

# HighPrep<sup>™</sup> Viral DNA/RNA Kit

Manual Revision v6.0 Catalog Nos. HPV-DR96, HPV-DR96X4, HPV-DR3840

- Total nucleic acid from whole blood serum, plasma saliva and other body fluids
- Magnetic beads based chemistry

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### **Product Description**

The HighPrep<sup>™</sup> Viral DNA/RNA kit is designed for rapid and reliable isolation of total nucleic acid from whole blood, serum, plasma, swabs, saliva 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.

#### Process

Samples are lysed in a specially formulated buffer containing detergent. Nucleic acid is bound to the surface of MAG-S1 particles under proper condition. Proteins and cellular debris are efficiently washed with 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.

HighPrep <sup>™</sup> Viral DNA/RNA Kit Catalog No.	HPV-DR96	HPV-DR96X4	HPV-DR3840	STORAGE
Number of Preps	96	384	3840	
Viral Lysis Buffer	30 ml	120 ml	1,200 ml	15-25°C
RDW Buffer <sup>1</sup>	30 ml	120 ml	1,200 ml	15-25°C
Nuclease-Free Water	35 ml	140 ml	1,400 ml	15-25°C
Pro K Solution <sup>2</sup>	1.1 ml	4.4 ml	44 ml	2-8°C
NBE <sup>3</sup>	2 ml	8 ml	80 ml	2-8°C
MAG-S1 Particles	1.1 ml	4.4 ml	44 ml	2-8°C

#### **Kit Contents and Storage**

<sup>1</sup>Ethanol must be added prior to use. See Preparation of Reagents Section.

# Stability

All components are stable for 14 months when stored accordingly.

<sup>2</sup> Pro K Solution comes in a ready to use solution. Pro K is stable for 12 months when stored at 15-25°C. For storage longer than 1 year, store at 2-8°C.

<sup>3</sup>NBE comes in a ready to use solution. NBE is stable at room temperature (15-25°C) for one week. For longer storage, keep at 2-8°C.

# **Safety Information**

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate material safety data sheets (MSDSs). MSDS can be downloaded from the "Product Resource" tab when viewing the product kit.

# **Preparation of Reagents**

Prepare the following components for each kit before use:

Catalog No.	Component	Add 100% Ethanol	Storage
HPV-DR96	RDW Buffer	20 ml	Room Temp 15-25°C
Components are stable for 14 months when stored accordingly.			

Catalog No.	Component	Add 100% Ethanol	Storage
HPV-DR96X4	RDW Buffer	80 ml	Room Temp 15-25°C
Components are stable for 14 months when stored accordingly.			

Catalog No.	Component	Add 100% Ethanol	Storage
HPV-DR3840	RDW Buffer	800 ml (400 ml per bottle)	Room Temp 15-25°C
Components are stable for 14 months when stored accordingly.			

#### Viral DNA/RNA - 50 μl sample volume (96 well format)

#### **Equipment and Reagents to Be Supplied by User**

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate material safety data sheets (MSDSs) from each product supplier.

- Ethanol (70%)
- □ Isopropanol
- □ Magnetic separation device for 96-well plate
- □ 96-well microplates (U or V bottom)

#### Things to do before starting

□ Prepare all reagents accordingly according to the instructions on page 2.

#### Protocol

- 1. Add 50µl of bodily fluid or cell culture lysate to each sample well. Note: If sample is less than 50µl, bring volume up to 50µl with nuclease-free water.
- 2. To 50µl of sample add 60µl of Viral Lysis Buffer and 5µl of Pro K Solution. Mix very well.
- 3. Incubate at 56°C 60°C for 10 min. May use a thermoshaker. If there's none in the lab, make sure to shake the samples once or twice during incubation.
- 4. Let the samples cool to room temperature and add 4μl of NBE, 70μl of Isopropanol, and 5μl of MAG-S1 Particles. Pipette mix 15 times.

 $\triangle$  Shake well to resuspend the MAG-S1 particles before use.

- 5. Let the samples sit at room temperature for 10 min.
- 6. Place the sample plate on the magnetic separation device for 5 min to magnetize the MAG-S1 particles or until the magnetic beads clear from solution.
- 7. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.

A Do not disturb the attracted beads while aspirating the supernatant.

- 8. Remove the plate from the magnetic separation device.
- 9. Add 200 μl RDW 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 min or until the magnetic beads clear from solution.

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

 $\triangle$  Do not disturb the attracted beads while aspirating the supernatant.

- 12. Remove the plate from the magnetic separation device.
- 13. Add 250 µl 70% ethanol to the sample and pipette mix 15 times to resuspend the MAG-S1 particles.

A 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 min or until the magnetic beads clear from solution.
- 15. With the plate on the magnetic separation device, remove and discard the supernatant.

 $\triangle$  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 7 min at room temperature with the plate still on the magnetic separation device.

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

18. Remove the plate from the magnetic separation device. Add 20-50 μl nuclease free water (pre-warmed to 60-65°C) to each well and pipette mix 25 times to completely resuspend the MAG-S1 magnetic particles.

Complete resuspension of the MAG-S1 particles is crucial for obtaining purity.

19. Incubate at room temperature for 10 min.

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

- 20. Place the sample plate back on the magnetic separation device and wait for 5 min or until the magnetic beads clear from solution.
- 21. Transfer the eluate (cleared supernatant containing the DNA or RNA) to a new microplate for storage. Store DNA at -20°C and RNA at -80°C.

#### Viral DNA/RNA - 200 μl sample volume (96 well format)

### **Equipment and Reagents to Be Supplied by User**

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate material safety data sheets (MSDSs) from each product supplier.

- □ Ethanol (70%)
- □ Isopropanol
- □ Magnetic separation device for 96-well plate
- □ 96-well microplates (U or V bottom)

#### Things to do before starting

□ Prepare all reagents accordingly according to the instructions on page 2.

#### Protocol

- 1. Add 200µl of bodily fluid or cell culture lysate to each sample well. Note: If sample is less than 200µl, bring volume up to 200µl with nuclease-free water.
- 2. To 200µl of sample add 240µl of Viral Lysis Buffer and 10µl of Pro K Solution. Mix very well.
- 3. Incubate at 56°C 60°C for 10 min. May use a thermoshaker. If there's none in the lab, make sure to shake the samples once or twice during incubation.
- 4. Let the samples cool to room temperature and add 8μl of NBE, 280μl of Isopropanol, and 10μl of MAG-S1 Particles. Pipette mix 15 times.

A Shake well to resuspend the MAG-S1 particles before use.

- 5. Let the samples sit at room temperature for 10 min.
- 6. Place the sample plate on the magnetic separation device for 5 min to magnetize the MAG-S1 particles or until the magnetic beads clear from solution.
- 7. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.

A Do not disturb the attracted beads while aspirating the supernatant.

- 8. Remove the plate from the magnetic separation device.
- 9. Add 400 μl RDW 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 min or until the magnetic beads clear from 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  $\mu l$  70% 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 min or until the magnetic beads clear from solution.
- 15. With the plate on the magnetic separation device, remove and discard the supernatant.

A 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 min at room temperature with the plate still on the magnetic separation device.

riangle 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 nuclease free water (pre-warmed to 60-65°C) to each well and pipette mix 25 times to complete resuspend the MAG-S1 magnetic particles.

A Complete resuspension of the MAG-S1 particles is crucial for obtaining purity.

19. Incubate at room temperature for 10 min.

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

- 20. Place the sample plate back on the magnetic separation device and wait for 5 min or until the magnetic beads clear from solution.
- 21. Transfer the eluate (cleared supernatant containing the DNA or RNA) to a new microplate for storage. Store DNA at -20°C and RNA at -80°C.

#### Viral DNA/RNA - 500 μl sample volume (96 well format)

#### **Equipment and Reagents to Be Supplied by User**

When working with chemicals, always wear a suitable lab coat, disposable gloves, and protective goggles. For more information, please consult the appropriate material safety data sheets (MSDSs) from each product supplier.

- □ Ethanol (70%)
- □ Isopropanol
- □ Magnetic separation device for 96-well plate
- □ 96-well deep plate

#### Things to do before starting

□ Prepare all reagents accordingly according to the instructions on page 2.

#### Protocol

- 1. Add 500µl of bodily fluid or cell culture lysate to each sample well. Note: If sample is less than 500µl, bring volume up to 500µl with nuclease-free water.
- 2. To 500µl of sample add 500µl of Viral Lysis Buffer and 20µl of Pro K Solution. Mix very well.
- 3. Incubate at 56°C 60°C for 10 min. May use a thermoshaker. If there's none in the lab, make sure to shake the samples once or twice during incubation.
- 4. Let the samples cool to room temperature and add 16μl of NBE, 500μl of Isopropanol, and 10μl of MAG-S1 Particles. Pipette mix 15 times.

A Shake well to resuspend the MAG-S1 particles before use.

- 5. Let the samples sit at room temperature for 10 min.
- 6. Place the sample plate on the magnetic separation device for 10 min to magnetize the MAG-S1 particles or until the magnetic beads clear from solution.
- 7. With the plate on the magnetic separation device, remove and discard the supernatant by pipetting.

 $\triangle$  Do not disturb the attracted beads while aspirating the supernatant.

- 8. Remove the plate from the magnetic separation device.
- 9. Add 500 μl RDW 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 min or until the magnetic beads clear from 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  $\mu l$  70% 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 min or until the magnetic beads clear from solution.
- 15. With the plate on the magnetic separation device, remove and discard the supernatant.

A 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 min 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 nuclease free water (pre-warmed to 60-65°C) to each well and pipette mix 25 times to complete resuspend the MAG-S1 magnetic particles.

A Complete resuspension of the MAG-S1 particles is crucial for obtaining purity.

19. Incubate at room temperature for 10 min.

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

- 20. Place the sample plate back on the magnetic separation device and wait for 5 min or until the magnetic beads clear from solution.
- 21. Transfer the eluate (cleared supernatant containing the DNA or RNA) to a new microplate for storage. Store 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: 301-302-0144 (in US), outside US, 1-855-262-4246

Email: support@magbiogenomics.com

Symptoms	Possible Causes	Comments	
	Incomplete resuspension of MAG-S1 particles.	Resuspend MAG-S1 particles by vortexing vigorously before use.	
Low DNA or RNA Yield	Loss of MAG-S1 particles during operation.	Avoid disturbing the MAG-S1 particles during aspiration of supernatant.	
	Ethanol is not added into RDW Buffer.	Add absolute 100% Ethanol to RDW 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 solution	Too short of magnetizing time.	Increase collection time on the magnet.	
Problems in downstream	Insufficient DNA/RNA in starting material	Use more starting material.	
applications	Ethanol carry-over.	Dry the MAG-S1 particles completely before elution.	
Carryover of MAG-S1 particles	The eluate has particles and is not fully clear.	Increase magnetization time. If small amount of carryover, place eluted sample on a magnetic separtion device and perform an additional 5 min magnetization.	

# **Ordering Information**

Product Description	Catalog No.	Preps
HighPrep <sup>™</sup> Viral DNA/RNA (96 preps)	HPV-DR96	96
HighPrep <sup>™</sup> Viral DNA/RNA (384 preps)	HPV-DR96X4	384
HighPrep <sup>™</sup> Viral DNA/RNA (3840 preps)	HPV-DR3840	3840

HighPrep™Viral DNA/RNA



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