



In addition to lentiviral elements, pLVX-DsRed-Express2-N1 contains a puromycin resistance gene (Puro<sup>r</sup>) under the control of the murine phosphoglycerate kinase (PGK) promoter ( $P_{PGK}$ ) for the selection of stable transductants. The vector also contains a pUC origin of replication and an *E. coli* ampicillin resistance gene (Amp<sup>r</sup>) for propagation and selection in bacteria.

### Use

To construct a fusion protein, the gene of interest must be cloned into pLVX-DsRed-Express2-N1 so that it is in-frame with the DsRed-Express2 coding sequence. The inserted sequence should include an initiation codon (ATG) and lack in-frame stop codons.

The fusion protein is constitutively expressed when pLVX-DsRed-Express2-N1 is transduced into target cells. Before the vector can be transduced, however, it must be transfected into 293T packaging cells with our Lenti-X™ HT Packaging System (Cat. Nos. 632160 and 632161). This packaging system allows you to safely produce high titer, infectious, replication-incompetent, VSV-G pseudotyped lentiviral particles that can infect a wide range of cell types, including non-dividing and primary cells (5).

### Location of Features

- 5' LTR: 1–635
- PBS (primer binding site): 636–653
- Ψ (packaging signal): 685–822
- RRE (Rev-response element): 1303–1536
- cPPT/CTS (central polypurine tract/central termination sequence): 2028–2151
- $P_{CMVIE}$  (human cytomegalovirus immediate early promoter): 2185–2787
- MCS (multiple cloning site): 2815–2867
- DsRed-Express2 fluorescent protein gene: 2875–3552
- $P_{PGK}$  (phosphoglycerate kinase promoter): 3571–4079
- Puro<sup>r</sup> (puromycin resistance gene): 4100–4699
- WPRE (woodchuck posttranscriptional regulatory element): 4713–5304
- 3' LTR: 5507–6143
- pUC origin of replication: 6613–7286 (complementary)
- Amp<sup>r</sup> (ampicillin resistance gene; β-lactamase): 7431–8427 (complementary)

### Selection of Stable Transfectants

- Selectable marker: plasmid confers resistance to puromycin.

### Propagation in *E. coli*

- Suitable host strains: DH5α™, DH10B and other general purpose strains.
- Selectable marker: plasmid confers resistance to ampicillin (100 μg/ml) in *E. coli* hosts.
- *E. coli* replication origin: pUC
- Copy number: high

### Excitation and emission maxima of DsRed-Express2

- Excitation maximum = 554 nm
- Emission maximum = 591 nm

### Notes:

The vector sequence was compiled from information in the sequence databases, published literature, and other sources, together with partial sequences obtained by Clontech. This vector has not been completely sequenced.

The viral supernatants produced by this lentiviral vector could contain potentially hazardous recombinant virus. Due caution must be exercised in the production and handling of recombinant lentivirus. Appropriate NIH, regional, and institutional guidelines apply.

## References

1. Shaner, N.C. *et al.* (2004) *Nat. Biotechnol.* **22**(12):1567–1572.
2. Zufferey, R. *et al.* (1999) *J. Virol.* **73**(4):2886–2892.
3. Cochrane, A. W. *et al.* (1990) *Proc. Natl. Acad. Sci. USA* **87**(3):1198–1202.
4. Zennou, V. *et al.* (2000) *Cell* **101**(2):173–185.
5. Wu, X. *et al.* (2000) *Mol. Ther.* **2**(1):47–55.

## Notice to Purchaser

Clontech products are to be used for research purposes only. They may not be used for any other purpose, including, but not limited to, use in drugs, *in vitro* diagnostic purposes, therapeutics, or in humans. Clontech products may not be transferred to third parties, resold, modified for resale, or used to manufacture commercial products or to provide a service to third parties without written approval of Clontech Laboratories, Inc.

DH5 $\alpha$ <sup>™</sup> is a trademark of Invitrogen Corporation.

### cPPT/CTS Element:

This product and its use are the subject of U.S. Pat. No. 6,682,907. The purchase of this product conveys to the buyer the non-transferable right to use the purchased amount of the product and components of the product in research conducted by the buyer (whether the buyer is an academic or for-profit entity). The buyer cannot disclose information, sell or otherwise transfer this product, its components or materials made using this product or its components to a third party or otherwise use this product or its components or materials made using this product or its components for any commercial purposes. If the buyer is not willing to accept the limitations of this limited use statement, Clontech is willing to accept return of the product with a full refund. For information on purchasing a license to the DNA-Flap technology for purposes other than research, contact the Transfer of Technology Office, Institut Pasteur, 28 rue du Docteur Roux, 75 724 Paris Cedex 15 ([www.pasteur.fr](http://www.pasteur.fr)).

### Reef Coral Fluorescent Proteins:

The RCFP's (including DsRedExpress and DsRedExpress2) are covered by one or more of the following U.S. patent Nos. 7,166,444; 7,157,565; 7,217,789; 7,338,784; 7,338,783; 7,537,915 and 7,442,522.

### Living Colors<sup>®</sup> Fluorescent Protein Products:

Not-For-Profit Entities: Orders may be placed in the normal manner by contacting your local representative or Clontech Customer Service at 650.919.7300. At its discretion, Clontech grants Not-For-Profit Entities a non-exclusive, personal, limited license to use this product for non-commercial life science research use only. Such license specifically excludes the right to sell or otherwise transfer this product, its components or derivatives thereof to third parties. No modifications to the protein coding sequence may be made without express written permission from Clontech. Any other use of this product requires a license from Clontech. For license information, please contact a licensing representative by phone at 650.919.7320 or by e-mail at [licensing@clontech.com](mailto:licensing@clontech.com).

For-Profit Entities wishing to use this product are required to obtain a license from Clontech. For license information, please contact a licensing representative by phone at 650.919.7320 or by e-mail at [licensing@clontech.com](mailto:licensing@clontech.com) or click here for more information.

### WPRE Purchaser Notification:

Clontech has a license to sell products containing WPRE, under the terms described below. Any use of WPRE outside of Clontech's product or the product's intended use, requires a license as detailed below. Before using the product containing WPRE, please read the following license agreement. If you do not agree to be bound by its terms, contact Clontech within 10 days for authorization to return the unused product containing WPRE and to receive a full credit.

Patents: The WPRE technology is covered by one or more of the following US patents and corresponding patent claims outside the US: 6,136,597 ; 6,284,469 ; 6,312,912 ; 6,287,814 , issued to The Salk Institute for Biological Studies

Individual License Agreement: Clontech grants you a non-exclusive license to use the enclosed product containing WPRE in its entirety for its intended use. The product is being transferred to you in furtherance of, and reliance on, such license. Any use of WPRE outside of Clontech's product or the product's intended use, requires a license from the Salk Institute for Biological Studies.

Termination of License: This license agreement is effective until terminated. You may terminate it at any time by destroying all products containing WPRE in your control. It will also terminate automatically if you fail to comply with the terms and conditions of the license agreement. You shall, upon termination of the license agreement, destroy all products containing WPRE in your control, and so notify Clontech in writing. This License shall be governed in its interpretation and enforcement by the laws of the State of California.

### Contact for WPRE Licensing:

The Salk Institute for Biological Studies  
10010 North Torrey Pines Road  
La Jolla, CA 92037  
Attn.: Office of Technology Management  
Phone: 858.453.4100 ext. 1275  
Fax: 858.546.8093

Clontech, the Clontech logo and all other trademarks are the property of Clontech Laboratories, Inc., unless noted otherwise. Clontech is a Takara Bio Company. ©2009 Clontech Laboratories, Inc