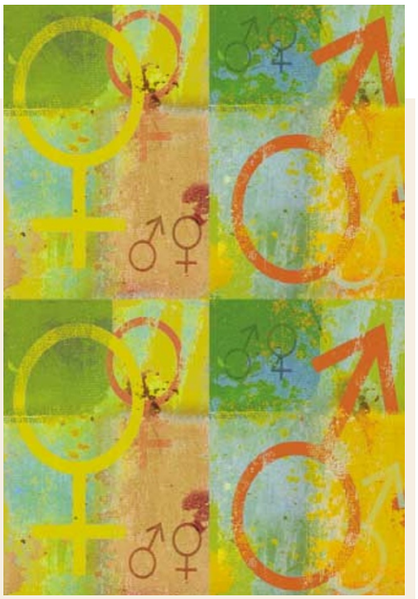
EPIGENETICS

Gender-dependent tumour suppression



As its name suggests, the *HIC1* (hypermethylated in cancer 1) gene is hypermethylated and hence transcriptionally silent in several types of cancer, indicating that it might act as a tumour suppressor. Now, Stephen Baylin and colleagues have developed a mouse model to further investigate this possibility, and show that complete loss of *Hic1* predisposes mice to a gender-dependent array of tumour types.

The authors generated *Hic1*$^{+/-}$ mice and investigated the incidence of tumour formation. The heterozygous mice began to develop tumours at 70 weeks, and by 100 weeks 34.2% had developed malignant tumours, compared with 14.2% of the wild-type mice. Surprisingly, the tumour types that were identified differed according to the sex of the mice — male heterozygotes developed carcinomas at an increased frequency, whereas female heterozygotes mostly developed lymphomas and sarcomas. The tumours that developed in *Hic1* heterozygotes were also more aggressive than those that developed in wild-type mice.

So, was the remaining wild-type allele lost in these cancers? Gross chromosomal deletions were not observed, so the authors investigated the methylation status of the *Hic1* promoter. As in humans, *Hic1* has two alternative promoters, 1a and 1b; it is 1b that seems to be hypermethylated in most human cancers. In mice, however, methylation-specific polymerase chain reaction revealed that the 1a promoter is predominantly methylated in most lymphomas and sarcomas. All carcinomas from heterozygous mice that do not have a methylated 1a promoter were found to contain dense methylation of the 1b promoter, and, in total, 87% of malignant tumours from the heterozygous mice had dense methylation of one of the alternative promoters. This promoter methylation corresponded with a lack of Hic1 protein, as it does in human cells.

HIC1 is therefore the first candidate tumour suppressor that is transcriptionally silenced, rather than mutated, and that acts as a tumour suppressor in mice. The identification of other such genes, and their confirmation using mouse models, will no doubt follow.

Emma Greenwood

References and links

ORIGINAL RESEARCH PAPER Chen, W. Y. *et al.* Heterozygous disruption of *Hic1* predisposes mice to a gender-dependent distribution of malignant tumors. *Nature Genet.* **33**, 197–202 (2003)

WEB SITE

Stephen Baylin's lab: http://www.hopkinsmedicine.org/graduateprograms/cmm/baylin.html

EPIGENETICS

Breaking the silence

Epigenetic modifications, such as DNA hypermethylation and histone deacetylation, are commonly detected in human cancer cells, where they have been shown to downregulate tumour-suppressor genes. Sidransky and colleagues have been using a combination of techniques to identify additional genes that are silenced in this way, and have subsequently uncovered three new tumour suppressors.

Sidransky's group treated oesophageal squamous-cell carcinoma (ESCC) cells with pharmacological agents to prevent the epigenetic silencing of genes — cells were treated with 5-aza-2′-deoxycytidine to block DNA methylation, and then with trichostatin A to inhibit histone deacetylase. This treatment reverses the formation of transcriptionally repressive chromatin structures at methylated promoters. Microarray analysis was then used to compare gene-expression patterns between transcriptionally silenced and unsilenced cancer cells. A total of 58 silenced genes were identified by this approach: 44 (76%) were found to contain dense CpG islands — well-known methylation sites — in their promoter regions. Some 13 of 22 tested gene promoters were methylated in the ESCC cell lines, and 10 of these genes were found to be downregulated at the mRNA level.

Three of the genes — *CRIP1*, *APOD* and *NMU* — had growth-suppressive activity when transfected into ESCC cells, as determined using colony-formation assays. NMU is proposed to be involved in normal oesophageal mucosa integrity, and its receptor, FM3, is a G-protein-coupled receptor that can signal through phosphatidylinositol 3-kinase-γ — recently shown to block the growth of human colon cancer cells. CRIP1 is believed to be a transcription factor that was shown to induce apoptosis in cancer cell lines, and APOD has been associated with cell growth arrest.

The epigenetically silenced genes were found to cluster in specific chromosomal regions. Many of these loci — such as 3q26, 4q12 and 14q24 — have also been shown to harbour chromosome deletions of loss of heterozygosity in cancer cells. This approach could therefore be used to identify new tumour-suppressor genes and loci in other types of cancer cell.

Kristine Novak

References and links

ORIGINAL RESEARCH PAPER Yamashita, K. *et al.* Pharmacological unmasking of epigenetically silenced tumor suppressor genes in esophageal squamous cell carcinoma. *Cancer Cell* **2**, 485–495 (2002)

FURTHER READING Sidransky, D. Emerging molecular markers of cancer. *Nature Rev. Cancer* **2**, 210–219 (2002)

WEB SITE

David Sidransky's lab: http://urology.jhu.edu/faculty/sidransky/