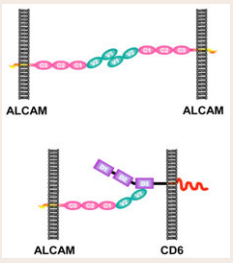
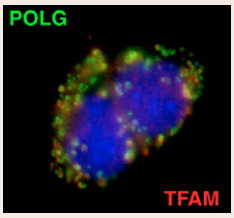
# In this issue

by Jenny Bangham



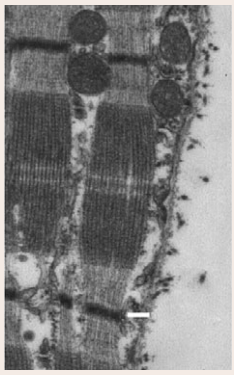

## Leukocyte sticking power

Cell adhesion molecules have diverse roles in development, tissue organisation and tumour progression. But are distinct characteristics of the molecule important in each case? To address this question, Carl Figdor and colleagues have taken the unusual step of using atomic force microscopy (AFM) on living cells to investigate the kinetics and mechanics of the activated leukocyte cell adhesion molecule (ALCAM). ALCAM is involved in several dynamic situations – it engages in both homotypic interactions (important for maintaining tissue architecture and in metastasis) and heterotypic interactions with CD6 (important for association between dendritic cells and T cells). The authors measured the strength of adhesion between single ALCAM-expressing cells on the tip of the probe used in AFM and surfaces, coated with either ALCAM or CD6 molecules, under different degrees of external loading (see p. 3965). They find that, at physiologically relevant forces, the ALCAM-ALCAM interaction is much more labile than the ALCAM-CD6 interaction. This has not been apparent from soluble-ligand-binding assays and indicates that the interactions have very different underlying kinetic and mechanical features. The authors suggest that the different strengths of the homotypic and heterotypic associations reflect their distinct roles in migration of melanoma cells, binding between early dendritic cells and T cells, and dendritic-cell-induced T-cell proliferation.



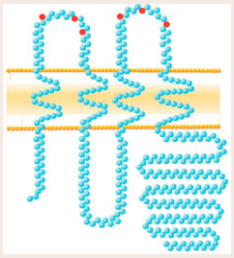

## Channelling stem cell energy

As embryonic stem cells (ESCs) differentiate, their metabolic requirements change – there is a shift from glycolysis to oxidative phosphorylation, and the number of mitochondria inside the cell increases in a lineage-specific way. On p. 4025, Justin St. John and colleagues investigate how ESCs coordinate these changes, analysing mitochondrial DNA (mtDNA) replication during differentiation of mouse ESCs. The researchers used real-time PCR to track the transcription of genes needed for mtDNA replication (such as POLG and Tfam) over this period. They find that the degree of ESC pluripotency correlates with mtDNA copy number and with the transcription of POLG and TFAM – and that this correlation varies in an ESC-line-specific way. They observe similar patterns when they use agents to direct differentiation towards certain cell fates. Using RNAi, they also show that POLG is required for the maintenance of pluripotency in ESCs. This supports the idea that a certain level of POLG is required to maintain pluripotency and that modulation of its activity can result in differentiation.



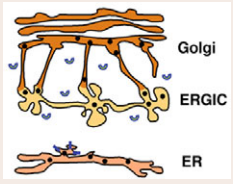
## Keratin shows its muscle

The strength and integrity of striated muscle cells depends crucially on links between the sarcolemma (the plasma membrane of muscle cells) and the underlying myofibrils (which orchestrate muscle contraction). The sarcolemma is organised into structures called costameres, which are linked to the contractile apparatus by several different cytoplasmic filaments, including intermediate filaments. On p. 3999, Robert Bloch and colleagues investigate the roles of the keratin component of these intermediate filaments by looking closely at the sarcolemma and costameres of keratin-19-null mice. These mice have mild myopathy; the link between the sarcolemma is disrupted, and there is a large gap between the sarcolemma and adjacent myofibrils, which becomes filled with mitochondria. Loss of keratin 19 also compromises costamere organisation, affecting in particular the cytoskeletal binding partners of the important dystrophin-dystroglycan complex. The authors propose that the myopathic phenotype of these mice is due in part to a shift in distribution of mitochondria – because subsarcolemmal mitochondria do not function as efficiently as intermyofibrillar mitochondria. They conclude that keratin 19 is important for organising costameres and their links to the contractile apparatus, and that it controls the distribution of mitochondria in skeletal muscle fibres.



## Connexin in the dock

Gap junctions – the channels that allow adjacent cells to exchange molecules – are made of two hemichannels that dock with one another through their extracellular domains. Undocked hemichannels are also thought to have roles in intercellular communication, but until now these had not been investigated in vivo. Gerald Kidder and colleagues (p. 4016) have been probing possible roles for undocked hemichannels, using mouse folliculogenesis as a model. During folliculogenesis, developing oocytes are surrounded and nurtured by granulosa cells that are connected to one another by gap junctions containing the protein connexin 43 (Cx43). To find out whether undocked hemichannels have a role in this process, the researchers produced constructs encoding wild-type Cx43 or a Cx43 mutant that can form hemichannels but not intercellular gap junctions. They then expressed these in Cx43-deficient granulosa cells and combined them with wild-type oocytes to make reaggregated ovaries. Whereas wild-type Cx43 constructs can rescue folliculogenesis, the mutant Cx43 construct cannot. So although in-vitro-based studies have shown evidence for gap-junction-independent roles, in this first in-vivo system undocked hemichannels cannot compensate for intercellular gap junctions.




## COPIng with selection

The vesicle coat protein COPI is at the heart of trafficking through the Golgi, but its precise roles are still the subject of some debate. The recruitment of COPI involves ADP-ribosylation factors (ARFs), which cycle from inactive to active states with the help of ARF-activating guanine nucleotide exchange factors (GEFs). On p. 3929, Elizabeth Sztul and colleagues investigate the role of COPI in Golgi structure and function by knocking down the ARF-GEF GBF1. They show that GBF1 is the major recruiter of COPI in the cell. Surprisingly, preventing recruitment of COPI to membranes does not cause the complete collapse of the secretory pathway but does cause selective tubulation of the cis-Golgi. Equally unexpected is the finding that, although depletion of GBF1 blocks the transport of transmembrane proteins, it does not affect the trafficking of soluble secretory proteins. The authors therefore suggest that GBF1-mediated COPI recruitment is required for trafficking of select types of cargo – transmembrane proteins in particular – through specific stages of the secretory pathway.

## *Development* in press

### FGF keeps segmentation clock ticking

The segmented pattern of the vertebrate spine is established during embryogenesis by the formation of somites (blocks of mesoderm that form the vertebrae and back muscles) at regular time intervals. In the 'clock and wavefront' model for somitogenesis, pulses of Notch, fibroblast growth factor (FGF) and Wnt signalling in the presomitic mesoderm (PSM) are somehow translated into a periodic array of somites at the so-called wavefront. In a paper published in *Development*, Olivier Pourquié and colleagues now provide the first genetic evidence that FGF signalling positions the wavefront and controls the segmentation clock in mice. They show that conditional deletion of the FGR receptor gene *Fgfr1* in the mesoderm abolishes FGF signalling and disrupts normal cyclic gene expression in the PSM. It also arrests somite formation. In addition, pharmacological inhibition of FGF signalling blocks the oscillations of both Wnt and Notch signalling in the PSM, but with different kinetics. The researchers conclude, therefore, that FGF signalling acts upstream of Wnt and Notch signalling to control the segmentation clock during somitogenesis.

**Wahl, M. B., Deng, C., Lewandoski, M. and Pourquié, O.** (2007). FGF signaling acts upstream of the NOTCH and WNT signaling pathways to control segmentation clock oscillations in mouse somitogenesis. *Development* **134**, 4033-4041.