TOOLS IN BRIEF

GENOMICS

CRISPR screens tackle the noncoding genome

The 98% of the human genome that does not encode proteins harbors many important regulatory regions, but identifying functions in such a vast genomic space is not trivial. Two research groups used CRISPR–Cas9-based screens to find regulatory elements in ~1 megabase of the human genome. Sanjana *et al.* targeted Cas9 to regions around three genes involved in resistance to a BRAF inhibition in melanoma. They identified loci that determine transcription factor binding and the deposition of active or repressive histone modifications around one of the genes. Fulco *et al.* targeted dead Cas9 fused to a transcriptional inhibitor to the vicinity of two genes encoding transcription factors and found several enhancers that regulate gene expression. They observed the emergence of regulatory networks in which multiple enhancers regulate the same gene or where multiple genes are regulated by the same enhancer.

Fulco, C.P. *et al. Science* http://dx.doi.org/10.1126/science.aag2445 (2016).
Sanjana, N.E. *et al. Science* **353**, 1545–1548 (2016).

IMAGING

Instant tracking of single-molecule position and orientation

Many biological structures depend on the proper orientation of molecular building blocks. However, studying an object's position and orientation in living cells can be challenging. Mehta *et al.* developed an instantaneous fluorescence polarizing microscope (instantaneous FluoPolScope) to enable tracking of single-molecule positions and orientations in living cells. The instantaneous FluoPolScope uses total internal reflection excitation with a custom image-splitting device that enables sorting of emitted fluorescence along four polarization orientations. The researchers show that their microscope can be used to track the positions and orientations of sparsely labeled proteins in order to study dynamic structural rearrangements, and it can also be used to track the position and orientation of protein subunits as they interact with higher-order assemblies. The microscope can potentially shape understanding of how relatively large ordered assemblies are formed from individual molecules.

Mehta, S.B. *et al. Proc. Natl. Acad. Sci. USA* **113**, E6352–E6361 (2016).

CHEMICAL BIOLOGY

Three steps to installing authentic protein modifications

Post-translational modifications (PTMs) such as methylation and acetylation endow proteins with diverse functions. In order for such functions to be studied in detail, methods for installing specific PTMs at specific residues are needed. Yang *et al.* now describe a general approach for this. They used their previously reported *O*-phosphoserine (Sep) orthogonal translation system to introduce Sep at a desired location in a target protein. Sep can be converted via phosphate removal into a reactive dehydroalanine (Dha), which acts as a radicalophile. PTM-containing alkyl-iodide moieties can then be coupled to Dha via a metal-mediated reaction, resulting in carbon–carbon bond formation. This tool provides a straightforward approach to installing authentic PTMs onto target proteins, which the authors demonstrated for histone H3K79 as well as for ubiquitin, showing that the chemically modified proteins displayed expected biological functions.

Yang, A. *et al. Science* **354**, 623–626 (2016).

NANOBIOTECHNOLOGY

Nanokits for single cells

Methods for analyzing small molecules and proteins at the single-cell level are crucial for understanding cellular heterogeneity. Pan *et al.* describe a 'nanokit' for probing single cells. The nanokit consists of a nanometer-sized capillary with a ring electrode at the tip. This nanocapillary, which is filled with components from traditional assay kits, is stuck into cells to introduce femtoliters of assay solution. The extent of assay kit reactions are then read out by coulometry, rather than fluorescence. The researchers demonstrated the utility of the device by examining the abundance of glucose, as a representative small molecule, and the activity of the protein sphingomyelase, as a representative enzyme, at the single-cell level. This nanokit should facilitate single-cell analysis of many important biological targets.

Pan, R. *et al. Proc. Natl. Acad. Sci. USA* **113**, 11436–11440 (2016).