nature reviews genetics

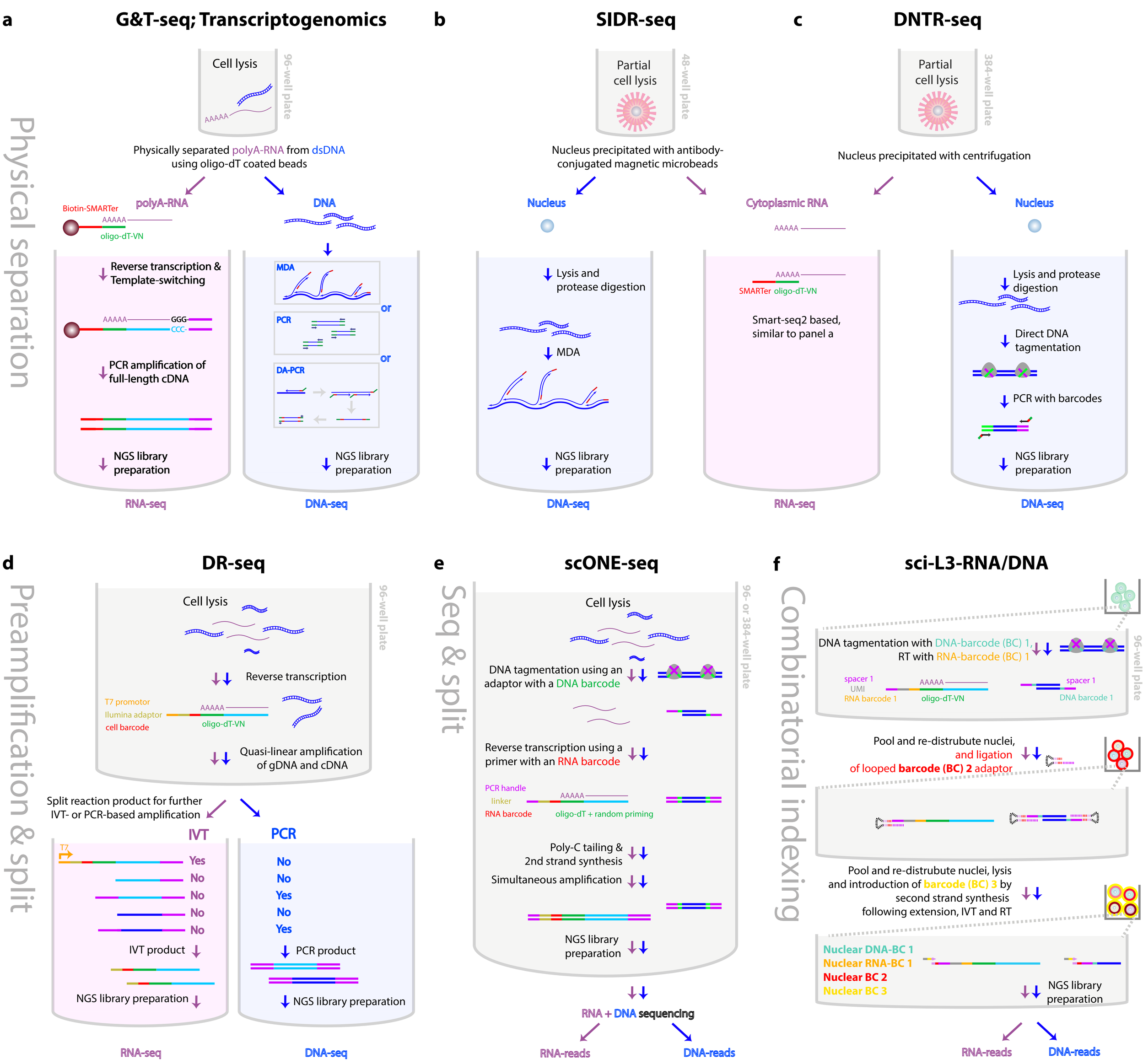
https://doi.org/10.1038/s41576-023-00580-2

Review article

Check for updates

Methods and applications for single-cell and spatial multi-omics

In the format provided by the authors and unedited



Supplementary Figure 1 | **Methods for single-cell genomics-plus-transcriptomics.**

(a-c) Methods relying on physical separation of DNA and RNA. **a |** Following oligo-dT bead-based separation, poly(A)-RNA is converted to cDNA using Smart-seq2. Separately, the DNA in the supernatant is precipitated and amplified with either MDA, PCR or DA-PCR before barcoded DNA-seq library preparation. **b |** In SIDR-seq, cytoplasmic poly(A)-RNA is subjected to Smart-seq2, while the nuclear DNA is amplified by MDA for DNA sequencing library preparation. **c |** In DNTR-seq, cytoplasmic RNA undergoes Smart-seq2, while the nuclear DNA is processed to an NGS-library using direct DNA tagmentation and PCR. **d |** In DR-seq, poly(A)-RNA is primed with oligo-dT (which also contains T7 promoter, NGS adaptor and cell-barcode sequences) for RT, and then quasi-linearly amplified together with single-cell DNA in a single-tube reaction. The resulting amplicons are then further amplified in two separate reactions: via IVT for RNA-seq, and via PCR for gDNA-seq library preparation. Note that in the latter reaction cDNA-derivative amplicons also co-amplify. **e |** In scONE-seq, DNA- and RNA-derived reads can be identified through their specific barcode incorporated during this single-tube reaction. **f |** In the sci-L3-RNA/DNA co-assay, no single cells are isolated, instead three rounds of pool-and-split based combinatorial indexing are applied to attain single-cell resolution of DNA- and RNA-measurements. In round 1, DNA and RNA are each equipped with a specific barcode 1 through tagmentation and RT, respectively. In round 2, a looped barcode 2-adaptor with T7 promoter is ligated and extended. Following IVT and first-strand cDNA synthesis, the 3rd barcode is incorporated via second-strand synthesis.