RNase-Free DNase I Set

For research use only

Application: In column DNase I digestion and DNA digestion in solution with Geneaid RNA extraction kits

Storage: -20ºC for extended periods

Unit Definition: One Kunitz unit is defined as the amount of enzyme required to produce an increase in absorbance of 260 nm of 0.001/min/ml at 25ºC of highly polymerized DNA

Geneaid

CERTIFICATE NO. QAIC/TW/50077
ISO 9001:2015 QMS

Introduction

The RNase-Free DNase I set ensures complete DNA removal from RNA extracted using Geneaid's RNA extraction kits for use in DNA sensitive downstream applications. This is an optional treatment as Geneaid's spin column technology yields RNA with the majority of DNA removed. Without using DNase treatment, the extracted RNA can be used in downstream applications which are not DNA sensitive. This set can be used for both in column DNase digestion and DNase digestion in solution.

Quality Control

The quality of the RNase-Free DNase I set is tested on a lot-to-lot basis according to Geneaid's ISO-certified quality management system.

Kit Contents

Component	DNS050	DNS100	DNS300
DNase I (2U/µl)	275 µl	550 µl	550 µl x 3
DNase I Reaction Buffer	2.5 ml	5 ml	15 ml
Stop Solution	110 µl	110 µl	350 µl

In Column DNase I Digestion Protocol (Following RNA Binding)

IMPORTANT: DNA contamination is significantly reduced following In Column DNase I Digestion. However, traces of residual DNA may be detected in very sensitive applications. In this situation, please perform DNA Digestion In Solution instead to efficiently remove trace amounts of DNA. Standard DNase buffers are incompatible with In Column DNase I Digestion and may effect RNA integrity and reduce yield.

1. Add 400 µl of Wash Buffer (make sure ethanol was added) to the RB Column.
2. Centrifuge at 14-16,000 x g for 30 seconds.
3. Discard the flow-through and place the RB Column back in the 2 ml Collection Tube.
4. Prepare DNase I reaction solution in a 1.5 ml microcentrifuge tube (RNase-free) as follows:

DNase I (2U/µl)	5 µl
DNase I Reaction Buffer	45 µl
Total volume	50 µl

5. Gently pipette the DNase I reaction solution to mix (DO NOT vortex) then add DNase I solution (50 µl) into the CENTER of the RB column matrix.
6. Incubate the column for 15 minutes at room temperature (20-30ºC) then proceed with the RNA Wash step.

DNA Digestion In Solution Protocol (Following RNA Elution)

1. Prepare DNase I reaction solution in a 1.5 ml microcentrifuge tube (RNase-free) as follows:

RNA in RNase-free water	1 - 40 µl
DNase I (2U/µl)	0.5 µl/µg RNA
DNase I Reaction Buffer	5 µl
RNase-free water	Add to final volume = 50 µl
Total volume	50 µl

2. Gently pipette the DNase I reaction solution to mix (DO NOT vortex) then incubate at 37ºC for 15-30 minutes.
3. Stop the reaction by adding 1 µl of Stop Solution then incubate the microcentrifuge tube at 75ºC for 10 minutes.

NOTE: DNase I Reaction Buffer may cause aberrant migration or smearing of RNA on gels. If analyzing RNA by gel electrophoresis, repurify the RNA sample using the Geneaid™ RNA Cleanup Kit instead of stopping the reaction with Stop Solution.