

Pre-made Lentiviral Particles for intracelular labeling: (LocLight[™] Living cell imaging lentivirus for sub-cellular localization)

Product Name	Produce Description	Product Catalog #
Nuc-GFP (Puro), Nuc-RFP (Puro), Nuc-CFP (Puro), Nuc-GFP (Neo), Nuc-RFP (Neo), Nuc-RFP (Bsd), Nuc-BFP (Puro)	Nucleus localized different fluorescent marker, with different antibiotic selection	LVP360-G-PBS LVP360-R-PBS LVP360-C-PBS LVP360-GN LVP360-RN LVP360-RN LVP360-RB LVP1495
Cyto-GFP (Puro), Cyto-RFP (Puro), Cyto-CFP (Puro), Cyto-BFP (Puro),	Cytoplasm area targeted fluorescent marker	LVP450-G LVP450-R LVP450-C LVP1496
ER-GFP (Puro), ER- <mark>RFP</mark> (Puro), ER-CFP (Puro), ER-BFP (Puro),	Endoplasmic Reticulum (ER) targeted fluorescent marker, by the C-terminal KDEL (ER retention signal).	<u>LVP606-G</u> <u>LVP606-R</u> <u>LVP606-C</u> <u>LVP1497</u>
Golgi-GFP (Bsd), Golgi-RFP (Bsd), Golgi-CFP (Bsd), Golgi-BFP (Puro),	Golgi targeted fluorescent marker, by the golgi retention signal from 1, 4-galactosyltransferase (GT).	LVP451-G LVP451-R LVP451-C LVP1498
Mito-GFP (Bsd), Mito-RFP (Bsd), Mito-CFP (Bsd), Mito-GFP (Puro), Mito-RFP (Puro), Mito-CFP (Puro), Mito-GFP (Neo), Mito-RFP (Neo), Mito-CFP (Neo), Mito-BFP (Puro),	Mitochondria targeted fluorescent marker, by the Leader sequence of E1 alpha pyruvate dehydrogenase.	LVP452-G LVP452-R LVP452-C LVP893-G LVP893-R LVP893-C LVP894-G LVP894-R LVP894-C LVP894-C LVP894-C
Nuc-membrane-GFP (Puro), Nuc-membrane-RFP (Puro), Nuc-membrane-CFP (Puro), Nuc-membrane-BFP (Puro),	Nuclear Membrane targeted fluorescent marker, by the inner nuclear membrane localization signal from lamin B membrane receptor.	LVP453-G-PBS LVP453-R LVP453-C LVP1500



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Peroxisome-GFP (Puro), Peroxisome-RFP (Puro), Peroxisome-CFP (Puro), Peroxisome-BFP (Puro),	Peroxisome targeted fluorescent marker, by the Peroxisomal C- terminal SKL targeting sequence.	<u>LVP454-G</u> <u>LVP454-R</u> <u>LVP454-C</u> <u>LVP1501</u>
Plasma-mem-GFP (Puro) Plasma-mem-RFP (Puro), Plasma-mem-CFP (Puro), Plasma-mem-BFP (Puro),	Plasma membrane targeted fluorescent marker, by ADP- ribosylation factor 6, a plasma membrane protein.	<u>LVP455-G</u> <u>LVP455-R</u> <u>LVP455-C</u>
Microtubule-GFP (Puro), Microtubule-RFP (Puro), Microtubule-CFP (Puro), Microtubule-BFP (Puro),	Microtubule targeted fluorescent marker, by microtubule-associated protein 4 (MAP4).	<u>LVP456-G</u> <u>LVP456-R</u> <u>LVP456-C</u> <u>LVP1503</u>
GFP-H2B (Puro), RFP-H2B (Puro), CFP-H2B (Puro), BFP-H2B (Puro),	mitotic chromosomes and interphase chromatin targeted fluorescent marker, by Histone 2B protein (H2B).	LVP444-G LVP444-R LVP444-C LVP1492
Lysosomes-GFP (Bsd), Lysosomes-RFP (Bsd), Lysosomes-CFP (Bsd), Lysosomes-BFP (Puro),	Lysosomes targeted fluorescent marker, by lysosomal associated membrane protein 1 (LAMP1).	LVP457-G LVP457-R LVP457-C
Endosomes-GFP (Puro), Endosomes-RFP (Puro), Endosomes-CFP (Puro), Endosomes-BFP (Puro),	Endosomes targeted fluorescent marker, by RAB5A that localized to early endosomes for endocytosis and endocytic-sorting pathways.	LVP458-G LVP458-R LVP458-C LVP1505
GFP LocLight control, RFP LocLight control, CFP LocLight control, BFP LocLight control,	Fluorescent marker fusioned with a non targeting sequence (Null), serves the non-targeted fluorescent signal controls.	<u>Null-G</u> (Puro) <u>Null-R</u> (Puro) <u>Null-C</u> (Puro) <u>Null-B (Puro)</u>

Storage: -80 °C, avoid repeat freeze/thaw cycles. Stable for 12 6 months.

1. Product Description:

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 longterm expression.

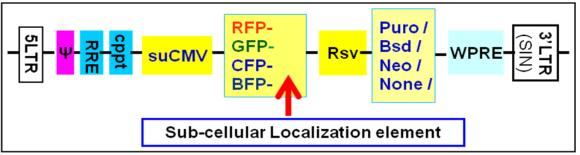
Utilizing our proprietary lentiviral vector systems, GenTarget has generated the **LocLight[™] Sub-Cellular Labeling Lentivirus** product line. These



premade lentiviruses contain a well-defined organelle targeting signal fusioned to a fluorescent protein (**GFP**, **RFP**, **CFP** or **BFP**) and expressed under our proprietary suCMV promoter. A non-targeting spacer sequence (Null) was fusioned with fluorescent which serves as the control lentivirus. The Null control virus demonstrate a evenly distributed fluorescent signal inside the cell. The lentivirus also contains an antibiotic marker (**Puromycin**, **Neomycin** or **Blasticidin**) under a Rsv promoter, or no any antibiotic selection (**None**) (see lentivector's core structure scheme below).



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The fluorescent proteins are non-toxic to cells, do not compromise cell structure, or interfere with signaling pathways. They are therefore ideal tools for live cell imaging and dynamic investigation of sub-cellular signal pathways.

Pre-made LocLight[™] sub-cellular labeling lentiviruses are extremely easy to use, and do not require any additives or substrates. Simply add the lentivirus into a mammalian cell culture. The expression of auto-fluorescent protein will be localized to the specific sub-cellular compartments at 1-3 days post-transduction and can be visualized by fluorescence microscopy. Each particle also contains a antibiotic selection which provides the selection for the transducd cells if desired. Or you can simply sort the transduced via its Fluorescent signal.

2. Key features:

- 1) **Robust and High Titer**: GenTarget's Premade Lentiviral Particles have the brightest fluorescence and the strongest transduction efficiency of any lentiviral particles on the market. Lentivirus are provided in two formats, crude lentivirus; and concentrated lentivirus in PBS which is best used for suspension cells, or "hard-to-transduced" cell types
- 2) **Long-Term Expression:** GenTarget's Premade Lentiviral Particles produce long-lasting expression of fluorescently-labeled target proteins even in hard-to-transfect cell lines such as primary and neuronal cells.





- 3) **Easy Transduction:** All premixed with 10x Polybrene (60ug/ml). Simply add the Particles to your cell culture, wait 48-92 hours, then visualize fluorescence. There is no need for any additives or medium changes.
- 4) **Multiple Colors:** Particles expressing different colors may be transduced into the same cells for **multi-color applications.**
- 5) **Easy Selection of the transduced Cells:** Use either fluorescence signal or antibiotic killing selection.
- 6) **Tested and Validated:** Each lot of Particles is validated and guaranteed to be of the highest quality.

For general questions about our ready-to-use lentiviral particles, please See *FAQs for pre-made lentiviral particles* (.pdf) on our website.

3. Transduction Protocols:

Note: Pre-made lentivirus is provided ready to use status, simply added into your cell culture. The amount of lentivirus to add depends on cell type. In general, add 50 µl of virus into one well in 24-well-plate where cell density is 50% to 75%. After 72 hours (no need to change medium), visualize positive transduction rate by fluorescence microscopy if applicable. For stable cell line generation, pass cells into medium containing antibiotic or perform fluorescence cell sorting followed by antibiotic selection. (**Note**: for suspension cells or the "hard-to-transduced" cell type, you may need to double the virus amount added.)

Day 0:

Seed cells in complete medium at the appropriate density and incubate overnight.

Note: at the time of transduction, cells should be 50%-75% confluent. For example, seed HeLa cells at 0.5×10^{5} /ml x 0.5ml in a well of a 24-well plate.

Day 1:

- Thaw the pre-made lentiviral stock at room temperature and add the appropriate amount of virus stock to obtain the desired MOI.
- Return cells to 37°C, CO₂ incubator. Do nothing.



Note: Try to avoid freezing and thawing. If you do not use all of the virus at one time, you may re-freeze the virus at -80 °C for future use; virus titer will decrease by $\sim 10\%$ for each freeze/thaw cycle.

Day 3:

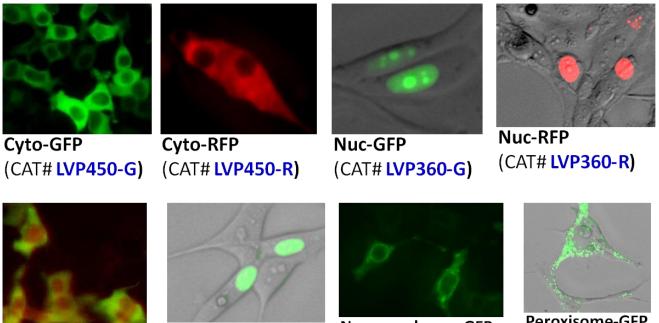
At 48hr~72hr (Depend upon cell type) after transduction, check the transduction rate by fluorescence microscopy or calculate the exact transduction rate by flow cytometry (FACS or Guava).

Day 3 + (optional):

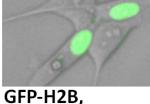
Sort transduced cells by FACS, or select by antibiotic killing. A pilot experiment should be done to determine the antibiotic's kill curve for your specific cell line (refer to the pertinent literature on generation of stable cell lines).

Note: Filter wavelength settings:

CFP filter: ~Ex436 ~Em480; GFP filter: ~Ex450-490 ~Em525; RFP filter: ~Ex550 ~Em583: BFP filter: ~Ex380 ~Em460:



Cyto-GFP + Nuc-RFP (CAT# LVP450-G + LVP360-R)



(CAT# LVP440-G)

Nuc-membrane-GFP, (CAT# LVP453-G)

Peroxisome-GFP (CAT# LVP454-G)



4. 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. Wear glove all the time when handling Lentiviral particles! Please refer CDC and NIH's guidelines for more details regarding to safety issues.

5. References:

- 1) J Virol. 2000 November; 74(22): 10778-10784.
- 2) Hum Gene Ther (2003) 14: 1089-105.
- 3) Mol Ther (2002) 6: 162-8.
- 4) NIH Guidelines for Biosafety Considerations for Research with Lentiviral Vectors. (Link).

6. Warranty:

This product is for research use only. It is warranted to meet its quality as described when used in 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.

Product Category	Product Description (please click into each category's page)
<u>Pathway</u> <u>Reporter</u>	Repoter Lentivirus for all kinds of pathway screening assays
<u>Cell</u> Immortalization	Lentivirus for cell immortalization: Large T-antigen, hTERT, EBNA1/EBNA2, HpV16-E6/E7, Adenovial E1A, Kras_G12V, HOXA9, et al.
<u>ImmunoOncology</u> <u>Research</u>	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;
<u>CAR-T, TCR</u> <u>Lentivirus</u>	CARs Lentivirus: Anti-CD19 /CD20 /CD22 /BCMA /hHER2 /HLA-A2 /TGFβ; TCRs : MART-1/ NY-ESO1/ CD1d-α-GalCer/ TRaV3-F2A-TRβV5-6;

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

Ready-to-use lentiviral Particles, "LocLight[™] Living cell imaging lentivirus" product manual, Page 6 of 9 <u>www.gentarget.com</u>; GenTarget Inc Copyrights, 2025



Product	Product Description
Category	(please click into each category's page)
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).
<u>Cell-Specific</u> <u>Reporter</u>	a set of reporter lentiviruses to express a luminescence or fluorescent reporter (firefly Luciferase, Renilla luciferase, RFP or GFP fluorescent marker) under a tissue specific promoter
<u>Infectious</u> <u>Antigens</u>	Llentivirus that express all kinds of infectious antigens with C-term 6His-tag.
<u>Virus Like</u> <u>Particles (VLP)</u>	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.
<u>shRNA</u> <u>Knockdown</u>	Knockdown verifeid and customized shRNA lentivirus for target knockdown,
<u>microRNA</u> lentivirus	Premade lentivirus expression human or mouse precursor miRNA . And anti-miRNA lentivector and virus for human and mouse miRNA.
<u>Anti-miNA</u> <u>lentivirus</u>	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



Product	Product Description	
Category	(please click into each category's page)	
Sub-cellular	Lentivirus contain a well-defined organelle targeting	
Imaging	signal fusioned to a fluorescent protein, great tools for	
	live-cell imaging and for dynamic investigation of sub-	
	cellular signal pathways.	
<u>Cytoskeleton</u>	A fluorescent marker (GFP, RFP or CFP) fusion with a	
<u>Imaging</u>	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	
	assay and in knockdown / knockout detection	
near-infrared RFP	The near-infrared Red fluorescent (niRFP) expression	
	Lentiviurs provides the whole-body images with better	
Elugroscopt ODE	contrast and brighter images Pre-made lentivirus expression a "GFP/RFP/CFP-ORF"	
Fluorescent-ORF fusion	fusion target.	
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CRE recombinase	Premade lentivirus for expressing nuclear permeant CRE recombinase with different flurescent and antibiotic	
	markers.	
CRE, Flp	Lentivirus expressing "LoxP-GFP-Stop-LoxP-RFP" or	
<u>ColorSwtich</u>	"FRT-GFP-Stop-FRT-RFP" cassette, used to monitor the	
	CRE or Flp recombination event in vivo.	
	lentivirus expressing SEAP under different promoters	
SEAP Reporter	(TetCMV, EF1a, CAG, Ubc, mPGK, Actin-beta or a signal	
	pathway responsive promoter),	
	Premade lentivirus expressin TetR (tetracycline	
<u>TetR Repressor</u>	regulator) protein, the repressor protein for the	
	inducible expression system.	
HTA Expression	rtTA binds to the tetracycline operator element (TetO) in	
rtTA Expression	the presence of doxycycline (Dox). Used for Tet-On /OFF inducible system.	
	Premde lentivirus for human and mouse iPS (Myc,	
iPS factors	NANOG, OCT4, SOX2, FLF4) factors with different	
	fluorescent and antibitoic markers	
LacZ expression	Express different full length β - galactosidase	
	(lacZ) with different selection markers	
	Premade negative control lentivirus with different	
Negative control	markers: serves as the negative control of lentivurs	
lentiviruses	treatment, for validation of the specificity of any	

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Product Category	Product Description (please click into each category's page)	
	lentivirus target expression effects.	
Other Enzyme	Ready-to-use lentivirus, expressing a specific enzymes	
expression	with different selection markers.	
<u>Ultra titer</u>	Ultra-titer lentivirus used for the hard-to-transduced	
<u>lentivirus</u>	cells and for in vivo manipulation of sperm cells, or stem	
	cells.	