Varizymes®
OPTIONS AMPLIFIED

SwifTag™ Tn5 2.0 Transposase

Next Generation Sequencing

Contents

SwifTag™ Tn5 2.0 enzyme is supplied at a concentration of 1 U/µL with 10X Tn5 reaction buffer (100 mM Tris-HCl, 100 mM MgCl2, pH 7.5) and 40X Stop solution (2% SDS).

Background

SwifTag Tn5 2.0 Transposase is a hyperactive form of the enzyme engineered for improved activity, speed, and robustness. Tn5 can be used to randomly insert unique oligonucleotide sequences into any target DNA *in vitro*. Efficient transposition requires the assembly of a specific 19-bp transposase recognition sequence (Mosaic End or ME sequence) with the Tn5 Transposase. The assembled transposon catalyzes a random “cut and paste” reaction that adds the ME sequence to the target DNA, creating a 9-bp sequence duplication immediately flanking the transposon insertion site.

Application Notes

Construction of random libraries for second-generation sequencing and *in vitro* transgenic experiments. SwifTag Tn5 2.0 is utilized in genome sequencing to covalently attach oligonucleotide adaptors and fragment the DNA in a single enzymatic reaction, reducing the time and input requirements over traditional NGS sequencing library preparation.

**These products are intended for research use only, not for diagnostic use. The safety and efficacy of these products in diagnostic or other clinical uses has not been established.*

Shipping & Storage

SwifTag Tn5 2.0 is shipped on dry or blue ice. On arrival store at -20 °C for optimum stability.

Quality Control

- SwifTag Tn5 2.0 activity: The activity of Tn5 2.0 is determined by fragmentation of 200 ng of λ phage DNA by 2 Units of Tn5 in 10 minutes at 55 °C to fragments of 200 - 500 bp in length as assessed by agarose gel electrophoresis
- Purity: >95% as determined by SDS-PAGE analysis
- SwifTag Tn5 2.0 is free of detectable RNase and DNase (exo- and endonuclease)
- <0.25 ng contaminating host DNA per Unit

Generating a Transposon for Illumina NGS

Preparation of Adapter Mix

The name and sequence of reference primers for Illumina platform:
Primer ME A: 5'-TCGTCGGCAGCGTCAGATGTGTATAAGAGACAG-3'
Primer ME B: 5'-GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAG-3'
Primer ME rev: 5'-pCTGTCTCTTATACACATCT-3'
Dissolve Primer ME A, Primer ME B, Primer ME rev with Annealing Buffer (10 mM Tris-HCl, 100 mM NaCl, 1 mM EDTA, pH 7.5) to 1 mM.

Example for mixing and annealing adapters **ME A/rev** **ME B/rev**

Component	Volume	Volume
Tn5 ME rev [1 mM]	5 µL	5 µL
Tn5 ME A [1 mM]	5 µL	
Tn5 ME B [1 mM]		5 µL
1X annealing buffer	40 µL	40 µL
Total volume	50 µL	50 µL

Use thermocycler for annealing adapters with the following program:

95 °C, 5 minutes
65 °C, 5 minutes
25 °C, 30 minutes

Mix A/rev and B/rev together in equal amounts after annealing.

Final concentration is 50 µM each.

Transposon Assembly

Component	Volume
50 µM each ME A/rev + ME B/rev	2 µL
Tn5 transposase	10 µL

Incubate at 25 °C with lid set to 55 °C for 30 min.

Transposon assembly may be stored at -20 °C until ready to use.

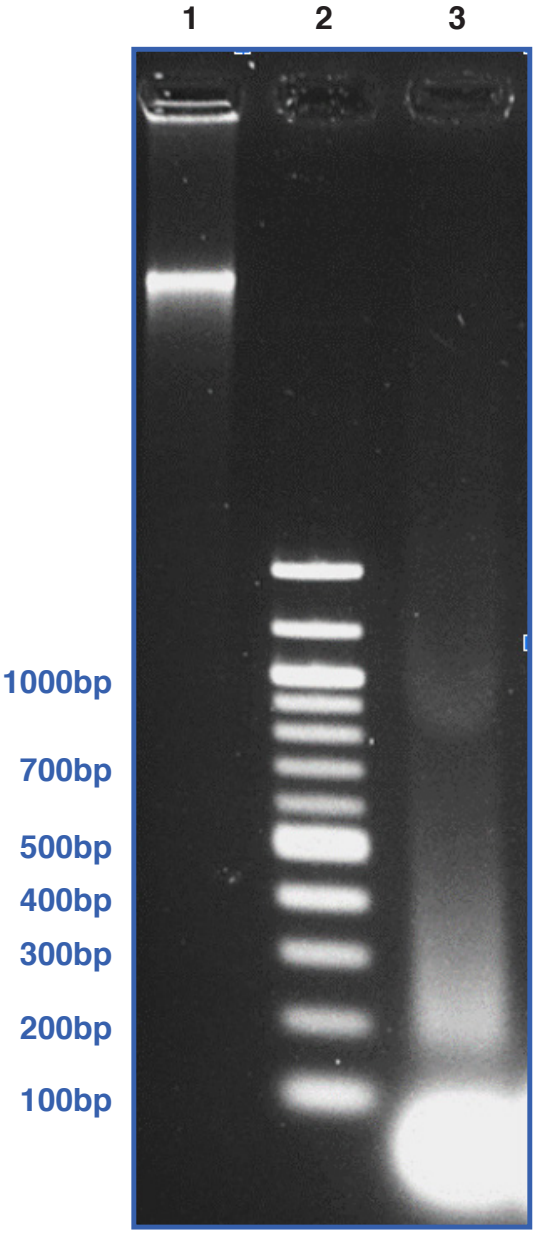
Component	Volume
10X Tn5 reaction buffer	2 µL
Lambda DNA [200 ng/µL]	1 µL
water	15 µL
Transposon assembly mix	2 µL
Total volume	20 µL

Incubate samples at 55 °C in thermocycler with lid set to 65 °C for 10 min.

For evaluation of fragment size distribution add 0.5 µL of 40X Stop Solution to samples, heat at 65 °C for 15 minutes, add loading dye and run on 1.2 % agarose gel for 1.5 hrs at 80V.

For downstream sequencing applications, samples can be used for PCR amplification immediately following tagmentation reaction. For tagmentation reactions deactivated by heat treatment with SDS it is necessary to remove the SDS prior to PCR using a suitable clean up kit (Zymo PCR Inhibitor Removal Kit or equivalent).

Performance of SwifTag Tn5 2.0 transposase on Lambda DNA



Lane:

1. Uncut λ DNA
2. GeneRuler 1 kb plus DNA Ladder
3. SwifTag Tn5 2.0-treated λ DNA