MACHEREY-NAGEL

Viral nucleic acid purification from wastewater

Reliable concentration and purification of viral nucleic acids from wastewater with the INNOVAPREP CP-Select™ sample concentration device

Introduction

Many researchers are working on developing wastewater surveillance strategies for COVID-19, aiming to develop a highly sensitive, accurate and reliable method to achieve real-time surveillance and predict future outbreaks.

As viruses are highly diluted in wastewater, it is crucially important to apply effective concentration methods before the subsequent RNA extraction and RT-qPCR detection.

However, most sample concentration methods like ultracentrifugation or precipitation are very time consuming and tedious as they require large centrifuges and expensive equipment. Furthermore, many concentration methods cause challenges for nucleic acid extraction since PCR inhibitors are concentrated by those methods as well.

In this application note, we show efficient and inhibition-free nucleic acid purification from synthetic wastewater samples with MACHEREY-NAGEL's NucleoMag® DNA/RNA Water and NucleoSpin® RNA Virus extraction kits in combination with sample concentration on the INNOVAPREP Concentrating Pipette Select.

Products at a glance

NucleoMag® DNA/RNA Water	
Technology	Magnetic bead technology
Sample material	10–1000 mL
Preparation time	40–120 min/96 preps (excl. lysis and sample concentration)
Fragment size	300 bp–approx. 50 kbp
Elution volume	50–250 µL

NucleoSpin® RNA Virus	
Technology	Silica membrane technology
Sample material	< 150 µL cell free fluid
Preparation time	30 min/4–6 preps (excl. lysis and sample concentration)
Fragment size	100 bp–approx. 50 kbp
Elution volume	50 µL

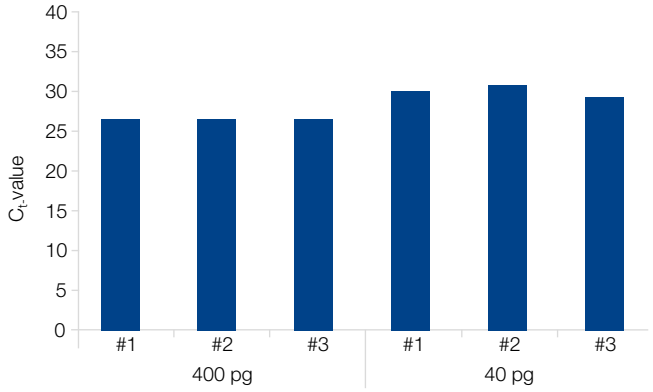
INNOVAPREP CP-Select™ sample concentration device	
Technology	Single-use Concentrating PipetteTips
Sample Volume	up to 5 L
Rate	up to 150 mL/min
Elution volumne	150 µL up to 1000 µL (Wet Foam Elution™)
Size / weight	13″ × 11″ × 7″ / 8 lbs

Material and methods

Standard synthetic wastewater was prepared according to ASTM standards (D5905). The wastewater samples were filtered through a 0.2 µm filter unit and 20–40 mL were concentrated via the INNOVAPREP CP-Select™ sample concentration device. For more information on sample concentration using the INNOVAPREP Concentrating Pipette Select please visit: https://drive.google.com/file/d/1TU0dT_vahGNc8HkFbFl9g4w85dSKm67Z/view.

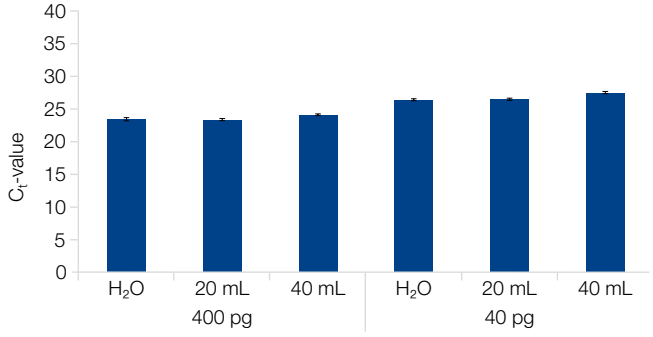
After sample concentration, nucleic acids were isolated with either NucleoMag® DNA/RNA Water or NucleoSpin® RNA Virus. First, 10 µL of Liquid Proteinase K (see ordering information) was mixed with 200 µL of the wastewater concentrate and the indicated volumes of lysis buffer. Samples were incubated at 56 °C for 10 min and processed according to the standard protocols.

Application data



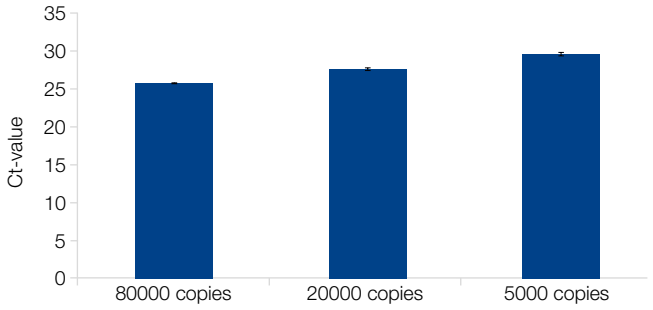
NucleoSpin® RNA Virus allows reliable, inhibition-free qPCR detection of MS2 phage RNA from wastewater concentrates

RNA was purified from synthetic wastewater concentrates spiked with different amounts of MS2 phage RNA using the NucleoSpin RNA Virus kit. qRT-PCR analysis was performed with a Taqman® probe for MS2 RNA using the SensiFastTM Probe One-Step Lo-ROX kit from Bioline on an Applied Biosystems® 7500 Real-Time PCR system. MS2 bacteriophage RNA was detected consistently and reliably.



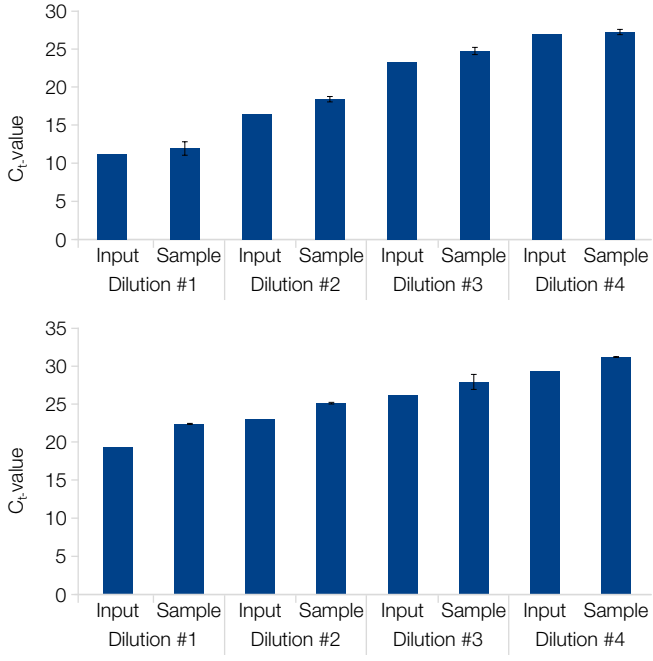
NucleoMag® DNA/RNA Water allows reliable, inhibition-free qPCR detection of MS2 phage RNA from wastewater concentrates.

MS2 bacteriophage RNA was spiked into wastewater concentrates from 20 or 40 ml or into water in two different concentrations. RNA was isolated using the NucleoMag® DNA/RNA Water kit. qRT-PCR analysis was performed with a Taqman® probe for MS2 RNA using the SensiFastTM Probe One-Step Lo-ROX kit from Bioline on an Applied Biosystems® 7500 Real-Time PCR System.



Detection of armored control RNA in wastewater concentrates

Different amounts of armored control RNA were spiked into wastewater concentrates from 40 ml sample before RNA isolation. qRT-PCR results demonstrate the reliable detection of different amounts of SARS-CoV-2 RNA purified with NucleoMag® DNA/RNA Water.



Superior RNA and DNA recovery from wastewater concentrates

NucleoMag® DNA/RNA Water was used to isolate DNA and RNA from fake wastewater concentrates spiked with different amounts of T7 DNA (upper data) or MS2 phage RNA (lower data). The recovered DNA and RNA were compared to the respective DNA and RNA input demonstrating superior DNA and RNA recovery from wastewater concentrates.

Convenient and reliable nucleic acid extraction from wastewater samples!

Combine INNOVAPREP's convenient sample concentration device CP-Select™ with MACHEREY-NAGEL's proven nucleic acid purification procedures for reliable nucleic acid concentration and purification from wastewater samples.

- Reliable performance, excellent removal of PCR inhibitors
- Convenient sample concentration without the need for time consuming and labor-intensive methods.

Application data

Product	Specifications	Pack of	REF
NucleoMag® DNA/RNA Water	Magnetic bead-based isolation of DNA and RNA from water and air samples	96 preps 384 preps	744220.1 744220.4
NucleoSpin® RNA Virus	Mini kit for viral RNA from cell-free fluids	50 preps 250 preps	740956.50 740956.250
Liquid Proteinase K	Ready-to-use Liquid Proteinase K, recombinant	5 mL 30 mL	740396 740396.30

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For more detailed information, please visit https://www.innovaprep.com/.