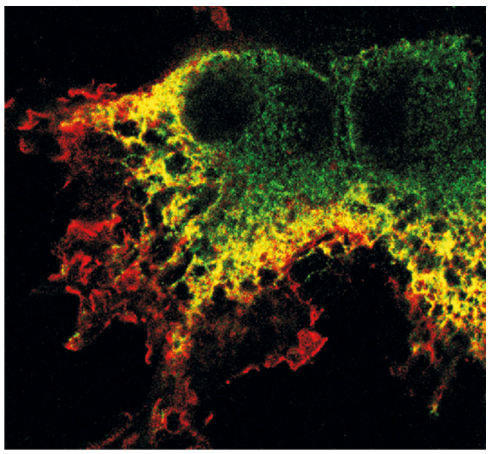
Research Roundup

Signaling together apart



USHKARYOV

Distinct latrophilin halves (red and green) wander away from each other.

After being cleaved apart, two halves of a split personality receptor protein are free to wander far away from each other, say Kirill Volynski, Yuri Ushkaryov, and colleagues (Imperial College, London, UK). But when signaling is needed the two halves reunite.

The receptor, called latrophilin, is known only as a binding site for the black widow poison latrotoxin. Although no endogenous ligand for latrophilin is known, a varied family of receptors exists with a similar organization. Each family member has two domains: an NH_2-terminal fragment (NTF) that interacts with other cell surface or possibly extracellular matrix proteins, and a COOH-terminal fragment (CTF) that resembles a G-protein–coupled receptor (GPCR). The two domains were known to be cleaved, but the persistence of the NTF on the cell surface led previous workers to assume that the transmembraneless NTF must remain bound to CTF.

The London group now shows that this is not the case. The two fragments have distinct localizations and can be aggregated away from each other using antibodies. Addition of a latrotoxin variant, however, induces clustering of the fragments and signaling.

For the cell, the fusion of two protein functions allows for coordinated expression, but cleavage allows divergent regulation. For example, the NTF may stay attached extracellularly even as the CTF is internalized to allow for GPCR desensitization. Ushkaryov now hopes to test whether different members of the protein family mix and match their respective halves. JCB

Reference: Volynski, K.E., et al. 2004. *EMBO J.* doi:10.1038/sj.embo.7600443.

Dormant tumors awaken

What happens during tumor remission? After chemotherapy, tumor cell numbers may be reduced, or surviving cells may enter an altered, dormant state. Evidence for dormancy now comes from Catherine Shachaf, Dean Felsher (Stanford University, Stanford, CA), and colleagues. In their mouse model, liver tumors can be forced into dormancy and then reawakened at will.

As in previous studies, regression was induced by shutting off the oversupply of MYC, in this case using a tetracycline-controlled promoter. Transgenic mice took ~12 weeks to develop the MYC-induced tumors, but within 4 days of MYC inactivation the cells died or differentiated into normal liver cells. Tumors regressed but could later be reactivated by switching MYC back on.

The persistence of dormant tumor cells was demonstrated by transplanting tumor cells containing transgenes for both luciferase and the tetracycline-controlled MYC. Even when MYC was inactivated the luciferase signal persisted in the transplanted cells, which looked like normal liver cells. Turning MYC back on restored tumorigenesis, increasing cell number and thus luciferase activity.

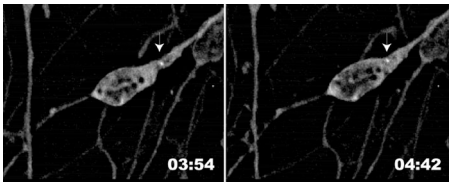
Reversible dormancy has not been seen in previous MYC inactivation studies. Felsher believes that the differences are explained by the properties of the tumors' originating tissues. Lacking MYC, tumor cells from a terminally differentiated tissue such as bone resort to terminal differentiation; those from the apoptotic-prone hematopoietic compartment die; and those from liver (a tissue that is rich in stem cells and can regenerate) differentiate but produce many stem cells. It is those stem cells that may perpetuate the dormant state and that Felsher wants to understand. JCB

Reference: Shachaf, C.M., et al. 2004. *Nature*. doi:10.1038/nature03043.

Neurons inch along

Centrosome and nucleus engage in an inchworm-like dance in certain migrating neurons, according to David Solecki, Mary Hatten, and colleagues (Rockefeller University, New York, NY).

The neurons under study migrate along glia to form the layered architecture of higher brain areas. Whereas primitive brain areas have a nuclear organization, the cortex builds its more complex circuitry by sending its neurons off on these treks.



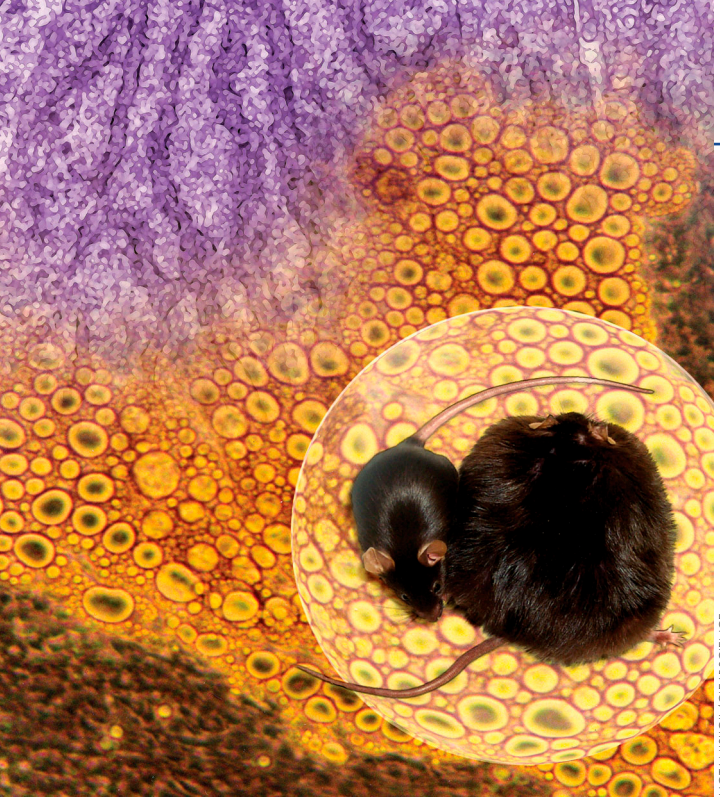
HATTEN/MACMILLAN

Centrosome (arrow) moves before nucleus.

The neurons were known to move and adhere in a periodic cycle, with a long process leading the way and the nucleus at the rear. But the Rockefeller group now shows that the centrosome moves forward first; the nucleus then closes the gap. This cycle has a similar period to the adhesion cycle. Although the relative timing of the events is not known, coordination may rely on mPar6α, which the Rockefeller group identifies as a centrosome component essential for centrosome and cell movement.

The nucleus is surrounded by a perinuclear microtubule cage whose shape is distorted during the cycle. The cage and nucleus are probably moved towards the centrosome by dynein. The movement of the centrosome itself is more of a mystery, both in terms of the responsible motor and the structure against which the motor pulls. JCB

Reference: Solecki, D.J., et al. 2004. *Nat. Neurosci.* doi:10.1038/nn1332.



HOTAMISLIGIL/CLEARSCIENCE

Stressed-out ER (top) may cause diabetes in overfed mice.

Text by William A. Wells
wellsw@rockefeller.edu

Diabetes as a stress response

The origin of adult-onset diabetes may be a counterproductive stress reaction in the endoplasmic reticulum (ER), according to Umut Özcan, Gökhan Hotamisligil (Harvard School of Public Health, Boston, MA), and colleagues.

Type II or adult-onset diabetes has remained a mystery because the body seemingly acts against its own interests. As caloric intake and obesity rise, the body does not increase insulin responses to pack away the excess energy but instead becomes resistant to insulin's energy-storing signal. This just makes the situation worse. "You develop a little bit of obesity and then everything starts going crazy," says Hotamisligil.

The Boston team now suggests that the body sees the stress of dealing with excess calories as analogous to an environmental or infectious stress, and responds appropriately. "Insulin is the most powerful signal opposing the mobilization of energy," says Hotamisligil. "You turn it off to mobilize energy against the pathogen or other stress."

The ER appears to be the site of this regulatory action. The Boston team found that indicators of ER stress such as the unfolded protein response were elevated in liver and adipose tissue but not muscle of diabetic mice. Drugs and genetic conditions that exacerbated ER stress, both in cultured cells and diabetic mice, resulted in increased insulin resistance. In cultured cells the converse was also true: an overdose of a stress-fighting protein reduced markers of insulin resistance.

ER stress may arise because extra calories have to be processed by the ER as they get turned into either extra proteins or more lipids. "Under the best conditions the [fat] cell uses all its capacity," says Hotamisligil. The stress signaling goes from the ER to the insulin receptor complex via the immune-activating JNK kinase, which may explain why the immune response is also turned on during type II diabetes.

Muscle is not a secretory cell type and thus may escape the original insult, but other studies suggest that it succumbs to a signal from fat cells that promotes insulin resistance. This signal may have been helpful during evolution but is now less useful, when the greater threat is not bugs but burgers. JCB

Reference: Özcan, U., et al. 2004. *Science.* 306:457–461.

A SNARE for fast endocytosis

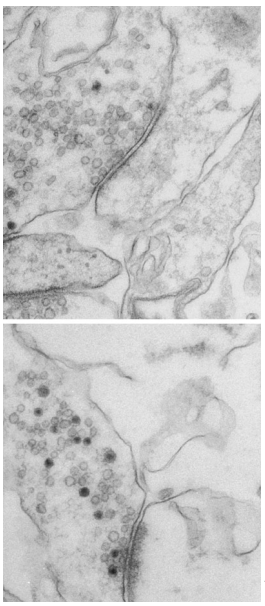
A fusion-promoting exocytic protein is also required for fast endocytosis, based on results from Ferenc Deák, Thomas Südhof, Ege Kavalali (UTSW, Dallas, TX), and colleagues. The requirement may give hints about how neurons recycle synaptic vesicles back into the cell so rapidly, and how exocytosis prepares the way for such a fast process.

Exocytosis is greatly reduced but not eliminated at synapses lacking the SNARE fusion-promoting protein synaptobrevin/VAMP. The group found that endocytosis was delayed in these neurons even if amounts of exocytosis were first equalized at wild-type and mutant synapses by using different stimulation protocols. Dyes provided a further suggestion that mutant synapses had selectively lost fast endocytosis, as mutant synapses released both fast- and slow-diffusing styryl dyes and wild-type synapses released only the fast-diffusing dye.

Hints of an endocytic SNARE role have been seen previously in yeast and flies. But in neither has the link been made to the special case of fast endocytosis. This process is essential for neurons, but the responsible molecules, unlike the clathrin used during slow endocytosis, remain obscure. Fast endocytosis may involve partial retention of vesicle structure ("kiss and run") between fusion and endocytosis.

What synaptobrevin is actually doing to promote endocytosis remains unclear. It may either act as a nucleator of endocytic proteins or, the authors suggest, be required to take the exocytic vesicle into a pathway that is primed for fast endocytosis. For example, synaptobrevin on the vesicle might attach to plasma membrane SNAREs to set up a hemi-fusion state, but then release the plasma membrane SNAREs before full fusion. Thus disengaged, the hemi-fusion structure could, when the pro-fusion calcium signal arrived, move into full fusion mode and back out to endocytosis without having to wait for SNARE disentanglement. JCB

Reference: Deák, F., et al. 2004. *Nat. Cell Biol.* doi: 10.1038/ncb1185.



DEÁK/MACMILLAN

Without synaptobrevin (top), endocytosis is slow.