

Bat SARS-Like Coronavirus, HKU5, Recombinant, Containing the SARS Coronavirus, Urbani Spike Glycoprotein Ectodomain

Catalog No. NR-48814

Product Description: Cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial cells¹ infected with synthetic recombinant bat severe acute respiratory syndrome (SARS)-like coronavirus (CoV), HKU5, containing the spike glycoprotein ectodomain of SARS-CoV, Urbani

Passage History: Submission laboratory: Vero (3); BEI Resources: Vero¹ (2)

Lot²: 63140622

Manufacturing Date: 02APR2015

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero Cells¹	Syncytial rounding and sloughing	Syncytial rounding and sloughing
Sequencing of Strain-Specific Regions Polyprotein (orf1ab) gene (878 nucleotides) Spike (S) glycoprotein gene (716 nucleotides)	Consistent with bat-CoV, HKU5 Consistent with SARS-CoV, MA15	99% identity with bat-CoV, HKU5 (GenBank: EF065512) 100% identity with SARS-CoV, MA15 (GenBank: DQ497008) ³
Titer by TCID₅₀ Assay in Vero Cells^{1,4,5}	Report results	8.9 x 10 ⁷ TCID ₅₀ per mL
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
Mycoplasma Contamination Agar and broth culture (14-day incubation at 37°C) DNA detection by PCR of extracted Test Article nucleic acid	None detected None detected	None detected None detected

¹Vero 76, clone E6 cells: ATCC® CRL-1586™

²Grown in Dulbecco's Modified Eagle's Medium (ATCC® 30-2002) supplemented with 5% fetal bovine serum (ATCC® 30-2020) for 2 days at 37°C and 5% CO₂

³Alignment of the S gene sequence of NR-48814 with the published sequence of the mouse-adapted SARS-CoV, MA15 strain confirmed both the SARS, Urbani origin of the S gene and the presence of the histidine for tyrosine substitution at position 436, previously shown to enhance replication in mice.

⁴6 days at 37°C and 5% CO₂

⁵TCID₅₀ (50% tissue culture infectious dose): The TCID₅₀ is the 50% infectious endpoint in tissue 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. A reciprocal of the dilution required to yield the TCID₅₀ provides a measure of the titer 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: 14 DEC 2015

Signature: 

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