TUMOUR SUPPRESSORS

A new class?

RIZ1 — a suspected protein methyltransferase — has long been thought to possess tumour-suppressor activity: its gene maps to a region that is frequently deleted in human cancers, and reintroducing the gene into tumours causes apoptosis and growth suppression in animal tumour models. Definitive proof has, however, not been available until now. Steele-Perkins *et al.*, in the September issue of *Genes & Development*, show that mice deleted for *Riz1* develop a range of tumours, and that several human tumours contain mutations in *RIZ1*. RIZ1 is therefore the founding member of a new class of tumour suppressors.

RIZ1 was initially isolated as an RB-binding protein and a transcriptional co-activator. It possesses two important motifs — an RB-binding motif and a nuclear hormone receptor binding motif — in addition to the PR domain, which places it within the PR subfamily of protein methyltransferases. RIZ1's substrates are a complete mystery, but the gene is deleted in many human cancers, and mRNA expression is also decreased or lost in many more, probably by epigenetic silencing. So, is RIZ1 a tumour suppressor?

Riz1$^{-/-}$ mice developed normally, but had a shorter lifespan than *Riz1*$^{+/-}$ and *Riz1*$^{+/+}$ mice, owing to an increase in tumour burden. In fact, 80% of *Riz1*$^{-/-}$ mice had tumours at 20 months (compared with 30% of *Riz1*$^{+/+}$ mice), and almost half of these had tumours in several organs. The predominant tumour type in *Riz1*$^{-/-}$ mice is malignant B-cell lymphoma, with a histology similar to diffuse large B-cell lymphoma (DLBL). Mutation of *Riz1* in a *Trp53*$^{+/-}$ background also increased tumour formation compared with *Riz1*$^{-/-}$ and *Trp53*$^{+/-}$ mice, indicating a cooperative effect on tumorigenesis and confirming that Riz1 acts as a tumour suppressor in mice.

So, does RIZ1 also participate in tumorigenesis in humans? Single-strand conformation polymorphism analysis of *RIZ1* in 35 human DLBL tumours revealed that ten of these had an abnormal band, corresponding to a single base substitution, C-terminal to the PR domain. Not one of the other 432 tumours tested had a mutation in this region.

The authors also found other mutations in this region of *RIZ1*, either within or C-terminal to the PR domain. Using its ability to stimulate oestrogen-receptor function in activating target gene promoters as a functional assay, the authors showed that none of the *RIZ1* mutations discussed were active.

RIZ1 therefore represents the first in a new class of tumour suppressors — the identification of many more will surely follow. Discovery of RIZ1's substrates should elucidate how this class acts.

Emma Greenwood

References and links

ORIGINAL RESEARCH PAPER Steele-Perkins, G. *et al*. Tumor formation and inactivation of RIZ1, an Rb-binding member of a nuclear protein–methyltransferase superfamily. *Genes Dev.* **15**, 2250–2262 (2001)

WEB SITE

Shi Huang's lab: www.burnham.org/reports/6.Huang.97.html

METASTASIS

Peering into a black box

The molecular determinants of metastasis are one of the great unsolved mysteries of cancer research. But expression analysis using microarrays is exposing some of the daemons that lurk within this black box. Tobey MacDonald and colleagues, reporting in the October issue of *Nature Genetics*, have used microarrays to identify consistent changes in gene expression in metastatic medulloblastoma — a brain cancer that most commonly affects children. The genes identified leave a remarkably consistent trail that might lead down some new therapeutic avenues.

The authors used some statistical methods that are fast becoming the gold standard for the molecular characterization of cancer. Having measured the expression levels of over 1900 transcripts for 10 metastatic and 13 non-metastatic medulloblastoma samples using commercially available oligonucleotide microarrays, they identified 85 genes — the predictor set — that differed significantly between metastatic and non-metastatic tumours. To determine the ability of these genes to categorize a tumour as metastatic or non-metastatic, the expression profile for each tumour was compared against those of the other 22 — a technique known as the 'leave one out' approach. The predictor set got it right for 90% of the non-metastatic tumours, but only for 50% of the metastatic ones, leading the authors to speculate that there might be subsets with different expression profiles within the metastatic group. Nevertheless, it correctly predicted the outcomes for four of another five blinded tumour samples, but categorized only one of three metastatic cell lines — the Daoy cell line — as metastatic.

But more exciting than the predictor's mixed successes was the finding that many of the genes that are overexpressed in the metastatic group are in the same growth-factor signalling pathway: the platelet-derived growth factor receptor PDGFRA was over-expressed in 85% of metastatic, but no non-metastatic tumours. Migration assays revealed that PDGFA (PDGFRA's ligand) stimulated movement of Daoy cells on fibronectin matrices, and this was blocked by neutralizing antibodies to PDGFRA. Activation of PDGFRA increased the phosphorylation of the mitogen-activated protein kinases MAPK1 and MAPK3, and their upstream kinases, MAPKK1 and MAPKK2. The antibodies blocked this increase in MAPK signalling, and pretreatment with a specific inhibitor of these MAPKKs, U0126, blocked PDGFA-mediated migration and MAPK activation in these cells.

Children with medulloblastoma are given radiotherapy to prevent or treat metastasis, but this has serious neurological consequences. Identifying individuals at low risk of metastasis would allow clinicians to tailor this aggressive therapy to children for whom the benefits would outweigh the risks. The prospect of treating metastatic medulloblatoma with tyrosine kinase inhibitors such as STI-571 (Gleevec), which inhibits PDGFRA's kinase activity as well as ABL's, is another potential outcome of this work. Further analysis of gene-expression patterns in metastatic medulloblastoma will doubtless improve the predictor set, perhaps yielding further promising therapeutic targets.

Cath Brooksbank

References and links

ORIGINAL RESEARCH PAPER MacDonald, T. J. *et al*. *Nature Genet.* **29**, 143–152 (2001)

FURTHER READING Quackenbush, J. Computational analysis of microarray data. *Nature Rev. Genet.* **2**, 418–427 (2001)

WEB SITE

Children's National Medical Center Microarray Center: http://microarray.cnmcresearch.org/microarray.html