Thermostable RNase T1

REF: EG24211-S/M

Storage Condition

-20℃

Components

Component	EG24211S	EG24211M
Thermostable RNase T1 (500 U/μl)	200 μl	5×200 μl
10× RNase T1 Buffer	1 ml	5×1 ml

Description

Thermostable RNase T1 is a thermostable mutant derived from RNase T1 through directed protein evolution. Compared to the wild-type RNase T1, Thermostable RNase T1 retains 100% of its activity at 50 ℃ , while the activity of the wild-type decreases by at least 50% at the same temperature. The higher reaction temperature facilitates the complete unfolding of RNA secondary structures, reducing or avoiding their interference with the enzymatic cleavage process, thereby making the digestion more thorough.

Definition of Activity Unit

One unit of the enzyme causes an increase in absorbance of 1.0 at 260 nm in 15 min when yeast RNA is hydrolyzed at 37℃ and pH7.5.

Applications

1. RNA removal from recombinant proteins.
2. RNA removal from recombinant proteins.
3. RNA sequencing.
4. RNA protection assay (in conjunction with RNase A).
5. Detecting the transcription level of G-free or low-G DNA templates.

Quality Control Assays

Protein Purity

The enzyme is ≥95% pure as determined by SDS-PAGE analysis using Coomassie Blue staining.

Endonuclease Activity

A 20 μl reaction containing 200 ng of supercoiled plasmid and 500 U of Thermostable RNase T1 incubated for 4 hours at 37 ℃ results in <20% conversion to the nicked or linearized form as determined by agarose gel electrophoresis.

Non-specific Nuclease Activity

A 20 μl reaction containing 15 ng of dsDNA fragments and 500 U of Thermostable RNase T1 incubated for 16 hours at 37 ℃ results in no detectable degradation of the dsDNA fragments as determined by agarose gel electrophoresis.

Protocol

1. Reaction system

Reagent	Amount	Final Concentration
ssRNA	1~10 μg	50~500 ng/μl
10× RNase T1 Buffer	2 μl	1×
RNase Inhibitor (40 U/μl)*	1 μl	2 U/μl
Thermostable RNase T1 (500 U/μl)	0.4 μl	10 U/μl
Nuclease-Free Water	up to 20 μl	-

Note: To prevent RNA degradation, it is recommended to add RNase Inhibitor (REF: EG20002) into the reaction mixture Substrate RNA should be added after the addition of RNase Inhibitor.

2. Recommended reaction conditions

Incubate at 50 ℃ for 15 minutes. The incubation time can be appropriately extended or shortened to ensure sufficient digestion.

Notice

1. Thermostable RNase T1 after heat-inactivation is reversible. It is recommended to remove Thermostable RNase T1 by column purification or phenol/chloroform extraction.
2. RNase-related experiments should be conducted in a fume hood or a designated area to avoid affecting other experiments.
3. For your safety and health, please wear a lab coat, disposable glovesand a mask while conducting the experiment.

Add: No.17 Huaguoshan Avenue,Lianyungang City, China
Tel: +86 (518) 8558 6628 • E-mail:support@best-enzymes.com • www.best-enzymes.com