

Product Information Reverse Transcription Kit

ExcelRT™ series

RP1300 100 RXN

Reverse Transcriptase (200 U/μl)	100 µl
RNase Inhibitor (20 U/μl)	100 µl
5X RT Buffer (DTT)	500 μl
dNTP Mix (10 mM each)	الم 200
Oligo (dT) ₂₀ (50 μM)	100 µl
Random Hexamers (100 μM)	100 μΙ
DEPC-Treated H ₂ O	1 ml x2

Storage

-20°C for 24 months

Description

ExcelRT™ Reverse Transcription Kit is a complete, efficient and convenient kit to synthesize high quality first strand cDNA. This kit contains ExcelRT™ Reverse Transcriptase, which is able to synthesize the first strand cDNA at 37~50°C. The ExcelRT™ Reverse Transcriptase is a recombinant Moloney Murine Leukemia Virus (M-MLV) reverse transcriptase, which is designed to reduce RNase H activity and create better thermal stability. This kit also contains RNAok™ RNase Inhibitor, which is active against RNase A, RNase B, and RNase C. This product is supplied with oligo (dT)₂₀ and random hexamers, which are used to synthesize cDNA from poly(A) tailed mRNA and total RNA, respectively.

Features

- Contains all components for reverse transcription
- High vield
- Thermostable, up to 50°C, during first strand synthesis
- High processivity, generating cDNA up to 8 kb
- · Reduced RNase H ribonuclease activity





Application

- Generation of first strand cDNA from total RNA or mRNA.
- Suitable for generating cDNA from RNA with strong secondary structure which can be reduced at temperature up to 50°C.

Storage Buffer

Reverse Transcriptase: 20 mM Tris-HCl (pH 7.5), 200 mM NaCl, 0.1 mM EDTA, 1 mM DTT, stabilizer and 50% (v/v) glycerol

RNase Inhibitor: 40 mM HEPES-KOH (pH 7.5), 100 mM KCI, 8 mM DTT, 0.1 mM EDTA, stabilizer and 50% (v/v) glycerol

5X RT Buffer (DTT)

250 mM Tris-HCl (pH 8.3 at 25°C), 375 mM KCl, 15 mM MgCl $_{
m 2}$ and 50 mM DTT

First Strand cDNA Synthesis Condition

1. Denature (Mixture A):

Total DNIA

IULAI KIN	A	Λ μι (IIIg Z μg)
dNTP Mix (10 mM each)		1 μΙ
Primers	50 μM Oligo (dT) ₂₀	
or	100 μM Random Hexamers	
or	10 μM Gene Specific Primers	
DEPC-Tr	eated H₂O	to 10 ul final vol.

Mix well; incubate at 70°C/5 minutes Place on ice for at least 1 minute

2. First strand cDNA buffer (Mixture B) per reaction:

(Master Mix can be prepared before or during the denaturing step)

5X RT Buffer (DTT)	4 μΙ
DEPC-Treated H ₂ O	4 μΙ
RNAok™ RNase Inhibitor	1 μΙ
ExcelRT™ Reverse Transcriptase	1 μΙ
Final volume	10 μΙ



V ul (1ng~2 ug)



First Strand cDNA Synthesis Condition (continued)

3. First strand cDNA synthesis:

Mixture A (RNA + primers + dNTPs) 10 μl Mixture B (First strand cDNA buffer) 10 μl

Final volume	20 μΙ	
Incubate	(25°C/10 minutes)*	
	37~50°C/50 minutes	

4. Termination: 85°C/5 minutes

Keep at 4°C

5. RNA removal*: add 1 μ l RNase H into each reaction 37°C/20 minutes

Store cDNA at -20°C or for immediate PCR reaction

Recommended PCR Condition

(SMOBIO's TP1000 ExcelTaq™ Taq DNA polymerase)

cDNA	2~10 μl
Forward primer	$0.1 - 0.5 \mu M$
Reverse primer	$0.1 - 0.5 \mu M$
10X <i>Taq</i> Buffer	5 μΙ
dNTPs	0.2 mM each
Taq DNA polymerase	0.25 μl (1.25 units)
H_2O	to 50 μl
Total volume	50 μl

Recommended PCR Program

94°C	2 min		
94°C	30 sec	<u> </u>	
50~68°C**	30 sec	}	25 ~ 40 cycles
72°C	30 sec/kb	J	•
72°C	1 min	-	

^{**}Optimal PCR conditions vary according to primers' thermodynamic properties.

^{*}For random hexamers, an additional 10 minutes of incubation at 25°C is suggested.

^{*}Optional step recommended for long range RT-PCR reaction.



Other Information

SMOBIO Technology, Inc. claims all warranties with respect to this document, expressed or implied, including but not limited to those of merchantability or fitness for a particular purpose. In no event shall SMOBIO Technology, Inc. be liable, whether in contract, tort, warranty, or under any statute or any other basis for special, incidental, indirect, punitive, multiple or consequential damages in connection with or arising from this document, including but not limited to the use thereof.

Caution: Not intended for human or animal diagnostic or therapeutic uses.

Champion E coli Transformation Kit

Related Products

Champion E. coil Transformation Kit
RNAok RNase Inhibitor, 2000 U
ExcelRT Reverse Transcriptase, 20,000 U
ExcelRT One-step RT-PCR Kit, 50 RXN
ExcelRT Reverse Transcription Kit II,
100 RXN
SMO-HiFi DNA Polymerase, 100 U
G-HiFi DNA Polymerase, 100 U
ExcelTaq Taq DNA Polymerase, 500 U × 1
ExcelTaq 5X PCR Master Dye Mix, 200 RXN
ExcelTaq Hot Start II DNA Polymerase,
500 U
ExcelTaq 2X Q-PCR Master Mix (SYBR, no
ROX), 200 RXN
ExcelTaq 2X Q-PCR Master Mix (SYBR,
ROX), 200 RXN
ExcelTaq 2X Q-PCR Master Mix (TaqMan,
ROX), 200 RXN
ExcelBand 100 bp DNA Ladder, 500 μl
ExcelBand 1 KB (0.25-10 kb) DNA Ladder,
500 μl
FluoroVue Nucleic Acid Gel Stain

For research use only

2019 ver. 2.1.1

P06-F11-A1



(10,000X), 500 ul