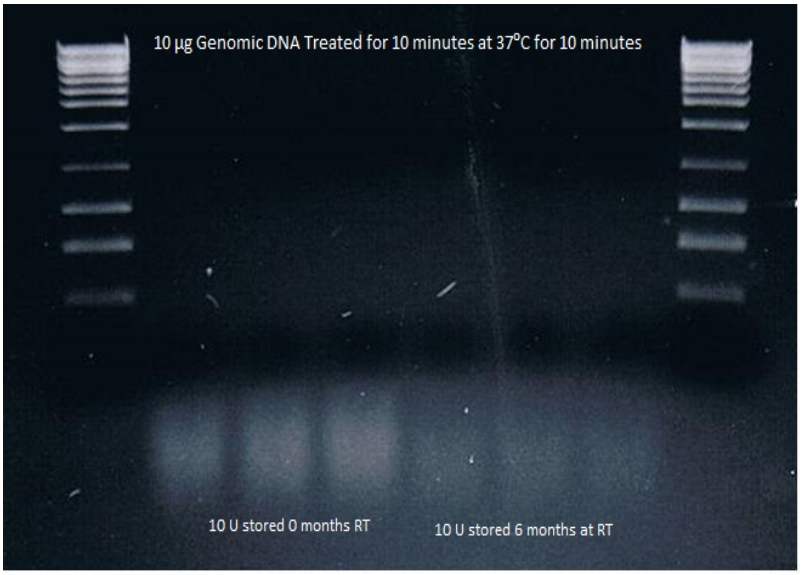
RT Stable DNase I System

Ideal For:

- Field Isolation of RNA where refrigeration is scarce.
- High Throughput Labs
- Users wanting to keep all reagents at RT together
- Any Application Needing Long DNase Shelf Life

Unique Features:

- Stable up to 6 months at RT
- Unique inactivation resin for removal of enzyme and divalents.



Concentration: 10 U/µl, 100 U//µl

Purity: No contaminating bands detected by loading 5 ug of RT DNase on a 4-20% Tris-Glycine SDS gel and staining with SimplyBlue™ (Invitrogen Corporation) gel stain for 1 hour.

Unit Definition: One unit is the amount of enzyme that degrades 3 µg of Lambda DNA completely at 37ºC in 10 minute in 1X RT DNase Reaction Buffer.

Storage Buffer: 50 mM Tris pH 7.5, 50% Glycerol, Stabilizing Cations. Store at -20ºC.

INTRODUCTION

RT DNase I System is a highly purified DNase formulated for extended stability at room temperature and 4ºC coupled with a highly active buffer and removal resin. RT DNase I is ideal for removal of any contaminating genomic DNA in RNA or protein preparations. The 10 U formulation can remove 30 µg of DNA in 10 minutes and the 100 U formulation can remove up to 300 µg of DNA in 10 minutes. RT DNase I is stable up to 6 months at standard room temperature (20-25ºC) and up to 2 years at 4ºC without detectible loss of activity (see photo above). The system can be purchased with a highly specific resin which is used to bind and remove the RT DNase and the subsequent cations from the reaction which saves an EDTA or heating step. The RNA or protein sample is ready to use after treatment.

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Protocol

Wear gloves at all times, especially when working with RNA

Do not vigorously vortex DNase Enzyme prior to or during reaction set up. This will cause loss of activity. Pipetting up and down several times is sufficient for reaction mixing.

For DNA Removal:

- Mix 1 µl of RT DNase I enzyme and 10X RT DNase Reaction Buffer to a final concentration of 1X in the reaction mix. For every 10 µl of reaction mix, this would be 1 µl of reaction buffer. It is highly recommended for optimal activity to perform these reactions in non-stick tubes.
- Bring the reaction to final volume with RNase-free water. Remember to mix by pipetting up and down and do not vortex.
- Incubate the reaction at 37º for 10 to 20 minutes.

DNase Removal with Resin:

- Add 5 µl of homogeneous DNase Max™ Removal Resin per 10 units of RT DNase in a 50 µl reaction, or 10 µl of slurry for every 100 µl of reaction volume, whichever is greater.
- Incubate for 10-20 minutes at room temperature. Invert or flick to resuspend every 1-2 minutes or place the tubes on a vortex adapter attached to a Vortex Genie 2 and set the vortex between speed 5-6 to agitate the resin and promote binding of the DNase to the resin in the reaction. The agitation should be thorough but not promote splashing.
- Centrifuge 13,000 x g for 1 minute to pellet the resin.
- Transfer the supernatant to a new tube, taking care not to transfer any of the resin.
- The sample is now ready for downstream use.

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