DNA METHYLATION

A question of self defence?

DNA methylation and its role in development has been the subject of much debate. Did methylation evolve solely to defend mammalian genomes from transposable elements or is it required for developmentally regulated gene silencing? The jury has been out on this question for several years, but a recent paper in *Nature Genetics* has provided crucial evidence that might help swing the vote towards a role for methylation in development. This study found that the conditional inactivation of *Dnmt1*, which encodes the 'maintenance' DNA methyltransferase 1, in mouse embryonic fibroblasts caused DNA demethylation and the subsequent widespread induction of gene expression — results that strongly indicate that DNA methylation has a role in developmentally regulated gene silencing.

Jackson-Grusby and colleagues turned to mouse embryonic fibroblasts for these studies for several reasons. Cells from *Dnmt1*$^{-/-}$ embryos were of limited use to the authors because they undergo stochastic cell death and incomplete demethylation before the embryos die during early development. Furthermore, differentiated *Dnmt1*$^{-/-}$ embryonic stem cells apoptose, although they proliferate normally while undifferentiated. So to study what causes differentiated cells to apoptose on demethylation and how this affects gene expression, the researchers used embryonic mouse fibroblasts derived from embryos carrying a 'floxed' *Dnmt1* allele. Infecting these cells with a *Cre*-carrying retrovirus created a non-functional *Dnmt1* transcript, and the fibroblasts subsequently underwent demethylation, drastically reduced proliferation and apoptotic cell death.

Reasoning that the cell might interpret DNA demethylation as a sign of DNA damage, the authors next assayed for the effects of inactivating *Trp53* by infecting the fibroblasts with a mutant *Trp53* allele and also by crossing the floxed *Dnmt1* mice to a *Trp53* knockout strain. The results were surprising. In double-mutant fibroblasts, proliferation increased and apoptosis was reduced fourfold. Whether demethylation directly activates *Trp53*-inducing genes or whether *Trp53* is induced indirectly by cell damage remains an open question. But these results strongly indicate that epigenetic deregulation leads to p53-dependent cell-cycle arrest and apoptosis.

Finally, the authors compared the expression profiles of these double-mutant fibroblasts with those of *Trp53* null primary fibroblasts using oligonucleotide microarrays. Of the ~13,000 sequences on this array, around 10% of them were upregulated in the *Dnmt1*-deficient cells — by contrast, only 1–2% of them were downregulated. Known genes that were deregulated include chromatin and silencing factors, transcription factors, DNA-repair proteins, tumour suppressors and oncogenes, signalling-pathway components, and cytoplasmic and nuclear enzymes. And eleven tissue-specific genes were induced, including the testis-specific *Dazl* and the placental-specific *Pl1* genes.

As Andrew Feinberg remarks in an accompanying News and Views article, like most good studies this paper raises more questions than it answers. Dnmt1 is a component of the replication complex — does its loss reduce proliferation through epigenetic deregulation or by destabilizing the complex? Is the widespread induction of gene expression a primary or secondary effect of demethylation? The jury is still out on these questions, but they can rest assured that more evidence will soon come rolling in.

Jane Alfred

References and links

ORIGINAL RESEARCH PAPER Jackson-Grusby, L. *et al.* Loss of genomic methylation causes p53-dependent apoptosis and epigenetic deregulation. *Nature Genet.* **27**, 31–39 (2001)

FURTHER READING Feinberg, A. Methylation meets genomics. *Nature Genet.* **27**, 9–10 (2001)

WEB SITE *Dnmt1* microarray dataset

IN THE NEWS

Cloning OK in UK

On 19 December 2000, the UK Parliament approved laws that will allow scientists to create embryos cloned from human cells, and keep them alive for 14 days to extract stem cells for therapeutic purposes. The proposal, backed by a majority of more than two to one, is very controversial. For example:

"HUMAN CLONES NEXT?"
The New York Times, US

This is the crux of the issue, whether cell-based treatments will offer cures for scores of diseases, ranging from Parkinson, diabetes and stroke, or whether we face a slippery slope towards human cloning.

"Ian Wilmut, the 'father' of the first cloned sheep, Dolly, is convinced [that tissues derived from stem cells could be used as 'spare parts' to save thousands of human lives], and was among the first scientists to congratulate the Blair government."
Il Giorno, Italy

Not all were pleased: "Pope John Paul II has judged this law to be 'morally unacceptable'."
***La Repubblica*, Italy**

"The [German] minister for research, Edelgard Bulmahn, said Germany would not breach this 'ethical boundary'. Instead, the country should focus on exploring alternatives to the cloning of human embryos."
Frankfurter Allgemeine, Germany

"The move would legally endorse human cloning 'for the first time in the world'."
***The Guardian*, UK**

However,
"Mike Dexter, director of the Wellcome Trust research charity, said: 'This is a vote for science, for health, and for the future.' "
The Guardian, UK

Tanita Casci