

Porcine Transmissible Gastroenteritis Virus (TGEV), Miller

Catalog No. NR-447

Product Description: Cell lysate and supernatant from swine testicular (ST) cells¹ infected with porcine TGEV, Miller.

Lot^{2,3}: 57689881

Manufacturing Date: 30MAY2008

TEST	SPECIFICATIONS	RESULTS
Identification by Infectivity in ST Cells ¹	Report results	Cell rounding and sloughing
Identification by Direct Fluorescent Antibody Assay ⁴	Fluorescence observed	Fluorescence observed
Sequencing of Species-Specific Region	Consistent with TGEV	Consistent with TGEV
Titer by TCID ₅₀ Assay ^{5,6} in ST Cells ¹	Report results	8.89 X 10 ⁶ TCID ₅₀ /mL
Functional Activity by RT-PCR Assay	~ 430 bp amplicon	~ 430 amplicon
Sterility (21-day incubation) Harpo's HTYE broth ⁷ , 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 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

¹ST cells: ATCC® CRL-1746™

²The inoculum for this lot was BEI Resources NR-447 (Lot 4462784), which was determined by PCR to be contaminated with *Mycoplasma orale*. Upon three passages in the presence of 0.5 µg/mL mycoplasma removal agent (MP 30-500-44), PCR tests showed the live virus to be clean and free of mycoplasma contamination. Source virus for NR-447 (Lot 4462784) was prepared in ST cells and provided by the Food Animal Health Research Program, Ohio Agricultural Research and Development Center, The Ohio State University, Wooster, Ohio.

³Grown in Minimum Essential Medium containing Earle's salts and non-essential amino acids (Invitrogen™ 10370-021), 2 mM L-glutamine (Invitrogen™ 25030-081), and 1 mM sodium pyruvate (Invitrogen™ 11360-070) for 3 days at 37°C with 5% CO₂

⁴Using fluorescein-isothiocyanate conjugate monoclonal antibody specific to TGEV (VMRD 210-50-TGE)

⁵The 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.

⁶2 days at 37°C with 5% CO₂

⁷Atlas, Ronald M. *Handbook of Microbiological Media*. 3rd ed. Ed. Lawrence C. Parks. Boca Raton: CRC Press, 2004, p. 798.

Date: 28AUG2008

Signature: Signature on file

Title: Technical Manager, BEI Authentication or designee

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