

Bear Canyon Virus, VH-C067

Catalog No. NR-12235

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Product Description: Clarified supernatant from *Cercopithecus aethiops* kidney epithelial cells (Vero E6)¹ infected with Bear Canyon virus (BCNV), VH-C067

Lot²: 60591585

Manufacturing Date: 13OCT2013

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero E6 Cells ¹ Using RT-PCR ³	Report results	Infectious, no cytopathic effect
Sequencing of Species-Specific Region (363 nucleotides)	Consistent with BCNV ⁴	100% identity with BCNV ⁴ (GenBank: AY924391)
Titer by TCID ₅₀ Assay ⁵ in Vero E6 Cells ¹ Using RT-PCR ³	Report results	1.6 × 10 ⁴ TCID ₅₀ per mL
Functional Activity by RT-PCR Assay	~ 370 bp amplicon	~ 370 bp amplicon
Sterility (21-day incubation) Harpo's HTYE broth ⁶ , 37°C and 26°C, aerobic Trypticase soy broth, 37°C and 26°C, aerobic Sabouraud broth, 37°C and 26°C, aerobic Sheep blood agar, 37°C, aerobic Sheep blood agar, 37°C, anaerobic Thioglycollate broth, 37°C, anaerobic DMEM with 10% FBS, 37°C and 5% CO ₂	No growth No growth No growth No growth No growth No growth No growth No growth	No growth No growth No growth No growth No growth No growth No growth No growth
Mycoplasma Contamination Agar and broth culture (14-day incubation at 37°C) DNA Detection by PCR of Test Article nucleic acid	None detected None detected	None detected None detected

¹Vero E6 cells; ATCC® CRL-1586™

²Grown in Eagle's Minimum Essential Medium (ATCC® 30-2003™) supplemented with 2% fetal bovine serum (ATCC® 30-2020™) for 10 days at 37°C and 5% CO₂

³RNA used for RT-PCR was extracted from infected cell lysate and supernatant after 13 days incubation at 37°C and 5% CO₂.

⁴There is no sequence information for BCNV, VH-C067 in the NCBI database. Nucleotide sequence obtained for NR-12235 is consistent with BCNV strains for which sequence data is available.

⁵The Tissue Culture Infectious Dose 50% (TCID₅₀) endpoint is the 50% infectious endpoint in cell culture. The TCID₅₀ is the dilution of virus that under the conditions of the assay can be expected to infect 50% of the culture vessels inoculated, just as a Lethal Dose 50% (LD₅₀) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the TCID₅₀ provides a measure of the titer (or infectivity) of a virus preparation.

⁶Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

Date: 12 MAR 2015

Signature: 

Title: Technical Manager, BEI Authentication or designee

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