MagMAX™ Pathogen RNA/DNA Kit

For whole blood samples

Catalog Numbers 4462359

Pub. No. 4466366 **Rev.** B

Note: For safety and biohazard guidelines, see the "Safety" appendix in the *MagMAX*™ *Pathogen RNA/DNA Kit User Guide* (Pub. No. 4463379). Read the Safety Data Sheets (SDSs) and follow the handling instructions. Wear appropriate protective eyewear, clothing, and gloves.

This document provides procedures for purifying nucleic acid on the KingFisher[™] Flex Magnetic Particle Processor or MagMAX[™] Express-96 Deep Well Magnetic Particle Processor (MME-96 processor). For manual purification procedures, see the $MagMAX^{™}$ Pathogen RNA/DNA Kit User Guide (Pub. No. 4463379).

Before each use of the kit

When preparing the reagents:

- Calculate the total volume that is required for each component: volume for 1 reaction × the total number of reactions.
- Include 10% excess volume to account for pipetting errors.

Prepare Lysis/Binding solution

1. Combine the components listed below in the order indicated.

Component	Volume
Lysis/Binding Solution Concentrate	200 μL
Carrier RNA (µg/µL)	2 μL
(Optional) ^{1]} Xeno™ RNA or Xeno™ DNA	2 μL
100% Isopropanol	200 μL
Total volume for 1 reaction	404 μL

 $^{^{[1]}}$ Add nuclease-free water if not adding Xeno $^{\text{\tiny{M}}}$ RNA or DNA.

2. Mix well by vortexing.

Prepare the Bead Mix

- 1. Vortex the Nucleic Acid Binding Beads well to ensure that the beads are fully resuspended.
- 2. On ice, combine the components that are listed below.

Component	Volume
Nucleic Acid Binding Beads	10 μL
Lysis ENHANCER	10 μL
Total volume for 1 reaction	20 μL

3. Mix well by vortexing.

Store on ice for up to 4 hours.



Prepare the whole blood samples

You can use the MagMAX[™] Pathogen RNA/DNA Kit to purify nucleic acid from 100 µL of whole blood samples.

- 1. Mix blood by inverting the blood samples at least 5 times.
- 2. Pulse spin for 1 second to collect the blood at the tube bottom.

Proceed to "Process samples on the KingFisher™ Flex/MagMAX™ Express-96 Deep Well Magnetic Particle Processor" on page 2.

Process samples on the KingFisher™ Flex/MagMAX™ Express-96 Deep Well Magnetic Particle Processor

For whole blood samples, we determined that 100 μ L is the optimal input volume. Because MME-96 Deep Well Plates are required for the volume that is used with this workflow, we have not included MagMAX Express Magnetic Particle Processor (MME-24) or manual purification procedures.

Note: It is critical that you prepare the sample plate last to reduce the time that the Bead Mix, sample, and Lysis/Binding Solution are unmixed. To ensure best results, after preparing the sample plate, immediately load it onto the processor for purification.

Complete the steps below at room temperature and in the order indicated.

1. Select the one of the following scripts.

Script	Instrument
4462359_DW_HV	 KingFisher[™] Flex Magnetic Particle Processor MagMAX[™] Express-96 Deep Well Magnetic Particle Processor
MagMAX [™] _Pathogen_High_Vol	 KingFisher[™] Flex Magnetic Particle Processor

- 2. Prepare the tip comb plate: Place an MME-96 Deep Well Tip Comb in one MME-96 Standard Plate.
- 3. Prepare the Wash Solution 1 plates, by adding 300 µL of prepared Wash Solution 1 to two MME-96 Deep Well Plates.
- 4. Prepare the Wash Solution 2 plates, by adding 450 µL of prepared Wash Solution 2 to two MME-96 Deep Well Plates.
- 5. Prepare the elution plate, by adding 90 µL of Elution Buffer to one MME-96 Standard Plate (not the tip comb plate).
- **6.** Prepare the sample plate:

Note: After you start preparing the sample plate, do not pause until all plates are loaded onto the MME-96 processor and you have started the script.

- a. Add 20 μ L of prepared Bead Mix to one MME-96 Deep Well Plate.
- **b.** Add 100 µL of prepared sample to the plate.
- c. Using a plate shaker, shake at moderate speed for 1 minute (see the MagMAX™ Pathogen RNA/DNA Kit User Guide, Part. No 4463379, for shaker settings).

Note: To avoid cross-contamination, do not pipet up and down.

d. Add 400 µL of prepared Lysis/Binding Solution to the plate.

- **e.** Using a plate shaker, shake at moderate speed for 1 minute.
- 7. Immediately start the MME-96 processor script, then load the plates onto the processor as directed.

 Table 1
 Processing plate setup : Whole blood (deep-well head configuration)

Plate ID	Plate position	Plate type	Reagent	Volume per well
Sample plate	1	MME-96 Deep Well Plate	Bead Mix	20 μL
			Sample	100 μL
			Lysis/Binding Solution	400 μL
First Wash 1	2	MME-96 Deep Well Plate	Wash Solution 1	300 μL
Second Wash 1	3	MME-96 Deep Well Plate	Wash Solution 1	300 μL
First Wash 2	4	MME-96 Deep Well Plate	Wash Solution 2	450 μL
Second Wash 2	5	MME-96 Deep Well Plate	Wash Solution 2	450 μL
Elution	6	MME-96 Standard Plate	Elution Buffer	90 μL
Tip comb plate	7	MME-96 Standard Plate	MME-96 Deep Well Tip Comb in plate	

STOPPING POINT Store the purified nucleic acid on ice for immediate use, at -20°C for up to 1 month, or at -80°C for long-term storage.

Limited product warranty

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Revision	Date	Description
В	12 January 2018	Added instructions for KingFisher™ Instruments
		Updated to the current document template, with associated updates to the warranty, trademarks, and logos.
A	June 2011	New document

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