

Certificate of Analysis for NR-48813

Middle East Respiratory Syndrome Coronavirus, Recombinant Infectious Clone with ORF5 Red Fluorescent Protein Replacement (icMERS-CoV-RFP-△ORF5)

Catalog No. NR-48813

Product Description: Cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial cells¹ infected with Middle East respiratory syndrome coronavirus, recombinant infectious clone with ORF5 red fluorescent protein replacement (icMERS-CoV-RFP-ΔORF5)

Passage History: Submission laboratory: Vero (3); BEI Resources: Vero (7)

Lot²: 63140603 Manufacturing Date: 27SEP2015

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in Vero Cells ¹	Syncytial rounding and sloughing	Syncytial rounding and sloughing
Identification by Direct Red Fluorescence	Fluorescence observed	Fluorescence observed
Sequencing of Strain-Specific Region (1132 nucleotides)	Consistent with MERS-CoV	100% identity with MERS-CoV, EMC/2012 (GenBank: JX869059)
Titer by TCID ₅₀ Assay in Vero Cells ^{1,3,4}	Report results	$8.9 \times 10^7 TCID_{50} per mL$
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 extracted Test Article nucleic acid	None detected None detected	None detected None detected

¹Vero cells: ATCC® CCL-81™

Date: 18 DEC 2015

Signature:

BEI Resources Authentication

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BEI Resources www.beiresources.org E-mail: contact@beiresources.org
Tel: 800-359-7370

Fax: 703-365-2898

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

³TCID₅₀ (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.

⁴6 days at 37°C and 5% CO₂

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