

Certificate of Analysis for NR-50132

Bourbon Virus, Original

Catalog No. NR-50132

This reagent is the property of the U.S. Government.

Product Description: Clarified supernatant from *Cercopithecus aethiops* kidney epithelial cells (Vero E6)¹ infected with Bourbon virus (BRBV), Original

Passage History: V1/V2 (Centers for Disease Control/BEI Resources); V# = Number of passages

in Vero cells

Lot²: 64077566 Manufacturing Date: 15APR2016

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero E6 Cells ²	Report results	Cell rounding and detachment
Sequencing of Species-Specific Region (672 nucleotides)	Consistent with BRBV	100% identity with BRBV (GenBank: KP657749)
Titer by TCID ₅₀ Assay ³ in Vero E6 Cells ⁴	Report results	$2.8 \times 10^7 \text{ TCID}_{50} \text{ per mL}$
Sterility (21-day incubation) Harpo's HTYE broth ⁵ , 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
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™

Date: 20 JUN 2016

Signature: ////

BEI Resources Authentication

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²Grown in Dulbecco's Modified Eagle's Medium (ATCC® 30-2002™) supplemented with 2% Fetal Bovine Serum (ATCC® 30-2020™) for 3 days at 37°C and 5% CO₂

³The 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.

⁴5 days at 37°C and 5% CO₂

⁵Atlas, Ronald M. <u>Handbook of Microbiological Media</u>. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798