TOOLS IN BRIEF

BIOCHEMISTRY

Capturing ubiquitin-binding interactions

Multiple cellular processes are influenced by ubiquitination, the covalent attachment of chains of ubiquitin molecules to proteins. Because ubiquitination is so prevalent in cells, and because different ubiquitin chain structures result in different cellular signals, this post-translational modification is challenging to study. Chojnacki *et al.* describe Ub-phototrap tools as an approach to detect ubiquitin-binding interactions. The Ub-phototrap is a ubiquitin variant containing a photoactivatable crosslinker as an unnatural side chain, allowing ubiquitin binding domains to be captured and then identified. Chojnacki *et al.* created a series of Ub-phototrap reagents with distinct linkages and lengths. Applying these tools to the 26S proteasome complex, the researchers trapped Rpn10 and Rpn13, known ubiquitin-binding domains, and Rpn1, which was previously unknown. They followed up with NMR experiments to identify the binding region.

Chojnacki, M. *et al. Cell Chem. Biol.* doi:10.1016/j.chembiol.2017.02.013 (2017).

CELL BIOLOGY

Optogenetic protein regulation at near-infrared wavelengths

Various optogenetic tools are available to control proteins. The BphP1–PpsR2 system is sensitive to near-infrared light and therefore suited for *in vivo* applications (Kaberniuk *et al.*, 2016). However, PpsR2 is a large protein and susceptible to oligomerization. Redchuk *et al.* report an improved version of the BphP1–PpsR2 system, in which RpsR2 is replaced by Q-PAS1, a version lacking the domains responsible for oligomerization (Redchuk *et al.*, 2017). When used to control transcription of a reporter gene in a light-dependent manner, the updated BphP1–Q-PAS1 system works as efficiently as the original. The researchers demonstrated the performance of the BphP1–Q-PAS1 pair in a variety of applications in cell culture, including light-dependent regulation of the epigenetic state, protein sequestration to the plasma membrane and recruitment of proteins to the nucleus. Furthermore, the near-infrared-controlled BphP1–Q-PAS1 system can be combined with the blue-sensitive LightON system for multiplexed protein regulation.

Redchuk, T.A. *et al. Nat. Chem. Biol.* doi:10.1038/nchembio.2343 (2017); Kaberniuk, A.A. *et al. Nat. Methods* **13**, 591–597 (2016).

SENSORS AND PROBES

Brighter and redder fluorescent rhodopsins

Near-infrared fluorescent proteins (NIFPs) have long excitation and emission wavelengths compatible with imaging deeply into tissues. However, despite substantial progress in discovering and engineering such tools, the photophysical properties of NIFPs still stand to be improved. Herwig *et al.* developed brighter, red-shifted variants of Archaerhodopsin-3 (Arch-3). Arch-3 is a microbial rhodopsin that emits red fluorescence when bound to its natural chromophore. The team substituted the natural chromophore for a derivative, which led to an ~200-nm red shift in fluorescence emission. They also introduced mutations to increase fluorescence brightness. The final tools excite and emit around 760 and 775 nm, respectively, which make them the reddest available NIFPs. They were shown to be brightly fluorescent in *Escherichia coli*.

Herwig, L. *et al. Cell Chem.Biol.* **24**, 415–425 (2017).

NEUROSCIENCE

Light-dependent inhibition of CaMKII

Ca^{2+}/calmodulin kinase II (CaMKII) plays a central role in synaptic plasticity. While the temporal requirements for CaMKII activation have been addressed by pharmacological inhibition, such measurements are not precise, as delays occur during bath application of these compounds. Murakoshi *et al.* report a genetically encoded and light-dependent inhibitor for CaMKII. The researchers fused an inhibitory peptide, the autocamtide inhibitory peptide 2 (AIP2), to LOV2-Jα, a domain that changes its conformation from closed to open in response to blue light, thus creating a photoactivatable CaMKII inhibitor. They applied this inhibitor, called paAIP2, to determine the precise temporal window in which CaMKII is required to exert its function during long-term potentiation in rat hippocampal slice preparations and in mice during a behavioral task.

Murakoshi, H. *et al. Neuron* **94**, 37–47 (2017).