MACHEREY-NAGEL

NucleoMag® VET

Comparison of NucleoMag® VET with other commercial nucleic acid extraction kits used in veterinary diagnostics



Introduction

The nucleic acid extraction in veterinary diagnostics is quite difficult as there is a large diversity of starting materials, such as whole blood, serum, swabs, tissue, and ear notches. Due to this large variety, different extraction methods are used and a sample specific lysis condition is needed to enable an efficient extraction of nucleic acids and a subsequent reliable down-stream application. In addition, the content of target molecules is often low in certain starting materials.

Here, we present a comparison of commercial nucleic acid extraction kits available for isolation of viral RNA and / or DNA. The extraction kits were compared by the detection of four different viruses: Bluetongue virus (BTV), Classical Swine Fever virus (CSFV), Porcine Circovirus Type 2 (PCV 2), and Schmallenberg virus (SBV).

For the comparison, NucleoMag® VET kit and other extraction kits from well-known manufacturers were used. The NucleoMag® VET kit is based on magnetic bead technology, therefore the comparing extractions were performed on KingFisher® Flex instrument. For comparison two manual kits based on silica membrane technology were tested additionally.

Material and methods

In the comparing extractions, four different viruses in typical sample material were tested. For each virus, 12 samples with different virus titers were tested.

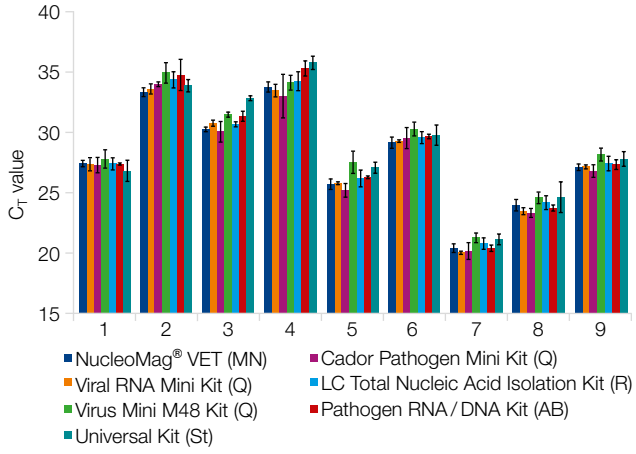
- Bluetongue virus in blood treated with EDTA (cattle)
- Classical Swine Fever virus in blood treated with EDTA (pig)
- Porcine Circovirus Type 2 in a tissue suspension (pig)
- Schmallenberg virus in serum (cattle)

All extractions were performed in duplicates (biological duplicates) and were applied in the real-time PCR in duplicates (technical duplicates, qScript XLT One-Step RT-qPCR Tough Mix (Quanta)).

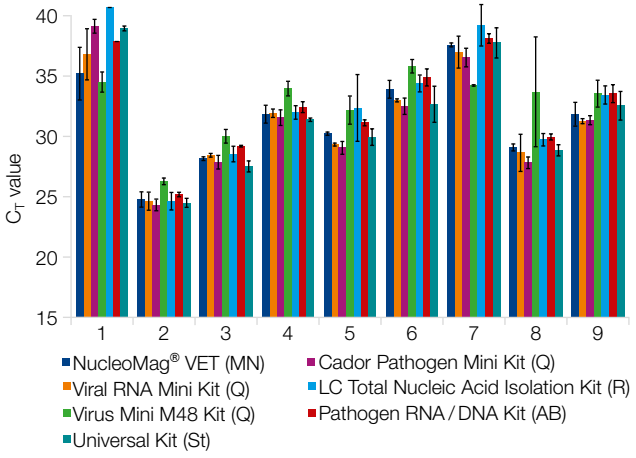
Product	Manufacturer	Technology	Processing
Viral RNA Mini Kit	Q	Silica membrane	Manual
Cador Pathogen Mini Kit	Q	Silica membrane	Manual
Virus Mini M48 Kit	Q	Magnetic bead	KingFisher® 96 Flex
LC Total Nucleic Acid Isolation Kit	R	Magnetic bead	MagNa Pure LC 2.0
NucleoMag® VET	MN	Magnetic bead	KingFisher® 96 Flex
Pathogen RNA / DNA Kit	T (AB)	Magnetic bead	KingFisher® 96 Flex
Universal Kit	St	Magnetic bead	KingFisher® 96 Flex

Application data

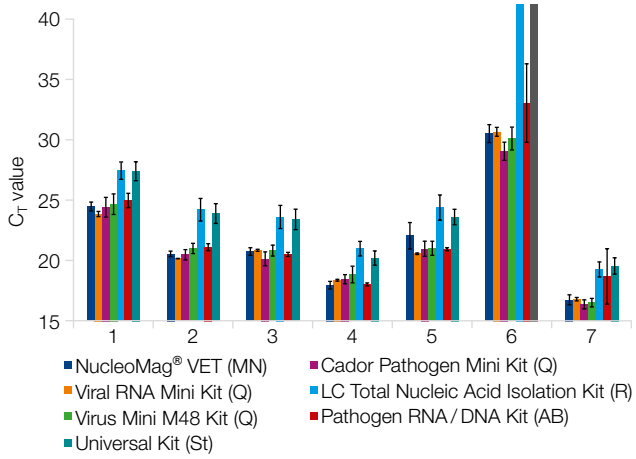
A: Detection of BTV



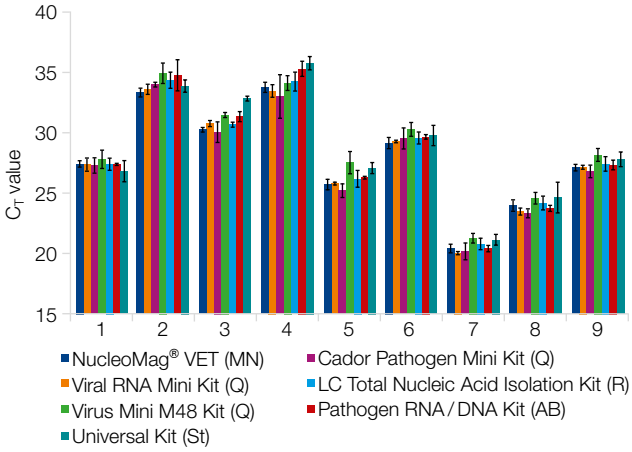
B: Detection of CSFV



C: Detection of PCV 2



D: Detection of SBV



Detection of BTV, CSFV, PCV 2, and SBV after DNA or RNA isolation using different extraction kits.

Specific qPCR was performed to determine the viral titer load in the different sample materials. (A) Detection of BTV (Bluetongue virus) in cattle blood. (B) Detection of CSFV (Classical Swine Fever virus) in pig blood. (C) Detection of PCV 2 (Porcine Circovirus Type 2) in pig tissue. (D) Detection of SBV (Schmallenberg virus) in cattle serum.

Data was kindly provided by Dr. Hoffmann, Friedrich-Loeffler-Institut, Germany

Conclusion

The qRT-PCR data show that DNA or RNA concentration varies dependent on the used extraction kit, which results in a different detection of BTV, CSFV, PCV 2, and SBV. For the detection of the different viruses in the different sample materials, it is shown that DNA or RNA concentration was always high using the NucleoMag® VET compared to the other kits.

The obtained C_T values for the viral nucleic acids and sample materials show that the NucleoMag® VET kit is a very consistent and reliable extraction kit getting low C_T values for a reliable virus detection independent from virus type or sample material.

Ordering information

Product	Preps	REF
NucleoMag® VET	1 x 96 / 4 x 96	744200.1 / .4

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