DNase Treatment of Total RNA

1. **Starting material: RNA sample at <10ug/50ul**
2. **DNase I (Ambion AM2222).**

Reaction Mix		**(x1)**
RNA	10ug	n ul
10X DNase I Reaction Buffer		5ul
DNase I	2U	1ul
RNase-free water		44-n ul
Total		**50ul**

3. Incubate at 37°C, 30mins.
4. **Qiagen RNeasy MinElute Clean up protocol (modified). Column capacity at 45ug.**
5. Add 50ul RNase-free water to sample (Total volume at 100ul).
6. Add 350ul Buffer RLT, mix well.
7. Add 700ul 100% ethanol, mix well.
8. Transfer sample to column. Spin at >8000g, 15s.
9. Repeat for the remaining.
10. Wash column with 500ul Buffer RPE.
11. Wash column with 500ul of 80% ethanol.
12. Place column in a new collection tube.
13. Spin at 14000g for 5mins to dry the column.
14. Place column in a new tube for eluate.
15. Add 14ul RNase-free water to the center of the spin column.
16. Spin at 14000g, 1 min.