



Exonuclease I (Exo I)

Manual

Catalog #	3405	3407
Package Size	5,000 units	20,000 units
Concentration	20 U/μL	20 U/μL
Volume	250 μL	1,000 μL

Custom formats and package sizes are available upon request



Important!

-20°C Storage Required

- * Immediately inspect packages
- * Freeze upon receipt



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Intact Genomics, Inc.

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Description:

Intact Genomics' Exonuclease I (Exo I) progressively degrades single-stranded DNA (ssDNA) in the 3'→5' direction, releasing deoxyribonucleoside 5'-monophosphates in a stepwise manner and leaving double-stranded DNA and the 5'-terminus intact. It does not cleave double-stranded DNA.

The cleavage of single-stranded DNA is blocked when the terminal 3'-OH group is phosphorylated or acetylated. Intact Genomics' Exonuclease I can work in a wide range of buffers from different manufacturers, as long as there is sufficient magnesium (1 mM—10 mM). Intact Genomics' Exonuclease I can be heat-inactivated by incubation at 80 °C for 15 minutes.

Applications and Benefits:

- PCR Cleanup – Removes residual primers while preserving dsDNA.
- Sequencing & Genotyping – Ideal for Sanger sequencing, SNP analysis, and cloning workflows.
- Selective ssDNA Removal – Specifically degrades ssDNA with a free 3'-OH end.
- Broad Buffer Compatibility – Compatible with a wide range of buffer systems from different brands.
- Heat Inactivation – Inactivated at 80 °C for 15 minutes.
- High-Throughput & OEM Ready – Suitable for automated workflows, bulk supply, and OEM applications.

Product Source:

Recombinant Exo I expressed from a plasmid carrying *E.coli* sbcB gene

Specifications:

- Purified recombinant Exo I with a molecular weight of 55 kDa
- Free of detectable endonuclease activity
- ≥95% pure from host proteins

Product Components and Storage Temperature:

The following reagents are supplied with this product:

IG Cat #	Package Size	Component Name	IG Part #	Amount	Concentration	Storage
3405	5000 units	Exonuclease I (Exo I)	P241210	1 x 250 µL	20 units/µL	-20 °C
		10X Reaction Buffer	P241216	1 x 300 µL	10X	-20 °C
3407	20000 units	Exonuclease I (Exo 1)	P241214	1 x 1 mL	20 units/µL	-20 °C
		10X Reaction Buffer	P241218	1 x 1.25 mL	10X	-20 °C

Custom formats and package sizes are available upon request.

Storage Buffer:

10 mM Tris-HCl, 100 mM NaCl, 5 mM 2--Mercaptoethanol, 0.5 mM EDTA, 50% glycerol (pH 7.5 at 25°C)

Quality Control:

Protein Purity is evaluated by SDS-PAGE.

Double-stranded DNA endonuclease activity is measured in a 10 μ L reaction containing 0.5 μ g of plasmid DNA and 0.5 μ L of enzyme solution incubated for 14 hours at 37°C.

ssDNase Activity: 20 units of Exonuclease I enzyme in a 10 μ L reaction digests 2 μ g of ssDNA in a 30 minute reaction at 37°C.

Definition of Activity Unit:

One unit is defined as amount of Exonuclease I that catalyzes the release of 10 nmol of acid soluble nucleotides in 30 min at 37°C from 3 H-labeled single-stranded DNA substrate.

The activity is assayed in a mixture containing 67 mM glycine-KOH (pH 9.5), 6.7 mM $MgCl_2$, 10 mM 2-Mercaptoethanol, and 10 nmol 3 H-labeled single-stranded DNA.

Protocol for the Digestion of Single-Stranded DNA:

The reaction will digest ssDNA including primers.

1. Use the table below to set up the reaction:
2. Incubate the mixture at for 10 to 20 minutes 37°C. Time may be extended if desired.
3. Incubate for 15 minutes at 80°C to inactivate enzyme/s.
4. PCR products can be used for direct sequencing or for other downstream applications.

Reagent	μ L of sample
ssDNA	X (up to 2 μ g)
H ₂ O-Nuclease-Free	8-X
10x Exonuclease I Buffer	1
Exonuclease I Enzyme	1

Total Volume: 10 μ L

References:

1. Lehman IR, Nussbaum AL. The Deoxyribonucleases of Escherichia Coli. V. On The Specificity of Exonuclease I (Phosphodiesterase). J Biol Chem. 1964 AUG;239:2628-36. PMID: 14235546.
2. Werle E, Schneider C, Renner M, Völker M, Fiehn W. Convenient single-step, one tube purification of PCR products for direct sequencing. Nucleic Acids Res. 1994 Oct 11;22(20):4354-5. doi: 10.1093/nar/22.20.4354. PMID: 7937169; PMCID: PMC331970.
3. Nabavi S, Nazar RN. Simplified one-tube "megaprimer" polymerase chain reaction mutagenesis. Anal Biochem. 2005 Oct 15;345(2):346-8. doi: 10.1016/j.ab.2005.06.033. PMID: 16061194.

Related Products

- T4 Polynucleotide Kinase (PNK) (Cat.# 3219)
- Taq DNA Ligase (Cat.# 3219)
- T4 DNA Ligase (Cat.# 3212)
- IG T5 Exonuclease (Cat.# 3236,3238)
- Exonuclease III (Cat.# 3412, 3415)
- ig-Fusion™ Cloning Enzyme Premix (Cat.# 4111-1,4115-1,4117-1)
- Hi-Fi *Tth* DNA Ligase (Cat.# 3223, 3225)

Ordering Information

- Order online within the USA. Place orders on **www.intactgenomics.com** using our secure Shopping Cart.
- Order by email, phone, or fax.
Email: **sales@intactgenomics.com**
Phone: (314) 942-3655 | Toll-free : 855-835-7172 | Fax: (314) 942-3656
- Order via our distributors.

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Our hours are Monday - Friday, 8 AM to 5 PM, U.S. central standard time (CST).

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