mission bio

tapestri
APPLICATION NOTE

BGI Sequencing Platforms Produce High-Quality Performance Metrics from Tapestri Single-cell Experiments

Takeaways

- Tapestri single-cell DNA libraries are compatible with BGI sequencing platforms
- Tapestri BGI sequencing data show high concordance with Illumina sequencing data across key performance metrics
- Tapestri single-cell DNA libraries enable accurate variant calling and subclone identification

Abstract

Next-generation sequencing (NGS) technology is the gold standard for genomics studies in a variety of clinical and research applications. Although Illumina sequencers dominate the NGS market, platforms developed by Beijing Genomics Institute (BGI) are rapidly growing in popularity. Here, we show that BGI and Illumina sequencing platforms are equally capable of Tapestri single-cell DNA sequencing. To this end, we designed a straightforward experiment that compares each sequencer's ability to profile single-cell genotypes in a 1:1 mixture of two cell lines. Data processing and single-cell analyses using Tapestri Pipeline and Tapestri Insights software, respectively, showed high concordance between the two sequencing platforms at every level of single-cell analysis (primary, secondary, and tertiary). These findings show that these sequencers can be used equivalently for single-cell DNA mutational profiling with the Tapestri Platform.

Experimental Methods and Results

In this study, we sequenced Tapestri single-cell DNA libraries using two sequencing platforms: an Illumina NextSeq 2000 and a BGI MGISEQ DNBSEQ-G400 (Figure 1). The libraries were generated from a 1:1 mixture of RAJI and K562 cells obtained from ATCC using the Mission Bio catalog Acute Myeloid Leukemia (AML) panel (MB03-0016), which covers genetic variants specific to each cell line. The goal was to compare the ability of each platform to sequence and profile the cell mixture in light of Illumina's established compatibility with the Tapestri Platform and BGI's rising popularity in the NGS market.

Table 1 summarizes the sequencing metrics obtained after data processing and analysis with Tapestri Pipeline and Tapestri Insights software. These metrics were highly concordant across the two sequencing platforms. Both sequencers produced enough total read pairs for robust single-cell genotype calling in downstream analyses. In the secondary analysis, the number of cells called and the panel uniformity were identical across the two platforms. In the tertiary analysis, both platforms called 36 pass-filter (PF) variants with comparably low allele dropout (ADO) rates. The performance metrics for both sequencers aligned with those specified in the AML Panel Data Sheet, indicating that both are suitable for sequencing Tapestri single-cell libraries.

To determine whether each platform could successfully identify cells and their respective genetic variants, we generated UMAP projections

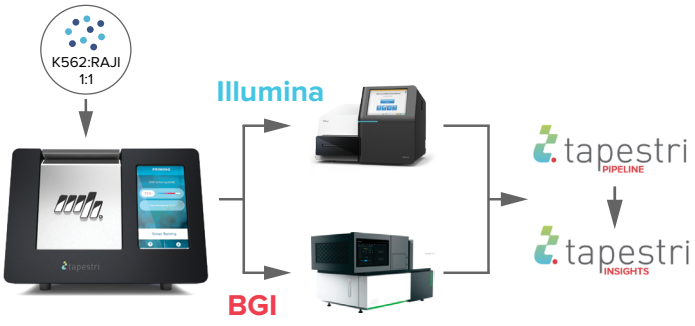

Figure 1 - Schematic overview of experimental workflow.

Table 1 - Primary, secondary, and tertiary analysis metrics.

Stage of NGS Analysis	Performance Metric	Illumina	BGI
Primary Analysis	Total PF Read Pairs (millions)	32.6	37.2
Secondary Analysis	Cell Number	2,484	2,484
	Panel Uniformity	96.9%	96.9%
	Coverage (per cell and amplicon)	83X	78X
Tertiary Analysis	PF Variants	36	36
	ADO Rate	2.5%	2.7%

of cells combined from the BGI and Illumina runs using Tapestri Insights software. This analysis revealed two cell populations, one corresponding to RAJI cells and the other to K562 cells. Both populations were consistently identified by the two sequencing platforms, as demonstrated by the overlap between BGI and Illumina associated cells (Figure 2A, left). The identity of these populations was confirmed by cell line-specific mutant variants in the *TET2* and *KIT* genes (Figure 2A, right). In addition, using hierarchical clustering and heatmap projections, the overall variant mutation patterns of each cell line were concordant across both platforms (Figure 2B). Thus, both sequencers were able to successfully profile single-cell genotypes with equal performance.

Conclusions

This application note demonstrates that BGI and Illumina sequencers perform equivalently when used to sequence Tapestri single-cell libraries. The performance of both sequencers was identical across several metrics, including the number of cells called, panel uniformity, the number of PF variants

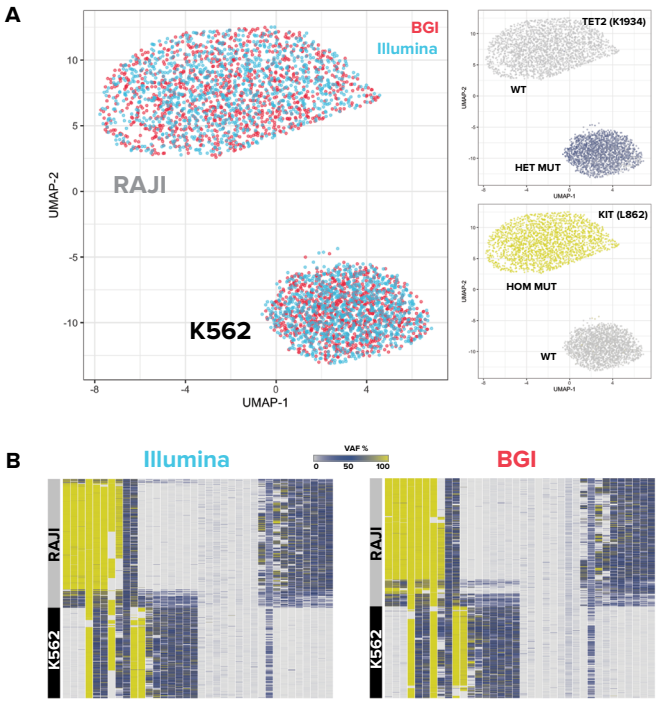

Figure 2 - UMAP projections of cells from both the Illumina and BGI sequencing runs (A) illustrate differentiation between RAJI and K562 cells, extensive overlap of BGI and Illumina cells (left), and cell type-specific variants known to be mutant in each cell type (right). Hierarchical clustering (B) of all cells and PF variants identified both cell types (rows) and variant mutation patterns (columns) for Illumina (left) and BGI (right).

detected, and the number of cell clusters identified. Differences in other sequencing read-associated metrics were negligible and did not meaningfully impact downstream analyses. In aggregate, these data concur with published comparisons showing highly similar performance between the two sequencing technologies.[1,2] Overall, Tapestri single-cell DNA libraries enable accurate variant calling and subclone identification regardless of whether they are sequenced with BGI or Illumina sequencers.

References

1. Senabouth et al., Comparative performance of the BGI and Illumina sequencing technology for single-cell RNA-sequencing, NAR Genomics and Bioinformatics, 2(2):lqaa034 (2020).
2. Natarajan et al., Comparative analysis of sequencing technologies for single-cell transcriptomics. Genome Biology, 20(1):70 (2019).