

OptoSeq™ Single Cell 3' mRNA Kit

The OptoSeq Single Cell 3' mRNA kit integrates single cell sequencing library preparation into the Opto™ Cell Therapy Development workflow on the Beacon® and Lightning™ optofluidic systems. Live cell phenotypic data can now be directly linked to differential gene expression to reveal genetic drivers behind effective therapeutic T cells.

KIT OVERVIEW

The OptoSeq Single Cell 3' mRNA kit includes all of the reagents required for on-chip cell lysis, mRNA capture, reverse transcription and barcode generation followed by off-chip cDNA amplification for single cells after they have been characterized on a Berkeley Lights system. The OptoSeq 3' mRNA kit can be performed on up to two OptoSelect™ 3500 chips on the Beacon system and on one 1500 chip on the Lightning system. Each kit provides enough reagent to perform up to four runs on four total chips on either system.

After a functional assay is performed, mRNA capture beads are loaded into NanoPen™ chambers. To initiate 3' mRNA capture single cells are lysed (**Figure 1A**). Captured mRNA is then reverse transcribed and bead barcodes are probed with complementary fluorescent oligos and imaged (**Figure 1B**). Beads with non-redundant barcode sequences are exported off-chip in batches for second strand cDNA amplification and library preparation. Following sequencing, PrimeSeq™ software is used to analyze the sequence data. The proprietary capture bead barcodes

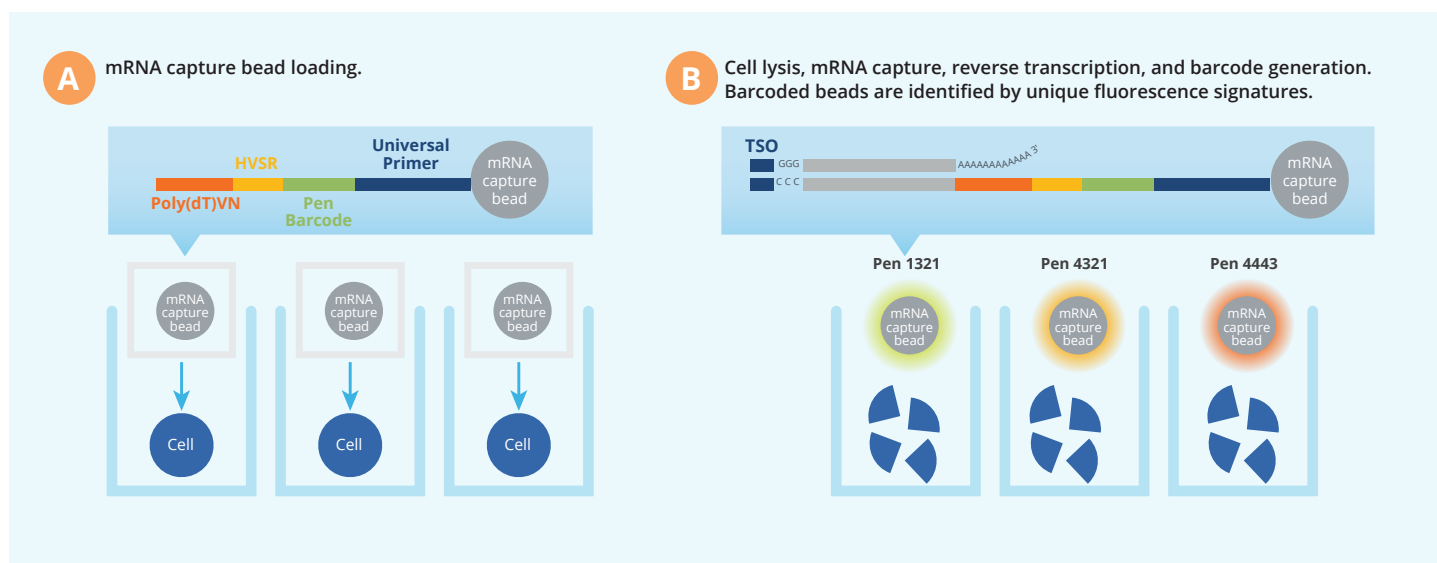


Figure 1. After characterization, the OptoSeq Single Cell 3' mRNA kit enables live cells of interest to be prepared for genetic analysis via cell lysis, mRNA capture, reverse transcription, and barcode association.

provide unique fluorescence signatures that allow PrimeSeq software to directly link the phenotypic history of a single cell in a specific NanoPen chamber to gene expression data associated with specific phenotypes (**Figure 2**).

Possible data analyses include identifying genes associated with phenotypes, generating and filtering gene expression data, and creating UMAP, volcano, and MA plots to visualize differential gene expression.

This powerful transcriptomic analysis, coupled with functional characterization, creates a direct link between phenotypic data and gene expression. This application has the potential to support development of more effective cell therapies, because the genetic drivers of ideal T cell therapeutics can now be fully understood.

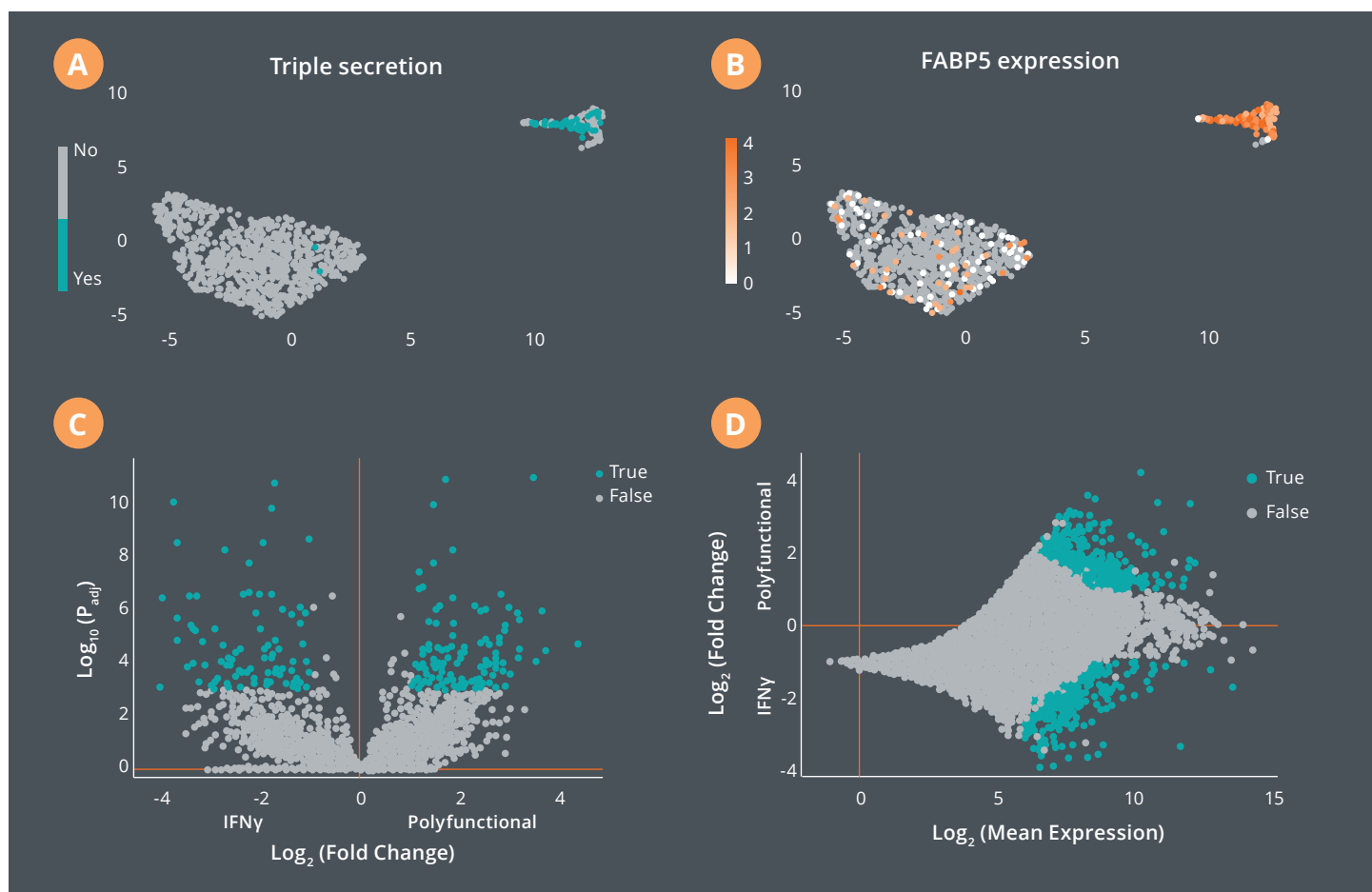


Figure 2. PrimeSeq software provides a powerful way to visually differentiate cell phenotypes by gene expression downstream from functional characterization.

KIT CONTENTS

DESCRIPTION	QUANTITY	VOLUME/UNIT	STORAGE CONDITIONS
OptoSeq™ Single Cell 3' mRNA Kit, Box 1			
RNase Inhibitor	4 vials	100 µL	-20°C
Probe Mix 1	1 vial	12 µL	
Probe Mix 2	1 vial	12 µL	
Probe Mix 3	1 vial	12 µL	
Probe Mix 4	1 vial	12 µL	
Cell Lysis Buffer	1 vial	60 µL	
RT Enzyme	1 vial	20 µL	
PCR Enzyme T	1 vial	160 µL	
RT Master Mix	1 vial	105 µL	
mRNA Primer 1	1 vial	110 µL	
mRNA Primer 2	1 vial	530 µL	
PCR Buffer TX	2 vials	2 mL	
OptoSeq™ Single Cell 3' mRNA Kit, Box 2			
Wetting Additive	2 vials	1.5 mL	4°C
mRNA Capture Beads	4 vials	400 µL	
Buffer T	3 bottles	50 mL	
Buffer NW	1 vial	2 mL	
OptoSeq™ Single Cell 3' mRNA Kit, Box 3			
Buffer P+	2 bottles	110 mL	4°C
Buffer S	2 bottles	20 mL	

ORDERING INFORMATION

PART NUMBER	DESCRIPTION
750-03000	OptoSeq™ Single Cell 3' mRNA kit
110-08004	Beacon® optofluidic system
110-02407	Lightning™ optofluidic system

REQUIRED SOFTWARE

SOFTWARE	VERSION
CAS™ software	Cell Analysis Suite software v2.2.0 or later
Assay Analyzer	Assay Analyzer v2.2 or later
PrimeSeq™ software	PrimeSeq analysis software v2.2.0 or later

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FOR MORE INFORMATION, VISIT
berkeleylights.com/cell-therapy-development

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5858 Horton Street | Suite 320 | Emeryville, CA 94608

info@berkeleylights.com

+1-510-858-2855

berkeleylights.com

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