b|e|i RESOURCES

SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-52318

Enterovirus Species D Type 111, ANG/2010-23294

Catalog No. NR-52318

Product Description:

Enterovirus species D type 111 (EV-D111), ANG/2010-23294 was isolated in 2010 from a stool sample from a human subject in Angola. The subject was not suffering from acute flaccid myelitis (AFM). NR-52318 lot 70043450 was produced by infecting rhabdomyosarcoma cells (RD; ATCC® CCL-136™) and incubating in Dulbecco's Modified Eagle's Medium (ATCC® 30-2002™) supplemented with 2% fetal bovine serum (ATCC® 30-2020™) for 8 days at 37°C with 5% CO_2. It was freeze-thawed three times before dispensing lot 70043450.

Passage History:

RD(3)/RD(2) (Prior to deposit at BEI Resources/BEI Resources); RD = rhabdomyosarcoma cells

Lot: 70043450 **Manufacturing Date: 28APR2021**

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in RD Cells	Cell rounding and detachment	Cell rounding and detachment
Sequencing of Species-Specific Region (~ 840 nucleotides)	≥ 98% identity with EV-D111, ANG/2010-23294 (GenBank: MT081371.1)	100% identity with EV-D111, ANG/2010-23294 (GenBank: MT081371.1)
Titer by $TCID_{50}$ Assay in RD Cells by Indirect Fluorescent Antibody[1,2] (12 days at 37°C with 5% CO_2)	Report results	5.0×10^4 $TCID_{50}$ per mL
Sterility (21-day incubation)		
Harpo's HTYE broth, 37°C and 26°C, aerobic[3]	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, aerobic	No growth	No growth
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

[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]Using Pan-enterovirus (Millipore 3360)
[3]Atlas, Ronald M. Handbook of Microbiological Media. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

/Heather Couch/
Heather Couch 15 DEC 2021
Program 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 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.

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BEI Resources
www.beiresources.org
E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898