

## **Certificate of Analysis for NR-4289**

## Genomic RNA from Dengue Virus Type 4, H241 (Tissue Culture Adapted)

## Catalog No. NR-4289

**Product Description:** Genomic RNA was isolated from a preparation of cell lysate and supernatant from Rhesus monkey kidney (LLC-MK2 derivative) cells<sup>1</sup> infected with dengue virus type 4 (DEN-4), H241 (tissue culture adapted). RT-PCR amplification of a sequence of approximately 1200 nucleotides was successful with dilutions of up to 10-fold for this lot of NR-4289.

Lot<sup>2</sup>: 57558393 Manufacturing Date: 19AUG2009

TEST	SPECIFICATIONS	RESULTS
Sequencing of DEN-4 Specific Sequence (~ 980 nucleotides)	Consistent with DEN-4	Consistent with DEN-4
Functional Activity by RT-PCR Amplification <sup>3</sup>	~ 1200 bp amplicon	~ 1200 bp amplicon (Figure 1)
Total RNA Content by RiboGreen <sup>®</sup> Measurement (Viral, Cellular, and Carrier)	Report results	36.4 ng per 100 µL
Virus Inactivation  10% of total yield inoculated on LLC-MK2 derivative cells and evaluated for immunofluorescence <sup>1,4</sup>	No viable virus detected	No viable virus detected
Sodium Azide Content	Report results	0.0004%

<sup>&</sup>lt;sup>1</sup>LLC-MK2 derivative cells: ATCC<sup>®</sup> CCL-7.1™

**Date:** 06 FEB 2015

Signature: Miller G. Gy

**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.

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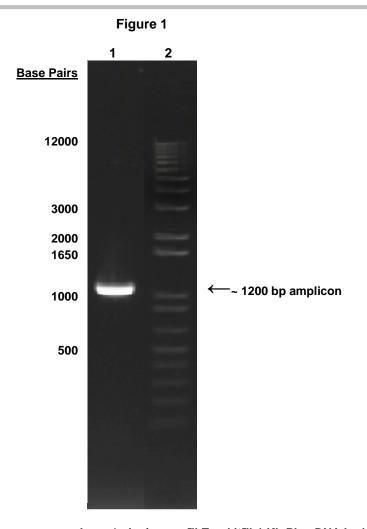
<sup>&</sup>lt;sup>2</sup>Nucleic acid was extracted from a preparation of DEN-4, H241 (tissue culture adapted) using a QIAGEN QIAamp<sup>®</sup> Viral RNA Mini Kit (Qiagen 52906).

<sup>&</sup>lt;sup>3</sup>Amplified using a 1:10 dilution of NR-4289 and a QIAGEN One-Step RT-PCR Kit (210212)

<sup>&</sup>lt;sup>4</sup>This extraction procedure has been shown to consistently inactivate 100% of dengue viruses using a number of methods designed to detect the virus in cells: 1) cytopathic effect (100% plating, 10% plating, blind passage sub-culturing) and 2) indirect immunofluorescence of viral antigens.



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Lane 1: Invitrogen™ TrackIt™ 1 Kb Plus DNA Ladder

Lane 2: 10-fold dilution of NR-4289

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