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SUPPORTING INFECTIOUS DISEASE RESEARCH

Certificate of Analysis for NR-51219

Genomic RNA from Usutu Virus, SAAR 1776

Catalog No. NR-51219

Product Description:

Genomic RNA was extracted from a preparation of cell lysate and supernatant from *Cercopithecus aethiops* kidney epithelial cells infected with Usutu virus (USUV), SAAR 1776.[1] USUV, SAAR 1776 was isolated in 1959 from a mosquito *(Culex univittatus)* in Ndumo, Natal, South Africa.

Lot: 70021062[2] **Manufacturing Date: 21JAN2019**

TEST	SPECIFICATIONS	RESULTS
Genotypic Analysis Sequencing of species-specific region (~ 980 nucleotides)	≥ 98% identity with USUV, SAAR 1776 complete genome (GenBank: AY453412)	99.5% identity with USUV, SAAR 1776 complete genome (GenBank: AY453412)
Functional Activity by RT-PCR Amplification[3] NS3-NS4 gene	~ 1100 base pair amplicon	~ 1100 base pair amplicon
Pre-Vial Concentration by RiboGreen® Measurement (Viral, Cellular and Carrier)[4]	Report results	3.9 ng per 100 µL (0.04 µg/mL)
Estimated Amount per Vial[4]	Report Results	3.9 ng
Virus Inactivation 10% of total yield inoculated on Vero cells and evaluated for cytopathic effect after serial passage[1,5]	No viable virus detected	No viable virus detected

[1]Vero cells: ATCC® CCL-81™
[2]Nucleic acid was extracted from a preparation of USUV, SAAR 1776 (BEI Resources NR-51184 lot 70014388), using a QIAamp® Viral RNA Mini Kit (Qiagen 52906).
[3]Amplified using 5 µL of NR-51219 in a 50 µL reaction
[4]Measurement is determined pre-vial prior to dilution due to the limit of detection of the quantification method
[5]Use of the QIAamp® Viral RNA Mini Kit has been demonstrated to consistently inactivate flaviviruses as shown by the absence of cytopathic effect (CPE) and viral RNA expression by RT-PCR after plating the entire extract on virus-susceptible cells for two passages.

/Heather Couch/
Heather Couch 16 SEP 2019
Program Manager or designee, ATCC Federal Solutions

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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