Efficient and sensitive high-throughput mouse immune repertoire profiling using SMART technology

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Objective: Immune receptor (BCR/TCR) repertoire profiling for biomarker discovery of host B cell and T cell responses are revealing more about the roles B cells and T cells play in viral and bacterial infections, autoimmune disorders, and cancer. Mice are one of the most common animal models used for both BCR - and TCR profiling. However, current technologies are limited in their ability to generate data accurately and reproducibly for all mouse BCR isotypes and for mouse TCR alpha and beta chains. To address this need, we developed two new mouse immune profiling kits: one to profile heavy and light-chains of all mouse BCR isotypes; the other to profile mouse TCR alpha and beta chains.

Methods: Both kits were developed as end-to-end solutions, streamlining library preparation to data analysis. Libraries were prepared using RNA purified from mouse spleens using our new mouse TCR repertoire profiling kit (1 ng – 1 ug purified mouse spleen RNA) or mouse BCR profiling kit (10 ng – 1 ug purified mouse spleen RNA). BCR and TCR libraries were sequenced on the Illumina® Miseq® benchtop sequencer with 300-bp paired-end reads. TCR libraries were sequenced on the Illumina® MiniSeq® and NextSeq® benchtop sequencer with 150-bp paired-end reads. Sequencing data were analyzed using our Cogent™ NGS Immune Profiler Software and MiXCR v3.

Results: Both the updated mouse TCR and BCR sequencing kits achieved a high on-target map rate across all RNA inputs tested. The updated mouse TCR sequencing kit demonstrated sequencing flexibility as clonotype counts were similar whether we generated full length (300bp x 2) or CDR3 only (150bp x 2) reads. The updated BCR-sequencing kit generated ~5x higher total clonotype count across various RNA inputs than the previous version of the kit. The most common clonotype was found to be consistent among technical duplicates regardless of the RNA input amount.

Conclusion: Our new mouse TCR profiling kit and mouse BCR profiling kit were observed to accurately and reproducibly profile T-cell and B-cell receptor sequences, respectively, and provide information on the diversity of immune repertoires in mouse samples.