Ribonuclease T1, *Aspergillus oryzae*

Cat. Nos. NT09100K and NT09500K
1. Introduction

Ribonuclease T1 (RNase T1, E.C. 3.1.27.3) is an endoribonuclease that specifically cuts RNA or deaminated RNA at the 3’ end of guanosine residues and adjacent nucleotides through a 2’,3’-cyclic phosphate intermediate mechanism. Unlike RNase A which degrades RNA through cleavage at pyrimidine residues, or RNase I which cleaves at many sites along the RNA molecule, RNase T1 specifically cuts RNA molecules at guanine residues, making it suitable for assays where base-specific cleavage is desirable, (e.g., RNA mapping, RNA structure studies and RNA sequencing, due to its cleavage specificity,\(^{1-3}\) or as a general RNase in extraction and purification of DNA from mammalian cells\(^{4}\)). Epicentre’s RNase T1 is cloned from *Aspergillus oryzae* and over expressed in *E. coli* to produce a highly pure enzyme without contaminating nuclease activities. The enzyme is active in a broad range of reaction buffers.

2. Product Specifications

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

**Storage Buffer:** RNase T1 is supplied in a 50% glycerol solution containing 50 mM Tris-HCl (pH 7.5), 100 mM NaCl, and 0.1 mM EDTA.

**Unit Definition:** One unit converts 100 ng of *E. coli* ribosomal RNA into acid-soluble nucleotides in 5 seconds at 37°C.

**Quality Control:** RNase T1 is function-tested in a 100-μl reaction containing: 120 μg *E. coli* ribosomal RNA in 50 mM Tris-HCl (pH 7.0) and 2 mM EDTA for 10 minutes at 37°C.

**Contaminating Activity Assays:** RNase T1 is free of detectable DNA exo- and endonuclease activities, and non-specific RNase activities.

**Reaction Buffer:** RNase T1 treatment can be performed simultaneously with the digestion of plasmid DNA by restriction endonucleases. RNase T1 maintains >90% of its activity in buffers containing 100-200 mM salt (NaCl or KOAc). The activity of RNase T1 is also relatively constant over a pH range of 7.0-8.8. Therefore, if the restriction endonuclease buffer is within these parameters, digestion of contaminating RNA using RNase T1 can be performed in the restriction endonuclease buffer.

Since divalent cations (Mn\(^2+\), Cu\(^2+\), Zn\(^2+\)) inhibit the activity of RNase T1, EDTA should be included in the reaction buffer. RNase T1 activity can be stopped by the addition of 1/10 volume 10% SDS followed by phenol extraction.

**Dilution Buffer:** RNase T1 may be diluted to a lower working concentration in the indicated Storage Buffer.

3. Kit contents

<table>
<thead>
<tr>
<th>Cat. #</th>
<th>Quantity</th>
</tr>
</thead>
<tbody>
<tr>
<td>NT09100K</td>
<td>100,000 Units</td>
</tr>
<tr>
<td>NT09500K</td>
<td>500,000 Units</td>
</tr>
</tbody>
</table>

Ribonuclease T1 is available in two sizes:

Each is supplied at a concentration of 1,000 Units/μl.
4. **Related Products**

The following products are also available:

- RiboShredder™ RNase Blend
- RNase I
- RNase III
- OmniCleave™ Endonuclease
- MasterPure™ DNA Purification Kit for Blood
- MasterPure™ Complete DNA and RNA Purification Kit

5. **References**


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