

Removal of Genomic DNA from RNA Preparations

This protocol is for the Removal of Genomic DNA from RNA Preparations

1. Add to an RNase-free tube:

RNA	1 µg
10X reaction buffer with MgCl₂	1 µl
DNase I, RNase-free	1 µl (1 u)
DEPC-treated Water	to 10 µl
Total volume	10 µl

2. Incubate 30 min at 37°C.
3. Add 1 µl of 50 mM EDTA and incubate 10 min at 65°C. RNA hydrolyzes if heated in the absence of a chelating agent (1).
4. Use the prepared RNA as a template for reverse transcriptase.

Note

- Do not use more than 1 u of DNase I, RNase-free per µg of RNA.
- Reaction mixture can be scaled up for larger amounts of RNA. The recommended final concentration of RNA is 0.1-0.2 µg/µl.
- RiboLock™ RNase Inhibitor (#EO0381), typically at 1 u/µl, can also be included in the reaction mixture to inactivate type A RNases potentially present in the initial RNA solution

Reference

1. Wiame, I., et al., Irreversible heat inactivation of DNase I without RNA degradation, *BioTechniques*, 29, 252-256, 2000.

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