MobiuSCOPE Single Cell Full Length RNA Library Kit

Accurate and comprehensive RNA sequencing at the single cell level is crucial for understanding cellular heterogeneity, gene regulation, and the complexities of disease mechanisms. Traditional short-read single cell RNA sequencing methods often capture either 5' or 3' end of the transcripts for their quantification, which can miss critical information such as alternative splicing events, gene fusion, and allele-specific gene expression. This limitation can obscure the true complexity of cellular transcriptomes and hinder the discovery of novel biomarkers and therapeutic targets.

The MobiuSCOPE Kit provides an innovative solution to obtain full length single cell mRNA information with short read sequencing.

Highlights:

- **Accuracy and Sensitivity:** Comprehensive full length RNA information acquired with the high accuracy and throughput of short read sequencing
- **Simple workflow:** straightforward protocol, manual or automated
- **High Throughput:** Full length RNA information from tens of thousands cells in parallel

Technical Principle

MobiuSCOPE is a full-length RNA sequencing solution that captures the entire mRNA transcript at single cell level. A microwell chip, SCOPE-Chip is used for partition thousands of cells into individual wells, followed by capturing and barcoding mRNA from each single cell. An optimized reverse transcriptase (RT) formulation which has improved enzyme processivity is then used to generate full length 3' barcoded cDNA, each with a unique cell barcode and UMI next to the 3' sequence of the mRNA. A circularization step combined with subsequent reverse PCR is then used to bring the 5' sequence of the same mRNA to the proximity of the cell barcode and UMI, generating a 5' barcoded cDNA (Figure 1) . Sequencing libraries are constructed by random fragmentation of both 3' and 5' cDNA pools and ligating the fragments to Illumina sequencing adaptors. All reads coming from the same transcript have the same cell barcode and UMI and can be assembled by a proprietary bioinformatics pipeline to yield full length transcript information (Figure 2).

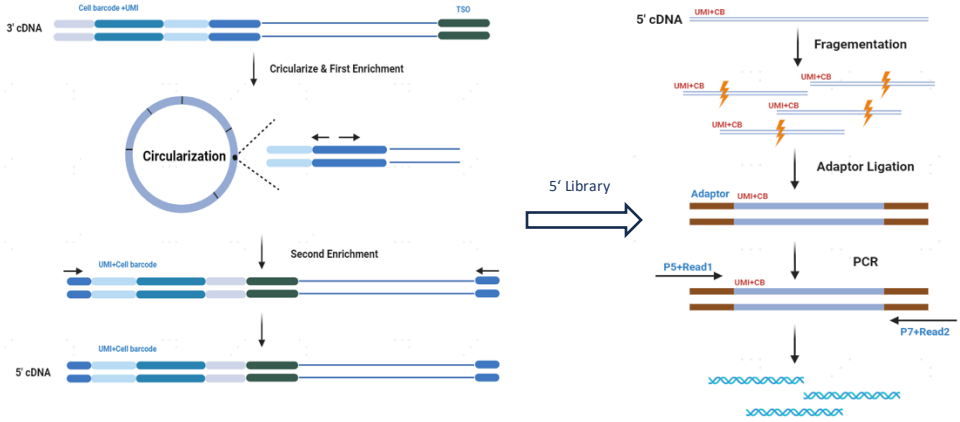

Figure 1. mRNA in each single cell is captured by the polyT oligos on cell barcoding beads and reversed transcribed to cDNA with unique barcode and UMI on the 3' end of the mRNA. The cDNA was then enzymatically circulated to bring the 5' region of the mRNA sequence close to the cell barcode and UMI. A reverse PCR is used to re-linearize the cDNA while keeping the 5' mRNA sequence in close proximity to the cell barcode and UMI. Sequencing library is then constructed by randomly fragmentation of the re-linearized cDNA and adaptor ligation.

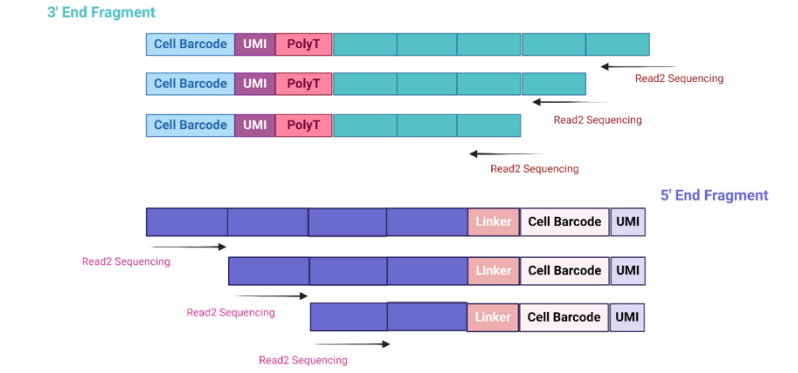

Figure 2. Randomly fragmented cDNA reads from both 3' and 5' of the same mRNA can be identified by the unique combination of cell barcode and UMI and assembled to generate full length mRNA information.

Full Length mRNA Sequence with Minimum Bias

Most high-throughput single cell RNA sequencing methods barcode either 5' or 3' end of the mRNA and have bias on one end or the other MobiuSCOPE circumvents this challenge by using circularization to bring both 3' and 5' regions of the mRNA close to the cell barcode to achieve full length mRNA information across the whole gene body with minimum bias (Figure 2). The comprehensive coverage of the RNA makes it possible to study allele-specific gene expression of SNPs and detect expressed genetic mutations at single cell level.

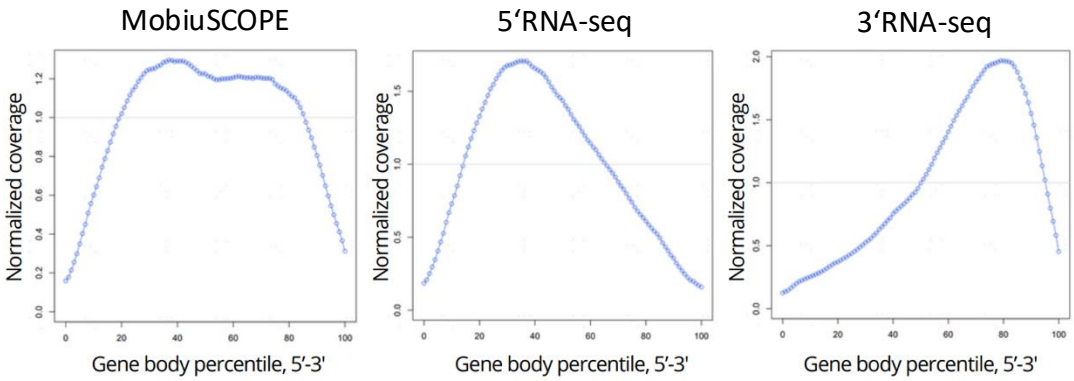

Figure 3. A mixture of mouse spleen cells and human PBMC was processed with MobiuSCOPE kit, or standard 3' or 5' scRNAseq method, and sequenced with PE150 on NovaSeq 6000 (Illumina). The RNA sequence information obtained with MobiuSCOPE has good coverage of the whole gene body, in contrast to the bias commonly seen with either 3' or 5' based single cell RNAseq methods.

Confident Detection of Splice Junctions

RNA splicing is an important step in the eukaryotic gene expression process that contributes to the functional diversity of the transcriptome. Aberrant regulation of the splicing events are often associated with diseases. With its comprehensive coverage across the whole gene body, MobiuSCOPE is able to detect significantly higher number of splice junctions of various types (Figure 4).

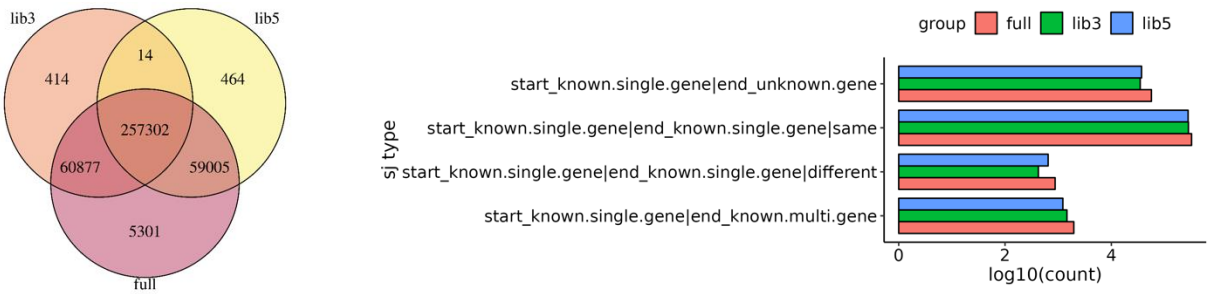

Figure 4. A mixture of mouse spleen cells and human PBMC was processed with MobiuSCOPE kit, or standard 3' or 5' scRNAseq method, and sequenced with PE150 on NovaSeq 6000 (Illumina). Significantly higher number of splice variants was detected with MobiuSCOPE method (‚full'), compared to either 3' scRNAseq method (‚lib3'), or 5' scRNAseq method (‚lib5').

Ordering information:

The MobiuSCOPE Single Cell Full Length RNA Library Kits can be used to generate full length single cell RNA sequencing library from cell suspension either manually, or with automated protocol on MatrixNEO instrument from Singleron.

Product	Reactions	Catalog number
MobiuSCOPE Single Cell Full Length RNA Library Kit (Manual)	2 RXNs	4503411
MobiuSCOPE Single Cell Full Length RNA Library Kit for NEO	4 RXNs 16 RXNs	45034251 45034221

Singleron Biotechnologies GmbH
Phone: +49 (0) 221 16824777
Address: Gottfried-Hagen-Strasse 60 51105 Cologne, Germany
Email: info@singleron.bio

Singleron Biotechnologies Pte. Ltd
Phone: +65 6990 5987
Address: 61 Science Park Rd, West Wing, the Galen 02-05 Singapore 117525
Email: sales.sg@singleron.bio

Singleron Biotechnologies US
Phone: +1 203-298-0604 (CT)
+1 734-249-0883 (MI)
Address: 1 Bradley Road, Suite 401 Woodbridge, CT 06525
333 Jackson plaza, Ann Arbor, MI 48103