

Certificate of Analysis for NR-55245

SARS-Related Coronavirus 2, Isolate USA/CA_CDC_5574/2020, Heat Inactivated

Catalog No. NR-55245

Product Description:

NR-55245 is a preparation of SARS-Related Coronavirus 2 (SARS-CoV-2), isolate USA/CA_CDC_5574/2020 that has been inactivated by heating to 65°C for 30 minutes, followed by 5-fold dilution prior to vialing.

The starting material (BEI Resources NRC-54022 lot 70043446) was produced by infecting *Cercopithecus aethiops* kidney cells (Vero E6; ATCC® CCL-81[™]) in Eagle's Minimum Essential Medium (ATCC® 30-2003[™]) supplemented with 2% fetal bovine serum (ATCC® 30-2020[™]) for 5 days at 37°C with 5% CO₂. Titer of spin clarified NRC-54022 was 5.0×10^4 TCID₅₀ per mL by TCID₅₀ assay in Vero E6 cells, determined by cytopathic effect in 5 days at 37°C with 5% CO₂, and sterility of the preparation was confirmed.¹

Lot: 70043111 Manufacturing Date: 24MAR2021

TEST	SPECIFICATIONS	RESULTS
Genome Copy Number Using BioRad QX200 Droplet Digital PCR (ddPCR™) System²	Report results	3.24 × 10 ⁸ genome equivalents per mL ³
Virus Inactivation		
10% of total bulk heat-treated preparation inoculated on Vero E6 cells and evaluated for cytopathic effect and presence of viral RNA by qRT-PCR ⁴	No viable virus detected	No viable virus detected

¹The Tissue Culture Infectious Dose 50% (TCID₅₀) 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.

/Heather Couch/

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Program Manager or designee, ATCC Federal Solutions

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²The genome copy number reported is obtained using Qiagen RNA extraction kit (Cat 52904).

³Two vials of NR-55245 lot 70043111 were used for RNA extraction. Average of the results obtained from nine replicates from one vial and eight replicates from the other is reported.

⁴The inactivated virus preparation was plated on Vero E6 cells and incubated for 14 days at 37°C and 5% CO₂; cell lysate and supernatant from these cultures were blind passaged on fresh monolayers of Vero E6 cells and again incubated for 14 days at 37°C and 5% CO₂. Samples from both passages were also tested by qPCR at the end of day 14 of the passages.