Product Information	
5X ER/A-Tailing Enzyme Mix	
Part Number	Y9420L
Unit Size	24 reactions
Volume	0.24mL
Storage Temperature	-25°C to -15°C
Lot Number	
Reference Number	

Product Description:

5X ER/A Tailing is a NGS library preparation module that uses a one-step reaction to combine end-repair and dA-tailing to convert fragmented DNA into 5´-phosphorylated and 3´-dA-tailed DNA fragments enabling direct ligation of Illumina sequencing adapters. When used in combination with the 5X WGS ligation module (L6030-W-L), the optimized chemistry ensures high sensitivity for low input DNA, high ligation efficiency for maximum library yield and a workflow that is under 3 hours with less than 45 minutes hands on time.

Product Specifications	
Assay	**ER/A Tailing Enzyme Mix Functional Assay**
Specification	Functional

Quality Control Analysis:

ER/A Tailing Enzyme Mix Functional Assay: QC Library length must be within 15% of the reference library length. Concentration of the QC library generated from 100 ng input DNA (average ~300 bp fragments) is >60 nm with mapped reads > 90%. For QC library, normalized coverage should be within 0.7 to 1.3 for most of the genome (10% - 80% GC content).

Notes:

Enzyme components were tested prior to assembly and free of contaminating endonucleases and exonucleases. Enzyme purity was >95% as determined by SDS-PAGE and negligible *E. coli* genomic DNA contamination was confirmed by qPCR.

Usage instructions:

1. Enter the following program into a thermal cycler (see table below). Be certain to use the instrument's heated lid, and if possible, set the temperature of the heated lid to ~70°C.

   When the thermal cycler block reaches 4°C, pause the program.

Compatible with DNA inputs as low as 250pg and up to 1µg in water, EB or 1X TE

Step	Incubation Temperature	Incubation Time
1	4°C	1 min
2	20°C	30 min
3	65°C	30 min
4	4°C	Hold

2. It is important to follow the procedure described below, in order to achieve optimal results. Prepare a reaction mix in a new thin-walled PCR tube on ice by combining ERA Buffer, DNA sample, and nuclease-free water as indicated in the table (per DNA sample). Mix well by gently pipetting (do not vortex to mix).
   The final reaction volume is 50 µl.

	1 reaction (µl)
10X ERA Buffer	5
DNA sample	X
Nuclease-free H_2O	(35 - X)
Total	40

3. Add 10 µl of 5X ER/A-Tailing Enzyme Mix to each reaction and gently mix well by pipetting up and down 6-8 times. It is recommended to keep the PCR tube on ice for the whole time during reaction setup.

4. Pulse-spin the sample tube and immediately transfer to the pre-chilled thermal cycler (4°C). Resume the cycling program.

5. When thermal cycler program is complete and sample block has returned to 4°C, remove samples from block and place on ice.

6. Proceed directly into Adapter Ligation. We recommend using Enzymatics' WGS Ligase (L6030-W-F or L6030-W-L)