

## **Certificate of Analysis for NR-50530**

## Genomic RNA from Dengue Virus Type 1, UIS 998

## Catalog No. NR-50530

## **Product Description:**

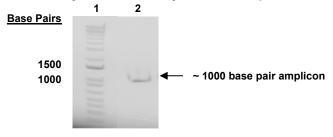
Genomic RNA was isolated from a preparation of cell lysate and supernatant from *Aedes albopictus* mosquito larval epithelial cells (clone C6/36; ATCC<sup>®</sup> CRL-1660<sup>™</sup>) infected with dengue virus type 1 (DEN-1), UIS 998 (BEI Resources lot 70047467) using QIAamp<sup>®</sup> Viral RNA Mini Kit (Qiagen<sup>®</sup> 52906). The viral genomic RNA is in a background of cellular nucleic acid and carrier RNA.

Lot: 70053410 Manufacturing Date: 13JUN2022

TEST	SPECIFICATIONS	RESULTS
Genotypic Analysis Sequencing of species-specific region (~ 900 nucleotides)	≥ 98% identity with DEN-1	99.1% identity with DENV-1/ CO/BID-V3390/2007 (GenBank: GQ868569) <sup>1</sup>
Functional Activity by RT-PCR Amplification <sup>2</sup>	~ 1000 base pair amplicon	~ 1000 base pair amplicon (Figure 1)
Estimated Concentration (post-dilution) by RiboGreen® Measurement (Viral, Cellular and Carrier) <sup>3</sup>	Report results	0.9 ng per 100 µL (0.009 µg per mL)
Estimated Amount per Vial <sup>3</sup>	Report results	0.9 ng
Virus Inactivation 10% of total yield inoculated on C6/36 cells and evaluated for cytopathic effect and fluorescent antibody after serial passage <sup>4</sup>	No viable virus detected	No viable virus detected

<sup>&</sup>lt;sup>1</sup>Sequence information for DEN-1, UIS 998 is not available in the NCBI database; nucleotide sequence obtained for lot 70053410 is ~ 99% identical to DENV-1/CO/BID-V3390/2007 (GenBank: GQ868569), a DEN-1 strain that was also isolated in Santander, Colombia in 2007.

Figure 1: Functional Activity of NR-50530 by RT-PCR Amplification



Lane 1: Invitrogen™ TrackIt™ 1 Kb Plus DNA Ladder Lane 2: PCR product from 1 µL of NR-50530

/Sonia Bjorum Brower/ Sonia Bjorum Brower

19 JAN 2023

Technical Manager or designee, ATCC Federal Solutions

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<sup>&</sup>lt;sup>2</sup>Amplified using iTaq™ Universal SYBR Green One-step Kit (Bio-Rad<sup>®</sup> 172-5151) with 5 µL of NR-50530 in a 50 µL reaction

<sup>&</sup>lt;sup>3</sup>Measurement is determined pre-vial prior to dilution due to the limit of detection of the quantification method

<sup>&</sup>lt;sup>4</sup>Use of the QIAamp<sup>®</sup> Viral RNA Mini Kit has been demonstrated to consistently inactivate 100% of DENV-1 as shown by the absence of cytopathic effect (CPE) and fluorescent antibody using Millipore mouse anti-dengue complex (cat no. MAB8705) after plating the entire extract on virus-susceptible cells for two passages.