METHODS IN BRIEF

IMAGING

Counting stoichiometry with super-resolution image data

Approaches for calculating the stoichiometry of complexes from single-molecule localization microscopy data have begun to make super-resolution imaging more quantitative. However, true quantitation is hindered by factors such as incomplete labeling and fluorophore blinking. Fricke *et al*. describe an approach for counting membrane-protein stoichiometry that takes fluorophore blinking kinetics into account. The team first measured the blinking behavior of mEos2 to determine the probability of the protein blinking. They then imaged mEos2-labeled structures of interest using photoactivated localization microscopy (PALM) and extracted the number of blinking events per complex. Using the probability of a single emitter blinking, they determined the number of emitters present in a given complex. They demonstrated their approach on monomeric and dimeric surface proteins.

Fricke, F. *et al. Sci. Rep.* **5**, 14072 (2015).

STEM CELLS

Mouse chimeras for testing human stem cell pluripotency

Human embryonic stem cells (hESCs) and human induced pluripotent stem cells (hiPSCs) are considered pluripotent because of their ability to differentiate into most embryonic cell types *in vitro*. But a more stringent test for pluripotency—the ability to contribute to all tissues in a developing embryo—has been assessed only in mouse pluripotent cells, for obvious reasons. Mascetti and Pedersen have now applied a similar standard to human cells by generating stage-matched interspecies chimeras in the mouse. They transplanted three hiPSC and two hESC fluorescently labeled lines into gastrula-stage embryos and found high (70–100%) tissue incorporation across all fetal subregions, similar to observations with murine epiblast stem cells. The work establishes the mapping of human tissue contribution to murine chimeras as an *in vivo* test for stem cell pluripotency.

Mascetti, V.L. and Pedersen, R.A. *Cell Stem Cell* **18**, 1–6 (2016).

GENOMICS

Comparing cancer genome analyses

Whole-genome sequencing (WGS) of tumor samples to find somatic variants that explain cancer biology is becoming increasingly common, but data obtained by different research groups can be difficult to compare. The International Cancer Genome Consortium (ICGC) has now assessed different aspects of WGS experiments and recommends PCR-free library construction and a sequencing depth of ~100× for these studies. In addition, the ICGC developed benchmarking data sets—manually curated single-nucleotide variants and insertion/deletions from tumor-normal pairs sequenced to ~300×—to test somatic mutation–calling pipelines. The consortium showed that their benchmark data can improve the quality of pipelines by optimizing the combination of read mapper and variant caller used, but they also stress that further improvements are needed before WGS can be adopted for clinical use.

Alioto, T.S. *et al. Nat. Commun*. **6**, 10001 (2015).

CHEMICAL BIOLOGY

Capturing lysine PTM–dependent interactions

Protein post-translational modifications (PTMs), such as those found on lysine residues in histones, have important jobs in regulating protein interactions. These interactions are challenging to capture, however, as they are often weak and the PTMs themselves are present at low levels. Yang *et al.* describe an approach for capturing interactions dependent on lysine PTMs by utilizing photo–cross-linking. They designed a photo-reactive lysine that, when provided as the exclusive lysine source in cell culture, is incorporated in place of the native lysine and undergoes modification after translation. The photo-lysine residue, which contains a diazirine moiety, will form a covalent bond with binding proteins in close proximity upon photo–cross-linking. Such binding partners then can be identified using proteomics methods. The tool enabled Yang *et al.* to identify histone modification readers and erasers in live cells.

Yang, T. *et al. Nat. Chem. Biol.* doi:10.1038/nchembio.1990 (21 December 2015).