Certificate of Analysis

ICELL8® TCR Chip

Catalog No.	Amount	Lot Number
640178	1 chip	Specified on product label.

Description

The ICELL8 TCR Chip is part of the ICELL8 Human TCR a/b Profiling workflow, which enables users to analyze T-cell receptor (TCR) diversity from single T cells using the ICELL8 Single-Cell System. Each well in the 5,184-nanowell chip contains a preprinted barcode. Each ICELL8 TCR Chip contains 1,728 unique barcodes, and each barcode is printed three times on the chip. CellSelect® Software is used to detect single-cell-bearing wells and direct nanowell-specific delivery of coupled SMART® RT-PCR reagents to only those wells on the ICELL8 TCR Chip that have been selected for further processing. Barcoded cDNA from the selected wells are PCR-amplified in-chip. The full-length barcoded amplicons are then pooled off-chip, and the purified cDNA is used as template in the first TCR-specific PCR. The product from the first TCR-specific PCR is used as a template in a second TCR-specific PCR to incorporate Illumina® indexed primers. Most importantly, the kit allows for efficient, cost-effective, high-throughput single-cell capture of complete V(D)J variable regions of TCR transcripts.

Package Contents

- 1 x ICELL8 TCR Chip

Storage Conditions

- Store at –20°C.

Shelf Life

- Specified on product label.

Shipping Conditions

- Dry ice (–70°C)

Product Documents

Documents for our products are available for download at **takarabio.com/manuals**
The following documents apply to this product:

- ICELL8 Human TCR a/b Profiling User Manual
- ICELL8 Imaging System User Manual
- ICELL8 MultiSample NanoDispenser User Manual
- CellSelect Software User Manual

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Quality Control Data

Three chips from each lot were tested as follows:

Chip 1 is tested to ensure appropriate imaging quality performance: 1X PBS was dispensed in wells of a chip. The wells in the chip were then imaged in the Hoechst channel using a ICELL8 Single-Cell System and analyzed using the CellSelect Software. A count of wells positive for images in Hoechst was made and compared to the total number of wells assayed. The percent of wells showing images positive for Hoechst was <5%.

Chip 2 is tested to ensure barcodes are yielding appropriate reads per barcode: 5 pg of Jurkat Total RNA in 1X PBS was dispensed in the 1,728 wells of a ICELL8 TCR Chip containing unique barcodes. The RT-PCR reagents are dispensed into 1,728 wells containing 5 pg of Jurkat Total RNA. The chip was processed as per the ICELL8 Human TCR a/b Profiling User Manual. The concentration of the cDNA was confirmed to be ≥4 ng/µl with an average size >600 bp. 500 pg of cDNA was used as template following two rounds of PCR as per the user manual. The concentration of the library was confirmed to be >4 nM. The resulting library was then pooled with the library from Chip 3. Pooled libraries were denatured and diluted to 13.5 pM with 5% PhiX then sequenced on a MiSeq® Sequencer using the 150-cycle (Read 1 = 75 cycles, Read 2 = 75 cycles, Index 1 = 8 cycles) MiSeq Reagent Kit v3 (Illumina, Cat No. MS-102-3001). Total reads per library were measured at >8 x 10^6 with sufficient median diversity per barcode measured at >90 normalized reads per barcode. >95% of barcodes must be functional with >0.2 normalized reads per barcode observed.

Chip 3 is tested to ensure barcode locations are as expected: 5 pg of Jurkat Total RNA in 1X PBS was dispensed into specific wells of a ICELL8 TCR chip. These wells are unique and should yield reads with expected barcodes only. The RT-PCR reagents were dispensed into 180 wells containing 5 pg of Jurkat Total RNA. The chip was processed as per the ICELL8 Human TCR a/b Profiling User Manual. The concentration of the cDNA was confirmed to be ≥0.5 ng/µl with an average size >600 bp. 500 pg of cDNA was used as template following two rounds of PCR as per the user manual. The concentration of the library was confirmed to be >4 nM. The resulting library was then pooled with the library from Chip 2. Pooled libraries were denatured and diluted to 13.5 pM with 2% PhiX and sequenced on a MiSeq Sequencer using the 150-cycle (Read 1 = 75 cycles, Read 2 = 75 cycles, Index 1 = 8 cycles) MiSeq Reagent Kit v3 (Illumina, Cat No. MS-102-3001). The library was mixed with Chip2 indexed libraries. Total reads per library were measured at >3 x 10^5 which was sufficient to determine that expected barcodes comprise the majority (>70%) of sequencing reads.

It is certified that this product meets the above specifications, as reviewed and approved by the Quality Department.

Notice to Purchaser

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640178

NOTICE TO PURCHASER:

Our products are to be used for **Research Use Only**. They may not be used for any other purpose, including, but not limited to, use in humans, therapeutic or diagnostic use, or commercial use of any kind. Our products may not be transferred to third parties, resold, modified for resale, or used to manufacture commercial products or to provide a service to third parties without our prior written approval.

Your use of this product is also subject to compliance with the licensing requirements, listed below if applicable, and described on the product´s web page at http://www.takarabio.com. It is your responsibility to review, understand and adhere to any restrictions imposed by these statements.

TRADEMARKS:

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