



**Lassa Pseudovirus**  
**Mouse/SierraLeone/Josiah/1976 Variant**

**Luciferase reporter**

**Lot #250804**



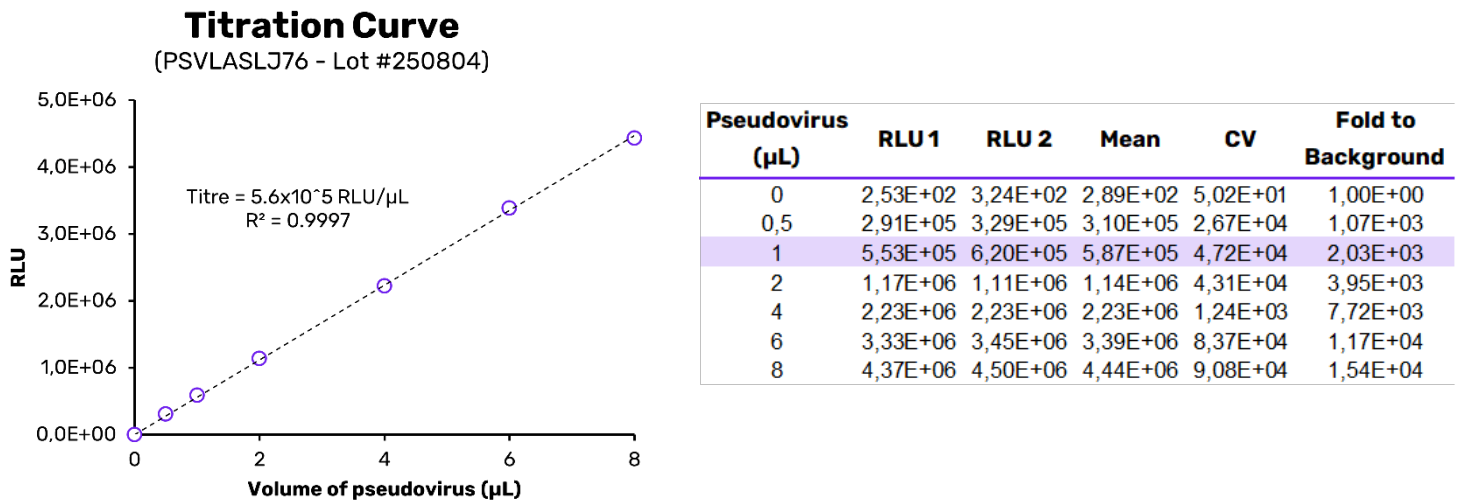
**Certificate of Analysis**

# 1. Summary

This certificate provides a functional validation of the Lassa pseudovirus, Mouse/SierraLeone/Josiah/1976, lot #250804. The titer is  $5.6 \times 10^5$  RLU/ $\mu$ L. A 1,000-fold-to-background ratio is obtained with 1  $\mu$ L per well (96-well plate). Hence, a volume of 1 mL is sufficient to perform approximately 1,000 reactions, or  $10 \times$  96-well plates. According to IVANO Bioscience’s protocol, available upon request.

# 2. Transduction efficiency assay

**Target cells** HEK293T cells  
**Volume of pseudovirus** 0 – 0.5 – 1 – 2 – 4 – 6 – 8 – 10  $\mu$ L/well  
**Detection signal** Luminescence (firefly luciferase)  
**Detection method** Microplate reader Biotek Synergy H1 (Gain: 135)



**Figure 1: Transduction efficiency curve**

A volume range of pseudovirus was mixed in a final volume of 50  $\mu$ L of medium, in a 96-well plate. Then, 50  $\mu$ L of medium containing 10,000 cells was seeded in each well. On the day of analysis, an additional 100  $\mu$ L of Bright-Glo Luciferase reagent was added in each well and incubated for 2 minutes. Data in relative unit luminescence (RLU) were obtained from the analysis of 150  $\mu$ L of the cell lysate, using a microplate reader. Data are expressed in relative unit luminescence (RLU).

**Conclusion**

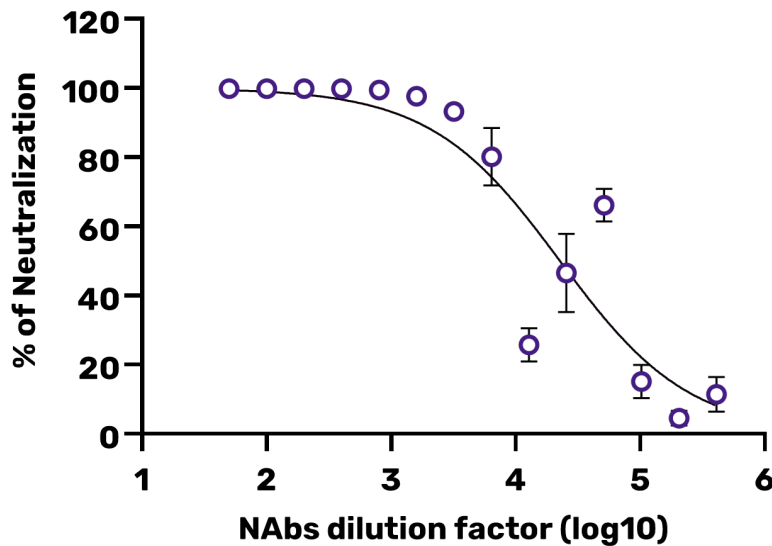
The Lassa pseudovirus, Mouse/SierraLeone/Josiah/1976 variant (lot #250804), is capable of transducing target cells. The titer is  $5.6 \times 10^5$  RLU/ $\mu$ L. Using 1 microliters of pseudovirus per reaction in a 96-well plate results in a 1,000-fold increase in RLU compared to the background. Accordingly, 1 mL of lot #250804 can be used to perform approximately 1,000 reactions, or  $10 \times$  96-well plates, according to IVANO Bioscience’s protocol (available upon request)  
 Note that a volume of 10  $\mu$ L of pseudovirus provided an overflow signal upon analysis.

### 3. Neutralization assay

<b>Target cells</b>	HEK293T cells
<b>Volume of pseudovirus</b>	1 $\mu$ L/well
<b>Neutralizing antibody (Nabs)</b>	Anti-Glycoprotein Complex [37.7H] , Ab01102-10.0
<b>Detection signal</b>	Luminescence (firefly luciferase)
<b>Detection method</b>	Microplate reader Biotek Synergy H1 (Gain: 135)

#### Neutralization curve

(PSVLASLJ76 - Lot #250804)



**Figure 2: Neutralization curve**

A monoclonal neutralizing antibody (Ab01102-10.0), at a starting dilution of 1/50, was serially diluted in a final volume of 50  $\mu$ L of complete medium and incubated for 1 hour at 37  $^{\circ}$ C, with 1  $\mu$ L of pseudovirus, in a 96-well plate. Then, an additional 50  $\mu$ L containing 10,000 cells was seeded in each well and incubated for 72 hours. Finally, an additional 100  $\mu$ L of Bright-Glo<sup>TM</sup> Luciferase buffer was added in each well and incubated for 2 minutes. Data in relative unit luminescence (RLU) were obtained from the analysis of 150  $\mu$ L of the cell lysate with a microplate reader. Raw data were analyzed using a log(inhibitor) vs normalized-response (variable slope) non-linear regression model in Prism v10 (GraphPad). Percentages of neutralization were normalized considering only cells in wells as 100% neutralization and cells transduced by pseudoviruses without any NABs as 0% neutralization. Data are representative of duplicates. ).

#### Conclusion

The Lassa pseudovirus, Mouse/SierraLeone/Josiah/1976 variant (lot #250804) can be efficiently neutralized by neutralizing antibodies.

## 4. Additional information

<b>Intruction of use</b>	We recommend determining the titer in your lab's conditions before performing any experiments. Handle under biosafety level-2.
<b>Pseudovirus</b>	3 <sup>rd</sup> generation lentiviral vector, incompetent replication and non-toxic.
<b>Pseudotyping</b>	Glycoprotein of the Lassa pseudovirus, Mouse/SierraLeone/Josiah/1976 variant. (Uniprot Accession Number: P08669)
<b>Pseudotyping sequence</b>	MGQIVTFFQEVPHVIEEVMNIVLIALSVLAVLKGLYNFATCGLVGLVTFLLLCGRS CTTSLYKGVYELQTLELNMETLNMTMPLSCTKNNSHHYIMVGNETGLELTLTNT SIINHKFCNLSDAHKKNLYDHALMSIISTFHLSIPNFNQYEAMSCDFNGGKISVQY NLSHSYAGDAANHCGTVANGVLQTFMRMAWGGSYIALDSGRGNWDCIMTSYQ YLIIQNTTWEDHCQFSRPSPIGYLGLLSQRTRDIYISRRLGTFWTLS DSEGKDT PGGYCLTRWMLIEAELKCFGNTAVAKCNEKHDEEFCDMLRFLDFNKQAIQRLKA EAQMSIQLINKAVNALINDQLIMKNHLRDIMGIPYCNYSKYWYLNHTTTGRTSLPK CWLVSNGSYLNETHFSDDIEQQADNMITEMLQKEYMERQGKTPLGLVDFVFS TSFYLISIFLHLVKIPTHRHRHIVGKSCPKPHRLNHMGICSCGLYKQPGVPVKWKR
<b>Glycosylation origin</b>	Human
<b>Reporter Protein</b>	Firefly luciferase
<b>Storage</b>	- 80 °C, avoid freeze/thaw cycles.
<b>For more information</b>	mathias.mangion@ivanobioscience.com Message object should contain: "PSVLASLJ76 – lot #250804".
<b>Intended use</b>	For Research Use Only. Not for Use in Diagnostic Procedures. Not Meant for Resale. Not Meant for Resale.