

OMNI Nuclease

Cat. No.	size
E1120-01	20 000 units
E1120-02	100 000 units

Unit Definition:

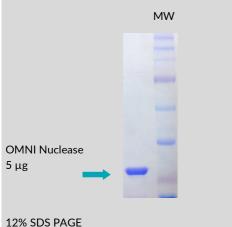
One unit is defined as the amount of enzyme that causes a $\Delta A260$ of 1.0 in 30 min, which corresponds to complete digestion of 37 μg of Herring sperm DNA.

Storage Conditions:

Store at -20°C. Do not store at -80°C.



OMNI Nuclease activity test (1% agarose gel). 37 μ g of Herring sperm DNA (1-4) and Torula Yeast RNA type IV (5-8) was incubated for 30 min at 37°C with increasing amount of OMNI Nuclease units.



MW: 240, 140, 100, 70, 50, 35, 25 and 20 kDa.

References:

- 1.Nestle M., Roberts W.K., (1969) J. Biol. Chem. 244, 5219-5225
- 2.Friedhoff P., Gimadutdinow O., (1994) Protein Expr.Purif. 5, 37-43
- 3.Meiss G., Friedhoff P., (1995) Biochemistry 34, 11979 -11988

OMNI Nuclease catalyzes the removal of all forms of DNA and RNA (linear, circular, double and single stranded) (1). Nuclease completely degrades nucleic acids to 5'-monophosphorate terminated oligonucleotides of 2-6 in length (2, 3).

Applications:

- Viscosity reduction in bacterial, yeast and mammalian protein extracts.
- Sample preparation for protein 2D electrophoresis.
- Removal of nucleic acids contaminants from protein preparations.

Assay Conditions:

50 mM Tris-HCl (pH 8.0), 1 mM MgCl₂, 100 μ g/ml BSA, 37 μ g of Herring sperm DNA. Incubation is carried out at 37°C for 30 min in a reaction volume of 30 μ l.

Storage Buffer:

20 mM Tris-HCl (pH 8.0), 20 mM NaCl, 2 mM MgCl₂, 50% glycerol.

Quality Control:

All preparations are assayed for contaminating protease activity.

Instructions for use:

For removal of nucleic acids:

The substrate DNA or RNA dilute in assay buffer (50 mM Tris-HCl pH 8.0, 1 mM MgCl $_2$, 100 µg/ml BSA) to 2 µg/µl. Incubate the substrate with different units of OMNI Nuclease at 37°C for 30 min. Analyze by gel electrophoresis. To remove trace DNA/RNA use 2 U of nuclease to 1 µg of nucleic acids and extend incubation time to 24 h.

For viscosity reduction:

It is recommended to use 25 U/ml of lysate. The ratio of lysis buffer (ml) to the gram of cells should be 1 g : 1-10 ml

Example reaction:

- 1. 5g of E. coli (wet weight) resuspend in 25 ml of Tris-HCl pH 8.0, 1 mM EDTA (lysozyme, protease inhibitors if needed).
- 2. Add MgCl₂ the final concentration of 5 mM.
- 3. Add 2 µl (500 U) of OMNI Nuclease.
- 4. Use disruption method of your choice (Ultrasonic disruption, French press, etc).
- 5. Obtain clear cell lysate supernatant by centrifugation ~12,000 rpm for 30 min at 4°C.

Notes:

- OMNI Nuclease is very stable, can be diluted with 20 mM Tris-HCl (pH 8.0 at 25°C), 20 mM NaCl, 2 mM MgCl₂ and stored at 4°C for many days without loss of activity.
- 2. Displays a broad pH tolerance (from 6 to 10) and has a wide temperature spectrum (0-44°C).
- 3. OMNI Nuclease is not recommended for purification of proteins that must be nuclease-free. However it can be separated from the target molecule using chromatography for example anion and cation exchange, hydrophobic interaction, hydroxyapatite or size exclusion. OMNI Nuclease does not bind to Ni-NTA resin.
- 4. OMNI Nuclease is inhibited (app. 50% of activity) by:
 - monovalent cation concentrations (Na⁺, K⁺, etc.) > 50 mM,
 - phosphorate concentration > 20 mM,
 - ammonium sulphate concentration > 25 mM.