HighPrep Viral-Pathogen DNA & RNA Kit - DX

Isolation of Nucleic Acids from Pathogens

Catalog Nos. HPP-DR5E, HPP-DR96E, HPP-DR96x4E
Manual Revision 1
WI-72-67

- Isolation of nucleic acids from viral, bacterial, and yeast pathogens from various sample matrices
- Recovery of DNA/RNA from pathogens using magnetic bead-based chemistry

Instructions For Use

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For *in vitro* diagnostic procedures.

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Product Description

The HighPrep Viral-Pathogen DNA & RNA Kit - DX is designed for rapid and reliable isolation of total nucleic acids from viruses, bacteria, and yeast 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-Pathogen DNA & RNA Kit - DX Catalog No.*	HPP-DR5E	HPP-DR96E	HPP-DR96x4E	Storage
Number of Preps	5	96	384	--
AS Buffer	500 µL	10 mL	40 mL	15-25°C
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]	250 µL	5 mL	20 mL	2-8°C
LES II[6] (Lyophilized)	5 mg	100 mg	400 mg	2-8°C
LES II Suspension Buffer[7]	500 µL	10 mL	40 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 with cold packs (2-8°C). Store at 2-8°C.
- [6]LES II ships at room temperature. Store lyophilized at 2-8°C. Store at -20°C after reconstitution.
- [7]LES II Suspension Buffer 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
HPP-DR5E	Wash Buffer	1 mL	Room Temp 15-25°C

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

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

**Ensure bottle/tube lid Is closed tightly when preparing and storing reagents.*

LES II Solution

Store lyophilized LES II at 2-8°C until the component is ready to be prepared. Centrifuge lyophilized LES II before reconstituting with LES II Suspension Buffer. Pour the LES II Suspension Buffer into the bottle containing the lyophilized LES II. Dissolve the LES II thoroughly, divide it into conveniently sized aliquots, and store at –20°C. Do not freeze–thaw the aliquots of LES II more than 3 times.

Create a 10 mg/mL solution using LES II Suspension Buffer*

Catalog No.	Component	Lyophilized LES II	LES II Suspension Buffer	Storage
HPP-DR5E	LES II	5 mg	500 µL	-20°C
HPP-DR96E	LES II	100 mg	10 mL	-20°C
HPP-DR96x4E	LES II	400 mg	40 mL	-20°C

**Final concentration of LES II should be 10 mg/mL.*

A - VIRAL NUCLEIC ACID ISOLATION PROTOCOLS

Protocol: Swab Samples/Nasal Washes 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.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

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.

11. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.

12. 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.*

13. 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.

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

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

16. 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 obtaining better yield.*

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

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

19. 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: Swab Samples/Nasal Washes 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.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

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.

11. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.

12. Remove the plate/tube from magnetic separation device. Add 500 μL of 80% Ethanol and mix to resuspend **MAG-S1 Particles**.

    ⚠ *Make sure the solution is homogeneous.*

13. 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.

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

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

16. 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 obtaining better yield.*

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

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

19. 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. Add 200 µL of the sample to each sample well.

   ⚠ *If the sample volume is less than 200 µL, bring the volume up to 200 µL with **Nuclease-Free Water**.*

2. To 200 µL of the sample add 100 µL of **AS Buffer**, 200 µL of **Lysis Buffer** and 20 µL of **Pro K Solution**. Mix very well.

3. Incubate at 60-65°C for 10 minutes.

4. Let the samples cool to room temperature and add 8 µL of **NBE**, 430 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.*

5. Let the samples sit at room temperature for 10 minutes.

6. Place the sample plate on the magnetic separation device for 5 minutes or more to magnetize the **MAG-S1 Particles** or until the magnetic beads clear from the solution.

7. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

8. Remove the plate from the magnetic separation device.

9. Add 400 µL of **Wash Buffer** to each sample and pipette mix 15 times to resuspend the **MAG-S1 Particles**.

10. Place the sample plate back on the magnetic separation device and wait for 5 minutes or until the magnetic beads clear from the solution.

11. With the plate on the magnetic separation device, remove and discard the supernatant.

    ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

12. Remove the plate from the magnetic separation device.

13. Add 500 µL of 80% Ethanol to the sample and pipette mix 15 times to resuspend the **MAG-S1 Particles**.

    ⚠ *Complete resuspension of the **MAG-S1 Particles** is crucial for obtaining quality DNA or RNA.*

14. Place the sample plate back on the magnetic separation device and wait for 5 minutes or until the magnetic beads clear from the solution.

15. With the plate on the magnetic separation device, remove and discard the supernatant.

    ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

16. Repeat steps 12-15 for a second wash.

17. Dry the beads by incubating for 10 minutes at room temperature with the plate still on the magnetic separation device.

    ⚠ *It is critical to completely remove any residual liquid from each well.*

18. Remove the plate from the magnetic separation device. Add 50-100 µL of **Nuclease-Free Water** (pre-warmed to 60-65°C) to each well and pipette mix 25 times to completely resuspend the **MAG-S1 Particles**.

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

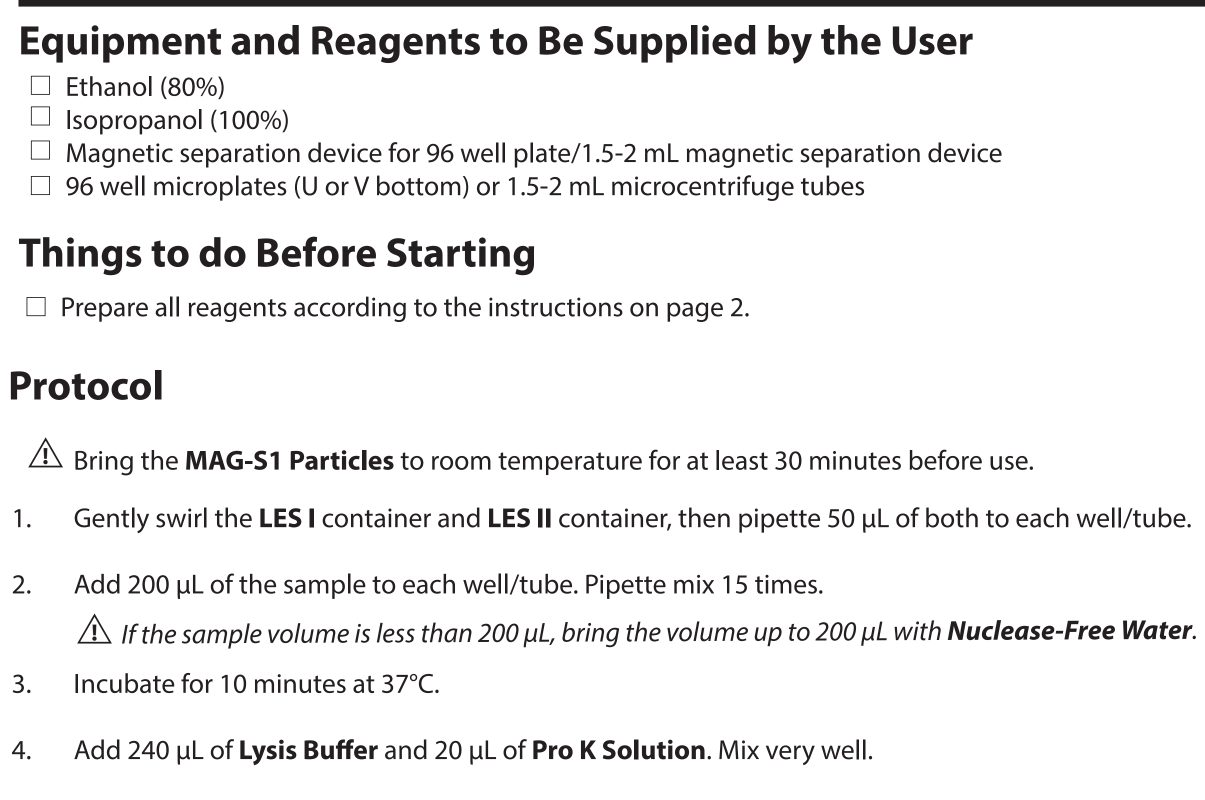
19. Incubate at room temperature for 10 minutes.

    ⚠ *Incubation at 65°C for 5 minutes may increase the DNA yield, but may affect the quality of the RNA.*

20. Place the sample plate back on the magnetic separation device and wait for 5 minutes or until the magnetic beads clear from the solution.

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

B - BACTERIAL/FUNGAL 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 and **LES II** container, then pipette 50 µL of both 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.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*
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.
11. Place the sample back on the magnetic separation device and wait for 3 minutess to magnetize the **MAG-S1 Particles**. Remove the supernatant.
12. 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.*

13. 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.

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

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

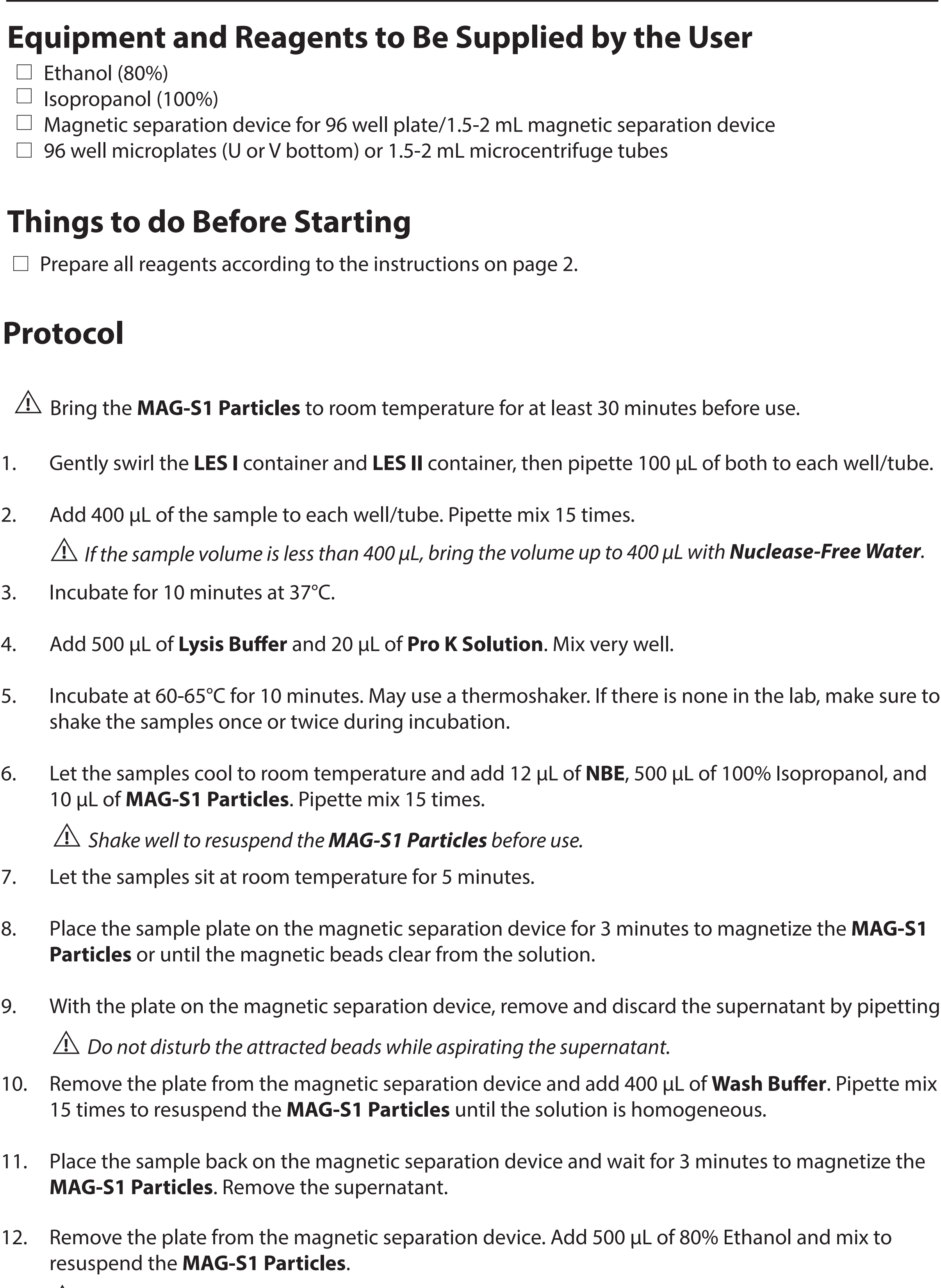
16. Remove the plate from the magnetic separation device. Add 30-100 µL of **Nuclease-Free Water** to each well/tube and pipette mix 15 times to completely resuspend the **MAG-S1 Particles**.

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

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

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

19. 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 and **LES II** container, then pipette 100 µL of both 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.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

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.

11. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.

12. Remove the plate 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.*

13. 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.

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

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

16. 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**.

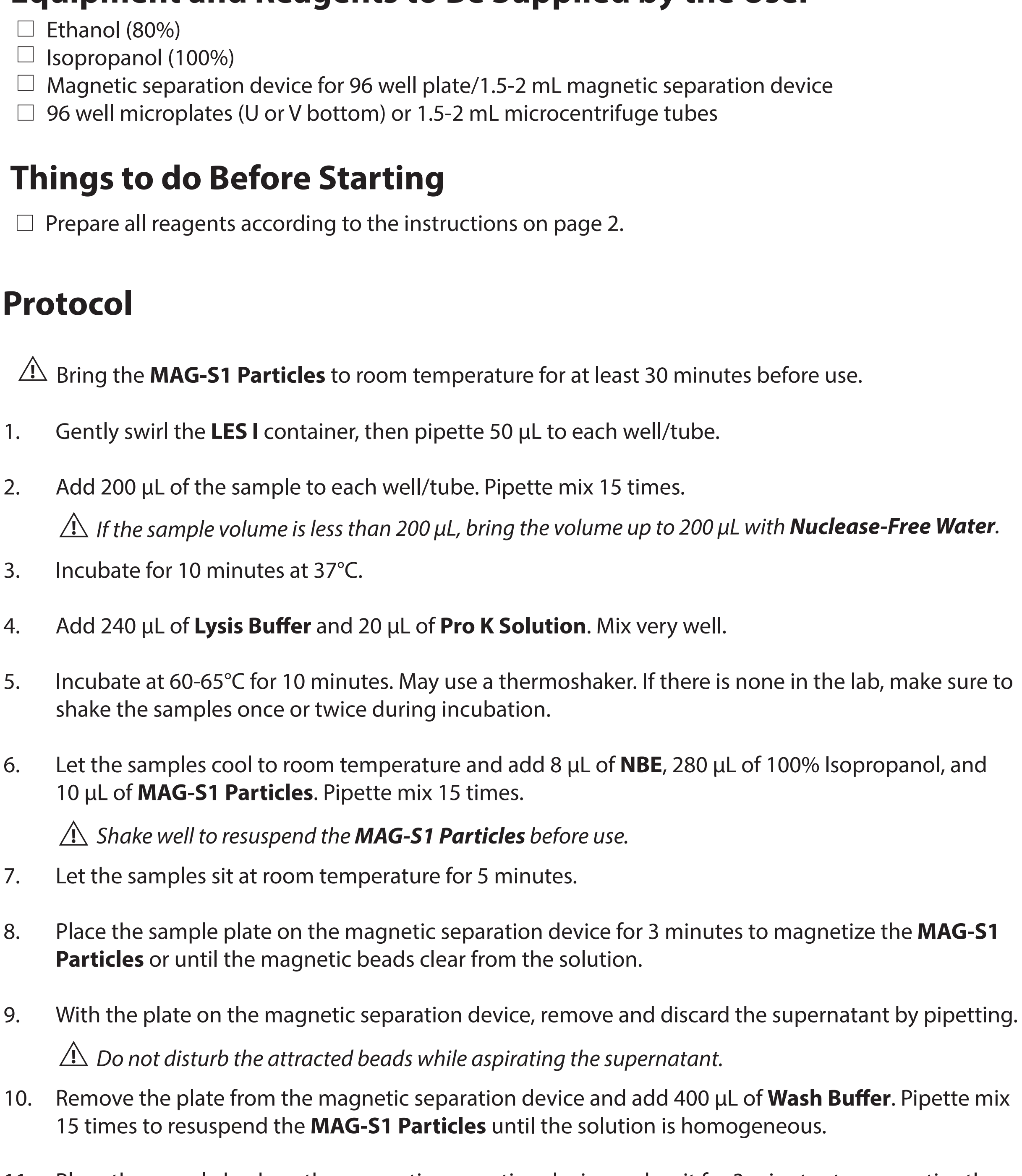
    ⚠ *Complete resuspension of the **MAG-S1 Particles** is crucial for obtaining better yield.*

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

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

19. 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.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

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.

11. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.

12. 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.*

13. 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.

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

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

16. 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 obtaining better yield.*

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

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

19. 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.

C - YEAST/FUNGAL 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 II** 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.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

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.

11. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.

12. 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.*

13. 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.

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

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

16. 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**.

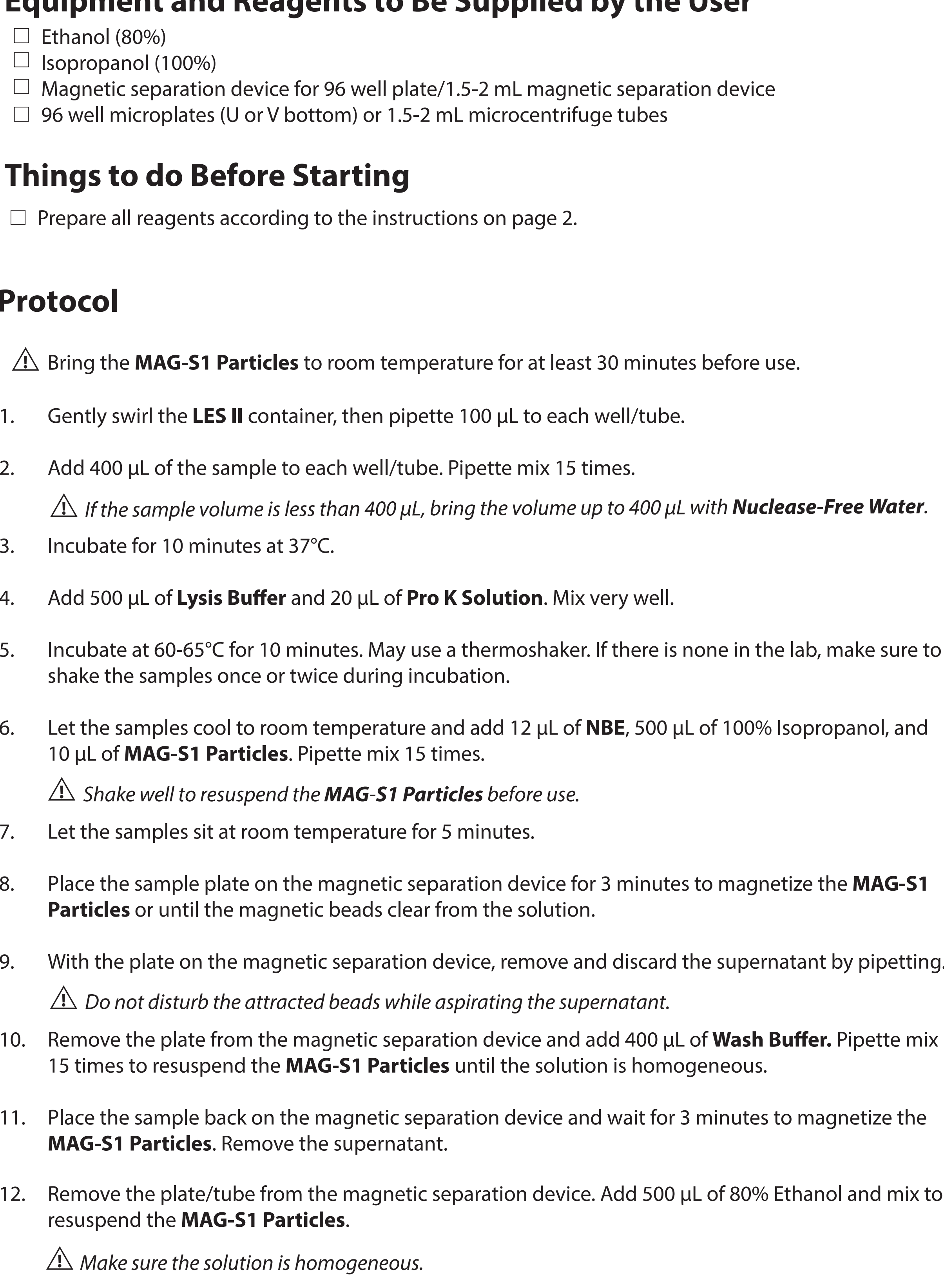
    ⚠ *Complete resuspension of the **MAG-S1 Particles** is crucial for obtaining better yield.*

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

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

19. 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 II** 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.

   ⚠ *Do not disturb the attracted beads while aspirating the supernatant.*

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.

11. Place the sample back on the magnetic separation device and wait for 3 minutes to magnetize the **MAG-S1 Particles**. Remove the supernatant.

12. 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.*

13. 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.

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

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

16. 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 obtaining better yield.*

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

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

19. 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 the Wash Buffer	Add absolute 100% Ethanol to the Wash Buffer (see page 2 for instructions)
	Inefficient cell lysis	Double the volume of Pro K Solution and increase the incubation time
MAG-S1 Particles do not completely clear from the solution	Magnetizing time too short	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 beads
Carryover of MAG-S1 Particles	The eluate contains particles and is not fully clear	Increase magnetization time. If a small amount of carryover is observed, place eluted sample on a magnetic separation device and perform an additional 5 minute magnetization

Ordering Information

HighPrep Viral-Pathogen DNA & RNA Kit - DX

Catalog No.	Product	Description	Preps
HPP-DR96E	HighPrep Viral-Pathogen DNA & RNA Kit - DX (96 preps)	Magnetic bead-based kit for isolation of total nucleic acids from viruses, bacteria, and yeast in whole blood, serum, plasma, swabs, saliva, urine, and other bodily fluids.	96
HPP-DR96x4E	HighPrep Viral-Pathogen 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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