HB220414

2 × Frag/Prime Buffer

Product Information

Product name	Cat#	Specification
2 × Frag/Prime Buffer	11377ES24	24 T
	11377ES96	96 T

Product Description

This product is connected to Hieff NGS™ Ultima Dual-mode RNA Library Prep Kit for Illumina® (Cat#12252) or Hieff NGS™ Ultima Dual-mode RNA Library Prep Kit for MGI® (Cat#13333), replaces Frag/ Prime buffer which is used for RNA fragmentation and one-strand cDNA synthesis steps. Input RNA can be a total RNA or a product of mRNA purification or rRNA removal, etc. Since the RNA from the mRNA purification or rRNA removal is directly eluted with 1× Frag/Prime Buffer in Cat#12252 or Cat#13333, there is no additional volume for adding RNA-containing solutions. If the input RNA has been dissolved in Nuclease free ddH_2O, this product can be used for RNA fragmentation or one-strand cDNA synthesis.

Product Component

Component	24 T	96 T
2 × Frag/Prime Buffer	204 μL	816 μL

Shipping and Storage

All the components are shipped with dry ice and can be stored at -20°C for one year.

Cautions

1. For your safety and health, please wear lab coats and disposable gloves for operation.
2. For research use only!

Instructions

Select Scheme A when input RNA does not require fragmentation, or Scheme B when input RNA requires fragmentation.

A1. If the input RNA does not need to be fragmented, the first-strand cDNA synthesis reaction system can be directly prepared according to Table 1 in combination with Cat#12252 or Cat#13333 reagent.

Table 1 Direct first-strand cDNA synthesis reaction

Components	Volume (μL)
Input RNA	8.5
2×Frag/Prime Buffer	8.5
Strand Specificity Reagent	6
1st Strand Enzyme Mix	2
Total	25

A2. Gently mix with a pipette, centrifuge the reaction solution to the bottom of the tube, and perform one-strand synthesis reaction in a PCR machine according to the reaction program in Table 2. One-strand synthesis products can be ligated to Cat#12252 or Cat#13333 for subsequent reactions.

Table 2 First-strand cDNA synthesis reaction

Temperature	Duration
Hot cover 105 °C	On
25 °C	10 min
42 °C	15 min
70 °C	15 min
4 °C	Hold

B1. If the input RNA needs to be fragmented, the fragmentation system can be prepared according to 3 for RNA fragmentation.

Table 3 Fragmentation reaction

Components	Volume(μL)
Input RNA	8.5
2 ×Frag/Prime Buffer	8.5
Total	17

B2. Use a pipette to gently mix, centrifuge the reaction solution to the bottom of the tube, and set appropriate fragmentation conditions according to the completeness of the input RNA and the purpose of the experiment, referring to the instructions of Cat#12252 or Cat#13333.

B3. One-strand cDNA synthesis was performed on the fragmented products, and the one-strand cDNA synthesis reaction system was prepared according to Table 4.

Table 4 First-strand cDNA synthesis reaction

Components	Volume(μL)
Fragmented RNA	17
Strand Specificity Reagent	6
1 st Strand Enzyme Mix	2
Total	25

B4. Gently mix with a pipette, centrifuge the reaction solution to the bottom of the tube, and perform one-strand synthesis reaction in a PCR machine according to the reaction program in Table 2. One-strand synthesis products can be ligated to Cat#12252 or Cat#13333 for subsequent reactions.