



singulator 100

Automated solid tissue dissociation
for single cell 'omics analyses

Livermore, CA

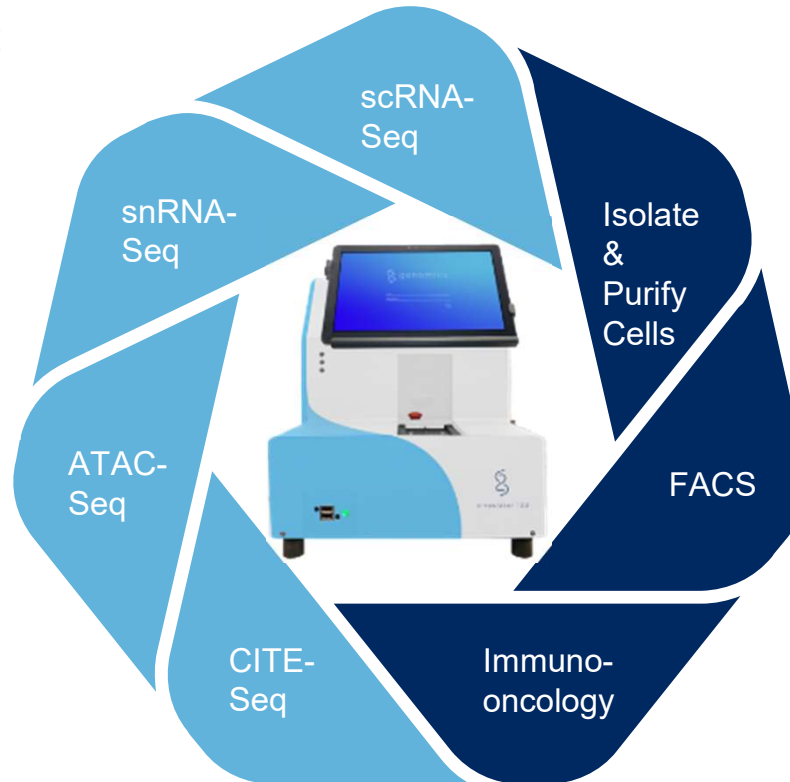
From Samples to Genomics

One tissue prep platform for omics applications

Single Cell Genomics:

Genomics Core Labs
Individual Researchers
Clinical Research Labs

Genomics



Stem Cell Biology:

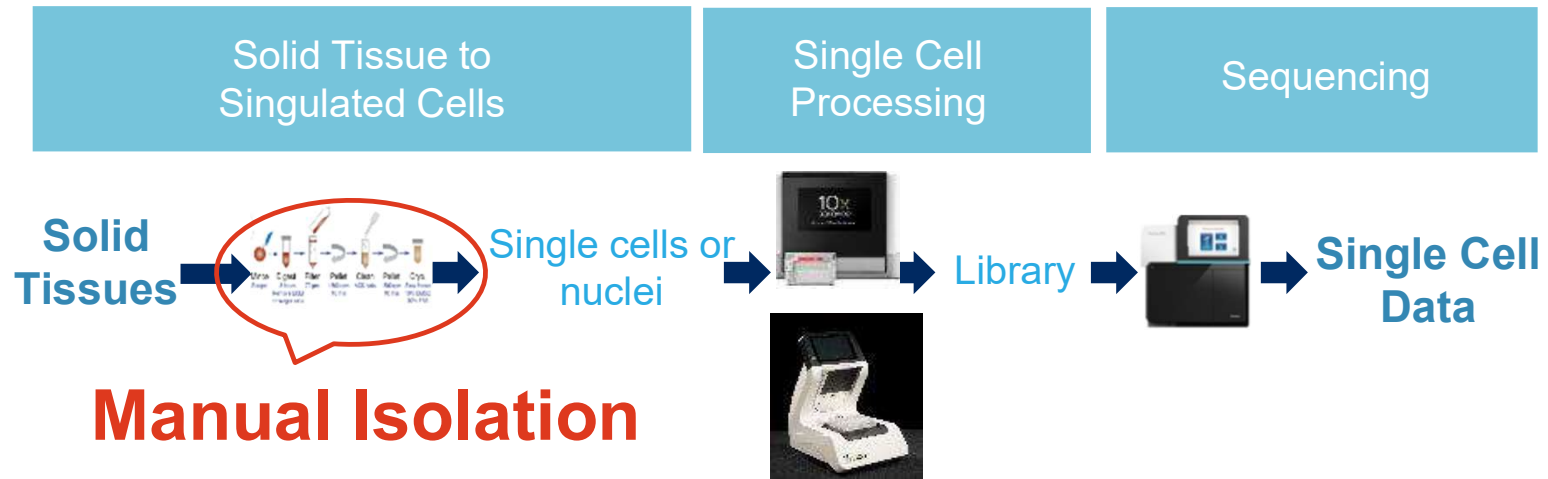
Biotechs
Clinical Research

Cell Biology

TILS or Tumor Cells:

Clinical Research Labs
Personalized Medicine
Cancer Vaccines

Manual cell isolation impairs single cell data quality



Cells

- Requires skilled experts
- No standard protocols
- Isolations affect transcriptome
- Biased cell-type representation

Nuclei

- Requires skilled experts
- Low, variable yields
- Compromised nuclear membranes
- Degraded chromatin

Sample Preparation Artifacts In → Data Artifacts Out

Simplify and Automate the Isolation Workflow

- Automated operation
- High quality cells and nuclei
- Customizable processes
- Temperature controlled
 - Cold operation for nuclei or cells
- Fast
 - Nuclei: 5 minutes
 - Cells: 20-60 minutes
- Samples below 10mg
 - (2mg supported 1/2022)



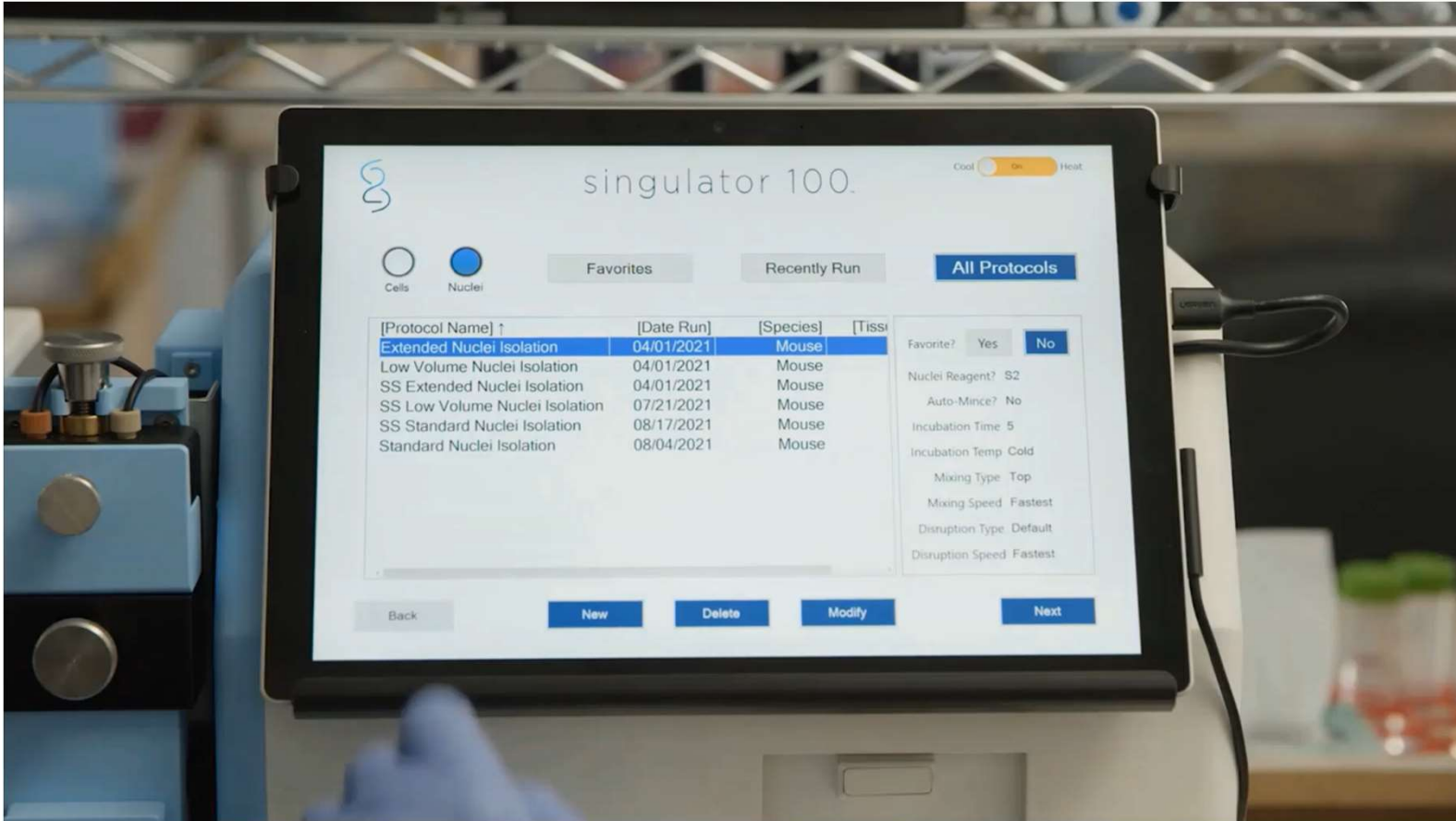
Genomics

- scRNA-Seq
- snRNA-Seq
- ATAC-Seq

Cell Biology

- FACS
- Organoids
- TILS
- Cell Health

Singulator Operation: Automated & Customizable



High Quality Cells and Nuclei

Heiko Yang, Franklin Huang, UCSF

Human Bladder Tumor

10x scRNA Seq Data: Singulator vs. Manual Cell Isolations

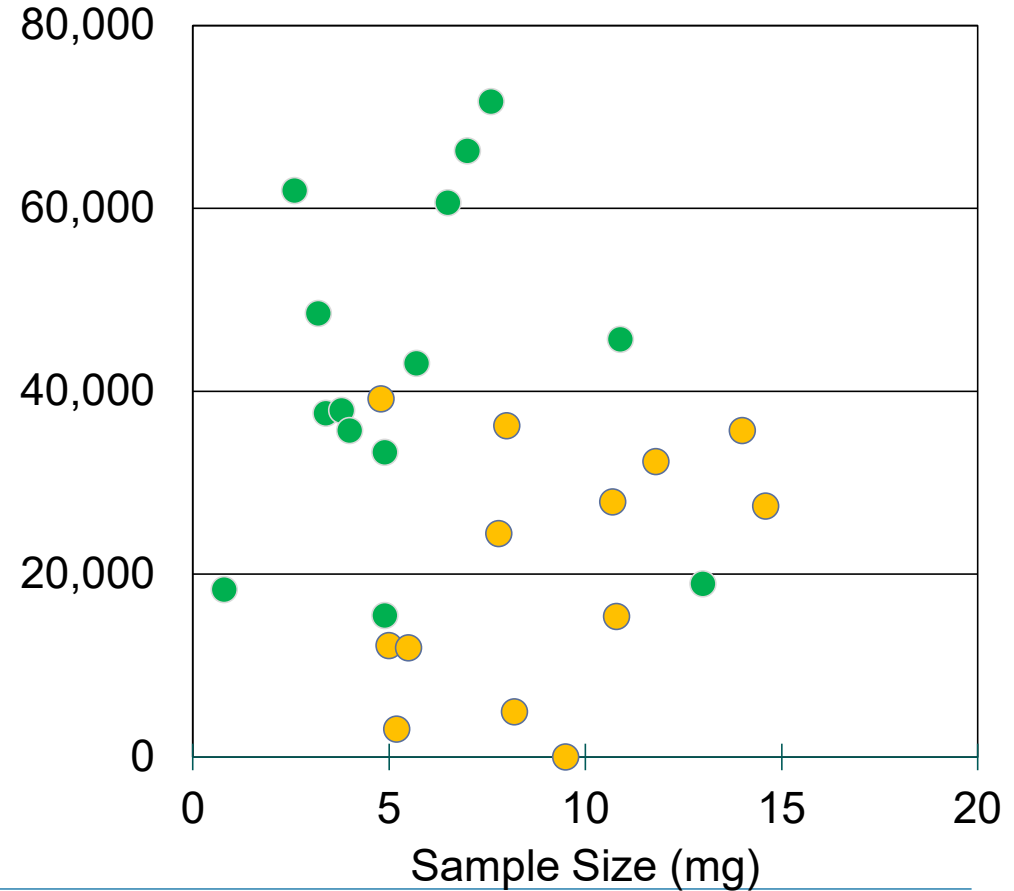
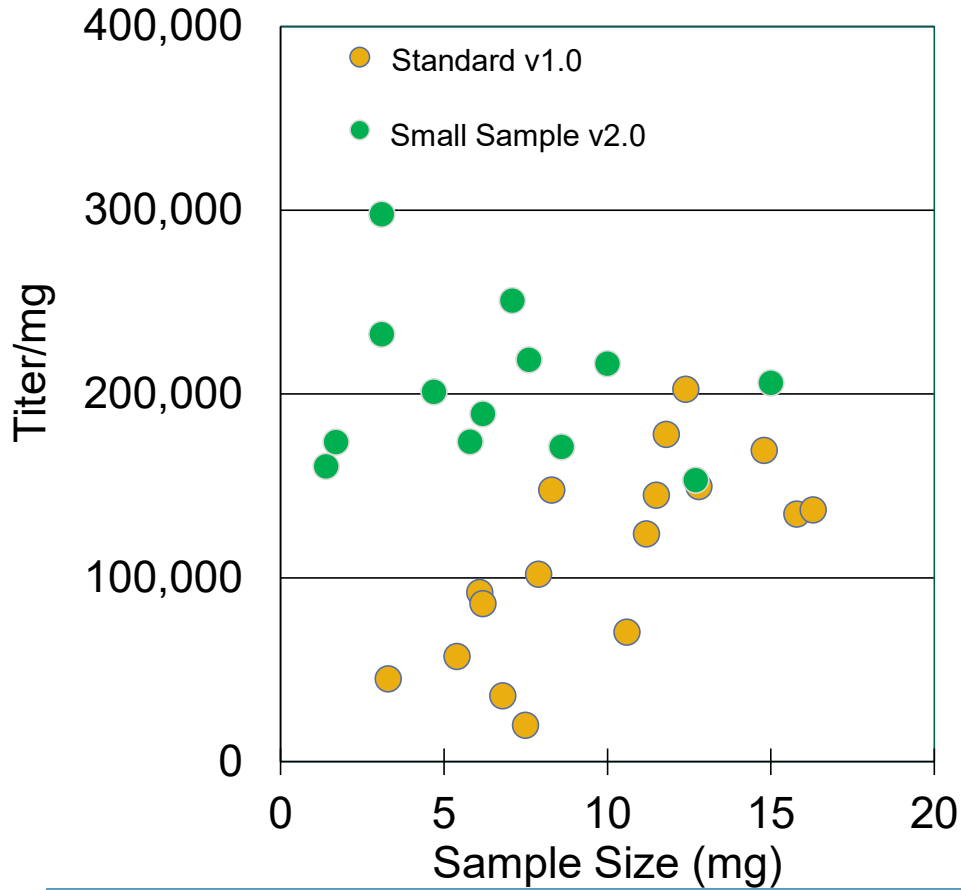
	Singulator	Manual
Initial Dissociation	226,000 cell, 96.8%	135,000 cells, 64.3%
Cells Captured	1185	889
After QC	1013	557
Yield	85.5%	62.7%

nFeature, nCount, % mt, % ribosomal all better with Singulator

Small Sample Processing

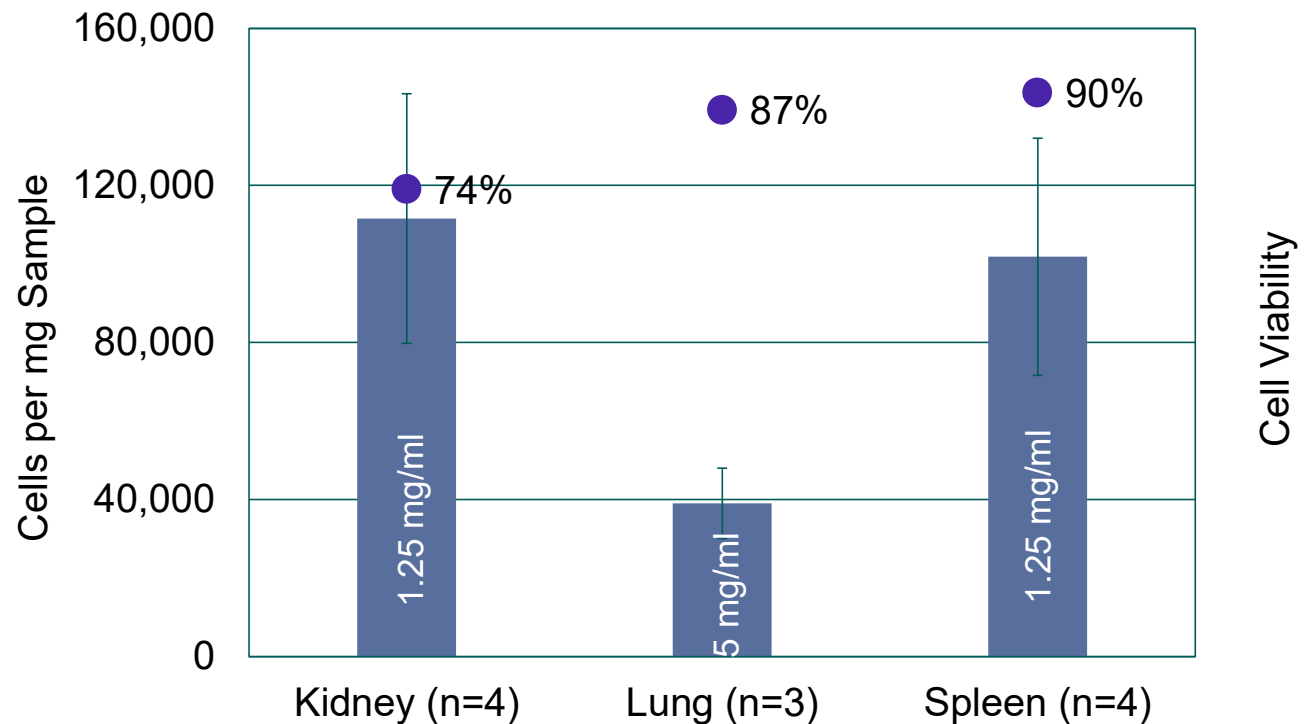
Mouse Kidney Nuclei

Mouse Lung Nuclei



Temperature Control: Cold Protocols for Cell Isolations

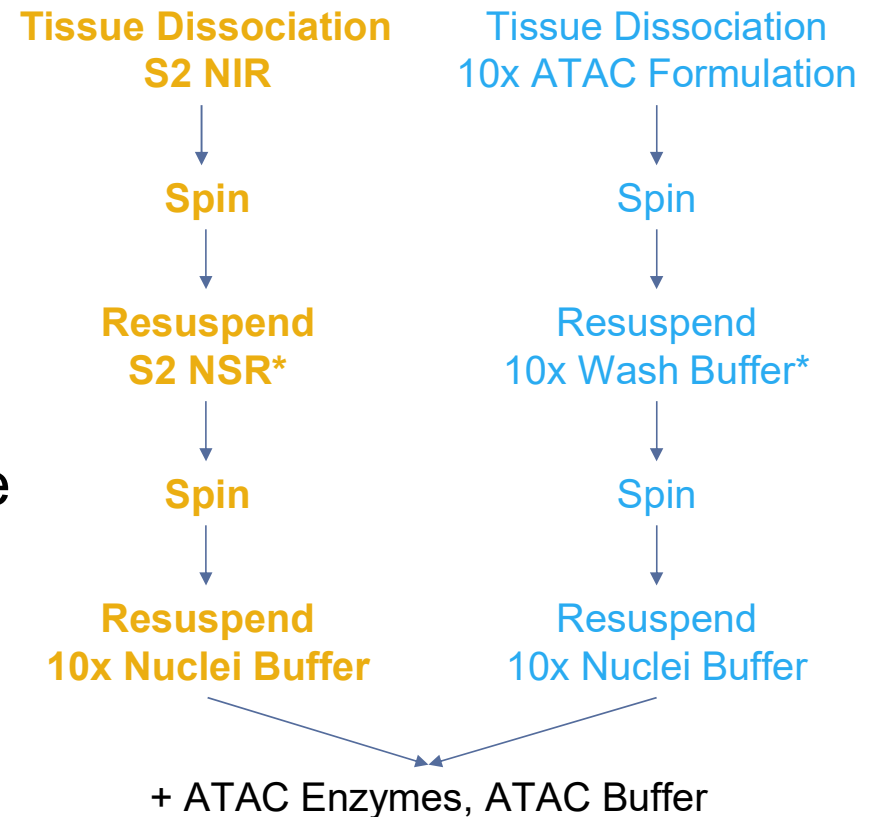
- Cold protocols same duration as 37°C
- Protease from *B. Licheniformis* (Sigma)
- Sequencing to confirm reduction in cell stress genes underway
- Dramatic reduction in enzyme over literature methods



ATAC SEQ: Impact of Dissociation Reagent

- Isolate Nuclei with the Singulator 100
 - S2 Genomics Reagent (NIR/NSR)
 - 10x ATAC (NP40/TWEEN/Digitonin)
- Compare ATAC data quality
- Fresh Mouse brain*, lung, kidney tissue

*Myelin removal in Percoll



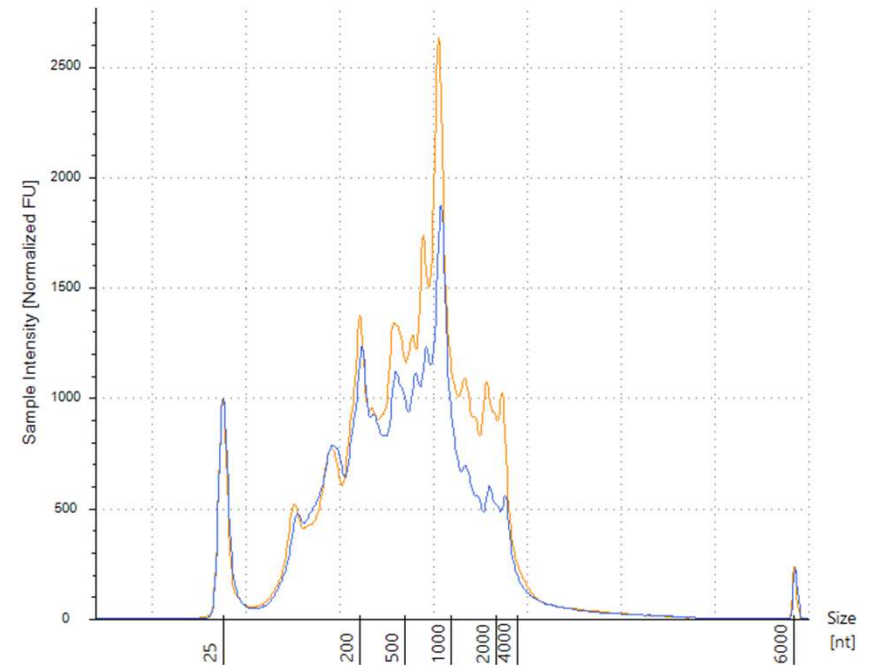
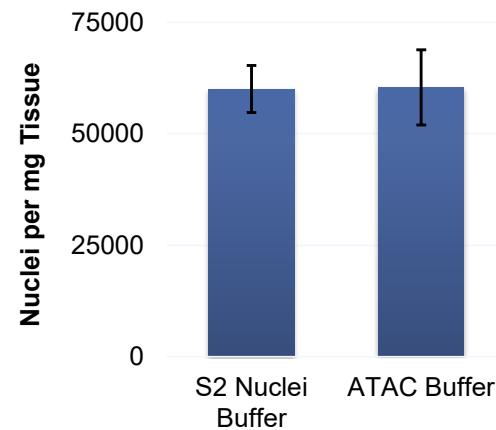
Nuclei Isolation and RNA Quality: Lung Tissue

- Standard Singulator Protocol
- +1u/ μ L Rnase inhibitor
 - 10x ATAC Formulation: 35mg
 - SG2 Genomics Reagents: 47mg

Reagents	DV200
S2	79%
10x	75%

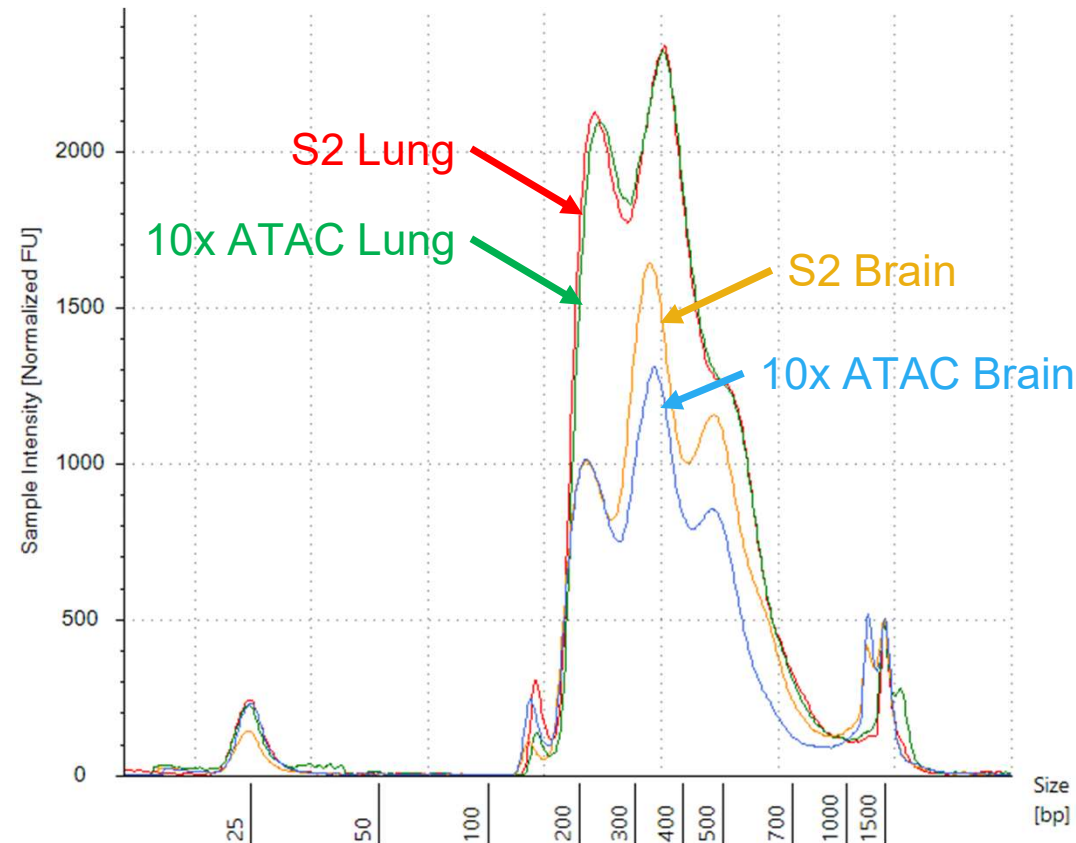


ATAC-Seq Buffer (40x)
Trypan Blue



Mouse Brain & Lung Library Quality

- **Comparing**
 - S2G Reagents vs. 10x ATAC Formulation
 - Tapestation Traces
 - S2 formulation has equivalent or better profiles

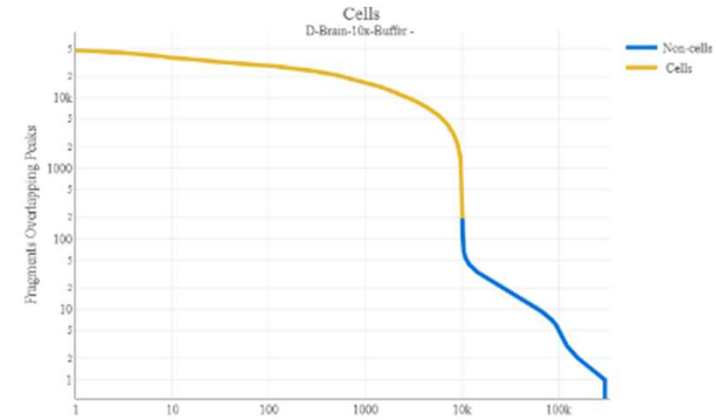
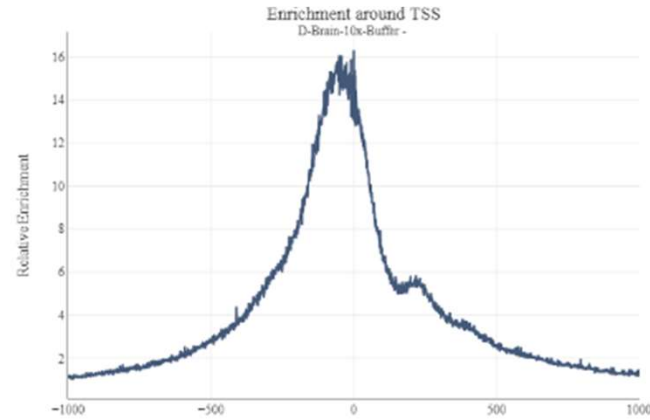
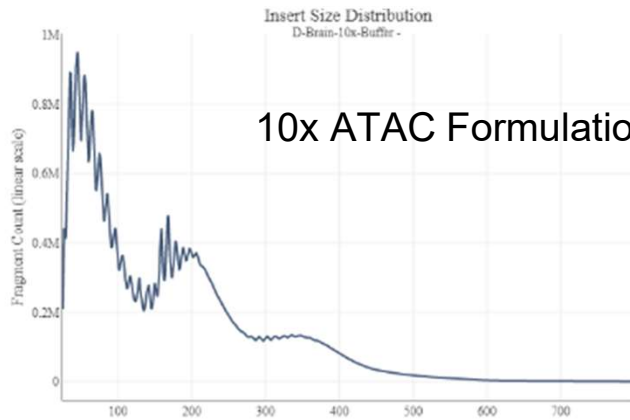


Mouse Brain & Lung ATAC Data

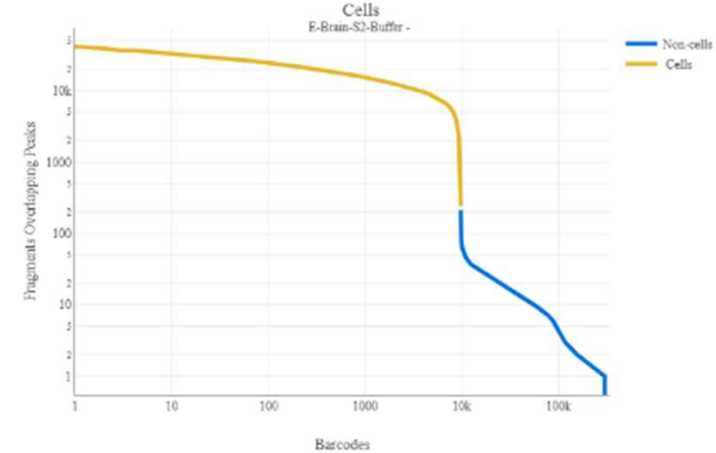
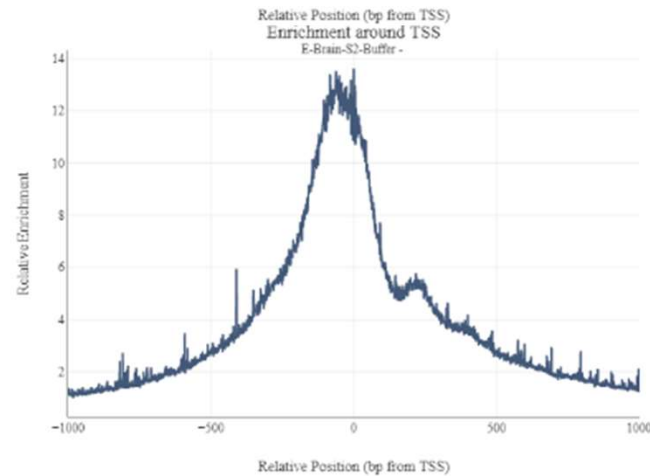
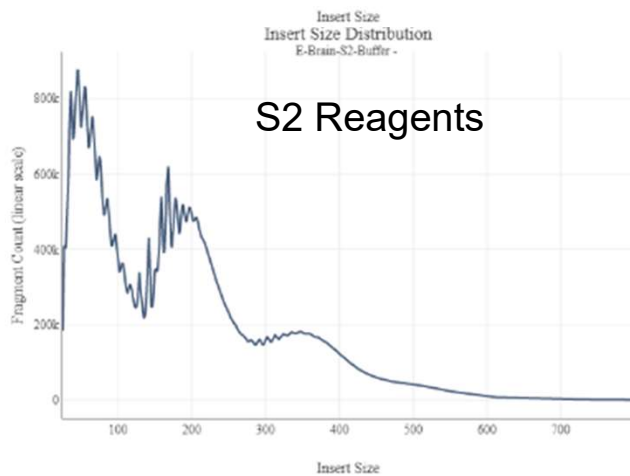
	S2 Brain	10x ATAC Brain	S2 Lung	10x ATAC Lung
Estimated # Cells	9,736	10,110	8,132	8,158
Median HQ Fragments/Cell	13,069	8,758	11,775	10,638
HQ Fragments Overlapping Peaks	56.4%	68.5%	49.1%	46.6%
Confidently mapped read pairs	92.9%	92.6%	88.3%	87.7%
Unmapped read pairs	2.7%	3.4%	2.2%	2.1%
Non-nuclear read pairs	0.3%	0.1%	1.3%	0.8%
Fragments in nucleosome-free regions	44.2%	51.6%	56.5%	57.6%
Fragments flanking a single nucleosome	36.4%	33.4%	31.5%	31.1%
TSS Enrichment Score	13.6	16.3	12.1	10.6
Fraction of HQ fragments overlapping TSS	34.8%	36.1%	31.1%	29.2%
Fraction of HQ fragments overlapping peaks	65.4%	68.5%	49.1%	46.6%

ATAC Seq Quality Metrics: Brain Tissue

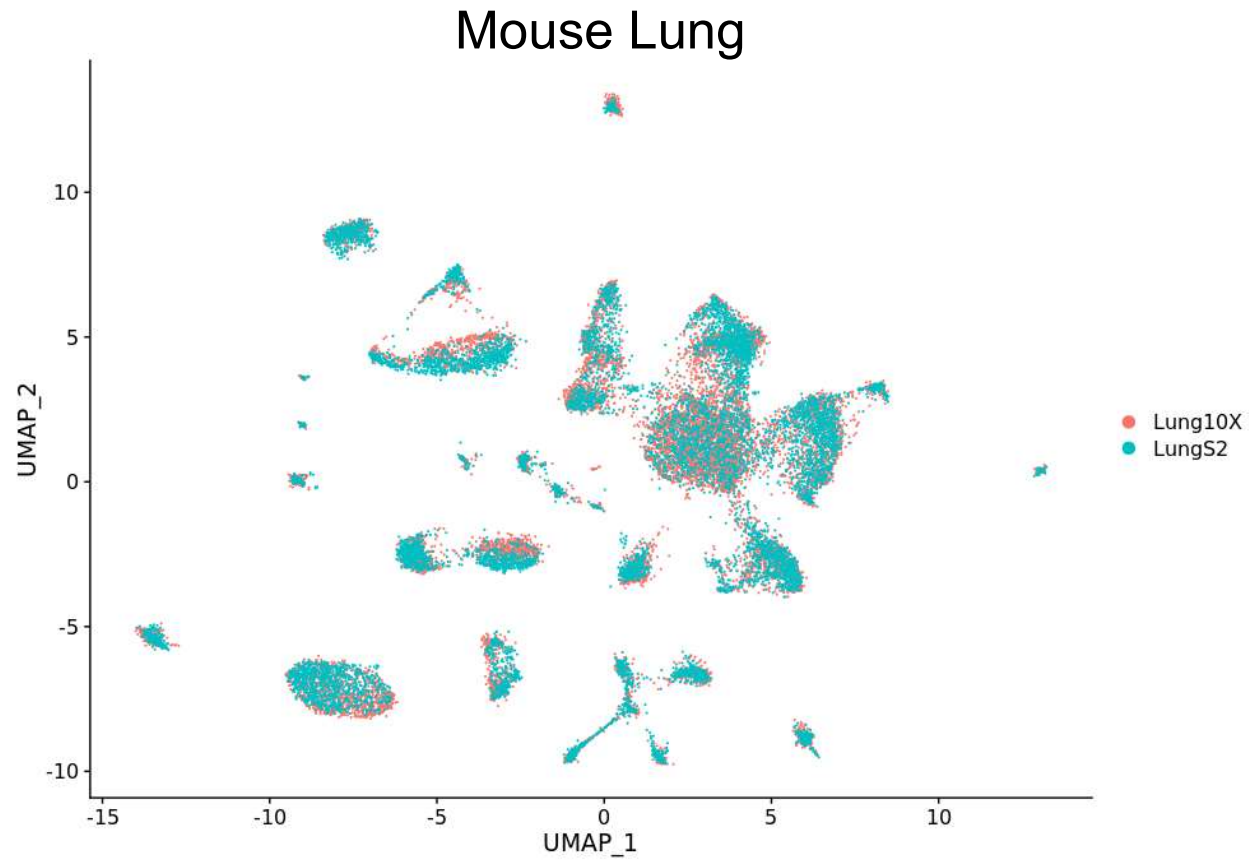
10x ATAC Formulation



S2 Reagents



Comparative Cell Clustering



Cells: Success with a wide range of tissues

Human

Colon

Colon Polyp

Colon Tumor

Neck Tumor

Colorectal Carcinoma

Bladder Tumor

Planaria

Whole

Cut

Mouse

Brain

Kidney

Liver

Lung

Spleen

Ovaries

Snail

Eye

Embryo

Intestine

Colon PDX Tumor

Lung PDX Tumor

Pancreatic Tumor

Rat

Brain

Kidney

Liver

Lung

Spleen

Zebrafish

Brain

Liver

Italics denotes processing in a customer's lab

Nuclei: Success with a wide range of tissues

Human

Aorta
Brain: Adult, Infant, Fetal
Breast Tumor
Colon: Normal, Polyps, Tumor
Heart - Adult, Fetal
Hemangioma
Hepatoblastoma
Intestine – Fetal, Adult
Kidney (biopsy)
Lung – Fetal
Muscle: TA and SA Muscles
Organoids: Retinal, Cerebral
Prostate: Normal, Tumor
Spleen – Fetal
Thymus – Fetal
Vascular Abnormality: Arterial, Lymphatic
PBMC's

Mouse

Brain
Colon PDX Tumor
Heart
Intestine
Kidney
Liver
Lung
Muscle
Pancreatic Tumor
Spinal Cord
Spleen

Rat

Brain
Kidney
Liver
Lung
Spleen

Acomi

Kidney

Honeybee

Thorax

Drosophila

Brain
Larvae
Ovaries

A. Thaliana

Whole Seedling
Root
Leaves

Italics denotes processing in a customer's lab

Automation with the Singulator improves results

- Immediate production of high-quality cells or nuclei
 - Increased viability
 - Reduced failures
 - Frozen tissue biobank samples for nuclei
- Automated filtration eases downstream cleanup
- Standardizes front-end process to reduce failures
 - Easy process optimizations
 - Precious samples
 - Costly downstream sequencing
 - Enables access to flash frozen tissue banks
- Works well on small samples
 - ~2mg with v2.0 cartridges
- Temp control for 6°C, RT, 37°C
 - Cold protease protocols
- Open reagent platform



Dr. John Bashkin

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For more information, visit www.S2Genomics.com