

# Exonuclease I

## 1 Packing list

Components	HY-KE8010-750 U
Exonuclease I (5 U/ $\mu$ L)	150 $\mu$ L
10 $\times$ Exo I Reaction Buffer	1 mL

## 2 Introduction

This product is obtained by expressing the Exo I gene (*E. coli*) in *Escherichia coli* and then purifying and isolating it multiple times. Exonuclease I (ExoI) is a single-stranded specific 3'  $\rightarrow$  5' exonuclease that cleaves the 3'-OH end of ssDNA to generate 5'-mononucleotides. Very high specificity for single-stranded DNA and does not break down double-stranded DNA and RNA. It can be used to clean PCR reaction products by enzymatic hydrolysis and remove ssDNA fragments from the reaction solution.

## 3 Unit definition

Using heat-denatured calf thymus DNA as the substrate, the amount of enzyme required to produce 10 nmol of acid-insoluble precipitate within 30min at 37°C and pH 9.5 is defined as one unit of activity.

## 4 General Protocol

Protocol for PCR product clean-up prior to sequencing

1) Prepare the following reaction mixture:

Components	Volume
PCR mixture	5 $\mu$ L
Exonuclease I	2 $\mu$ L
Shrimp Alkaline Phosphatase (SAP)	1 $\mu$ L

2) Mix well and incubate at 37 °C for 15min.

3) Stop the reaction by heating the mixture at 85 °C for 15min.

## 5 Storage

-20°C, 1 year

## 6 Precautions

1. These instructions are applicable to PCR products produced by any DNA polymerase. The purified PCR products can be directly used for DNA sequencing, but it is not recommended to be used directly for subsequent cloning.
2. If there are other specific experiments to remove ssDNA alone, it is recommended to use it with 10× Exonuclease I Reaction Buffer.
3. This product is for R&D use only, not for drug, household, or other uses.
4. For your safety and health, please wear a lab coat and disposable gloves to operate.