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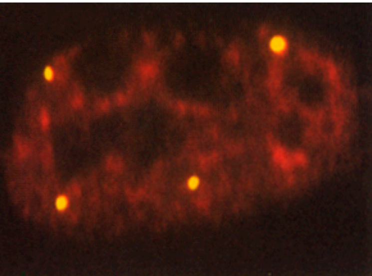
The Hunting of the snRNP

The protein–RNA complexes known as snRNPs are the cell's editors. In 1992, researchers knew from many in vitro studies that different snRNPs band together to form the "spliceosome," which splices premessenger RNA molecules to form functional mRNAs. But they didn't know where in the nucleus snRNPs congregated. Using a new technique, Angus Lamond (now at the University of Dundee, UK) and colleagues tracked snRNPs to two mysterious structures, the interchromatin granules and the Cajal bodies (formerly coiled bodies). Follow-up work suggests that both objects are important for snRNP production and activity.



Foci rich in snRNPs (red) also contain the coiled body protein coilin (green).

When cell biologists first detected snRNPs using immunofluorescence, they observed glowing "speckles" strewn around the nucleus. Although this technique could reveal the presence of snRNPs, the antibodies initially available couldn't determine which kinds of snRNP mustered in a particular location. So Lamond and his team added another ingredient: antisense probes that could discriminate among different snRNPs. The combination of antisense and antibodies revealed that "not all speckles were equal," says Lamond. Some, which the researchers dubbed "nuclear foci," glowed brighter (Carmo-Fonseca et al., 1991a,b).

The next year, Lamond's group confirmed the existence of two kinds of speckles and pinned down their identities (Carmo-Fonseca et al., 1992). Electron microscopy studies from David Spector's group at the Cold Spring Harbor Lab in New York had suggested that antibodies against snRNPs detected the structures known as interchromatin granules. Lamond's group, then at the European Molecular Biology Laboratory, had access to one of the first confocal microscopes, allowing them to analyze combinations of labels and test this possibility. When the team tagged cells with antibodies against snRNPs and against the granules, the staining patterns corresponded, confirming that some speckles were granules.

However, the antigranule antibody didn't cling to the bright foci, suggesting that they were different. The researchers suspected that they were Cajal bodies. Another group at the Scripps Research Institute in California had just discovered antibodies in autoimmune patients that recognize a Cajal body protein called coilin (Raska et al., 1991). When Lamond's lab tagged nuclei with antisense strands and this anticoilin antibody, labeling overlapped, showing that snRNPs were loitering in the Cajal bodies. Huang and Spector (1992) obtained similar results around the same time. These findings "gave us a much higher resolution picture of what was going on [in the nucleus]," says Lamond.

But they didn't explain why snRNPs were parking in the Cajal bodies and interchromatin granules. Sleeman and Lamond (1999) provided one clue by showing that snRNPs that have just entered the nucleus gather in the Cajal bodies. The snRNPs then travel to the interchromatin granules and acquire their finishing touches, maturing into functional particles (Jády et al., 2003). The granules may serve as storage depots for inactive snRNPs, says Lamond, while active snRNPs likely bind to pre-mRNA molecules at the genes themselves. Their dispersal around the nucleus probably accounts for the diffuse glow researchers noted in some labeling experiments, says Lamond. **ML**

Carmo-Fonseca, M., et al. 1991a. *EMBO J.* 10:195–206.

Carmo-Fonseca, M., et al. 1991b. *EMBO J.* 10:1863–1873.

Carmo-Fonseca, M., et al. 1992. *J. Cell Biol.* 117:1–14.

Huang, S., and D.L. Spector. 1992. *Proc. Natl. Acad. Sci. USA.* 89:305–308.

Jády, B.E., et al. 2003. *EMBO J.* 22:1878–1888.

Raska, I., et al. 1991. *Exp. Cell Res.* 195:27–37.

Sleeman, J.E., and A.I. Lamond. 1999. *Curr. Biol.* 9:1065–1074.

ECM determines fate

Biologists long thought that the extracellular matrix (ECM) provided only support and protection for cells. But from the early 1980s on, Mina Bissell of the Lawrence Berkeley Laboratory in California contended that the ECM was a prime influence on cells, transmitting signals that direct gene expression and differentiation (Bissell et al., 1982). A 1991 paper from her lab (Streuli et al., 1991) clinched the case for this view, showing that single mammary cells growing in ECM could fashion a milk protein without stimulation from other cells. Later studies from her group identified the signal-sending component of ECM and revealed how it affected cancer cells.

By 1991, Bissell's lab and others had demonstrated that mammary cells reared on basement membrane, the ECM underlying epithelial layers, form bulbs—just like those that abound in breast tissue and exude milk proteins such as β-casein (Li et al., 1987; Barcellos-Hoff et al., 1989). By contrast, cells nurtured on plastic or collagen did neither, unless they could synthesize and assemble their own basement membrane.

But because cells growing in these cultures were in contact with each other, the possibility remained that cell–cell interactions, not ECM signals, provoked differentiation, Bissell recalls. So instead of growing the mammary cells at high density, her group embedded them in basement membrane gels, leaving many cells with no close neighbors. Most single cells in these gels oozed β-casein, the researchers reported (Streuli et al., 1991), but no single cells embedded in collagen did. By adding antibodies that jammed one of the integrins, the surface receptors that receive messages from the ECM, Bissell slashed β-casein output in the basement membrane gels. But blocking other surface proteins that don't heed the ECM had no effect. "It was the first clear demonstration of the role