

## USING THE AGILENT BIOANALYZER

**Purpose:** to analyze RNA for quality prior to use in hybridization experiments

### Materials Needed:

- RNA 6000 Nano LabChip Kit (Agilent, cat#5065-4476)
  - RNA gel matrix
  - RNA dye concentrate
  - RNA chip
  - RNA 6000 Nano Markers
- Spin filters
- RNA 6000 Ladder (Ambion, cat#7152)

### Preparation:

1. Prepare the gel-dye mix.
2. Put 400 ul of RNA gel matrix (red top) into a spin filter.
3. Centrifuge at 1500g $\pm$ 20% for 10 minutes. USE FILTERED GEL WITHIN 4 WEEKS.
4. Mix 130ul of filtered RNA gel matrix with 2 ul of RNA dye concentrate (blue top) in a RNase free 1.5ml microcentrifuge tube.
5. Vortex solution well. PROTECT SOLUTION FROM LIGHT. STORE AT 4 DEGREES C. USE WITHIN ONE WEEK.

### Procedure:

Loading the gel-dye mix

1. Put a new RNA chip on the Chip Priming Station.
2. Pipette 9.0 ul of gel-dye mix into the well marked "G" in a black circle.
3. Close the Chip Priming Station.
4. Press plunger until it is held by the clip.
5. Wait for exactly 30 seconds, then release clip.
6. Open the Chip Priming Station and check chip for air bubbles.
7. Pipette 9 ul of gel-dye mix in the wells Marked "G" in a purple square.

Loading the RNA 6000 Nano Marker

1. Pipette 5 ul of RNA 6000 Nano Marker (green top) into the well with the ladder picture next to it.
2. Pipette 5 ul of RNA 600 Nano Marker into all twelve sample wells.

Loading the Ladder

1. Pipette 1ul of RNA 6000 Ladder into the well with the ladder picture next to it.

## Loading the Samples

1. Pipette 1ul of sample in each of the twelve sample wells. Pipette 1ul of RNA 6000 Nano marker in each unused sample well.
2. Put the chip in the adapter and vortex for one minute at the set-point of the IKA vortexer.
3. Run the chip in the Bioanalyzer within 5 minutes.

## Comments:

## Contacts:

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For frequently asked questions go to the following address:  
<http://www.grc.nia.nih.gov/branches/rrb/dna/protocolFAQs.htm>

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