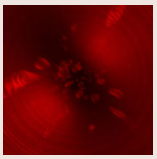
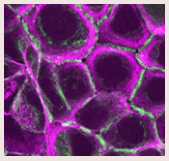
In this issue



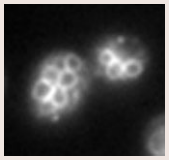
MINIFOCUS: the ESCRT machinery

In this issue, we are pleased to present a collection of articles that focus on the endosomal sorting complex required for transport (ESCRT) machinery. These complexes were initially identified on the basis of their important function in the sorting of ubiquitylated receptors into multivesicular bodies (MVBs). Since this discovery, cell biologists have made significant progress in understanding the structure and function of the ESCRT machinery at the molecular level, and it is now clear that its function extends to many other topologically equivalent cellular processes that involve a membrane-scission event. This issue contains three articles that bring these new ideas together: on page 2163, James Hurley and colleagues provide a schematic overview in their Cell Science at a Glance article, and examine the molecular interactions of the ESCRT machinery that enable its membrane-scission activity. In a Commentary on page 2167, Bethan McDonald and Juan Martin-Serrano discuss how the ESCRT machinery is involved in the diverse processes of MVB formation, cytokinesis and viral budding. Finally, Tor Erik Rusten and Harald Stenmark discuss the involvement of the ESCRT machinery in autophagy in an Opinion article on page 2179.



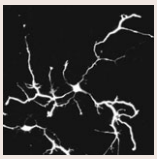
CD151: a two-faced tetraspanin?

Tetraspanins regulate tumour-cell metastasis through effects on signalling pathways and integrin-dependent interactions with the extracellular matrix. On page 2263, Christopher Stipp and colleagues investigate how the tetraspanin CD151 regulates interactions between tumour cells, and show that it is important for maintaining the stability of E-cadherin-based cell-cell junctions. By knocking down CD151 expression in epithelial carcinoma cells, they show that CD151 silencing causes an increase in the rate of collective cell migration, despite a decrease in the velocity of single cells. Microscopic analysis reveals that the junctional localisation of E-cadherin is perturbed in CD151-silenced cells, and adhesion assays show that, although E-cadherin-based junctions can initially form in the absence of CD151 expression, they are unstable compared with junctions in wild-type cultures. But CD151 and E-cadherin do not physically interact, so how does CD151 mediate its effect? CD151-silenced cells are shown to have increased activity of the small GTPase RhoA, and to form more dynamic monolayers than wild-type cells. CD151 has been implicated as a promoter of metastasis in some settings, but these results suggest that it also negatively regulates metastasis by suppressing RhoA activity, thereby stabilising junctions between E-cadherin-expressing tumour cells.



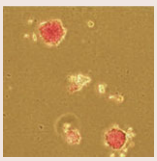
Switching on syntaxins

Membrane fusion in all eukaryotes is regulated by SNARE complexes, which comprise a four-helical bundle of SNARE motifs. Syntaxins form part of this complex, and contain in their structure an N-terminal Habc domain in addition to a SNARE motif. It has been previously shown that some syntaxins exist in a closed conformation, in which the Habc domain binds intramolecularly to the SNARE motif, preventing the formation of SNARE complexes. However, whether this is a conserved feature of syntaxins has been a contentious issue. Now, Nia Bryant and colleagues (p. 2292) show that mammalian syntaxin 16 (Sx16) is a functional homologue of yeast Tlg2p, and that these syntaxins are regulated in an evolutionarily conserved manner. More specifically, they show that the conserved Sec1p/Munc18 (SM) protein Vps45p regulates the assembly of SNARE complexes by relieving the inhibition mediated by the syntaxin Habc domain. Although more detailed structural data will be required to confirm the conformational changes of these proteins at the molecular level, this study indicates that SM proteins can facilitate the switch of syntaxins from a closed to an open conformation and thereby regulate SNARE-complex assembly.



Shaping neurons with Nek3

NIMA-related kinases (Neks) have known roles in cell-cycle regulation and ciliogenesis, but the recent finding that Neks are expressed in the nervous system suggests that they might also have a role in other cellular processes. On page 2274, Jeffrey Milbrandt and colleagues investigate the role of Nek3 in post-mitotic mouse neurons, and show that this kinase regulates neuronal morphology and polarity through effects on microtubules. Previously, the Thr475 residue was identified as a highly conserved phosphorylation site located in the PEST domain of Nek3; in this study, the authors show that the expression of Nek3 mutants lacking either the PEST domain or the Thr475 residue induces abnormal neuronal morphology, suggesting that Nek3 phosphorylation is essential for its normal function. Consistent with the known role of Neks in regulating microtubule dynamics, the authors show here that the expression of Nek3 mutants causes a decrease in the level of acetylated α-tubulin in neurons, although the overall α-tubulin network was not disrupted. Finally, they use inhibitors to show that the effects of the Nek3 mutants on α-tubulin are mediated by histone deacetylase 6 (HDAC6). The identity of the kinase that phosphorylates Nek3 at Thr475, and the nature of Nek3 downstream substrates, remain to be discovered.



Teasing out PI3Ks in ESCs

Defining the underlying pathways that control self-renewal and proliferation is an important goal in embryonic stem cell (ESC) research. On the basis of previous work showing that phosphoinositide 3-kinase (PI3K) family members are involved in regulating the behaviour of ESCs, Emmajayne Kingham and Melanie Welham (p. 2311) now seek to understand the role of specific class-IA PI3K isoforms in mouse ESC self-renewal and proliferation. Each of the three class-IA PI3Ks comprises a 110-kDa catalytic subunit (p110α, p110β or p110δ) and a regulatory subunit; differential roles of these three catalytic isoforms have been reported in many cellular processes. In this study, the authors specifically inhibit these three different p110 isoforms using isoform-selective small-molecule inhibitors or small interfering RNAs to investigate their respective roles in regulating ESC behaviour. They find that p110β, but not p110δ or p110α, is required to maintain optimal ESC self-renewal. By contrast, p110α regulates ESC proliferation. The authors conclude that different isoforms of the class-IA PI3K catalytic subunit are coupled to self-renewal and proliferation in mouse ESCs. However, because there is also evidence for crosstalk between the different isoforms, they hypothesise that PI3K signalling might link self-renewal with proliferation in ESCs.

Development in press

Keeping neuroblast self-renewal in check

Drosophila larval brain neuroblasts normally divide asymmetrically to generate another self-renewing neuroblast and a more fate-restricted cell, and failure of asymmetric division results in neuroblast overgrowth that resembles some vertebrate brain tumours. In a paper published in *Development*, Hongyan Wang and colleagues now identify protein phosphatase 2A (Pp2A) as a novel tumour suppressor that limits neuroblast self-renewal. The authors report that defects in, or the depletion of, Pp2A cause larval neuroblast overgrowth, which occurs at the expense of normal neuronal differentiation. Pp2A also regulates neuroblast polarity, and the defects caused by Pp2A disruption resemble those caused by mutations in *polo*, which encodes a kinase that phosphorylates and thereby activates the cell-fate determinant Numb. The authors go on to show that Pp2A regulates Numb activity by promoting Polo expression. On the basis of these and other findings, the authors propose that Pp2A acts upstream of Polo and Numb to block excessive neuroblast self-renewal. Whether this function of Pp2A is conserved in other stem-cell systems awaits future investigation.

Wang, C., Chang, K. C., Somers, G., Virshup, D., Ang, B. T., Tang, C., Yu, F. and Wang, H. (2009). Protein phosphatase 2A regulates self-renewal of *Drosophila* neural stem cells. *Development* **136**, 2287-2296.