

Micrococcal Nuclease



1-800-632-7799
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www.neb.com



M0247S 010131015101

M0247S

RRR BSA 37°

320,000 gel units Lot: 0101310 Exp: 10/15
2x10⁶ gel units/ml Store at -20°C

Description: Micrococcal nuclease is derived from *Staphylococcus aureus* and is a relatively non-specific endo-exonuclease. It is purified from a recombinant *E. coli* strain that digests double-stranded, single-stranded, circular and linear nucleic acids. The enzyme is active in the pH range of 7.0–10.0, with optimal activity at pH 9.2 for both RNA and DNA substrates. Cleavage preferences have been observed at sites rich in adenylate, deoxyadenylate or thymidylate (1).

More Units

Both DNA and RNA are degraded to 3' phosphomononucleotides and dinucleotides.

Source: An *E. coli* strain containing a genetic fusion of the micrococcal nuclease gene (Gene ID: 3238436) and the gene coding for maltose binding protein, or MBP. The micrococcal nuclease is cleaved from the fusion protein and purified away from MBP.

Applications:

- Degrade nucleic acids present in protein preparations
- *In vitro* translation (2)
- Reduce the viscosity of cell lysates during non-mechanic cell lysis preparation
- Chromatin structure analysis (3)
- Rapid RNA sequencing

Supplied in: 10 mM Tris-HCl (pH 7.5), 50 mM NaCl, 1 mM EDTA and 50% glycerol.

Reagents Supplied with Enzyme:

10X Micrococcal Nuclease Reaction Buffer
100X BSA

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Reaction Conditions: 1X Micrococcal Nuclease Reaction Buffer, supplemented with 100 µg/ml BSA. Incubate at 37°C.

1X Micrococcal Nuclease Reaction Buffer:

50 mM Tris-HCl
5 mM CaCl₂
pH 7.9 @ 25°C

Notes: This enzyme does not work in NEBuffer 1, 2, 3 or 4 due to the lack of Ca²⁺. Additional Ca²⁺ in NEBuffer only shows 10% activity. 1–5 mM Ca²⁺ is required for activity.

The enzyme is active in the pH range 7–10 as long as salt concentration is less than 100 mM.

Enzyme can be inactivated by addition of excess EGTA.

Unit Definition: (Kunitz Unit) One unit is defined as the amount of enzyme required to release acid soluble oligonucleotides that produce an absorbance increase of O.D. 1.0 at 260 nm in 30 minutes at 37°C.

(Agarose Gel Unit) One gel unit is defined as the amount of enzyme required to digest 1 µg of lambda genomic DNA in 15 minutes at 37°C, to the extent that the accumulation of low

molecular DNA fragments (100–400 base pairs) disappears on a 1.2% agarose gel.

Note: 10,000 Gel Units is approximately equal to 1,000 Kunitz Units.

Unit Assay Conditions: (Kunitz Unit) 1X Micrococcal Nuclease Buffer, 0.1mg/ml BSA and 500 µg sonicated Salmon testis genomic DNA in a total volume of 500 µl.

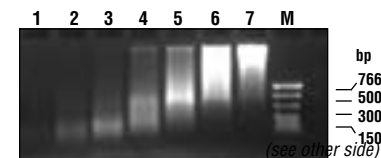


Figure 1: Digestion of 1 µg of Lambda genomic DNA with Micrococcal Nuclease in a 3-fold dilution series. The amount of enzyme used in Lane 2 is defined as 1 gel unit. Lane M is the PCR Marker (NEB #N3234).

CERTIFICATE OF ANALYSIS

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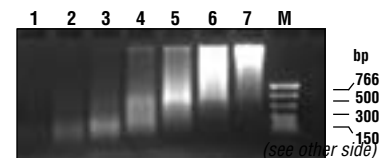


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Quality Assurance: Free of detectable protease activity.

References:

1. Cuatrecasas, S.F., and Anfinsen, C.B. (1967) *J. Biol. Chem.*, 244, 1541–1547.
2. Craig, D. et al. (1992) *Nucl. Acids Res.*, 20, 4987–4955.
3. O'Neill, L.P. and Turner B.M. (2003) *Methods*, 31, 76–82.

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