Protocol

# Guidelines to Avoid RNase Contamination

## This protocol is for the Guidelines to Avoid RNase Contamination

RNA purity and integrity is essential for synthesis of full-length cDNA, which results in high quality RT-PCR products. Therefore, RNase contamination is always a concern when working with RNA. The RNA quality can be affected by RNase A, which is a highly stable contaminant of any laboratory environment. All components of the kit have been rigorously tested to ensure that they are RNase free. To prevent contamination both the laboratory environment and all prepared solutions must be free of RNases.

- DEPC-treat all tubes and pipette tips to be used in the cDNA synthesis or use certified nuclease-free labware.
- Use pipettes dedicated for RNA work.
- Wear gloves when handling RNA and all reagents, as skin is an common source of RNases. Change gloves frequently.
- Use certified reagents, including high quality water (e.g., nuclease-free or DEPC-treated Water).
- Use an RNase inhibitor, such as RiboLock™ RNase Inhibitor, to protect template RNA.
- Always assess the integrity of RNA prior to cDNA synthesis. For example, if sharp bands of both the human 18S rRNA (runs at approx. 1.9 kb) and the 28S rRNA (runs at approx. 5 kb) are formed during denaturing agarose gel electrophoresis of total RNA, the mRNA in the sample is considered to be intact.

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**North America**
**Technical Services:**
techservice.genomics@thermofisher.com
**Customer Services:**
customerservice.genomics@
thermofisher.com
**Tel** 800 235 9880
**Fax** 800 292 6088

**Europe and Asia**
**Technical Services:**
techservice.emea.genomics@
thermofisher.com
**Customer Services:**
customerservice.emea.genomics@
thermofisher.com

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