Technology Watch

RNA DOMAIN CHARACTERIZATION

The topology and function of long non-coding RNAs (lncRNAs) have been poorly characterized, because it usually involves the laborious generation of deletion mutants. To circumvent this drawback, Quinn *et al.* developed domain-specific chromatin isolation by RNA purification (dChIRP). This technique simultaneously maps RNA–chromatin interactions and chromatin-based RNA–RNA and RNA–protein interactions, from which RNA domain functionality can be inferred. In dChIRP, biotinylated antisense oligonucleotides are designed in pools that target distinct regions of the RNA, cells are crosslinked to preserve protein–nucleic acid interactions, nucleic acids are sheared (their size delineates the domain-mapping resolution) and the different oligonucleotide pools are then added separately. Oligonucleotides that hybridized to their target RNA fragments are purified, and RNA, protein and DNA are extracted. Using dChIRP, the authors found that the fly lncRNA roX1, which is essential for X chromosome dosage compensation, has many target sites clustered in a dosage compensation territory on the chromosome. roX1 is organized into modular, functional subunits — a core ('palm'), and extending 'fingers' that bind to proteins and chromatin. The finger domains rescue the phenotype of roX1-null male flies as efficiently as does full-length roX1.

ORIGINAL RESEARCH PAPER Quinn J. J. *et al.* Revealing long noncoding RNA architecture and functions using domain-specific chromatin isolation by RNA purification. *Nature Biotechnol.* http://dx.doi.org/10.1038/nbt.2943 (2014)

MULTIPLEXED ENZYME SCREENING

Enzyme inhibitors are widely screened for potency; however, selectivity is often tested only on lead candidates, resulting in many clinical failures. The authors developed EnPlex for multiplexed, high-throughput screening of compound potency and specificity. In EnPlex, small quantities of purified enzymes are coupled to colour-coded microbeads. Multiplexed bead–enzyme complexes are then incubated with an inhibitor that competes with activity-based fluorescent probes for binding to enzyme active sites. These mixtures are scanned by flow cytometry, in which one laser detects bead colour (enzyme identity) and another detects fluorescent signals (enzyme activity). EnPlex was validated by simultaneously screening >100 Ser hydrolases, a clinically relevant enzyme superfamily, against a panel of known inhibitors. The results confirmed known enzyme–inhibitor interactions and revealed hundreds of new interactions. EnPlex was then used to screen a library of electrophilic compounds, identifying lead molecules on the basis of both selectivity and potency. Unlike EnPlex, low-throughput methods can confirm enzyme–compound interactions *in situ* and do not require protein purification and immobilization; however, EnPlex offers both high-throughput coverage and increased sensitivity, and could be used to prioritize compounds during primary screening based on both potency and specificity.

ORIGINAL RESEARCH PAPER Bachovchin, D. A. *et al.* A high-throughput, multiplexed assay for superfamily-wide profiling of enzyme activity. *Nature Chem. Biol.* http://dx.doi.org/10.1038/nchembio.1578 (2014)