

How can we help you?

Answers to questions about

Benzonase® Nuclease

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What type of nucleic acids does Benzonase® Nuclease work on? Can I use it when isolating RNA?

Benzonase Nuclease is a promiscuous endonuclease that attacks and degrades all forms of DNA and RNA (single stranded, double stranded, linear and circular).

For what purposes would I want to use Benzonase Nuclease?

Applications include: reduction of viscosity to facilitate processing (e.g., recombinant protein purification, viscosity-sensitive applications using mammalian cell lysates, etc.); reduction of clumping in stored peripheral blood mononuclear cell (PBMC) samples prior to processing (see Perrin 2008 *inNovations* 28, 21); preparation of inclusion bodies to allow successful protein renaturation; and removal of negatively charged nucleic acids from samples prior to two-dimensional SDS-PAGE.

How can Benzonase Nuclease be inactivated? How can it be removed?

Reversible inhibition can be achieved using EDTA to chelate essential metal ions. Irreversible inactivation can only be accomplished with extreme conditions (100 mM NaOH at 70°C for 30 minutes). Benzonase can be separated from the target product using chromatography. However, because of the robust nature of this endonuclease, we recommend that Benzonase not be used if a nuclease-free end product is required.

My Benzonase Nuclease was left out on the bench. Is it still good?

We have done extensive stability testing on Benzonase Nuclease, and find that it is extremely stable. Even with extended incubations at 37°C, Benzonase Nuclease maintained > 90% activity for several months. For more information on stability testing, see: http://www.merck.de/servlet/PB/show/1249030/w220911_Benzonase_WF.pdf

I want to use a different buffer. What conditions are absolutely required for full activity of Benzonase Nuclease? What will reduce its activity?

Benzonase Nuclease requires 1-2 mM Mg²⁺ for activity. Benzonase is inhibited (approximately 50% activity) by monovalent cation concentrations >50%, phosphate concentrations >20 mM, and by ammonium sulfate concentrations >25 mM.

Is Benzonase Nuclease compatible with protease inhibitor cocktails?

Yes. However, caution should be exercised, since many protease inhibitor cocktails include EDTA. Concentrations of greater than 1 mM EDTA will inhibit Benzonase activity.

My protein is insoluble and I need to perform purification under denaturing conditions. Will Benzonase Nuclease still work in urea?

Benzonase Nuclease activity actually increases in presence of urea at concentrations up to 6 M. At 6 M urea, enzyme activity first increases,

then decreases over time. At 7 M urea, Benzonase Nuclease denatures after 15 minutes, and activity is lost. However, significant degradation of nucleic acids occurs before inactivation. Higher initial concentrations of Benzonase Nuclease can partially compensate the effects of 7 M urea.

Why do you have so many varieties of Benzonase Nuclease? What does HC mean? What impact does 90% versus 99% purity have?

To meet the widest possible range of processing and cost requirements, Benzonase Nuclease is available in two different purity grades: Purity grade I (>99% pure) and Purity grade II (>90% pure). Both grades are available at 25 U/μl or at a high concentration (HC), which is defined as 250 U/μl. For bulk purchases, contact Custom Services.

What is the end result of complete nucleic acid degradation by Benzonase nuclease?

Benzonase nuclease completely digests nucleic acids to 5'-monophosphate terminated oligonucleotides that are 2-5 bases in length.

Product	Size	Cat. No.	Price
Benzonase® Nuclease, Purity > 99%	10 KU	70664-3	\$118
Benzonase® Nuclease HC, Purity > 99%	25 KU	71206-3	\$270
Benzonase® Nuclease, Purity > 90%	2.5 KU 10 KU	70746-4 70746-3	\$31 \$92
Benzonase® Nuclease HC, Purity > 90%	25 KU	71205-3	\$140