The addition of an ER signal sequence and an ER-retention tag resulted in the D1 cameleon being specifically localized to the ER, and Palmer showed that the D1 cameleon can detect both increases and decreases in $[Ca^{2+}]_{ER}$ with a much greater sensitivity than previous cameleons. Furthermore, she showed that the D1 cameleon and the fluorescent dye fura-2 can be used together to allow the simultaneous monitoring of $[Ca^{2+}]_{ER}$ and $[Ca^{2+}]_{cytosol}$, respectively.

Moving to a biological application for the improved cameleon, Palmer collaborated with Can Jin in the Reed laboratory to study the relationship between Bcl2, $[Ca^{2+}]_{ER}$ and apoptosis in a breast cancer cell line. Over-expressing Bcl2 in these cells, which is known to make them resistant to apoptosis, lowered $[Ca^{2+}]_{ER}$ under resting conditions by increasing Ca^{2+} leakage and altered the Ca^{2+} oscillations that were induced by ATP. In addition, the authors showed that a green-tea compound, epigallocatechin gallate, induced apoptosis in Bcl2-overexpressing breast cancer cells in a dose-dependent manner. Epigallocatechin gallate binds to Bcl2 and increases $[Ca^{2+}]_{ER}$ by blocking Bcl2-mediated leakage. They are now trying to elucidate the exact link between Bcl2 inhibition, increased $[Ca^{2+}]_{ER}$ and apoptosis.

Rachel Smallridge

References and links

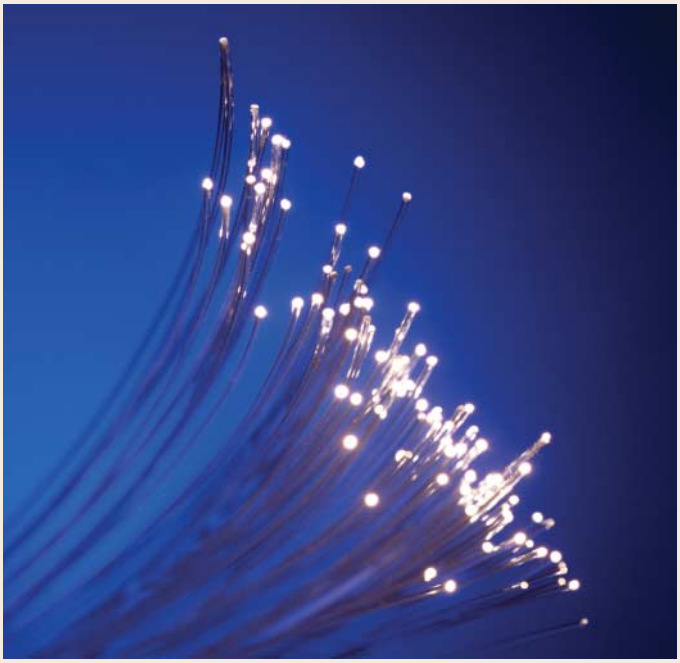
ORIGINAL RESEARCH PAPER Palmer, A. E. *et al.* Bcl-2-mediated alterations in endoplasmic reticulum Ca^{2+} analyzed with an improved genetically encoded fluorescent sensor. *Proc. Natl Acad. Sci. USA* **101**, 17404–17409 (2004)

FURTHER READING Rudolf, R. *et al.* Looking forward to seeing calcium. *Nature Rev. Mol. Cell Biol.* **4**, 579–586 (2003)

WEB SITES

John Reed's laboratory: http://www.burnham.org/FacultyAndResearch/faculty/john_reed_bio.asp

Roger Tsien's laboratory: http://www.tsienlab.ucsd.edu/



nucleus follows. This brings the number of classes of actin-nucleation factor to three.

Katrin Bussell

References and links

ORIGINAL RESEARCH PAPER Quinlan, M. E. *et al.* *Drosophila* Spire is an actin nucleation factor. *Nature* 27 Jan 2005 (doi:10.1038/nature03241)

IN BRIEF

PROTEOMICS

Nucleolar proteome dynamics.

Andersen, J. S. *et al. Nature* **433**, 77–83 (2005)

Using mass-spectrometry-based organellar proteomics and stable-isotope labelling, Mann, Lamond and colleagues quantitatively analysed the proteome of human nucleoli. By monitoring 489 endogenous nucleolar proteins, they showed that the nucleolar proteome varies considerably over time in response to changes in cellular growth conditions. They therefore "…conclude that there is no unique, complete proteome for the nucleolus, or probably for any other organelle, but rather an overlapping set of proteomes that are relevant to different cell states or conditions."

MITOSIS

Stabilization of microtubule dynamics at anaphase onset promotes chromosome segregation.

Higuchi, T. & Uhlmann, F. *Nature* **433**, 171–176 (2005)

Microtubules (MTs) form the bipolar mitotic spindle, which captures chromosomes during metaphase and segregates chromosomes during anaphase. At the onset of anaphase, the highly dynamic MTs become more stable, and the authors showed that the phosphatase Cdc14, which is activated by separase at the onset of anaphase, is required for this. Without Cdc14, MTs remain dynamic, and this interferes with the transport of chromosomes towards the spindle poles and with spindle elongation.

SIGNALLING

EGF receptor signaling regulates pulses of cell delamination from the *Drosophila* ectoderm.

Brodu, V., Elstob, P. R. & Gould, A. P. *Dev. Cell* **7**, 885–895 (2004)

The authors studied the separation of oenocyte precursors from the ectoderm of developing *Drosophila melanogaster* embryos and report an example of oscillatory cell behaviour. The precursors delaminate in discrete, well-separated bursts of three. The epidermal growth factor receptor (EGFR) ligand Spitz specifies the final number of delamination pulses, but has only a permissive role in generating pulses. Before delamination, several EGFR targets, some of which might underlie pulse generation, are switched on in the three cells.

CYTOSKELETON

TSC2 modulates actin cytoskeleton and focal adhesion through TSC1-binding domain and the Rac1 GTPase.

Goncharova, E. *et al. J. Cell Biol.* **167**, 1171–1182 (2004)

Tuberosis sclerosis complex (TSC) protein-1 and -2 regulate protein translation and cell growth, but are also implicated in cell motility. Goncharova *et al.* found that TSC2 regulates stress-fibre disassembly and focal-adhesion remodelling through its TSC1-binding domain by Rac1 activation and subsequent Rho inhibition. The TSC1-binding domain, however, is not needed for TSC2 to regulate the ribosomal protein S6 or DNA synthesis.