

Heat-Labile Double-Strand Specific DNase (HL-dsDNase)

Product Data

Source

Produced in a *Pichia pastoris* strain expressing a gene for a recombinant DNase.

Concentration

1-50 U/μl

Specific activity

Ca. 220 000 Unit/mg

Unit definition

One unit increases the absorbance at 260 nm by 0.001 OD per min at 25°C and pH 5.0 with large molecular weight DNA as the substrate according to the assay method of Kunitz.

Storage

Store at -20°C

Storage buffer

25 mM Tris-HCl (pH 7.5 at 25°C)

2.0 mM MgCl₂

10 mM NaCl

0.01% (v/v) Triton X-100

50% (v/v) glycerol

Protocol recommendations

For preferential degradation of dsDNA, and no RNase activity, use a reaction buffer containing 20 mM Tris-HCl (pH 8.0) and 2.5 mM MgCl₂.

Activators

Mg²⁺ ions (up to 10 mM) are required for maximum activity.

Optimum pH: 7.5

Optimum reaction temperature: 40°C

Properties

Heat-Labile dsDNase is an endonuclease that cleaves phosphodiester bonds in DNA to yield oligonucleotides with 5'-phosphate and 3'-hydroxyl termini. Heat-Labile dsDNase has a high specific activity, and it is easily inactivated by heat. It has a particularly strong preference for double-stranded DNA (dsDNA). In the presence of magnesium as only divalent cation and using oligos as a substrate; the activity towards dsDNA is

minimum 5000-fold higher than towards ssDNA. The enzyme can therefore be used to specifically degrade dsDNA, leaving ssDNA essentially intact.

Application

Removal of contaminating DNA from master mixes or enzymes before PCR or RT-PCR leaving single-stranded primers and RNA intact. Removal of DNA from enzymes and mastermixes before PCR.

Heat inactivation

The Heat-Labile dsDNase can be heat inactivated by heat treatment at 55°C for 15 min. If proceeding directly to PCR, 55°C for 5 min is sufficient. The enzyme requires 1 mM DTT for inactivation.

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