ViralSEQ Vesivirus Detection Assay

Integrated sample preparation and real-time PCR assay for the detection of vesivirus in cell culture samples

- Detection of known vesivirus strains
- Rapid time-to-results of less than 5 hours
- Applied Biosystems™ TaqMan™ assay–based real-time PCR
- Demonstrated sensitivity and specificity
- Proprietary optimized sample preparation for high RNA recovery

Viral contamination in mammalian product manufacturing presents a serious risk to the manufacturing process, the manufacturing facility, and the integrity of the product. Vesivirus 2117 is an RNA virus that has reemerged as a potential threat to mammalian cell culture production. To meet the challenge of rapid, specific, and sensitive detection of vesivirus, we have developed the first commercially available integrated kit for isolation and detection of vesivirus from mammalian cell culture samples. The Applied Biosystems™ ViralSEQ™ Vesivirus Detection Assay is a PCR-based test that enables users to quickly isolate and detect vesivirus 2117 and all other known strains with high sensitivity and specificity. The test utilizes Applied Biosystems™ PrepSEQ™ sample preparation magnetic beads to isolate viral RNA from a wide range of sample types, combined with TaqMan assay–based technology for sensitive and specific detection of the viral RNA. The kit also includes a proprietary positive control template that helps eliminate the possibility of false positive results arising from inadvertent contamination of test samples with the positive control.

Table 1. Exclusion panel. The ViralSEQ Vesivirus Detection Assay has been demonstrated to be specific for vesivirus and does not detect RNA purified from other organisms.

Sample	RNA quantity	Sample	RNA quantity
Bacillus	2.5 ng	Chicken	5 ng
Clostridium	2.5 ng	Dog	5 ng
E. coli	2.5 ng	Human	5 ng, 1 µg
Lactococcus	2.5 ng	MDCK	5 ng
Shigella	2.5 ng	Rat	5 ng
Yeast	5 ng	CHO	500 ng

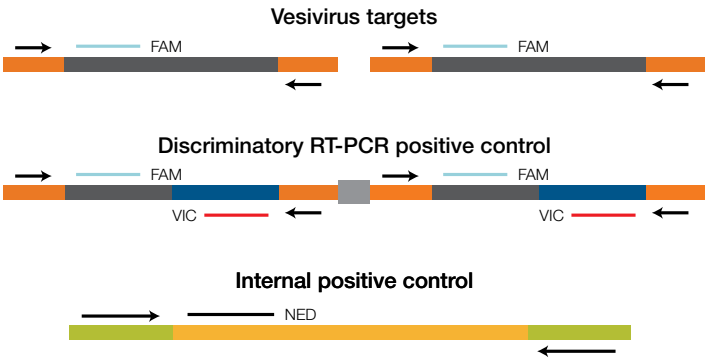

Figure 1. Multiplex assay and control design. The unique design of the ViralSEQ Vesivirus Detection Assay includes two viral targets and discriminatory positive controls. This allows detection of known vesivirus genomes. The discriminatory positive control helps monitor the real-time PCR reaction and cross-contamination of samples to reduce false positive results.

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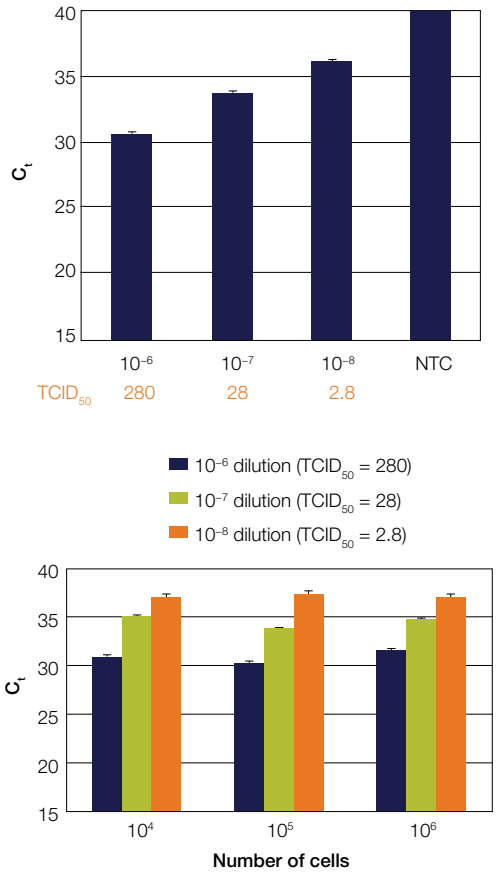

Figure 2. Applied Biosystems™ PrepSEQ™ 1-2-3 Nucleic Acid Extraction Kit. Optimized for use with the ViralSEQ Vesivirus Detection Assay, the sample preparation procedure allows detection of $TCID_{50}$ = 2.8 in 100 µL of cell culture and serum. In this example, 10^{-6} to 10^{-8} dilutions of feline calicivirus (used as a surrogate for vesivirus) were spiked into 100 µL of fetal bovine serum (top panel) and cell culture (bottom panel). The serial dilutions of the virus correspond to $TCID_{50}$ of 280, 28, and 2.8.

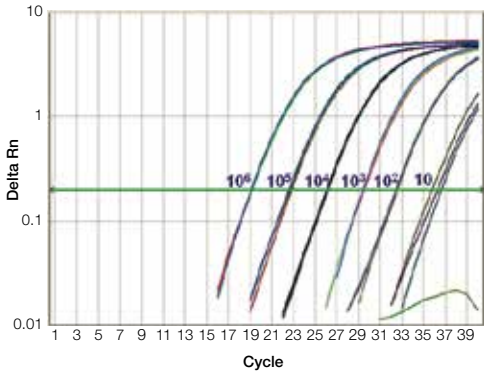

Figure 3. High sensitivity and broad dynamic range. The ViralSEQ Vesivirus Detection Assay can detect from 10^6 copies down to 10 copies of *in vitro*–transcribed vesivirus RNA per reaction.

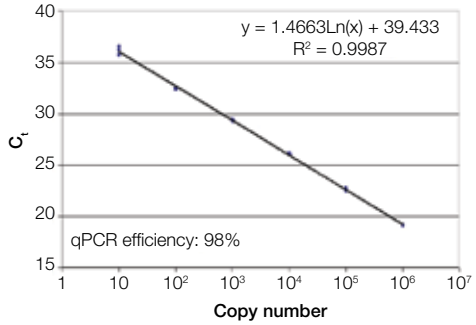

Figure 4. High PCR efficiency. Using the viral RNA template, the ViralSEQ Vesivirus Detection Assay demonstrated high real-time PCR efficiency and a broad linear detection range.

Ordering information

Product	Quantity	Cat. No.
ViralSEQ Vesivirus Detection Assay Contains real-time PCR assay, buffer and enzyme mix, positive control, and negative control	100 rxns	4448398C
PrepSEQ 1-2-3 Nucleic Acid Extraction Kit Contains buffers, magnetic beads, and enzymes	100 rxns	4452222

Find out more at **thermofisher.com/viralseq**

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