

Certificate of Analysis for NR-10176

Pirital Virus, VAV-488

Catalog No. NR-10176

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

Product Description: Cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial cells (Vero E6)¹ infected with Pirital virus, VAV-488

Lot²: 60505363 Manufacturing Date: 21JUN2013

| TEST | SPECIFICATIONS | RESULTS |
|---|---|---|
| Identification by Infectivity in Vero E6 Cells ¹ Using RT-PCR ³ | Report results | Infectious, no cytopathic effect |
| Sequencing of Species-Specific Region (335 nucleotides) | Consistent with Pirital virus, VAV-488 | 99% identity with Pirital virus, VAV-488 (GenBank: AF485262) |
| Titer by TCID ₅₀ Assay ⁴ in Vero E6 Cells ¹ Using RT-PCR ³ | Report results | 8.9×10^5 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 Test Article nucleic acid | None detected None detected | None detected None detected |

¹Vero E6 cells; ATCC[®] CRL-1586™

Date: 18 MAR 2014 Signature: Michael Q. Gypthe

Title: Technical Manager, BEI Authentication or designee

ATCC®, on behalf of BEI Resources, hereby represents and warrants that the material provided under this certificate has been subjected 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.

ATCC® is a trademark of the American Type Culture Collection.

You are authorized to use this product for research use only. It is not intended for human use.

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

Fax: 703-365-2898

²Grown in Eagle's Minimum Essential Medium (ATCC[®] 30-2003[™]) supplemented with 2% fetal bovine serum (ATCC[®] 30-2020[™]) for 14 days at 37°C and 5% CO₂

³RNA used for RT-PCR was extracted from infected cell lysate and supernatant after 13 days incubation at 37°C and 5% CO₂.

⁴The 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.

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