

EcoZyme RNase A

Troubleshooting Guide

1 mL, 5 x 1 mL

Cat No: EZRA1, EZRA5

Use this guide for common RNA-digestion and handling problems encountered with EcoZyme RNase A solution. The product is supplied as a 10 mg/mL molecular biology grade solution and is intended for RNA digestion in plasmid and genomic DNA purification procedures.

Before You Start - Quick Checks

1. The ready-to-use solution is supplied at 10 mg/mL in 10 mM Tris-HCl, pH 8.
2. Store at -20°C.
3. EcoZyme RNase A is described as DNase- and protease-free and is pretreated to remove DNase I.
4. The enzyme specifically degrades single-stranded RNA at C and U residues.
5. Use the RNase A amount and incubation conditions specified by the DNA-purification workflow in which it is being used.

Problem	Most likely cause	Recommended action
Residual RNA remains after treatment	The amount of active RNase A or the exposure used in the purification workflow may be insufficient.	Verify that the 10 mg/mL stock was added at the amount specified by the validated plasmid or genomic DNA purification protocol.
RNA contamination varies between samples	RNase A dosing or sample input may not be consistent.	Keep sample input and RNase A addition consistent across comparable samples and mix the reaction uniformly.
Low performance after storage	The enzyme may not have been stored at the specified temperature.	Store at -20°C and compare with a properly stored aliquot if storage history is uncertain.
DNA yield or integrity is unexpectedly poor	The datasheet describes the product as DNase-free, so the cause may be elsewhere in the DNA purification workflow.	Verify the RNase A identity and amount, then review lysis, binding, wash, and handling steps for DNA loss or damage.
Unexpected proteolytic effect is suspected	The product is described as protease-free.	Check for contamination or other reagents in the workflow rather than attributing proteolysis to the RNase A stock.
Results are inconsistent after repeated use	Aliquots may have been handled or pipetted inconsistently.	Use clean tips, keep the stock at the specified storage temperature, and dispense the same calculated volume for each reaction.
RNA removal is poor in high-input samples	The amount of RNase A may be insufficient relative to the sample load.	Use the sample input and RNase A amount validated for the purification protocol; reduce sample load or optimize the RNase A dose if required.
RNase A was added to the wrong workflow step	Timing of RNase treatment may not match the purification procedure.	Follow the plasmid or genomic DNA purification protocol for the point of RNase A addition rather than adding it arbitrarily.
Unexpected contamination between samples	RNase A or sample may have been transferred with a reused tip or contaminated pipette.	Use a fresh tip for each sample and keep the stock tube and work area clean.

Still having trouble? Verify -20°C storage, confirm that the 10 mg/mL stock was used at the amount specified by the DNA-purification workflow, and check sample input consistency. If DNA quality is unexpectedly poor, investigate the broader purification workflow because the product is specified as DNase- and protease-free.