MAGBIO

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HighPrep Viral-Bacterial DNA & RNA Kit - DX

Isolation of Viral and Bacterial Nucleic Acids

Catalog Nos. HVB-DR5E, HVB-DR96E, HVB-DR96x4E
Manual Revision 0
WI-72-118

- Isolation of viral and bacterial total nucleic acids from different types of viral transport media, plasma, swabs, saliva, urine, whole blood, and other bodily fluids
- Magnetic bead-based chemistry

Instructions For Use

Contents

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CE IVD MHRA

For *in vitro* diagnostic procedures.

Information in this document is subject to change without notice.

Product Description

The HighPrep Viral-Bacterial DNA & RNA Kit - DX is designed for rapid and reliable isolation of total nucleic acids from viruses and bacteria in whole blood, serum, plasma, swabs, saliva, urine, and other bodily fluids. The kit extracts high quality DNA and RNA that is suitable for direct use in most downstream applications such as amplification and enzymatic reactions. It can be adapted to most major liquid handling workstations in the market.

This product is intended to be used by qualified and trained laboratory professionals only.

Process

Samples are lysed in a specially formulated lysis buffer. Nucleic acids are bound to the surface of MAG-S1 Particles under special conditions. Proteins and cellular debris are efficiently washed with a few wash steps. Pure RNA and DNA are then eluted in Nuclease-Free Water or low ionic strength buffer. Purified RNA or DNA can be directly used in downstream applications without the need for further purification.

Kit Contents and Storage

HighPrep Viral-Bacterial DNA & RNA Kit - DX Catalog No.*	**HVB-DR5E**	**HVB-DR96E**	**HVB-DR96x4E**	**Storage**
Number of Preps	5	96	384	
Lysis Buffer	1.6 mL	30 mL	120 mL	15-25°C
Wash Buffer[1]	1.6 mL	30 mL	120 mL	15-25°C
Nuclease-Free Water	5 mL	35 mL	140 mL	15-25°C
Pro K Solution[2]	110 µL	2.2 mL	8.8 mL	2-8°C
NBE[3]	50 µL	2 mL	8 mL	2-8°C
MAG-S1 Particles[4]	55 µL	1.1 mL	4.4 mL	2-8°C
LES I[5]	255 µL	5 mL	20 mL	2-8°C

[1]Ethanol must be added prior to use. See preparation of reagents section.
*Once opened, reagents are usable until the expiration date on the product label. Be sure to close the lid firmly before storing reagents for later use.

Shipping and Storage

- [2]Pro K Solution comes in a ready to use solution. Ships at room temperature. Store at 2-8°C.
- [3]NBE ships at room temperature. Store at 2-8°C.
- [4]MAG-S1 Particles ship at room temperature. Store at 2-8°C.
- [5]LES I ships at room temperature. Store at 2-8°C.

Safety Information

Any consumables, including plates, tubes, etc., used to process samples with infectious or microbial hazards should be disposed of in an appropriate biohazard waste bin. When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate safety data sheets (SDSs). The SDS can be downloaded from the "Product Documents" tab when viewing this product at www.magbiogenomics.com.

Preparation of Reagents

Prepare the following components for each kit before use:

Catalog No.	Component	Add 100% Ethanol*	Storage
HVB-DR5E	Wash Buffer	1 mL	Room Temp 15-25°C

Catalog No.	Component	Add 100% Ethanol*	Storage
HVB-DR96E	Wash Buffer	20 mL	Room Temp 15-25°C

Catalog No.	Component	Add 100% Ethanol*	Storage
HVB-DR96x4E	Wash Buffer	80 mL	Room Temp 15-25°C

A - VIRAL NUCLEIC ACID ISOLATION PROTOCOLS

Protocol: 200 µL sample volume (96 well plate format/single tube)

Equipment and Reagents to Be Supplied by the User

- ☐ Ethanol (80%)
- ☐ Isopropanol (100%)
- ☐ Magnetic separation device for 96 well plate/1.5 - 2 mL magnetic separation device
- ☐ 96 well microplates (U or V bottom) or 1.5 - 2 mL microcentrifuge tubes

Things to do Before Starting

☐ Prepare all reagents according to the instructions on page 2.

Protocol

⚠ Bring the **MAG-S1 Particles** to room temperature for at least 30 minutes before use.

1. Gently swirl the **LES I** container, then pipette 50 µL to each well/tube.
2. Add 200 µL of the sample to each well/tube. Pipette mix 15 times.

   ⚠ *If the sample volume is less than 200 µL, bring the volume up to 200 µL with **Nuclease-Free Water**.*
3. Incubate for 10 minutes at 37°C.
4. Add 240 µL of **Lysis Buffer** and 10 µL of **Pro K Solution**. Mix very well.
5. Incubate at 56-60°C for 5 minutes. May use a thermoshaker. If there is none in the lab, make sure to shake the samples once or twice during incubation.
6. Let the samples cool to room temperature and add 8 µL of **NBE**, 280 µL of 100% Isopropanol, and 10 µL of **MAG-S1 Particles**. Pipette mix 15 times.

   ⚠ *Shake well to resuspend the **MAG-S1 Particles** before use.*
7. Let the samples sit at room temperature for 5 minutes.
8. Place the sample plate on the magnetic separation device for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.
9. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.
10. Remove the plate from the magnetic separation device and add 400 µL of **Wash Buffer**. Pipette mix 15 times to resuspend the **MAG-S1 Particles** until solution is homogeneous. Then, place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.
11. Remove the plate/tube from the magnetic separation device. Add 500 µL of 80% Ethanol and mix to resuspend the **MAG-S1 Particles**.

    ⚠ *Make sure the solution is homogeneous.*

12. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.

13. Discard the supernatant and repeat steps 11-12 for a 2nd wash.

14. Discard the supernatant and air-dry the beads for 7 minutes.

15. Remove the plate from the magnetic separation device. Add 30-100 µL of **Nuclease-Free Water** to each well/tube and pipette mix 15 times to completely resuspend the **MAG-S1 Particles**.

    ⚠ *Complete resuspension of the **MAG-S1 Particles** is crucial for better yield.*

16. Incubate at 56-60°C for 5 minutes.

17. Place the sample plate back on the magnetic separation device and wait for 3 minutes or until the magnetic beads clear from the solution.

18. Transfer the eluate (cleared supernatant containing DNA or RNA) to a new microplate for storage. Store the DNA at -20°C and RNA at -80°C.

Protocol: 400 µL sample volume (96 well plate format/single tube)

Equipment and Reagents to Be Supplied by the User

- ☐ Ethanol (80%)
- ☐ Isopropanol (100%)
- ☐ Magnetic separation device for 96 well plate/1.5 - 2 mL magnetic separation device
- ☐ 96 well microplates (U or V bottom) or 1.5 - 2 mL microcentrifuge tubes

Things to do Before Starting

☐ Prepare all reagents according to the instructions on page 2.

Protocol

⚠ Bring the **MAG-S1 Particles** to room temperature for at least 30 minutes before use.

1. Gently swirl the **LES I** container, then pipette 100 µL to each well/tube.
2. Add 400 µL of the sample to each well/tube. Pipette mix 15 times.

   ⚠ *If the sample volume is less than 400 µL, bring the volume up to 400 µL with **Nuclease-Free Water**.*
3. Incubate for 10 minutes at 37°C.
4. Add 400 µL of **Lysis Buffer** and 20 µL of **Pro K Solution**. Mix very well.
5. Incubate at 56-60°C for 5 minutes. May use a thermoshaker. If there is none in the lab, make sure to shake the samples once or twice during incubation.
6. Let the samples cool to room temperature and add 12 µL of **NBE**, 400 µL of 100% Isopropanol, and 20 µL of **MAG-S1 Particles.** Pipette mix 15 times.

   ⚠ *Shake well to resuspend the **MAG-S1 Particles** before use.*
7. Let the samples sit at room temperature for 5 minutes.
8. Place the sample plate on the magnetic separation device for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.
9. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.
10. Remove the plate from the magnetic separation device and add 400 µL of **Wash Buffer.** Pipette mix 15 times to resuspend the **MAG-S1 Particles** until the solution is homogeneous. Then, place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.
11. Remove the plate/tube from the magnetic separation device. Add 400 µL of 80% Ethanol and mix to resuspend the **MAG-S1 Particles**.

    ⚠ *Make sure the solution is homogeneous.*
12. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.

13. Discard the supernatant and repeat steps 11-12 for a 2nd wash.

14. Discard the supernatant and air-dry the beads for 7 minutes.

15. Remove the plate from the magnetic separation device. Add 30-100 µL of **Nuclease-Free Water** to each well/tube and pipette mix 15 times to completely re-suspend the **MAG-S1 Particles**.

    ⚠ *Complete resuspension of the **MAG-S1 Particles** is crucial for better yield.*

16. Incubate at 56-60°C for 5 minutes.

17. Place the sample plate back on the magnetic separation device and wait for 3 minutes or until the magnetic beads clear from the solution.

18. Transfer the eluate (cleared supernatant containing DNA or RNA) to a new microplate for storage. Store the DNA at -20°C and RNA at -80°C

B - BACTERIAL NUCLEIC ACID ISOLATION PROTOCOLS

Protocol: Urine 200 µL sample volume (96 well plate format/single tube)

Equipment and Reagents to Be Supplied by the User

- ☐ Ethanol (80%)
- ☐ Isopropanol (100%)
- ☐ Magnetic separation device for 96 well plate/1.5 - 2 mL magnetic separation device
- ☐ 96 well microplates (U or V bottom) or 1.5 - 2 mL microcentrifuge tubes

Things to do Before Starting

☐ Prepare all reagents according to the instructions on page 2.

Protocol

⚠ Bring the **MAG-S1 Particles** to room temperature for at least 30 minutes before use.

1. Gently swirl the **LES I** container, then pipette 50 µL to each well/tube.
2. Add 200 µL of the sample to each well/tube. Pipette mix 15 times.

   ⚠ *If the sample volume is less than 200 µL, bring the volume up to 200 µL with **Nuclease-Free Water**.*
3. Incubate for 10 minutes at 37°C.
4. Add 240 µL of **Lysis Buffer** and 20 µL of **Pro K Solution**. Mix very well.
5. Incubate at 60-65°C for 10 minutes. May use a thermoshaker. If there is none in the lab, make sure to shake the samples once or twice during incubation.
6. Let the samples cool to room temperature and add 8 µL of **NBE**, 280 µL of 100% Isopropanol, and 10 µL of **MAG-S1 Particles**. Pipette mix 15 times.

   ⚠ *Shake well to resuspend the **MAG-S1 Particles** before use.*
7. Let the samples sit at room temperature for 5 minutes.
8. Place the sample plate on the magnetic separation device for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.
9. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.
10. Remove the plate from the magnetic separation device and add 400 µL of **Wash Buffer**. Pipette mix 15 times to resuspend the **MAG-S1 Particles** until the solution is homogeneous. Then, place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.
11. Remove the plate/tube from the magnetic separation device. Add 500 µL of 80% Ethanol and mix to resuspend the **MAG-S1 Particles**.

    ⚠ *Make sure the solution is homogeneous.*

12. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.

13. Discard the supernatant and repeat steps 11-12 for a 2nd wash.

14. Discard the supernatant and air-dry the beads for 7 minutes.

15. Remove the plate from the magnetic separation device. Add 30-100 µL of **Nuclease-Free Water** to each well/tube and pipette mix 15 times to completely resuspend the **MAG-S1 Particles.**

    ⚠ *Complete resuspension of the **MAG-S1 Particles** is crucial for better yield.*

16. Incubate at 56-60°C for 5 minutes.

17. Place the sample plate back on the magnetic separation device and wait for 3 minutes or until the magnetic beads clear from the solution.

18. Transfer the eluate (cleared supernatant containing DNA or RNA) to a new microplate for storage. Store the DNA at -20°C and RNA at -80°C.

Protocol: Urine 400 µL sample volume (96 well plate format/single tube)

Equipment and Reagents to Be Supplied by the User

- ☐ Ethanol (80%)
- ☐ Isopropanol (100%)
- ☐ Magnetic separation device for 96 well plate/1.5 - 2 mL magnetic separation device
- ☐ 96 well microplates (U or V bottom) or 1.5 - 2 mL microcentrifuge tubes

Things to do Before Starting

☐ Prepare all reagents according to the instructions on page 2.

Protocol

⚠ Bring the **MAG-S1 Particles** to room temperature for at least 30 minutes before use.

1. Gently swirl the **LES I** container, then pipette 100 µL to each well/tube.
2. Add 400 µL of the sample to each well/tube. Pipette mix 15 times.

   ⚠ *If the sample volume is less than 400 µL, bring the volume up to 400 µL with **Nuclease-Free Water**.*
3. Incubate for 10 minutes at 37°C.
4. Add 500 µL of **Lysis Buffer** and 20 µL of **Pro K Solution**. Mix very well.
5. Incubate at 60-65°C for 10 minutes. May use a thermoshaker. If there is none in the lab, make sure to shake the samples once or twice during incubation.
6. Let the samples cool to room temperature and add 12 µL of **NBE**, 500 µL of 100% Isopropanol, and 10 µL of **MAG-S1 Particles**. Pipette mix 15 times.

   ⚠ *Shake well to resuspend the **MAG-S1 Particles** before use.*
7. Let the samples sit at room temperature for 5 minutes.
8. Place the sample plate on the magnetic separation device for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.
9. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.
10. Remove the plate from the magnetic separation device and add 400 µL of **Wash Buffer**. Pipette mix 15 times to resuspend the **MAG-S1 Particles** until the solution is homogeneous. Then, place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.
11. Remove the plate/tube from the magnetic separation device. Add 500 µL of 80% Ethanol and mix to resuspend the **MAG-S1 Particles**.

    ⚠ *Make sure the solution is homogeneous.*

12. Place sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.

13. Discard the supernatant and repeat steps 11-12 for a 2nd wash.

14. Discard the supernatant and air-dry the beads for 7 minutes.

15. Remove the plate from the magnetic separation device. Add 30-100 µL of **Nuclease-Free Water** to each well/tube and pipette mix 15 times to completely resuspend the **MAG-S1 Particles.**

    ⚠ *Complete resuspension of the **MAG-S1 Particles** is crucial for better yield.*

16. Incubate at 56-60°C for 5 minutes.

17. Place the sample plate back on the magnetic separation device and wait for 3 minutes or until the magnetic beads clear from the solution.

18. Transfer the eluate (cleared supernatant containing DNA or RNA) to a new microplate for storage. Store the DNA at -20°C and RNA at -80°C.

Protocol: Whole Blood 200 µL sample volume (96 well plate format/single tube)

Equipment and Reagents to Be Supplied by the User

- ☐ Ethanol (80%)
- ☐ Isopropanol (100%)
- ☐ Magnetic separation device for 96 well plate/1.5 - 2 mL magnetic separation device
- ☐ 96 well microplates (U or V bottom) or 1.5 - 2 mL microcentrifuge tubes

Things to do Before Starting

☐ Prepare all reagents according to the instructions on page 2.

Protocol

⚠ Bring the **MAG-S1 Particles** to room temperature for at least 30 minutes before use.

1. Gently swirl the **LES I** container, then pipette 50 µL to each well/tube.
2. Add 200 µL of the sample to each well/tube. Pipette mix 15 times.

   ⚠ *If the sample volume is less than 200 µL, bring the volume up to 200 µL with **Nuclease-Free Water**.*
3. Incubate for 10 minutes at 37°C.
4. Add 240 µL of **Lysis Buffer** and 20 µL of **Pro K Solution**. Mix very well.
5. Incubate at 60-65°C for 10 minutes. May use a thermoshaker. If there is none in the lab, make sure to shake the samples once or twice during incubation.
6. Let the samples cool to room temperature and add 8 µL of **NBE**, 280 µL of 100% Isopropanol, and 10 µL of **MAG-S1 Particles**. Pipette mix 15 times.

   ⚠ *Shake well to resuspend the **MAG-S1 Particles** before use.*
7. Let the samples sit at room temperature for 5 minutes.
8. Place the sample plate on the magnetic separation device for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.
9. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.
10. Remove the plate from the magnetic separation device and add 400 µL of **Wash Buffer**. Pipette mix 15 times to resuspend the **MAG-S1 Particles** until the solution is homogeneous. Then, place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles.** Remove the supernatant.
11. Remove the plate/tube from the magnetic separation device. Add 500 µL of 80% Ethanol and mix to resuspend the **MAG-S1 Particles**.

    ⚠ *Make sure the solution is homogeneous.*

12. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.

13. Discard the supernatant and repeat steps 11-12 for a 2nd wash.

14. Discard the supernatant and air-dry the beads for 7 minutes.

15. Remove the plate from the magnetic separation device. Add 30-100 µL of **Nuclease-Free Water** to each well/tube and pipette mix 15 times to completely resuspend the **MAG-S1 Particles**.

    ⚠ *Complete resuspension of the **MAG-S1 Particles** is crucial for better yield.*

16. Incubate at 56-60°C for 5 minutes.

17. Place the sample plate back on the magnetic separation device and wait for 3 minutes or until the magnetic beads clear from the solution.

18. Transfer the eluate (cleared supernatant containing DNA or RNA) to a new microplate for storage. Store the DNA at -20°C and RNA at -80°C.

Troubleshooting Guide

Please use this guide to troubleshoot any problems that may arise. For further assistance, please contact technical support via:
Phone: US/Canada, +1 301-302-0144. Europe, +49 7250 33 13 403
Email: US/Canada, support@magbiogenomics.com. Europe, info.europe@magbiogenomics.com

Symptoms	Possible Causes	Comments
Low DNA or RNA yield	Incomplete resuspension of the MAG-S1 Particles	Resuspend the MAG-S1 Particles by vortexing vigorously before use
	Loss of MAG-S1 Particles during operation	Avoid disturbing the MAG-S1 Particles during aspiration of the supernatant
	Ethanol is not added into Wash Buffer.	Add absolute 100% Ethanol to Wash Buffer (see page 2 for instructions)
	Inefficient cell lysis.	Double the volume of Pro K Solution and incubate longer
MAG-S1 Particles do not completely clear from the solution	Too short of magnetizing time	Increase collection time on the magnet. Ensure that the solution is completely clear before discarding the supernatant
Problems in downstream applications	Insufficient DNA/RNA in starting material	Use a higher volume of starting material
	Ethanol carry-over	Dry the MAG-S1 Particles completely before elution. Use a fine pipette tip to remove any residual liquid during the drying of the beads
Carryover of MAG-S1 Particles	The eluate contains particles and is not fully clear	Increase the magnetization time. If a small amount of carryover is observed, place the eluted sample on a magnetic separation device and perform an additional 5 minute magnetization

Ordering Information

HighPrep Viral-Bacterial DNA & RNA Kit - DX

Catalog No.	Product	Description	Preps
HVB-DR96E	HighPrep Viral-Bacterial DNA & RNA Kit - DX (96 preps)	Magnetic bead-based kit for isolation of total nucleic acids from viruses and bacteria in different types of viral transport media, plasma, swabs, saliva, urine, whole blood, and other bodily fluids	96
HVB-DR96x4E	HighPrep Viral-Bacterial DNA & RNA Kit - DX (384 preps)		384

Related Products

HighPrep PCR - DX

Catalog No.	Product
AC-60005E	HighPrep PCR - DX (5 mL)
AC-60050E	HighPrep PCR - DX (50 mL)
AC-60250E	HighPrep PCR - DX (250 mL)
AC-60500E	HighPrep PCR - DX (500 mL)

HighPrep RNA Elite - DX

Catalog No.	Product
RC-90005E	HighPrep RNA Elite - DX (5 mL)
RC-90050E	HighPrep RNA Elite - DX (50 mL)
RC-90250E	HighPrep RNA Elite - DX (250 mL)
RC-90500E	HighPrep RNA Elite - DX (500 mL)

Magnetic Separation Devices

Catalog No.	Product
MYMAG-96	Handheld Magnetic Separation Device (96 well microplate format)
MYMAG-96X	Magnetic Separation Device (96 well ring format)
MBMS-12	MagStrip Magnet Stand (1.5 mL x 12)
MBMS-31550	15 mL and 50 mL Magnetic Stand Combo (3 x 15 mL and 3 x 50 mL)

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