

Ectromelia Virus Expressing Murine Interleukin-4

Catalog No. NR-791

Product Description:

Ectromelia virus (ECTV) expressing murine interleukin-4 (IL-4) was prepared by recombining the murine IL-4 gene, under the control of the 11K late promoter of vaccinia virus, into the Chinese hamster ovary cell host-range gene of ECTV, Moscow. This strain produces a lethal infection in mouse strains that are genetically resistant to ECTV, Moscow. NR-791 lot 70026737 was produced by infecting *Cercopithecus aethiops* cells (BS-C-1; ATCC® CCL-26™) and incubating in Eagle's Minimum Essential Medium (ATCC® 30-2003) supplemented with 2% fetal bovine serum (ATCC® 30-2020) for 2 days at 37°C with 5% CO₂. The first three virus passages were in the presence of Mycoplasma Removal Agent (MRA; MP Biomedicals 3050044) to remove contaminating mycoplasma.

Passage History:

X(?)/B(6) (Prior to deposit at BEI Resources/BEI Resources); X = Unknown; B = BS-C-1 cells

Lot: 70026737

Manufacturing Date: 08AUG2019

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in BS-C-1 Cells	Cell rounding and detachment	Cell rounding and detachment
Sequencing of Hemagglutinin (HA) Gene (590 nucleotides)	≥ 98% identity with ECTV, Moscow (GenBank: AF012825.2)	100% identity with ECTV, Moscow (GenBank: AF012825.2)
Confirmation of IL-4 Gene by PCR Amplification of Extracted DNA	~ 680 base pair amplicon	~ 680 base pair amplicon
Titer by TCID₅₀ Assay in BS-C-1 Cells by Cytopathic Effect¹	Report results	2.8 × 10 ⁶ TCID ₅₀ per mL in 4 days at 37°C with 5% CO ₂
Sterility (21-day incubation) Harpo's HTYE broth, 37°C and 26°C, aerobic ² 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 No growth No growth No growth No growth No growth	No growth No growth No growth No growth No growth 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

¹The 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.

²Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

/Heather Couch/

Heather Couch

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Program Manager or designee, ATCC Federal Solutions

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