MiSeq FGx™ Reagent Kit Reference Guide

FOR RESEARCH, FORENSIC, OR PATERNITY USE ONLY

January 2015

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Part # 15055483 Rev B TG-143-9001DOC January 2015

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Revision History

Part #	Revision	Date	Description of Change
15055483	В	January 2015	Updated guide name from MiSeq ForenSeq Sequencing Kit Reagent Prep Guide to MiSeq FGx Reagent Kit Reference Guide. Updated the reagent kit name from MiSeq ForenSeq Sequencing Kit to MiSeq FGx Reagent Kit. Updated the catalog number of the reagent kit from MS-201-1001 to TG-143-1001. Updated the guide catalog number from MS-201-9001DOC to TG-143-9001DOC. Reorganized the description of reagent cartridge contents.
15055483	A	September 2014	Initial release.

MiSeq FGx Reagent Kits

To perform a run on the MiSeq FGx^{TM} , you need one single-use MiSeq FGx Reagent Kit. Each MiSeq FGx Reagent Kit includes a kit-specific flow cell type and reagents required for performing one run.

Kit Type	Flow Cell Type	Available Kit Sizes
MiSeq FGx Reagent Kit	Includes a MiSeq FGx flow cell	380 cycles

The reagent cartridge provided in the kit uses radio-frequency identification (RFID) for accurate consumable tracking and compatibility.

Reagent Kit Size

Reagent kit size is based on the number of cycles that can be performed with one kit. The available size is 380 cycles.

Kit Size	Number of Cycles
380-Cycle Kit	Provides reagents for up to 398 cycles of sequencing, which is sufficient for 351 + 31 cycles plus two eight-cycle index reads.

MiSeq MiSeq FGx Reagent Kit

Consumable	Catalog #
MiSeq FGx Reagent Kit, 380 Cycles	TG-143-1001

Kit Contents and Storage Requirements

All MiSeq FGx reagent kits are packaged in two boxes. As soon as you receive your kit, promptly store the kit components at the indicated temperature to ensure proper performance.

Box 1: Store at -25°C to -15°C

Quantity	Component	Storage	Description
1	Reagent Cartridge	-25°C to -15°C	Single-use prefilled cartridge
1	HT1	-25°C to -15°C	5 ml tube, Hybridization Buffer

Box 2: Store at 2°C to 8°C

Quantity	Component	Storage	Description
1	PR2 Bottle	2°C to 8°C	500 ml bottle, Incorporation Buffer
1	MiSeq FGx Flow Cell	2°C to 8°C	Single-use PE flow cell

Flow Cell Overview

The flow cell is immersed in storage buffer in a flow cell container.

The flow cell is a single-lane glass-based substrate on which clusters are generated and the sequencing reaction is performed. During the sequencing run, the single lane of the flow cell is imaged in small areas called tiles.

Flow Cell Tiles

Flow Cell	Tiles	Imaging Surface	Total Tiles Imaged
FGx Flow Cell	19 tiles	Тор	19 tiles total

Software Requirements

You must use a compatible version of MiSeq FGx Control Software to read the RFID of the flow cell.

Kit Name	Required Control Software Version
MiSeq FGx Reagent Kit	MiSeq FGx Control Software v1 or later

Reagent Cartridge Overview

The MiSeq FGx reagent cartridge is a single-use consumable consisting of foil-sealed reservoirs prefilled with clustering and sequencing reagents sufficient for sequencing one flow cell. Each reservoir on the cartridge is numbered.

Before beginning the sequencing run, sample libraries are loaded onto the cartridge in position 17, which is labeled **Load Sample**.

Figure 1 Reagent Cartridge

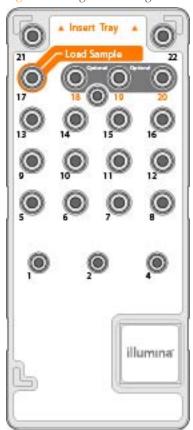


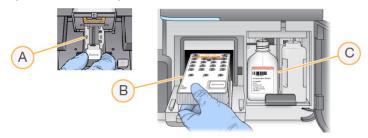
 Table 1
 Reagent Cartridge Reservoirs

Position	Reagent Name	Description
8	LDR	Denaturation Reagent (contains formamide)
17	Reserved	Load Sample (Reserved for sample libraries)
18	Reserved	Reserved for custom Read 1 primer [Optional]
19	Reserved	Reserved for custom Index Read primer [Optional]
20	Reserved	Reserved for custom Read 2 primer [Optional]

Prepare Reagents

The MiSeq FGx Reagent Kit contains a tube of HT1, the reagent cartridge, and a bottle of PR2. For best results, carefully follow the reagent preparation instructions.

Preparation Requirements



- A Flow Cell—Must be thoroughly cleaned before use. See the MiSeq FGx Instrument Reference Guide (part # 15050524).
- **B** Reagent Cartridge—Must be thoroughly thawed and inspected before use.
- **C** PR2 Bottle—No preparation required. Use directly from 2°C to 8°C storage.
- **D** HT1 (not shown)—Must be thawed and chilled before using to denature and dilute libraries. See *ForenSeq DNA Signature Prep Guide* (part # 15049528).

Prepare HT1

The tube of HT1 (Hybridization Buffer) is used to dilute denatured libraries before loading libraries onto the reagent cartridge for sequencing.

- 1 Remove the tube of HT1 (Hybridization Buffer) from -25°C to -15°C storage and set aside at room temperature to thaw.
- When thawed, store at 2°C to 8°C until you are ready to dilute denatured libraries.

Thaw Reagent Cartridge

The following instructions describe how to thaw the reagent cartridge using a room temperature water bath.



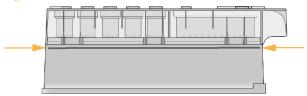
NOTE

Alternatively, you can thaw reagents overnight in 2°C to 8°C storage. Reagents are stable up to one week when stored at this temperature.

1 Remove the reagent cartridge from -25°C to -15°C storage.

2 Place the reagent cartridge in a water bath containing enough room temperature deionized water to submerge the base of the reagent cartridge. Do not allow the water to exceed the maximum water line printed on the reagent cartridge.

Figure 2 Maximum Water Line



- 3 Allow the reagent cartridge to thaw in the room temperature water bath for approximately 60–90 minutes or until it is thawed completely.
- 4 Remove the cartridge from the water bath and gently tap it on the bench to dislodge water from the base of the cartridge. Dry the base of the cartridge.

Inspect the Reagent Cartridge

- 1 Invert the reagent cartridge ten times to mix the thawed reagents, and then visually inspect that all positions are thawed.
- 2 Visually inspect the reagents in positions 1, 2, and 4 to make sure that they are fully mixed and free of precipitates.
- 3 Gently tap the cartridge on the bench to reduce air bubbles in the reagents.



NOTE

The MiSeq FGx sipper tubes go to the bottom of each reservoir to aspirate the reagents, so it is important that the reservoirs are free of air bubbles.

4 Place the reagent cartridge on ice for up to 6 hours, or set aside at 2°C to 8°C until ready to set up the run. For best results, proceed directly to loading the sample and setting up the run.



WARNING

This set of reagents contains formamide, an aliphatic amide that is a probable reproductive toxin. Personal injury can occur through inhalation, ingestion, skin contact, and eye contact. Dispose of containers and any unused contents in accordance with the governmental safety standards for your region. For more information, see the SDS for this kit at support.illumina.com/sds.html.

Next Steps

After you have properly thawed the reagent cartridge, you are ready to proceed to the following steps described in the *MiSeq FGx Instrument Reference Guide* (part # 15050524):

- Denature and dilute your libraries for cluster generation and sequencing. See the *ForenSeq DNA Signature Prep Guide (part # 15049528)*.
- 2 Load your libraries onto the reagent cartridge in the reservoir labeled Load Sample.
- 3 Using the MiSeq Forensic Control Software interface, follow the run setup steps to load the flow cell and reagents, and then start the run.

Visit the Forensics support pages on the Illumina website for access to documentation, software downloads, and frequently asked questions. To view a comprehensive list of MiSeq training courses, go to www.illumina.com/training/miseq.

Notes

Technical Assistance

For technical assistance, contact Illumina Technical Support.

Table 2 Illumina General Contact Information

Website	www.illumina.com
Email	techsupport@illumina.com

Table 3 Illumina Customer Support Telephone Numbers

Region	Contact Number	Region	Contact Number
North America	1.800.809.4566	Italy	800.874909
Australia	1.800.775.688	Netherlands	0800.0223859
Austria	0800.296575	New Zealand	0800.451.650
Belgium	0800.81102	Norway	800.16836
Denmark	80882346	Spain	900.812168
Finland	0800.918363	Sweden	020790181
France	0800.911850	Switzerland	0800.563118
Germany	0800.180.8994	United Kingdom	0800.917.0041
Ireland	1.800.812949	Other countries	+44.1799.534000

Safety Data Sheets

Safety data sheets (SDSs) are available on the Illumina website at support.illumina.com/sds.html.

Product Documentation

Product documentation in PDF is available for download from the Illumina website. Go to support.illumina.com, select a product, then click **Documentation & Literature**.



Part # 15055483 Rev E



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