

AmpliSeq™ for Illumina Immune Repertoire Plus, TCR beta Panel

Targeted RNA panel for characterizing T cell receptor diversity.

Highlights

- High-value gene content**
 Obtain comprehensive coverage of T cell receptor beta chain sequences
- Fast, streamlined workflow**
 Prepare sequencing-ready libraries in a single day from whole blood, fresh/frozen tissue, or fluorescence-activated cell sorting (FACS) cells
- Low input requirement**
 Generate high-quality sequencing libraries from as little as 10 ng of input RNA

Introduction

T cell receptor (TCR) is a transmembrane heterodimer that enables T cells to recognize and respond to foreign "nonself" material. The vast majority of TCRs consist of an alpha and beta chain that contain complementary determining regions (CDRs). T cell receptor (TCR) diversity describes the potential of a small set of genes encoding the CDRs within the TCR to create in excess of 10^{12} T cell clonotypes (populations of T cells expressing identical TCRs) by recombination, random insertion, deletion, and substitution.¹ TCR diversity plays a vital role in host defense. Investigation of this diversity may be useful in understanding immune function, autoimmune diseases, and immune-mediated adverse effects (IMAEs).

The AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel is a highly multiplexed targeted resequencing panel designed to measure T cell diversity and clonal expansion by sequencing TCR beta chain rearrangements (Table 1). Using a single pool of multiplex PCR primers, library reagents, and sample barcodes, libraries can be generated from RNA extracted from whole blood, fresh/frozen tissue, or FACS-sorted cells for sequencing on compatible Illumina sequencing systems.

High-value gene content

The AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel offers coverage of the TCR beta chain, with ~ 325 bp read-length amplicons for comprehensive characterization of all three CDRs (CDR1, CDR2, and CDR3). This ready-to-use panel saves researchers the time and effort of identifying targets, designing amplicons, and optimizing performance.

Table 1: AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel at a glance

Parameter	Specification
No. of genes	Variable
Targets	TCR beta chain, including CDR1, CDR2, and CDR3
Amplicon size	~ 325 bp
No. of amplicons	Variable
Input DNA/RNA requirement	10–1000 ng
No. of pools per panel	1
Supported sample types	Blood, fresh/frozen tissue, FACS cells
Total assay time ^a	5–6 hours
Hands-on time	< 1.5 hours
DNA-to-data time	2.5 days

a. Time represents library preparation only and does not include library quantification, normalization, or pooling

Simple, streamlined workflow

The AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel is part of an RNA-to-results solution that offers easy-to-perform library preparation, push-button sequencing systems, and simplified data analysis.

Library preparation begins with the conversion of total RNA to cDNA, followed by a straightforward, PCR-based protocol that can be completed in as little as 5 hours, with < 1.5 hours hands-on time. Resulting libraries can be normalized, pooled, and then loaded on to a flow cell for sequencing. Prepared libraries are sequenced using proven sequencing by synthesis (SBS) chemistry on a compatible Illumina sequencing system (Table 2).

Resulting data can be easily streamed into BaseSpace™ Sequence Hub for analysis. The MiXCR Immune Repertoire Analyzer app in BaseSpace Sequence Hub enables fast and accurate processing of sequencing data from T and B cell receptor libraries. It aligns reads against germline segments, assembles clonotypes, and corrects for PCR and sequencing errors. Output provides detailed information on germline segment assignments, alignment, and mutations.

Table 2: Illumina sequencing systems recommended for use with the AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel

Instrument
MiniSeq™ System (mid output)
MiniSeq System (high output)
MiSeq™ System (v2 chemistry)
MiSeq System (v3 chemistry)
NextSeq™ 550 System

Accurate data

The AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel enables investigation of TCR diversity in immune function. To demonstrate assay capabilities, samples were analyzed using the AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel, the NextSeq 550 System, and the MiXCR Immune Repertoire Analyzer app. Results show that samples with productive immune repertoire clones (leukocytes and Jurkat cells) have high read utilization, while a sample type with little to no expression of immune repertoire targets (brain tissue) has low read utilization (Figure 1).

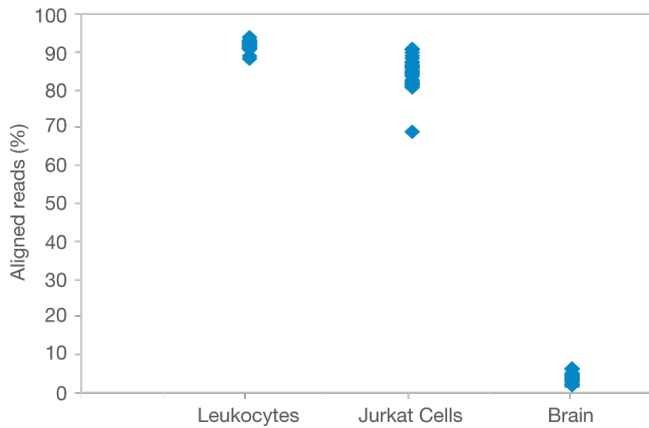


Figure 1: Productive read utilization—Libraries were prepared with the AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel, sequenced on the NextSeq 550 System and analyzed with the MiXCR app. Input DNA from productive immune repertoire clones (leukocytes, Jurkat cells) show high read utilization, in contrast to input DNA from a sample with little to no expression of immune repertoire targets (brain tissue), which shows low read utilization. Error bars indicate variability of technical replicates.

Learn More

To learn more about the AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel, visit www.illumina.com/products/by-type/sequencing-kits/library-prep-kits/ampliseq-immune-repertoire-panel.html

To learn more about the AmpliSeq for Illumina targeted resequencing solution, read the overview at www.illumina.com/content/dam/illumina-marketing/documents/products/datasheets/ampliseq-for-illumina-targeted-resequencing-solution-data-sheet-770-2017-022.pdf

Ordering Information

Order AmpliSeq for Illumina products online at www.illumina.com

Product	Catalog no.
AmpliSeq for Illumina Immune Repertoire Plus, TCR beta Panel (24 reactions)	20024479
AmpliSeq for Illumina Library PLUS (24 reactions)	20019101
AmpliSeq for Illumina Library PLUS (96 reactions)	20019102
AmpliSeq for Illumina Library PLUS (384 reactions)	20019103
AmpliSeq for Illumina CD Indexes Set A (96 indexes, 96 samples)	20019105
AmpliSeq for Illumina cDNA Synthesis (96 reactions)	20022654

References

1. Laydon DJ, Bangham CRM, Asquith B. *Estimating T-cell repertoire diversity: limitations of classical estimators and a new approach.* *Phil. Trans. R. Soc. B.* 2015;370(1675):20140291.