

Lentiviral Expression System

Obtain high-level expression in virtually any cell type with our complete Lenti-X™ Expression System

- **Optimized lentiviral vector and packaging system for high titers and high expression**
- **Transfer genes into dividing and nondividing cells and stem cells**
- **Puromycin resistance allows rapid selection of transduced cells**
- **Safe, replication-incompetent virus**

Recombinant lentiviruses derived from HIV-1 are able to deliver genes into almost any mammalian cell type, including primary cultures, dividing or nondividing cells, and stem cells. Clontech has developed a highly advanced lentiviral expression system that provides the broad cellular tropism of VSV-G pseudotyped lentivirus, high viral titers, and excellent transgene expression. The **Lenti-X Expression System**, which includes the **pLVX-Puro** expression vector and our **Lenti-X HT Packaging System**, enables you to produce exceptionally high titers of safe, replication-incompetent lentivirus from your customized pLVX-Puro vector (Figure 1).

Superior Lenti-X Vectors

Like all our Lenti-X vectors, pLVX-Puro not only carries the LTRs and packaging sequence required for lentivirus production and replication, but it also contains elements that improve transgene expression, titer, and overall vector function. Its WPRE element, believed to promote RNA processing events and nuclear export, imparts a dual benefit (1). First, it acts within the context of viral genomic transcripts to enhance vector packaging and increase the titers of viral supernatants produced from 293T packaging cells. Second, it boosts expression of your gene of interest in target cells by facilitating the production of mature mRNA from transcripts initiated by the vector's internal CMV promoter. Lenti-X vectors also contain a cPPT element that increases nuclear importation of the viral genome during target cell infection, resulting in improved vector integration and more efficient transduction (2).

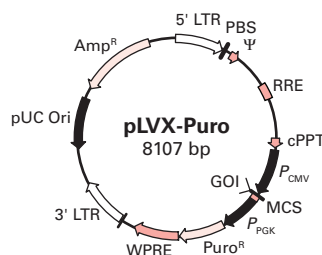


Figure 1. Map of pLVX-Puro. The vector contains the lentiviral-specific LTRs and packaging sequence (Ψ); a multiple cloning site (MCS) to insert your gene of interest (GOI); puromycin resistance; and WPRE and cPPT elements to boost packaging, viral titers, and transgene expression.

High-Efficiency Packaging

Our Lenti-X HT Packaging System produces outstanding viral titers due to a synergism of highly optimized components (3; pages 8–9). The Lenti-X HT Packaging Mix safely provides all the essential lentiviral packaging and replication gene products *in trans* on a proprietary suite of separate vectors. Selected plasmids in the mixture generate high expression levels for critical viral proteins as a result of Tet-Off® transactivation. For added safety, a split gag-pol gene delivery strategy thoroughly prevents viral replicative functions from being transferred to target cells (3). Finally, the included **Lentiphos™ HT** transfection reagents transfer the Lenti-X HT Packaging Mix, along with your pLVX-Puro vector, into 293T cells with unprecedented efficiency (see page 15). The resulting high-titer viral supernatants can be used directly, without concentration.

High Titers & Rapid Selection

We used the Lenti-X Expression System to generate a high-titer pLVX-Puro supernatant, serial dilutions of which were used to infect naïve cultures of 293T cells (Figure 2). After replating the infected cells on 10 cm dishes and selecting transductants with puromycin, the resulting colonies of stable transductants were stained for detection. Cells infected with only 0.1 μ l of supernatant produced hundreds of colonies, while colonies from cells infected with 1 μ l virtually covered

Product	Size	Cat. No.
Lenti-X Expression System*	each	632164
Puromycin	25 mg	631305
	100 mg	631306

* 本製品の購入には拡散防止措置に関する確認書が必要です。詳細は3ページ【ご注意】をご覧ください。

Components

- pLVX-Puro Vector
- Lenti-X™ HT Packaging Mix
- Lentiphos™ HT
- Lenti-X™ Lentiviral Expression Systems User Manual (PT3983-1)

Related Products

- Lenti-X™ Fluorescent Vectors (Cat. Nos. 632152, 632153, 632154 & 632155)
- Lenti-X™ HT Packaging System (Cat. Nos. 632160 & 632161)

Notice to Purchaser

Please see the bGH Poly A, CMV Sequence, cPPT Element, IRES Sequence, Lentiviral Expression Products, Tet-Based Expression Products, VSV-G Technology, and WPRE Technology licensing statements on page 42.

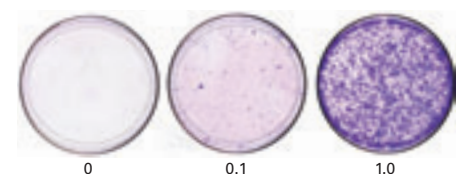


Figure 2. Puromycin selection of transduced cells. 293T cells were infected with the indicated volumes (μ l) of pLVX-Puro supernatant and selected with puromycin for 9 days to allow the formation of colonies, which were then stained with crystal violet.

the entire plate. These results demonstrate the high titer and infectivity of a typical pLVX-Puro supernatant.

The Lenti-X Expression System is a comprehensive system for preparing recombinant lentivirus to express any cDNA in any cell type susceptible to lentivirus transduction. It easily produces high-titer lentiviral supernatants suitable for safe use with virtually any downstream application.

References

1. Zufferey, R. *et al.* (1999) *J. Virol.* **73**(4):2886–2892.
2. Zennou, V. *et al.* (2000) *Cell* **101**(2):173–185.
3. Wu, X. *et al.* (2000) *Mol. Ther.* **2**(1):47–55.