

# **TIANSeq Single- Index Adapter (Illumina)**

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# TIANSeq Single-Index Adapter (Illumina)

Cat.no. 4992641/4992642/4992378

## Kit Contents

Contents	4992641 (Set A)	4992642 (Set B)	4992378 (Set A+B)
TIANSeq Single-Indexed Adapter (30 $\mu$ M)	12 $\times$ 40 $\mu$ l	12 $\times$ 40 $\mu$ l	24 $\times$ 40 $\mu$ l
Adapter Dilution Buffer	5 $\times$ 1.8 ml	5 $\times$ 1.8 ml	10 $\times$ 1.8 ml

## Storage and Working Conditions

Upon receiving the kit, please store at -30~-15°C. The shelf life of the kit is one year.

TIANSeq Single - Indexed Adapter should not be kept at more than 25°C. Avoid repeated freezing and thawing. Please store at -30~-15°C after use.

Adapter Dilution Buffer can be stored at 2-8°C for a month. For long-term storage, please keep at -30~-15°C.

## Product Description

TIANSeq Single-Indexed Adapter is specifically developed for illumina High-throughput sequencing platform. It can be used to construct DNA and RNA libraries for illumina High-throughput sequencing platform. The kit is provided in two forms: 12 kinds of adapters mix and 24 kinds of adapters mix, and each adapter in the mix contains a unique 6 base index sequence (barcode) to identify different samples in multi-sample sequencing.

The concentration of the adapter mastermix of this kit is 30  $\mu$ M. The working concentration varies with the kit used for the library construction, the initial DNA input and the size of DNA fragments. Please refer to the protocol for specific information. In addition, the Index sequences of the 24 adapters mix are shown in the Adapter Sequence Information section.

## Applications:

1. Generally, this product is used for the construction of DNA and RNA libraries for NGS of illumina High-throughput sequencing platform.
2. Specific applications of this product include exon sequencing, target region sequencing, RNA-Seq, CHIP-Seq, directed sequencing and whole genome sequencing.
3. It should be noted that this product is not suitable for Illumina HiSeq X<sup>TM</sup> instruments and methylation-related sequencing.

## Precautions Please carefully read these precautions before using this kit

1. It is recommended to keep the TIANSeq Single - Indexed Adapter on ice or in an ice box during the experiment. Please do not place the product at more than 25°C, otherwise the advanced structure of adapter might be damaged.
2. If adapter needs to be diluted, please use the Adapter Dilution Buffer supplied in the kit. Don't use ultrapure water or other buffer.
3. The diluted adapter should be prepared and used within one day. It is not recommended to store the diluted adapter for a long time, or repeatedly freeze and thaw the diluted adapter.
4. Please use nuclease- and nucleic acid-free consumables. Make sure to use the consumables made by low nucleic acids adsorption materials. In addition, cross-contamination between adapters should be avoided during the experiment.

### Recommended Alternative Reagents:

1. TIANSeq DirectFast DNA Library Prep Kit (illumina)
2. TIANSeq Fast DNA Library Prep Kit (illumina)

### Protocol

1. If the volume of TIANSeq single-indexed Adapter added in the ligation system is fixed at 5  $\mu$ l, then the working concentrations of the adapter for different DNA input and DNA fragment sizes are shown in the following table:

DNA Input	Adaptor working concentration for different sizes of DNA fragments			Adapter: Insert (molar ratio)
	200 bp	300 bp	400 bp	
1 $\mu$ g	15 $\mu$ M	10 $\mu$ M	7.5 $\mu$ M	10 : 1
500 ng	19 $\mu$ M	12.5 $\mu$ M	9.5 $\mu$ M	25 : 1
250 ng	15 $\mu$ M	10 $\mu$ M	7.5 $\mu$ M	40 : 1
100 ng	15 $\mu$ M	10 $\mu$ M	7.5 $\mu$ M	100 : 1
50 ng	15 $\mu$ M	10 $\mu$ M	7.5 $\mu$ M	200 : 1
25 ng	7.5 $\mu$ M	5 $\mu$ M	3.75 $\mu$ M	200 : 1
10 ng	3 $\mu$ M	2 $\mu$ M	1.5 $\mu$ M	200 : 1
5 ng	1.5 $\mu$ M	1 $\mu$ M	750 nM	200 : 1
2.5 ng	750 nM	500 nM	375 nM	200 : 1
1 ng	300 nM	200 nM	150 nM	200 : 1
0.25 ng	75 nM	50 nM	37.5 nM	200 : 1

2. When the TIANGEN library construction products are applied TIANSeq DirectFast DNA Library Prep Kit (illumina) and TIANSeq Fast DNA Library Prep Kit (illumina) , please dilute the adapter master mix according to the DNA input range of the relevant products using the above table as reference.
3. When applying the library construction products from other suppliers, if the DNA input is greater than 1  $\mu$ g, dilute the adapter based on the standard of 10:1 mole ratio between the adapter and the inserted fragment; if the DNA input is less than 0.25 ng, dilute adapter based on the standard of 200:1 molar ratio between the adapter and the inserted fragment. The mole number of DNA input is calculated according to the following formula:

$$\text{Mole number of DNA input (pmol)} = \frac{\text{Mass of DNA (ng)}}{660} \times \frac{1000}{\text{DNA fragment size (bp)}}$$

4. For the DNA fragments with sizes not shown in the table, the mole number of DNA input can also be calculated according to the above formula.

### Adapter Sequence Information

The sequence information of the adapters in this kit is listed below:

#### 1. Universal Sequence

5'-AATGATACGGCGACCACCGAGATCTACACTCTTTCCCTACACGACGCTCTTCCGA  
TCT-3'

#### 2. Index Including Sequence

5'-GATCGGAAGAGCACACGTCTGAACTCCAGTCAC [index1-27] ATCTCGTATGCCGTCTTCTGCTTG-3'

#### 3. Index Number and Sequences

Set A		Set B	
Index	Sequence	Index	Sequence
1	ATCACG	13	AGTCAA
2	CGATGT	14	AGTTCC
3	TTAGGC	15	ATGTCA
4	TGACCA	16	CCGTCC
5	ACAGTG	18	GTCCGC
6	GCCAAT	19	GTGAAA
7	CAGATC	20	GTGGCC
8	ACTTGA	21	GTTTCG
9	GATCAG	22	CGTACG
10	TAGCTT	23	GAGTGG
11	GGCTAC	25	ACTGAT
12	CTTGTA	27	ATTCTT

### Ingredients of the Adapter Dilution Buffer

10 mM Tris-HCl, 10 mM NaCl, 1 mM EDTA, pH 8.0~8.5 @ 25°C.