TOOLS IN BRIEF

SENSORS AND PROBES

Tracking β-galactosidase activity *in vivo*

β-galactosidase (β-gal) is a widely used enzyme in molecular biology. But it also has important physiological roles and can function as a cancer biomarker, generating a need for tools to image β-gal activity *in vivo*. Gu *et al*. describe a fluorescent dye that allows ratiometric measurement of β-gal activity *in vivo*. The dye, called DCM-β-gal, undergoes a large red-shift upon reaction with β-gal to emit maximally in the near-infrared region, making it particularly useful for deep-tissue imaging, as tissues are relatively transparent at these wavelengths. The researchers showed that the fluorescence ratio is linear over a broad range of enzyme concentrations, and they demonstrated utility by imaging tumor xenografts in living mice.

Gu, K. *et al. J. Am. Chem. Soc.* **138**, 5334–5340 (2016).

BIOINFORMATICS

Mapping nanopore sequence reads

The recent availability of the first protein-nanopore-based commercial sequencer has enabled the production of very long reads from single-molecule DNA templates. Taking advantage of this output is made complicated by the high random error rate in individual reads and the variability in quality and read lengths. Sović *et al*. introduce GraphMap, an efficient computational tool for mapping long sequence reads that is largely robust to error profile and error rate. On benchmark synthetic data modeled on a number of organisms, including human, GraphMap shows sensitivity similar to that of the BLAST gold standard, but it is several orders of magnitude faster. The method also performs well on real data relative to competing tools, and it enables downstream applications such as pathogen detection and robust calling of single-nucleotide polymorphisms and structural variants.

Sović, I. *et al. Nat. Commun.* **7**, 11307 (2016).

NEUROSCIENCE

Optimized tracing of neural circuits

Direct inputs into particular neurons can be identified through retrograde monosynaptic tracing with rabies-virus-based tools. To improve the efficiency of trans-synaptic tracing, Kim *et al*. optimized both trans-synaptic spreading and initial expression levels in starter neurons. The trans-synaptic infection process depends on virus particle packaging in the postsynaptic cell and on uptake in the presynaptic cell, which involves viral glycoprotein G. The researchers tested the trans-synaptic spread of rabies virus in the presence of chimeric G proteins that consisted of the extracellular portion of various rabies strains and the transmembrane and cytoplasmic parts of the commonly used G protein of the SAD B19 strain. Furthermore, they increased expression levels through optimized codon usage. The resulting optimized G protein (oG) resulted in up to twentyfold more efficient trans-synaptic tracing.

Kim, E.J. *et al. Cell Rep.* **15**, 692–699 (2016).

BIOINFORMATICS

Fast RNA-seq quantification

The volume of RNA-seq data keeps growing, creating the need for ever faster read-mapping and quantification tools. Rather than aligning reads to positions in each transcript, Bray *et al*. designed kallisto, a software tool based on the principle of pseudoalignment, in which reads or sequence fragments are mapped back to their transcript of origin rather than to their precise location within a transcript. The method relies on accessing an index of short sequences known as *k*-mers, along with a de Bruijn graph representation of the transcriptome that is also generated from *k*-mers. kallisto was the fastest among a number of popular methods tested, has a small memory footprint and retains high accuracy. The improved speed enables bootstrapping to assign confidence to transcript abundance estimates.

Bray, N.L. *et al. Nat. Biotechnol.* **34**, 525–527 (2016).