

Agilent HS Large Fragment 50 kb Kit

5200 Fragment Analyzer System

Quick Guide

For Research Use Only.

Not for use in diagnostic procedures.

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Kit Specifications

Specifications	Description
DNA sizing range	75 bp to 48,500 bp
DNA sizing accuracy ¹	± 15% or better
DNA fragment concentration range ²	5 pg/μL to 600 pg/μL input DNA (optimal concentration 500 to 600 pg/μL)
DNA smear concentration range ²	50 pg/μL to 5 ng/μL input DNA (optimal concentration 1 ng/μL)
DNA quantification accuracy ²	± 25%
DNA quantification precision ²	20% CV

¹ Using DNA fragment standards at 600 pg/μL and DNA smears at 1 ng/μL prepared in 1x TE Buffer.

² Using DNA fragment standards and DNA smears prepared in 1x TE Buffer.

Kit Components

Agilent HS Large Fragment 50 kb kit (DNF-464)

Part Number	Name
DNF-220	Large Fragment Separation Gel
DNF-600-U030	Intercalating Dye
DNF-355	5x 930 dsDNA Inlet Buffer (Dilute to 1x)
DNF-475	5x Capillary Conditioning Solution (Dilute to 1x)
DNF-497	0.25x TE Rinse Buffer
DNF-381	HS Large Fragment Diluent Marker
DNF-365	HS Extended Large Frag DNA Ladder
DNF-300	BF-25 Blank Solution
DNF-495	Dilution Buffer 1x TE
GP-440-0100	Capillary Storage Solution (sold separately)

Analysis Protocol

Gel preparation

Prepare gel/dye mixture for 5200 Fragment Analyzer system:

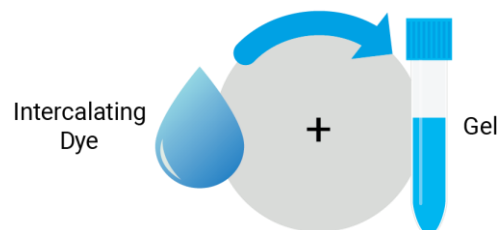
# of samples to be analyzed ¹	Volume of Intercalating Dye	Volume of gel
12	1.0 µL	10 mL
24	1.5 µL	15 mL
36	2.0 µL	20 mL
48	2.5 µL	25 mL
96	4.5 µL	45 mL

¹ Typically one sample well per separation is dedicated to the ladder.

Instrument and sample preparation

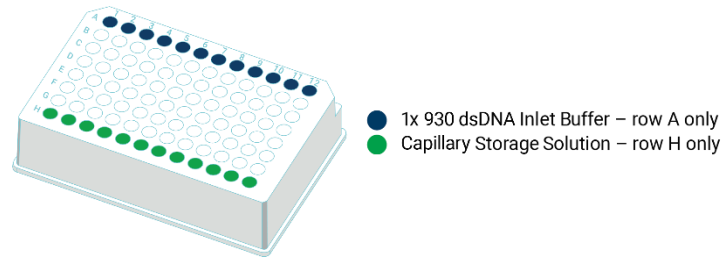
- 1 Mix fresh gel and dye. Refill 1x Capillary Conditioning Solution as needed.

1.0 µL dye / 10 mL gel

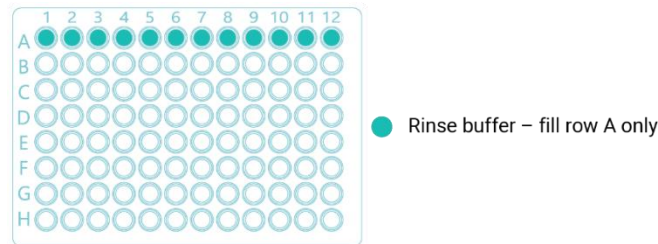


- 2 Place a fresh 1x 930 dsDNA Inlet Buffer tray on the 5200 Fragment Analyzer system. Fill row A (1.0 mL/well). Replace 1x 930 dsDNA Inlet Buffer daily.

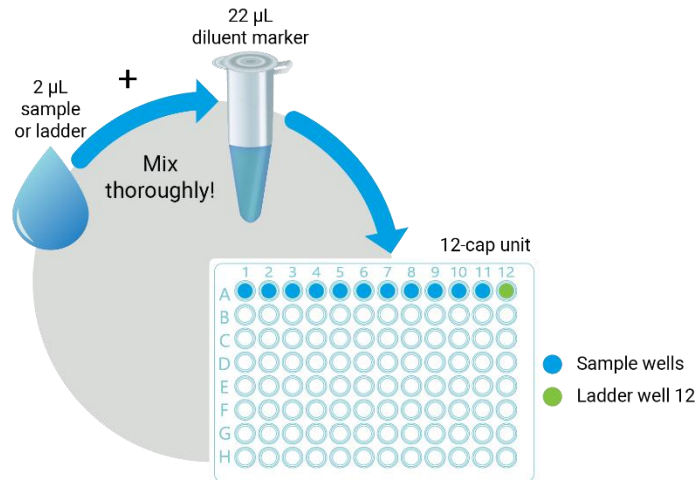
- Place Capillary Storage Solution in row H (1.0 mL/well). Replace Capillary Storage Solution every 2-4 weeks.



- Place 0.25x TE Rinse Buffer plate in marker drawer location. Fill row A (200 μ L/well).



- Dilute samples to optimal concentration range using Dilution Buffer 1x TE. Mix samples or HS Extended Large Frag DNA Ladder with HS Large Fragment Diluent Marker in sample plate. Add 24 μ L of BF-25 Blank Solution to unused wells.




WARNING

Working with Chemicals

The handling of reagents and chemicals might hold health risks.

- Refer to product material safety datasheets for further chemical and biological safety information.
- Follow the appropriate safety procedures such as wearing goggles, safety gloves and protective clothing

Fragment Analyzer software operating procedure

- 1 Select **Tray** and **Row** to run for 12-Cap.
- 2 Enter **Sample ID** and **Tray ID** (optional).
- 3 Select **Add to Queue**, select the **DNF-464-(22 or 33) HS Large Fragment 50Kb** method from the Dropdown menu.
- 4 Enter **Tray Name**, **Folder Prefix**, and **Notes** (optional).
- 5 Select **OK** to add method to the queue.
- 6 Select  to start the separation.

NOTE

Please refer to the Kit Guide for additional details.

Technical Support and Further Information

For technical support, please visit www.agilent.com.

It offers useful information, support and current developments about the products and technology.

www.agilent.com

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