Certificate of Analysis



SMARTer® Mouse BCR IgG H/K/L Profiling Kit Components

Catalog Nos.

634425 (Not sold separately; sold as a part of 634422) 634426 (Not sold separately; sold as a part of 634423) 634427 (Not sold separately; sold as a part of 634424) **Amount** 12 rxns 48 rxns 96 rxns **Lot Number** Specified on product label. Specified on product label. Specified on product label.

Description

The SMARTer Mouse BCR IgG H/K/L Profiling Kit Components are part of the SMARTer Mouse BCR IgG H/K/L Profiling Kit, which enables users to analyze B-cell receptor (BCR) diversity from RNA input samples. The full kit is designed to work with a range of RNA input amounts depending on the sample type and has been shown to generate high-quality libraries from as little as 10 ng–3 µg of total RNA obtained from spleen, lymph node, or PBMCs or from 1,000–10,000 purified B cells. As the name suggests, the kit can be used to generate data for heavy-chain or light-chain (kappa or lambda) diversity.

The full kit leverages SMART® (Switching Mechanism at 5' end of RNA Template) technology and employs a 5' RACE-like approach to capture complete V(D)J variable regions of BCR transcripts. Included in the full kit are primers that incorporate Illumina®-specific adaptor sequences during cDNA amplification. The protocol generates indexed libraries that are ready for sequencing on Illumina platforms.

Package Contents

Package 1:

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	<u>634425</u> (12 rxns)	<u>634426</u> (48 rxns)	<u>634427</u> (96 rxns)				
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	5 µl	5 µl	5 µl	Control RNA (1 µg/µl)			
	12 µl	48 µl	96 µl	BCR Oligonucleotide (24 µM)			
Package 2:							
	<u>634425</u>	<u>634426</u>	<u>634427</u>				
	(12 rxns)	(48 rxns)	(96 rxns)				
	24 µl	96 µl	192 µl	BCR dT Primer (12 µM)			
	48 µl	192 µl	384 µl	5X Ultra® Low First-Strand Buffer			
	24 µl	96 µl	192 µl	SMARTScribe [™] Reverse Transcriptase (100 U/µl)			
	5 ml	20 ml	2 x 20 ml	Nuclease-Free Water			
	30 µl	60 µl	60 µl	RNase Inhibitor (40 U/µl)			
	230 µl	920 µl	1.85 ml	10X Lysis Buffer			
2	2 x 1.7 ml	8 ml	2 x 8 ml	Elution Buffer (10 mM)			
	6 µl	24 µl	48 µl	mBCR Primer 1H (12 µM)			
	6 µl	24 µl	48 µl	mBCR Primer 1K (12 µM)			
	6 µl	24 µl	48 µl	mBCR Primer 1L (12 µM)			
	18 µl	72 µl	144 µl	BCR Primer 1V (12 µM)			
	200 µl	600 µl	2 x 600 µl	PrimeSTAR® GXL SP DNA Polymerase (1.25 U/µl)			
	2 x 1 ml	6 ml	2 x 6 ml	5X PrimeSTAR GXL SP Buffer			
	800 µl	2.4 ml	4.8 ml	dNTP Mixture (2.5 mM each)			

Takara Bio USA, Inc.

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# Certificate of Analysis

SMARTer Mouse BCR IgG H/K/L Profiling Kit Components (Not sold separately)

#### **Storage Conditions**

- Store Control RNA and BCR Oligonucleotide at –70°C.
- Store 10X Lysis Buffer at –20°C. Once thawed, the buffer can be stored at 4°C.
- Store Nuclease-Free Water at -20°C. Once thawed, the water can be stored at 4°C.
- Store Elution Buffer at –20°C. Once thawed, the buffer can be stored at room temperature.
- Store all other reagents at  $-20^{\circ}$ C.

#### **Expiration Date**

• Specified on product label.

#### **Shipping Conditions**

• Dry ice

#### **Product Documents**

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

• SMARTer Mouse BCR IgG H/K/L Profiling Kit User Manual

### **Quality Control Data**

A sample kit from each lot was tested as follows: 10 ng of Control RNA (Mouse spleen total RNA) was subjected to firststrand cDNA synthesis as described in the SMARTer Mouse BCR IgG H/K/L Profiling Kit User Manual. The first-strand cDNA was then used as a template in three different PCRs for heavy chain and kappa and lambda light chains, each for 18 cycles of PCR. 1 µl of the resulting double-stranded cDNA was amplified by nested PCR for 19 cycles for heavy chain, 13 cycles for kappa chain, and 16 cycles for lambda chain. The final sequencing library was purified with an Agencourt AMPure XP Kit (Beckman Coulter, Part No. A63880 or A63881) and resuspended in 17 µl of Elution Buffer. 1 µl of the sequencing library was analyzed with an Agilent 2100 Bioanalyzer and a DNA 1000 Kit (Agilent, Cat. No. 5067-1504). The analysis indicated that the BCR library profile produced a broad peak spanning 650–1,100 bp, with the peak maximum at approximately 700 bp. The area containing the peak was manually integrated from 500 bp– 1,200 bp, and the concentration of this region in nmol/L (nM) was determined by the Bioanalyzer software. Final library outputs were  $\geq$ 4 nmol/L (nM).

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



# SMARTer® Mouse BCR IgG H/K/L Profiling Kit Components

CATALOG NOS.

634425, 634426 & 634427

#### NOTICE TO PURCHASER:

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