ssDNA 7K Assay Quick Guide LabChip[®] GX Touch/GXII Touch

Notes:

- Allow the DNA 5K/RNA/CZE HT LabChip (P/N 760435) and all refrigerated ssDNA 7K reagents (Reagent Kit P/N CLS158169) to equilibrate to room temperature for at least 30 minutes before use.
- The ssDNA Dye Concentrate must be thawed completely and vortexed before use. Protect the Dye and Gel-Dye Solution from light.
- Thaw the ssDNA 7K Ladder on ice.
- Upload the ssDNA 7K.asyx file (P/N CLS157952) into the "C:\Program Files\...\LabChip GX Touch\Assay" folder on the LabChip GX Touch instrument before running the assay.

Preparing the Gel-Dye Solution

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

Warning: The dye is light sensitive. Do not expose the Dye or Gel-Dye solution to light for any length of time. Keep the prepared Gel-Dye Solution in the dark.

- 1. Allow the chip and all refrigerated reagents to equilibrate to room temperature for at least 30 minutes before use.
- 2. Vortex the thawed ssDNA Dye Concentrate for 10 15 seconds before use.
- Transfer 90 μL of ssDNA Dye Concentrate (blue cap) to a tube with 510 μL ssDNA Gel Matrix (red cap).
- 4. Vortex and invert this dye-gel tube several times until the solution is well mixed and then spin down for a few seconds.
- 5. Transfer the Gel-Dye Solution into a spin filter and centrifuge at 9300 rcf for 10 min at RT.
- 6. Discard the filter. Label and date the tube, and then store in the dark at 2-8°C. Use within 5 days.

Low-Throughput (LT) Chip Preparation - up to 48 samples and

High-Throughput (HT) Chip Preparation - up to 96 samples

- 1. Rinse and completely aspirate each active well (1, 3, 4, 7, 8, and 10) twice with nuclease-free water (Milli-Q[®] or equivalent).
- 2. Using a Reverse Pipetting Technique, add Gel-Dye solution to chip wells 3, 7, 8, and 10 as shown in **Figure 1 (LT)** or **Figure 2 (HT)**.
- 3. Add **50 μL (LT)** or **100 μL (HT) of ssDNA Marker** to chip well **4** as shown in **Figure 1 (LT)** or **Figure 2 (HT)**.
- 4. Clean both sides of the chip window with the supplied clean room cloth dampened with 70% isopropanol.
- 5. Make sure the rims of the chip wells are clean and dry.
- 6. **IMPORTANT**: Ensure chip well 1 (waste well) is empty before placing the chip into the LabChip GX Touch/GXII Touch.



Figure 1. Low-Throughput Chip Preparation



Figure 2. High-Throughput Chip Preparation



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Sample, Ladder and Buffer Preparation



Figure 3. Sample, Ladder Tube, and Buffer Tube Preparation

- 1. Prepare **1X Sample Buffer** by adding **200 μL ssDNA Sample Buffer Concentrate** ^Ο to **1800 μL DEPC treated or nuclease-free water**. (**Note:** The 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. Prepare sample, Ladder Tube, and Buffer Tube according to Figure 3.
- 3. Load the prepared plate, chip, ladder tube, and buffer tube into the instrument.
- 4. In the LabChip[®] GX Touch[™] software, select "HT DNA5K" assay, then click "**ssDNA 7K**" program to RUN.
- **Note:** There is no need for heating treatment of ssDNA Ladder. Denature process by chemical or heat might be required for samples which are depended on sample preparation process and storage buffer.

Chip Cleaning and Storage

After use, the chip must be cleaned and then stored in the chip container.

- 1. Place the chip into the chip storage container. Verify the sipper is submerged in the fluid reservoir.
- 2. Remove reagents from each well using vacuum.
- 3. Rinse and completely aspirate each active well (1, 3, 4, 7, 8, and 10) twice with water (Milli-Q[®] or equivalent).
- 4. Add **120 μL** of **Chip Storage Buffer** ^O to the active wells.
- 5. Place the chip back into the LabChip GX Touch/GXII Touch. Ensure a Buffer Tube with **750 µL Chip Storage Buffer** \bigcirc is in the buffer slot.
- 6. Touch the **Wash** button on the Home screen.
- 7. Touch the **Wash** button on the Wash screen.
- 8. When the chip wash is complete, remove the chip from the instrument and place the chip into the chip storage container.
- 9. Add an additional **50 \muL** of **Chip Storage Buffer** \bigcirc to well **1**.
- 10. Cover the wells with Parafilm[®] to prevent evaporation and store at 2-8°C. Allowing chip wells to dry may lead to changes in chip performance. If using the chip again within 24 hours, the chip can be stored at room temperature.



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Assay Specifications

The LabChip ssDNA 7K Assay is for use with LabChip GXII Touch instruments. LabChip GXII Touch instruments are for research use only and not for use in diagnostic procedures.

Sensitivity ¹	500 pg/μL total fragments 19 pg/μL 1.1 kb fragment 89 pg/μL 5.1 kb fragment
Quantitation Reproducibility	20% CV
Quantitation Accuracy	± 30% (for ladder as sample)
Size Range	Up to 7200 nucleotides
Sizing reproducibility	CV < 15%
Maximum Salt	10 mM Tris
Run Time	100 seconds per sample (About 3 hours for 96 samples)
Compatible Plate Types	384-well, 96-well plate
Chip Lifetime ²	HT: 2000 samples 24: 750 samples
Samples per Chip Prep	Up to 96 samples per HT chip prep Up to 48 samples per LT chip prep
Chip Preps per Reagent Kit	5 HT chip preps or 10 LT chip preps

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For the complete ssDNA 7K Assay User Guide, go to: http://www.Revvity.com/

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¹ Estimated by using ssDNA 7K Ladder as sample diluted in TE buffer.

² Estimated from RNA Pico Assay Reagent (P/N CLS960012).