

## RNA/DNA Purification Kit and RNA/DNA Purification Micro Kits Supplementary Protocol

RNA/DNA Purification Kit Cat# 48600, 48700  
RNA/DNA Purification Micro Kit Cat# 50300

### Protocol for Optional On-Column DNA Removal

Norgen's RNA/DNA Purification Kits isolates total RNA with minimal amounts of genomic DNA contamination. However, an optional protocol is provided below for maximum removal of residual DNA that may affect sensitive downstream applications. It is recommended that Norgen's RNase-Free DNase I Kit (Product # 25710) be used for this step.

1. For every on-column reaction to be performed, prepare a mix of 15  $\mu$ L of **DNase I** and 100  $\mu$ L of **Enzyme Incubation Buffer** using Norgen's RNase-Free DNase I Kit (Product # 25710). Mix gently by inverting the tube a few times. **DO NOT VORTEX**.

**Note:** If using an alternative DNase I, prepare a working stock of 0.25 Kunitz unit/ $\mu$ L RNase-free DNase I solution according to the manufacturer's instructions. A 100  $\mu$ L aliquot is required for each column to be treated.

2. Perform the appropriate DNA and RNA Purification Procedure for your starting material up to and including "**Binding RNA to Column**" (Steps 1 through Step 6 of all protocols)
3. Apply 400  $\mu$ L of **Wash Solution** to the column and centrifuge for 2 minutes. Discard the flowthrough. Reassemble the spin column with its collection tube.
4. Apply 100  $\mu$ L of the RNase-free DNase I solution prepared in Step 1 to the column and centrifuge at 14, 000 x g (~14 000 RPM) for 1 minute.

**Note:** Ensure that the entire DNase I solution passes through the column. If needed, spin at 14, 000 x g (~14 000 RPM) for an additional minute.

5. After the centrifugation in Step 4, pipette the flowthrough that is present in the collection tube back onto the top of the column.

**Note:** Ensure Step 5 is performed in order to ensure maximum DNase activity and to obtain maximum yields of RNA, in particular for small RNA species.

6. Incubate the column assembly at 25 - 30°C for 15 minutes.
7. Without any further centrifugation, proceed directly to the second wash step in the "**RNA Wash**" section (Step 7c).

Related Products	Product #
RNase-Free DNase I Kit	25710

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