

1. Mix the following components in a PCR tube:

X	20 μ L
B	20 μ L
Total RNA (1 ng – 1 μ g)	Up to 60 μ L
H₂O	Fill to 100 μ L

2. Vortex and **spin down**.

3. Anneal the samples in a thermocycler:

- 95°C, 3 min
- Cool from 80°C to 20°C over 20 min
- Hold at 20°C until centrifugation

4. Centrifuge at $\geq 10,000 \times G$ for 30 min.*

5. Obtain up to 70 μ L of the **supernatant** (rRNA-depleted library).

The final rRNA-depleted library contains 5 mM Tris, 1 mM EDTA, and 150 mM NaCl. If necessary, remove leftover DNA probes with DNase I; isolate purified RNA with a silica spin tube.

* You may use Dynamic Matrices Microcentrifuge Inserts (Cat. no. MI08)

LASSO workflow

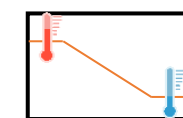
1. Mix



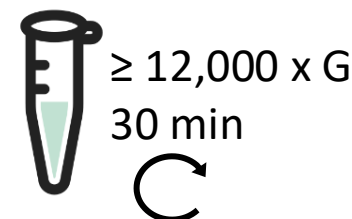
2. Vortex



3. Anneal



4. Spin



5. Obtain supernatant



Annealing protocol

