

## **Certificate of Analysis for NR-50369**

## Zika Virus, PRVABC59, Heat-Inactivated

Catalog No. NR-50369

**Product Description:** Clarified cell lysate and supernatant from Vero E6 cells infected with Zika virus, PRVABC59 that has been inactivated by heating to 65°C for 30 minutes

Lot: 64493946 Manufacturing Date: 28JUN2016

TEST	SPECIFICATIONS	RESULTS
Pre-Inactivation Titer by TCID <sub>50</sub> Assay in Vero E6 Cells <sup>1,2</sup>	Report results	2.8 x 10 <sup>7</sup> TCID <sub>50</sub> per mL
Pre-Inactivation Sterility iNST culture bottle, 32°C, anaerobic, 14-day incubation iAST culture bottle, 32°C, aerobic, 14-day incubation	No growth No growth	No growth No growth
Pre-Inactivation Mycoplasma Contamination Agar and broth culture (14-day incubation at 37°C) DNA Detection by PCR of Test Article nucleic acid	None detected None detected	None detected None detected
Viral Genome Copy Number Droplet Digital RT-PCR Quantitative Real-Time RT-PCR	Report results Report results	$4.6 \times 10^6$ copies per $\mu$ L $1.1 \times 10^7$ copies per $\mu$ L
Altona Diagnostics RealStar® Zika Virus RT-PCR Test	Zika virus RNA detected	Zika virus RNA detected
CDC Trioplex Real-Time RT-PCR Assay Chikungunya virus Dengue virus Zika virus	None detected None detected Detected	None detected None detected Detected
Virus Inactivation  10% of total bulk heat-treated preparation inoculated on Vero E6 cells and evaluated for cytopathic effect, viral antigen expression by indirect immunofluorescence assay, and presence of viral RNA by real-time RT-PCR <sup>3</sup>	No viable virus detected	No viable virus detected
Amplification of Zika Virus NS3/NS4 Coding Region by RT-PCR Pre-inactivation Post-Inactivation <sup>4</sup>	Report results Report results	~ 1100 bp amplicon None detected

¹The Tissue Culture Infectious Dose 50% (TCID<sub>50</sub>) is the 50% infectious endpoint in cell culture. The TCID<sub>50</sub> 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<sub>50</sub>) is expected to kill half of the animals exposed. A reciprocal of the dilution required to yield the TCID<sub>50</sub> provides a measure of the titer (or infectivity) of a virus preparation. <sup>27</sup> days at 37°C and 5% CO.

**Date:** 21 OCT 2016

Signature:

**BEI Resources Authentication** 

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<sup>&</sup>lt;sup>3</sup>The inactivated virus preparation was plated on Vero E6 cells and incubated for 14 days at 37°C and 5% CO<sub>2</sub>; cell lysate and supernatant from these cultures was blind passaged on fresh monolayers of Vero E6 cells and again incubated for 7 days at 37°C and 5% CO<sub>2</sub>.

<sup>&</sup>lt;sup>4</sup>NR-50369 does not contain intact Zika virus genomić RNA, and should not be used as a source of nucleic acid for transfection or for RT-PCR of "gene-length" amplicons.