VV_(Li)-RlucGFP-lacZ-tRFP

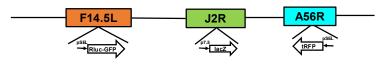


Product Name:	VV _(Li) -RlucGFP-lacZ-tRFP (Standard Titer, 0.25 mL)	
Catalog Number: Lot Number:	OV4006 OV-IM18	
Shipping conditions: Storage conditions:	Dry ice Store at ≤ -70°C upon receipt, avoid subjecting to multiple freeze-thaw cycles	
Species: Virus strain: Transgenes:	Vaccinia Lister (Li) Green fluorescent protein (GFP) Renilla luciferase (Rluc) β-galactosidase (lacZ) TurboFP635-like protein (tRFP)	
Production Cell Line: Vero		

Storage Media:1mM TrisCl, pH 9Propagation:Inoculate producer cells (e.g. Vero) at an MOIof 0.1 for 2-3 hours. After 48 hours harvestculture supernatant and cells and freeze-thawor sonicate 3X. Clarify the supernatant by low-speed centrifugation and purify through asucrose cushion. Resuspend the viral pellet in1mM TrisCl, pH 9.

VV_(Li)-RlucGFP-lacZ-tRFP is a recombinant vaccinia virus (VV) encoding the following additional transcription units: 1) the Renilla luciferase (Rluc)-Aequorea green fluorescent protein (GFP) fusion cDNA under transcriptional control of the VV synthetic early late (pSEL) promoter, 2) the β-galactosidase (lacZ) cDNA under transcriptional control of the VV early/late (p7.5) promoter, and 3) the TurboFP635-like protein (tRFP) cDNA under transcriptional control of the VV synthetic early-late (pSEL) promoter, and 3) the TurboFP635-like protein (tRFP) cDNA under transcriptional control of the VV synthetic early-late (pSEL) promoter. These additional transcription units are inserted into, and disrupt the function of, the viral F14.5L, J2R (thymidine kinase), and A56R (hemagglutinin) genes, respectively (see diagram below). The purchased virus is ready for use in cell-killing assays or can be amplified to create additional virus stocks

Genome:



References:

Zhang et al. Cancer Res 2007, 67:10038-10046 Stritzker et al, J. Virol. 2014, 88: 11556-11567 Duggal et al, J. Transl. Med. 2013, 11:155. Puhlmann, et al., Cancer Gene Therapy, 2000 7, 66–73 Gholami et al. Breast Cancer Research, 2013, 15:R26

Safety Precaution:

All culture work with Vaccinia should be performed by trained personnel and performed under BSL2 containment following NIH guidelines.

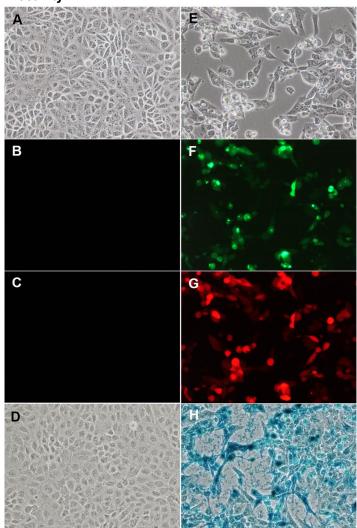


Certificate of Analysis

Testing performed by Imanis Life Sciences

Test description	Result
Titer (on Vero)	6.85x10 ⁶ TCID ₅₀ units/mL
Sterility	Pass QC
Cytopathic Effect	Pass QC
Endotoxin Test	Pass QC
Fluorescence Expression	Pass QC
Luciferase Expression	Pass QC

Infectivity



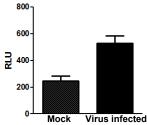
A-D: Mock-infected Vero cells.

E-H: VV_(L)-RlucGFP-lacZ-tRFP infected Vero cells (MOI 0.1) at 48 h.p.i. **D&H:** β-galactosidase activity: Cells were incubated with media containing X-Gal (5-Bromo-4-Chloro-3-Indolyl β-D-Galactopyranoside).



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Luciferase Expression



Vero cells were mock-infected or infected with VV_(Li)-RlucGFP-lacZ-tRFP at an MOI of 0.1. After 48 hours, 1.25 µg of coelenterazine was added to the wells and luminescence (RLU) was immediately measured using a microplate reader.

Quality Control by: MT Quality Assurance by: RLV Effective Date: 17-Nov-2017

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