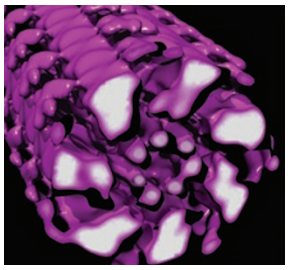
Stealth flagella



Flagella are long filamentous appendages used by bacteria to move around. Flagellar filaments are polymers of the protein flagellin. The sequence conservation of flagellin from different bacteria might be due to selective pressure to maintain function, and this has led to the assumption that all flagellar filaments have the same ultrastructure. Flagellins are usually recognized by the vertebrate innate immune system, binding and activating the Toll-like receptor 5 (TLR5). However, flagella from *Campylobacter jejuni* and *Helicobacter pylori* (human pathogens that cause diarrhea and peptic ulcer, respectively) do not activate TLR5, due to the lack of conservation of the motif recognized by this receptor. However, mutation of this motif in *Salmonella* led to a loss of motility, raising the question of how Campylobacter flagella could be functional. Now, EM analysis from Egelman and colleagues shows that flagellar filaments from *Campylobacter* are quite different from those from *Salmonella enterica*, previously shown to have 11 protofilaments. *Campylobacter* flagella are formed by seven protofilaments, and the coiled-coil regions of flagellin subunits are packed more loosely compared to those of *Salmonella*. These changes seem to reflect small sequence changes in the flagellin domain responsible for helical symmetry, suggesting that large changes in quaternary structure resulting from small changes in amino acid sequence may be a general mechanism for evolutionary divergence. (*Science* **320**, 382–385, 2008) *IC*

Viral splitting

Vaccine development against flaviviruses, a group of positive-strand RNA viruses that include West Nile virus (WNV) and dengue virus, has been limited. Traditional vaccination methods using live-attenuated virus risk the development of full infection, which can be fatal. DNA vectors encoding structural proteins, such as the premembrane (prM) or envelope (E) proteins, cannot replicate within the host as a live virus does, so large or repeated doses are required to cause protective immunity. Khromykh and colleagues have now developed a new DNA vaccine concept that holds much promise. The DNA plasmid used for vaccination encodes two separate mRNAs from two cytomegalovirus promoters placed back to back. The first promoter directs transcription of the entire WNV replicon RNA, except that the capsid protein gene is truncated, resulting in a nonfunctional protein. The second promoter directs transcription of mRNA for mature, full-length capsid protein. Together, the two RNAs have everything required for RNA replication and packaging, but only the replicon will be amplified by the nonstructural viral proteins and packaged into infectious particles. This system creates single-round infectious particles (SRIPs) that can be secreted to infect neighboring cells in a second round of amplification, but because the SRIPs lack a functional capsid gene, the infectious cycle stops there. As secreted structural (prM and E) proteins induce neutralizing antibodies and nonstructural proteins are targeted by cellular immune responses, the vaccine mimics viral infection. The authors found their vaccine was protective against WNV in mice and induced sufficient amounts of neutralizing antibodies required for protection in horses. This approach may be useful in vaccine design against other medically important flaviviruses. (*Nat. Biotech.*, advance online publication 20 April 2008, doi:10.1038/nbt1400) *MM*

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Translating semaphorin signals

Semaphorins (SMPs) are Plexin (PLX) receptor ligands involved in developmental processes. Translational regulation has been suggested to mediate SMP signaling, but the mechanism and targets of such regulation are unclear. Takagi and colleagues now address this question in *Caenorhabditis elegans*. Because the stereotypical tail fan morphology of males is altered when SMP–PLX-1 signaling is defective, the authors looked for suppressors of the *plx-1* mutant and identified *gcn-1*, an activator of the GCN-2 kinase, which phosphorylates and thus inactivates translation initiation factor eIF2α. Following up on this finding, the authors showed that forced SMP and PLX-1 expression decreases eIF2α phosphorylation and thus probably activates translation. Furthermore, eIF2α carrying a phosphomimic residue phenocopies *plx-1*, also suggesting that SMP–PLX-1 signaling antagonizes eIF2α inactivation. Knocking down other translation factors led to the *plx-1*–like tail fan phenotype, indicating that translational activation is a major conduit for mediating the SMP signaling response. The cofilin, or UNC-60, homolog was a plausible candidate for a target of translational control, given that actin regulation is a known outcome of SMP signaling. Further analysis found that the *unc-60* transcript's 3′ untranslated region (3′ UTR) is a target of translational regulation. Thus, regulation of eIF2α phosphorylation, and thereby UNC-60 synthesis, seems to be a major response to SMP–PLX signaling, at least in this developmental system. Further analysis will elucidate the mechanism underlying altered translation and the role of the *unc-60* 3′ UTR in this process. (*Genes Dev.* **22**, 1025–1036, 2008) *SL*

No nonsense

Genetic mutations can alter the primary sequence of a protein, and are linked to a broad range of diseases. Such mutations can also produce a premature termination codon (PTC); translation of an mRNA containing one of these PTCs would yield truncated, and possibly deleterious, proteins. Thus, mRNAs are carefully screened for PTCs: in mammalian cells, the nonsense-mediated mRNA decay (NMD) pathway is responsible for identifying PTCs and ensuring that mRNAs containing these mutations are promptly degraded. Maquat and co-workers recently probed the role of one of these proteins, the RNA helicase Upf1, in NMD. Upf1 was known to be part of a multiprotein complex that recognizes PTCs, and the authors determined that after PTC recognition, Upf1 phosphorylation increases its affinity for several NMD 'degradative factors', including the mRNA decapping enzyme Dcp1a. Experiments involving hepatitis C virus (HCV) internal ribosome entry site (IRES)-dependent translational initiation (which requires eIFs) and cricket paralysis virus (CrRV) IRES-dependent translational initiation (which does not require eIFs) suggested that phosphorylated Upf1 interacts with eIF2, eIF3 or both proteins. The authors then showed that after PTC recognition, phosphorylated Upf1 directly binds to eIF3 and prevents the 60S ribosomal subunit from binding to 40S-bound mRNA. As PTCs are detected during the 'pioneer' round of translation, these experiments suggest an elegant mechanism by which the aberrant mRNA is detected: Upf1 is phosphorylated after PTC recognition, translation of that mRNA by a subsequently initiating ribosome is inhibited and key members of the degradative pathway are recruited to the PTC-containing mRNA by phosphorylated Upf1. (*Cell* **133**, 314–327, 2008) *JMF*