

Vector pLVX-EF1 α -IRES-Puro Containing the Enhanced Green Fluorescent Protein

Catalog No. NR-52977

Product Description:

Note: The vial label indicates this product contains a TST tag. This nomenclature refers to a 2X Strep tag. This product does not express the Twin-Strep-tag[®] that is commonly referred to as a TST tag. The enhanced green fluorescent protein (eGFP) was codon optimized and modified by the addition of a C-terminal 2X Strep tag and cloned into the [pLVX-EF1 \$\alpha\$ -IRES-Puro](#) lentiviral expression plasmid. The vector contains an internal ribosomal entry site (IRES) that allows a gene-of-interest and a puromycin resistance gene to be simultaneously co-expressed from a single mRNA transcript. Expression of the transcript is driven by the human elongation factor 1 alpha (EF1 α) promoter. The beta-lactamase gene, *bla*, provides transformant selection through ampicillin resistance in *Escherichia coli* (*E. coli*) and the puromycin resistance gene, *pac*, provides transformant selection through puromycin resistance in eukaryotic cells. The deposited plasmid was transformed into NEB[®] Stable Competent *E. coli* cells (New England Biolabs[®] C3040H), grown in Luria-Bertani broth with ampicillin (100 μ g per mL) for 1 day at 30°C in an aerobic atmosphere, extracted using a Plasmid *Plus* Maxi Kit (QIAGEN[®] 12963) and vialled in TE buffer (10 mM Tris-HCl, 1 mM EDTA, pH 8.0).

Lot: 70037932

Manufacturing Date: 05AUG2020

TEST	SPECIFICATIONS	RESULTS
Next-Generation DNA Sequencing	~ 9640 base pairs	9642 base pairs ¹
Genotypic Analysis Sequencing of eGFP insert (~ 820 base pairs)	100% sequence identity to depositor's sequence 2X Strep tag sequence confirmed	100% sequence identity to depositor's sequence ² 2X Strep tag sequence confirmed ³
Antibiotic Resistance Ampicillin (encoded by beta-lactamase gene <i>bla</i>) ⁴ Puromycin (encoded by puromycin n-acetyltransferase gene <i>pac</i>)	<i>bla</i> sequence present <i>pac</i> sequence present	<i>bla</i> sequence present <i>pac</i> sequence present
Concentration by PicoGreen[®] Measurement	≥ 2 μ g/mL	0.2 μ g in 20 μ L per vial (12 μ g/mL)
Amount per Vial	Report results	0.2 μ g per vial
OD₂₆₀/OD₂₈₀ Ratio (pre-vial)	1.7 to 2.1	1.9
Effective Bacterial Transformation NEB [®] Stable Competent <i>E. coli</i>	≥ 50 colonies per ng	169 colonies per ng

¹The sequence was assembled pre-vial using the predicted sequence as the reference sequence. The complete plasmid sequence and map are provided on the BEI Resources webpage.

²The NR-52977 insert was codon optimized but is 100% identical with the eGFP (GenPept: AAB02572).

³This 2X Strep tag is defined by the sequence N-WSHPQFEKGGGSGGGSGGGWSHPQFEK-C. For more information, please see Busby, M., et. al. "Optimisation of a Multivalent Strep Tag for Protein Detection." *Biophys. Chem.* 152 (2010): 170-177. PubMed: 20970240.

⁴The antibiotic ampicillin degrades quickly during growth. Bacterial stationary phase should be minimized during plasmid expansion to avoid plasmid loss and increased antibiotic concentrations may be necessary.

/Heather Couch/

Heather Couch

18 OCT 2020

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 by ATCC® and the contributor 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.

