

RiboShredder™ RNase Blend

Cat. Nos. RS12100 and RS12500

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1. Introduction

RiboShredder™ RNase Blend is a proprietary blend of potent RNases that completely degrade unwanted RNA in DNA purification procedures. Unlike other RNase cocktails, RiboShredder RNase Blend completely degrades all RNA. RiboShredder RNase Blend uses recombinant, highly purified ribonucleases and thus does not require boiling to remove unwanted DNase activities prior to use. RiboShredder RNase Blend is useful in a number of applications. When used for DNA purification, all unwanted RNA can be removed using a simple 10-minute procedure. After the reaction is complete, RiboShredder RNase Blend can be removed using a phenol-chloroform extraction procedure. It's broad range of salt tolerance makes it ideal for use in many kinds of DNA purification protocols (plasmid, cosmid, BAC, and genomic DNA). For protein purification, RiboShredder RNase Blend degrades RNA efficiently and can be used in concert with DNases (i.e., DNase I) to remove all nucleic acids from a protein preparation, thus reducing the viscosity of the protein solution for easier handling and further purification.

RiboShredder RNase Blend is available in two sizes: 100 Units and 500 Units at 1 U/μl.

2. Product Specifications

Storage: Store only at -20°C in a freezer without a defrost cycle.

Storage Buffer: RiboShredder RNase Blend is supplied in a 50% glycerol solution containing 50 mM Tris-HCl (pH 7.5), 100 mM NaCl, and 0.1 mM EDTA.

Dilution Buffer: RiboShredder RNase Blend may be diluted to a lower working concentration in the indicated Storage Buffer. Dilutions may be stored up to two months at -20°C.

Unit Definition: One unit completely degrades the total RNA component in a reaction consisting of 1 μl of RiboShredder RNase Blend and 10 μl of substrate (13 μg/μl of nucleic acid in T₁₀E₁ buffer) in 10 minutes at 37°C. The assay substrate is the total nucleic acid recovered from a standard alkaline lysis plasmid DNA preparation, (RNA+DNA concentration is 13 μg/μl and the plasmid DNA concentration is ~0.5 μg/μl).

Contaminating Activity Assays: RiboShredder RNase Blend is free of detectable DNase activities as judged by incubation of 10 U of enzyme blend in overdigestion assays on 1 μg of DNA substrate from various sources.

Reaction Buffer: Treatment with RiboShredder RNase Blend can be performed simultaneously with the digestion of plasmid DNA by restriction endonucleases. RiboShredder RNase Blend maintains >90% of its activity in buffers containing 100-200 mM salt (NaCl or KOAc), and the activity is relatively constant over a pH range of 7.0-8.8. If the restriction endonuclease buffer is within these parameters, simultaneous DNA restriction and RNA digestion may be performed.

3. Related Products

The following products are also available:

- OmniCleave™ Endonuclease
- RNase I
- RNase III
- RNase T1
- RNase H
- RNase-Free DNase I

4. RiboShredder Protocol

(removal of RNA from DNA Preparations)

1. Extract the DNA from a 1- to 2-ml overnight bacterial culture using the nucleic acid purification method of your choice.
2. After ethanol precipitation, resuspend the final nucleic acid pellet in T₁₀E₁ Buffer (10 mM Tris-HCl [pH 7.5], 1 mM EDTA) at a concentration appropriate for subsequent applications.
3. Add 1 µl of RiboShredder RNase Blend per 130 µg total nucleic acids to each reaction.
4. Incubate the reaction at 37°C for 10 minutes.
5. Remove RiboShredder RNase Blend by phenol/chloroform extraction. Precipitate the DNA using an ethanol precipitation.

Note: Reactions may be scaled up as needed. Use 1 U of RiboShredder RNase Blend/130 µg total nucleic acids in the sample.

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