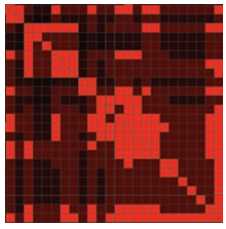
hOGG tied

DNA glycosylases function in DNA repair by recognizing and removing damaged DNA bases; human 8-oxoguanine DNA glycosylase (hOGG1) is specific for 8-oxoguanine lesions. Structural studies have shown that these enzymes swivel their target base out from the DNA duplex, with the base pausing at an "exosite" next to the active site before making its way into the active site; however, structures that reveal how the base transits from the exosite to the active site have remained elusive. Lee *et al.* surmised that the addition of a bulky photocleavable group to 8-oxoguanine would prevent the insertion of the nucleotide into the active site until photolysis, thus making it possible to capture the transit of oxoguanine from the exosite to the active site. The development of a straightforward synthesis for a photocaged 8-oxoguanine and its incorporation into DNA led to a crystal structure of the unirradiated complex in which the modified base was not resolved, whereas a brief laser pulse followed by cryotrapping successfully captured a complex with 8-oxoguanine inserted into the active site. Interestingly, the structure shows that, although the nucleotide has inserted into the active site and formed one key hydrogen bond between the N7 hydrogen and the protein, the remainder of the network of active site interactions necessary for catalysis has not yet formed. Since this N7 hydrogen is not present in guanine itself, the structure may represent the last biochemical checkpoint prior to base excision. (*J. Am. Chem. Soc.*, published online 29 May 2008, doi:10.1021/ja800821t) *CG*

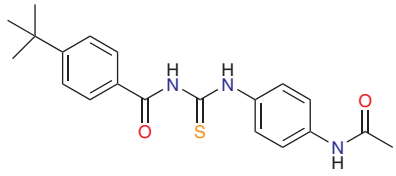
PINpointing *in vivo* interactions



Identifying protein-protein interaction networks (PINs) provides a critical foundation for understanding biological systems. Some previous efforts to catalog PINs have involved yeast two-hybrid screens, which detect direct protein-protein interactions, but only those that can be translocated to the nucleus. Tandem affinity purification followed by mass spectrometry has also been used to identify components of protein complexes, but this can only detect complexes that are stable to the purification conditions. Protein-fragment complementation assays (PCAs) are an alternative method in which an N- or C-terminal fragment of a reporter protein is attached to a 'bait' protein and the complementary fragment to the potential 'prey'. If the bait and prey proteins interact, then the reporter protein folds from the fragments, becomes functional and provides a readout. Using a growth-based PCA, Tarassov *et al.* screened 15 million combinations of proteins, covering 93% of yeast open reading frames, to characterize the yeast PIN *in vivo* under endogenous conditions. With this method, the authors report 2,770 interactions among 1,124 proteins. Although this new glimpse at the yeast interactome contained many known interactions, 80% of the results were previously unreported protein-protein interactions and included weaker and more transient connections. This approach can now be used to investigate the remodeling of PINs in response to different perturbations, which will provide an important tool for understanding the plasticity of cellular pathways. (*Science*, published online 8 May 2008, doi:10.1126/science.1153878) *JK*

Written by Amy Donner, Catherine Goodman, Joanne Kotz & Terry L. Sheppard

p53 activation for sirtuin



p53's function as a tumor suppressor protein is deficient in numerous cancers. Thus, approaches that activate p53 function offer potential for treatment of diverse cancer types. Lain *et al.* now report the identification of a class of small molecules, called tenovins, which enhance p53 function in mammalian cells by targeting sirtuin proteins. The authors performed a cell-based screen of 30,000 'drug-like' molecules, searching for compounds that activate p53-dependent transcription. The screen identified tenovin-1, an aromatic thiourea compound that inhibited cell growth in numerous cancer cell lines. Subsequent structure-activity relationship studies identified more water-soluble analogs, including tenovin-6. The antiproliferative effects of the tenovins were enhanced by, but did not require, functional p53, which prompted the authors to search for potential tenovin targets that were coupled to p53 action. A haploinsufficiency screen implicated the histone deacetylase Sir2p as a target in yeast. Subsequent *in vitro* assays demonstrated that tenovins inhibit the protein deacetylase activities of purified human sirtuins SirT1 and SirT2. In human cells, tenovins blocked deacetylation of SirT1 substrates, including p53, and of SirT2 substrates such as α-tubulin. Since acetylation of p53 is essential for its function as a transcription factor, it is very likely that tenovins enhance p53-dependent transcription in cells through SirT1 inhibition. Although additional studies are required to understand the mode of tenovin binding to sirtuins, this new class of p53 activators offers potential probes of p53 biology and leads for the development of chemotherapeutic agents. (*Cancer Cell* **13**, 454–463, 2008) *TLS*

MicroRNAs on TOP of translation

MicroRNAs (miRNAs) are short noncoding RNAs that often regulate gene expression by moderating transcript stability or repressing translation through direct association with the 3′ untranslated region (UTR). miRNA dysfunction correlates with some human cancers, but there is little to no mechanistic understanding of these observations. Ørom *et al.* used a novel affinity-based target purification protocol to identify ribosomal protein mRNAs as targets for the mir-10a miRNA in mouse embryonic stem cells. Surprisingly, the 3′ UTRs of these putative targets did not impose mir-10a sensitivity on transcripts. However, introduction of mir-10a in embryonic stem cells increased the amount of newly synthesized ribosomal proteins. Ribosomal protein mRNAs contain a 5′ TOP motif in the 5′ UTR that promotes their translation during amino acid deprivation. The authors demonstrated that mir-10a associates directly with these transcripts adjacent to the 5′ TOP sequence and promotes ribosome association during amino acid starvation. Enhanced ribosomal protein translation was dependent on the integrity of both the identified mir-10a binding site and the 5′ TOP sequence. The introduction of mir-10a into Ras-transformed cells augmented both global translation and colony formation in soft agar, whereas repression of mir-10a decreased both, thus providing a possible explanation for miRNA-dependent cellular transformation and cancer progression. (*Mol. Cell* **30**, 460–471, 2008) *AD*