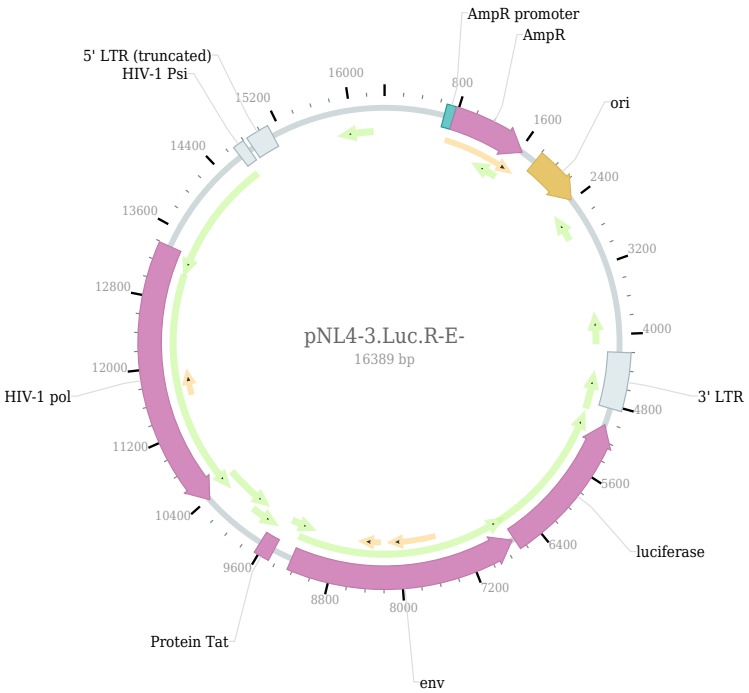
pNL4-3.Luc.R-E-

Vector ID	V012797
Antibiotic Resistance	Ampicillin
Length	16389 bp
Vector Type	Viral Expression & Packaging Vectors

Product Information

Replication Origin	ori
Source/Author	Dr. Nathaniel Landau, Aaron Diamond AIDS Research Center, The Rockefeller University.
Copy Number	High copy number
Growth Strain(s)	Stbl3
Growth Temperature	37℃
Expression Method	Constiutive, Stable
Comment	Firefly luciferase gene was inserted into the pNL4-3 nef gene. Two frameshifts (5'Env and Vpr aa 26) render this clone Env- and Vpr-. Competent for a single round of replication. Requires cotransfection with env expression vector to produce infectious virus. Virus can be produced by transfecting 2x10^6 293 or 293T cells with 20 ug of the NL4-3 DNA, or with 10 ug NL4-3 DNA and 10 ug env expression vector DNA. Transfections can be performed in a 10 cm^2 tissue culture dish using standard calcium phosphate protocols. Virus is typically harvested 48 hours post-transfection. Infections should be performed in a total volume of 0.5 ml. Amphotropic pseudotypes generally have much higher infectivity than those bearing HIV-1 env. Cultures infected with the luciferase viruses can be lysed 2-5 days post-infection and assayed using commercial lysis buffer and luciferase reagents

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 ℃.