Certificate of Analysis for NR-52287

SARS-Related Coronavirus 2, Isolate USA-WA1/2020, Gamma-Irradiated

Catalog No. NR-52287

Product Description:

NR-52287 is a gamma-irradiated preparation of severe acute respiratory syndrome-related coronavirus 2 (SARS-CoV-2), isolate USA-WA1/2020. NR-52287 was gamma-irradiated (5 × 10^6 RADs) on dry ice, followed by sonication to clarify the supernatant prior to vialing.

The starting material (BEI Resources lot 70047442) was produced by infecting *Chlorocebus aethiops* kidney cells (Vero E6; ATCC® CRL-1586™) in Eagle's Minimum Essential Medium (ATCC® 30-2003™) supplemented with 2% fetal bovine serum (ATCC® 30-2020™) for 4 days at 37°C with 5% CO_2.

Lot: 70069600 **Manufacturing Date: 07AUG2024**

TEST	SPECIFICATIONS	RESULTS
Pre-Inactivation Titer by $TCID_{50}$ Assay in Vero E6 Cells[1]	Report results	7.9 × 10^5 $TCID_{50}$ per mL at 8 days at 37°C and 5% CO_2
Pre-Inactivation Sterility (21-day incubation)		
Harpo's HTYE broth, 37°C and 26°C, aerobic[2]	No growth	No growth
Trypticase soy broth, 37°C and 26°C, aerobic	No growth	No growth
Sabouraud broth, 37°C and 26°C, aerobic	No growth	No growth
Sheep blood agar, 37°C, aerobic	No growth	No growth
Sheep blood agar, 37°C, anaerobic	No growth	No growth
Thioglycollate broth, 37°C, anaerobic	No growth	No growth
DMEM with 10% FBS, 37°C and 5% CO_2	No growth	No growth
Pre-Inactivation Mycoplasma Contamination		
Agar and broth culture (14-day incubation at 37°C)	None detected	None detected
DNA detection by PCR of extracted Test Article nucleic acid	None detected	None detected
Genome Copy Number Using BioRad QX200 Droplet Digital PCR (ddPCR™) System[3] (15 replicates)	Report results	1.52 × 10^9 genome copies per mL
Virus Inactivation 10% of total bulk gamma-irradiated preparation inoculated on Vero E6-TMPRSS2-T2A-ACE2 cells and evaluated for cytopathic effect and expression of viral antigen by indirect immunofluorescence assay (IFA)[4,5]	No viable virus detected	No viable virus detected

[1]The Tissue Culture Infectious Dose 50% ($TCID_{50}$) endpoint is the 50% infectious endpoint in cell culture. The $TCID_{50}$ 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% (LD_{50}) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the $TCID_{50}$ provides a measure of the titer (or infectivity) of a virus preparation.
[2]Atlas, Ronald M. Handbook of Microbiological Media. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.
[3]The genome copy number reported is obtained using Qiagen RNA extraction kit (Cat 52906). The extraction efficiency of the kit used will influence the final quantitation results.
[4]The gamma-irradiated virus preparation was plated on Vero E6-TMPRSS2-T2A-ACE2 cells and incubated for 14 days at 37°C and 5% CO_2; cell lysate and supernatant from these cultures were blind passaged on fresh monolayers of Vero E6-TMPRSS2-T2A-ACE2 cells and again incubated for 14 days at 37°C and 5% CO_2.
[5]Indirect immunofluorescence assay (IFA) was performed at University of Texas Medical Branch, Galveston, Texas, USA

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/Sonia Bjorum Brower/
Sonia Bjorum Brower 30 MAY 2025
Technical Manager or designee, ATCC Federal Solutions

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected by ATCC® or the contractor to the tests and procedures specified and that the results described, along with any other data provided in this certificate, are true and accurate to the best of ATCC®'s knowledge.

Support Provided by NIAID

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