

Tinzyme Co., Limited

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NGS DNA Library Prep End Repair Kit for llumina

Product Number: EK0513

Shipping and Storage

Store at -20°C and transport on dry ice.

Components

Component	96 rxns
ERAT Mix	192μL
10×ERAT Buffer	$480 \mu L$

Description

The second-generation sequencing library end repair kit (Illumina) provides the enzyme end repair enzymes and reaction buffer required for constructing DNA libraries. It can repair the ends of mechanically processed fragmented DNA or small fragments, forming flat ends of DNA double strands. The resulting product does not require purification and can be directly used for Adaptor ligation of DNA fragments using the second-generation sequencing library connection kit. Easy to operate, higher efficiency in library conversion.

Prepare instruments, reagents, and consumables

- 1. Magnetic frame: It is recommended to use DynaMag^{TM-2}.
- 2. Adaptor Connection: It is recommended to use the second-generation sequencing library connection kit.
- 3. DNA purification and recovery: It is recommended to use the magnetic bead method DNA purification and recovery kit.
- 4. PCR enrichment: HiFi PCR Mix for NGS.
- 5. Sample adapter primer kit: It is recommended to use the second-generation sequencing multi sample adapter primer kit I/II.
- 6. Reaction tube: It is recommended to use low adsorption PCR tubes and 1.5mL centrifuge tubes; Gun head: It is recommended to use high-quality filtering gun heads to prevent contamination of reagent kits and library samples.

Preparation and important precautions before the experiment

To avoid repeated freezing and thawing of reagents, it is recommended that you pack and store the remaining reagents after the first use of the reagent kit.

DNA library construction process

- 1. DNA Terminal Repair
- 2. Adaptor Connection
- 3. Connect product purification
- 4. per amplification
- PCR Product Purification

Starting material: 5ng -1µg of broken double stranded DNA, dissolved in EB (10mM Tris-HCl pH 8.0) or deionized water, DNA purity requirement: OD260/OD 280=1.8-2.0.

DNA Terminal Repair Reaction

1. According to the sample concentration, take an appropriate amount of sample (recommended 100 ng) into a new PCR tube and add deionized water to a total volume of 43μL. DNA Control can be added as a quality control product for each batch of library construction. The sample size of DNA Control is 2μL, and 41μL of deionized water is added to make the total system 43μL. The following end repair reaction mixture is prepared in the PCR tube:



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Reagents	Volume
10×ERAT Buffer	5μL
ERAT Mix	$2\mu L$
fragmented DNA	$X(5ng-1\mu g)$
RNase Free Water	Up to50μL

Table 1 Configuration of End Repair Reaction Solution

- 2. Gently blow and mix the above solution with a gun tip, centrifuge briefly, and collect all components to the bottom of the tube to ensure that there are no bubbles in the reaction solution.
- 3. Place the PCR tube containing the mixture from the previous reaction on the PCR instrument and react according to the following conditions:

Temperature	Time
Hot lid (85°C)	
37°C	15min
65°C	15min
4°C	Hold