RE1001-25KU

Benzonase (Recombinant, Molecular Biology Grade)

Size	**25'000 units** (RE1001-25KU) Bulk sizes available, please inquire
Form	Liquid, in storage buffer with 50% glycerol
Activity	1'000 units per microgram
Storage	Store at -20°C.
Shelf life	At least 12 months when stored at -20°C.
CAS No.	9025-65-4

Properties

Source	Purified from yeast cells transformed with a cloned gene encoding genetically engineered endonuclease *from Serratia marcescens*.
Molecular mass	30 kDa
pI	8.9
Activity (pH range)	6-10.0 Optimum pH range 8.0-9.2
Activity (temperature range)	0-42°C Optimum temperature 37°C
Unit Definition	One unit is defined as the amount of enzyme that causes a ΔA_{260} of 1.0 in 30 minutes, which corresponds to complete digestion of 37 µg DNA.
Purity and Quality	2-step purification, > 95% pure, as determined by SDS-PAGE. Proteases: none detected.

Product Description

Serratia marcescens endonuclease, also known as Benzonase, is a non-specific nuclease that degrades both single- and double-stranded nucleic acids, including DNA and RNA, but exhibits no proteolytic activity (1). Therefore, it is ideal for the removal of nucleic acid contaminants from protein samples and applications where complete digestion of nucleic acids is desirable. Benzonase is also commonly used in bioprocessing applications to reduce viscosity of samples caused by genomic DNA. The optimum pH for enzyme activity is 8.0-9.2. It hydrolyzes internal phosphodiester bonds between nucleotides in nucleic acids to produce 5'-monophosphate oligonucleotides of 3-8 bases in length (2). The active enzyme is a homodimer with two disulfide bonds in each monomer that are crucial to the enzyme activity and stability (3). The absolute activity of the recombinant enzyme is measured by a phosphatase coupled assay (4), where the 5'-phosphate of oligosaccharides generated by the enzyme is further released by non-specific alkaline phosphatase and quantitated by Malachite reagents (5).

Preparation Notes

Proprietary storage buffer containing 50% glycerol, pH 7.4.

Applications

- Viscosity reduction in protein extracts
- Sample preparation for 2D gel electrophoresis
- Removal of nucleic acid contaminants from recombinant protein preparations

Benzonase retains its activity under a wide range of conditions as follows.

Condition	Optimal	Effective
Mg^{2+} concentration	1-2 mM	1-10 mM
pH	8.0-9.0	6.0-10.0
Temperature	37°C	0-42°C
Dithiothreitol	0-100 mM	> 100 mM
2-meracptoethanol	0-100 mM	> 100 mM
Monovalent cations (Na^+, K^+, etc)	0-20 mM	0-150 mM

Notes

- Benzonase is inhibited (approximately 50% reduction in relative activity) by monovalent cation concentrations > 50 mM, phosphate concentrations > 20 mM, and by ammonium sulfate concentrations > 25 mM.
- Benzonase can be diluted for ease of handling small quantities with 50 mM Tris-HCl, 20 mM NaCl, 2 mM $MgCl_2$, pH 8.0. Diluted samples can be stored at 4°C for several days without loss of activity.
- Although Benzonase requires Mg^{2+} for activation, it does not appear to require additional Mg^{2+} under many conditions.
- Benzonase treatment is not generally recommended for purification of proteins that must be nuclease free. However, depending on the processing methods, Benzonase may be removed during purification. Residual nuclease activity can be checked by incubation of the purified protein with RNA or DNA markers followed by gel analysis.

References

1. Benedik, MJ and Strych, U. (1998) FEMS Microbiol Lett. 165:1.
2. Nestle, M, et al. (1999) J. Biol. Chem. 274:825.
3. Ball, T.K. et al. (1992) Nucleic Acids Res. 20:4971.
4. Wu, Z.L. et al. (2011) Glycobiology 21:727.
5. Van Veldhoven, P.P. and G.P. Mannaerts (1987) Anal. Biochem. 161:45.

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