

Tips for analyzing results (from Applied Biosystems/Ambion technical resources):
http://www.ambion.com/techlib/append/supp/rna_gel.html

Intact total RNA run on a denaturing gel will have sharp 28S and 18S rRNA bands (eukaryotic samples). The 28S rRNA band should be approximately twice as intense as the 18S rRNA band (Figure 1, lane 3). This 2:1 ratio (28S:18S) is a good indication that the RNA is intact. Partially degraded RNA will have a smeared appearance, will lack the sharp rRNA bands, or will not exhibit a 2:1 ratio. Completely degraded RNA will appear as a very low molecular weight smear (Figure 1, lane 2). Inclusion of RNA size markers on the gel will allow the size of any bands or smears to be determined and will also serve as a good control to ensure the gel was run properly (Figure 1, lane 1). Note: Poly(A) selected samples will not contain strong rRNA bands and will appear as a smear from approximately 6 kb to 0.5 kb (resulting from the population of mRNAs, and depending on exposure times and conditions), with the area between 1.5 and 2 kb being the most intense (this smear is sometimes apparent in total RNA samples as well).

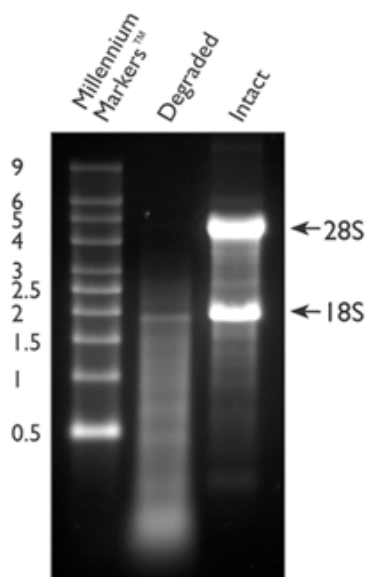


Figure 1. Intact vs. Degraded RNA. Two μg of degraded total RNA and intact total RNA were run beside Ambion's RNA Millennium Markers™ on a 1.5% denaturing agarose gel. The 18S and 28S ribosomal RNA bands are clearly visible in the intact RNA sample. The degraded RNA appears as a lower molecular weight smear.