cell. Gene expression in the forespore is governed by the transcription factor σ^F whose activation in a cell-specific manner is the consequence of two pathways (*40*). One pathway involves SpoIIE, which (in addition to its role in the formation of the FtsZ helix) incorporates into the polar septum during cytokinesis (*41*, *42*) and contributes to the selective activation of σ^F in the forespore (*43*–*45*). The second pathway explicitly exploits the inherent polarity of the chromosome (*46*–*48*). Before asymmetric division, *B. subtilis* contains two chromosomes, each oriented with its single origin of replication at an extreme pole of the cell (*49*) (see below). Initially, when the polar septum is formed, only the region near the origin is trapped in the forespore; the remaining two-thirds of the chromosome remain in the mother cell (Fig. 4). A DNA translocase in the septum pumps the origin-distal portion of the chromosome into the forespore (*49*). Thus, the forespore briefly lacks a full complement of genes. One of these genes located near the terminus of the chromosome encodes the anti-sigma factor SpoIIAB, an unstable inhibitor of σ^F. Therefore, when the forespore is formed it initially lacks the *spoIIAB* gene, whereas the mother cell has two copies. The resulting delay in bringing the anti-σ^F factor gene into the forespore (and hence the delay in replenishing the unstable inhibitor) permits σ^F activation in the forespore (*40*).

The sporangium also exploits the polar septum as a beacon to attract proteins that orchestrate subsequent morphogenesis. After division, the membrane on the mother-cell face of the septum migrates around and fully engulfs the forespore (Fig. 4). Proteins, such as SpoVM, are carried along on the membrane during engulfment, first localizing to the polar septum and then traveling with the mother-cell membrane as it envelops the forespore (*50*). Mutants that have a uniform distribution of SpoVM in the mother cell are impaired in sporulation, which suggests that proper sporulation function is dependent upon proper localization.

Localization by Diffusion and Capture

One possible means for membrane proteins to reach the polar septum is by direct insertion into the septal membrane by a dedicated secretory apparatus. Alternatively, they could insert anywhere in the membrane and reach their ultimate destination by diffusion. A revealing example is the sporulation membrane protein SpoIVFB. As in the case of SpoVM, SpoIVFB is normally first seen at the polar septum. It then migrates with the septal membrane during engulfment. Experiments in which the timing of SpoIVFB synthesis was altered indicate that SpoIVFB actually initially localizes to the cytoplasmic membrane and then rapidly reaches the polar septum by diffusion, where it is captured (*51*) (Fig. 1). *Caulobacter* chemoreceptors reach the cell pole by a similar diffusion and capture mechanism (*52*).

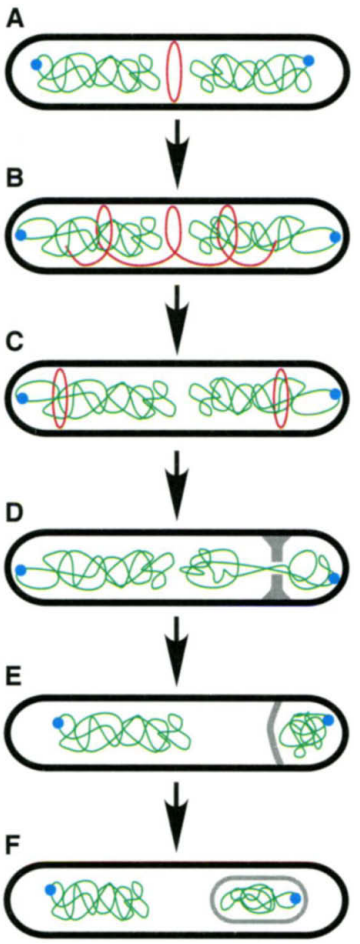

Fig. 4. Polarization during sporulation. (**A** to **C**) The origin regions (blue) of the two chromosomes (green) localize to extreme opposite poles while the medial Z ring (red) is redeployed into two polar rings via a helical intermediate. (**D**) One of the polar Z rings is converted into a polar septum (gray), creating forespore and mother cell compartments and trapping the origin-proximal region of a chromosome in the forespore. The remainder of the chromosome is pumped across the septum. (**E** and **F**). The septal membranes migrate around the forespore, engulfing it within the mother cell.

Polar Segregation of the Origins of DNA Replication

The pole can be the target for chromosome regions as well as for proteins. Visualizing specific sites on the chromosome shows that the origins of replication (each bacterial chromosome has a single bidirectional origin) rapidly move apart toward opposite cell poles after replication starts (*53*). Usually origin regions migrate only partway to the poles, but there are two cases in which origin regions reach the extreme ends of the cell. Before initiation of replication, the origin is localized to the stalked pole of the *Caulobacter* stalked cell (*54*). Then, during DNA replication, one of the two origin regions moves to the extreme opposite pole of the cell. The other case is sporulation in *B. subtilis* when newly duplicated origin regions migrate to extreme opposite poles of the sporangium (*55*, *56*).

Summary

Bacteria exploit polar positioning for many essential functions. To survive in dilute aquatic environments, *Caulobacter* uses polarity to generate an asymmetric predivisional cell, which upon division yields a daughter cell that swims away to find food. Bacterial pathogens such as *Shigella* and *Listeria* use proteins located at one cell pole to nucleate an actin tail to propel it through an invaded host cell. Forward motion in liquid or solid surfaces is facilitated by polar organelles such as flagella or pili. Recently discovered polar nozzles secrete slime to allow bacteria such as *Myxococcus* to glide along a surface. To survive under harsh conditions, *B. subtilis* produces a spore near one cell pole after an asymmetric cell division. To carry out these specialized functions, bacterial cells exhibit a high level of spatial organization with complex structures placed at or near the poles or at midcell exactly when needed. Regulation of gene expression is coupled to progressive changes at the cell poles, and spatial deployment of the chromosome itself is coupled to these regulatory mechanisms. Orchestrating these changes are dynamic mechanisms involving as-yet-unidentified polar cues and/or self-contained, reaction-diffusion systems that oscillate from pole-to-pole.

References and Notes

1. M. A. de Pedro, J. C. Quintela, J. V. Holtje, H. Schwarz, *J. Bacteriol.* **179**, 2823 (1997).
2. E. Mileykovskaya, W. Dowhan, *J. Bacteriol.* **182**, 1172 (2000).
3. J. T. Loh, S. C. Ho, A. W. de Feijter, J. L. Wang, M. Schindler, *Proc. Natl. Acad. Sci. U.S.A* **90**, 3033 (1993).
4. D. E. Zoutman *et al.*, *Scanning Microsc.* **5**, 109 (1991).
5. C. Wolgemuth, E. Hoiczyk, D. Kaiser, G. Oster, *Curr. Biol.* **12**, 369 (2002).
6. M. B. Goldberg, O. Barzu, C. Parsot, P. J. Sansonetti, *J. Bacteriol.* **175**, 2189 (1993).
7. G. A. Smith, D. A. Portnoy, J. A. Theriot, *Mol. Microbiol.* **17**, 945 (1995).
8. M. Charles, M. Perez, J. H. Kobil, M. B. Goldberg, *Proc. Natl. Acad. Sci. U.S.A.* **98**, 9871 (2001).
9. K. D. Shere, S. Sallustio, A. Manessis, T. G. D'Aversa, M. B. Goldberg, *Mol. Microbiol.* **25**, 451 (1997).
10. C. Egile, H. d'Hauteville, C. Parsot, P. J. Sansonetti, *Mol. Microbiol.* **23**, 1063 (1997).
11. C. Kocks, R. Hellio, P. Gounon, H. Ohayon, P. Cossart, *J. Cell Sci.* **105** (Part 3), 699 (1993).

12. J. A. Hoch, *Curr. Opin. Microbiol.* **3**, 165 (2000).
13. J. M. Boyd, *Mol. Microbiol.* **36**, 153 (2000).
14. R. B. Jensen, S. C. Wang, L. Shapiro, *Nature Rev. Mol. Cell. Biol.* **3**, 167 (2002).
15. P. Viollier, N. Sternheim, L. Shapiro, *Proc. Natl. Acad. Sci. U.S.A.* **99**, 13831 (2002).
16. ———, *EMBO J.* **21**, 4420 (2002).
17. P. Aldridge, U. Jenal, *Mol. Microbiol.* **32**, 379 (1999).
18. U. Jenal, *FEMS Microbiol. Rev.* **24**, 177 (2000).
19. C. Jacobs, D. Hung, L. Shapiro, *Proc. Natl. Acad. Sci. U.S.A.* **98**, 4095 (2001).
20. M. T. Laub, H. H. McAdams, T. Feldblyum, C. M. Fraser, L. Shapiro, *Science* **290**, 2144 (2000).
21. I. J. Domian, K. C. Quon, L. Shapiro, *Cell* **90**, 415 (1997).
22. J. R. Maddock, L. Shapiro, *Science* **259**, 1717 (1993).
23. M. R. Alley, J. R. Maddock, L. Shapiro, *Genes Dev.* **6**, 825 (1992).
24. P. Ames, C. A. Studdert, R. H. Reiser, J. S. Parkinson, *Proc. Natl. Acad. Sci. U.S.A.* **99**, 7060 (2002).
25. T. S. Shimizu *et al.*, *Nature Cell Biol.* **2**, 792 (2000).
26. N. Maki, J. E. Gestwicki, E. M. Lake, L. L. Kiessling, J. Adler, *J. Bacteriol.* **182**, 4337 (2000).
27. E. F. Bi, J. Lutkenhaus, *Nature* **354**, 161 (1991).
28. P. A. de Boer, R. E. Crossley, L. I. Rothfield, *Cell* **56**, 641 (1989).
29. C. A. Hale, H. Meinhardt, P. A. de Boer, *EMBO J.* **20**, 1563 (2001).
30. Z. Hu, J. Lutkenhaus, *Mol. Microbiol.* **34**, 82 (1999).
31. Z. Hu, E. P. Gogol, J. Lutkenhaus, *Proc. Natl. Acad. Sci. U.S.A.* **99**, 6761 (2002).
32. M. Howard, A. D. Rutenberg, S. de Vet, *Phys. Rev. Lett.* **87**, 278102 (2001).
33. F. J. Trueba, *Arch. Microbiol.* **131**, 55 (1982).
34. C. L. Woldringh, E. Mulder, P. G. Huls, N. Vischer, *Res. Microbiol.* **142**, 309 (1991).
35. A. L. Marston, H. B. Thomaides, D. H. Edwards, M. E. Sharpe, J. Errington, *Genes Dev.* **12**, 3419 (1998).
36. S. Ben-Yehuda, R. Losick, *Cell* **109**, 257 (2002).
37. P. A. Levin, R. Losick, *Genes Dev.* **10**, 478 (1996).
38. P. Eichenberger, P. Fawcett, R. Losick, *Mol. Microbiol.* **42**, 1147 (2001).
39. J. Pogliano *et al.*, *Mol. Microbiol.* **31**, 1149 (1999).
40. J. Dworkin, R. Losick, *Cell* **107**, 339 (2001).
41. F. Arigoni, K. Pogliano, C. D. Webb, P. Stragier, R. Losick, *Science* **270**, 637 (1995).
42. A. Feucht, T. Magnin, M. D. Yudkin, J. Errington, *Genes Dev.* **10**, 794 (1996).
43. N. King, O. Dreesen, P. Stragier, K. Pogliano, R. Losick, *Genes Dev.* **13**, 1156 (1999).
44. A. Feucht, R. A. Daniel, J. Errington, *Mol. Microbiol.* **33**, 1015 (1999).
45. L. Duncan, S. Alper, F. Arigoni, R. Losick, P. Stragier, *Science* **270**, 641 (1995).
46. N. Frandsen, I. Barak, C. Karmazyn-Campelli, P. Stragier, *Genes Dev.* **13**, 394 (1999).
47. A. Khvorova, V. K. Chary, D. W. Hilbert, P. J. Piggot, *J. Bacteriol.* **182**, 4425 (2000).
48. M. L. Zupancic, H. Tran, A. E. Hofmeister, *Mol. Microbiol.* **39**, 1471 (2001).
49. L. J. Wu, J. Errington, *Science* **264**, 572 (1994).
50. C. van Ooij, R. Losick, *J. Bacteriol.*, in press.
51. D. Rudner, R. Losick, *Proc. Natl. Acad. Sci. U.S.A.* **99**, 8701 (2002).
52. M. R. Alley, personal communication.
53. G. S. Gordon, A. Wright, *Annu. Rev. Microbiol.* **54**, 681 (2000).
54. R. B. Jensen, L. Shapiro, *Proc. Natl. Acad. Sci. U.S.A.* **96**, 10661 (1999).
55. C. D. Webb *et al.*, *Cell* **88**, 667 (1997).
56. P. Glaser *et al.*, *Genes Dev.* **11**, 1160 (1997).
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REVIEW

Anterior-Posterior Polarity in *C. elegans* and *Drosophila*—PARallels and Differences

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The eggs of *Caenorhabditis elegans* and *Drosophila* bear little similarity to each other, yet both depend on the *par* genes for control of anterior-posterior polarity. Here we explore possible common roles for the *par* genes (*pars*) in converting transient asymmetries into stably polarized axes. Although clear mechanistic parallels remain to be established, *par*-dependent regulation of microtubule dynamics and protein stability emerge as common themes.

The key to every biological problem must finally be sought in the cell, for every living organism is, or at some time has been, a cell.
E. B. Wilson, 1925 [(*1*), p. 1]

A major challenge in developmental biology is to understand how asymmetries are elaborated along the main body axes. How are heads made different from tails and everything in between? Remarkably, in many organisms these morphological differences can be traced back to the one-cell stage, where axis determinants localize to opposite ends of the egg. For many biologists, this realization has meant that to understand axis formation, one must first understand how asymmetries arise within a single cell: the egg.

Genetic screens in *Drosophila* and *C. elegans* have identified several regulators of egg polarity. These two models were long thought to bear little resemblance to one another. In *Drosophila*, polarization of the egg begins during oogenesis and requires microtubules. In contrast, in *C. elegans* polarization begins after fertilization and requires the actin cytoskeleton. The discovery of a group of genes essential for polarization of the *C. elegans* embryo ("*par*" genes) proved to be a turning point in the field (*2*). *par-3* and *par-6* encode two PDZ domain proteins, which together with the atypical protein kinase C PKC-3, form a complex in the anterior half of the *C. elegans* zygote (*3–5*). The serine threonine kinase PAR-1 and the ring finger protein PAR-2 occupy the posterior half (*6*, *7*). Two other genes, *par-4* and *par-5*, encode proteins that are uniformly distributed (*8*, *9*). Mutations in any one of these genes disrupt polarization of the zygote. Homologs of the *par* genes were soon discovered in mammals, where they regulate the polarization of epithelial cells, and in *Drosophila*, where they regulate epithelial and neuronal polarity [reviewed in (*10*, *11*)]. These observations prompted several groups to investigate whether the *par* genes might also regulate polarity in the *Drosophila* egg. Thus far, results indicate that this is the case. In fact, except for *par-2*, homologs of all the *par* genes have now been identified in *Drosophila* and are required for egg polarity (*12–20*). This remarkable conservation raises an apparent paradox: how can the same group of genes regulate polarity in such dissimilar cells (Fig. 1)? Here, we explore this issue by focusing on the role of the *par* genes in regulating anterior-posterior (A/P) polarity. We refer the reader to (*21*) and (*22*) for comprehensive reviews of axis formation in *Drosophila* and *C. elegans*.

Establishment of A/P Polarity in *C. elegans*

In *C. elegans*, polarization of the egg begins after fertilization and is initiated by the sperm asters, which marks the future posterior end of the embryo (*23–26*). The first sign of polarity is seen when contractions of the egg cortex suddenly cease in a small area near the sperm pronucleus and internal cytoplasm begins to flow toward that area (*27*). The cue that initiates these rearrangements is not known but appears linked to the nucleation of microtubules by the sperm-derived centrosomes (*24–26*). The actin cytoskeleton is also involved: cytochalasin treatment and depletion of the nonmuscle myosin NMY-2 block polarization (*28–30*). Close contact between the sperm asters and the cortex has been correlated with the onset of polarity (*31*),

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