GenTarget Inc

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Pre-made inducible lentivirus, fluorescent controls:

for validation of tetracycline inducbile expression system

Cat#	Product Name	Amounts
LVP800	GFP (TetCMV, Puro) lentiviral particles	$1x10^7$ IFU/ml x 200ul
LVP800-PBS	GFP (TetCMV, Puro) lentiviral particles in PBS	$1x10^8$ IFU/ml x 200ul
LVP1399	GFP (TetCMV, Bsd) lentiviral particles	$1x10^7$ IFU/ml x 200ul
LVP1399-PBS	GFP (TetCMV, Bsd) lentiviral particles in PBS	$1x10^8$ IFU/ml x 200ul
LVP801	RFP (TetCMV, Puro) lentiviral particles	$1x10^7$ IFU/ml x 200ul
LVP801-PBS	RFP (TetCMV, Puro) lentiviral particles in PBS	$1x10^8$ IFU/ml x 200ul
LVP802	CFP (TetCMV, Puro) lentiviral particles	$1x10^7$ IFU/ml x 200ul
LVP802-PBS	CFP (TetCMV, Puro) lentiviral particles in PBS	$1x10^8$ IFU/ml x 200ul
LVP803	BFP (TetCMV, Puro) lentiviral particles	$1x10^7$ IFU/ml x 200ul
LVP803-PBS	BFP (TetCMV, Puro) lentiviral particles in PBS	$1x10^8$ IFU/ml x 200ul
LVP024	GFP inducible control lentivirus	$1x10^7$ IFU/ml x 200ul
LVP024-PBS	GFP inducible control lentivirus in PBS	$5x10^7$ IFU/ml x 200ul
LVP357	YFP inducible control lentivirus	$1x10^7$ IFU/ml x 200ul
LVP357-PBS	YFP inducible control lentivirus in PBS	$5x10^7$ IFU/ml x 200ul
LVP531	RFP inducible control lentivirus	$1x10^7$ IFU/ml x 200ul
LVP531-PBS	RFP inducible control lentivirus in PBS	$5x10^7$ IFU/ml x 200ul

Storage: <-70 °C, avoid repeat freeze/thaw cycles. Stable for 6 months at <-70oC.

1. Product Description:

Lentivectors are HIV-1 (Human Immunodeficiency Virus 1) derived plasmids. They generate replication-incompetent lentivirus that can be transduced into almost all types of mammalian cells, including primary and non-dividing cells. Lentiviral particles (LP) are lentivirus supernatant generated from lentivectors express a specific gene or RNAi construction.

Lentivirus is the easiest and most effective method for delivering genes into the majority of mammalian cell types, including non-dividing and primary cells. It allows genes to be integrated into the host cell genome for long-term expression.

GenTarget provides pre-made optional inducible expression lentiviruses for fluorescent proteins or specific human or mouse genes with various selection markers. To validate the inducible system in your cell lines, GenTarget provides **inducible lentivirus controls** which have been generated from GenTarget's **optional inducible lentiviral system**.

The inducible controls contain a fluorescent protein under control of the optional inducible promoter (**TetCMV**), enabling convenient verification of tetracycline-inducible effects by simple monitoring of the **GFP, RFP YFP or BFP** signal under a fluorescence microscope. The controls also contain an antibiotic or a fluorescent-antibiotic fusion dual selection marker under a RSV promoter which is a constitutive promoter and not affected by induction. The marker provides a convenient way to monitor viral transduction efficiency and select positive cells by either antibiotic killing or fluorescence sorting (See vector scheme above)

The VSV-G pseudotyped lentiviral particles are generated in 293T cells, and are provided in two formats:

- DMEM containing 10% FBS and 60 ug/ml polybrene (10x);
- PBS without any additives (suitable for serum sensitive culture or for the hard-to-transduce cells, or for *in vivo* applications.

For more details about premade particles, please see **FAQ for pre-made lentiviral particles** . (**http://www.gentarget.com/pdf/FAQ-Premade-Lentiviral-particles.pdf**). **Note:** all GenTarget pre-made lentivirus are intended for research use only, not for therapeutic or clinic usage.

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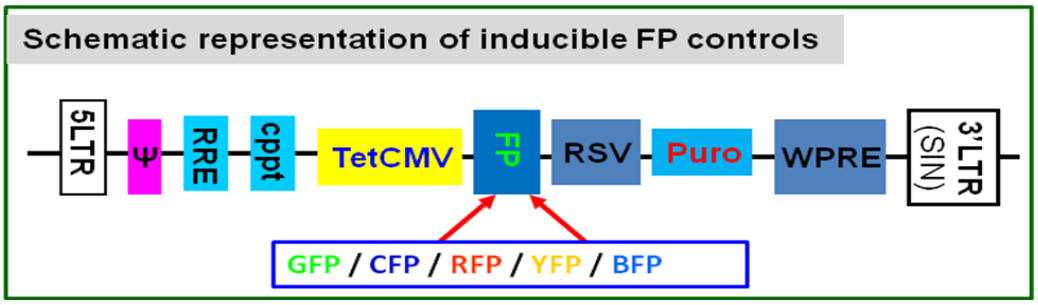

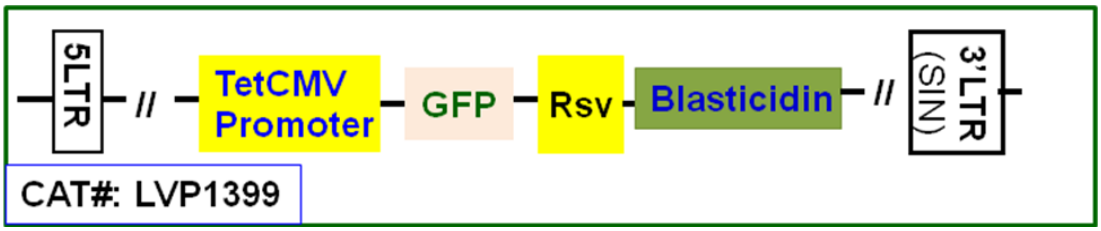

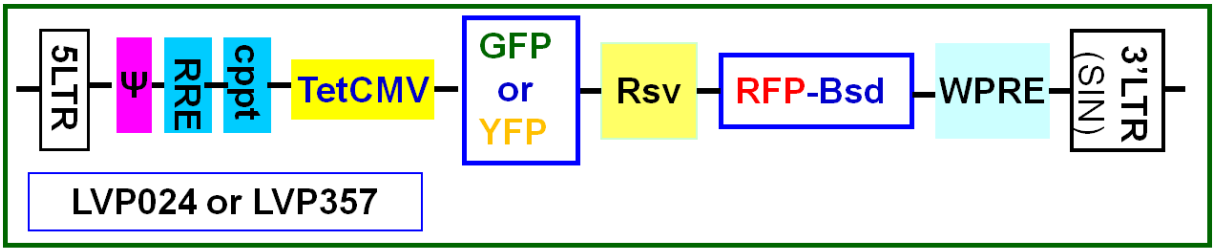

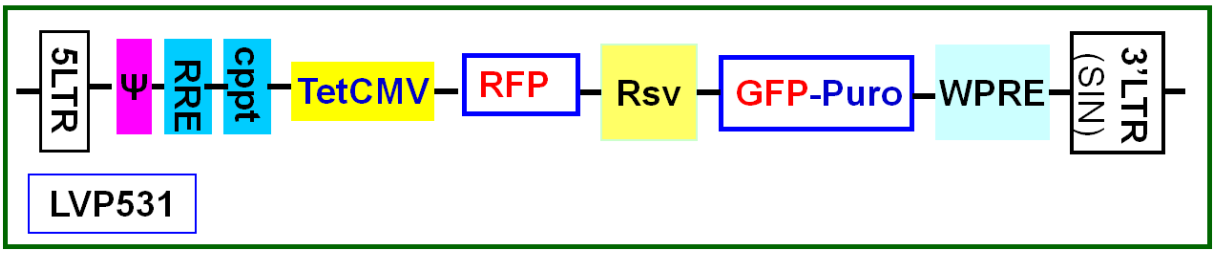

2. About optional tetracycline inducible expression:

Like GenTarget's target-specific lentiviruses, the inducible lentivirus controls can be used for regular constitutive high expression without the requirement for tetracycline induction, or they can also be used for tetracycline inducible expression. The inducible CMV promoter (TetCMV) has two copies of the tetracycline operator sequence. Expression becomes inducible only when the tetracycline regulator protein (TetR) is present in advance; in this case, TetR stops transcription, which can be activated by the addition of tetracycline.

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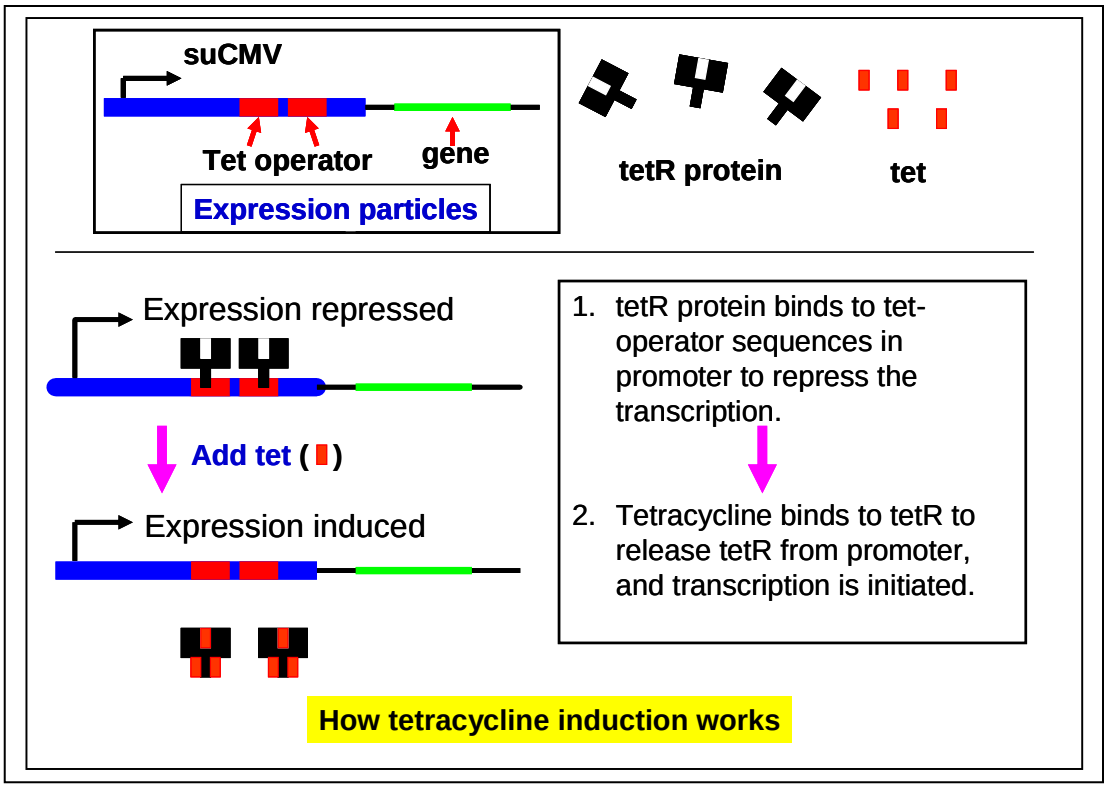
Inducible expression is tetracycline dose dependent; generally, 1-2 µg/ml of tetracycline is used.

If inducible expression is desired, the repressor regulator protein (TetR) must be expressed in advance or at the same time as transduction. The presence of tetR can be achieved by the following methods:

- **TetR stable cell lines** that constitutively express the TetR protein
- Co-transfection with a TetR expression plasmid and a target-inducible expression vector
- Co-transduction with TetR lentiviral particles and inducible gene expression lentiviral particles

Co-transduction with GenTarget's premade TetR lentiviral particles is the best method for delivering the TetR protein.

The image below illustrates how inducible expression works.



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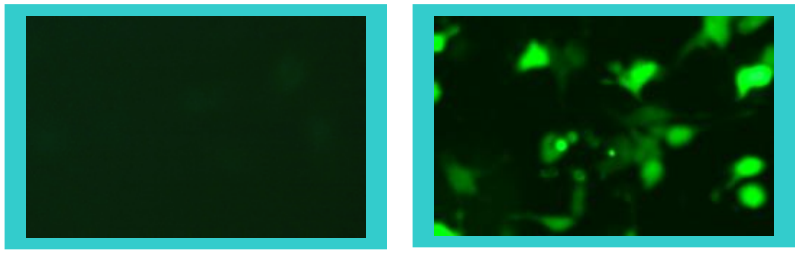
3. Key features:

- High GFP, YFP, and RFP inducible expression levels and high viral titer
- Convenient monitoring of transduction efficiency by the constitutively expressed RFP or GFP signal.
- Dual markers: transduced cells can be sorted by fluorescent signal or selected for antibiotic resistance.

Inducible expression sample images:



Sample image for inducible expression: 50ul of GFP inducible particles (Cat#: **LVP024**) were added into **TetR-293 stable cells** (Cat#: **SC005-Bsd**). And 2ug/ml final of tetracycline was added at 3 days post transduction. Image were taken at 24 hours after induction, the left image without induced, the right image induced by 2 ug/ml final Tetracycline.

4. Related products:

Products	Name	Applications
Product series (>500 CAT#)	**Premade expression ready lentiviral particle for human and mouse genes**	• Used as constitutive expression of a human or mouse target. • Or used with TetR particles together for inducible expression of a human or mouse target.
TetR expression stable cell lines	Premade TetR Stable cell lines in 293 host cells with different antibiotic selection markers (CAT#: **SC005**) .	Used for tetracycline inducible expression of any constructs with TetR binding sequence in their promoter.

5. Safety Precaution:

Gentarget lentiviral particles adapts must advanced lentiviral safety features (using the third generation vectors with self-inactivation SIN-3UTR), and the premade lentivirus is replication incompetent. However, please use extra caution when using lentiviral particles. Use the lentiviral particles in Bio-safety II cabinet. Ware glove all the time at handling Lentiviral particles! Please refer CDC and NIH's guidelines for more details regarding to safety issues.

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6. References:

1. OGorman et al., 1991; Sauer, 1994).
2. Molecular Therapy (2003) 7, 460–466;
3. Annu Rev Microbiol. 1994;48:345-69.
4. Microbiol Mol Biol Rev. 2005 Jun;69(2):326-56.
5. NIH Guidelines for Biosafety Considerations for Research with Lentiviral Vectors. (Link).
6. CDC guidelines for Lab Biosafety levels (Link).

Warranty:

This product is warranted to meet its quality as described when used accordance with its instructions. Gentarget disclaims any implied warranty of this product for particular application. In no event shall GenTarget be liable for any incidental or consequential damages in connection with the products. Gentarget's sole remedy for breach of this warranty should be, at Gentarget's option, to replace the products.

Attachment: GenTarget's pre-made lentivirus product categories.

Product Category	Product Description (please click into each category's page)
Pathway Reporter	Repoter Lentivirus for all kinds of pathway screening assays
Cell Immortalization	Lentivirus for cell immortalization: Large T-antigen, hTERT, EBNA1/EBNA2, HpV16-E6/E7, Adenovial E1A, Kras_G12V, HOXA9, et al.
ImmunoOncology Research	Lentivirus products for immuno therapy research: CAR and TCR; Assay Cell Lines for T-cell targeted killing assay and other cell-based assays; over-expression lentivirus products for the immune response targets; Cell surface antigens (CDs); immune checkpoint / Receptors; CRISPR gene Repair and knock-IN lentivirus; CRISPR knockout lentivirus;
CAR-T, TCR Lentivirus	**CARs** Lentivirus: Anti-CD19 /CD20 /CD22 /BCMA /hHER2 /HLA-A2 /TGFβ; **TCRs**: MART-1/ NY-ESO1/ CD1d-α-GalCer/ TRαV3-F2A-TRβV5-6;
CRISPR Gene Editing	Preamde lentivirus express humanzied wild-type **Cas9** endonuclease, the **dCas9**, gRNAs, **CRISPR** gene editing research
Epigenomic: CRISPRi and CRISPRa	"**dCas9-Protein**" fusion Lentivirus for epigenomic modification, resulted in CRISPR interference (CRISPRi) or activation (CRISPRa).
Cell-Specific Reporter	a set of reporter lentiviruses to express a luminescence or fluorescent reporter (firefly Luciferase, Renilla luciferase, RFP or GFP fluorescent marker) under a

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Product Category	Product Description (please click into each category's page)
	tissue specific promoter
Infectious Antigens	Llentivirus that express all kinds of infectious antigens with C-term 6His-tag.
Virus Like Particles (VLP)	Lentiviral Like Particles, pseudo-typed with a different envelope proteins.
Non-integrating LV	Integration Defective Lentivirus, express different targets for transient expression without the unwanted insertional mutagenesis.
shRNA Knockdown	Knockdown verifeid and customized shRNA lentivirus for target knockdown,
microRNA lentivirus	Premade lentivirus expression human or mouse **precursor miRNA**. And **anti-miRNA** lentivector and virus for human and mouse miRNA.
Anti-miNA lentivirus	Pre-made lentivirus expression a specific anti-miRNA cassette.
Human and mouse ORFs	Premade lentivirus expressin a **human, mouse or rat** gene with RFP-Blastididin fusion dual markers.
Luciferase expression	Premade lentivirus for all kinds of luciferase protein expression**: firefly and Renilla, Red-Luc and more,** with different antibiotic selection markers.
Fluorescent Markers	Lentivirus express all commonly used fluorescent proteins: GFP, RFP, CFP, BFP YFP, niRFP, unstable GFP and others.
Luminescent Imaging	Lentivirus express Nano-Latern as Bio-probes for in vivo imaging of sub-cellular structural organization and dynamic processes in living cells and organisms
Sub-cellular Imaging	Lentivirus contain a well-defined organelle targeting signal fusioned to a fluorescent protein, great tools for live-cell imaging and for dynamic investigation of sub-cellular signal pathways.
Cytoskeleton Imaging	A fluorescent marker (GFP, RFP or CFP) fusion with a cellular structure protein, provides a convenient tool for visualization of cytoskeletal structure
Unstable GFP	Lentivirus express the the destabilized GFP (uGFP) which provides fast turnover responses in signal pathway

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Product Category	Product Description (please click into each category's page)
	assay and in knockdown / knockout detection
near-infrared RFP	The near-infrared Red fluorescent (niRFP) expression Lentiviurs provides the whole-body images with better contrast and brighter images
Fluorescent-ORF fusion	Pre-made lentivirus expression a "**GFP/RFP/CFP-ORF**" fusion target.
CRE recombinase	Premade lentivirus for expressing **nuclear permeant CRE** recombinase with different flurescent and antibiotic markers.
CRE, Flp ColorSwtich	Lentivirus expressing "LoxP-GFP-Stop-LoxP-RFP" or "FRT-GFP-Stop-FRT-RFP" cassette, used to monitor the CRE or Flp recombination event in vivo.
SEAP Reporter	lentivirus expressing SEAP under different promoters (TetCMV, EF1a, CAG, Ubc, mPGK, Actin-beta or a signal pathway responsive promoter),
TetR Repressor	Premade lentivirus expressin TetR (tetracycline regulator) protein, the repressor protein for the inducible expression system.
rtTA Expression	rtTA binds to the tetracycline operator element (TetO) in the presence of doxycycline (Dox). Used for Tet-On /OFF inducible system.
iPS factors	Premde lentivirus for human and mouse iPS **(Myc, NANOG, OCT4, SOX2, FLF4**) factors with different fluorescent and antibitoic markers
LacZ expression	Express different full length **β- galactosidase (lacZ)** with different selection markers
Negative control lentiviruses	Premade **negative control lentivirus with different markers**: serves as the negative control of lentivurs treatment, for validation of the specificity of any lentivirus target expression effects.
Other Enzyme expression	Ready-to-use lentivirus, expressing a specific enzymes with different selection markers.
Ultra titer lentivirus	Ultra-titer lentivirus used for the hard-to-transduced cells and for in vivo manipulation of sperm cells, or stem cells.