b|e|i RESOURCES
SUPPORTING INFECTIOUS DISEASE RESEARCH

# Certificate of Analysis for NR-17606

## Vaccinia Virus, Western Reserve Genome, VAC(WR)-LoxP-GFP-BAC/Zeo, Recombinant in *Escherichia coli*

## Catalog No. NR-17606

This reagent is the property of the U.S. Government.

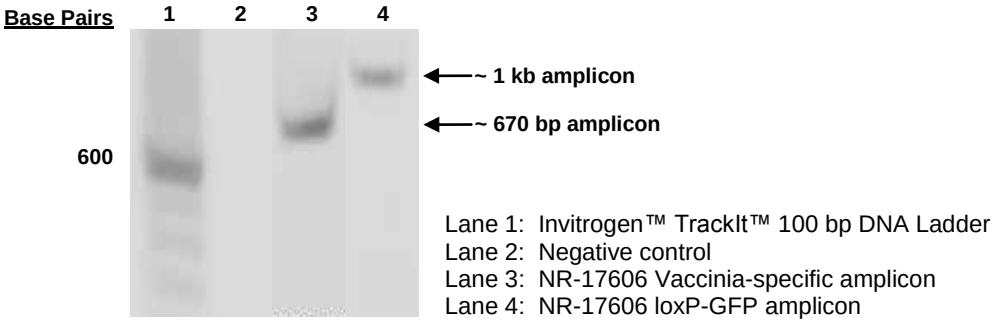
**Product Description:** The entire vaccinia virus (VACV) Western Reserve (WR) genome with a green fluorescent protein (GFP) sequence, two loxP sites, and a zeomycin resistance gene was cloned as a bacterial artificial chromosome (BAC) and grown in *Escherichia coli* DH10β cells harboring a mini-lambda prophage.[1]

**Lot: 61922857** **Manufacturing Date: 28AUG2013**

| TEST | SPECIFICATIONS | RESULTS |
|---|---|---|
| **Functional Activity by PCR Amplification**[2] | Vaccinia-specific amplicon, region of high variability (~670 bp)<br>GFP amplicon (~1 Kb) | Vaccinia-specific amplicon, region of high variability (~670 bp) (Figure 1)<br>GFP amplicon (~1 Kb) (Figure 1) |
| **Sequencing of Vaccinia-Specific Region (655 bp)** | Consistent with VACV WR | 100% identity with VACV WR (Genbank: AY243312) |
| **Sequencing of GFP Amplicon (829 bp)** | Consistent with GFP gene | 100% identity with EGFP sequence from pEGFP.N1 vector (Genbank U55762) |
| **Viability (post-vialing)**[3] | Growth | Growth |

[1]Grown in Luria-Bertani (LB) broth containing 25 µg/mL Zeocin™ (Invitrogen R25001) at 32°C in an aerobic atmosphere with shaking at 200 rpm
[2]Plasmid extracted using QIAprep Spin Miniprep Kit (QIAGEN® 27104) and amplified using iTaq™ DNA Polymerase (Bio-Rad 170-8870) and dNTP Mix (Bio-Rad 170-8874)
[3]Cultured overnight in LB broth containing 25 µg/mL Zeocin™ at 32°C in an aerobic atmosphere with shaking at 180 rpm, and on nutrient agar plates containing 25 µg/mL Zeocin™ at 32°C in an aerobic atmosphere

**Figure 1**




Lane 1: Invitrogen™ TrackIt™ 100 bp DNA Ladder
Lane 2: Negative control
Lane 3: NR-17606 Vaccinia-specific amplicon
Lane 4: NR-17606 loxP-GFP amplicon

**Date:** 14 NOV 2014 **Signature:** Michael R. Gyrla

**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.

ATCC® is a trademark of the American Type Culture Collection.
You are authorized to use this product for research use only. It is not intended for human use.

Support Provided by NIAID

**BEI Resources**
www.beiresources.org

E-mail: contact@beiresources.org
Tel: 800-359-7370
Fax: 703-365-2898