METHODS IN BRIEF

PROTEOMICS

Proteome changes upon infection

Human cytomegalovirus (HCMV) is a common human pathogen that can persist for the lifetime of an infected individual because of its ability to evade immune response. To study how the virus achieves this, Weekes *et al*. used quantitative temporal viromics, involving a multiplexed tandem-mass-tag–based mass spectrometry method for following viral and host-cell protein production over time. The researchers profiled cytoplasmic proteins from whole-cell lysates as well as plasma-membrane proteins in HCMV-infected fibroblasts over a three-day time course. The proteomic data reveal the dynamics of ~80% of viral proteins and over 8,000 host proteins during infection, identifying host factors that may play a role in viral defense and that, as candidate targets of HCMV, may permit immune evasion.

Weekes, M.P. *et al. Cell* **157**, 1460–1472 (2014).

CHEMICAL BIOLOGY

Two peptides from one mRNA

Genetic code reprogramming methods enable researchers to take control of protein translation, allowing site-specific labeling of proteins with designer amino acids. Applications range from a variety of basic biological experiments to the synthesis of new types of polymer materials. In new work, Terasaka *et al.* focused on engineering the translation machinery itself. Knowing that Watson-Crick base-pairing between the 3' end of tRNA and rRNA in the peptidyl transferase center is conserved, the researchers introduced pairs of complementary mutations into *Escherichia coli* tRNA and rRNA. This resulted in the generation of an engineered translation system that, along with the native translation system, could in parallel produce two different peptide sequences from a single mRNA template.

Terasaka, N. *et al. Nat. Chem. Biol.* **10**, 555–557 (2014).

IMAGING

Keeping an eye on the retina

The transparency of the vertebrate eye to infrared light makes it possible to image the retina at the back of the eye with two-photon microscopy. To counteract aberrations introduced by the anterior part of the eye, Palczewska *et al.* increased imaging quality and efficiency with the help of adaptive optics, a short-pulsed laser and highly sensitive detectors. With these improvements, the researchers could visualize and spectrally characterize intrinsically fluorescent products of the visual chromophore regeneration pathway, even in living mice. Depending on the excitation wavelength, different fluorophores can be detected. This technique will be helpful for analyzing the impact of drug therapies, disorders affecting the retina and environmental stress on the biochemical processes in the eye.

Palczewska, G. *et al. Nat. Med.* **20**, 785–789 (2014).

SINGLE MOLECULE

Four-color FRET follows Hsp90 states

Single-molecule fluorescence resonance energy transfer (smFRET) is an important tool in structural biology, used to report the proximity of two probes whose fluorescence readout is modulated by distance. Ratzke *et al*. now expand the use of smFRET to four colors to study conformational changes in the Hsp90 protein-complex chaperone from yeast. They used fluorescent Atto dyes to label the two Hsp90 monomers, cochaperone p23 and ATP in order to detect p23 docking, changes in Hsp90 configuration, ATP binding and the presence of all three components. The researchers found that p23 binds to Hsp90 in the presence of ATP and that ATP hydrolysis causes a rearrangement of the binding partners in a directional manner.

Ratzke, C. *et al. Nat. Commun*. **5**, 4192 (2014).