YeaCell™ Reverse Transcriptase for Single Cell Full Length cDNA

Product description

YeaCell™ Reverse Transcriptase for Single Cell Full Length cDNA can use Oligo (dT)18 or TSO Primer to sythesize a complementary DNA strand from single cell or low input cells. The cDNA of the product can be enriched by PCR reaction with universal primers, and then constructed a DNA library using Yeasen's enzymatic library construction kit. This product uses a new reverse transcriptase based on M-MLV(RNase H-)Reverse Transcriptase through multi-point mutation, which has the advantages of high reverse transcriptase efficiency, low mismatch rate and high fidelity.

Specifications

Cat.No.	13589ES08 / 13589ES95 / 13589ES96
Size	10000 U / 96×10000 U / 100×10000 U

Components

Components No.	Name	13589ES08	13589ES95	13589ES96
13589-A	5×RT Buffer	600 μL	47 mL	60 mL
13589-B	Reverse Transcriptase for Single Cell Full Length cDNA	50 μL	4.8 mL	5 mL

Storage

This product should be stored at -25~-15°C for 1 years.

Application

1. High-throughput single-cell full-length cDNA synthesis for mammalian or eukaryotic cells without cell walls.
2. 10 pg~1 μg total RNA with poly (A).
3. This product is not suitable for prokaryotic total RNA and degraded RNA, such as FFPE RNA.

Instructions

Step 1 RNA denaturation

1. Prepare the reaction liquid according to Table 1:

Table 1 RNA denaturation reaction

Name	Value（μL）
Oligo (dT)18（20~50 μM）(customer)	1
Lysis cell or RNA	12
Total	13

2. Mix thoroughly by gently pipetting up and down at least 10 times. Briefly spin down the tube in a microcentrifuge to collect the liquid from the side of the tube.

3. Place tube in a thermocycler and run the following program: (heated lid 80°C on) 70°C, 5 min,then place it on ice immediatly.

Step 2 1st Strand Synthesis

1. Remove the first chain of synthetic reagents from -20°C, thaw at room temperature, mix thoroughly and spin down. Prepared the 1st strand synthesis reagents according to the Table 2.

Table 2 1st Strand Synthesis reaction

Name	Value (μL)
Above step	13
5×RT Buffer	4
TSO Primer (20~50 μM) (customer)	1
RNase Inhibitor(40 U/μL) (customer)	1
Reverse Transcriptase for Single Cell Full Length cDNA	1
Total	20

2. Mix thoroughly by gently pipetting up and down at least 10 times. Briefly spin down the tube in a microcentrifuge to collect the liquid from the side of the tube.

3. Place tube in a thermocycler and run the following program: (heated lid 105°C on) 42°C, 90 min; 70°C, 15min; 4°C, hold。

4. The product can be directly used for two-strand cDNA synthesis or temporary storage at -80°C.

Notes

1. This product is for research use only.

2. Please operate with lab coats and disposable gloves, for your safety.

3.Supplies free of RNase contamination and cleaning the experimental area regularly are necessary. ThermoFisher's RNAZap™ high-efficiency nucleic acid removal spray was recommended to remove RNase contamination.

4. Other materials should be asked if we want to construct a DNA library.