pA-TN5 transposase (loaded)

Cat. No. C01070001
Format: 15 µl (32 rxns) / 30 µl (64 rxns)
Concentration: 1.05 mg/ml
Molecular weight: 67 kD

Product description
pA-Tn5 Transposase is a fusion protein of hyperactive Tn5 transposase and protein A developed for the CUT&Tag assay. For ease of use, the fusion protein is pre-loaded with sequencing adapters suitable for single or dual indexing in single or paired-end Illumina platforms. The adapters contain 19-mer Tn5 mosaic ends and the sequences for PCR amplification with barcoded i7/i5.

Mosaic end_reverse: [PHO]CTGTCTCTTATACACATCT

Mosaic end_Adapter A: TCGTCGCAAGC6TCAGATGTGTATAAGAGACAG

Mosaic end_Adapter B: GTCTCGTGGGCTCGGAGATGTGTATAAGAGACAG

Underlined regions correspond to the double-stranded part of the adapter, recognized by the transposase. Bold regions represent sequences required per PCR amplification with barcoded i7/i5 primers as described by Buenrostro et al (2015).

The protein A has a high affinity to rabbit polyclonal antibodies, mouse IgG2a, IgG2b and IgA, guinea pig IgG, dog IgG, pig IgG. However, the use of secondary antibody (e.g. guinea pig anti-rabbit) is recommended for a higher sensitivity of CUT&Tag assay.

Suggested dilution:
For the standard CUT&Tag assay, the recommended dilution of pA-Tn5 Transposase (loaded) is 1:250 (0.4 µl of pA-Tn5 for 100 µl of buffer). Please note that depending on the starting amount of cells and/or primary antibody, different dilution in a range 1:50-1:500 might be tested.

Storage conditions
Store at -20°C. Guaranteed stable for 6 months from date of receipt when stored properly.

Storage buffer
Supplied in solution containing 50% v/v glycerol.

Precautions
This product is for research use only. Not for use in diagnostic or therapeutic procedures.
Quality control

Each lot of pA-Tn5 transposase is quality checked by an in vitro activity test (cleavage of human genomic DNA) [Figure 1, A] and by a CUT&Tag assay using H3K27me3 polyclonal ChIP-seq grade antibody [Cat. No. C15410195] [Figure 1, B].

Figure 1: Quality control of pA-Tn5 transposase loaded with sequencing adapters

A: The Fragment Analyzer trace showing the representative cleavage pattern of gDNA. The pA-Tn5 fusion protein [Cat. No. C01070001] efficiently digests gDNA to a smear. 500 ng of human genomic DNA were incubated for 7 min at 55°C with 1 µl of pA-Tn5 fusion protein loaded with appropriated adaptors in a tagmentation buffer (40mM Tris-HCl pH7.5, 40mM MgCl2 and 12.5% DMF). The reaction was stopped by adding SDS, cleaned-up and resolved on the Fragment Analyzer to assess the cleavage.

B: Representative screenshot at selected locus obtained using Diagenode pA-Tn5 fusion protein [Cat. No. C01070001] and H3K27me3 polyclonal ChIP-seq grade antibody [Cat. No. C15410195] following CUT&Tag protocol [Kaya-Okur, H.S., Nat Commun 10, 1930 (2019)].