WEB WATCH

• http://npd.hgu.mrc.ac.uk/

Centre of attention

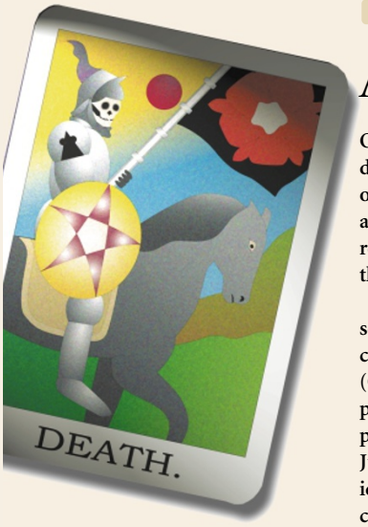
Proteins that localize to the nucleus are the centre of attention in the Nuclear Protein Database (NPD). This searchable database contains information on more than 1,000 vertebrate nuclear proteins, mainly from mice and humans.

The web site was an initiative undertaken by Wendy Bickmore's Group at the MRC-Human Genetics Unit, Edinburgh, United Kingdom, with the aim of making data available on new nuclear proteins. However, it soon became clear that such a database would be valuable to the entire community, so it was expanded to include published data on nuclear proteins. Knowledge of the subnuclear localization of proteins can be important in understanding the regulation and function of the genome, and can also provide clues to protein function.

You can search the entire database, or you can browse by subnuclear compartment or by domain/motif. The compartment browsing option takes you to a visually appealing, 'clickable' graphic of the nucleus, and gives you information on each compartment and the associated proteins. There are also useful links to nuclear structure and function resources, other nuclear protein databases and bioinformatics resources.

For each protein, the subnuclear compartment is reported, if known, together with information on the isoelectric point, protein size and sequence (including any repeats, motifs or domains). Protein functions are described using Gene Ontology™ terms, and links to other databases — for example, SwissProt and PubMed — are included where possible. In the future, the Bickmore Group would like to include more data from other groups, and hope to develop partnerships with other database projects.

Rachel Smallridge



APOPTOSIS

A marked CARD

One characteristic of apoptotic cell death is the extensive fragmentation of nuclear DNA, which depends on a DNase called CAD. According to a report in *Current Biology*, however, this might not be the whole story.

The twist in the tale began with a screen for proteins that contain the caspase-recruitment domain (CARD), which mediates protein–protein interactions in pro-apoptotic signalling pathways. Jürg Tschopp and colleagues identified a new protein that contains two amino-terminal CARD domains and a predicted helicase domain, hence its name — Helicard.

Transient transfection of 293T cells with Helicard led to the expression of full-length Helicard, but also to the appearance of a 45-kDa fragment. Then, when FasL was used to induce apoptosis in these cells, two further fragments were generated. The authors calculated that these processing events occurred in the region between the CARD and helicase domains.

Confocal microscopy showed that, on cleavage of Helicard in apoptotic cells, the CARD-containing fragment remains in the cytoplasm, whereas the helicase

APOPTOSIS

Getting rid of obstacles

The activation of caspases — which are essential effectors of apoptosis — is regulated by IAP ('inhibitor of apoptosis') proteins. In *Drosophila*, cell death and caspase activation require proteins of the Reaper, Hid, Grim and Sickle family (referred to here as RHG proteins). A series of reports published in *Nature Cell Biology* now sheds light on the relationship between these essential components of the apoptotic pathway.

The reports show that regulation of the levels of DIAP1 (*Drosophila* IAP1) is important for the initiation of apoptosis by RHG proteins *in vivo*. All six groups saw that RHG proteins reduce DIAP1 levels. In mammals, it has been reported that IAPs can be degraded by the proteasome after treatment with an apoptotic stimulus. Also, the RING-finger domain of IAPs has E3 ligase activity, which allows the polymerization of polyubiquitin chains on target proteins, and thereby targets them for degradation by the proteasome.

So, each group checked whether RHG proteins could induce ubiquitylation of DIAP1 and its degradation by the proteasome. They found that RHG proteins can indeed induce the autoubiquitylation of DIAP1 and its degradation in a proteasome-dependent manner. This requires the presence of the RING-finger domain of DIAP1, which is also essential for RHG-induced killing.

Ubiquitylation of a protein substrate requires three successive enzymatic reactions that are controlled by an E1 ubiquitin-activating enzyme, an E2 ubiquitin-conjugating enzyme and an E3 ubiquitin protein ligase. As mentioned before, DIAP1 has E3-ligase activity, and Hermann Steller's group identified an E2 enzyme, UBCD1, that is required for Reaper- and Grim-induced DIAP1 ubiquitylation and degradation, and for Reaper- and Grim-induced killing. The groups of Ross Cagan and John Nambu identified another protein, Morgue, that is required for the ubiquitin-conjugating reaction. Morgue is related to E2 enzymes, yet lacks the consensus active site, so it could work in association with another E2 enzyme. John Nambu's lab provides preliminary evidence that it might bind SkpA, a component of the SCF–E3 ubiquitin ligase complex.

Sally Kornbluth and Bruce Hay made another interesting observation: they saw that RHG proteins could reduce levels of general protein translation, which would contribute further to reducing DIAP1 levels. And finally, Pascal Meier's lab found that DIAP1 also induces the ubiquitylation of the *Drosophila* caspase Dronc and