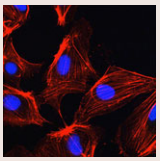
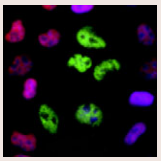
In this issue



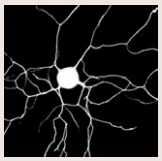
VEGF branches out with PLCβ3

The binding of vascular endothelial growth factor (VEGF) to its receptors (VEGFR1 and VEGFR2) on endothelial cells can induce many effects, including proliferation, migration and angiogenesis, through the activation of several intracellular signalling pathways. For example, Debabrata Mukhopadhyay and colleagues showed in a previous study that VEGF induces the lipase activity of phospholipase Cγ1 (PLCγ1) and PLCβ3. Whereas PLCγ1 activation is known to have a role in VEGF-induced DNA synthesis, the role of PLCβ3 downstream of VEGF has been unclear. Now, Mukhopadhyay and colleagues (p. 1025) use RNA-interference techniques to show that PLCβ3 promotes directional migration and inhibits proliferation downstream of VEGFR2 in endothelial cells. They show that the pro-migratory function of PLCβ3 involves its effects on actin reorganisation and its capacity to activate the small GTPase Cdc42, and that PLCβ3 inhibits proliferation through effects on the cell cycle. Therefore, the authors conclude that PLCβ3 is important in the context of angiogenesis by promoting a pro-migratory and anti-proliferative phenotype in endothelial cells.



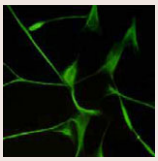
Mapping a COP9-signalosome network

Cullin-RING E3 ligases (CRLs) are the largest known family of ubiquitin ligases. The activity of some CRLs is inhibited by an eight-subunit complex known as the COP9 signalosome, which can act on the cullin subunit of CRLs and thereby influence the ubiquitylation of CRL substrates. On p. 1035, Lionel Pintard and colleagues use mass spectrometry to identify the subset of CRLs that might be regulated by the COP9 signalosome. By determining the adaptor proteins with which six of the COP9-signalosome subunits interact, the authors identify 15 CRLs that associate with the COP9 signalosome. Interestingly, most of these CRLs have a role in DNA metabolism; the authors propose that their coordinated regulation might ensure rapid CRL-mediated responses to specific physiological cues. The authors' mass spectrometry analysis also reveals that Dda1, a small protein that is thought to associate with CRL4 complexes, positively regulates their ubiquitin-ligase activity. Interestingly, the expression of Dda1 and its association with chromatin occurs in a cell-cycle-dependent manner, and the authors speculate that Dda1 might regulate the function of CRL4 complexes that have a role in DNA replication and repair.



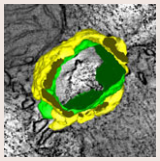
Matrilin 2 hits a nerve

Matrilins 1-4 are extracellular-matrix proteins that are involved in the formation of collagen-dependent and collagen-independent filamentous networks, but their specific functions are mostly unknown. In this study, Dieter Riethmacher and colleagues (p. 995) study matrilin-2-deficient mice (which were previously found to have a normal phenotype) to investigate whether matrilin 2 is important in nervous tissue, in which it is reportedly expressed. They show that matrilin 2 is expressed in mouse Schwann cells (SCs) during development and following peripheral-nerve injury in adult mice. Two types of migration assays reveal that matrilin-2-deficient SCs have defective migration and adhesion abilities in vitro. In addition, they report the remarkable finding that matrilin 2 supports axonal growth in vitro as efficiently as laminin, the most potent known enhancer of axonal growth. In vivo experiments show that, although matrilin 2 is expressed at low levels in adult mice, its expression is induced following peripheral-nerve injury. Furthermore, nerve regeneration following injury was slowed in matrilin-2-deficient mice. These findings characterise a novel SC-derived factor that is important for optimal peripheral-nerve regeneration.



E2-2 factors in EMT

Epithelial-to-mesenchymal transition (EMT) occurs in physiological settings that involve cell migration and invasion, such as embryonic development, wound healing and cancer. Reduced expression of the cell-cell-adhesion protein E-cadherin is an indicator of EMT, and it is known that its repression can be mediated by several transcription factors (including Snail1, Snail2 and E47). On p. 1014, Amparo Cano and colleagues now show that the E-proteins E2-2A and E2-2B (which are related isoforms that are both encoded by the *Tcf4* gene) can also induce EMT. They show that the overexpression of either E2-2A or E2-2B in MDCK cells leads to the acquisition of a mesenchymal phenotype and repression of E-cadherin expression. Furthermore, MDCK cells overexpressing E2-2 factors have increased migratory and invasive behaviour in vitro. However, chromatin immunoprecipitation analysis indicates that, unlike Snail1, E2-2 factors do not bind directly to the gene encoding E-cadherin, suggesting an indirect mechanism of repression. In addition, the gene-expression profile induced by the overexpression of E2-2 factors only partially overlaps with that induced by E47. Therefore, the authors conclude that the role of E2-2 factors is complementary to that of other transcription factors that repress E-cadherin transcription and induce EMT.



Seeing cardiomyocyte signalling in 3D

Cardiomyocytes contain defined membranous structures known as dyadic clefts, where short-lived increases in local Ca^{2+} concentration (known as Ca^{2+} sparks) occur; the accumulation of Ca^{2+} sparks following stimulation regulates excitation-contraction (EC) coupling. Microscopy techniques have been useful for understanding the microarchitecture and signalling events in cardiomyocyte membrane systems, although technical limitations have made it difficult to resolve the three-dimensional (3D) structure of dyadic clefts. On p. 1005, Masahiko Hoshijima and colleagues now show that this is possible by using electron tomography to visualise the 3D structure of dyadic clefts across multiple sarcomeres in adult mouse cardiac muscle. In contrast to previous studies, they show that the size of dyadic clefts is highly variable and that many are smaller ($<1.5\times10^5$ nm^3) than previously reported. In addition, observations of Ca^{2+}-signalling events lead the authors to speculate that the asymmetrically clustered dyadic clefts defined using electron tomography act as 'hot spots' of spontaneous intracellular Ca^{2+} release that stimulate neighbouring Ca^{2+}-releasing sites. This work provides insight into the anatomical basis of Ca^{2+}-signalling events and EC coupling in cardiomyocytes.

Development in press

Planar cell polarity regulates bone growth

During long-bone growth, proliferative chondrocytes in the growth-plate cartilage form clonal columns of discoid cells, which enlarge to form the hypertrophic chondrocytes that make bone – but what regulates column formation and is this columnar organisation crucial for bone morphogenesis? In a paper published in *Development*, Yuwei Li and Andrew Dudley investigate these questions and, for the first time, suggest that a planar cell polarity (PCP)-like pathway is involved in regulating bone morphogenesis. They show that the plane of cell division in proliferative chondrocytes in chick long bones is orthogonal to the direction of growth and that the resultant daughter cells, which are initially displaced laterally, intercalate to form a single column of cells. Both the division plane and orientation of the chondrocytes depend on β-catenin-independent, noncanonical Wnt/frizzled signalling, and the disruption of this signalling pathway produces abnormally short and thick long bones. Thus, by regulating the cell polarity of growth-plate chondrocytes, noncanonical frizzled signalling (probably via a PCP-like pathway) plays a crucial role in bone morphogenesis.

Li, Y. and Dudley, A. T. (2009). Noncanonical frizzled signaling regulates cell polarity of growth plate chondrocytes. *Development* **136**, 1083-1092.