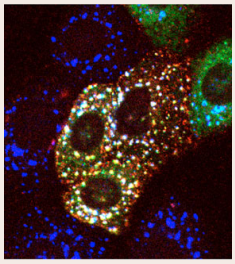
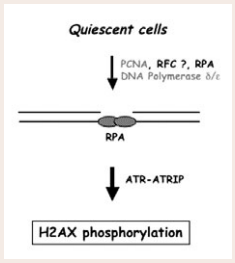
In this issue

by Jane Bradbury



Resetting a SNARE

Fusion of membrane-bound vesicles with target compartments is crucial for intracellular trafficking. v-SNAREs (soluble N-ethyl maleimide sensitive factor adaptor protein receptors) on vesicles and t-SNAREs on target compartments mediate each fusion event. Because the v-SNARE is transferred into the target compartment during fusion, it must be recycled to the donor compartment, but how? On p. 1028, Wanjin Hong and co-authors reveal the trafficking itinerary of VAMP4, a v-SNARE enriched in the trans-Golgi network (TGN) that functions in endosome-to-TGN trafficking. Using GFP-tagged VAMP4 and an anti-GFP antibody, the authors show that VAMP4 is endocytosed from the cell surface by clathrin-dependent pathways and passes through the early and recycling endosomes before being directly transported to the TGN. That is, VAMP4 – i.e. it is recycled to the endosomes from the TGN via the cell surface. Internalisation of VAMP requires the double double-leucine motif in its TGN-targeting signal whereas its transport from endosomes to the TGN involves the signal's acidic cluster. Thus, the authors conclude, the TGN-targeting signal of VAMP4 contains the sorting information necessary for efficient recycling of VAMP4 via a route that fits in with its role in trafficking.

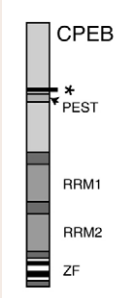


Histone H2AX minds the gap

DNA damage must be rapidly detected and repaired to avoid genomic instability. In human cells, the ATM (ataxia-telangiectasia mutated) protein phosphorylates histone H2AX at double-stranded DNA breaks; the phosphorylated histone then tethers DNA-repair proteins at the break. Replication stress caused by UV irradiation also induces H2AX phosphorylation – this time by ATR (ATM and Rad-3 related) – in cycling cells, but what happens in quiescent cells exposed to UV irradiation? Tsukasa Matsunaga and colleagues now report that ATR-dependent H2AX phosphorylation also occurs in quiescent human cells after UV irradiation but via a distinct pathway (see p. 1104). Their experiments indicate that low levels of replication factors cause the perturbation of nucleotide excision repair (the major system for removing UV-induced DNA lesions) at a gap-filling step, which leads to the formation of single-stranded DNA gaps and the subsequent phosphorylation of H2AX by ATR. Because most cells do not cycle in vivo, this route to H2AX phosphorylation could be crucial for the maintenance of genomic stability in many tissues.

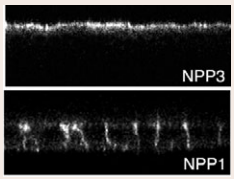
Neuronal death by consumption

Autophagy – a process in which eukaryotic cells digest their own organelles during development or starvation – has been implicated in the necrotic cell death that occurs in many human neurodegenerative disorders. To investigate this possibility, Tibor Vellai and co-workers have turned to *C. elegans* and, on p. 1134, they reveal a role for autophagy genes in ion-channel-dependent neurodegeneration in this organism. The authors show that mutational or RNAi-mediated inactivation of the worm autophagy genes *unc-51*, *bec-1* and *lgg-1* partly suppresses the degeneration of neurons that is induced by mutations affecting ion channel activity or by neurotoxin treatment. They also show that starvation promotes ion-channel-dependent necrosis whereas knocking down TOR (a kinase required for nutrition signalling) downregulates the autophagy gene cascade and protects neurons from necrotic cell death. Thus, the authors conclude, autophagy genes seem to be involved in neuronal necrotic cell death but, they add, further research is needed to determine whether autophagy itself is involved in neurodegeneration.



Oocyte maturation: new route MAP(K)

During steroid-induced maturation of *Xenopus* oocytes, activation of the cytoplasmic polyadenylation element binding protein (CPEB) by phosphorylation induces the translation of maternal mRNAs that encode cell-cycle regulators. But what controls CPEB activation? The answer, claim Laura Hake and colleagues, is a complex containing mitogen-activated protein kinase (MAPK) and the Rho-family guanine nucleotide exchange factor XGef (see p. 1093). XGef overexpression is known to accelerate meiotic progression and XGef interacts with CPEB throughout meiosis. Given previous reports that Aurora kinase A (Aur-A) activates CPEB by phosphorylation, the authors asked whether XGef controls the activity of Aur-A towards CPEB. Unexpectedly, they found that inhibition of Aur-A does not impair CPEB phosphorylation but that inhibition of MAPK does. In addition, they report that XGef forms a complex with MAPK and CPEB, and that MAPK phosphorylates several residues in CPEB although not S174, which is key to activating CPEB function. The authors propose, therefore, that MAPK drives meiotic progression by priming CPEB for phosphorylation on S174 by an as-yet-unidentified kinase.



Trans-membrane proteins sorted!

In epithelial cells, sorting of glycosylphosphatidylinositol-anchored proteins to the apical surface requires their interaction with lipid rafts – Triton-X-100-resistant membrane microdomains. Apical transmembrane proteins, although insoluble in weaker detergents such as Lubrol, tend to be soluble in Triton X-100. So are detergent-resistant rafts important for sorting of these proteins? On p. 1009, Jean-Louis Delaunay and colleagues report that for nucleotide pyrophosphatases/phosphodiesterases (NPPases), type II transmembrane proteins, there is not a strict correlation between detergent resistance and apical targeting. NPP1 is basolaterally localised and detergent soluble, whereas NPP3 is apically localised and Lubrol-insoluble. To examine the relationship between detergent resistance and apical targeting of these proteins, the authors stably transfected MDCK cells with wild-type NPP1 and NPP3, tail mutants, and chimeric constructs that combined different domains of the two proteins. Their results indicate that Lubrol resistance is an intrinsic property of NPP3 that does not determine its final destination. This is therefore strong evidence against the theory that Lubrol-resistant rafts target NPP3 and other transmembrane proteins to the apical membrane.

Development in press

Metamorphosis – the end of a tail

How a tadpole metamorphoses into an adult is a captivating process. But how does the tail disappear? In a paper published in *Development*, Chambon et al. describe a gene network required for apoptosis in the regressing tail of *Ciona* tadpoles. Using microarray analysis of *Ciona* larvae treated with inhibitors of the MAP kinase and JNK pathways, which regulate apoptosis, these investigators identified genes whose expression changes during metamorphosis. *Ci-sushi*, one such gene, is expressed in the tail epithelia and is downregulated in the presence of JNK inhibitors. When *Ci-sushi* is knocked down by a morpholino, tail cell death and regression are prevented. Since it encodes a protein important for cell-cell communication, the authors propose that JNK activity in the CNS (which escapes cell death) causes apoptosis in adjacent cells through Ci-sushi activity. Similar studies of *Ciona* larvae might shed light on the regulation of apoptosis in other vertebral column tissues, such as the neural tube.

Chambon, J.-P., Nakayama, A., Takamura, K., McDougall, A. and Satoh, N. (2007). ERK- and JNK-signalling regulate gene networks that stimulate metamorphosis and apoptosis in tail tissues of ascidian tadpoles. *Development* **134**, 1203-1219.