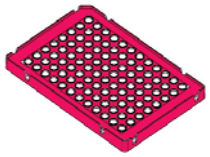
ProfilerPro Glycan Profiling Quick Guide
LabChip® GXII Touch

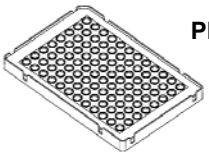
Sample Preparation

Denature
1. Thaw and spin Denaturing Plate at 1200g for 1 minute.
2. Carefully remove plate seal.
3. Add 8µL of sample (monoclonal antibody) with concentration range of 1.25mg/mL to 7.5mg/mL (10µg to 60µg total protein) to Denaturing Plate. Mix by pipetting up and down or with a plate shaker.
4. Seal plate carefully with an adhesive plate seal.
5. Spin plate at 1200g for 1 minute.
6. Incubate for 10 minutes at 70C using a PCR machine or heat block.



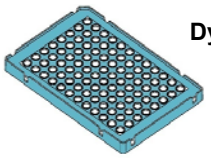
Denaturing Plate

Digestion
1. Thaw PNGase F Plate.
2. Spin both PNGase F Plate and Denaturing Plate at 1200g for 1 minute.
3. Carefully remove plate seals.
4. Transfer all denatured sample to PNGase F Plate. Mix by pipetting up and down or with a plate shaker.
5. Seal plate carefully with an adhesive plate seal.
6. Spin PNGase F Plate at 1200g for 1 minute.
7. Incubate for 1 hour at 37C using a PCR machine or heat block.



PNGase F Plate

Labeling
1. Thaw Dye Plate.
2. Spin both Dye Plate and PNGase F Plate at 1200g for 1 minute.
3. Carefully remove plate seals.
4. Transfer 8µL of Digested sample to the Dye Plate. Mix by pipetting up and down or with a plate shaker.
5. Spin Dye Plate at 1200g for 1 minute.
6. Incubate the unsealed plate for 2 hours at 55C, or until dry using a PCR machine (lid open) or heat block.



Dye Plate

Reconstitution
1. Add 100µL of molecular grade water to dried samples.
2. Seal plate carefully with an adhesive plate seal.
3. Mix samples on a plate shaker at maximum speed for at least 1 minute.
4. Spin plate at 1200g for 1 minute.
5. Carefully remove plate seal.
6. Run plate on LabChip GXII Touch.

LabChip GXII Touch

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PN CLS140171, Rev. B

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Chip Preparation Procedures

Note: Allow the chip and all reagent kit components to equilibrate to room temperature for about 20-30 minutes before use.

Preparing the Buffer Tube

1. Add 750µL of molecular grade water to the 0.75mL Buffer Tube.
2. Insert the Buffer Tube into the buffer slot on the LabChip GXII Touch instrument.


Buffer Tube

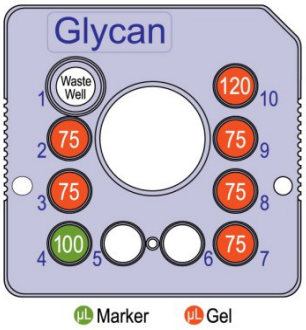
Preparing the Ladder Tube

1. Add 145µL of Ladder Diluent to one of the Ladder tubes.
2. Vortex at highest speed for about 30 seconds and spin down.
3. Transfer 120µL of prepared ladder to the 0.2mL Ladder Tube.
4. Insert the Ladder Tube into the ladder slot on the LabChip GXII Touch instrument.


Ladder Tube

Preparing the Chip

1. Remove reagents from all wells of the chip using a vacuum.
2. Rinse and aspirate all wells with molecular grade water. Repeat, and be careful to aspirate all the water in the wells and any water that may have spilled onto the outside of the chip.
3. Add 75µL of Gel Matrix to chip wells 2, 3, 7, 8 and 9 and 120µL in well 10 using a Reverse Pipetting Technique.
4. Prepare marker solution by adding 125µL of Marker Diluent to one of the Marker tubes. Vortex at highest speed for about 30 seconds and spin down. Transfer 100µL of prepared marker solution to chip well 4. (*Note: Prepare marker solution just before loading the chip in the LabChip GXII Touch and starting the assay. Do not prepare marker solution in advance as the marker signal degrades over time.*)
5. Place the chip in the LabChip GXII Touch instrument to begin the assay.



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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 **120 µL** of biology-grade water to active wells.
4. Make sure to cover all wells with Parafilm® and store at RT.

Assay Specifications

Amount of Sample Required	8 µL with concentration range of 1.25-7.5 mg/mL (10- 60 µg of MAb total)
Reproducibility of %Area	CV < 10% for a peak >= 2.5% of total glycan
Limit of Detection	1 ng of G0f standard (smallest amount of labeled G0f standard that can be detected)
Deglycosylation	>95% of all N-linked glycans will be released from MAb
Usable Size Range	Appropriate for neutral glycans found on MAbs, some charged glycans may run outside of our usable range
Sizing Reproducibility	CV < 2.5%
Sample Prep, Chip Prep, and Analysis Time	< 8 hours for a 96-well plate
Number of Samples per Chip Prep	Up to 192 samples before chip needs to be reloaded with fresh gel
For Research Use Only	

For complete ProfilerPro Glycan Profiling Assay User Guide, go to:

http://www.perkinelmer.com/labchipsystems

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