IN THIS ISSUE

Mining GRO-seq data for regulatory sites

A number of approaches aim to find regulatory regions in the genome. For example, global (and precision) run-on and sequencing (GRO-seq and PRO-seq) detect nascent transcription and can be used to pinpoint active promoters. A modified protocol that enriches for 5' capped RNAs (GRO-cap) has higher sensitivity for identifying signal from the short-lived enhancer RNAs transcribed at stimulus-dependent enhancers, but it is not used as widely. Siepel, Lis, Danko and colleagues now present the computational tool dREG for identifying both active enhancers and promoters on the basis of GRO-seq or PRO-seq data. dREG is a machine learning approach, trained on GRO-cap data, that uncovers a potent class of regulatory elements predicted by both DNase I-hypersensitive sites and active chromatin signatures, and that is highly enriched for functional indicators such as quantitative trait loci and phenotype-associated polymorphic sites.

Article p465

Epigenome editing to dissect enhancer function

Cell type-specific gene expression, which is essential during development, relies on the action of enhancers. Several techniques allow for the identification of enhancer elements; Maehr and colleagues have now developed a method to functionally characterize them. They use the clustered, regularly interspaced, short palindromic repeats (CRISPR) system whereby guide RNAs and a nuclease-'dead' version of Cas9 fused to a histone demethylase are used to target enhancers and remove methyl marks from core histones, thereby decommissioning them. Of eight targeted enhancers thought to regulate pluripotency-specific genes, four proved to be critical for the maintenance of pluripotency in embryonic stem cells. This epigenome-editing tool will enable the rapid functional characterization of cis-regulatory elements.

Brief Communication p401

Listening to the brain

In photoacoustic microscopy, excitation light is absorbed by the illuminated tissue and converted into ultrasound emission. Tissue properties, as well as the wavelength or the pulse width of the laser used for excitation, influence the characteristics of the emitted ultrasound waves. Wang and colleagues make use of these differences to measure blood oxygenation in the mouse brain. They excite hemoglobin with picosecond and nanosecond lasers of the same wavelength and energy and record the emitted signals at high speed. Because the ratio between the nanosecond and the picosecond signal varies with oxygen saturation, Wang and colleagues can map oxygen saturation as a proxy for neural activity across the brain in response to activities such as electrical stimulation of the hindlimbs.

Brief Communication p407

Imaging newborn proteins

The spatiotemporal regulation of translation is crucial for normal cellular function. Methods to study newly synthesized proteins, such as pulse-labeling with radioactive amino acids, label the entire newly synthesized proteome. Schuman and colleagues overcome this limitation to enable imaging of nascent proteins of interest using two complementary methods: cells are pulse labeled with either a methionine analog or puromycin, which are incorporated into newly synthesized proteins. Next, cells are probed with antibodies against the pulse label as well as the target protein. The antibodies colocalize only at sites of new target protein synthesis, giving rise to signal from the proximity ligation assay (PLA). The two methods—fluorescence noncanonical amino acid tagging–PLA (FUNCAT-PLA) and Puro-PLA—were used to study local translation in neurons.

Brief Communication p411

An expanded TALE repertoire

Designer transcription activator–like effector (TALE) proteins can be targeted to specific genomic sequences for either genome editing or transcriptional modulation. Targeting is achieved by the modular assembly of TALE repeat sequences, each recognizing a single DNA base via its repeat variable diresidue (RVD). Rebar and colleagues now greatly expand the set of RVDs available for TALE design by going beyond the RVD code that occurs in nature. In a test of all 400 possible RVDs, the researchers identified many non-natural sequences that effect specific and strong binding to DNA. They show examples of how these RVD modules may be used to improve the performance of TALENs for specific genome editing.

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