**Amplification-free RNA-seq**

The analysis of RNA by high-throughput sequencing has enabled researchers to draw a much more comprehensive picture of transcriptional patterns, but the most widely used protocol is bias-prone because it relies on the PCR amplification of a cDNA library before sequencing. In this issue of *Nature Methods*, Turner and colleagues now move the reverse transcription step directly to the flowcell of the sequencer (FRT-seq), thus avoiding the need for cDNA library preparation. This also allows them to retain strand specificity in the resulting sequences, as the template is mRNA.

**Brief Communication p123**

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**Western blot on a microarray**

Although it is indispensable for protein detection, the western blot is an inherently low-throughput method. High-throughput methods, such as mass spectrometry, require large amounts of sample and still cannot always detect low-abundance proteins. Reverse-phase protein microarrays, in which lysates are arrayed on a surface and probed with antibodies, can be used for multiplexed protein detection from limited sample amounts. Jones and colleagues now describe an advance to the reverse-phase microarray approach, which they call microwestern arrays. After arraying cell lysates on a gel, they perform an electrophoresis step to separate the proteins and thus reduce the complexity of the sample. This allows more sensitive antibody-based detection, for up to 96 different protein targets at a time. The authors used this approach along with network-modeling methods to infer connectivities between receptor tyrosine kinase phosphosites in human cancer cells.

**Article p148**

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**Uncaging in two colors**

The use of light to activate, or uncage, biological compounds that have been made biologically inert through the attachment of a 'caging' group via a light-sensitive bond has been a powerful tool to study neuronal signaling with high spatial and temporal precision. But up to now, it has not been possible to use this method to concurrently stimulate and inhibit neurons. By combining a new caged GABA with a specific caged glutamate, Ellis-Davies and colleagues demonstrated two-color, two-photon uncaging of these crucial neurotransmitters. This allows more in-depth investigation of neuronal signaling mediated by activation of endogenous receptors rather than relying on exogenously expressed light-sensitive channels.

**Brief Communication p130**

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**Enriching targets**

Often it is not the entire genome sequence that is of interest to researchers but only specific regions, and the question is how to best enrich for these targets while avoiding the rest. Turner and colleagues present a Review that discusses three popular target-enrichment strategies: PCR approaches including the RainStorm platform, molecular inversion probes and hybrid-capture methods. The authors advise users on the suitability of different strategies given target size and sample number, and provide optimized protocols for on-array and in-solution hybrid capture.

**Review p111**

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**Crystal-clear imaging**

The use of adaptive optics to correct for atmospheric optical aberrations has allowed astronomers to see details people thought would require space-based telescopes. Although similar optical aberrations degrade microscopic imaging, particularly in thick samples, the approaches commonly used by astronomers are not feasible for microscopists. Ji and colleagues overcame these difficulties by separating the excitation light from a microscope into subregions that can be individually shifted to correct for aberrations and recover nearly perfect imaging performance. They applied the method to two-photon imaging of fluorescently labeled neurons at depths of up to 400 µm in brain slices. The method promises to be a boon to many imaging applications in biology and beyond.

**Article p141, News and Views p108**