The total RNA was isolated using a modified guanidinium thiocyanate method. Our RNA isolation method has been validated to co-purify both miRNA and mRNA and delivers RNA suitable for qRT-PCR, miRNA detection, Northern blot analysis, cDNA synthesis, RT-PCR, in vitro translation, ribonuclease protection assays, S1 nuclease analysis, and microarray target labeling.

Total RNA is provided in 0.1 mM EDTA, pH 8.0. The small amount of EDTA does not interfere with downstream enzymatic manipulations. **Note:** To avoid any possible RNase contamination, always wear gloves when handling RNA.

**Test Conditions**

The quality of the RNA is assessed visually by observing distinct 28S and 18S ribosomal bands on a non-denaturing 1× MOPS gel. The purity of the RNA is assessed by spectrophotometry (A_{260}/A_{280} ≥ 1.8). The RNA is shown to be free of contaminating RNases by incubation in a suitable buffer at 37°C. The RNA is further tested functionally by Northern analysis using a human β-actin probe and by RT-PCR.

**Reference**