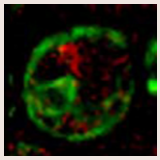
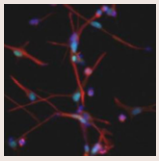
In this issue



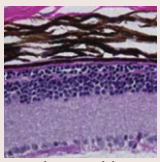
Stand up and be counted: Atg proteins in autophagosome formation

Autophagy is a system of bulk degradation of macromolecules and organelles that is conserved in eukaryotes from yeast to mammals, and is essential for maintaining cell homeostasis. The crucial event in this process is the formation of an autophagosome that contains the cytoplasmic materials and delivers them to the lysosome for degradation. In yeast, there are 18 autophagy-related (*ATG*) genes that are essential for autophagosome formation in response to starvation. Autophagy requires the generation of a cup-shaped membrane sac, called the isolation membrane (IM), from the pre-autophagosomal structure (PAS). Progress in understanding the detailed functions of the Atg proteins during IM expansion has been obstructed because it had not been possible to discriminate between the IM and PAS, owing to the resolution limit of light microscopy. Now (p. 2534), Kuninori Suzuki, Yoshinori Ohsumi and colleagues address this problem. Using fluorescent microscopy, the authors visualise expanding IMs as cup-shaped structures in *Saccharomyces cerevisiae* by enlarging a selective cargo of autophagosomes, and then finely map the localisation of individual Atg proteins. For example, they show that the IM is closely associated with the endoplasmic reticulum (ER) at ER exit sites, and that three Atg proteins localise adjacently to the ER exit sites at the IM. Taken together, these data suggest that Atg proteins have individual roles at spatially distinct sites during IM expansion.



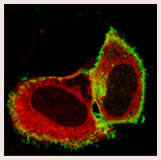
Genomic imprinting during reprogramming and redifferentiation

Pluripotent stem cells can differentiate into all cell types and self-renew without losing their differentiation potential. Induced pluripotent stem cells (iPSCs) can be generated from somatic cells by overexpression of certain transcription factors and have the same characteristics as embryonic stem cells (ESCs). During their reprogramming, iPSCs undergo epigenetic changes, including DNA methylation and histone modification, and ultimately acquire an epigenetic state similar to that of ESCs. However, it is unclear whether imprinted genes are maintained or revert back to the parthenogenetic state when iPSCs are redifferentiated into specialised cell types. Here (p. 2516), Jeong Tae Do and colleagues analyse genomic imprinting during reprogramming and redifferentiation by comparing biparental female neural stem cells (fNSCs), parental NSCs (pNSCs) and NSCs that have been obtained by redifferentiation from maternal iPSCs (miPSCs). They show that reprogrammed miPSCs can differentiate into a stable NSC line, and that these cells are very similar to normal fNSCs but not to the donor pNSCs. In particular, they find that some maternally imprinted genes, which are fully methylated in pNSCs, are demethylated in NSCs that have been derived from miPSCs, suggesting that parthenogenetic DNA methylation patterns in imprinted genes are reset following pluripotent reprogramming. These findings might have important implications for the use of iPSCs in stem-cell-based therapies.



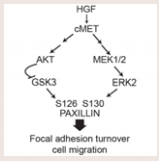
BBS7 is essential for BBSome formation

Bardet–Biedl syndrome (BBS) is an autosomal recessive ciliopathy that is associated with mutations in a number of genes encoding cilia proteins. Of the 17 genes identified so far, seven encode highly conserved core BBS proteins that form a stable complex called the BBSome, which functions in cilia membrane biogenesis. Although BBS mutant mice share common BBS phenotypes, phenotypic differences are observed among different BBS gene mutations, and physiological differences among BBS patients caused by mutations in distinct genes have been documented; thus, different BBS genes might have unique functions. BBS7 is an integral part of the BBSome, and here (p. 2372), Val Sheffield and colleagues examine the *in vivo* function of BBS7 by generating *Bbs7*-knockout mice. These mice developed BBS-associated phenotypes, including retinal degeneration, obesity, male infertility and hydrocephalus. The authors show that BBS7 is required for BBSome formation, and BBS7 and BBS2 are dependent on each other for stability. The BBSome is involved in cilia membrane biogenesis but, in this study, BBS7 was not required for ciliary membrane localisation or the function of bitter taste receptors. The absence of BBS7, however, led to an abnormal accumulation of the dopamine D1 receptor on the ciliary membrane. This work reveals, for the first time, the ciliopathy phenotypes associated with the loss of BBS7, and highlights the importance of the BBSome.



The secret life of STIM1 conformational change revealed

Stromal interaction molecule 1 (STIM1) acts as the Ca^{2+} sensor on the endoplasmic reticulum (ER) membrane. It monitors Ca^{2+} store content and, when stores are depleted, the SOAR/CAD domain of STIM1 undergoes a transition from an inactive conformation to an active exposed conformation, allowing it to couple with the plasma membrane channel Orai1, thereby activating the store-operated Ca^{2+} entry pathway. The molecular details of the transition of the SOAR/CAD domain, however, are not yet well understood. Khaled Machaca and colleagues (p. 2401) now use novel FRET sensors within the context of full-length membrane-anchored STIM1 to visualise the conformational change that STIM1 undergoes in response to Ca^{2+} store depletion. These sensors show that, at rest, STIM1 folds into a 'closed' conformation in which the active domain of SOAR/CAD is shielded. Depletion of Ca^{2+} stores induces STIM1 to cluster and adopt a stretched 'open' conformation that exposes SOAR/CAD so it can interact with Orai1. Moreover, mutational analyses show that, contrary to a previously proposed model, electrostatic interactions between the first and third coiled-coils of STIM1 are not involved in maintaining its closed conformation. The authors instead argue that stabilisation of this conformation requires an amphipathic α-helix between residues 317–336 in the first coiled-coil of STIM1. Further, a mutation in this helix results in an inactive STIM1 that does not respond to Ca^{2+} store depletion. These results provide important new insights into the structure–function regulation of STIM1.



ERK2 in the driver's seat for HGF-induced motility

The receptor tyrosine kinase (RTK) Met and its ligand hepatocyte growth factor (HGF) are thought to have a major role in the progression of tumour metastasis, owing in part to the ability of Met to induce a strong migratory response. Understanding the molecular mechanisms behind this process is therefore important for the design and application of specific cancer therapies. On page 2381, Peter Parker, Stéphanie Kermorgant and colleagues perform a small interfering RNA (siRNA)-based wound healing screen using the lung carcinoma cell line A549 to investigate the signalling pathways important for Met-mediated migration. Unexpectedly, they find that extracellular-signal-regulated kinase 2 (ERK2), but not ERK1, is a strong mediator of HGF-induced motility, and confirm this finding in other cell lines. Another hit in this screen was the focal adhesion protein paxillin, a known mediator of HGF-induced migration. The authors find that ERK2 is required for the HGF-induced phosphorylation of paxillin on serine 126. siRNA-mediated depletion of endogenous paxillin decreased HGF-mediated migration, and this effect was rescued by wild-type paxillin but not by a mutant in which serine 126 and the adjacent serine 130 were mutated to alanine residues. Finally, HGF stimulation was shown to increase paxillin turnover at focal adhesions, and this effect was inhibited by ERK2 (but not ERK1) knockdown. This study provides strong evidence for a functional distinction between ERK1 and ERK2 isoforms in HGF-dependent motility, and identifies paxillin as an important downstream mediator.

From *Development*

Pygopus, chromatin and Wnt signalling

The mouse genome contains two genes encoding for Pygopus (Pygo), which was identified as a Wnt signalling component in *Drosophila melanogaster*. All Pygo proteins contain a conserved plant homology domain (PHD) that allows them to bind di- and tri-methylated lysine 4 on histone H3, but it was not known whether histone binding is required for Pygo to function in Wnt signalling. In *Development*, Konrad Basler and colleagues investigate this question by generating mice homozygous for a PYGO2 mutation that abolishes chromatin binding. Surprisingly, PYGO2–chromatin binding is not needed to maintain the function of PYGO2 during mouse development, and abrogation of the PYGO2–chromatin interaction has little effect on Wnt signalling-related transcription during tissue homeostasis. Compromised PYGO2–chromatin binding leads to male sterility, however, and the researchers report that PHD-dependent recruitment of Pygo to regulatory regions in the testes is important for the recruitment of the histone acetylase GCN5 to chromatin. These results identify a testis-specific role for PYGO2 as a chromatin remodeler that is unrelated to its role as a modulator of Wnt signalling.

Cantù, C., Valenta, T., Hausmann, G., Vilain, N., Aguet, M. and Basler, K. (2013). The Pygo2-H3K4me2/3 interaction is dispensable for mouse development and Wnt signaling-dependent transcription. *Development* **140**, 2377-2386.