WEB WATCH

Face lift for mouse models

• http://emice.nci.nih.gov/

Mouse models are frequently used by cancer researchers, as they successfully recapitulate many aspects of human cancer. But with the number of models now on offer, how do researchers find out which models are available and most appropriate for their work? Enter the Mouse Models of Human Cancers Consortium (MMHCC) web site, stage right.

The MMHCC web site, hosted by the National Cancer Institute, has recently been given a major overhaul. It has merged with the private emice site, and some sections of the web site remain secure. So, does this new and improved site achieve all it sets out to?

The home page links through to seven sections, the most useful of which should prove to be 'Mouse models'. Much of this section is still under construction, but its potential is apparent. The models are arranged under subheadings that include organ type, genes, pathways and therapeutics, which should greatly facilitate the ease with which researchers can choose the best model for them. Organ types is the most complete section — it provides details on models for 12 organ sites. The Resources section complements this as — among other useful tips — it includes protocols that are relevant for work on specific organ sites.

Learning tools (which aims to "provide 'how to' information for all aspects of mouse model engineering") and Applications (which will include comparisons of human and mouse cancers and details on clinical trials) hold much promise but, again, are far from complete. The site should prove immensely valuable to researchers, so let's hope these sections are completed soon.

Emma Greenwood

ONCOGENES

Breaking and entering

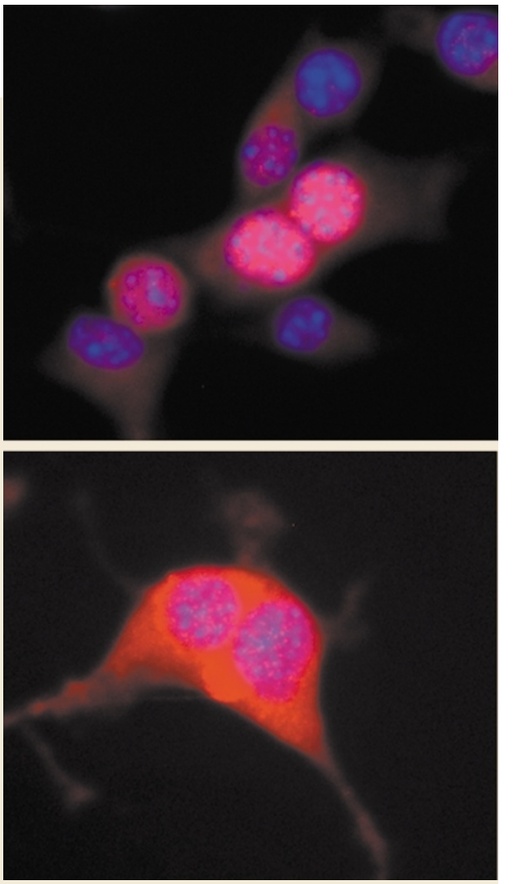
Avoiding cell death is something that many cancer cells are experts at — and they don't leave anything to chance. A common mechanism for dodging death is stimulation of the phosphatidylinositol 3-kinase (PI3K) pathway. This activates the serine/threonine kinase AKT which, in turn, switches on a slew of cellular survival mechanisms. Two papers, in *Nature Cell Biology* and *Proceedings of the National Academy of Sciences*, add a new tool to the AKT survival kit: inactivation of p53.

Binhua Zhou and colleagues wanted to know why cells that overexpress ERBB2 (also known as HER-2 or Neu), an oncogenic receptor tyrosine kinase that is often overexpressed in breast cancer, are resistant to DNA-damaging drugs such as etoposide. They suspected that AKT might be involved because it can confer resistance to other pro-apoptotic agents. They confirmed their suspicions by transfecting NIH-3T3 cells overexpressing ERBB2 — which are etoposide resistant — with a dominant-negative mutant of AKT (DN-AKT). This sensitized the cells to etoposide-mediated apoptosis, both *in vitro* and in nude mice harbouring tumours derived from these cells. This sensitization was dependent on p53: $p53^{-/-}$ cells could not be made sensitive to etoposide by blocking the AKT pathway.

Was etoposide inducing p53 in the DN-AKT-expressing cells? Surprisingly, these cells had high levels of p53 even in the absence of etoposide, leading Zhou and colleagues to suspect that ubiquitin-mediated p53 degradation, which is catalysed by MDM2 and usually keeps p53 levels low in unstressed cells, was blocked. Immunoprecipitations revealed that the levels of ubiquitin, and of MDM2, associated with p53 were much higher in cells expressing ERBB2 alone than in those expressing ERBB2 and DN-AKT. So AKT, activated by ERBB2, is somehow increasing MDM2's ability to dispose of p53.

Is the effect of AKT on MDM2 direct? Lindsey Mayo and David Donner, as well as Zhou and colleagues, discovered two AKT phosphorylation sites, centred around serines 166 and 186, in MDM2. Mayo and Donner could immunoprecipitate a complex containing MDM2 and AKT from serum-starved MCF-7 cells (a breast-cancer-derived cell line), and insulin-like growth factor 1 (IGF-1), which activates PI3K, caused dissociation of this complex. Similarly, Zhou *et al.* found more of the complex in DN-AKT-expressing cells than in cells expressing ERBB2 alone. Both groups found that phosphorylation of MDM2 on residues 166 and 186 depended on activation of AKT.

The position of the phosphorylation sites — close to MDM2's nuclear localization sequence — provided a clue as to what might be achieved by their phosphorylation. It is widely believed that MDM2 needs to enter the nucleus to ubiquitylate p53. Could AKT-mediated phosphorylation open the door to the nucleus? Both groups showed that transfection of their chosen systems with a constitutively active form of AKT (CA-AKT) caused MDM2 to relocalize to the nucleus.



Localization of MDM2 (red) in NIH 3T3 cells expressing ERBB2 alone (top) or ERBB2 plus DN-AKT (bottom). Nuclei are stained blue. Image reproduced with permission from *Nature Cell Biology* © (2001) Macmillan Magazines Ltd.

Treating MCF-7 cells with IGF-1 achieved the same end, and this could be blocked with a PI3K inhibitor. Similarly, MDM2 was constitutively nuclear in ERBB2-expressing cells, but not when DN-AKT was expressed (see picture). Further evidence that phosphorylation of serines 166 and 186 is important for nuclear entry of MDM2 came from mutating these residues: Mayo and Donner mutated both residues to alanine, yielding a form of MDM2 that couldn't get into the nucleus. Zhou and colleagues mimicked phosphorylation by mutating both residues to aspartate, producing an MDM2 that resided exclusively in the nucleus.

But is this mechanism operative in human cancer? Zhou and colleagues found that MDM2 was nuclear in tumour samples in which ERBB2 was overexpressed or AKT was activated, so nuclear shuttling of MDM2 could be causing resistance to DNA-damaging agents in p53 wild-type breast tumours. If AKT could be blocked in these tumours, we might have a chance of resensitizing them.

Cath Brooksbank

References and links

ORIGINAL RESEARCH PAPER Mayo, L. D. & Donner, D. B. A phosphatidylinositol 3-kinase/Akt pathway promotes translocatiion of Mdm2 from the cytoplasm to the nucleus. *Proc. Natl Acad. Sci. USA* **98**, 11598–11603 (2001) | Zhou, B. P. *et al.* HER-2/neu induces p53 ubiquitination via Akt-mediated MDM2 phosphorylation. *Nature Cell Biol.* **3**, 972–982 (2001)

FURTHER READING

Testa, J. R. & Bellacosa, A. AKT plays a central role in tumorigenesis. *Proc. Natl Acad. Sci. USA* **98**, 10983–10985 (2001) | Scheid, M. P. & Woodgett, J. P. PKB/AKT: functional insights from genetic models. *Nature Rev. Mol. Cell Biol.* **2**, 760–768 (2001)