

MiSeq[®] Reagent Kit v2

Reagent Preparation Guide

FOR RESEARCH USE ONLY

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Introduction

This guide explains how to prepare the MiSeq® Reagent Kit for a run on the MiSeq System, and includes an overview of kit contents, storage requirements, and instructions for thawing the reagent cartridge.

See the *MiSeq System User Guide*, Part # 15027617 for the following instructions:

- ▶ Preparing your libraries for cluster generation and sequencing
- ▶ Loading your sample onto the reagent cartridge
- ▶ Setting up a run on the MiSeq using the software interface
- ▶ Operating and maintaining the MiSeq

MiSeq Reagent Kit v2

To perform a run on the MiSeq, you need one MiSeq Reagent Kit. One kit contains the reagents required for sequencing one flow cell.

- ▶ The MiSeq Reagent Kit is packaged in two boxes. As soon as you receive your kit, promptly store the kit components at the indicated temperature to ensure proper performance.
- ▶ Each reagent cartridge uses radio-frequency identification (RFID) for accurate consumable tracking.
- ▶ **500 Cycle Kit**—Provides kitted reagents for up to 526 cycles of sequencing, which is sufficient for up to a 251-cycle paired-end run (2 x 251), plus two eight-cycle index reads.
- ▶ **300 Cycle Kit**—Provides kitted reagents for up to 326 cycles of sequencing, which is sufficient for up to a 151-cycle paired-end run (2 x 151), plus two eight-cycle index reads.
- ▶ **50 Cycle Kit**—Provides kitted reagents for up to 76 cycles of sequencing, which is sufficient for up to a 26-cycle paired-end run (2 x 26), or a 51-cycle single-read run, plus two eight-cycle index reads.

Table 1 MiSeq Reagent Kit v2

Consumable	Catalog #
MiSeq Reagent Kit v2, 500 Cycles (PE)	Catalog # MS-102-2003
MiSeq Reagent Kit v2, 300 Cycles (PE)	Catalog # MS-102-2002
MiSeq Reagent Kit v2, 50 Cycles (PE)	Catalog # MS-102-2001

Reagent Cartridge Contents

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

Figure 1 Reagent Cartridge

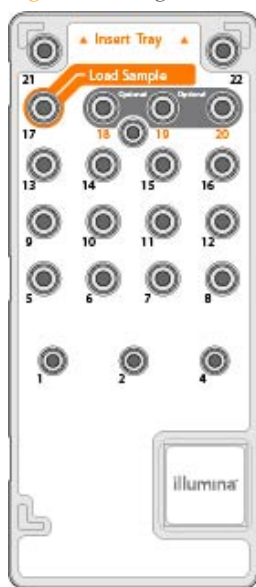


Table 2 MiSeq Reagent Kit v2, Cartridge Contents

Position	Reagent Name	Description
1	IMS	Incorporation Mix
2	SRE	Scan Mix
4	CMS	Cleavage Mix
5	AMS1	Amplification Mix, Read 1
6	AMS2	Amplification Mix, Read 2
7	LPM	Linearization Premix
8	LDR	Formamide
9	LMX1	Linearization Mix
10	LMX2	Read 2 Linearization Mix
11	RMF	Resynthesis Mix
12	HP10	Read 1 Primer Mix
13	HP12	Index Primer Mix
14	HP11	Read 2 Primer Mix
15	PW1	Laboratory-grade water
16	PW1	Laboratory-grade water

Position	Reagent Name	Description
17	Sample	Your sample libraries
18	Optional	Optional use for custom Read 1 primer
19	Optional	Optional use for custom Index Read primer
20	Optional	Optional use for custom Read 2 primer
21	PW1	Laboratory-grade water
22	Empty	Empty



NOTE

For more information about using custom primers on the MiSeq reagent cartridge, see the *MiSeq System User Guide*, Part # 15027617 Rev. C (or later).

Preparing Reagents

The following instructions describe how to thaw reagents using a room temperature water bath. This method requires approximately one hour.

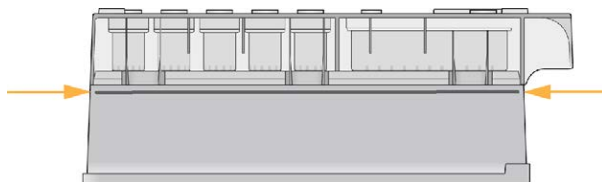


NOTE

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

- 1 Remove the tube of HT1 (Hybridization Buffer) from -15° to -25°C storage and set aside at room temperature to thaw. When thawed, store at 2° to 8°C until you are ready to dilute denatured libraries. (This step is not required for TruSeq Amplicon libraries.)
For more information, see the *MiSeq System User Guide*, Part # 15027617.
- 2 Remove the reagent cartridge from -15° to -25°C storage.
- 3 Place the reagent cartridge in a water bath containing enough room temperature deionized water to submerge the base of the reagent cartridge up to the water line printed on the reagent cartridge. Do not allow the water to exceed the maximum water line.

Figure 2 Maximum Water Line



- 4 Allow the reagent cartridge to thaw in the room temperature water bath for approximately one hour or until completely thawed.
- 5 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. Make sure that no water has splashed on the top of the reagent 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 reagent marked IMS (Position 1) to make sure that it is fully mixed and free of precipitates.
- 3 Gently tap the cartridge on the bench to reduce air bubbles in the reagents.



NOTE

The MiSeq 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 or set aside at 2° to 8°C until you are ready to set up your run. For best results, proceed directly to loading your 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 MSDS for this kit, at <http://www.illumina.com/msds>.

What's Next

After you have properly thawed the reagent cartridge, you are ready to proceed to the following steps described in the *MiSeq System User Guide*, Part # 15027617:

- 1 Prepare your libraries for cluster generation and sequencing (unless you are sequencing TruSeq Amplicon libraries).
- 2 Load your libraries onto the reagent cartridge in the reservoir labeled **Load Samples**.
- 3 Using the MiSeq Control Software (MCS) interface, follow the run setup steps to load the flow cell and reagents, and then start the run.

Visit the MiSeq 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 Customer Support.

Table 3 Illumina General Contact Information

Illumina Website	http://www.illumina.com
Email	techsupport@illumina.com

Table 4 Illumina Customer Support Telephone Numbers

Region	Contact Number	Region	Contact Number
North America	1.800.809.4566	Italy	800.874909
Austria	0800.296575	Netherlands	0800.0223859
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

MSDSs

Material safety data sheets (MSDSs) are available on the Illumina website at <http://www.illumina.com/msds>.

Product Documentation

You can obtain PDFs of additional product documentation from the Illumina website. Go to <http://www.illumina.com/support> and select a product. To download documentation, you will be asked to log in to MyIllumina. After you log in, you can view or save the PDF. To register for a MyIllumina account, please visit <https://my.illumina.com/Account/Register>.

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