

MBead Virus Nucleic acid Kit

Cat No. PDM04-0100(S)

Size: 100 Reactions



Description

This MBead Virus Nucleic acid Kit is designed specifically for the simultaneous virus DNA/RNA purification from the plasma, serum, body fluid or supernatant of virus-infected cell cultures. Its unique buffer system will efficiently lyse cells and degrade proteins, allowing for the nucleic acid to be easily bound by the surface of the magnetic beads. The other non-specific binding particles are removed with a wash buffer, and the nucleic acid is then released into the Release Buffer. The nucleic acid can be purified manually within 10~15 minutes (using most magnetic separators) or the kit can be easily adapted to satisfy most automated nucleic acid purification systems.

Kit Contents

Contents	PDM04-0100	PDM04-0100S
Magnetic Bead	2 ml	80 μ l
Lysis Buffer	30 ml	1.5 ml
Wash Buffer	80 ml	4 ml
Release Buffer	20 ml	1 ml

Specifications

- Sample: Up to 300 μ l of the virus sample
- Operation time: Within 10~15 minutes
- Applications: Restriction Enzyme Digestion, Southern Blotting, PCR and qPCR assays
- Storage: Room temperature

Required Materials

- Absolute EtOH
- Magnetic separator
- 1.5 ml microcentrifuge tubes
- Water bath / Dry bath

NOTE: Be sure and allow the magnetic beads to disperse completely during the binding, wash and elution steps.

Protocol

Step 1 Lysis

1. Transfer up to 300 μ l of the virus sample into a 1.5 ml microcentrifuge tube and add 300 μ l of the Lysis Buffer.
2. Mix well and incubate at 65°C for 5 minutes. During this time, pre-heat the Release Buffer to 65°C for the Step 4.
3. Add 300 μ l of the absolute EtOH to the lysate and mix well.

Step 2 DNA Binding

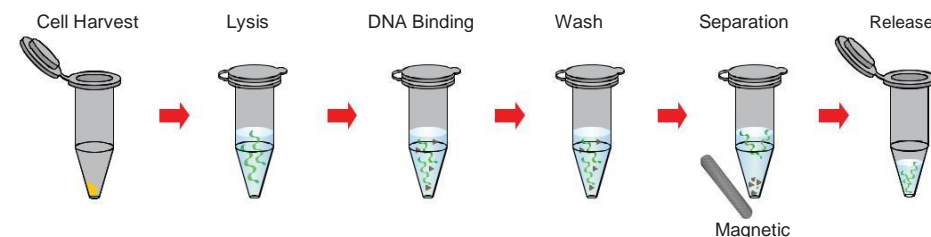
1. Add 20 μ l of the Magnetic Beads. Mix well by gently shaking for 3 minutes.
2. Place the tube in a magnetic separator for 30 seconds.
3. Remove the solution (If the mixture becomes viscous, increase magnetic bead separation time).

Step 3 Wash

1. Add 800 μ l of the Wash Buffer and mix well (Following the wash, the mixture will no longer be viscous).
2. Place the tube in a magnetic separator for 30 seconds. Remove the solution.

Step 4 Release

1. Add 200 μ l of the Release Buffer (pre-heated to 65°C) and mix well.
2. Incubate for 3 minutes at 65°C (During the incubation, shake the tube vigorously every minute).
3. Place the tube in a magnetic separator for 1 minute.
4. Carefully transfer ONLY the clean portion of the solution to a clean tube.



Troubleshooting

Refer to the table below to troubleshoot problems that you may encounter when purifying the genomic DNA with the kit.

Problem	Cause	Solution
DNA is sheared or degraded	Lysate mixed too vigorously	Use the appropriate pipette tip set. For the required volume, lower under the reading line of the solution to mix the sample. Pipet up and down gently to mix.
	DNase contamination	Maintain a sterile environment while working (e.g. wear gloves and use DNase-free reagents).
	Incomplete removal of the RNase	RNase A treatment
RNA containment	Incomplete lysis and homogenization	Reduce the amount of the starting material.
Low yields of gDNA	Incorrect handling of Magnetic Beads	Vortex the tube containing the magnetic beads to fully resuspend the beads before adding them to your sample.
	Incorrect elution conditions	During the Elution Step, incubate at 65°C for 3min, and vortex every minute.
	The quality of the starting material may not be optimal	Use the fresh sample and process immediately after collection, or freeze the sample at -80°C or in liquid nitrogen.
High background on UV measurement	Residual beads released	Repeat magnetic separation and transfer the eluate to a clean tube.

Caution

- During the operation, always wear a lab coat, disposable gloves, and protective equipment.
- Research Use Only. Not intended for any animal or human therapeutic or diagnostic uses.