In This Issue

Hoarding bug

A bug that causes meningitis does not like to share. On page 627, Doulet et al. show that it hoards actin-binding proteins away from immune cells. The hoarding allows the bacterium to cross the blood–brain barrier and to prevent immune cells from responding.

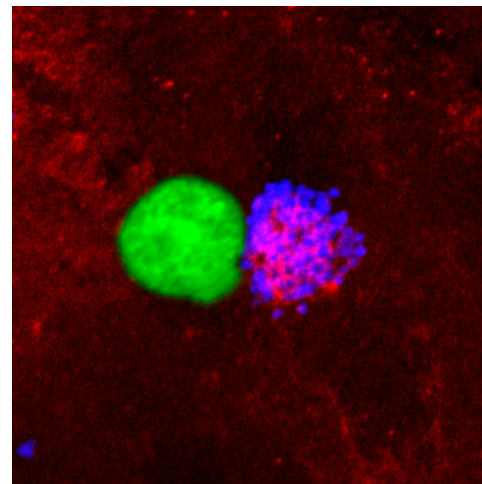
Many inflammation-inducing leukocytes pass from the bloodstream to infected tissues through loosened endothelial cell–cell junctions. The endothelial cells form cup-like actin structures that help leukocytes adhere and migrate. But these cups did not form on endothelial cells where extracellular colonies of *Neisseria meningitidis* grew, the authors found. Leukocytes thus failed to migrate to cell junctions and were easily detached from the surface by flow.

Cups were absent because the bug sequestered away host ezrin and moesin. These actin-binding proteins, which link adhesion molecules in the plasma membrane to the cytoskeleton, were required for cup formation and seem to be in limited quantities. Overexpression of either ezrin or moesin rescued leukocyte migration between infected cells.

The bug uses ezrin, moesin, and cytoskeletal molecules to adhere tightly to host cells in large, stable colonies. The cytoskeletal changes also allow a few of the bacteria to cross the blood–brain barrier via internalization at the apical surface and exocytosis out the other side, thus leading to meningitis.

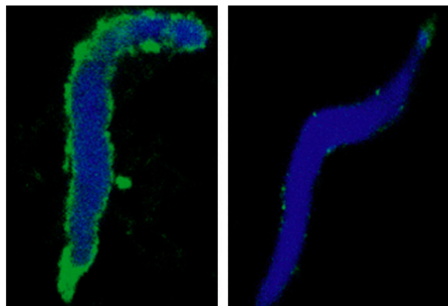
By usurping the same molecules that the immune cells use, *N. meningitidis* probably delays its pursuit by neutrophils, the host's first line of defense. No animal model of meningitis exists, however, to test whether this putative delay is a selective advantage for the bacterium.

This mechanism of immune interference may be specific to *N. meningitidis*. Most other human pathogenic bacteria, such as *Listeria* or *Shigella*, are quickly internalized, and thus have no lasting hold on ezrin or moesin. JCB



Infection of endothelial cells by *N. meningitidis* (blue) induces the massive recruitment of ezrin and adhesion molecules (red), preventing their accumulation at leukocyte (green) contact sites.

DNA recruits nuclear envelope



The reformation of the NE (green) around chromatin (blue) is impaired (right) by the addition of pure DNA.

DNA calls for the return of the nuclear envelope (NE) after mitosis, according to Ulbert et al. (page 469). The abundance of DNA may thus be one reason why the NE reforms so quickly.

NE reformation occurs so quickly that breaking down the process in vivo has so far been impossible. Many scientists instead use in vitro reconstitution assays, which indicate that chromatin decondensation (as occurs at the end of mitosis) initiates the recruitment of vesicle populations to chromatin. In the new work, the authors aimed to identify membrane and chromatin components that mediate this recruitment and thus NE assembly.

The main component of chromatin is, of course, DNA. The group found that excess naked DNA prevented in vitro NE reformation by titrating vesicles away from chromatin at early stages. DNA alone was a more potent inhibitor than chromatin, although chromatin proteins might also contribute, particularly later on as the envelope matures.

DNA does not bind pure liposomes, so the authors next sought the membrane proteins that mediate vesicle recruitment. Two transmembrane proteins of the inner nuclear envelope were already known to bind to DNA in vitro, and the authors have now identified several more. Most of them had highly basic nucleoplasmic regions, which are well-suited to binding negatively charged DNA. In fact, large basic tails were found in half of the tested nuclear membrane proteins but only 4% of ER and Golgi membrane proteins.

Loss of any particular transmembrane NE protein in vivo does not interfere with envelope reformation, so maybe they all work together. This redundancy and the prevalence of DNA might explain the speed of NE reformation.

Premature reformation is probably prevented by the inaccessibility of DNA in highly condensed mitotic chromatin. Additionally, many NE proteins are phosphorylated during mitosis. The added negative charge probably hinders their interaction with DNA. JCB

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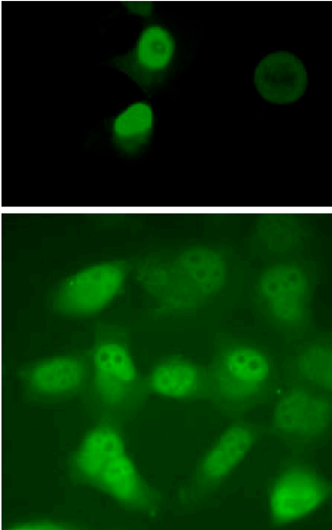

More p53 (green) is cytoplasmic when K320 is acetylated (bottom).

Acetylate to kill or save

Mammalian p53 can kill, or it can save. The end result, according to Knights et al. (page 533), depends on opposing acetylation events that send p53 down disparate paths.

The stabilization of p53 following cellular damage can trigger either apoptosis or a reversible cell cycle checkpoint that probably gives cells time to recover. As either savior or killer, p53 is subject to a battery of posttranslational modifications, including phosphorylation and acetylation. One such acetylation event (on K373) is now shown to trigger apoptosis, whereas another (on K320) works against death.

To understand how, the authors expressed mutant versions of p53 that mimic the acetylation events. After a brief treatment with a mild DNA-damaging agent, only cells with K320-modified p53 resumed proliferation; the rest died. The promoters bound, and genes activated, by the two p53 mimics correlated with their outcomes.

Many proapoptotic genes have promoters that might be too low in affinity for K320-acetylated p53. This form of p53 was in a slightly denatured state with less intrinsic DNA-binding ability. It was also more readily exported from the nucleus due to a block in serine-15 phosphorylation.

The K320 site is not conserved in fly and worm p53, which are solely death-inducing proteins. Mammalian tissues lacking regenerative abilities might have evolved the survival effect as a way to help maintain their numbers. Indeed, the group recently found that only K320 is acetylated during the neuronal maturation and neurite outgrowth that accompany regeneration. JCB

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Milton for motile mitochondria

Mitochondria are brought from the cell body to distant axons by kinesin. On page 545, Glater et al. identify two proteins that offer a regulatable link between motor and organelle.

The same group had already shown that the accumulation of mitochondria in photoreceptor axons required a cytoplasmic protein called milton. They now show why milton is so important—it recruits kinesin heavy chain to the mitochondria.

Milton hooks onto a mitochondrial protein called miro, as shown by immunoprecipitation assays and expression of miro mutants. Both miro and milton might offer opportunities for the cell to tune microtubule motility. One alternatively spliced version of milton, for instance, includes the kinesin-binding domain yet does not recruit the motor to mitochondria. Perhaps it is folded into an inert form that blocks transport. This form might even be opened by various cellular signals.

Miro contains calcium-binding and GTPase-like domains. Calcium is abundant at active synapses and requires large amounts of energy to be expelled. The authors therefore speculate that high calcium levels inhibit the transport complex so that mitochondria pause where they are needed.

Kinesin light chain was not found in the transport complex. Although this domain is usually the motor's cargo adaptor, milton seems to substitute for that job here. In theory, then, kinesin could have many adaptors specific to various cargo. JCB

Titinless mice

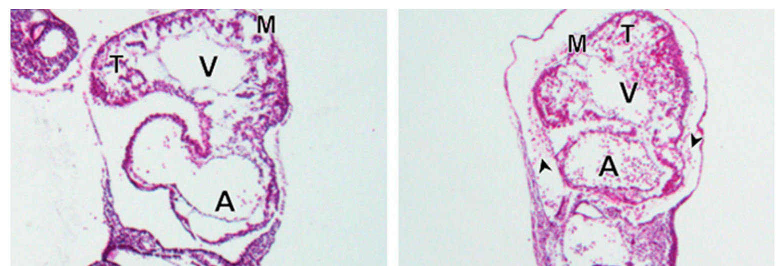
Titin needs no kinase activity to assemble sarcomeres, the building blocks of muscle tissue, according to Weinert et al. (page 559).

The giant protein called titin is a major component of muscles. Its COOH terminus, also known as the M-line region, includes a kinase-like domain. In yeast two-hybrid assays, titin's M-line interacts with proteins that shuttle to and from the nucleus, leading scientists to believe that it might initiate sarcomere assembly via signaling pathways.

A mouse strain lacking the kinase-like domain and nearby regions, however, reveals that the domain is not necessary for sarcomere assembly. These mice died during embryogenesis, but histological images revealed that sarcomere assembly and heart formation were initially normal. The lethal defect was heart failure due to a lack of sarcomere thickening—the addition of more actin and myosin filaments.

Recently identified signaling proteins that control muscle gene expression, and protein turnover via the kinase domain were not expressed during embryogenesis in wild-type or mutant mice. The effects thus seem to be mostly structural, unless as-yet unidentified kinase targets are required. In the deletion strain muscles, titin tails failed to overlap with neighboring molecules and were swinging freely, locked down on only one end.

Myomesin, a protein that links actin filaments, also binds to a titin region lost in the deletion. Weinert believes its lost binding site may lead to loosely packed filaments that impair the stabililty of the sarcomere. JCB



Without titin's kinase domain, the heart initially develops properly (left), but degenerates under strain (right).