

MACHEREY-NAGEL Application Note 05/2023 · Bioanalysis

NucleoMag[®] VET

Automated nucleic acid extraction from veterinary samples using the MagnetaPure 32+

Application benefits

The integration of the well-established NucleoMag[®] VET technology and the MagnetaPure 32+ system offers multiple benefits, optimizing molecular veterinary diagnostic workflows, while the utillization of NucleoProtect Swab and Blood collection tubes enhances biosafety by reducing the potential exposure to zoonotic pathogens:

- Verified automation method for RNA / DNA isolation from various veterinary sample materials
- Simultaneous processing of up to 32 samples in parallel
- Consistent and reliable results
- No programming required: Verified and pre-installed methods available

Keywords

Veterinary diagnostics, animal health, testing, viral RNA / DNA, bacterial DNA, biosafety, blood, swab, NucleoMag[®], NucleoProtect[®] VET, magnetic beads, magnetic rod system, MagnetaPure



Introduction

Veterinarians commonly utilize molecular detection tools to screen apparently healthy animals, diagnose infections, monitor disease progression, and assess therapeutic responses. These techniques, such as qRT-PCR and next-generation sequencing, rely on efficient nucleic acid purification as a crucial step in the diagnostic process. With the growing demand for processing and analyzing numerous samples, especially during disease outbreaks, automation has become indispensable in modern veterinary diagnostic laboratories.

The NucleoMag[®] VET kit by MACHEREY-NAGEL was specifically developed for the automated and rapid purification of viral DNA/RNA and bacterial DNA from diverse animal sample materials, employing a magnetic bead-based format that is well-suited for automation. This purification method ensures reliable and consistent results, delivering nucleic acids with high yields and purity.

This application note showcases the automated extraction of pathogen nucleic acids from veterinary samples using the NucleoMag[®] VET kit on the MagnetaPure 32+ system, which is a compact nucleic acid extraction platform based on magnetic bead technology. With the capacity to process up to 32 samples simultaneously, this user-friendly instrument offers pre-installed verified scripts and detailed protocol information, streamlining the mixing, magnetic bead transfer, washing, and elution steps and saving valuable hands-on time. For more information on the MagnetaPure 32+ and additional application notes, please visit www.mn-net.com/MagnetaPure32.

NucleoMag [®] VET	
Technology	Magnetic beads
Sample material	≤ 200 µL whole blood, serum, plasma; ≤ 200 µL swab wash solution; ≤ 25 mg tissue (e.g. ear notches); ≤ 200 µL feces; NucleoProtect [®] VET-collected blood and swab samples
Elution volume	50 – 200 µL
Fragment size	300 bp – approx- 50 kbp
Max. sample number on the MagnetaPure 32+	32 samples

MagnetaPure 32+	
Technology	Automated magnetic rod system
Display	7 inch-color touch screen
Capacity / volume per well	1 – 32 samples / 50 μL to 1000 μL
Dimensions	417 x 410 x 426 mm
Weight	20 kg
Contamination control	UV lamp, internal filter system
Website	www.mn-net.com/MagnetaPure32

Material and Methods

Porcine chewing cord and blood samples were collected and preserved overnight using the NucleoProtect[®] VET stabilizing and inactivating reagent. The subsequent extraction of the samples followed the instructions provided in the manual for the NucleoMag[®] VET kit, utilizing lysis buffer VL1 and Proteinase K for porcine chewing cord samples, and buffer VIA for NucleoProtect[®] VET-stabilized porcine blood samples. Nucleic acids were reversibly bound to paramagnetic NucleoMag[®] B-beads by

Application Data



adjusting with Binding Buffer VEB. The magnetic separation was followed by washing the NucleoMag[®] B-beads with Wash Buffer VEW1 and VEW2 to remove contaminants and salts. After a final washing step with 80% ethanol, the magnetic beads were air-dried and subsequently eluted in elution buffer VEL.

All binding, washing, and magnetic bead separation steps were carried out by the MagnetaPure 32+ magnetic rod device.

High sensitivity detection of viral RNA recovered from NucleoProtect® VET-stabilized chewing cord samples

Porcine chewing cord samples of 200 µL were collected using NucleoProtect[®] VET Swab collection tubes and spiked with 5 µL of a dilution series of PEV virus stock solution (undiluted, 1:10, 1:100, 1:1000, with dilution performed in PBS). After overnight storage at room temperature, viral PEV nucleic acids (ss(-)RNA) were extracted using the NucleoMag[®] VET kit on the MagnetaPure 32+ magnetic rod extraction robot (n=4 for each dilution). Subsequent qRT-PCR analysis was conducted using the AgPath-ID[™] One-Step RT-PCR reagent on a BioRad CFX96 Real-Time PCR System. The protocol demonstrates highly sensitive detection of RNA and excellent reproducibility in the log10 dilution series.

High sensitivity detection of viral RNA recovered from NucleoProtect® VET-collected porcine blood samples

Porcine blood samples, collected using NucleoProtect[®] VET Blood collection tubes, were spiked with series of dilutions of PEV virus stock solution (undiluted, 1:10, 1:100, 1:1000, with dilution performed in PBS). After overnight storage at room temperature, viral PEV nucleic acids (ss(-) RNA) were extracted using the NucleoMag[®] VET kit on the MagnetaPure 32+ extraction robot (n=4 for each dilution). The extraction followed the manual guidelines for NucleoProtect[®] VET-stabilized blood samples on magnetic rod systems, using buffer VIA. Subsequent qRT-PCR analysis was conducted using the AgPath-ID[™] One-Step RT-PCR reagent on a BioRad CFX96 Real-Time PCR System. The protocol demonstrates highly sensitive detection of RNA and excellent reproducibility in the log10 dilution series.

NucleoProtect® VET Swab and Blood sample tubes

NucleoProtect[®] VET, developed by MACHEREY-NAGEL, is a reliable and effective stabilizing and inactivating reagent designed for preserving nucleic acids in veterinary samples. It ensures the integrity and quality of the nucleic acids during sample collection and storage, even for extended periods. NucleoProtect[®] VET enhances biosafety by reducing the risk of exposure to zoonotic pathogens, making it an essential tool for molecular veterinary diagnostics. NucleoProtect[®] VET reagent is offered in three distinct formats, including pre-filled and barcoded swab or blood collection tubes, as well as a bottled reagent option. More information available at *www.mn-net.com/animalhealth*

Ordering information

Product	Specifications	Quantity	REF
NucleoMag [®] VET	Magnetic bead-based kit for the isolation of viral nucleic acids and bacterial DNA from veterinary samples; including NucleoMag [®] B-Beads, buffers, Carrier RNA, and Proteinase K	1 x 96 preps 4 x 96 preps 100 x 96 preps	744200.1 744200.4 744200.100
NucleoProtect [®] VET	DNA / RNA stabilization and inactivation reagent for veterinary swab and blood samples. NucleoProtect [®] VET is available as bottled reagent or as prefilled blood and swab collection tubes. More information at www.mn-net.com/animalhealth	50 blood collection tubes 50 swab collection tubes 50 / 500 mL bottled reagent	740755 740760 740750.50 / .500
Buffer VIA	Buffer VIA is needed for the nucleic acid extraction from NucleoProtect® VET-collected blood samples using the NucleoMag® VET kit	150 mL	744206
MagnetaPure 32+	Magnetic rod system for automated nucleic acid extraction using MACHEREY-NAGEL NucleoMag [®] kits, parallel processing of up to 32 samples	1	747010
96 Deep-well plates	96 deep-well plates for MagnetaPure 32+	25	744955
Tip combs	8-place magnetic tip combs for MagnetaPure 32+	50	744960

NucleoMag® and NucleoProtect® are registered trademarks of MACHEREY-NAGEL; AgPath-IDTM is a registered trademark of Thermo Fisher Scientific; MagnetaPure is a brand of Dominique Dutscher.