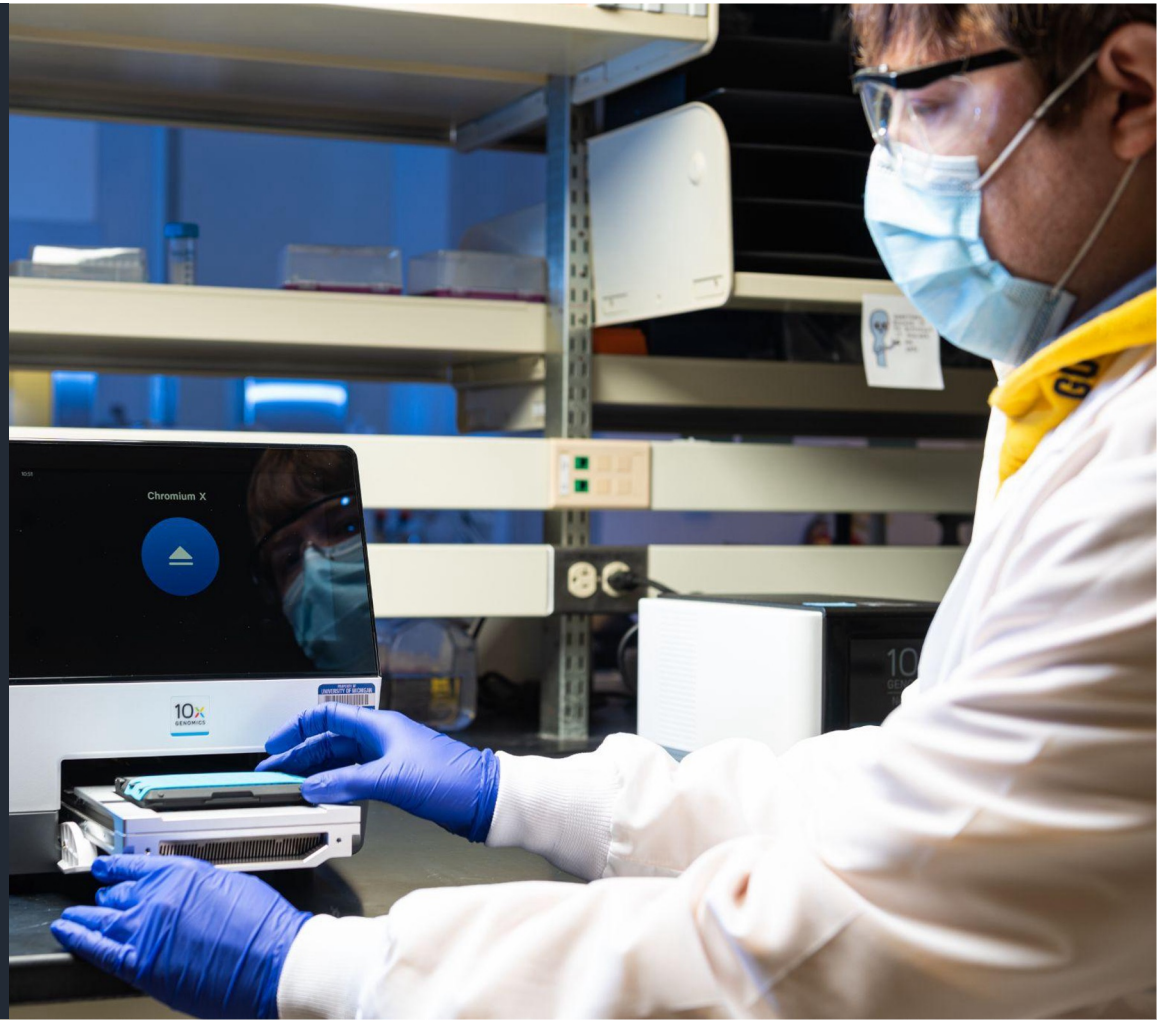
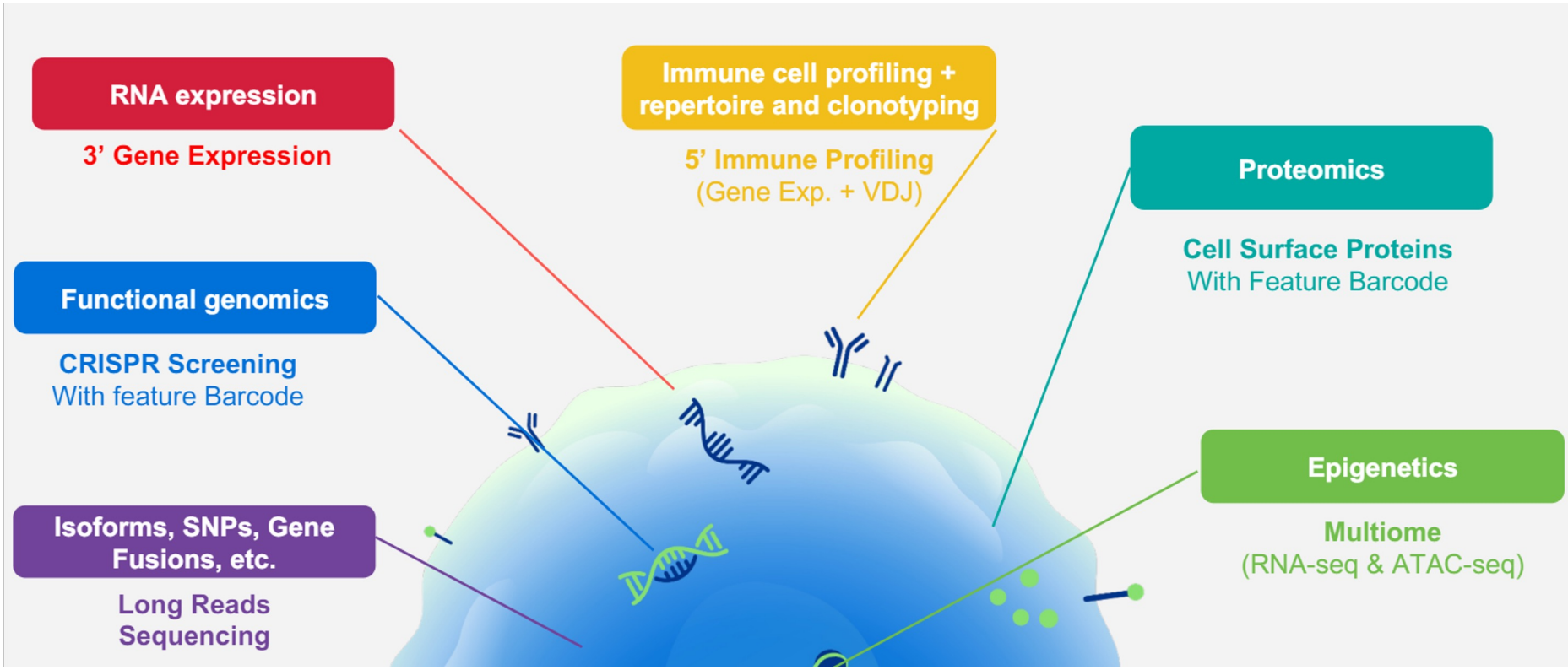
Intro to Single Cell Technologies at the AGC

advanced-genomics@umich.edu

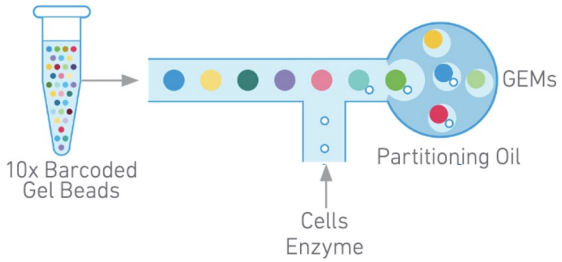


Flavors of Single Cell Sequencing



Approaches to Single Cell Sequencing

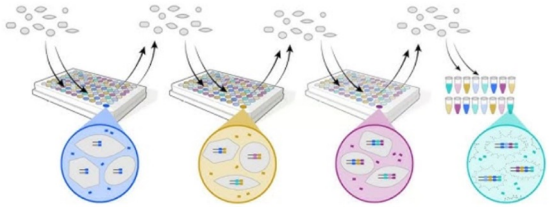
Microfluidic Droplet-Based



Benefits:
- throughput
- performance consistency
- low per cell cost
- multi-modal compatible

Limitations:
- size limitation (<30um)
- 3’ or 5’ bias or targeted

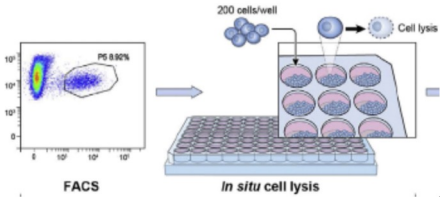
Split-pooling



Benefits:
- throughput
- reduced 3’ bias
- size agnostic

Limitations:
- fixation
- capture efficiency
- labor intensive

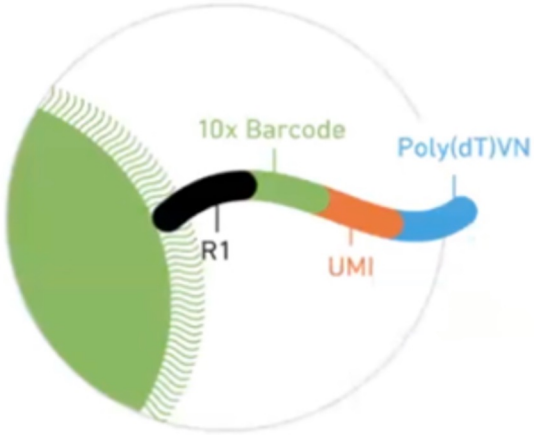
Direct Cell Lysis

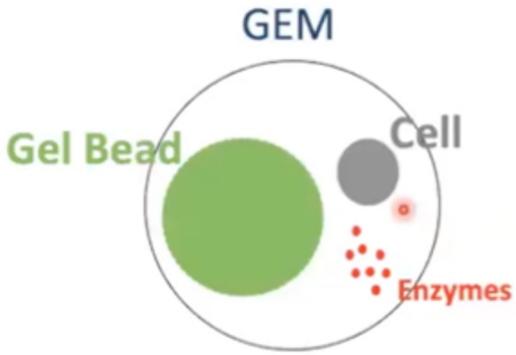


Benefits:
- full-length

Limitations:
- expense
- throughput

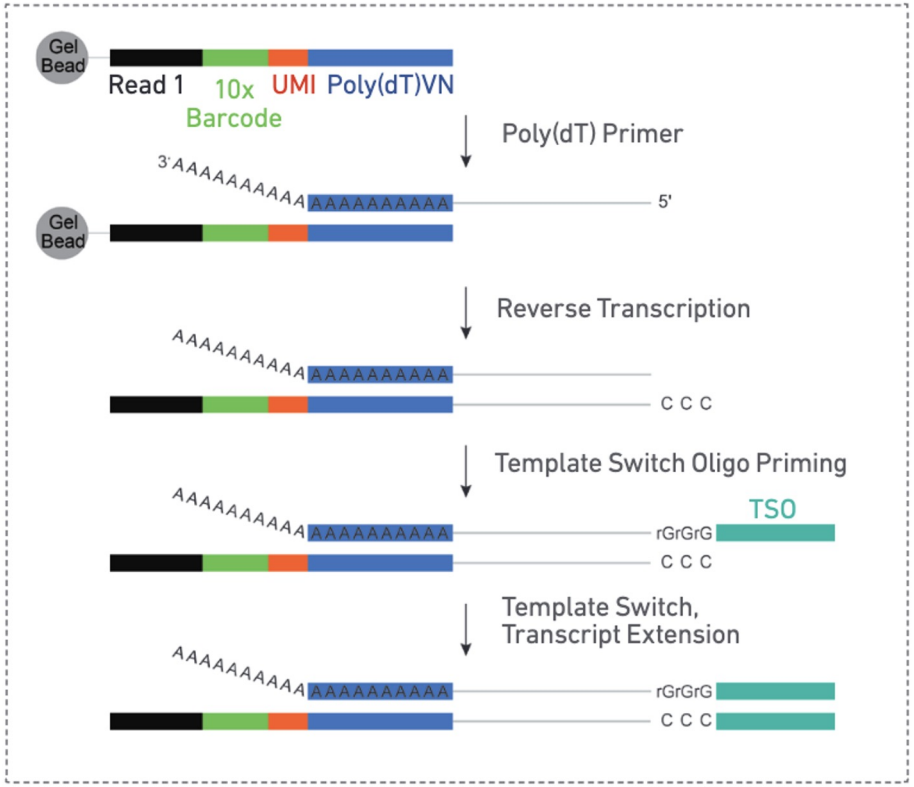
Single Cell RNA Sequencing



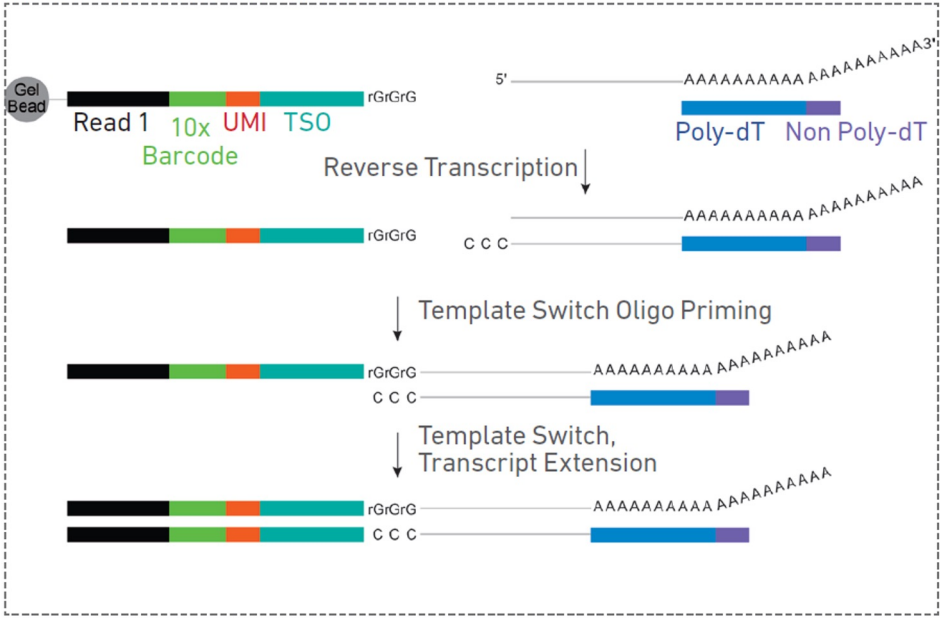


Transcript Capture Mechanisms

Inside individual GEMs 3’ GEX

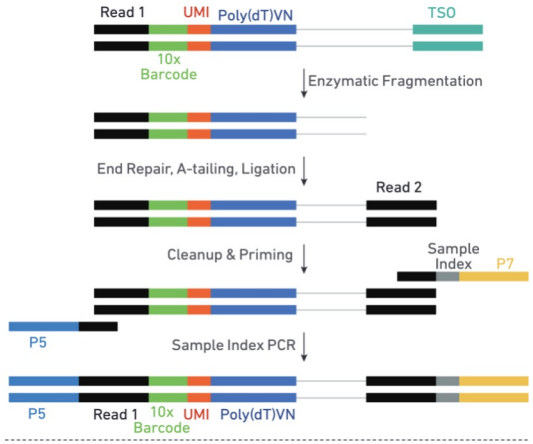


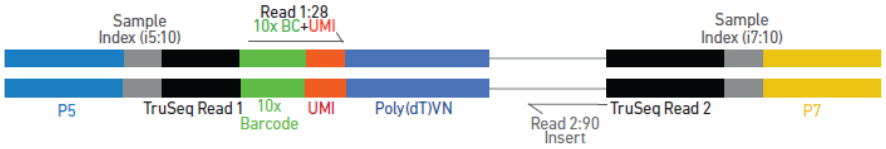
Inside individual GEMs 5’ GEX



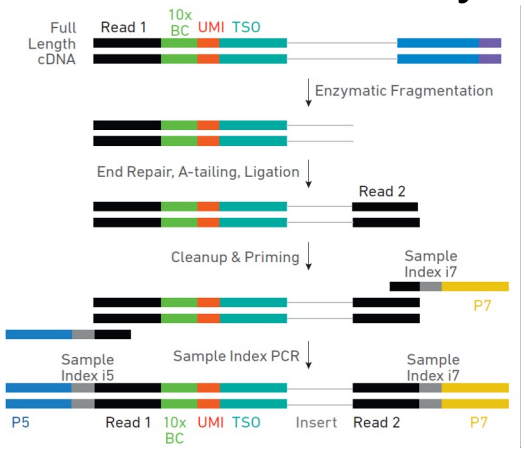
10x Capture Mechanisms

3’ GEX Library





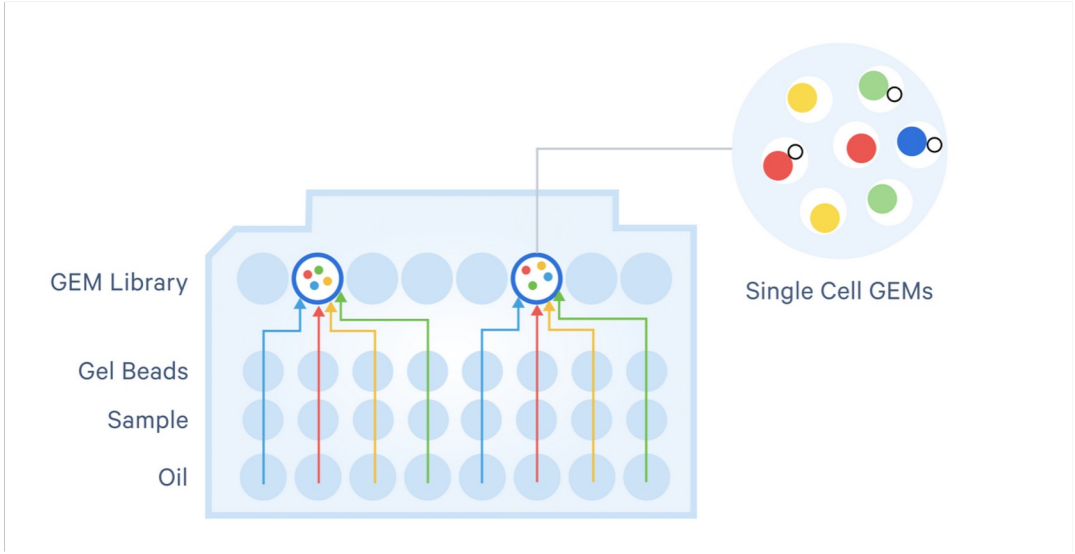
5’ GEX Library



BIOMEDICAL RESEARCH CORE FACILITIES
ADVANCED GENOMICS CORE
UNIVERSITY OF MICHIGAN

3’ and 5’ OCM: On-Chip Multiplexing

- Pool 4 samples
  - targeting 5000 cells/sample
- Compatible with
  - protein detection
  - TCR/BCR profiling
  - CRISPR screens



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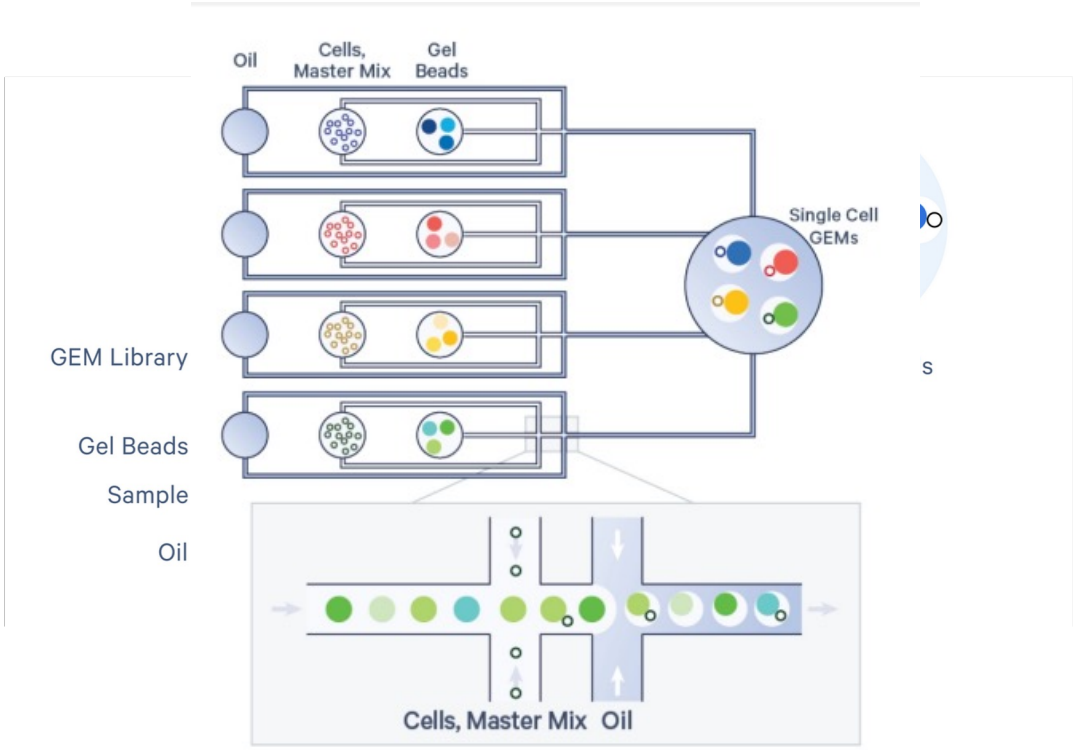

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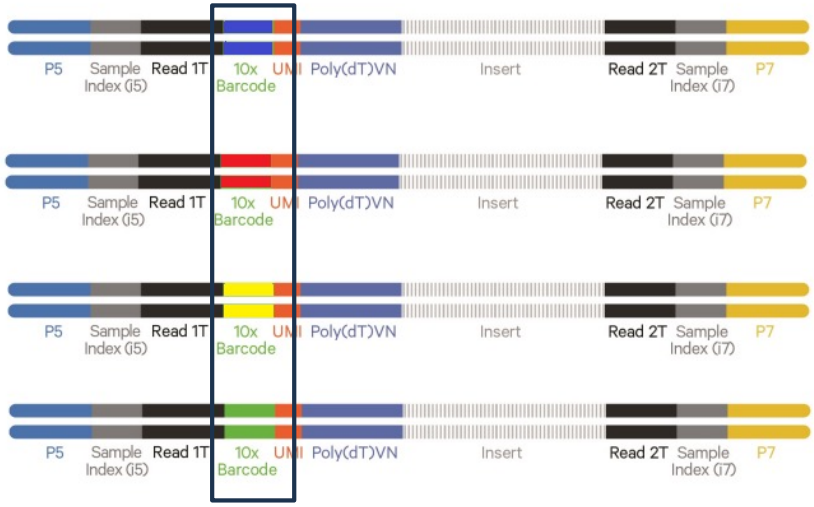
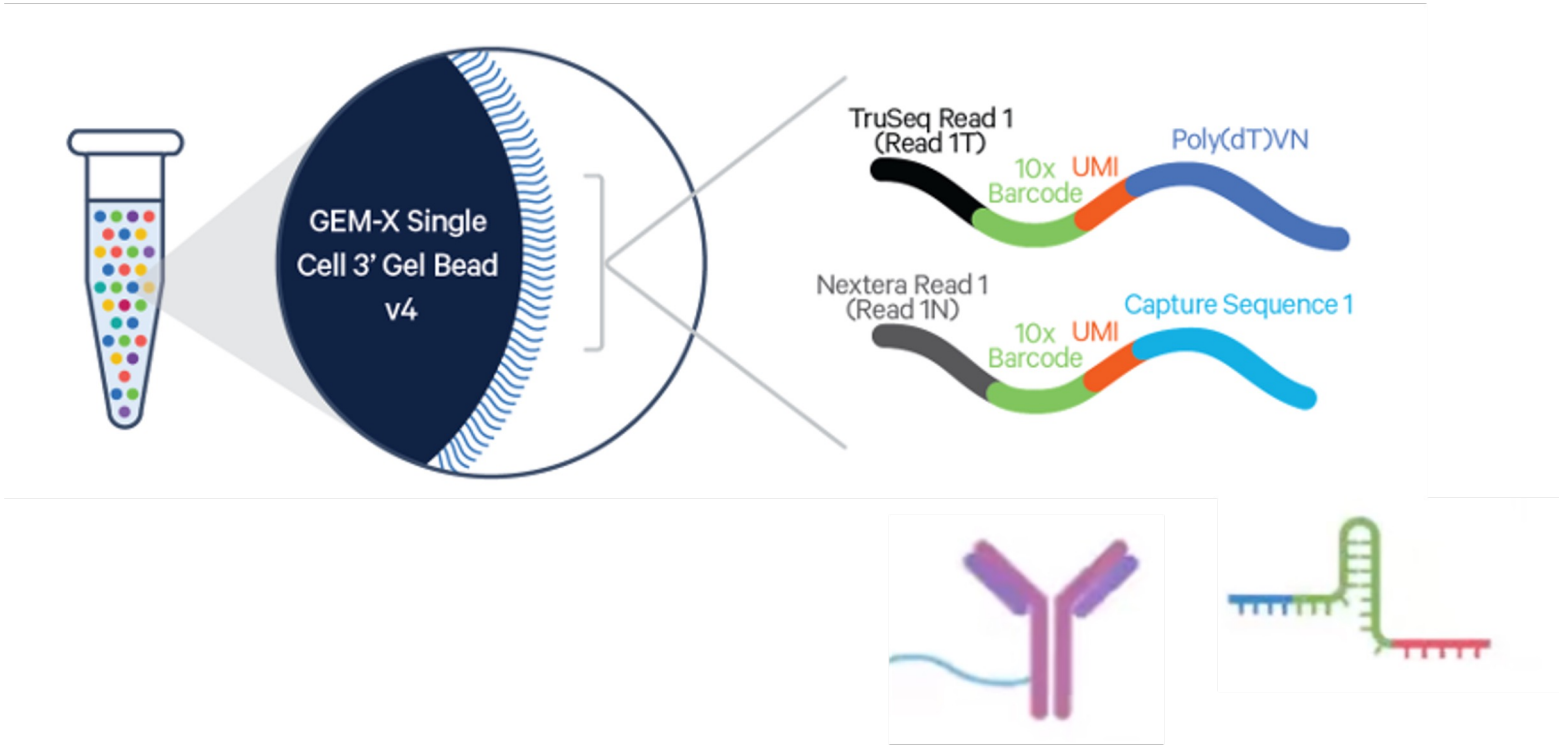
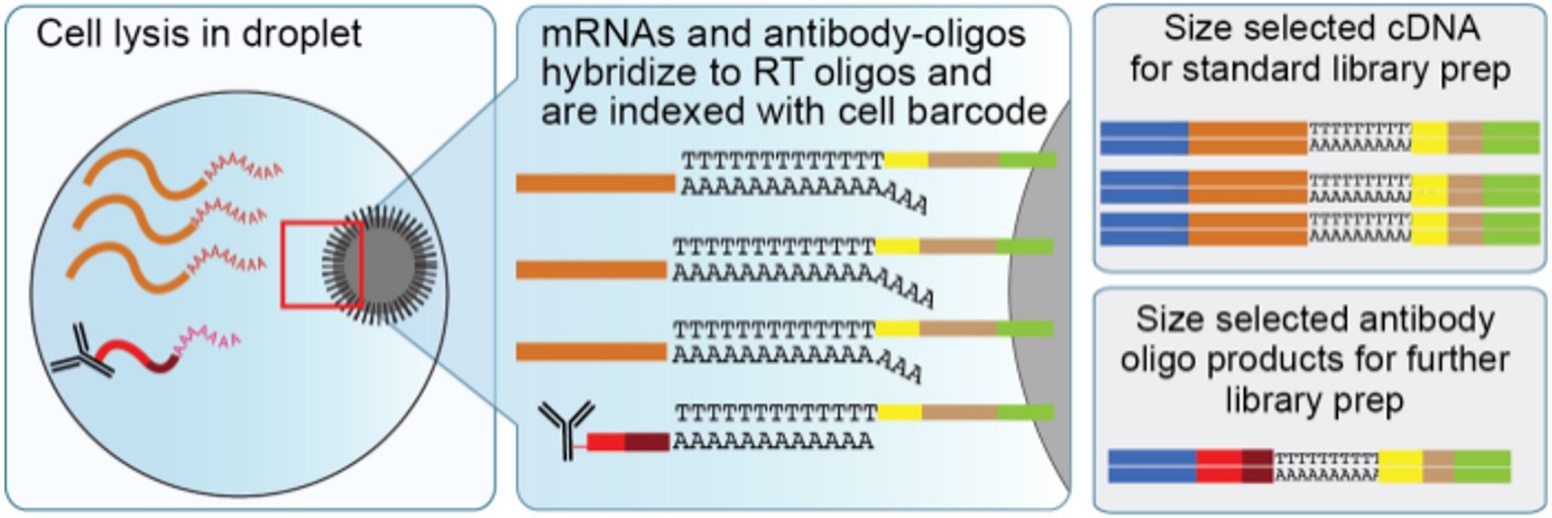


Proteomics

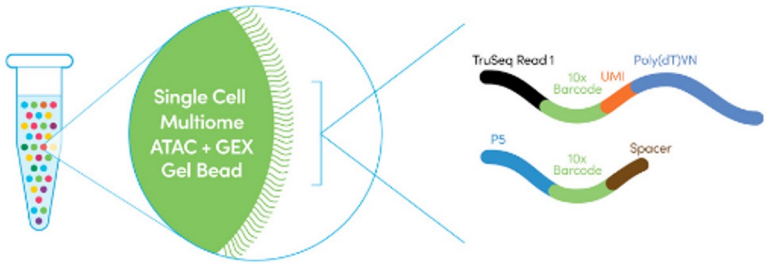

GEM-X Single Cell 3' Gel Bead v4
TruSeq Read 1 (Read 1T)
10x Barcode
UMI
Poly(dT)VN
Nextera Read 1 (Read 1N)
10x Barcode
UMI
Capture Sequence 1

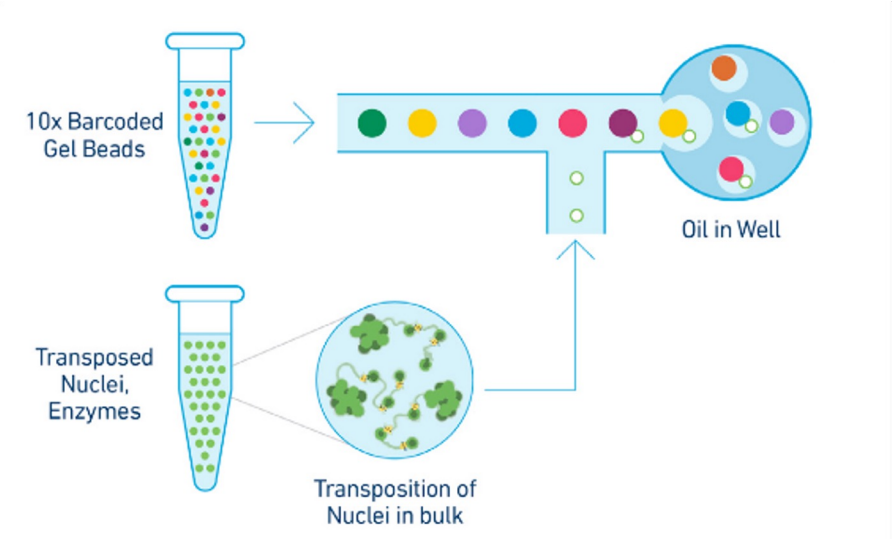
Cellular Indexing of Transcriptomes and Epitopes



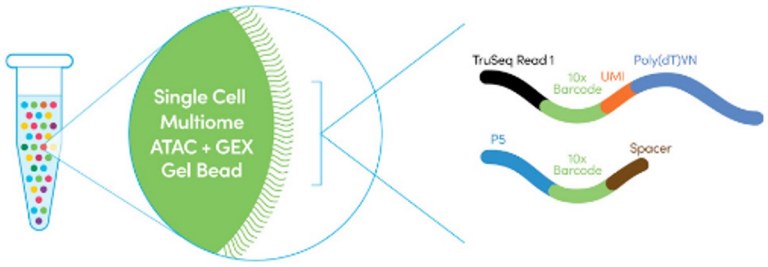
source: cite-seq.com

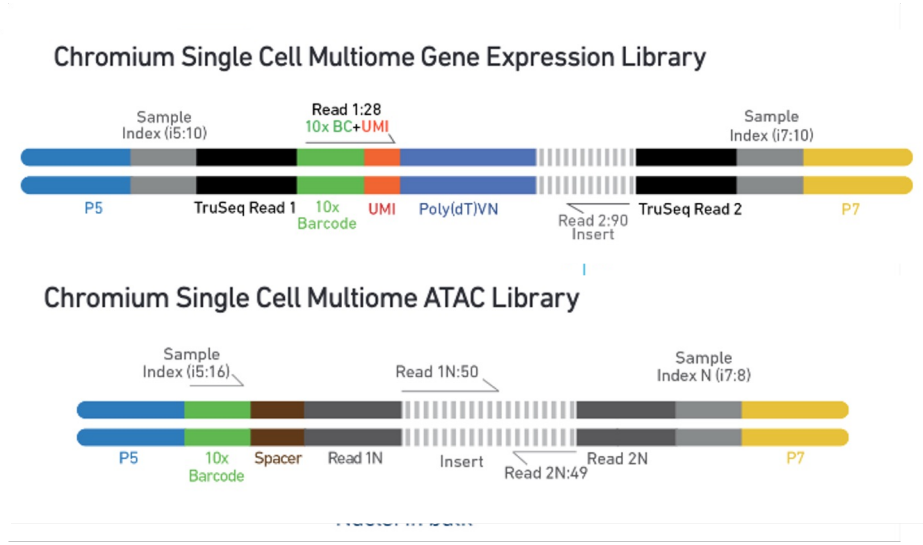
Epi Multiome (ATAC + Gene Expression)





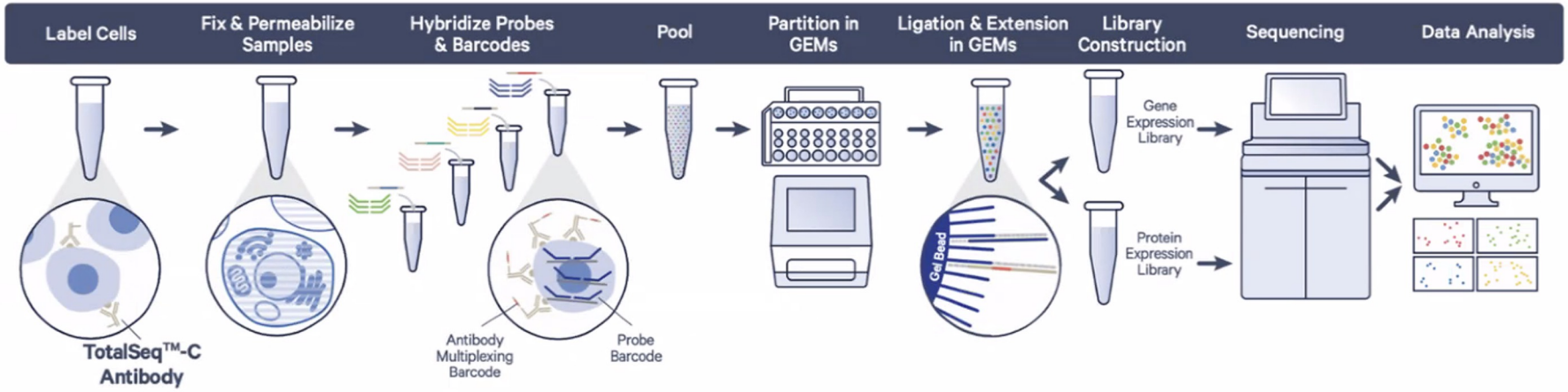
Epi Multiome (ATAC + Gene Expression)





Single Cell RNA Flex System

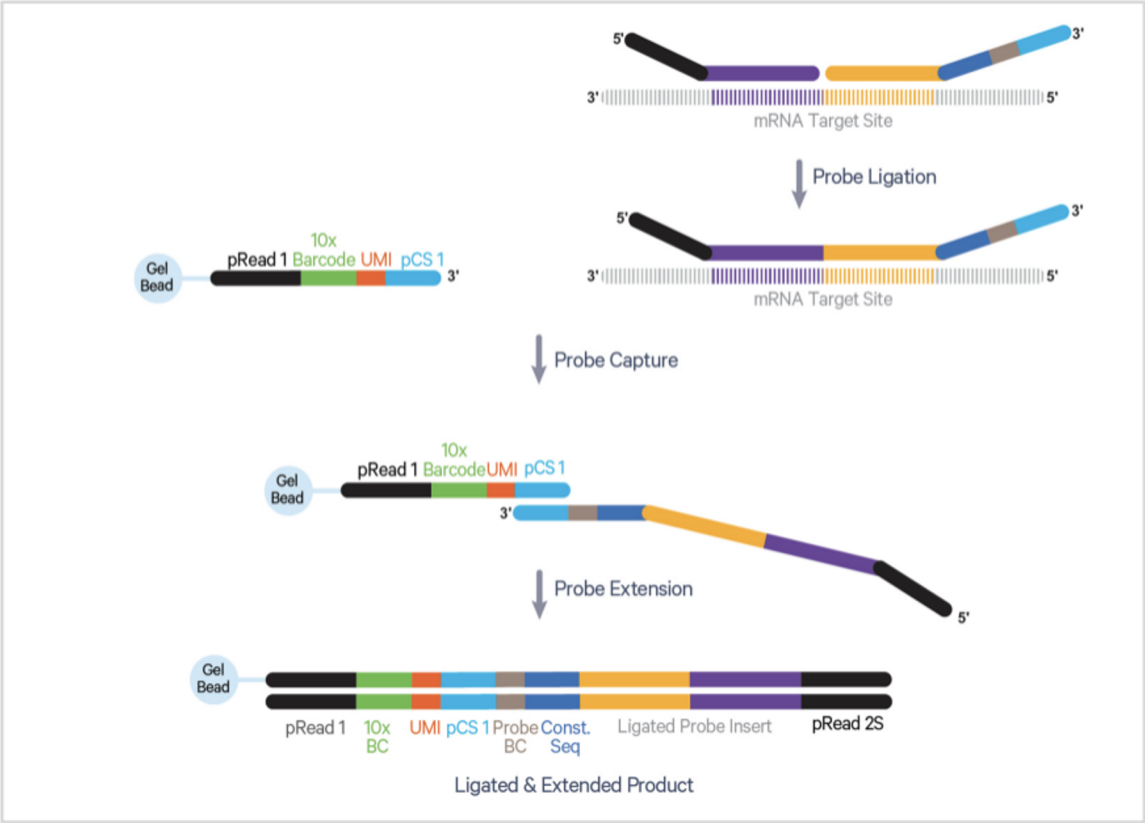
Same workflow - More cost effective and scalable



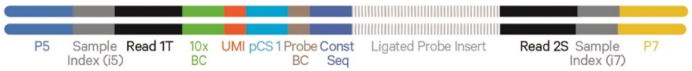
- Sensitive, probe-based **whole transcriptome assay**
- Compatible with Feature Barcode technology for **profiling cell surface and intracellular proteins** using singleplex or multiplex workflows
- Does not depend on polyA capture; covers more than 18,000 **human or mouse** genes

How it works:

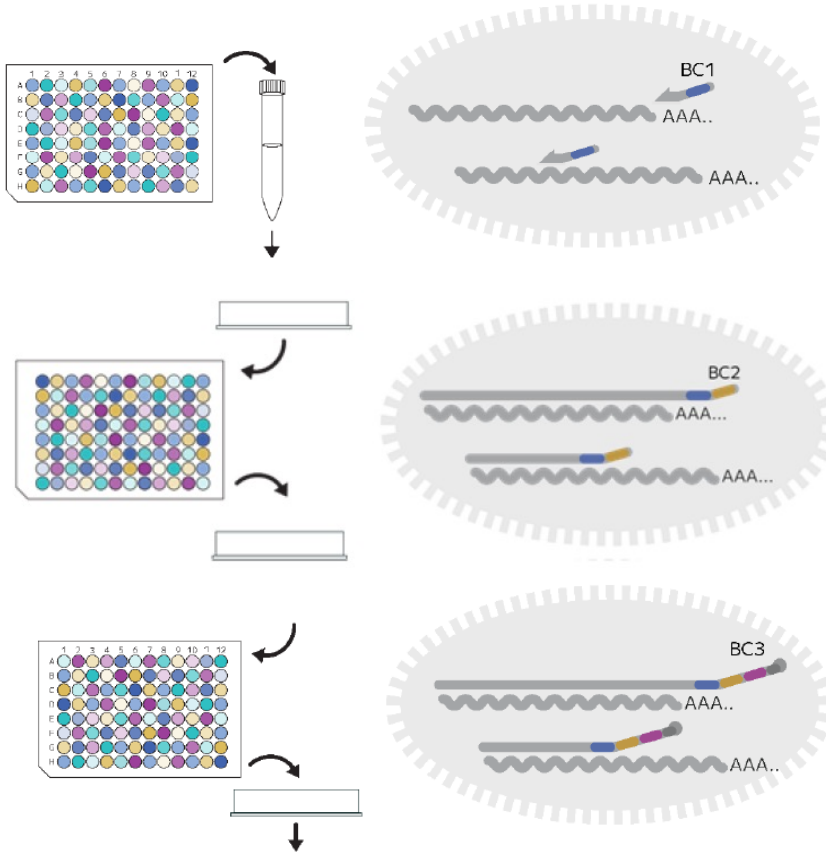
Direct capture of ligated probes

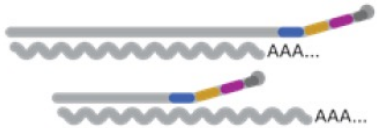


Followed by in-bulk amplification of products, and library construction and QC



SPLiT-seq


BC1
AAA..
AAA..
BC2
AAA...
AAA...
BC3
AAA..
AAA..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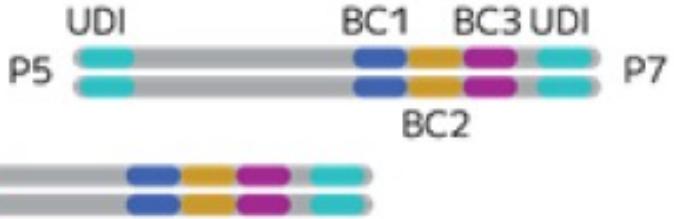

UDI
BC1
BC3
UDI
P5
P7
BC2

SPLiT-seq

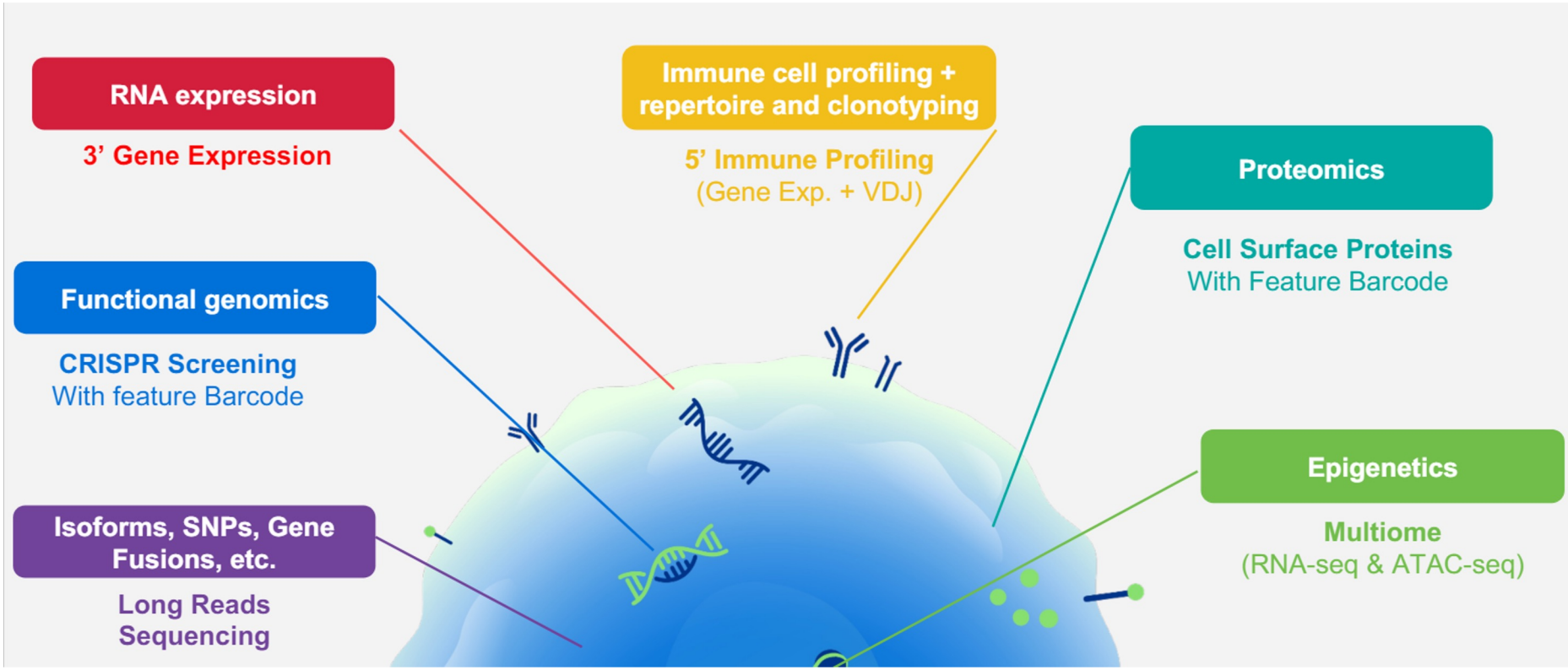
PARSE BIOSCIENCES

- Fix 100K-4 million cells or nuclei
- 5 million cells/96 or 384 samples
- RNA with TCR and CRISPR add-ons

SCALE biosciences

- Fix 100K-2.5million cells or nuclei
- 4 million cells/96 samples
- RNA, Methylation, ATAC, CRISPR, Proteomics

Flavors of Single Cell Sequencing



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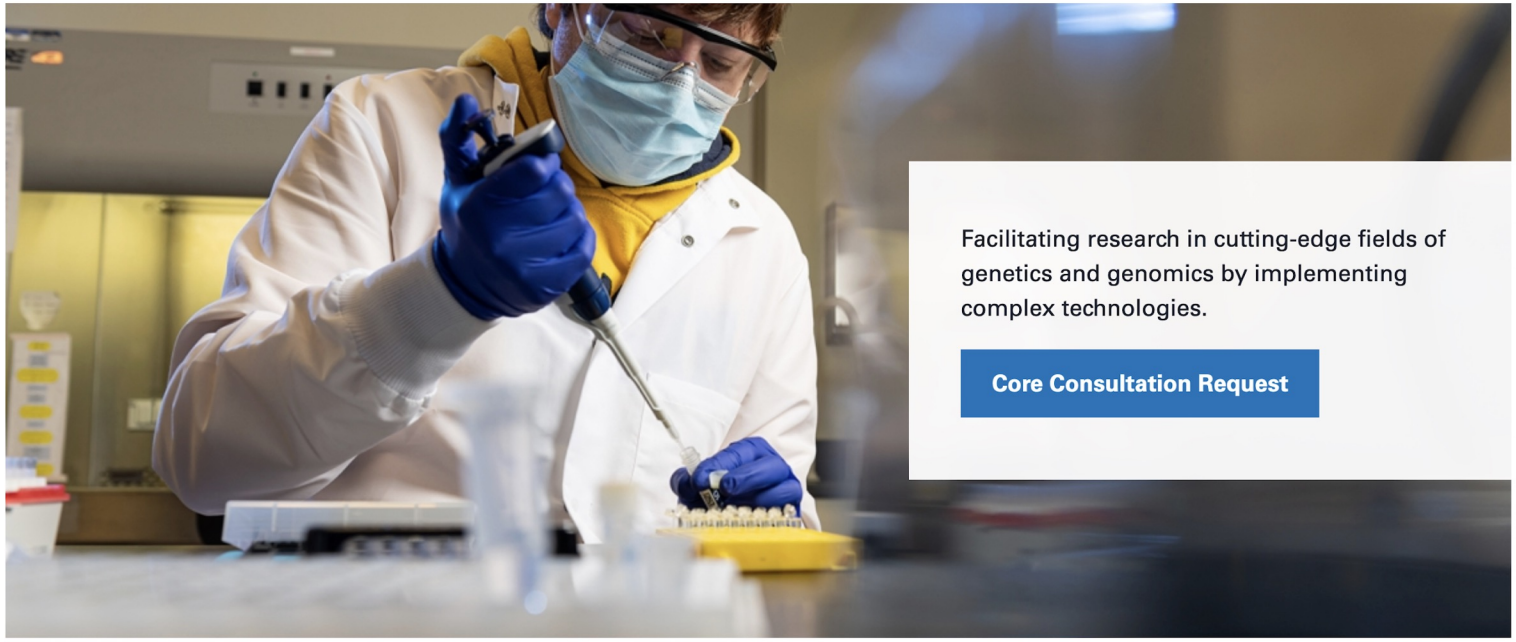

Introductory Workshops

Upcoming Workshop Dates:

Short Read Sequencing
August 5, September 2

Long Read Sequencing
August 19, September 16

Spatial Analysis
August 5, September 2

Single Cell Sequencing
August 19, September 16

Website: http://michmed.org/agc
Email: advanced-genomics@umich.edu

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