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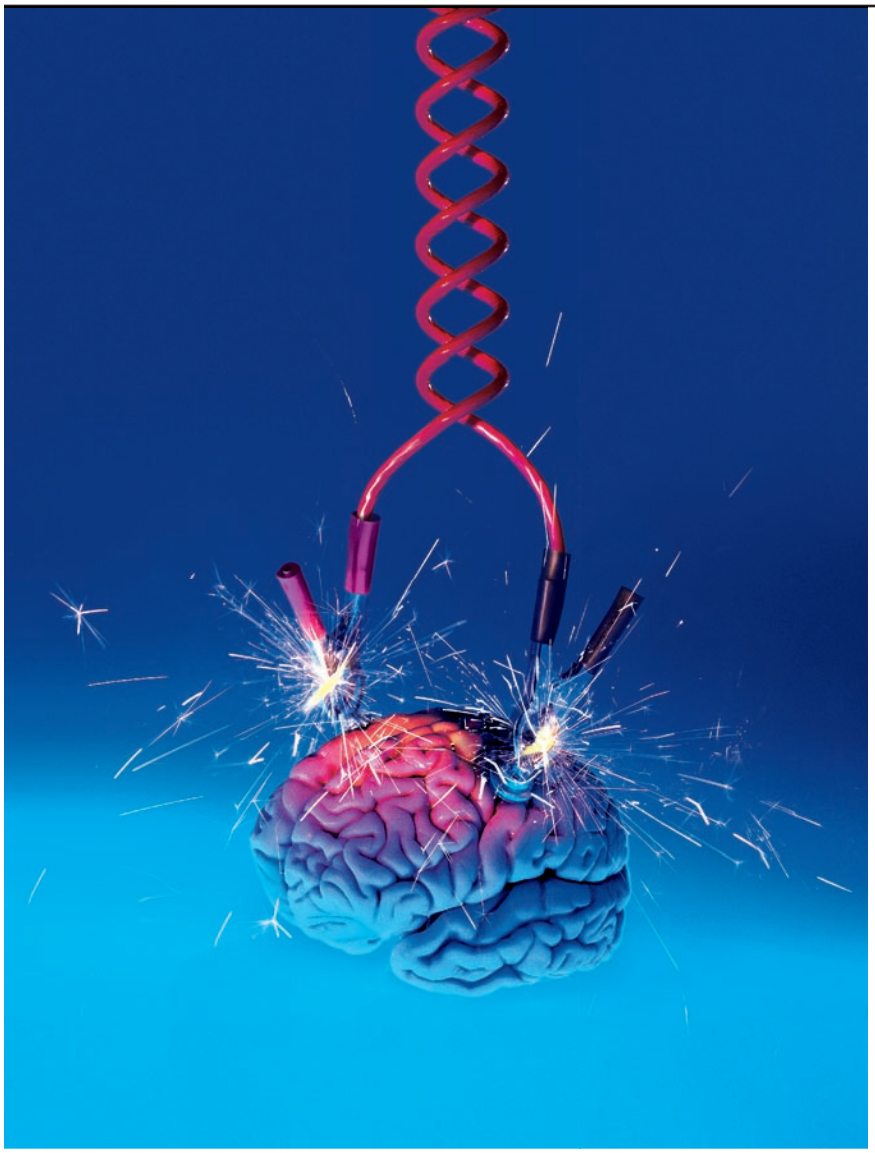
Jumpstart your research with ViraPower™ Lentiviral Expression Systems



With ViraPower™ Lentiviral Systems you can:

- Efficiently transduce both dividing and non-dividing cells
- Study long-term gene expression
- Reproducibly transduce cell populations
- Choose the most physiologically relevant promoter for your experiments

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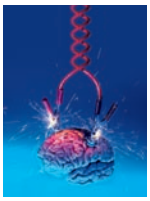
Stable gene expression in any cell type



Achieve stable gene expression at reproducible levels—regardless of mammalian cell type—with the ViraPower™ Lentiviral Expression System. Using the ViraPower™ Lentiviral System, you'll produce non-replicating viral particles that can efficiently transduce nearly any dividing or non-dividing cell type. In addition, virus-mediated expression allows precise and reproducible control over the number of cells transduced in a population and the number of stably integrated copies of your gene. This allows you to perform your studies at the expression levels you need, in the cells you want.

Powerful expression

Whether you're using a hard-to-transfect mammalian cell line, an animal model, or simply want efficient gene delivery, the ViraPower™ Lentiviral Expression System provides stable gene expression and reproducible delivery to both dividing and non-dividing cells (Table 1). Even in mammalian cell types that challenge standard transfection or other viral transduction experiments, the ViraPower™ Lentiviral Expression System provides the high levels of gene expression necessary for valid results.

Table 1 — Choose the best viral system for your experiments

Viral system	Transient expression		Stable expression			
	Dividing cells	Non-dividing cells	Dividing cells	Neuronal cells	Drug- or growth-arrested cells	Contact-inhibited cells
Adenovirus	●	●				
Retrovirus	●		●			
Lentivirus	●	●	●	●	●	●

How it works

Stable gene expression is only a few steps away with the ViraPower™ Lentiviral Expression System (Figure 1). Just transfect, harvest, and titer to produce sufficient viral supernatant for performing many transduction* experiments. Use the supernatant immediately or store it for up to one year. No need to buy additional reagents, the ViraPower™ Lentiviral Expression System has everything you need.

Figure 1 — How the ViraPower™ Lentiviral Expression System works

Generate the pLenti expression construct containing your gene of interest.

Cotransfect the ViraPower™ 293FT™ producer cell line with your pLenti expression construct and the optimized ViraPower™ packaging mix.

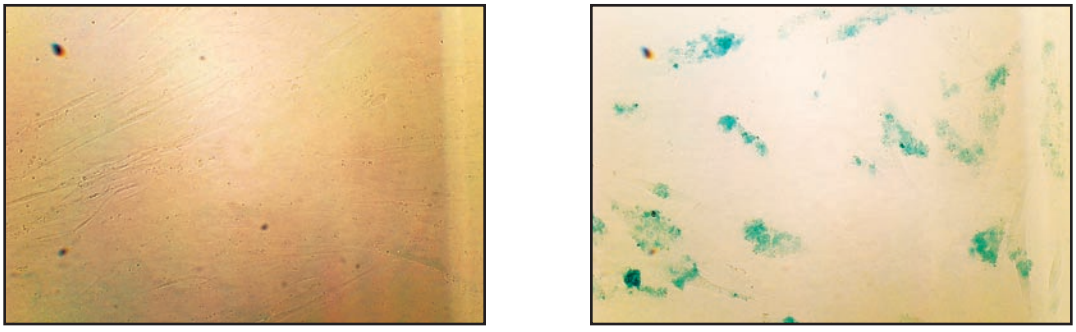
Harvest viral supernatant and determine the titer.

*The virus particles transduce cells only once and will not produce infectious progeny.

Expand your research options

In contrast to traditional Moloney (MLV)-based retroviral systems, the ViraPower™ Lentiviral System readily transduces non-dividing cells without a round of replication. Now even growth- or drug-arrested cells and non-dividing primary cultures are easily transduced by the ViraPower™ Lentiviral System (Figure 2).

Figure 2 — Lentivirus transduces primary fibroblasts



Traditional retroviral vector

pLenti6/V5-GW/*lacZ*™ lentiviral vector

Contact-inhibited non-dividing quiescent primary human foreskin fibroblasts were transduced with retroviral and lentiviral vectors at an MOI of 1 and stained for β-galactosidase activity 48 hours post-transduction.

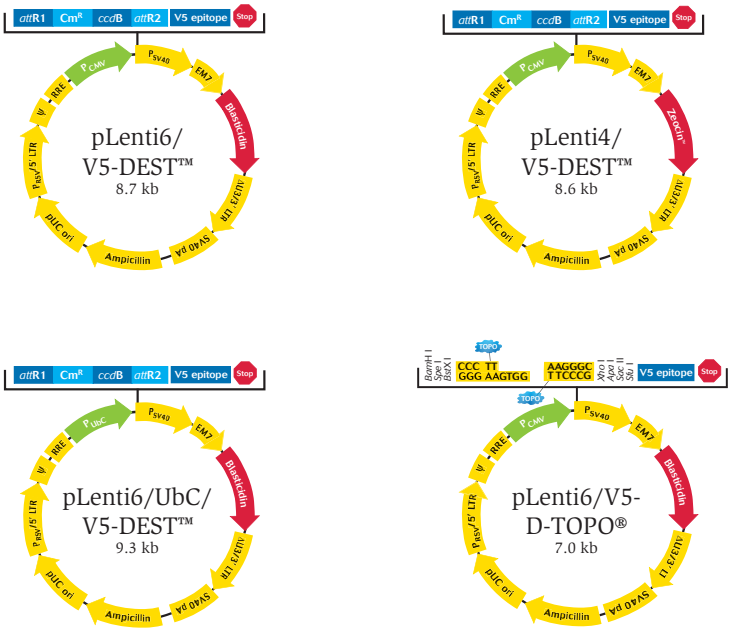
Expression vectors to meet any experimental design

A number of ViraPower™ Lentiviral Vectors are available, offering options for cloning method and promoter choice. This allows you to optimize the experiment for your cell line or animal model. The various selections are discussed on the following pages.

Flexible cloning options

You can enter into a ViraPower™ vector using either convenient Gateway® Technology‡ or the fast TOPO® Cloning reaction. If you want the flexibility to transfer your gene of interest between different expression vectors or multiple expression systems, choose the Gateway® pLenti6/V5-DEST™, pLenti4/V5-DEST™, or pLenti6/UbC/V5-DEST™ vectors (Figure 3). For simple, 5-minute TOPO® cloning, choose the pLenti6/V5-D-TOPO® vector (Figure 3). Vectors are available separately or as part of complete ViraPower™ Lentiviral Gateway® or Directional TOPO® Expression Systems.

Figure 3 — ViraPower™ Lentiviral expression vectors



‡To learn more about the flexibility of Gateway® Technology, visit the online seminar at www.invitrogen.com/gateway.

Two choices for constitutive expression

Different promoters work better in different cell lines. To help you achieve the expression levels you need, the ViraPower™ vectors are available with either the CMV or Ubiquitin C (UbC) promoters:

- The pLenti6/V5-DEST™, pLenti4/V5-DEST™, and pLenti6/V5-D-TOPO® vectors include the CMV promoter for high-level expression in most cell lines
- The pLenti6/UbC/V5-DEST™ vector carries the UbC promoter for efficient *in vivo* expression or when using cell lines that downregulate the CMV promoter

High-level regulated expression in any mammalian cell type

Turn expression of your gene on or off in nearly any mammalian cell type with the ViraPower™ T-REx™ Lentiviral Expression System (Figure 4). T-REx™ technology takes advantage of a tetracycline derepression mechanism to tightly control protein levels by the presence/absence of tetracycline†. Combining this with reliable delivery to dividing and non-dividing mammalian cells and long-term stable expression achieved with the ViraPower™ Lentiviral System (Figure 5) allows you to get the results you need—even with toxic genes. The ViraPower™ Lentiviral T-REx™ System allows you to:

- Induce high-level expression of the gene of interest from the CMV promoter
- Achieve low basal gene expression through tight control of the tetracycline operator sequences
- Control dose and timing of expression using a cost-efficient inducer
- Generate stable cell lines expressing the tetracycline repressor (TetR) from pLenti6/TR

Figure 4 — ViraPower™ T-REx™ Lentiviral vectors

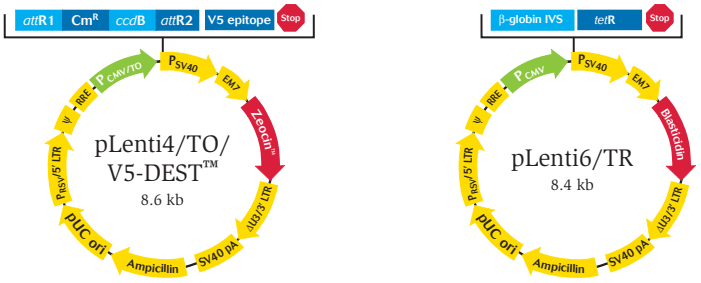
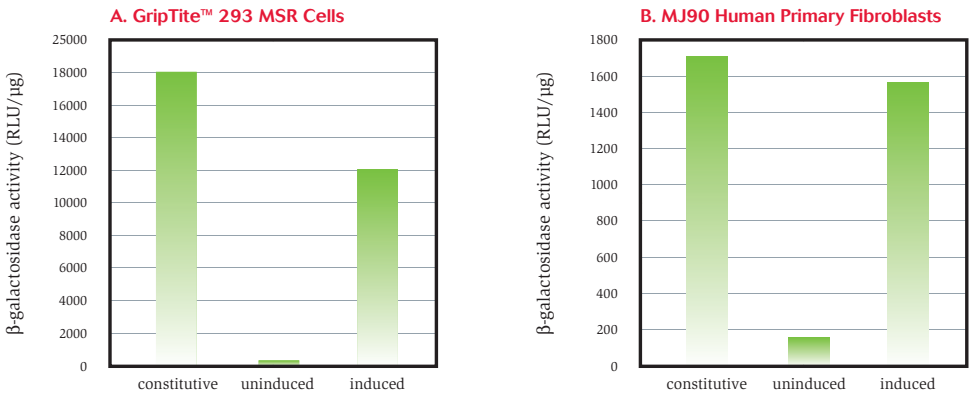


Figure 5 — High-level regulated expression in dividing and non-dividing cell types



(A) Actively growing GripTite™ 293 MSR cells were transduced with pLenti6/TR virus (MOI = 32) followed by transduction with pLenti4/TO/V5-GW/*lac*Z virus (MOI = 5) (B) Confluent, growth-arrested primary human fibroblasts were transduced with pLenti6/TR virus (MOI = 7.5) followed by transduction with pLenti4/TO/V5-GW/*lac*Z virus (MOI = 7.5). For all samples, cells were induced 24 hours post-transduction with tetracycline at 1 µg/ml for 48 hr. Cell lysates were assayed for *lac*Z expression and activity.

†To learn more about regulating expression with the T-REx™ System, visit www.invitrogen.com.

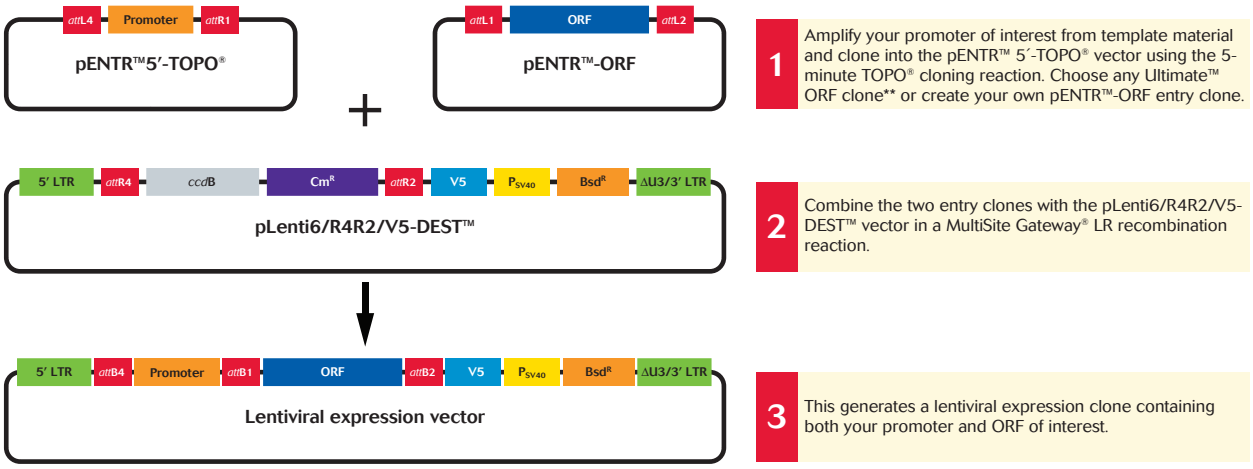
Expression vectors to meet any experimental design, continued

Add your own promoter

Advanced systems, such as primary cells, stem cells, and animal models, may require you to express your gene of interest using a physiologically relevant promoter or to regulate transgene expression. These advanced applications require a greater degree of flexibility in experimental design. You'll get this flexibility with the ViraPower™ Promoterless Lentiviral Gateway® System. By harnessing the cloning efficiency of MultiSite Gateway® Technology (Figure 6), you can:

- Rapidly and efficiently clone both promoter and gene of interest into a ViraPower™ Lentiviral Expression Vector
- Choose the best promoter for your cell type by efficiently expressing your gene of interest from multiple promoter constructs
- Achieve tissue-specific expression (Figure 7)

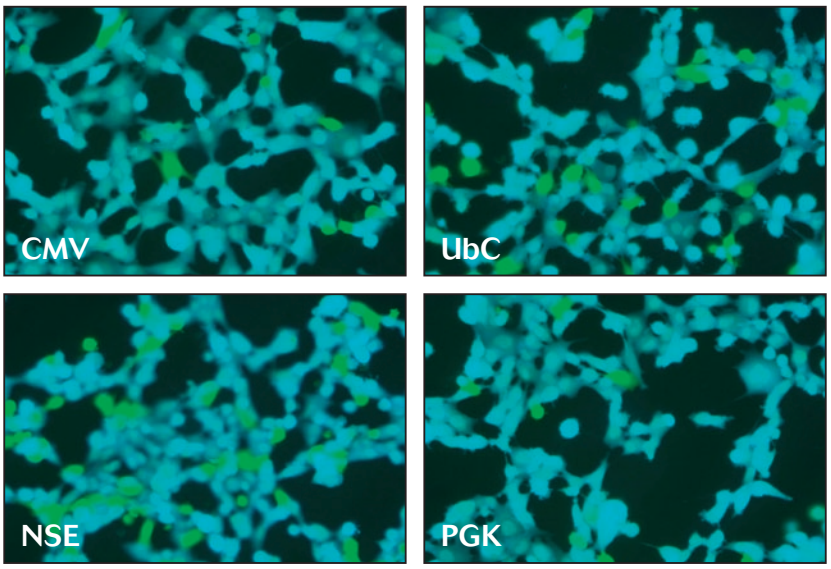
Figure 6 — The ultimate flexibility in experimental design



The ViraPower™ Promoterless Lentiviral Gateway® System uses three vectors and a highly efficient MultiSite Gateway® LR recombination reaction to allow you to create a lentiviral expression vector for your optimal experimental design.

**For more information on the Ultimate™ ORF clones available, visit http://orf.invitrogen.com.

Figure 7 — β-lactamase expression from a panel of promoters using pLenti6/R4R2/V5-DEST™ viruses



HT1080 cells were transduced at MOI = 1 with lentiviruses expressing beta-lactamase from the indicated promoters. Transduced cells were loaded with CCF2 48 hours after transduction. Imaging was performed on a fluorescence microscope using an Omega Optical Filter Set XF106-2. Cells that express β-lactamase fluoresce blue. Cells that do not express β-lactamase fluoresce green.
CMV: Cytomegalovirus immediate early gene; UbC: Ubiquitin C; PGK: Phosphoglycerate kinase; NSE: Neuron Specific Enolase.

Your experiment, your way

Whether you are working with a primary cell line, studying a toxic gene, or need tissue-specific expression of your gene of interest in an animal model, the ViraPower™ Lentiviral Gateway® Expression System is designed to meet your experimental needs.

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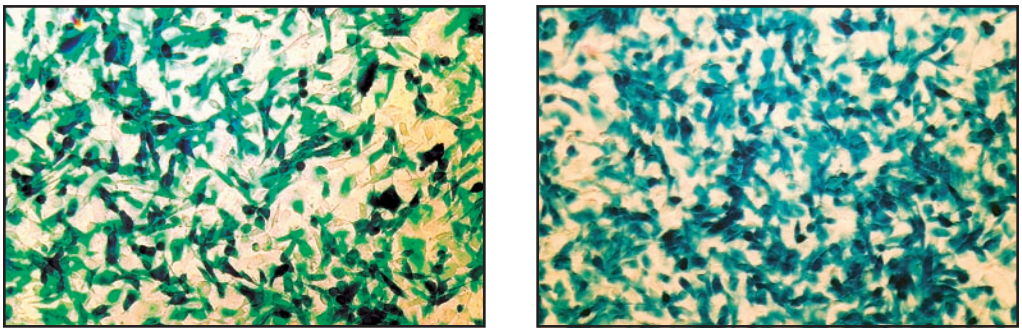
High-level, stable gene expression

Get long-term, stable gene expression in any mammalian cell type. Unlike traditional retroviruses, the lentivirus is actively imported into the nuclei of non-dividing cells by *cis*-acting elements. Gene expression occurs as soon as the gene is integrated into the genome. Expression is stable, allowing you to continuously achieve high protein yields (Figure 8).

Figures 8 — Stable, high-level lentiviral expression



10 days post-transduction **6 weeks post-transduction**

HT1080 cells were transduced with pLenti6/V5-GW/*lac*Z™ lentiviral expression vector and stably selected with 10 µg/ml blasticidin. Cultures were stained for β-galactosidase expression at 10 days and 6 weeks post-transduction.

Designed for safety

For your protection, key safety features are built into the ViraPower™ Lentiviral System§. With the majority of viral proteins removed, the gene delivery vector is safe and far removed from wild-type virus. Similar to other retroviral expression systems, the packaging functions are supplied *in trans*. Other safety features include:

- Absence of LTRs in the ViraPower™ Packaging Mix—packaging vectors are only expressed in the producer cell and never packaged into virions
- Viral particles are replication incompetent and only carry the gene of interest—no other viral species are produced
- Modified to be self-inactivating—transduced and integrated lentiviral vectors are no longer capable of producing a packageable viral genome

ViraPower™ delivers safe, powerful expression for all of your experiments.

§ Despite the inclusion of these safety features, we highly recommend that you strictly adhere to all published guidelines for Biosafety Level 2 (BL-2) organisms when working with lentiviral stocks generated using this system, as with all your other mammalian cells and reagents.

Complete kits

Each ViraPower™ Lentiviral Expression System provides everything you need to perform a lentiviral expression experiment, including a kit to clone your gene of interest into a pLenti™ expression vector, the 293FT™ producer cell line, and a 20-reaction ViraPower™ Lentiviral Support Kit with Lipofectamine™ 2000 Transfection Reagent, ViraPower™ Packaging Mix, and either Blasticidin or Zeocin™ selection agent. Use Table 2 to choose the system that best fits your experimental needs. Select components are also available separately.

Table 2 — Choose the ViraPower™ Lentiviral configuration to optimize your experimental design

Lentiviral product	Configuration		Cloning option		Promoter option			
	Vector Kit	Complete Kit	Gateway®	TOPO®	CMV	UbC	Inducible	User-defined
ViraPower™ Lentiviral Gateway® Expression System		●	●		●			
pLenti6/V5-DEST™ Gateway® Vector	●		●		●			
ViraPower™ Lentiviral Directional TOPO® Expression System		●		●	●			
pLenti6/V5™ Directional TOPO® Cloning Kit	●			●	●			
ViraPower™ UbC Lentiviral Gateway® Expression System		●	●			●		
pLenti6/UbC/V5-DEST™ Gateway® Vector	●		●			●		
ViraPower™ Zeo Lentiviral Gateway® Expression Kit		●	●		●			
pLenti4/V5-DEST™ Directional Gateway® Vector	●		●		●			
ViraPower™ Promoterless Lentiviral Gateway® Expression System		●	●					●
ViraPower™ Promoterless Lentiviral Gateway® Vector Kit	●		●					●
ViraPower™ T-REx™ Lentiviral Expression System		●	●				●	
ViraPower™ T-REx™ Lentiviral Gateway® Vector Kit	●		●				●	

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Get results with the ViraPower™ System

Now you can get reproducible results in every cell line, and high levels of stable gene expression in entire cell populations. The ViraPower™ Lentiviral Expression System even works on previously difficult-to-use cell types. Order today for powerful expression analysis.

Gateway® Technology or TOPO® Cloning

Product	Quantity	Cat. no.
ViraPower™ Lentiviral Gateway® Expression System	1 kit	K4960-00
pLenti6/V5-DEST™ Gateway® Vector	6 µg	V496-10
ViraPower™ Lentiviral Directional TOPO® Expression System	1 kit	K4950-00
pLenti6/V5™ Directional TOPO® Cloning Kit	20 rxns	K4955-10
ViraPower™ UbC Lentiviral Gateway® Expression Kit	1 kit	K4990-00
pLenti6/UbC/V5-DEST™ Gateway® Vector	6 µg	V499-10
ViraPower™ Zeo Lentiviral Gateway® Expression Kit	1 kit	K4980-00
pLenti4/V5-DEST™ Gateway® Vector	6 µg	V498-10

Promoterless Lentiviral Vector

Product	Quantity	Cat. no.
ViraPower™ Promoterless Lentiviral Gateway® Expression System *with MultiSite Gateway® Technology*	1 kit	K5910-00
ViraPower™ Promoterless Lentiviral Gateway® Vector Kit *with MultiSite Gateway® Technology*	1 kit	K591-10
pENTR™5′-TOPO® TA Cloning® Kit *with One Shot® TOP10 Chemically Competent Cells*	20 rxns	K591-20

Regulatable Expression

Product	Quantity	Cat. no.
ViraPower™ T-REx™ Lentiviral Expression System	20 rxns	K4965-00
ViraPower™ T-REx™ Lentiviral Gateway® Vector Kit	20 rxns	K4967-00

Support Products

Product	Quantity	Cat. no.
ViraPower™ Bsd Lentiviral Support Kit	20 rxns	K4970-00
ViraPower™ Zeo Lentiviral Support Kit	20 rxns	K4985-00
ViraPower™ 293FT™ Cell Line	3 x 10^6 cells	R700-07
pENTR™/D-TOPO® Cloning Kit	20 rxns	K2400-20
One Shot® Stbl3™ Chemically Competent *E. coli*	20 rxns	C7373-03
ViraPower™ Lentiviral Packaging Mix	60 rxns	K4975-00
Lipofectamine™ 2000 Transfection Reagent	1.5 ml	11668-019
	0.75 ml	11668-027
Blasticidin S HCl	50 mg	R210-01
Zeocin™	1 g	R250-01
	5 g	R250-05

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