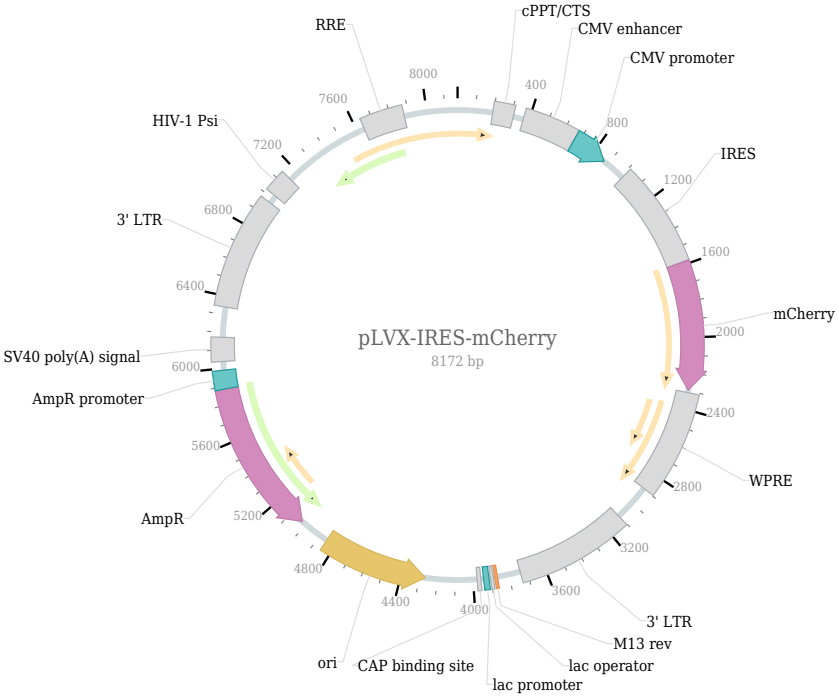
pLVX-IRES-mCherry

Vector ID	V012723
Antibiotic Resistance	Ampicillin
Length	8172 bp
Vector Type	Viral Expression & Packaging Vectors

Product Information

Replication Origin	ori
Source/Author	Clontech
Selection Marker	mCherry
Copy Number	High copy number
Promoter	CMV
Cloning Method	Enzyme Cut
Growth Strain(s)	DH10B
Growth Temperature	37℃
Expression Method	Constiutive, Stable
Comment	pLVX-IRES-mCherry is an HIV-1-based, lentiviral expression vector that allows the simultaneous expression of your protein of interest and mCherry in virtually any mammalian cell type, including primary cells. mCherry is a mutant fluorescent protein derived from the tetrameric Discosoma sp. red fluorescent protein, DsRed (1). The vector expresses the two proteins from a bicistronic mRNA transcript, allowing mCherry to be used as an indicator of transduction efficiency and a marker for selection by flow cytometry. Expression of the bicistronic transcript is driven by the constitutively active human cytomegalovirus immediate early promoter (PCMV IE) located just upstream of the MCS. An encephalomyocarditis virus (EMCV) internal ribosome entry site (IRES), positioned between the MCS and mCherry, facilitates cap-independent translation of mCherry from an internal start site at the IRES/mCherry junction (1). pLVX-IRES-mCherry contains all of the viral processing elements necessary for the production of replication-incompetent lentivirus, as well as elements to improve viral titer, transgene expression, and overall vector function. The woodchuck hepatitis virus posttranscriptional regulatory element (WPRE) promotes RNA processing events and enhances nuclear export of viral RNA (2), leading to increased viral titers from packaging cells. In addition, the vector includes a Rev-response element (RRE), which further increases viral titers by enhancing the transport of unspliced viral RNA out of the nucleus (3). Finally, pLVX-IRES-mCherry also contains a central polypurine tract/central termination sequence element (cPPT/CTS). During target cell infection, this element creates a central DNA flap that increases nuclear import of the viral genome, resulting in improved vector integration and more efficient transduction (4). The vector also contains a pUC origin of replication and an E. coli ampicillin resistance gene (Ampr) for propagation and selection in bacteria.

Vector Map



Plasmid Resuspension Protocol

1. Centrifuge at 5,000×g for 5 min.
2. Carefully open the tube and add sterile water to dissolve the DNA: add 20 µl for 5 µg plasmid, and 100 µl for 100 µg plasmid.
3. Close the tube and incubate for 10 minutes at room temperature.
4. Briefly vortex the tube and then do a quick spin to concentrate the liquid at the bottom. Speed is less than 5000×g.
5. Store the plasmid at -20 ℃.