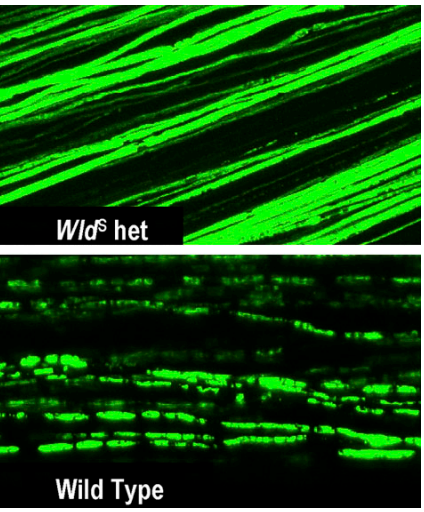
In This Issue



The chimeric Wld^S protein (top) protects neurons from degrading after insult, and it needs both its domains to do so.

How Wld^S neuroprotects

The neuronal guardian protein, Wld^S, like a true superhero, gained its powers by chance mutation. Now, thanks to two studies, we are closer to understanding how Wld^S exerts its protective force.

The slow Wallerian degeneration (Wld^S) protein and its neuroprotective powers came to exist thanks to a spontaneous mutation that fused the genes for Nmnat1 (a NAD^+ synthesizing enzyme) and Ube4b (a ubiquitination factor). The mutation was discovered by Coleman et al. in 1998 as the reason why a particular mouse strain, described by Hugh Perry a decade earlier, has exceptionally resistant neurons. Given Wld^S's ability, and thus its therapeutic potential for neurodegenerative diseases, it has become the focus of intense scrutiny.

Two groups, Conforti et al. and Avery et al., have now pulled apart Wld^S to learn the secret of its power. They show that both the enzymatic function of Nmnat1 and the ability of the Ube4-derived portion to bind a protein called VCP (or TER94 in flies) are indispensible for Wld^S's neuroprotective effect. VCP is abundant throughout the cell whereas Wld^S localizes primarily to the nucleus. However, when Avery et al. expressed mouse Nmnat3—a relative of Nmnat1 with the same enzyme activity, but that in fly neurons fails to localize to the nucleus—it conferred Wld^S-like protection. Both groups therefore believe that binding of Wld^S to VCP localizes Nmnat1 enzyme function to a region of the cytoplasm it wouldn't normally encounter. The question now is, where. **RW**

Avery, M.A., et al. 2009. *J. Cell Biol.* doi:10.1083/jcb.200808042.
Conforti, L., et al. 2009. *J. Cell Biol.* doi:10.1083/jcb.200807175.

1D is like 3D, but 2D isn't

Doyle et al. report that the movement of cells in a straight line is very similar to that in three dimensions (3D), whereas both differ markedly from 2D movement. Analyses in 1D might therefore provide useful information about cell movement in vivo.

The team made this discovery when they were investigating a new way to apply micropatterns of proteins onto coverslips (for analysis of cell attachment). Previous micropatterning techniques involved the printing of proteins onto gold-plated coverslips. However, while gold is great for protein binding, it's not so great for fluorescent microscopy—it readily absorbs fluorescent emissions. Doyle et al.'s new fluorescence-friendly approach was to coat coverslips with polyvinyl alcohol (PVA), which resists protein and cell binding, and then remove regions of the PVA by laser ablation. Proteins added to the coverslip will then stick to the ablated patterns only.

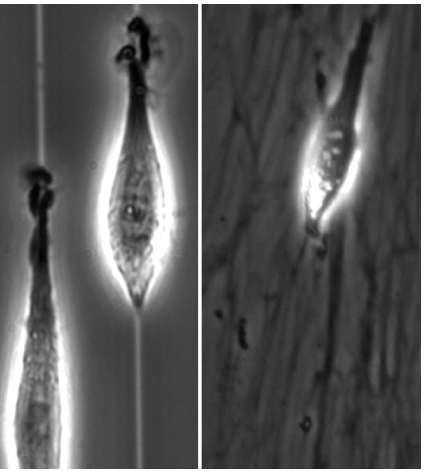
Among the micropatterns that the team drew were simple straight lines. When extracellular matrix proteins, and then cells, were added to such coverslips the cells started to migrate along the matrix-containing lines. Importantly, the cells' movement looked just like that on 3D fibrillar matrices.

Cells migrating in 2D have a spreading, multi-axial morphology, but on the 1D lines, as in 3D matrices, the cells were uniaxial. Their migration was also more rapid, even at high ligand densities (on 2D surfaces high ligand density slows cells, as they have trouble detaching).

The faster speed is probably explained by the fact that less of the cell is in contact with the surface, so more of its machinery is available to drive the cell forward. Indeed this would explain why Doyle et al. found that movement in 1D and 3D was dependent on myosin II contractility. Cells on 2D surfaces, on the other hand, use most of their myosin to make surface contacts, so actually speed up if contractility is lost.

So is there a future in 2D analyses? Doyle diplomatically suggests that researchers "have to take a closer look at whether or not what's happening in the dish is really what's happening in vivo." **RW**

Doyle, A.D., et al. 2009. *J. Cell Biol.* doi:10.1083/jcb.200810041.



1D cell migration (left) resembles 3D migration on a matrix (right).

Portioning up PIE-1

Asymmetry in the embryo starts with the first cell division. Daniels et al. show that one protein's asymmetrical distribution at this division is maintained by differential diffusion behavior.

In worms, PIE-1 is a germline-determining factor that represses somatic cell transcription programs. It starts off distributed symmetrically in the egg, but after fertilization it must be divvied up with each division. So what keeps PIE-1 enriched on one side of the cell?

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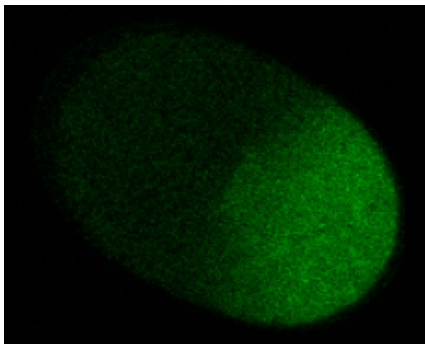
Daniels et al. ruled out immobilization and compartmentalization of the protein by analyzing the mobility of fluorescent PIE-1. They also showed that although localization of PIE-1 requires the reciprocal localization of a suspected PIE-1 degrader called MEX-5/6, this protein in fact does not degrade PIE-1 in the zygote.

Instead, PIE-1's asymmetry is down to diffusion kinetics. Analysis of single molecule movement revealed most PIE-1 molecules at the front of the cell moved quickly. At the rear, slow diffusers made up the majority and there was even a group of super-slow diffusers. Based on these findings, the team developed a model that explains PIE-1's distribution.

So what causes the difference in diffusion? The kinetics of the slowest group fits well with that of P granules, which are known to be enriched at the back of the cell and with which PIE-1 has been reported to interact. As for the slow diffusers, which account for the majority of the PIE-1 molecules at the rear, the authors suggest that PIE-1 is interacting with some type of RNA species. The next step is to determine whether this prediction is correct, and if so, what spatially restricts the RNA binding. **RW**

Daniels, B.R., et al. 2009. *J. Cell Biol.* doi:10.1083/jcb.200809077.



PIE-1 (green) diffuses slowly at the back of the zygote where it's enriched.

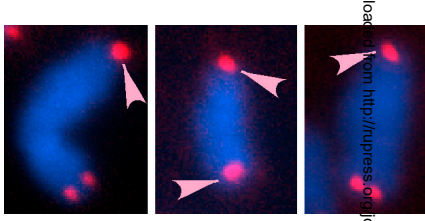
Tankyrase: A new breed of telomere security guard

At mitosis, a protein called tankyrase 1 serves as protector by separating sister chromatid strands to prevent their telomeres from fusing, conclude Hsiao and Smith.

The team had previously shown that cells lacking tankyrase 1 arrest at mitosis when their sister chromatid telomeres remain associated via protein–protein interactions—even though normal separation occurs along the rest of the chromatid pair. That work highlighted the unique nature of telomere cohesion and led the team to investigate more closely how and why tankyrase 1 prompts telomere separation.

For the how, the team's studies indicate that tankyrase prompts the removal of a telomere-specific TIN2/TRF1 complex that binds to chromatid cohesion proteins. As for the why, the authors demonstrate that promoting sister telomere separation prevents their fusion. After DNA replication, telomeres are processed to generate 3′ overhangs that form protective t loop structures by strand invasion. If chromatid separation does not occur, the DNA ends are at risk of invading the sister strand, and end joining. Indeed, many of the unseparated telomeres in tankyrase-lacking cells were fused together (by nonhomologous end joining). The team would now like to dissect the timing of 3′ end processing and sister telomere separation to understand when tankyrase 1 acts. **RW**

Hsiao, S.J., and S. Smith. 2009. *J. Cell Biol.* doi:10.1083/jcb.200810132.



Tankyrase 1 deficiency can prompt sister telomere fusion (arrowheads).

Bmper sticker sends Bmp to lysosomes

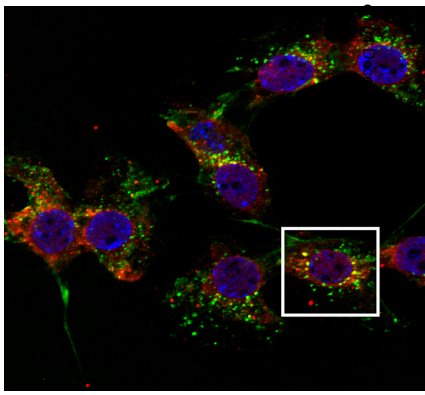
Kelley et al. describe how a protein called Bmper switches off Bmp signaling by targeting the growth factor to lysosomes for degradation.

Many developmental processes are regulated by members of the Bmp family, and Bmp activity can be modulated by secreted binding partners. Bmper is one such modulator, but has the unusual ability of both enhancing and diminishing Bmp activity, depending on its concentration. Small amounts of Bmper promote Bmp signaling, whereas higher concentrations are inhibitory. How Bmper exerts these opposing effects was unclear.

Kelley et al. found that high concentrations of Bmper relative to Bmp prompted both proteins to be internalized and trafficked to lysosomes for destruction. The N terminus of Bmper trapped and inhibited Bmp at the cell surface, while the C terminus—along with the Bmp receptor—drove the complex's internalization. Lower concentrations of Bmper, in contrast, didn't induce Bmp uptake. The authors don't yet know what triggers the switch from activation to internalization, but think that saturated binding of Bmper might induce a structural change in either Bmp or its receptor.

Author Cam Patterson speculates that Bmper's dual role serves to temporally restrict Bmp signaling by initially activating the pathway before rapidly down-regulating it. Importantly, the team found that two other Bmp inhibitors also stimulated Bmp endocytosis, so Patterson thinks this new mechanism may represent a general method by which modulating proteins inhibit Bmp. **BS**

Kelley, R., et al. 2009. *J. Cell Biol.* doi:10.1083/jcb.200808064.



High levels of Bmper (green) inhibit Bmp (red) by triggering its internalization and degradation.