Utrophin gets a new look

Utrophin, a member of the spectrin superfamily of actin-binding proteins ubiquitously expressed in human cells, helps link the actin cytoskeleton to the extracellular matrix. Galkin et al. report on page 231 that a newly developed image analysis method identifies two different modes of utrophin binding to F-actin. The findings contradict earlier studies on utrophin–actin interactions and provide a clearer explanation of how relatively few proteins may generate the diverse interactions and structures seen in the actin cytoskeleton.

The amino-terminal domain of utrophin contains a tandem pair of calponin homology (CH) domains, which are important for binding to actin. Unfortunately, disorder in binding and the variable twist of F-actin obscure the three-dimensional structure of utrophin–actin complexes seen under the electron microscope, making detailed studies problematic. Galkin et al. tackled this problem with their recently developed image analysis algorithm, which can separate classes of polymorphic structures that are indistinguishable by traditional techniques. First reported last year, the new method is rapidly gaining acceptance. It is now being used to study several other actin-binding proteins, as well as protein–DNA interactions during recombination.

Previous work identified only one type of utrophin binding to actin, but the new study shows that the utrophin actin-binding domain binds to F-actin in two distinct modes with different stoichiometries. The CH domains appear to bind in an extended conformation in both modes, as earlier X-ray crystallography studies had predicted. The two binding modes, and their fit within structural models, suggest that utrophin can establish different interactions with actin on multiple surfaces of actin subunits. Previous work has shown that other actin-binding proteins may exhibit similar diversity in their interactions, suggesting that multiple binding modes are a general theme allowing a small number of actin-binding proteins to create a diverse array of structures.

Consequences of a traffic jam

Identifying the genetic basis of a disease is only a small step toward understanding its pathogenesis, as Simons et al. clearly demonstrate on page 327. The dysmyelinating condition Pelizaeus-Merzbacher disease (PMD) is caused by duplication or overexpression of the myelin proteolipid protein (PLP) gene, but it was not clear if excess PLP caused dysmyelination directly or indirectly. Simons et al. characterized the changes in intracellular trafficking caused by PLP overexpression in three systems. Their findings help define a pathway that may transport myelin rafts in oligodendrocytes, and suggest that PLP overexpression causes PMD by a multistep, indirect mechanism.

The authors examined BHK cells, oligodendrocytes, and transgenic mice in which PLP was overexpressed. In these systems, PLP is not incorporated into lipid rafts, but is routed to late endosomes/lysosomes. Cholesterol then accumulates in these compartments, which normally maintain low cholesterol levels. Two fluorescently labeled sphingolipid analogues and GPI-YFP are also misrouted to late endosomes/lysosomes in PLP-overexpressing cells.

Based on these and earlier results, the authors propose that PLP overexpression saturates the myelin raft transport pathway, causing the surplus PLP to be routed to the degradative compartment. This rerouting also causes the misdirection of cholesterol, and eventually other raft components, to the late endosome/lysosome.

Thus PLP overexpression may trigger a chain of events that could trap myelin lipids, impair normal trafficking of late endosomes/lysosomes, and interfere with myelination-regulating signaling molecules that localize to lipid rafts. These defects may trigger the premature oligodendrocyte death seen in mouse models of PMD. The authors are now trying to identify the mechanism by which cell death is induced in PLP-overexpressing cells.