

Single Cell Sequencing Using the Parse Biosciences Evercode™ WT Kit on the G4™ Sequencing Platform

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Background

Single cell RNA sequencing (scRNA-Seq) has revolutionized basic and translational research in immunology, developmental biology, and cancer by enabling the resolution of distinct cell populations within heterogeneous samples. However, there remains a need for scRNA-Seq library preparation technologies that support greater cell throughput and sensitivity with lower background noise. Equally, there is a need for next generation sequencing (NGS) platforms that combine speed with flexible throughput to provide cost-effective sequencing while reducing batching related delays. In this work, we describe the application of the Parse Biosciences Evercode WT kit with the Singular Genomics G4™ Sequencing Platform to enable rapid, flexible, and accurate scRNA-seq.

Methods

Evercode WT scRNA-Seq libraries were prepared from previously fixed and frozen peripheral blood mononuclear cells (PBMCs) of four healthy donors. A 15,000 cell sublibrary was split for both sequencing arms of the study. One library was converted for sequencing on the G4 platform using the Singular Genomics library conversion kit with non-indexed PCR primers, then sequenced on the G4 instrument using a single F2 flow cell to yield 251M paired reads. The Illumina library was sequenced on the NovaSeq 6000 using two lanes of an S4 flow cell, then downsampled to 251M reads prior to downstream processing of each dataset using Parse Biosciences data analysis solution.

Results

Clustering revealed nearly identical grouping of single cell transcriptomes across platforms, with strongly correlated pseudo-bulk transcription profiles ($R^2 = 0.9932$). Likewise, CellTypist unsupervised clustering and automated cell type annotation yielded nearly identical cell type abundance across the platforms, with cell population frequencies and gene expression profiles matching expectations (Adjusted Rand Index= 0.9932).

Conclusions

scRNA-Seq data generated using the Parse Biosciences Evercode WT kit with the G4 Sequencing Platform demonstrates high accuracy, sensitivity and reproducibility. Combined with the rapid turnaround and flexible throughput of the G4, we expect the Parse Biosciences Evercode WT kit to accelerate single cell research.