RNA Assay Quick Guide
LabChip® GX Touch/GXII Touch

Note: Allow the chip and reagents to equilibrate to room temperature for about 20 minutes before use. ***The Dye Concentrate must be completely thawed and vortexed before use***.

Preparation of Gel-Dye Solution

1. Vortex the thawed Dye Solution for 20 seconds before use.
2. Transfer **90 µL** of RNA Dye Solution (blue cap) to **1 vial** of the RNA Gel Matrix (red cap).
3. Vortex and invert the tube several times until the solution is well mixed and spin it down for a few seconds.
4. Transfer the solution into one spin filter and centrifuge at 9300 rcf for 5 min at RT.
5. Remove the filter in the tube and discard the filter.

Low-Throughput Chip Preparation - up to 48 samples

1. Rinse and aspirate each active well (1, 3, 4, 7, 8 and 10) twice with nuclease free water.
2. Add prepared Gel-Dye to chip wells 3, 7, 8 and 10 (as shown in Figure 1) using a Reverse Pipetting Technique.
3. Add **50 µL** RNA Marker to chip well 4 (as shown in Figure 1).
4. Clean both sides of the chip window with the supplied clean room cloth dampened with 70% isopropanol.

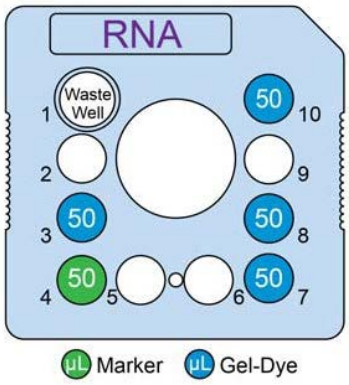

Figure 1. Low-Throughput Chip Preparation

High-Throughput Chip Preparation - up to 192 samples (well 10 must be refreshed after 96 samples*)

***NOTE:** When running > 96 samples, refresh well 10 by removing the contents and replacing with 120 µL unused Gel-Dye remaining from the earlier filtered Gel-Dye prep. See the *RNA Assay User Guide* for details.

1. Rinse and aspirate each active well (1, 3, 4, 7, 8 and 10) twice with nuclease-free water.
2. Add prepared Gel-Dye to chip wells 3, 7, 8 and 10 (as shown in Figure 2) using a Reverse Pipetting Technique.
3. Add **100 µL** RNA Marker to chip well 4 (as shown in Figure 2).
4. Clean both sides of the chip window with the supplied clean room cloth dampened with 70% isopropanol.

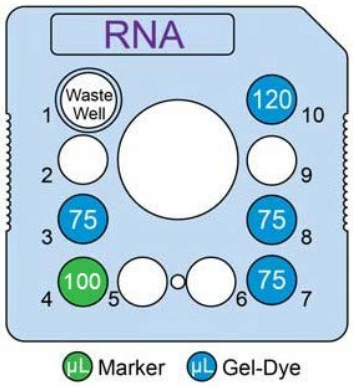

Figure 2. High-Throughput Chip Preparation

NOTE: The volume of Gel-Dye solution prepared is the amount required for one HT (High-Throughput) or two LT (Low-Throughput) chip preps.

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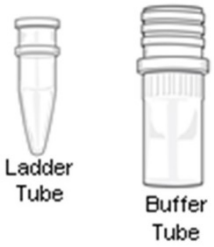
RNA Sample, Ladder and Buffer Preparation

Note: The RNA ladder should be kept on ice. Avoid multiple freeze/thaws.
It is recommended that you aliquot the RNA ladder into five 4 µL lots for individual use.

1. Prepare Sample Buffer by adding **620 µL** RNA Sample Buffer Concentrate to **5580 µL** DEPC treated or nuclease-free water.

   Note: The RNA Sample Buffer Concentrate is a 10X solution. Sample Buffer is stable after dilution, but to avoid RNase contamination, sample buffer should be prepared fresh.

2. Thaw RNA ladder on ice.
3. Transfer **4 µL** RNA Ladder into RNase-free microcentrifuge tube or a well of the microtiter plate.
4. For each sample to be analyzed, pipette **2 µL** (RNA Std Sens) or **6 µL** (RNA High Sens) sample into individual microtiter plate wells (cover with PCR strip caps) or RNase-free microcentrifuge tubes.
5. Cover and heat the ladder and samples at **70°C for 2 minutes**.
6. Snap cool the samples and ladder by immediately placing the tubes and/or microtiter plate on **ice for 5 minutes**.
7. Add **46 µL** (RNA Std Sens) or **19 µL** (RNA High Sens) prepared sample buffer to each sample. Cover the samples with PCR strip caps and spin down the plate.
8. Add **96 µL** prepared sample buffer to the Ladder Tube.
9. Add **750 µL** prepared sample buffer to the provided Buffer Tube.



Chip Cleaning and Storage

After use, the chip must be cleaned and stored in the chip container. The cleaning procedure can be conducted the following day, when running overnight.

1. Remove reagents from each well using a vacuum.
2. Rinse and thoroughly aspirate each active well (1, 3, 4, 7, 8 and 10) twice with molecular biology-grade water.
3. Add **100 µL** of Storage Buffer to active wells.
4. Place the chip back on the LabChip GX Touch/GXII Touch and touch the **Wash** button. Ensure a Buffer Tube with **750 µL** buffer or molecular biology-grade water is in the buffer slot.
5. Remove the chip from the LabChip GX Touch/GXII Touch and place in chip container.
6. Add an additional **50 µL** of Storage Buffer to wells 1 and 4.
7. Make sure to cover all wells with Parafilm® and store at 4°C.

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Assay Specifications

Linear Range	25 ng/μL – 250 ng/μL (Std Sens) 5 ng/μL – 50 ng/μL (High Sens)
Quantitation Reproducibility	20% CV
Size Range	100 – 6000 nucleotides (suitable for total RNA)
RNA Sample Volume	2 μL of user sample (Std Sens) 6 μL of user sample (High Sens)
Run Time	80 seconds per sample (about 2.5 hours for 96-well plate)
Setup Time	Approximately 30 minutes to prepare chip and samples
Number of Samples per Chip Prep	Up to 192 per HT chip prep Up to 48 per LT chip prep
Number of Chip Preps per Reagent Kit	5 (HT chip prep), 10 (LT chip prep)
For Research Use Only	

For complete RNA Assay User Guide, go to:

http://www.perkinelmer.com/labchipsystems

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