# METHODS IN BRIEF

STRUCTURAL BIOLOGY

## Cryo-EM with the Volta phase plate

Recent technology developments are enabling researchers to solve more and more challenging macromolecular structures at near-atomic resolution using cryo-electron microscopy (cryo-EM). One advance has been the introduction of an improved phase plate, the Volta phase plate, which enhances image contrast and allows in-focus data collection, thereby increasing signal-to-noise ratio. Danev and Baumeister recently showcased the advantages of the new Volta phase plate by using it in determining the structure of the *Thermoplasma acidophilum* 20S proteasome structure at 3.2-Å resolution. The Volta phase plate solves most technical problems with previous phase plates. Notably, precise centering of the electron beam is not required; a phase shift can be created on the fly at any place on the phase plate, which enables automated data collection, a real practical boon for the cryo-EM field.

Danev, R. & Baumeister, W. *eLife* http://dx.doi.org/10.7554/eLife.13046 (2016).

NEUROSCIENCE

## Mutational burden of neurons

It has been speculated that somatic mutations in neurons are involved in generating cellular diversity. However, single-cell genomic sequencing approaches lead to artifacts that preclude a detailed analysis of somatic mutations in postmitotic cells such as neurons. Hazen *et al.* created embryonic stem cells from neurons of the mouse olfactory bulb via somatic cell nuclear transfer, which resulted in sufficient cellular material for high-resolution genome sequencing. Using a bioinformatics pipeline, the researchers detected single-nucleotide variants, deletions, mobile element insertions and other mutational events. On the basis of their sample size of six cell lines, they concluded that individual neurons accumulate about 100 somatic mutations. Despite this mutational load, neurons can be reprogrammed to produce viable and fertile mice, indicating that neuronal diversity might not depend on somatic mutations.

Hazen, J.L. *et al. Neuron* **89**, 1223–1236 (2016).

GENE REGULATION

## Long noncoding RNA networks of suppression

Of the approximately 10,000 long noncoding (lnc) RNAs encoded in the human genome, the functions of only a few have been characterized. Some lncRNAs have been shown to harbor binding sites for microRNAs and thus act as their repressors, by preventing microRNAs from binding their targets on protein-coding genes (PCGs). To learn about the regulatory networks formed by these so-called sponge lncRNAs, Du *et al.* used gene expression data on lncRNAs and PCGs with shared microRNA binding domains. Their network consisted of 52 lncRNAs and 17 PCGs in prostate cancer cells. lncRNAs that effectively repressed microRNAs resided in the cytoplasm, and some, such as the lncRNA that binds a microRNA targeting PTEN, were shown to have tumor-suppressive effects.

Du, Z. *et al. Nat. Commun.* **7**, 10982 (2016).

IMAGING

## Zooming in on single tumor cells

Intravital imaging of cancer cells can yield important insights into processes such as metastasis. However, metastatic events are rare, and thus it is a challenge to identify them with high-resolution imaging. Karreman *et al.* describe a multimodal correlative microscopy approach that enables rapid and precise targeting of cancer cells for high-resolution imaging by 3D electron microscopy (3DEM). In their approach, a cell of interest is first identified by fluorescence microscopy. Next, the precise position of the cell is determined by registration with X-ray computed tomography data. Finally, the cell is imaged using 3DEM. To demonstrate the method, they injected fluorescently labeled cancer cells into a mouse heart and imaged cells that metastasized to the brain. Their approach improves the throughput of rare cell ultrastructural analysis by allowing experiments to be completed from start to finish in around two weeks.

Karreman, M.A. *et al. J. Cell Sci.* **129**, 444–456 (2016).