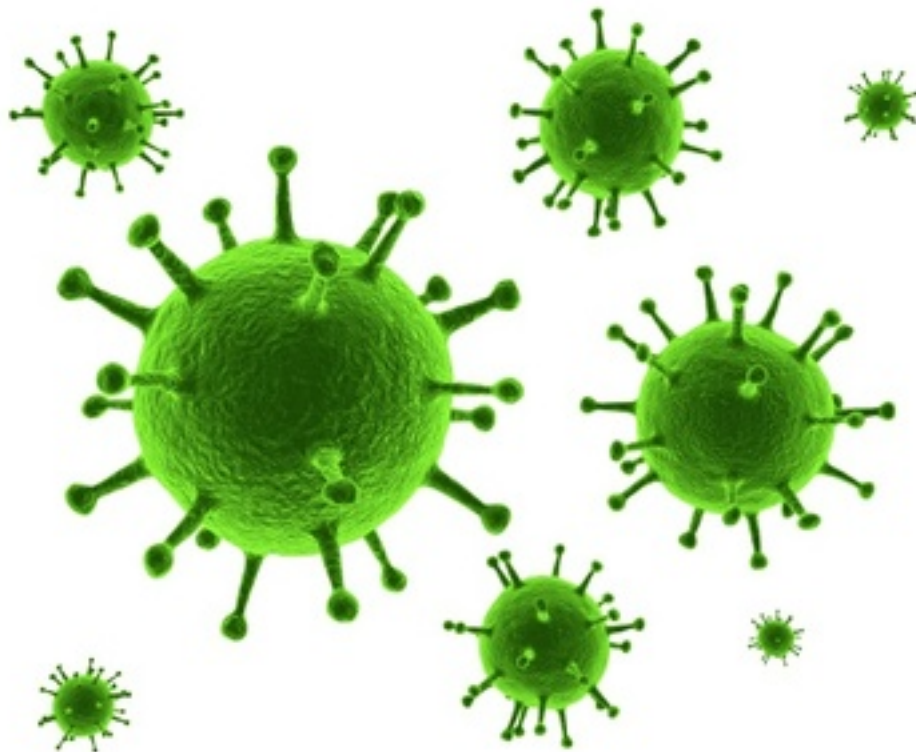


Viral Expression

Overview of Viral Gene Delivery	1
AAV (Adeno-Associated Virus) Expression	2
Adenoviral Expression	10
Lentiviral Expression	18
Retroviral Expression	25



Recombinant Viral Gene Delivery

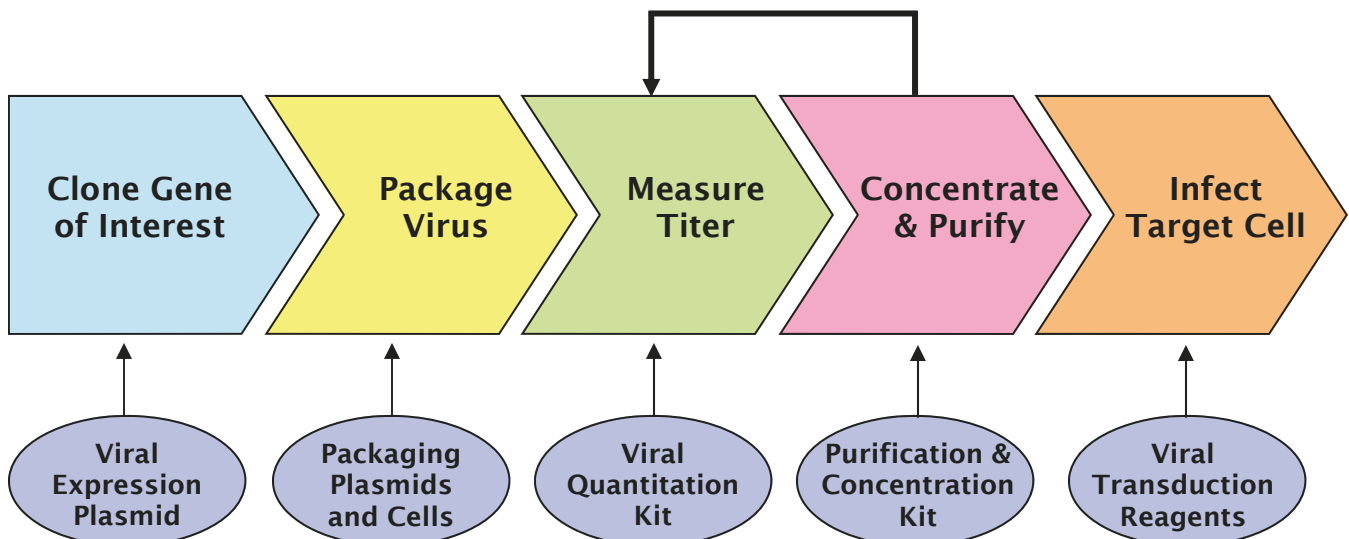
Recombinant viral vectors provide a powerful means of delivering a gene into a target cell. There are many viral vectors available, and there are pros and cons to each. Use the following table to select the best viral vector for your research.

Comparison of Viral Vectors for Gene Delivery

	Adeno-Associated Virus (AAV) (p. 41-48)	Adenovirus (p. 49-56)	Lentivirus (HIV-1, FIV, SIV) (p. 57-63)	Retrovirus (MMLV) (p. 64-72)
Gene Expression	Transient or Stable	Transient	Transient or Stable	Stable
Will Infect Dividing Cells	Yes	Yes	Yes	Yes
Will Infect Non-Dividing Cells	Yes	Yes	Yes	No
Integrates into Target Cell Genome	No*	No	Yes	Yes
Immune Response in Target Cells	Very Low	High	Low	Moderate
Relative Viral Titer	XXX	XXXX	XXX	XX
Relative Transduction Efficiency	XXX	XXXX	XXX	XX

*Native AAV can integrate, but recombinant AAV rarely does.

Typical Workflow for Viral Gene Delivery



Cell Biolabs offers kits and reagents for every step in your workflow.

Adeno-Associated Virus Kits & Reagents

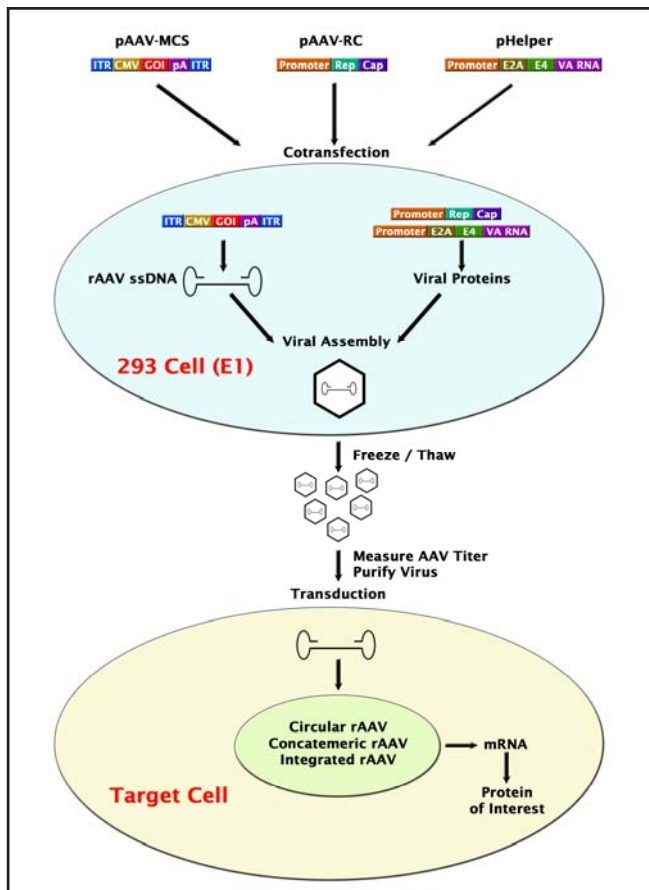
Adeno-associated virus (AAV) is less immunogenic than adenovirus or retrovirus. We offer a comprehensive line of AAV kits and reagents to ensure you get the best expression from your AAV expression studies:

- Helper Free Expression Systems
- Helper Free Packaging Systems
- Expression & Control Vectors
- Viral Packaging Cell Line
- Premade AAV Controls
- Purification Kits
- Quantitation / Titer Kit
- Transduction Kits

AAV Helper Free Systems

Production of recombinant AAV requires certain genes from the adenovirus genome, which means that an adenovirus usually needs to be present. The AAV Helper Free System eliminates the need for a helper adenovirus. Most of the required adenoviral genes (E2A, E4 and VA RNA) are provided in a pHelper plasmid, while the required E1 gene is provided by the 293 packaging cells.

- **Safer:** pHelper plasmid eliminates the need for a helper virus
- **Flexible:** Packaging vectors and expression vectors available separately or as one complete system, so you only order what you need
- **Expandable:** All plasmids are provided individually, not in a mixture, so you can amplify in competent cells



Gene Delivery using the AAV Helper Free System.

AAV Helper Free Systems are available for a variety of formats and serotypes:

- **AAV Complete Expression Systems** contain all packaging plasmids plus an expression vector and a GFP control vector: **p. 3-6**
- **AAV Packaging Systems** contain the pHelper plasmid and a serotype-specific Rep-Cap plasmid for use with your own expression construct: **p. 6**
- If you have an AAV packaging system for one serotype and want to try another, choose one of 8 different **AAV Rep-Cap Plasmids** from native serotypes 1 through 6 plus AAV-DJ and AAV-DJ/8: **p. 6**
- If you already have an AAV packaging system and need a cloning vector, choose one of 10 different **AAV Expression Vectors** available individually: **p. 7**
- Want to make a control virus? Choose one of our **AAV Control Vectors**: **p. 7**

AAV Helper Free Complete Expression Systems

AAV Helper Free Complete Expression Systems contain everything you need to produce high-titer recombinant adeno-associated virus:

- pHelper Plasmid
- Rep-Cap Plasmid (serotype specific)
- GFP Control Vector
- Choice of 10 AAV Expression Vectors:
 - Gene Expression (CMV or no promoter)
 - shRNA (U6 promoter)
 - Self complementary (scAAV)

AAV Helper Free Expression Systems are available for the following serotypes:

- Native serotypes 1-6
- AAV-DJ, engineered by DNA family shuffling to form a hybrid capsid from 8 different native serotypes; provides significantly higher infectivity rates *in vitro* (see table below)
- AAV-DJ/8, a mutant of AAV-DJ that exhibits increased uptake in brain and other tissues *in vivo*, similar to serotypes 8 and 9

Cell Line	Cell or Tissue Source	AAV-1	AAV-2	AAV-3	AAV-4	AAV-5	AAV-6	AAV-8	AAV-9	AAV-DJ	AAV-DJ/8
Huh-7	Hu Liver	13	100	2.5	0.0	0.1	10	0.7	0.0	500	0.2
HEK293	Hu Kidney	25	100	2.5	0.1	0.1	5	0.7	0.1	500	0.3
HeLa	Hu Cervix	3	100	2.0	0.1	3.7	1.0	0.2	0.1	667	0.2
HepG2	Hu Liver	3	100	16.7	0.3	1.7	5	0.3	ND	1250	0.5
Hep1A	Ms Liver	20	100	0.2	1.0	0.1	1.0	0.2	0.0	400	0.1
911	Hu Retina	17	100	11.1	0.2	0.1	17	0.1	ND	500	0.0
CHO	Hm Ovary	100	100	14.3	1.4	333	50	10.0	1.0	25000	5.0
COS	Si Kidney	33	100	33	3.3	5.0	14	2.0	0.5	500	0.3
MeWo	Hu Skin	10	100	20	0.3	6.7	10	1.0	0.2	2857	1.0
NIH3T3	Ms Fibroblasts	10	100	2.9	2.9	0.3	10	0.3	ND	500	0.1
A549	Hu Lung	14	100	20	ND	0.5	10	0.5	0.1	1000	0.1
HT1180	Hu Fibroblasts	20	100	10.0	0.1	0.3	33	0.5	0.1	333	0.2
Monocytes	Hu Primary Monocytes	1111	100	ND	ND	125	1429	ND	ND	100	ND
Immature DC	Hu Monocyte-derived DC	2500	100	ND	ND	222	2857	ND	ND	200	ND
Mature DC	Hu Monocyte-derived DC	2222	100	ND	ND	333	3333	ND	ND	100	ND

Relative Infectivity Rates of AAV Serotypes. Normalized to AAV-2 = 100. ND = Not determined.

AAV-DJ Helper Free Complete Expression Systems

Product Name	Size	Catalog Number
AAV-DJ Helper Free Expression System	1 kit	VPK-410-DJ
AAV-DJ Helper Free Bicistronic Expression System (Puro)	1 kit	VPK-415-DJ
AAV-DJ Helper Free Bicistronic Expression System (Neo)	1 kit	VPK-416-DJ
AAV-DJ Helper Free Bicistronic Expression System (Hygro)	1 kit	VPK-417-DJ
AAV-DJ Helper Free Bicistronic Expression System (GFP)	1 kit	VPK-418-DJ
AAV-DJ Helper Free Bicistronic Expression System (Blasticidin)	1 kit	VPK-419-DJ
AAV-DJ Helper Free Promoterless Expression System	1 kit	VPK-411-DJ
AAV-DJ Helper Free shRNA Expression System (Puro)	1 kit	VPK-412-DJ
AAV-DJ/Helper Free shRNA Expression System (GFP)	1 kit	VPK-413-DJ
scAAV-DJ Helper Free Expression System	1 kit	VPK-430-DJ

AAV-DJ/8 Helper Free Complete Expression Systems

Product Name	Size	Catalog Number
AAV-DJ/8 Helper Free Expression System	1 kit	VPK-410-DJ-8
AAV-DJ/8 Helper Free Bicistronic Expression System (Puro)	1 kit	VPK-415-DJ-8
AAV-DJ/8 Helper Free Bicistronic Expression System (Neo)	1 kit	VPK-416-DJ-8
AAV-DJ/8 Helper Free Bicistronic Expression System (Hygro)	1 kit	VPK-417-DJ-8
AAV-DJ/8 Helper Free Bicistronic Expression System (GFP)	1 kit	VPK-418-DJ-8
AAV-DJ/8 Helper Free Bicistronic Expression System (Blasticidin)	1 kit	VPK-419-DJ-8
AAV-DJ/8 Helper Free Promoterless Expression System	1 kit	VPK-411-DJ-8
AAV-DJ/8 Helper Free shRNA Expression System (Puro)	1 kit	VPK-412-DJ-8
AAV-DJ/8 Helper Free shRNA Expression System (GFP)	1 kit	VPK-413-DJ-8
scAAV-DJ/8 Helper Free Expression System	1 kit	VPK-430-DJ-8

AAV-1 Helper Free Complete Expression Systems

Product Name	Size	Catalog Number
AAV-1 Helper Free Expression System	1 kit	VPK-410-SER1
AAV-1 Helper Free Bicistronic Expression System (Puro)	1 kit	VPK-415-SER1
AAV-1 Helper Free Bicistronic Expression System (Neo)	1 kit	VPK-416-SER1
AAV-1 Helper Free Bicistronic Expression System (Hygro)	1 kit	VPK-417-SER1
AAV-1 Helper Free Bicistronic Expression System (GFP)	1 kit	VPK-418-SER1
AAV-1 Helper Free Bicistronic Expression System (Blasticidin)	1 kit	VPK-419-SER1
AAV-1 Helper Free Promoterless Expression System	1 kit	VPK-411-SER1
AAV-1 Helper Free shRNA Expression System (Puro)	1 kit	VPK-412-SER1
AAV-1 Helper Free shRNA Expression System (GFP)	1 kit	VPK-413-SER1
scAAV-1 Helper Free Expression System	1 kit	VPK-430-SER1

AAV-2 Helper Free Complete Expression Systems

Product Name	Size	Catalog Number
AAV-2 Helper Free Expression System	1 kit	VPK-410-SER2
AAV-2 Helper Free Bicistronic Expression System (Puro)	1 kit	VPK-415-SER2
AAV-2 Helper Free Bicistronic Expression System (Neo)	1 kit	VPK-416-SER2
AAV-2 Helper Free Bicistronic Expression System (Hygro)	1 kit	VPK-417-SER2
AAV-2 Helper Free Bicistronic Expression System (GFP)	1 kit	VPK-418-SER2
AAV-2 Helper Free Bicistronic Expression System (Blasticidin)	1 kit	VPK-419-SER2
AAV-2 Helper Free Promoterless Expression System	1 kit	VPK-411-SER2
AAV-2 Helper Free shRNA Expression System (Puro)	1 kit	VPK-412-SER2
AAV-2 Helper Free shRNA Expression System (GFP)	1 kit	VPK-413-SER2
scAAV-2 Helper Free Expression System	1 kit	VPK-430-SER2

AAV-3 Helper Free Complete Expression Systems

Product Name	Size	Catalog Number
AAV-3 Helper Free Expression System	1 kit	VPK-410-SER3
AAV-3 Helper Free Bicistronic Expression System (Puro)	1 kit	VPK-415-SER3
AAV-3 Helper Free Bicistronic Expression System (Neo)	1 kit	VPK-416-SER3
AAV-3 Helper Free Bicistronic Expression System (Hygro)	1 kit	VPK-417-SER3
AAV-3 Helper Free Bicistronic Expression System (GFP)	1 kit	VPK-418-SER3
AAV-3 Helper Free Bicistronic Expression System (Blasticidin)	1 kit	VPK-419-SER3
AAV-3 Helper Free Promoterless Expression System	1 kit	VPK-411-SER3
AAV-3 Helper Free shRNA Expression System (Puro)	1 kit	VPK-412-SER3
AAV-3 Helper Free shRNA Expression System (GFP)	1 kit	VPK-413-SER3
scAAV-3 Helper Free Expression System	1 kit	VPK-430-SER3

AAV-4 Helper Free Complete Expression Systems

Product Name	Size	Catalog Number
AAV-4 Helper Free Expression System	1 kit	VPK-410-SER4
AAV-4 Helper Free Bicistronic Expression System (Puro)	1 kit	VPK-415-SER4
AAV-4 Helper Free Bicistronic Expression System (Neo)	1 kit	VPK-416-SER4
AAV-4 Helper Free Bicistronic Expression System (Hygro)	1 kit	VPK-417-SER4
AAV-4 Helper Free Bicistronic Expression System (GFP)	1 kit	VPK-418-SER4
AAV-4 Helper Free Bicistronic Expression System (Blasticidin)	1 kit	VPK-419-SER4
AAV-4 Helper Free Promoterless Expression System	1 kit	VPK-411-SER4
AAV-4 Helper Free shRNA Expression System (Puro)	1 kit	VPK-412-SER4
AAV-4 Helper Free shRNA Expression System (GFP)	1 kit	VPK-413-SER4
scAAV-4 Helper Free Expression System	1 kit	VPK-430-SER4

AAV-5 Helper Free Complete Expression Systems

Product Name	Size	Catalog Number
AAV-5 Helper Free Expression System	1 kit	VPK-410-SER5
AAV-5 Helper Free Bicistronic Expression System (Puro)	1 kit	VPK-415-SER5
AAV-5 Helper Free Bicistronic Expression System (Neo)	1 kit	VPK-416-SER5
AAV-5 Helper Free Bicistronic Expression System (Hygro)	1 kit	VPK-417-SER5
AAV-5 Helper Free Bicistronic Expression System (GFP)	1 kit	VPK-418-SER5
AAV-5 Helper Free Bicistronic Expression System (Blasticidin)	1 kit	VPK-419-SER5
AAV-5 Helper Free Promoterless Expression System	1 kit	VPK-411-SER5
AAV-5 Helper Free shRNA Expression System (Puro)	1 kit	VPK-412-SER5
AAV-5 Helper Free shRNA Expression System (GFP)	1 kit	VPK-413-SER5
scAAV-5 Helper Free Expression System	1 kit	VPK-430-SER5

AAV-6 Helper Free Complete Expression Systems

Product Name	Size	Catalog Number
AAV-6 Helper Free Expression System	1 kit	VPK-410-SER6
AAV-6 Helper Free Bicistronic Expression System (Puro)	1 kit	VPK-415-SER6
AAV-6 Helper Free Bicistronic Expression System (Neo)	1 kit	VPK-416-SER6
AAV-6 Helper Free Bicistronic Expression System (Hygro)	1 kit	VPK-417-SER6
AAV-6 Helper Free Bicistronic Expression System (GFP)	1 kit	VPK-418-SER6
AAV-6 Helper Free Bicistronic Expression System (Blasticidin)	1 kit	VPK-419-SER6
AAV-6 Helper Free Promoterless Expression System	1 kit	VPK-411-SER6
AAV-6 Helper Free shRNA Expression System (Puro)	1 kit	VPK-412-SER6
AAV-6 Helper Free shRNA Expression System (GFP)	1 kit	VPK-413-SER6
scAAV-6 Helper Free Expression System	1 kit	VPK-430-SER6

AAV Helper Free Packaging Systems

AAV Helper Free Packaging Systems contain everything found in the Complete Expression Systems, with the exception of the AAV expression vector. This is an ideal choice if you already have an AAV construct containing your gene of interest. All plasmids are provided individually, not as a packaging mixture.

Product Name	Size	Catalog Number
AAV-DJ Helper Free Packaging System	1 kit	VPK-400-DJ
AAV-DJ/8 Helper Free Packaging System	1 kit	VPK-400-DJ-8
AAV-1 Helper Free Packaging System	1 kit	VPK-401
AAV-2 Helper Free Packaging System	1 kit	VPK-402
AAV-3 Helper Free Packaging System	1 kit	VPK-403
AAV-4 Helper Free Packaging System	1 kit	VPK-404
AAV-5 Helper Free Packaging System	1 kit	VPK-405
AAV-6 Helper Free Packaging System	1 kit	VPK-406

AAV Rep-Cap Plasmids (Serotype-Specific)

AAV Rep-Cap plasmids allow you to make recombinant AAV of a specific serotype. These plasmids are ideal if you already have an AAV packaging system for a different serotype. Just substitute one of these plasmids into your AAV Helper Free Packaging System or Expression System.

Product Name	Catalog Number
pAAV-DJ Vector	VPK-420-DJ
pAAV-DJ/8 Vector	VPK-420-DJ-8
pAAV-RC1 Vector	VPK-421
pAAV-RC2 Vector	VPK-422

Product Name	Catalog Number
pAAV-DJ Vector	VPK-420-DJ
pAAV-DJ/8 Vector	VPK-420-DJ-8
pAAV-RC1 Vector	VPK-421
pAAV-RC2 Vector	VPK-422

AAV Expression Vectors

Each of our AAV Expression Vectors may be used with any of our AAV Helper Free Systems, regardless of AAV serotype. Choose one of these vectors when you already have an AAV Packaging System but may want to use a different promoter or selection marker.

Recent Product Citation

Sen, Y. et al. (2014). TDP-43 causes differential pathology in neuronal versus glial cells in the mouse brain. *Human Mol. Genet.* 10.1093/hmg/ddt662. (VPK-410)

Product Name	Cloning Capacity	Size	Catalog Number
pAAV-MCS Expression Vector	3 kb	10 µg	VPK-410
pAAV-IRES-Puro Expression Vector	1.8 kb	10 µg	VPK-415
pAAV-IRES-Neo Expression Vector	1.6 kb	10 µg	VPK-416
pAAV-IRES-Hygro Expression Vector	1.4 kb	10 µg	VPK-417
pAAV-IRES-GFP Expression Vector	1.7 kb	10 µg	VPK-418
pAAV-IRES-Bsd Expression Vector	2 kb	10 µg	VPK-419
pAAV-MCS Promoterless Expression Vector	3.9 kb	10 µg	VPK-411
pAAV-U6-Puro Expression Vector	2.2 kb	10 µg	VPK-412
pAAV-U6-GFP Expression Vector	2.1 kb	10 µg	VPK-413
pscAAV-MCS Expression Vector	1.5 kb	10 µg	VPK-430

AAV Control Plasmids

Choose one of our AAV control vectors when you already have an AAV Packaging System and want to make a transduction control virus.

Product Name	Size	Catalog Number
pAAV-GFP Control Vector	10 µg	AAV-400
pAAV-Cre Control Vector	10 µg	AAV-401
pAAV-LacZ Control Vector	10 µg	AAV-402
pscAAV-GFP Control Vector	10 µg	AAV-410

AAV Premade Control Viruses

All AAV premade viruses are provided at a concentration of 1×10^{12} GC/mL.

Product Name	Size	Catalog Number
AAV1-GFP Control Virus	50 µL	AAV-301
AAV2 Null Control Virus	50 µL	AAV-300
AAV2-Cre Control Virus	50 µL	AAV-310
AAV2-GFP Control Virus	50 µL	AAV-302
AAV2-Luc Control Virus	50 µL	AAV-320
AAV3-GFP Control Virus	50 µL	AAV-303
AAV5-GFP Control Virus	50 µL	AAV-305
AAV6-GFP Control Virus	50 µL	AAV-306

293AAV Cell Line

Our 293AAV cell line is selected from the parental 293 cell line for larger surface area, flattened morphology, and firmer attachment to culture plates, resulting in production of higher yields of AAV.

Product Name	Size	Catalog Number
293AAV Cell Line	1 x 10 ⁶ cells	AAV-100

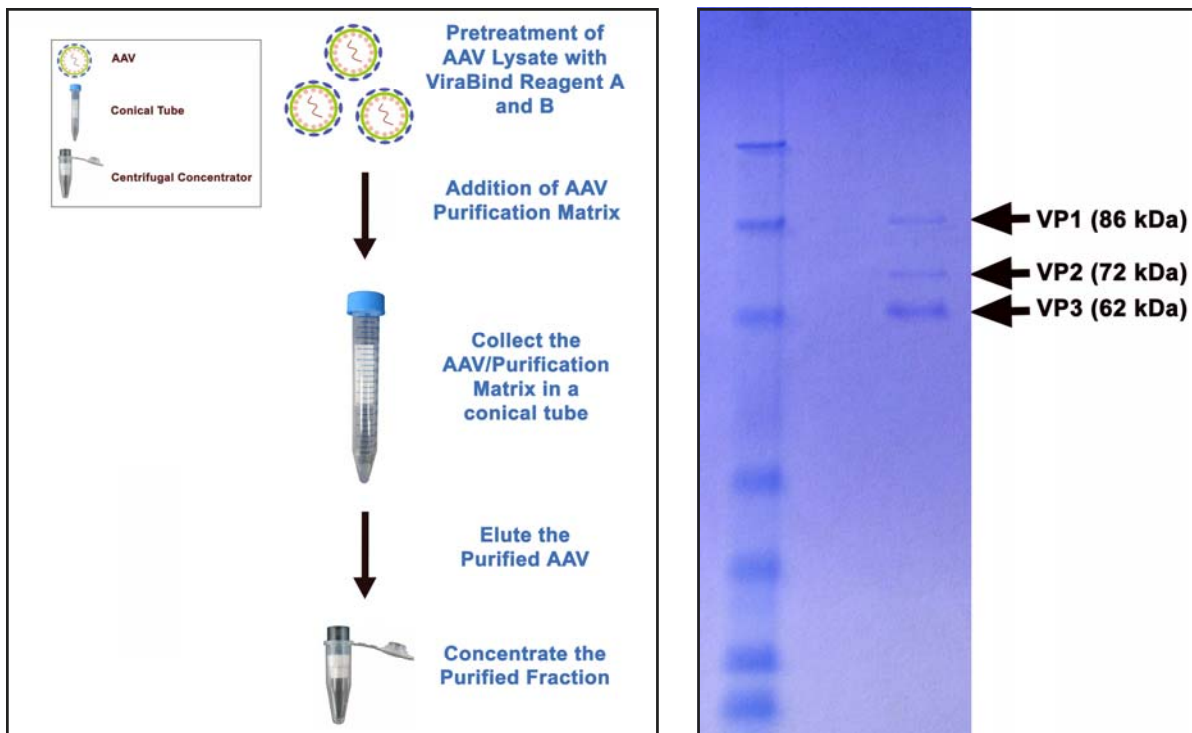
ViraBind™ AAV Purification Kits

Purification of AAV via ultracentrifugation can be tedious and time-consuming, and may result in low yields. ViraBind™ AAV Purification Kits use a one-step proprietary matrix followed by further purification and concentration using a centrifugal concentrator. The result is a higher AAV yield with high purity in a fraction of the time. Kits are suitable for AAV-2 or AAV-DJ; they will not work with other AAV serotypes.

- **High Purity:** No contamination bands as seen on SDS gel
- **Fast Results:** Obtain purified virus in about 3 hours
- **High Yields:** Recovery rate >60%

Recent Product Citation

Uchida, S. et al. (2010). Early life stress enhances behavioral vulnerability to stress through the activation of REST4-mediated gene transcription in the medial prefrontal cortex of rodents. *J. Neurosci.* **30**:15007-15018. (VPK-140)



Purification Procedure for the ViraBind™ AAV Purification Kit.

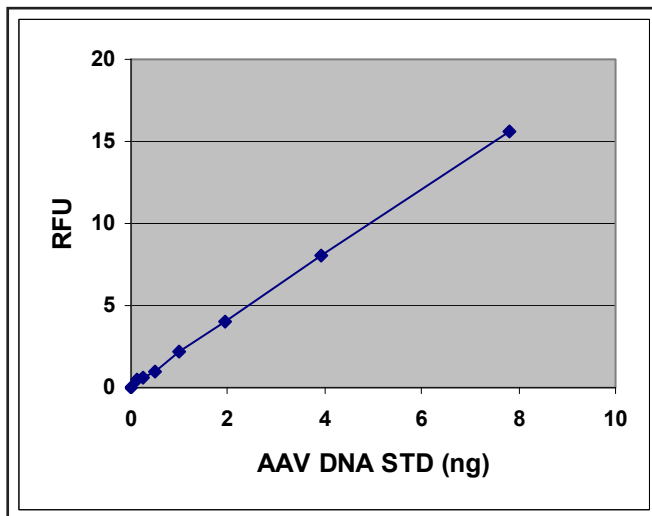
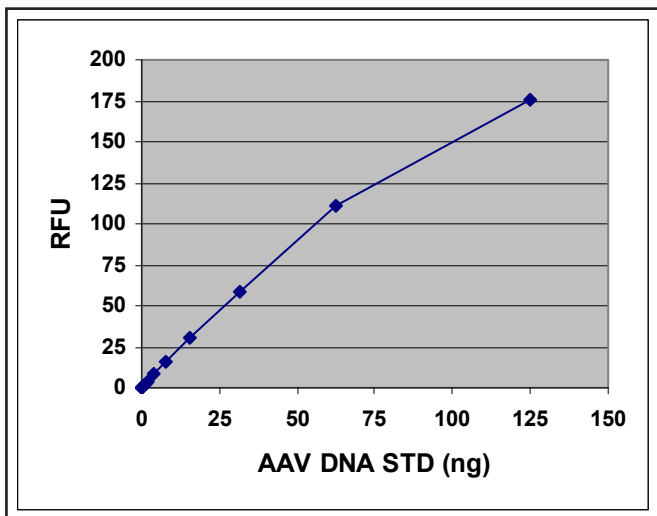
Electrophoretic Profile of Purified AAV2-GFP.

Product Name	Capacity/Prep	Size	Catalog Number
ViraBind™ AAV Purification Kit	Two 10-cm dishes	10 Preps	VPK-140
ViraBind™ AAV Purification Mega Kit	Ten 15-cm dishes	2 Preps	VPK-141
		10 Preps	VPK-141-5

QuickTiter™ AAV Quantitation Kit

Traditional AAV Quantitation by dot blot can be tedious, time consuming, and suffer from high inter-assay variability. Our QuickTiter™ AAV Quantitation Kit uses a proprietary technology to quantify AAV nucleic acid content of unpurified AAV-2 or AAV-DJ, or from purified AAV of any serotype.

- **Fast Results:** Obtain purified virus in less than 2 hours
- **High Sensitivity:** Limit of detection 1×10^9 GC/mL from unpurified supernatant or 5×10^{10} GC/mL from purified AAV



AAV-2 DNA Standard Curve. The QuickTiter™ AAV-2 DNA Standard was diluted as described in the assay protocol. Fluorescence was measured on a SpectraMax Gemini XS Fluorometer (Molecular Devices) with a 485/538 nm filter set and 530 nm cutoff.

Product Name	Capacity/Prep	Size	Catalog Number
QuickTiter™ AAV Quantitation Kit	Fluorometric	20 Assays	VPK-145

ViraDuctin™ AAV Transduction Reagent

Successful gene expression studies using AAV depend on high transduction efficiencies into host cells. Infection rates appear to be highest in S-phase cells, which can account for a very small fraction of a cell population.

Our ViraDuctin™ AAV Transduction Reagent can significantly increase the transduction efficiency of AAV vectors in both dividing and non-dividing cells. Increases are greatest in non-dividing cells, but even cells in S-phase show a noticeable increase in transduction efficiencies.

- **Higher Efficiencies:** Significantly increase rate of infection of host cells
- **Low Toxicity:** No noticeable effect on cell viability
- **Universal:** Suitable for use with both dividing and non-dividing cells

Product Name	Size*	Catalog Number
ViraDuctin™ AAV Transduction Reagent	10 Transductions	AAV-200
	50 Transductions	AAV-201

*Number of transductions performed in 35mm culture dishes. May be modified for use in culture plates or larger dishes. See product insert.

Adenoviral Expression Kits & Reagents

Recombinant adenoviruses are excellent tools for introducing genetic material into host cells, since they can infect a variety of mammalian cell types with high efficiency. They remain epichromosomal upon infection, so they are only suitable for transient gene expression. We offer a complete workflow solution to your adenoviral expression studies:

- Viral Expression Systems
- Viral Packaging Cell Line
- Premade Recombinant Adenoviruses
- Purification Kits
- Quantitation / Titer Kits
- Transduction Reagents

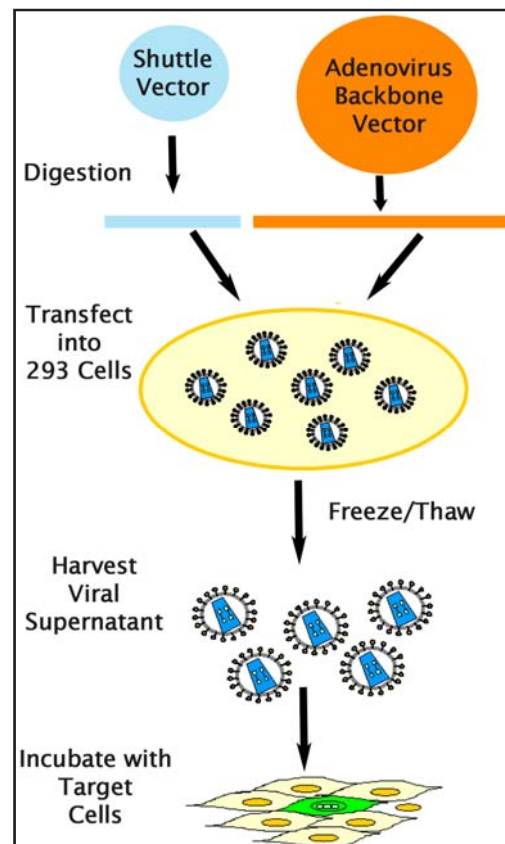
RAPAd® Adenoviral Expression Systems

Compared to other adenoviral expression systems, RAPAd® Adenoviral Expression Systems produce recombinant adenovirus in a much shorter time (about 2-3 weeks) with a substantial reduction in wild-type adenovirus. The RAPAd systems use a backbone vector from which the 5' ITR, packaging signal and E1 sequences have been removed. Additionally, serial amplification of the recombinant adenovirus does not increase the level of replication-competent adenovirus.

- **Virtually No Wild-Type Virus:** Backbone vector engineered to produce <300 wild-type plaques per 10^9 particles, compared with 10^4 - 10^6 WT plaques per 10^9 VP with most other methods
- **Faster Production:** Virus generated in 2-3 weeks compared to a few months with traditional methods
- **7 Complete Systems:** Choose CMV or RSV for gene expression, EF-1 for miRNA expression, U6 for shRNA, or clone your own promoter along with your gene of interest using our Universal system

Recent Product Citations

1. Kothari, H. et al. (2010). Cystine 186-cystine 209 disulfide bond is not essential for the procoagulant activity of tissue factor or for its de-encryption. *Blood* **115**:4273-4283. (VPK-252)
2. Li, P. et al. (2013). MicroRNA-638 is highly expressed in human vascular smooth muscle cells and inhibits PDGF-BB-induced cell proliferation and migration through targeting orphan nuclear receptor NOR1. *Cardiovasc. Res.* 10.1093/cvr/cvt082. (VPK-253)



Product Name	Promoter	Size	Catalog Number
RAPAd® Universal Adenoviral Expression System	None	1 Kit	VPK-250
RAPAd® RSV Adenoviral Expression System	RSV	1 Kit	VPK-251
RAPAd® CMV Adenoviral Expression System	CMV	1 Kit	VPK-252
RAPAd® Bicistronic Adenoviral Expression System (GFP)	CMV	1 Kit	VPK-254
RAPAd® miRNA Adenoviral Expression System	EF-1	1 Kit	VPK-253
RAPAd® shRNA Adenoviral Expression System (Puro)	U6	1 Kit	VPK-255
RAPAd® shRNA Adenoviral Expression System (GFP)	U6	1 Kit	VPK-256

293AD Cell Line for Adenoviral Packaging and Amplification

The 293AD cell line is derived from the parental 293 cell line, but has been specifically selected for adenovirus applications and offers advantages over conventional 293 cells: flattened morphology, firm attachment to culture plates, and a larger surface area for superior transfection and greater viral yields.

Recent Product Citations

- Peng, D. et al. (2011). Glutathione peroxidase 7 protects against oxidative DNA damage in oesophageal cells. *Gut* **61**:1250-1260. (AD-100)
- Kothari, H. et al. (2010). Cystine 186-cystine 209 disulfide bond is not essential for the procoagulant activity of tissue factor or for its de-encryption. *Blood* **115**:4273-4283. (AD-100)

Product Name	Size	Catalog Number
293AD Cell Line	1 x 10 ⁶ Cells	AD-100

Premade Recombinant Adenoviruses

Don't have time to make your own adenovirus? Are you studying the expression of multiple genes? Rely on our premade recombinant adenoviruses that already contain a gene of interest. All of Cell Biolabs' premade recombinant adenoviruses are provided as 50 µl aliquots at a concentration of 1 x 10¹¹ viral particles/mL in TBS with 10% glycerol.

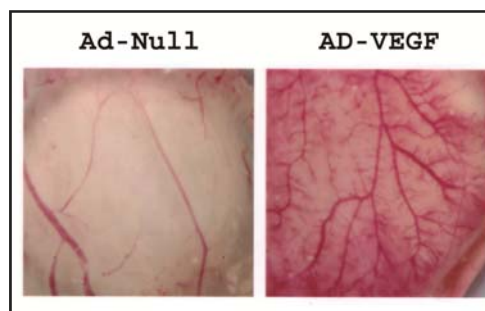
Controls and Reporter Genes

Recent Product Citations

- Matsushita, T. et al. (2009). FGFR3 promotes synchondrosis closure and fusion of ossification centers through the MAPK pathway. *Hum. Mol. Genet.* **18**:227-240. (ADV-001)
- Zhang, Z. et al. (2013). MEK inhibition leads to lysosome-mediated Na⁺/I⁻ symporter protein degradation in human breast cancer cells. *Endocr. Relat. Cancer* **20**:241-250. (ADV-002)
- Schramm, C. et al. (2012). The PTPN11 loss-of-function mutation Q510E-Shp2 causes hypertrophic cardiomyopathy by dysregulating mTOR signaling. *Am. J. Physiol. Heart Circ. Physiol.* **302**:H231-H243. (ADV-002)
- Salvati, E. et al. (2014). Evidence for G-quadruplex in the promoter of vegfr-2 and its targeting to inhibit tumor angiogenesis. *Nucleic Acids Res.* **42**:2945-2957. (ADV-004)
- Lu, D. et al. (2012). Peroxisome proliferator-activated receptor-coactivator-1alpha enhances engraftment and angiogenesis of mesenchymal stem cells in diabetic hindlimb ischemia. *Diabetes* **61**:1153-1159. (ADV-004)
- Kato, H. et al. (2011). Wnt/β-Catenin pathway in podocytes integrates cell adhesion, differentiation and survival. *J. Biol. Chem.* **286**:26003-26015. (ADV-005)

Target Name	Catalog Number
Null Control (No gene)	ADV-001
β-Galactosidase	ADV-002
Cre	ADV-005
Firefly Luciferase	ADV-008
GFP	ADV-004
SEAP (Secretory Alkaline Phosphatase)	ADV-003

Angiogenesis



Blood Vessel Formation After 3 Days. Purified Ad-Null or Ad-VEGF viruses were applied to a 10-day old CAM (chick chorioallantoic membrane). Results were visualized by stereomicroscope.

Recent Product Citations

- Kelber, J.A. et al. (2012). KRas induces a Src/PEAK1/ErbB2 kinase amplification loop that drives metastatic growth and therapy resistance in pancreatic cancer. *Cancer Res.* **72**:2554-2564. (ADV-101)
- Qiu, X. et al. (2012). Combined strategy of mesenchymal stem cell injection with vascular endothelial growth factor gene therapy for the treatment of diabetes-associated erectile dysfunction. *J. Androl.* **33**:37-44. (ADV-101)
- Stoletov, K. et al. (2010). Visualizing extravasation dynamics of metastatic tumor cells. *J. Cell Sci.* **123**:2332-2341. (ADV-101)
- Serban, D. et al. (2008). H-ras regulates angiogenesis and vascular permeability by activation of distinct downstream effectors. *Circ. Res.* **102**(11):1350-1358. (ADV-101)

Target Name	Catalog Number
HIF-1α	ADV-100
VEGF	ADV-101

Cancer/Tumor Antigens

Target Name	Catalog Number
Carbonic Anhydrase 9 (CA9)	ADV-602
Carcinoembryonic Antigen (CEA)	ADV-604
NY-ESO-1	ADV-601

Premade Recombinant Adenoviruses, continued

Cell Cycle & Transcription Regulation

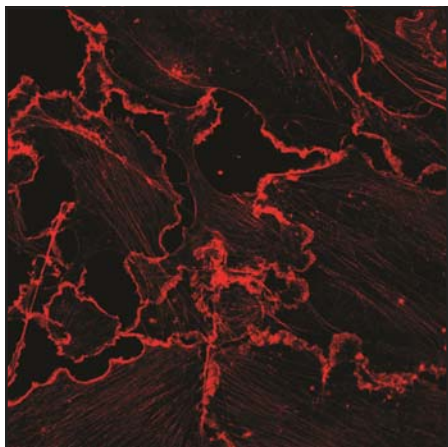
Recent Product Citation

Nguyen, N. et al. (2014). Mitsugumin 53 (MG53) ligase ubiquitinates focal adhesion kinase during skeletal myogenesis. *J. Biol. Chem.* **289**:3209-3216. (ADV-508)

Target Name	Catalog Number
DCC	ADV-504
MyoD	ADV-508

Target Name	Catalog Number
Myogenin	ADV-509
p53	ADV-501
p53 (Temperature Sensitive Mutant)	ADV-502
p68 RNA Helicase	ADV-505

Cytoskeleton Regulation / Small GTPase



Actin Cytoskeleton Staining. Cos-7 cells were infected with purified Ad-Ras V12 (ADV-146) at 50 MOI (multiplicity of infection). Membrane ruffling was visualized by staining the actin cytoskeleton with Rhodamine-coupled Phalloidin.

Target Name	Catalog Number
Cdc42	ADV-152
Cdc42 L61 (Constitutively Active)	ADV-154
Cdc42 N17 (Dominant Negative)	ADV-153
PAK1	ADV-202
PAK1 (H83L, H86L)	ADV-203
PAK1 (H83L, H86L, K299R)	ADV-205
PAK1 (K299R)	ADV-207
PAK1 (L107E, T423E)	ADV-206
PAK1 (T423E)	ADV-204
PAK1 (Kinase Domain)	ADV-209
PAK1 (Regulatory Domain)	ADV-208
Rac1	ADV-149
Rac1 L61 (Constitutively Active)	ADV-151
Rac1 N17 (Dominant Negative)	ADV-150

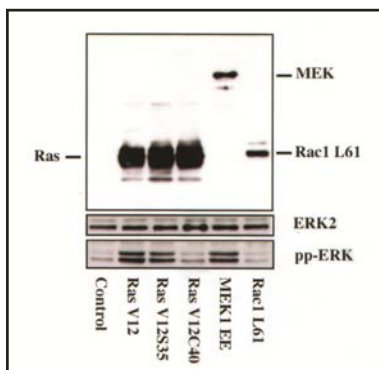
Recent Product Citations

- Black, S.A. et al. (2008). TGF β 1 stimulates connective tissue growth factor (CCN2/CTGF) expression in human gingival fibroblasts through a RhoA-independent, Rac1/Cdc42-dependent mechanism: statins with forskolin block TGF β 1-induced CCN2/CTGF expression. *J. Biol. Chem.* **283**:10835-10847. (ADV-145, ADV-150, ADV-153, ADV-156)
- Mao, Y. et al. (2012). Essential diurnal Rac1 activation during retinal phagocytosis requires α v β 5 integrin but not tyrosine kinases focal adhesion kinase or Mer tyrosine kinase. *Mol. Cell Biol.* **23**:1104-1114. (ADV-150)
- Thomas, M.A. et al. (2009). E4orf1 limits the oncolytic potential of the E1B-55K-deleted adenovirus. *J. Virol.* **83**:2406-2416. (ADV-150)
- Yu, W.-M. et al. (2009). Laminin is required for Schwann cell morphogenesis. *J. Cell Sci.* **122**:929-936. (ADV-150, ADV-153, ADV-154)
- Salvati, E. et al. (2014). Evidence for G-quadruplex in the promoter of vegfr-2 and its targeting to inhibit tumor angiogenesis. *Nucleic Acids Res.* **42**:2945-2957. (ADV-151, ADV-157)
- Cheng, Z.-J. et al. (2010). Co-regulation of caveolar and Cdc42-dependent fluid phase endocytosis by phosphocaveolin-1. *J. Biol. Chem.* **285**:15119-15125. (ADV-153)
- Neal M. et al. (2013). A critical role for TLR4 induction of autophagy in the regulation of enterocyte migration and the pathogenesis of necrotizing enterocolitis. *J. Immunol.* **190**:3541-3551. (ADV-156, ADV-157)
- Nie, J. et al. (2013). SAD-A kinase controls islet β -cell size and function as a mediator of mTORC1 signaling. *PNAS* **110**:13857-13862. (ADV-204, ADV-207)
- Nie, J. et al. (2013). Synapses of amphids defective (SAD-A) kinase promotes glucose-stimulated insulin secretion through activation of p21-activated kinase (PAK1) in pancreatic β -cells. *J. Biol. Chem.* **287**:26435-26444. (ADV-204, ADV-207)

Target Name	Catalog Number
Ras N17 (Dominant Negative)	ADV-145
Ras V12 (Constitutively Active)	ADV-146
Ras V12C40	ADV-148
Ras V12S35	ADV-147
Rho L63 (Constitutively Active)	ADV-157
Rho N19 (Dominant Negative)	ADV-156
SDF-1 α	ADV-210

Premade Recombinant Adenoviruses, continued

MAP Kinase Signaling



Immunoblot of Various Cell Signaling Targets.

HUVEC cells were infected with purified Ad-Null (ADV-001), Ad-Ras V12 (ADV-146), Ad-Ras V12S35 (ADV-147), Ad-Ras V12C40 (ADV-148), Ad-MEK1 (ADV-119), and Ad-Rac1 L61 (ADV-151) at 10 MOI (multiplicity of infection). Cell lysates were analyzed for gene expression and ERK activation.

See following page for recent citations using these adenoviruses

Target Name	Catalog Number
Cdc42	ADV-152
Cdc42 L61 (Constitutively Active)	ADV-154
Cdc42 N17 (Dominant Negative)	ADV-153
ERK2	ADV-112
ERK2 (Dominant Negative)	ADV-113
ERK5 (BMK1)	ADV-116
ERK5 (Dominant Negative)	ADV-117
Interferon- γ	ADV-103
Interleukin-2	ADV-102
JNK1	ADV-114
JNK1 (Dominant Negative)	ADV-115
MAPKAPK2	ADV-137
MAPKAPK2 (Dominant Negative)	ADV-138
MAPKAPK2 (Constitutively Active)	ADV-139
MEK1 (Dominant Negative)	ADV-118
MEK1 (Constitutively Active)	ADV-119
MEK5	ADV-129
MEK5 (Dominant Negative)	ADV-130
MEK5 (Constitutively Active)	ADV-131
MEKK1	ADV-135
MEKK1 (Dominant Negative)	ADV-136
MEKK3	ADV-162

Target Name	Catalog Number
MKK3	ADV-120
MKK3 (Dominant Negative)	ADV-121
MKK3 (Constitutively Active)	ADV-122
MKK4 (Dominant Negative)	ADV-160
MKK4 (Constitutively Active)	ADV-161
MKK6	ADV-123
MKK6 (Dominant Negative)	ADV-124
MKK6 (Constitutively Active)	ADV-125
MKK7	ADV-126
MKK7 (Dominant Negative)	ADV-127
MKK7 (Constitutively Active)	ADV-128
myr-Rac1	ADV-163
p38 α	ADV-104
p38 α (Dominant Negative)	ADV-105
p38 β	ADV-106
p38 β (Dominant Negative)	ADV-107
p38 γ	ADV-108
p38 γ (Dominant Negative)	ADV-109
p38 δ (Dominant Negative)	ADV-111
PRAK (Dominant Negative)	ADV-141
Rac1	ADV-149
Rac1 L61 (Constitutively Active)	ADV-151
Rac1 N17 (Dominant Negative)	ADV-150
Raf1	ADV-132
Raf1 (Dominant Negative)	ADV-133
Raf1 (Constitutively Active)	ADV-134
Ras N17 (Dominant Negative)	ADV-145
Ras V12 (Constitutively Active)	ADV-146
Rho L63 (Constitutively Active)	ADV-157
Rho N19 (Dominant Negative)	ADV-156
SOK	ADV-142
SOK (Dominant Negative)	ADV-143
SOK (Constitutively Active)	ADV-144
Tac-Rac1 (Membrane Targeting)	ADV-164

Premade Recombinant Adenoviruses, continued

MAP Kinase Signaling, continued

Recent Product Citations

- Harbrecht, B.G. et al. (2012). Insulin inhibits hepatocyte iNOS expression induced by cytokines by an Akt-dependent mechanism. *Am. J. Physiol. Gastrointest. Liver Physiol* **302**:G116-G122. (ADV-105)
- Jones, S.W. et al. (2009). Mitogen-activated protein kinase-activated protein kinase (MK2) modulates key biological pathways associated with OA disease pathology. *Osteoarthritis and Cartilage* **17**:124-131. (ADV-105)
- Kim, J.M. et al. (2008). Inhibition of apoptosis in *Bacteroides fragilis* enterotoxin-stimulated intestinal epithelial cells through the induction of c-IAP-2. *Eur. J. Immunol.* **38**(8):2190-2199. (ADV-105)
- Lee, J.Y. et al. (2007). Effects of transcription factor activator protein-1 on interleukin-8 expression and enteritis in response to *Clostridium difficile* toxin A. *J. Mol. Med.* **85**:1393-1404. (ADV-105, ADV-115)
- Monick, M. et al. (2008). Constitutive ERK MAPK activity regulates macrophage ATP production and mitochondrial integrity. *J. Immunol.* **180**:7485-7496. (ADV-112, ADV-113, ADV-118, ADV-119)
- Samuel, I. et al. (2008). Enteral exclusion increases MAP kinase activation and cytokine production in a model of gallstone pancreatitis. *Pancreatology* **8**(1):6-14. (ADV-113)
- Wang, X. et al. (2007). Human immunodeficiency virus protease inhibitor ritonavir inhibits cholesterol efflux from human macrophage-derived foam cells. *Am. J. of Pathology* **171**:304-314. (ADV-113)
- Jiang, S. et al. (2011). Role of inhibitory kB kinase and c-Jun NH2-terminal kinase in the development of hepatic insulin resistance in critical illness diabetes. *Am. J. Physiol. Gastrointest. Liver Physiol* **301**:G454-G463. (ADV-115)
- Zhang, Z. et al. (2013). MEK inhibition leads to lysosome-mediated Na⁺/I⁻ symporter protein degradation in human breast cancer cells. *Endocr. Relat. Cancer* **20**:241-250. (ADV-118)
- Matsushita, T. et al. (2009). FGFR3 promotes synchondrosis closure and fusion of ossification centers through the MAPK pathway. *Hum. Mol. Genet.* **18**:227-240. (ADV-119)
- Yoon, C-H. et al. (2009). Activation of p38 mitogen-activated protein kinase is required for death receptor-independent caspase-8 activation and cell death in response to sphingosine. *Mol. Cancer Res.* **7**(3):361-370. (ADV-119)
- Tan, S.H. et al. (2009). Regulation of cell proliferation and migration by TAK1 via transcriptional control of von Hippel-Lindau tumor suppressor. *J. Biol. Chem.* **284**:18047-18058. (ADV-128)
- Wu, Y. et al. (2012). ERK5 regulates glucose-induced increased fibronectin production in the endothelial cells and in the retina in diabetes. *Invest. Ophthalmol. Vis. Sci.* **53**:8405-8413. (ADV-130)
- Mao, Y. et al. (2012). Essential diurnal Rac1 activation during retinal phagocytosis requires α v β 5 integrin but not tyrosine kinases focal adhesion kinase or Mer tyrosine kinase. *Mol. Cell Biol.* **23**:1104-1114. (ADV-150)
- Yu, W.-M. et al. (2009). Laminin is required for Schwann cell morphogenesis. *J. Cell Sci.* **122**:929-936. (ADV-150)
- Neal, M. et al. (2013). A critical role for TLR4 induction of autophagy in the regulation of enterocyte migration and the pathogenesis of necrotizing enterocolitis. *J. Immunol.* **190**:3541-3551. (ADV-156, ADV-157)
- Taniguchi, C. et al. (2007). The p85a regulatory subunit of phosphoinositide 3-kinase potentiates c-Jun N-terminal kinase-mediated insulin resistance. *Mol. Cell Biol.* **27**:2830-2840. (ADV-161)

NF κ B SignalingRecent Product Citations

- Johnston, R.K. et al. (2009). β 3-integrin mediated ubiquitination activates survival signaling during myocardial hypertrophy. *FASEB J.* **23**(8):2759-2771. (ADV-302)
- Martin, A.P. et al. (2008). Lapatinib resistance in HCT116 cells is mediated by elevated MCL-1 expression and decreased BAK activation and not by ERBB receptor kinase mutation. *Mol. Pharmacol.* **74**:807-822. (ADV-302)
- Richardson, W.M. et al. (2010). Nucleotide-binding oligomerization domain-2 inhibits toll-like receptor-4 signaling in the intestinal epithelium. *Gastroenterology* **139**(3):904-917. (ADV-308, ADV-309)

Target Name	Catalog Number
I κ B- α	ADV-301
I κ B- α S32A (Dominant Negative)	ADV-302
IKK- β	ADV-305
IKK- β (Dominant Negative)	ADV-303
NOD2	ADV-308
Rel B	ADV-304

Tyrosine Kinases and PKCs

Recent Product Citations

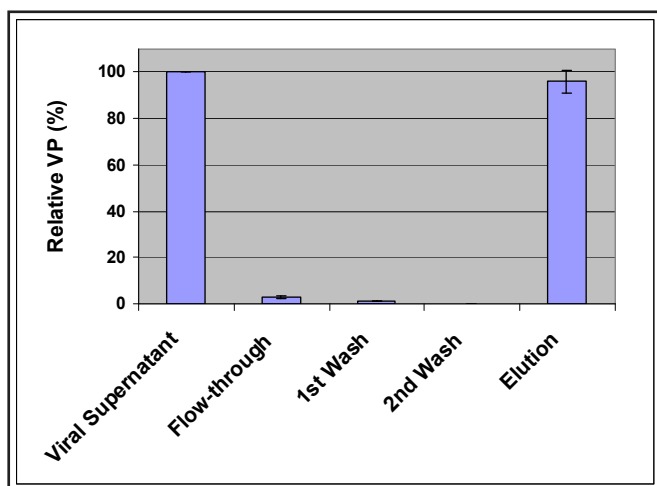
- Koh, W. et al. (2009). Formation of endothelial lumens requires a PKC-, Src-, Pak-, and Raf-kinase dependent signaling cascade downstream of Cdc42 activation. *J. Cell Sci.* **122**:1812-1822. (ADV-401, ADV-405, ADV-406)
- Lecuona, E. et al. (2009). Ubiquitination participates in the lysosomal degradation of the Na,K-ATPase in steady state conditions. *Am. J. Respir. Cell Mol. Biol.* **41**(6):617-679. (ADV-412)
- Vadasz, I. et al. (2008). AMP-activated protein kinase regulates CO₂-induced alveolar epithelial dysfunction in rats and human cells by promoting Na,K-ATPase endocytosis. *J. Clin. Invest.* **118**(2):752-762. (ADV-412)
- Briva, A. et al. (2007). High CO₂ levels impair alveolar epithelial function independent of pH. *PLoS ONE* **2**(11):e1238. (ADV-412)

Target Name	Catalog Number
CSK	ADV-405
CSK (Dominant Negative)	ADV-406
Fyn	ADV-403
Fyn (Dominant Negative)	ADV-404
PKC- α (Dominant Negative)	ADV-410
PKC- θ (Dominant Negative)	ADV-411
PKC- ζ (Dominant Negative)	ADV-412
shAkt1	ADV-417
shAkt2	ADV-418
Src	ADV-401

ViraBind™ Adenovirus Purification Kits

Purification of viruses via cesium chloride (CsCl) ultracentrifugation procedures can be tedious and time-consuming. ViraBind™ Adenovirus Purification Kits use an efficient system for quick adenoviral purification with high recovery. No ultracentrifugation is required. Kits use either a spin column or syringe filter for high purity adenovirus (see selection guide).

- **High Viral Yield:** >90% recovery
- **High Quality:** Provides quality of CsCl procedures, but in much less time
- **Faster Results:** 30 minutes (1-2 hrs for Mega kit)
- **User-Friendly Protocol:** No gradient preparation or ultracentrifugation steps



Purification of Recombinant Ad-β-Gal. Ad-β-Gal was purified according to the assay protocol. Each purification fraction was used to infect A549 cells in a 12-well plate. After 48 hr, cells were scored using our β-Galactosidase Staining Kit (p. 92).

Recent Product Citations

1. Wilkins, H. et al. (2013). Mitochondrial glutathione transport is a key determinant of neuronal susceptibility to oxidative and nitrosative stress. *J. Biol. Chem.* **288**:5091-5101. (VPK-099)
2. Wang, Y.S. et al. (2012). MicroRNA-195 regulates vascular smooth muscle cell phenotype and prevents neointimal formation. *Cardiovasc. Res.* **95**:517-526. (VPK-099)
3. Kirui, J.K. et al. (2010). Gβγ signaling promotes breast cancer cell migration and invasion. *J. Pharmacol. Exp. Ther.* **333**:393-403. (VPK-099)
4. Scallan, C. et al. (2013). An adenovirus-based vaccine with a double-stranded RNA adjuvant protects mice and ferrets against H5N1 avian influenza in oral delivery models. *Clin. Vaccine Immunol.* **20**:85-94. (VPK-100)
5. Haidar, M. et al. (2012). Integrin α2β1 mediates tyrosine phosphorylation of vascular endothelial cadherin induced by invasive breast cancer cells. *J. Biol. Chem.* **287**:32981-32992. (VPK-100)
6. Chen, F. et al. (2011). Dynamic regulation of PDX-1 and FoxO1 expression by FoxA2 in dexamethasone-induced pancreatic β-cells dysfunction. *Endocrinology* **152**:1779-1788. (VPK-100)
7. Prasad, S.S. et al. (2011). Enzymatic activities of the human AGPAT isoform 3 and isoform 5: localization of AGPAT5 to mitochondria. *J. Lipid Res.* **52**:451-462. (VPK-100)
8. Agarwal, A.K. et al. (2010). Enzymatic activity of the human 1-acylglycerol-3-phosphate-O-acyltransferase isoform 11: upregulated in breast and cervical cancers. *J. Lipid Res.* **51**:2143-2152. (VPK-100)
9. Triulzi, C. et al. (2010). Antibody-dependent natural killer cell-mediated cytotoxicity engendered by a kinase-inactive HER2 adenovirus-based vaccination mediates resistance to breast tumors. *Cancer Res.* **70**:7431-7441. (VPK-100)
10. Sen, P. et al. (2010). Zinc modulates the interaction of protein C and activated protein C with endothelial protein C receptor. *J. Biol. Chem.* **285**:20410-20420. (VPK-100)
11. Sabbatini, M. E. et al. (2010). CCK activates RhoA and Rac1 differentially through G-alpha-13 and G-alpha-q in mouse pancreatic acini. *Am. J. Physiol. Cell Physiol.* **298**:C592-C605. (VPK-100)
12. Lam, Y.W. et al. (2010). Proteomics analysis of the nucleolus in adenovirus-infected cells. *Mol. Cell. Proteomics* **9**:117-130. (VPK-100)

Selection Guide for ViraBind™ Adenovirus Purification Kits

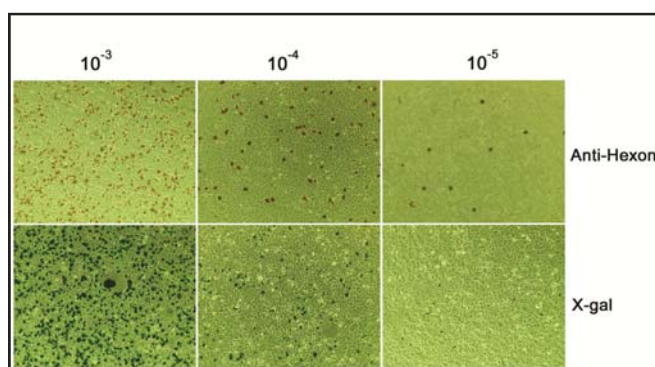
	ViraBind™ Adenovirus Miniprep Kit	ViraBind™ Adenovirus Purification Kit
Purification Method	Spin column	Syringe filter
Purification Time	30 minutes	30 minutes
Capacity/Prep (Viral Particles)	1 x 10 ¹¹ VP	2.5 x 10 ¹² VP
Capacity/Prep (Supernatant)	One T75 flask or one 10cm dish	Four T75 flasks

Product Name	Capacity/Prep	Size	Catalog Number
ViraBind™ Adenovirus Miniprep Kit	1 x 10 ¹¹ VP	10 Preps	VPK-099
ViraBind™ Adenovirus Purification Kit	2.5 x 10 ¹² VP	10 Preps	VPK-100

QuickTiter™ Adenoviral Titer & Quantitation Kits

Accurate measurement of virus titer is critical for viral gene delivery. Traditional plaque-forming unit (PFU) assays are long and have high inter-assay variability. The QuickTiter™ Adenovirus Titer Kits provide a complete system to functionally titer virus infectivity with greater accuracy in a fraction of the time. The assays may be used with any adenovirus system that can amplify in 293 cells. Assays are available for ICC staining or 96-well ELISA.

For a quick test of physical titer, our QuickTiter™ Adenovirus Quantitation Kit measures the concentration of your adenovirus prep in about one hour.



QuickTiter™ Adenovirus Titer Immunoassay Kit. 293AD cells (p. 42) were infected with different dilutions of purified Ad-β-Gal for 48 hours. Immunostaining was performed according to the assay protocol. X-gal staining was performed with β-Galactosidase Staining Kit (p. 106).

- **Faster, More Accurate and Precise:** Compared to traditional plaque-forming unit assays
- **User-Friendly Protocol:** No agar overlay steps
- **Versatile:** Recognize all 41 adenovirus serotypes

Recent Product Citations

1. Smith, M. et al. (2010). PRDM1/Blimp-1 controls effector cytokine production in human NK cells. *J. Immunol.* **185**:6058-6067. (VPK-106)
2. Reiter, C.E.N. et al. (2010). Green tea polyphenol epigallocatechin gallate reduces endothelin-1 expression and secretion in vascular endothelial cells: roles for AMP-activated protein kinase, Akt, and FOXO1. *Endocrinology* **151**:103-114. (VPK-106)
3. Wilkins, H. et al. (2013). Mitochondrial glutathione transport is a key determinant of neuronal susceptibility to oxidative and nitrosative stress. *J. Biol. Chem.* **288**:5091-5101. (VPK-109)
4. Scallan, C. et al. (2013). An adenovirus-based vaccine with a double-stranded RNA adjuvant protects mice and ferrets against H5N1 avian influenza in oral delivery models. *Clin. Vaccine Immunol.* **20**:85-94. (VPK-109)
5. Xiong, X. et al. (2012). The autophagy-related gene 14 (Atg14) is regulated by forkhead box O transcription factors and circadian rhythms and plays a critical role in hepatic autophagy and lipid metabolism. *J. Biol. Chem.* **287**:39107-39114. (VPK-109)
6. Haidar, M. et al. (2012). Integrin α2β1 mediates tyrosine phosphorylation of vascular endothelial cadherin induced by invasive breast cancer cells. *J. Biol. Chem.* **287**:32981-32992. (VPK-109)
7. Hisamitsu, T. et al. (2012). Na⁺/H⁺ exchanger 1 directly binds to calcineurin A and activates downstream NFAT signaling, leading to cardiomyocyte hypertrophy. *Mol. Cell Biol.* **32**:3265-3280. (VPK-109)
8. Lee, S. et al. (2012). Adiponectin abates diabetes-induced endothelial dysfunction by suppressing oxidative stress, adhesion molecules, and inflammation in type 2 diabetic mice. *Am. J. Heart Circ. Physiol.* **303**:H106-H115. (VPK-109)

Selection Guide for QuickTiter™ Adenoviral Quantitation Kits

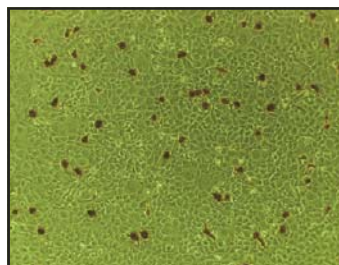
	QuickTiter™ Adenovirus Titer Immunoassay Kit	QuickTiter™ Adenovirus Titer ELISA Kit	QuickTiter™ Adenovirus Quantitation Kit
Functional or Physical Titer	Functional (Infectious units)	Functional (Infectious units)	Physical (Viral particles)
Assay Time	2.5 days	2.5 days	45-60 minutes
Assay Principle	Antibody-based	Antibody-based	Total nucleic acid content
Detection Method	Immunocytochemical staining	Colorimetric (ELISA) plate reader	Fluorescence plate reader
Key Benefit	Accuracy	Accuracy	Speed

Product Name	Detection	Size	Catalog Number
QuickTiter™ Adenovirus Titer Immunoassay Kit	ICC Staining	100 Assays	VPK-109
QuickTiter™ Adenovirus Titer ELISA Kit	Colorimetric	2 x 96 Assays	VPK-110
QuickTiter™ Adenovirus Quantitation Kit	Fluorometric	20 Assays	VPK-106

Rapid Replication Competent Adenovirus (RCA) Assay Kit

This kit uses the assay principles of the QuickTiter™ Adenovirus Titer Immunoassay Kit (see **page 47**), but is designed specifically to measure the level of replication-competent virus in your adenoviral prep.

- **Faster Results:** 2.5 days vs. 10 days with plaque assay
- **Versatile:** Recognizes all 41 adenovirus serotypes



Immunostaining of Wild Type Ad5 using the Rapid RCA Assay Kit.

Product Name	Detection	Size	Catalog Number
Rapid RCA Assay Kit	ICC Staining	30 Assays	VPK-111
		5 x 30 Assays	VPK-111-5

ViraDuctin™ Adenovirus Transduction Reagent, CAR-Independent

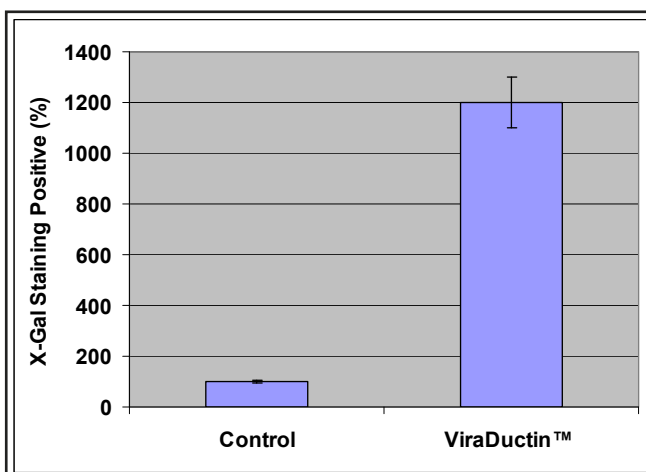
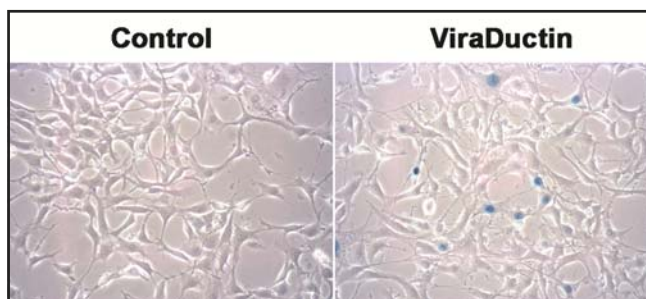
Adenovirus infection of target cells is mediated largely by the coxsackievirus-adenovirus receptor (CAR). Generally adenoviral transduction of many immortalized cell lines proceeds with a high level of efficiency. However, in many primary cells this receptor is either absent or present at extremely low-levels. This can reduce the efficiency of adenovirus transduction into your cell of choice.

ViraDuctin™ Adenovirus Transduction Reagent is designed specifically to increase the efficiency of adenoviral transduction, without regard to the level of CAR expression on the surface of the target cells.

- **Higher Transduction Efficiency:** Up to 12-fold increase in adenoviral uptake
- **User-Friendly:** Short incubation step prior to host cell infection
- **Versatile:** Ideal for target cells expressing little or no CAR, but may also improve transduction efficiency for CAR-expressing cells

Recent Product Citations

1. Haidar, M. et al. (2012). Integrin $\alpha 2\beta 1$ mediates tyrosine phosphorylation of vascular endothelial cadherin induced by invasive breast cancer cells. *J. Biol. Chem.* **287**:32981-32992.
2. Ackerman, W. et al (2008). Nuclear Factor-kappa B regulates inducible prostaglandin E synthase expression in human amnion mesenchymal cells. *Biol. Reprod.* **78**:68-76.
3. Monick, M. et al. (2008). Constitutive ERK MAPK activity regulates macrophage ATP production and mitochondrial integrity. *J. Immunol.* **180**:7485-7496.



Enhanced Transduction using ViraDuctin™ Adenovirus Transduction Reagent. Infection of NIH3T3 cells with recombinant Ad- β -gal (ADV-002). **Top:** X-gal staining under microscope. **Bottom:** scoring of infection with ViraDuctin™ reagent as a percentage of infection with control.

Product Name	Size*	Catalog Number
ViraDuctin™ Adenovirus Transduction Reagent (CAR-Independent)	10 Transductions	AD-200
	50 Transductions	AD-201

*Based on using 6-well plates or 35mm culture dishes; may also be used with 96-,24- or 12-well plates or 60mm or 100mm dishes.

Lentiviral Expression Kits & Reagents

As a sub-class of retroviruses, lentiviruses based on HIV-1 have the unique advantage of being able to infect both proliferating and non-proliferating cells, and they can be used for both transient and stable gene expression.

We offer a complete workflow solution to your lentiviral expression studies:

- Expression Systems & Vectors
- Premade Controls
- Viral Host Cell Line
- Concentration / Purification Kits
- Quantitation / Titer Kits
- Transduction Reagents

ViraSafe™ Lentiviral Expression Systems

Lentiviruses based on HIV-1 may infect both dividing and non-dividing cells. Recently developed third-generation lentiviral expression systems have reduced the risk of creating replication-competent virus upon recombination, but the risk is still present.

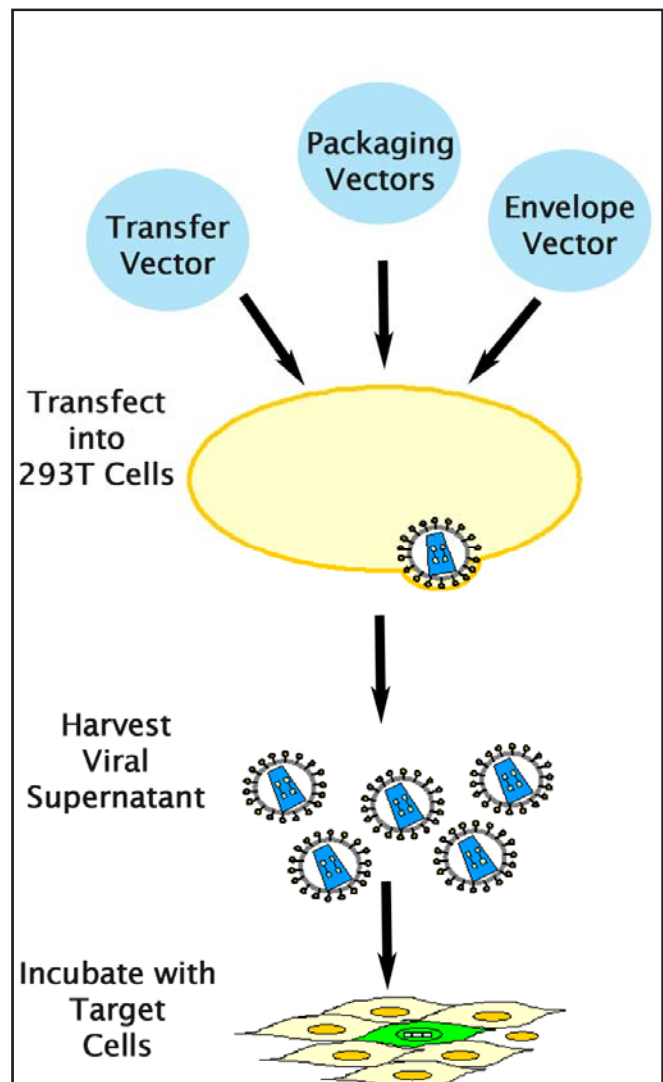
Our ViraSafe™ Lentiviral Expression Systems provide a safer and more flexible method to package your lentivirus, even compared to other third-generation lentivirus systems.

- **Safer:** 80-90% less sequence homology compared to other 3rd-generation lentiviral systems; ecotropic systems provide even more safety*
- **High Titters:** Incorporates elements that provide titers comparable to other 3rd-generation systems
- **Flexible:** Packaging vectors provided separately for increased safety and optimization of vector ratios

*Lentiviruses made with a ViraSafe™ Ecotropic Expression System will only readily infect mouse and rat cells. Pantropic lentiviruses are VSVG-pseudotyped and may infect cells of any species.

ViraSafe™ Lentiviral Technology is available in three formats (see next two pages for ordering information):

- **Complete Expression Systems:** Include 3 packaging plasmids, expression vector and control vector
- **Packaging Systems:** Include the 3 individual packaging plasmids; ideal if you already have a 3rd-generation lentiviral expression construct
- **Expression Vectors:** 11 cloning vectors to choose from; compatible with any 2nd or 3rd generation packaging system, but produce the highest titers with the ViraSafe™ packaging system



Lentivirus Production using the ViraSafe™ Lentiviral Expression System.

ViraSafe™ Lentiviral Expression Systems, continued

Complete ViraSafe™ Expression Systems include three individual packaging plasmids, an expression vector, and a control vector. Choose an ecotropic system for infection of mouse or rat cells, or a pan-tropic system to produce VSVG-pseudotyped lentivirus for infection of cells from any species.

Recent Product Citation

Davis, M. et al. (2013). RAC1P29S is a spontaneously activating cancer-associated GTPase. *PNAS* **110**:912-917. (VPK-214-PAN)

Product Name	Envelope	Size	Catalog Number
ViraSafe™ Universal Lentiviral Expression System (Promoterless)	Ecotropic	1 Kit	VPK-211-ECO
	Pantropic (VSVG)	1 Kit	VPK-211-PAN
ViraSafe™ Lentiviral Expression System (Puro)	Ecotropic	1 Kit	VPK-212-ECO
	Pantropic (VSVG)	1 Kit	VPK-212-PAN
ViraSafe™ Lentiviral Expression System (Neo)	Ecotropic	1 Kit	VPK-213-ECO
	Pantropic (VSVG)	1 Kit	VPK-213-PAN
ViraSafe™ Lentiviral Expression System (Hygro)	Ecotropic	1 Kit	VPK-214-ECO
	Pantropic (VSVG)	1 Kit	VPK-214-PAN
ViraSafe™ Lentiviral Bicistronic Expression System (Puro)	Ecotropic	1 Kit	VPK-215-ECO
	Pantropic (VSVG)	1 Kit	VPK-215-PAN
ViraSafe™ Lentiviral Bicistronic Expression System (Neo)	Ecotropic	1 Kit	VPK-216-ECO
	Pantropic (VSVG)	1 Kit	VPK-216-PAN
ViraSafe™ Lentiviral Bicistronic Expression System (Hygro)	Ecotropic	1 Kit	VPK-217-ECO
	Pantropic (VSVG)	1 Kit	VPK-217-PAN
ViraSafe™ Lentiviral Bicistronic Expression System (GFP)	Ecotropic	1 Kit	VPK-218-ECO
	Pantropic (VSVG)	1 Kit	VPK-218-PAN
ViraSafe™ Lentiviral Bicistronic Expression System (Blasticidin)	Ecotropic	1 Kit	VPK-219-ECO
	Pantropic (VSVG)	1 Kit	VPK-219-PAN
ViraSafe™ shRNA Lentiviral Expression System (Puro)	Ecotropic	1 Kit	VPK-221-ECO
	Pantropic (VSVG)	1 Kit	VPK-221-PAN
ViraSafe™ shRNA Lentiviral Expression System (GFP)	Ecotropic	1 Kit	VPK-222-ECO
	Pantropic (VSVG)	1 Kit	VPK-222-PAN

ViraSafe™ Lentiviral Packaging Systems

ViraSafe™ Packaging Systems contain 3 packaging plasmids for use with any 3rd-generation lentiviral expression vector. These systems are perfect if you already have a lentiviral construct containing your gene of interest.

Recent Product Citation

Vogt, J. et al. (2014). Protein associated with SMAD1 (PAWS1/FAM83G) is a substrate for type I bone morphogenetic protein receptors and modulates bone morphogenetic protein signaling. *Open Bio*. **4**:130210. (VPK-206)

Product Name	Envelope	Size	Catalog Number
ViraSafe™ Lentiviral Packaging System	Ecotropic	1 Kit	VPK-205
	Pantropic (VSVG)	1 Kit	VPK-206

ViraSafe™ Lentiviral Expression Vectors

These lentiviral expression vectors may be used with any 2nd or 3rd generation lentiviral packaging system, but best results are achieved when used with our ViraSafe™ Lentiviral Packaging Systems.

Recent Product Citations

1. Sankaran, V.G. et al. (2011). MicroRNA-15a and -16-1 act via MYB to elevate hemoglobin expression in human trisomy 13. *PNAS* **108**(4):1519-1524. (VPK-212)
2. Davis, M. et al. (2013). RAC1P29S is a spontaneously activating cancer-associated GTPase. *PNAS* **110**:912-917. (VPK-214)

Product Name	Cloning Capacity	Size	Catalog Number
pSMPUW Universal Lentiviral Expression Vector (Promoterless)	9.4 kb	10 µg	VPK-211
pSMPUW-Puro Lentiviral Expression Vector	7.9 kb	10 µg	VPK-212
pSMPUW-Neo Lentiviral Expression Vector	7.7 kb	10 µg	VPK-213
pSMPUW-Hygro Lentiviral Expression Vector	7.4 kb	10 µg	VPK-214
pSMPUW-IRES-Puro Lentiviral Expression Vector	7.8 kb	10 µg	VPK-215
pSMPUW-IRES-Neo Lentiviral Expression Vector	7.5 kb	10 µg	VPK-216
pSMPUW-IRES-Hygro Lentiviral Expression Vector	7.3 kb	10 µg	VPK-217
pSMPUW-IRES-GFP Lentiviral Expression Vector	7.6 kb	10 µg	VPK-218
pSMPUW-IRES-Bsd Lentiviral Expression Vector	7.9 kb	10 µg	VPK-219
pSMPUW-U6-Puro Lentiviral Expression Vector	7.7 kb	10 µg	VPK-221
pSMPUW-U6-GFP Lentiviral Expression Vector	7.6 kb	10 µg	VPK-222

Lentiviral Control Plasmids

Product Name	Size	Catalog Number
pLenti-GFP Lentiviral Control Vector	10 µg	LTV-400
pSMPUW-GFP-Puro Lentiviral Control Vector	10 µg	LTV-401
pSMPUW-MNDnLacZ Lentiviral Control Vector	10 µg	LTV-402
pLenti-RFP-Puro Lentiviral Control Vector	100 µL	LTV-403

Premade Reporter Lentivirus Controls

Product Name	Concentration	Size	Catalog Number
GFP Lentivirus Control	1 x 10 ⁶ TU/mL	200 µL	LTV-300
RFP Lentivirus Control	1 x 10 ⁶ TU/mL	200 µL	LTV-301

293LTV Lentiviral Cell Line

Our 293LTV cell line was selected from the parental 293T cell line for firmer attachment to culture plates and larger, rounder morphology for greater lentiviral production.

Recent Product Citations

1. Rossello, R.A. et al. (2013). Mammalian genes induce partially reprogrammed pluripotent stem cells in non-mammalian vertebrate and invertebrate species. *eLife Sci.* **2**:e00036.
2. Dillahunt, S. et al. (2013). Usage of sphingosine kinase isoforms in mast cells is species and/or cell type determined. *J. Immunol.* **190**:2058-2067.

Product Name	Size	Catalog Number
293LTV Cell Line	1 x 10 ⁶ Cells	LTV-100

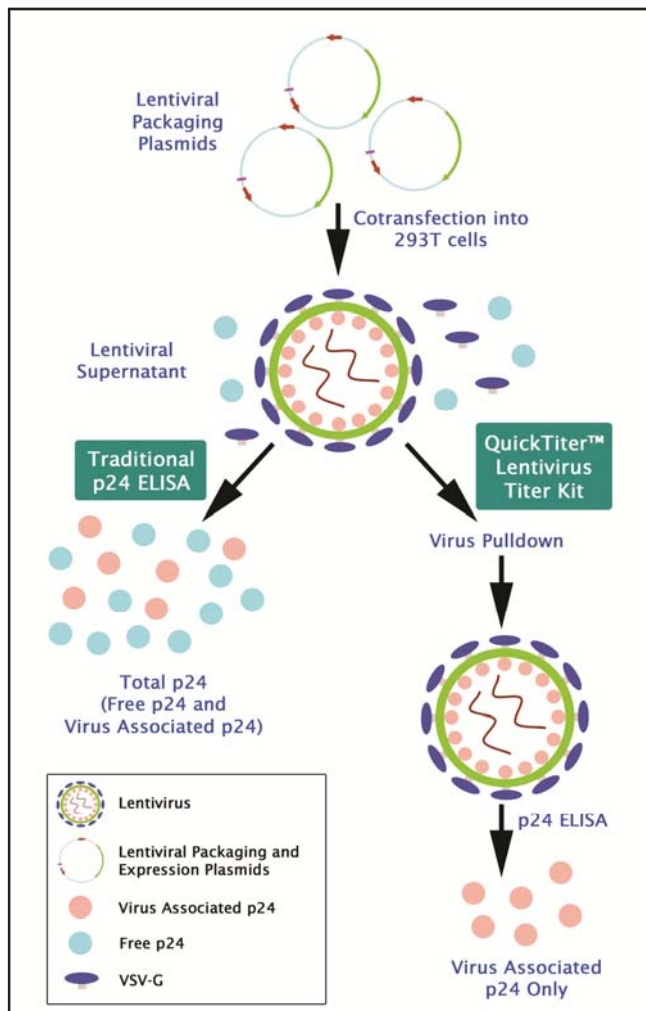
QuickTiter™ Lentivirus Titer / Quantitation Kits

Measuring lentiviral titer is important prior to infection of your target cells, and one of the most published methods is the p24 ELISA. Our traditional p24 ELISA kit provides a quick, convenient way to quantify the concentration of your HIV-1 based lentivirus.

One disadvantage of using a traditional p24 ELISA to quantify lentivirus is the overexpression of p24 during lentiviral packaging. Free p24 protein may account for a substantial portion of total p24 in lentiviral supernatant. The traditional p24 ELISA detects both virus-associated p24 and free p24 generated by 293T cells during transient transfection. Our QuickTiter™ Lentivirus Titer Kit minimizes the overestimation of p24 in lentivirus supernatant. Our proprietary technology separates the lentivirus-associated p24 from free p24 protein prior to performing the ELISA.

If you need a very quick estimate of your lentiviral concentration, try the QuickTiter™ Lentivirus Quantitation Kit. This kit specifically measures the viral nucleic acid content of purified virus or unpurified viral supernatant. This method is ideal for a quick measurement of viral titer, either before or after purification of your lentivirus.

- **More Accurate:** Exclusive technology in QuickTiter™ Lentivirus Titer Kit minimizes overestimation of virus titer
- **User-Friendly:** Read results on a standard microplate reader

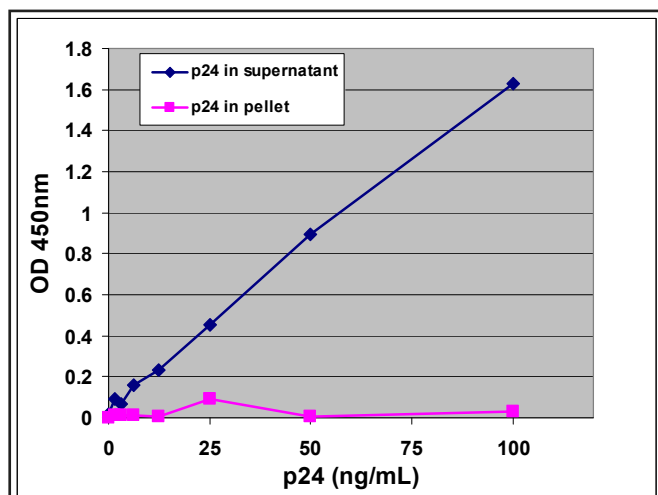


Assay Principle for the QuickTiter™ Lentivirus Titer Kit. Lentivirus particles are packaged with p24 protein, but additional free p24 protein is present in viral supernatant. A traditional p24 ELISA detects both sources of p24 which overestimates viral titer. The QuickTiter™ Lentivirus Titer Kit uses technology to pull the virus out of solution prior to quantitation for a more accurate viral titer.

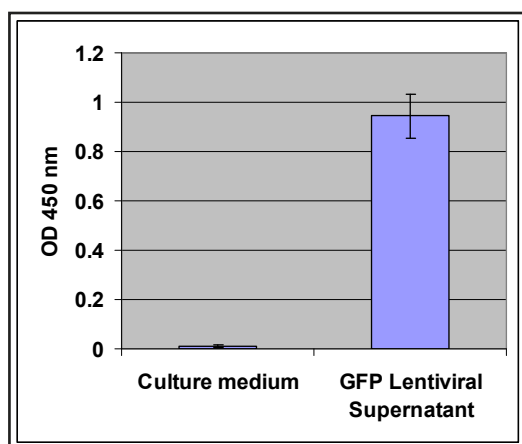
Selection Guide for QuickTiter™ Lentivirus Quantitation & Titer Kits

	QuickTiter™ Lentivirus Titer Kit (Lentivirus-Associated p24 ELISA)	QuickTiter™ Lentivirus Quantitation Kits (Traditional p24 ELISA)	QuickTiter™ Lentivirus Quantitation Kit
Assay Principle	p24 ELISA with proprietary technology to separate free p24 from viral p24	p24 ELISA	Measures nucleic acid content
Suitable Viruses	Recombinant HIV-1	Recombinant or native HIV-1	HIV-1, FIV, SIV
Detection Method	Colorimetric (ELISA) plate reader	Colorimetric (ELISA) plate reader	Fluorescence plate reader
Key Benefit	Accuracy	Most Published	Speed (45-60 min.)

QuickTiter™ Lentivirus Titer / Quantitation Kits, continued



Free p24 Does not Complex with ViraBind™ Reagents. Recombinant p24 was diluted in culture medium and treated with ViraBind™ Lentivirus Reagents A and B found in the QuickTiter™ Lentivirus Titer Kit. The amount of p24 in the supernatant and the pellet was measured according to the assay protocol.



p24 Titer of GFP Lentiviral Supernatant. GFP lentiviral construct was cotransfected with a packaging mix into 293 cells. The conditioned medium was harvested 48 hrs after transfection and used to further infect 293 cells. The p24 level of the diluted lentiviral supernatant (1:10 dilution) was determined as described in the assay protocol.

Recent Product Citations

- Belaner, K. et al. (2013). Binding of RNA by APOBEC3G controls deamination-independent restriction of retroviruses. *J. Exp. Biol.* **216**:2213-2220. (VPK-107)
- Yu, X. et al. (2012). Identification of Hepatitis B virus inhibitors targeting different aspects of infection using a cell-based assay. *Antimicrob. Agents Chemother.* **56**:6109-6120. (VPK-107)
- Walker, K. et al. (2012). Depletion of GGA1 and GGA3 mediates postinjury elevation of BACE1. *J. Neurosci.* **32**:10423-10437. (VPK-107)
- Zhou, B. et al. (2012). Interactions between β -catenin and Transforming growth factor- β signaling pathways mediate epithelial-mesenchymal transition and are dependent on the transcriptional co-activator cAMP-response element-binding protein (CREB)-binding. *J. Biol. Chem.* **287**:7026-7038. (VPK-107)
- Nedelec, A.D. et al. (2012). Noonan Syndrome-causing SHP2 mutants inhibit insulin-like growth factor 1 release via growth hormone-induced ERK hyperactivation, which contributes to short stature. *PNAS* **109**:4257-4262. (VPK-107)
- Lavender, H. et al. (2012). In vitro characterization of the activity of PF-05095808, a novel biological agent for Hepatitis C virus therapy. *Antimicrob. Agents Chemother.* **56**:1364-1375. (VPK-107)
- Keck, Z.Y. et al. (2011). Mapping a region of Hepatitis C virus E2 that is responsible for escape from neutralizing antibodies and a core CD81-binding region that does not tolerate neutralization escape mutations. *J. Virol.* **85**:10451-10463. (VPK-107)
- Sanchez-Antequera, Y. et al. (2011). Magslectofection: an integrated method of nanomagnetic separation and genetic modification of target cells. *Blood* **117**:e171-e181. (VPK-107)
- Yi, S.H. et al. (2014). Foxa2 acts as a co-activator potentiating expression of the Nurr1-induced DA phenotype via epigenetic regulation. *Development* **141**:761-772. (VPK-108-H)
- Smith, B. et al. (2013). Targeting the PyMT oncogene to diverse mammary cell populations enhances tumor heterogeneity and generates rare breast cancer subtypes. *Genes & Cancer* **10**.1177/1947601913475359. (VPK-108-H)
- Iftikhar, M. et al. (2011). Lysyl oxidase-like-2 (LOXL2) is a major isoform in chondrocytes and is critically required for differentiation. *J. Biol. Chem.* **286**:909-918. (VPK-108-H)
- Agrawal-Gamse, C. et al. (2010). Yeast-elicited cross-reactive to HIV Env glycans efficiently neutralize virions expressing exclusively high mannose N-linked glycans. *J. Virol.* **85**(1):470-480. (VPK-108-H)
- Rossello, R.A. et al. (2013). Mammalian genes induce partially reprogrammed pluripotent stem cells in non-mammalian vertebrate and invertebrate species. *eLife Sci.* **2**:e00036. (VPK-112)
- Fan, X. et al. (2012). Transient, inducible, placenta-specific gene expression in mice. *Endocrinology* **153**:5637-5644. (VPK-112)
- Veeraraghavalu, K. et al. (2010). Presenilin 1 mutants impair the self-renewal and differentiation of adult murine subventricular zone-neuronal progenitors via cell-autonomous mechanisms involving notch signaling. *J. Neurosci.* **30**:6903-6915. (VPK-112)

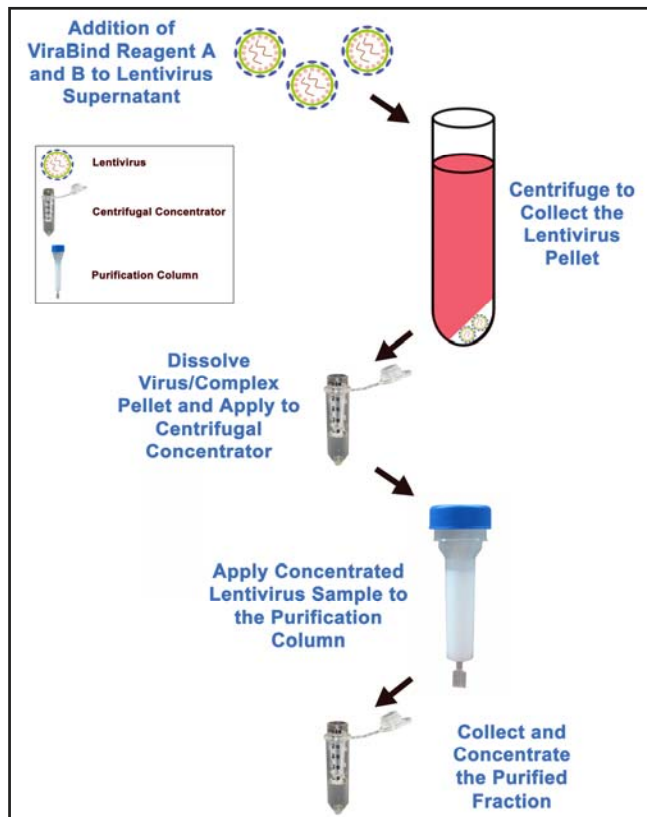
Product Name	Detection	Size	Catalog Number
QuickTiter™ Lentivirus Titer Kit (Lentivirus-Associated HIV p24 ELISA)	Colorimetric	96 Assays	VPK-107
		5 x 96 Assays	VPK-107-5
QuickTiter™ Lentivirus Quantitation Kit (HIV-1 p24 ELISA)	Colorimetric	96 Assays	VPK-108-H
		5 x 96 Assays	VPK-108-H-5
QuickTiter™ Lentivirus Quantitation Kit	Fluorometric	20 Assays	VPK-112

ViraBind™ Lentivirus Concentration & Purification Kits

Ultracentrifugation methods used for lentiviral supernatants are tedious and time-consuming and usually only partially purify your virus. Alternatively, filter-based methods have low sample capacity and do not substantially concentrate the virus.

ViraBind™ Lentivirus Concentration and Purification Kits produce purified lentivirus with extremely high titer without the need for ultracentrifugation. The virus is pelleted from solution using our proprietary isolation technology, then resuspended in a smaller volume. The resuspended lentivirus is then applied to either a purification column or a dialysis device for purification. If desired, the purified virus may be further concentrated using a centrifugal concentrator.

- **Fast:** Obtain purified virus in about 4-6 hours with column-based kits and 10-24 hours with dialysis-based kits
- **High Titer:** Concentrate up to 500-fold to as high as 10^8 - 10^{10} TU/ml, sufficient for *in vivo* studies
- **High Yield:** Recover >60%



Lentivirus Concentration and Purification Procedure.

Selection Guide for Lentivirus Concentration & Purification Kits

	ViraBind™ Lentivirus Concentration and Purification Kit	ViraBind™ PLUS Lentivirus Concentration and Purification Kit	ViraBind™ PLUS Lentivirus Concentration and Purification Mega Kit
Purification Method	Proprietary Reagent Cocktail + Purification Column	Proprietary Reagent Cocktail + Dialysis	Proprietary Reagent Cocktail + Dialysis
Total Time	6-8 hours	10-24 hours	10-24 hours
Capacity per Prep (Supernatant)	100 mL	50 mL	500 mL

Product Name	Size	Catalog Number
ViraBind™ Lentivirus Concentration and Purification Kit (100 ml/prep)	2 Preps	VPK-090
	5 Preps	VPK-091
	25 Preps	VPK-091-5
ViraBind™ PLUS Lentivirus Concentration and Purification Kit (50 ml/prep)	2 Preps	VPK-095
ViraBind™ PLUS Lentivirus Concentration and Purification Mega Kit (500 ml/prep)	2 Preps	VPK-096
	10 Preps	VPK-096-5

ViraDuctin™ Lentivirus Transduction Kit

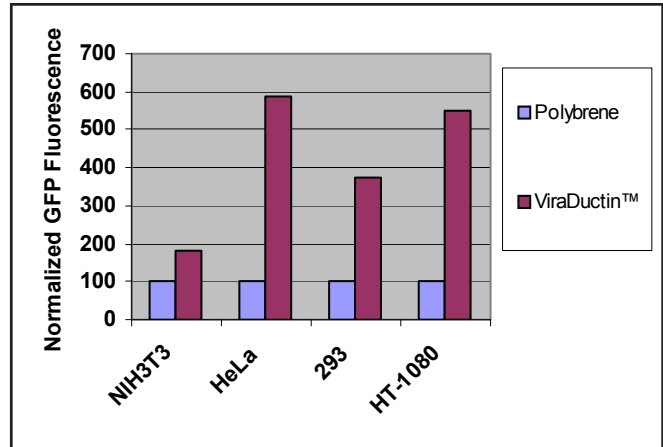
Lentivirus transduction efficiency is typically low. Additives such as Polybrene® can boost transduction efficiencies, but even then only a small fraction of lentiviral vectors can transduce many target cell lines.

Our ViraDuctin™ Lentivirus Transduction Kit provides superior transduction efficiencies in a variety of cell lines, even when compared to transductions in the presence of Polybrene®.

- **Higher Transduction Efficiency:** 2-6x higher in many cell lines compared to Polybrene
- **More Robust:** Useful for transduction of nonpermissive cells, including primary cells and stem cells

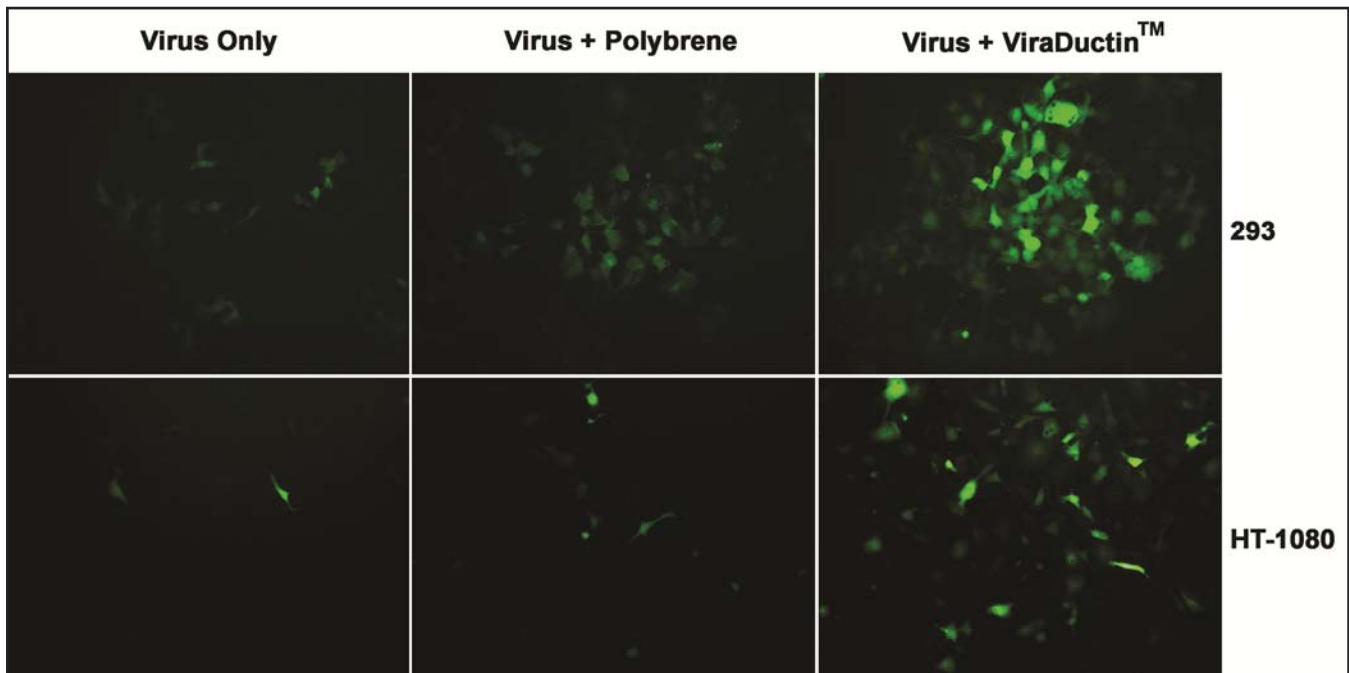
Recent Product Citations

1. Rossello, R.A. et al. (2013). Mammalian genes induce partially reprogrammed pluripotent stem cells in non-mammalian vertebrate and invertebrate species. *eLife Sci.* 2:e00036.
2. McEachron, T.A. et al. (2010). Protease-activated receptors mediate crosstalk between coagulation and fibrinolysis. *Blood* 116:5037-5044.
3. Zemskova, M. et al. (2010). p53-dependent induction of prostate cancer cell senescence by the PIM1 protein kinase. *Mol. Cancer Res.* 8:1126-1141.



Transduction Efficiencies in Various Cell Lines. NIH3T3 cells, HeLa cells, our own 293AD cells (page 36) and HT-1080 cells were each seeded at 50,000 cells/well in a 24-well plate overnight. Cells were infected with GFP lentivirus for 48 hours in the presence of Polybrene® or ViraDuctin™ Lentivirus Transduction Kit. For each cell line, fluorescence levels using the ViraDuctin™ system are depicted relative to a normalized fluorescence of 100 for Polybrene®.

Polybrene is a registered trademark of Abbott Laboratories.



Transduction of 293AD and HT-1080 Cells. 293AD cells (p. 36) and HT-1080 cells were each seeded at 50,000 cells/well in a 24-well plate overnight. Cells were infected with GFP lentivirus for 48 hours in the presence of no additive (left), Polybrene® (middle) or the ViraDuctin™ reagent cocktail (right).

Product Name	Size*	Catalog Number
ViraDuctin™ Lentivirus Transduction Kit	40 Transductions	LTV-200
	200 Transductions	LTV-201

*Based on a 24-well plate. Can also be used with 96-well, 12-well or 6-well plates, as well as 60mm or 100mm dishes. See product insert.

Retroviral Expression Kits & Reagents

Traditional retroviral vectors based on MMLV are useful for integrating genetic material into the host cell genome. However, retrovirus titer tends to be significantly lower than that of adenovirus, which can lead to a lower infection efficiency.

Our retroviral reagents and kits incorporate technologies that increase your chances of successful retroviral expression. We offer a comprehensive solution from start to finish:

- Retroviral Expression Systems
- Retroviral Packaging Cell Lines
- Retroviral Cloning & Expression Vectors
- Gene-Specific Retroviral Vectors
- Concentration / Purification Kits
- Quantitation / Titer Kits
- Transduction Reagents

Platinum Retroviral Packaging Cell Lines

Generate high titers of recombinant retrovirus with a single plasmid transfection* using these extremely powerful, stable cell lines. Platinum Retroviral Packaging Cells are based on the 293T cell line and exhibit greater stability and produce higher yields of retroviral structure proteins, resulting in higher retroviral titers.

The Platinum cell lines were invented in the laboratory of Dr. Toshio Kitamura at the University of Tokyo and are available exclusively from Cell Biolabs. They were first described in the following paper:

Morita, S. et al. (2000). *Gene Therapy* 7:1063-1066.

Recent Product Citations

1. Schmidt, T. et al. (2013). CXCR4 promotes B cell egress from Peyer's patches. *J. Exp. Med.* 10.1084/jem.20122574. (RV-101)
2. Wahlestedt, M. et al. (2013). An epigenetic component of hematopoietic stem cell aging amenable to reprogramming into a young state. *Blood* 121:4257-4264. (RV-101)
3. Zhong, S. et al. (2013). T-cell receptor affinity and avidity defines antitumor response and autoimmunity in T-cell immunotherapy. *PNAS* 110:6973-6978. (RV-101)
4. Nam, Y.J. et al. (2013). Reprogramming of human fibroblasts toward a cardiac fate. *PNAS* 110:5588-5593. (RV-102)
5. Hrdlickova, R. et al. (2012). Alternatively spliced telomerase reverse transcriptase variants lacking telomerase activity stimulate cell proliferation. *Mol. Cell Biol.* 32:4283-4296. (RV-102)
6. Nowakowski, T. et al. (2013). MicroRNA-92b regulates the development of intermediate cortical progenitors in embryonic mouse brain. *PNAS* 110:7056-7061. (RV-103)
7. Cavnar, P.J. et al. (2012). The actin regulatory protein HS1 interacts with Arp2/3 and mediates efficient neutrophil chemotaxis. *J. Biol. Chem.* 287:25466-25477. (RV-103)

Not sure which Platinum Expression System is right for you? See the table below for a selection guide based on the host species of your target cell.

	Plat-A Cells (Amphotropic)	Plat-E Cells (Ecotropic)	Plat-GP Cells (Pantropic*)
Human	+++	N.S.	+++
Mouse	+++	+++	+++
Rat	+++	+++	+++
Monkey	+++	N.S.	+++
Cat	+++	N.S.	+++
Dog	+++	N.S.	+++
Hamster	+	N.S.	+++
Bird	N.S.	N.S.	+++
Fish	N.S.	N.S.	+++
Frog	N.S.	N.S.	+++
Insect	N.S.	N.S.	+++
Mollusk	N.S.	N.S.	+++

*Plat-GP cells must be co-transfected with a pantropic envelope protein such as VSV-G.
N.S. = Not Suitable

Suitability of Platinum Retroviral Packaging Cell Lines by Host Species.

Product Name	Size	Catalog Number
Platinum-E Retroviral Packaging Cell Line, Ecotropic	≥3 x 10 ⁶ cells	RV-101
Platinum-A Retroviral Packaging Cell Line, Amphotropic	≥3 x 10 ⁶ cells	RV-102
Platinum-GP Retroviral Packaging Cell Line, Pantropic	≥3 x 10 ⁶ cells	RV-103
pVSV-G Packaging Vector	10 µg	RV-110

Platinum Retroviral Packaging Cells and Expression Systems

Our Platinum Retroviral Expression Systems incorporate superior packaging cell lines and vector technologies to produce high-titer virus with a single plasmid transfection. Each Platinum Expression System includes one of our exclusive Platinum Packaging Cell Lines which stably express the gag and pol genes. In the Ecotropic and Amphotropic systems, the packaging cells also express the envelope protein.* Simply clone your gene of interest into the vector provided and transfect into the Platinum cells.

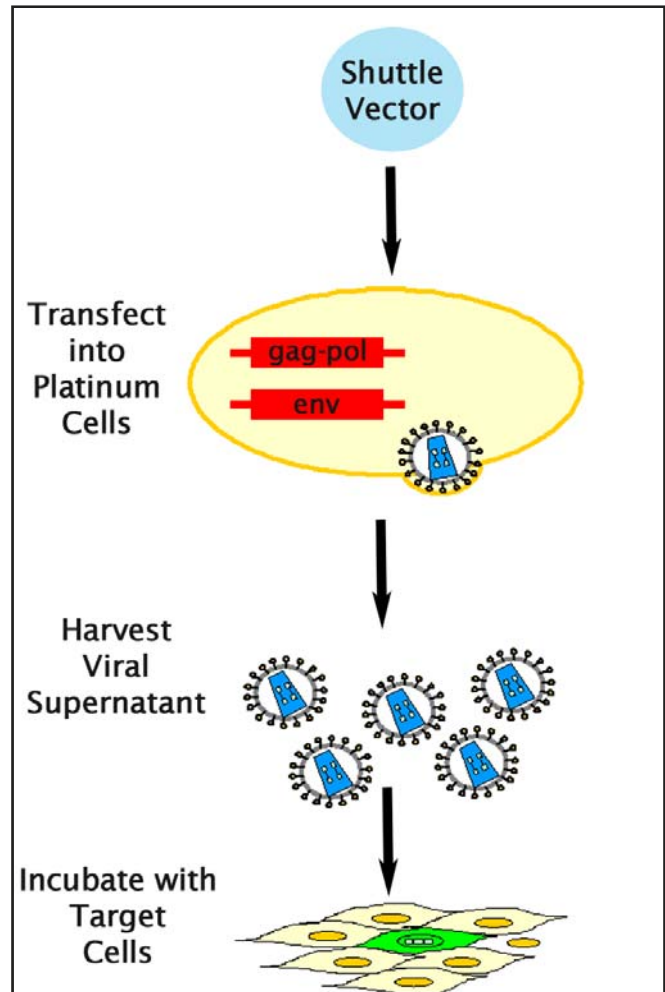
Platinum Retroviral Expression Systems contain everything you need to generate your recombinant retrovirus: packaging cell line, expression vector, and GFP control vector. Our pantropic systems also contain a VSVG envelope vector.

- **Higher Viral Yields:** Average titer 10^7 infectious units/mL with transient transfection
- **Longer Stability:** Expression up to 4 months in the presence of drug selection
- **Optimized Systems:** 3 packaging cell lines for infection of various species; 3 vector backbones (two specifically for infection of stem cells)
- **Flexible:** Order complete systems or cells and vectors separately

*Pantropic systems require co-transfection with the provided VSVG envelope vector.

Recent Product Citations

1. Aoi, N. et al. (2012). 1 α ,25-dihydroxyvitamin D3 modulates the hair-inductive capacity of dermal papilla cells: therapeutic potential for hair regeneration. *Stem Cells Trans Med.* 1:615-626. (VPK-301)
2. Wang, N. et al. (2013). Lacritin rescues stressed epithelia via rapid forkhead box O3 (FOXO3)-associated autophagy that restores metabolism. *J. Biol. Chem.* **288**:18146-18161. (VPK-302)
3. Tanaka, T. et al. (2012). Anthracycline inhibits recruitment of hypoxia-inducible transcription factors and suppresses tumor cell migration and cardiac angiogenic response in the host. *J. Biol. Chem.* **287**:34866-34882. (VPK-302)



Retrovirus Production Using the Platinum Expression Systems (Ecotropic and Amphotropic).

Product Name	Expression Vector	Packaging Cell	Catalog Number
Platinum Retroviral Expression System, Ecotropic	pMXs-Puro	Plat-E	VPK-300
Platinum Retroviral Expression System, Amphotropic	pMXs-Puro	Plat-A	VPK-301
Platinum Retroviral Expression System, Pantropic	pMXs-Puro	Plat-GP	VPK-302
Platinum ES/EC Retroviral Expression System, Ecotropic	pMCs-Puro	Plat-E	VPK-303
Platinum ES/EC Retroviral Expression System, Amphotropic	pMCs-Puro	Plat-A	VPK-304
Platinum ES/EC Retroviral Expression System, Pantropic	pMCs-Puro	Plat-GP	VPK-305
Platinum HSC Retroviral Expression System, Ecotropic	pMYs-Puro	Plat-E	VPK-306
Platinum HSC Retroviral Expression System, Amphotropic	pMYs-Puro	Plat-A	VPK-307
Platinum HSC Retroviral Expression System, Pantropic	pMYs-Puro	Plat-GP	VPK-308

Retroviral Cloning & Expression Vectors

Our Retroviral Expression Vectors are based on backbones derived from Moloney murine leukemia virus (MMLV). We offer the traditional pBABE system and the novel pMXs system, which has been shown to be useful in induced pluripotent stem cell (iPS) studies. pMYs vectors are optimal for use with hematopoietic stem cells, and pMCs vectors are optimal for ES and EC cells. All cloning vectors are supplied as 10 µg in TE buffer.

Retroviral Cloning Vectors for General Gene Expression (driven by 5' LTR)

Recent Product Citations

1. Duran, P.P. et al. (2012). UNG shapes the specificity of AID-induced somatic hypermutation. *J. Exp. Med.* **209**:1379-1389. (RTV-001-HYGRO)
2. Miyoshi, N. et al. (2010). Defined factors induce reprogramming of gastrointestinal cancer cells. *PNAS* **107**:40-45. (RTV-010)
3. Parikh, C. et al. (2012). Disruption of PH-kinase domain interactions leads to oncogenic activation of AKT in human cancers. *PNAS* **109**:19368-19373. (RTV-012)
4. Zhang, Q. et al. (2013). TNF-α impairs differentiation and function of TGF-β-induced Treg cells in autoimmune diseases through Akr and Smad3 signaling pathway. *J. Mol. Cell Biol.* 10.1093/jmcb/jms063. (RTV-013)
5. Sugatani, T. et al. (2011). A microRNA expression signature of osteoclastogenesis. *Blood* **117**:3648-3657. (RTV-014, RTV-016)

Vector Name	Cloning Capacity	Catalog Number
pBABEhygro	5.6 kb	RTV-001-HYGRO
pBABEneo	5.9 kb	RTV-003
pBABEpuro	6 kb	RTV-001-PURO
pBABEzeo	6.3 kb	RTV-004
pMXs	5.4 kb	RTV-010
pMXs-IRES-Bsd	5.6 kb	RTV-016
pMXs-IRES-GFP	5.3 kb	RTV-013
pMXs-IRES-Neo	5.2 kb	RTV-015
pMXs-IRES-Puro	5.4 kb	RTV-014
pMXs-Neo	3.8 kb	RTV-011
pMXs-Puro	4.4 kb	RTV-012
pMZs	5.3 kb	RTV-030

Retroviral Cloning Vector for miRNA

Recent Product Citation

Mansour, M. et al. (2013). The TAL1 complex targets the FBXW7 tumor suppressor by activating miR-223 in human T cell acute lymphoblastic leukemia. *J. Exp. Med.* **210**:1545-1557. (RTV-017)

Vector Name	Cloning Capacity	Catalog Number
pMXs-miR-GFP/Puro	4.2 kb	RTV-017

Retroviral Cloning Vectors with Strong Promoters for Overexpression

Vector Name	Cloning Capacity	Catalog Number
pMXs-CAG	5.2 kb	RTV-064
pMXs-CMV	5.5 kb	RTV-065
pMXs-EF1α	5.5 kb	RTV-063
pMXs-EF1-Bsd	4.2 kb	RTV-062
pMXs-EF1-GFP	3.9 kb	RTV-061
pMXs-EF1-Puro	4 kb	RTV-060
pMXs-SRα	5.4 kb	RTV-066

Retroviral Cloning Vectors for use with ES/EC Cells

Recent Product Citations

1. Malicet, C. et al. (2011). Distinct properties of human HMGN5 reveal a rapidly evolving but functionally conserved nucleosome binding protein. *Mol. Cell Biol.* **31**:2742-2755. (RTV-040)
2. Mochizuki, Y. et al. (2013). Phosphatidylinositol 3-phosphate myotubularin-related protein 6 (MTMR6) is regulated by small GTPase Rab1b in the early secretory and autophagic pathways. *J. Biol. Chem.* **288**:1009-1021. (RTV-041)

Vector Name	Cloning Capacity	Catalog Number
pMCs-IRES-GFP	5.2 kb	RTV-040
pMCs-Puro	4.3 kb	RTV-041

Retroviral Cloning Vectors for use with Hematopoietic Cells

Vector Name	Cloning Capacity	Catalog Number
pMYs	5.2 kb	RTV-020
pMYs-IRES-GFP	5.2 kb	RTV-021
pMYs-IRES-Neo	5.2 kb	RTV-023
pMYs-IRES-Puro	5.4 kb	RTV-022
pMYs-Puro	4.3 kb	RTV-024

Retroviral Cloning Vectors for shRNA

Vector Name	Cloning Capacity	Catalog Number
pMXs-U6-GFP	5 kb	RTV-071
pMXs-U6-Puro	5.1 kb	RTV-070
pMXs-U6-Puro-shGFP		RTV-055
pMXs-U6-Puro-shLuc		RTV-056

Retroviral Packaging Vectors and Cells

These constructs are ideal for researchers who prefer a traditional multi-plasmid transfection of 293 cells for packaging recombinant retrovirus.

Recent Product Citations

- Amagai, Y. et al. (2013). Stem cell factor contributes to tumorigenesis of mast cells via an autocrine/paracrine mechanism. *J. Leukoc. Biol.* **93**:245-250. (RV-110)
- Okamoto, K. et al. (2012). Dengue virus strain DEN2 16681 utilizes a specific glycochain of syndecan-2 proteoglycan as a receptor. *J. Gen. Virol.* **93**:761-770. (RV-110, RV-111)

Product Name	Size	Catalog Number
pCMV-10A1 Envelope Vector	100 µL	RV-114
pCMV-Ampho Envelope Vector	100 µL	RV-113
pCMV-Eco Envelope Vector	100 µL	RV-112
pCMV-Gag-Pol Retroviral Vector	10 µg	RV-111
pCMV-VSV-G Envelope Vector	10 µg	RV-110

293RTV Cell Line

Our 293RTV cells are derived from the 293 parental cell line, but are selected for firmer attachment to culture plates, faster growth and higher yields of retrovirus produced.

Product Name	Size	Catalog Number
293RTV Cell Line	≥1 x 10 ⁶ cells	RV-100

Gene-Specific Recombinant Retroviral Vectors

These constructs are based on backbones derived from MMLV, Vectors with GFP or stem cell factors are supplied as 10 µg of plasmid in TE buffer. All other vectors are supplied as 100 µL of bacterial glycerol stock. Product listing continues on the following pages.

Cell Cycle

Recent Product Citation

Huang, J. et al. (2009). Regulation of the leucocyte chemoattractant receptor FPR in glioblastoma cells by cell differentiation. *Carcinogenesis* **30**(2):348-355. (RTV-401)

Target Name	Vector Backbone	Catalog Number
c-Abl	pBABEpuro	RTV-402
c-Abl-TM	pBABEpuro	RTV-403
c-Abl (1-565)	pBABEpuro	RTV-404
c-Abl (1-958)	pBABEpuro	RTV-405
hTERT	pBABEhygro	RTV-007
	pBABEneo	RTV-005
	pBABEpuro	RTV-006
p53	pBABEpuro	RTV-401

Autophagy

This vector is supplied with a separate pMXs-GFP control vector at no additional cost.

Target Name	Vector Backbone	Catalog Number
GFP-LC3	pMXs	RTV-801

Reporter Genes

Recent Product Citations

- Hrdlickova, R. et al. (2012). Alternatively spliced telomerase reverse transcriptase variants lacking telomerase activity stimulate cell proliferation. *Mol. Cell Biol.* **32**:4283-4296. (RTV-002)
- Wahlestedt, M. et al. (2013). An epigenetic component of hematopoietic stem cell aging amenable to reprogramming into a young state. *Blood* **121**:4257-4264. (RTV-050)

Target Name	Vector Backbone	Catalog Number
GFP	pBABE	RTV-002
GFP	pMCs	RTV-051
GFP	pMX	RTV-050
GFP	pMYs	RTV-052
GFP-Puro	pMX	RTV-053

Gene-Specific Recombinant Retroviral Vectors, continued

MAP Kinase Signaling

Vector Name	Vector Backbone	Mutation State	Catalog Number
ERK2	pBABEhygro	Dominant Negative	RTV-109
JNK1	pBABEpuro	Dominant Negative	RTV-110
MAPKAPK2	pBABEpuro	Constitutively Active	RTV-118
	pBABEpuro	Dominant Negative	RTV-119
MAPKAPK3	pBABEpuro	Constitutively Active	RTV-120
	pBABEpuro	Dominant Negative	RTV-121
MEK1	pBABEhygro	Constitutively Active	RTV-112
	pBABEhygro	Dominant Negative	RTV-111
MKK3	pBABEpuro	Constitutively Active	RTV-114
	pBABEhygro	Dominant Negative	RTV-115
MKK6	pBABEpuro	Constitutively Active	RTV-116
	pBABEhygro	Dominant Negative	RTV-117
myr-Akt1	pWZLneo	Constitutively Active	RTV-125
p38 α	pBABEhygro	Dominant Negative	RTV-105
p38 β	pBABEhygro	Dominant Negative	RTV-106
p38 γ	pBABEhygro	Dominant Negative	RTV-107
p38 δ	pBABEhygro	Dominant Negative	RTV-108
PI3K p110 α -CAAX	pWZLneo	Constitutively Active	RTV-124
PRAK	pBABEpuro	Constitutively Active	RTV-122
	pBABEpuro	Dominant Negative	RTV-123
Raf1-CAAX	pWZLneo	Constitutively Active	RTV-113

Transcription Regulation

Target Name	Vector Backbone	Catalog Number
AUF1	pBABEpuro	RTV-305
hnRNPA0	pBABEpuro	RTV-310
hnRNP-A2	pBABEpuro	RTV-340
HuB	pBABEpuro	RTV-302
HuC	pBABEpuro	RTV-303
HuD	pBABEpuro	RTV-301
HuR	pBABEpuro	RTV-304
PABP	pBABEpuro	RTV-307
Stat5A	pMXs	RTV-330
Stat5A(1*6)	pMXs	RTV-331

Recent Product Citation

Yu, Y. et al. (2012). Bcl11a is essential for lymphoid development and negatively regulates p53. *J. Exp. Med.* **209**:2467-2483. (RTV-331)

Target Name	Vector Backbone	Catalog Number
Stat5A-IRES-GFP	pMXs	RTV-332
Stat5A(1*6)-IRES-GFP	pMXs	RTV-333
Stat5B	pMXs	RTV-334
Stat5B(1*6)	pMXs	RTV-335
TIA-1	pBABEpuro	RTV-309
TIAR	pBABEpuro	RTV-308
TTP	pBABEpuro	RTV-306

Gene-Specific Recombinant Retroviral Vectors, continued

Cytoskeleton Regulation

Recent Product Citation

Zhao, B. et al. (2012). TNF-induced osteoclastogenesis and inflammatory bone resorption are inhibited by transcription factor RBP-J. *J. Exp. Med.* **209**:2467-2483. (RTV-101)

Target Name	Vector Backbone	Mutation State	Catalog Number
Cdc42	pBABEhygro	L61	RTV-203
K-Ras	pBABEpuro	N/A	RTV-220
	pWZLhygro	Q61	RTV-221
myr-Rac1	pBABEpuro	N/A	RTV-201
		V12	RTV-206
Rac1	pBABEhygro	V12	RTV-202
N-Ras	pBABEpuro	K61	RTV-222
Rac3	pBABEhygro	V12	RTV-205
Ras	pBABEpuro	V12	RTV-101
	pBABEpuro	V12C40	RTV-104
	pBABEpuro	V12G37	RTV-103
	pBABEpuro	V12S35	RTV-102
RhoA	pBABEhygro	L63	RTV-204

iPS / Stem Cell Factors

Human iPS Genes

Target Name	Vector Backbone	Catalog Number
4-Vector Set*	pMXs	RTV-701-C
6-Vector Set**	pMXs	RTV-709-C
c-Myc	pMXs	RTV-703
Klf4	pMXs	RTV-704
Lin-28	pMXs	RTV-710
NANOG	pMXs	RTV-709
Oct-3/4	pMXs	RTV-701
Sox2	pMXs	RTV-702
p53 shRNA	pRetro	RTV-410

Mouse iPS Genes

Target Name	Vector Backbone	Catalog Number
4-Vector Set*	pMXs	RTV-705-C
6-Vector Set**	pMXs	RTV-711-C
c-Myc	pMXs	RTV-707
Klf4	pMXs	RTV-708
Lin-28	pMXs	RTV-712
NANOG	pMXs	RTV-711
Oct-3/4	pMXs	RTV-705
Sox2	pMXs	RTV-706
p53 shRNA	pRetro	RTV-400

*4-Vector sets contain individual constructs with the following genes: c-Myc, Klf4, Oct-3/4 and Sox2.

**6-Vector sets contain individual constructs with the following genes: c-Myc, Klf4, Oct-3/4, Sox2, Lin-28 and NANOG.

Proteases and Related Molecules

Target Name	Vector Backbone	Catalog Number
uPA	pBABEpuro	RTV-501
uPAR	pBABEhygro	RTV-502

Recent Product Citation

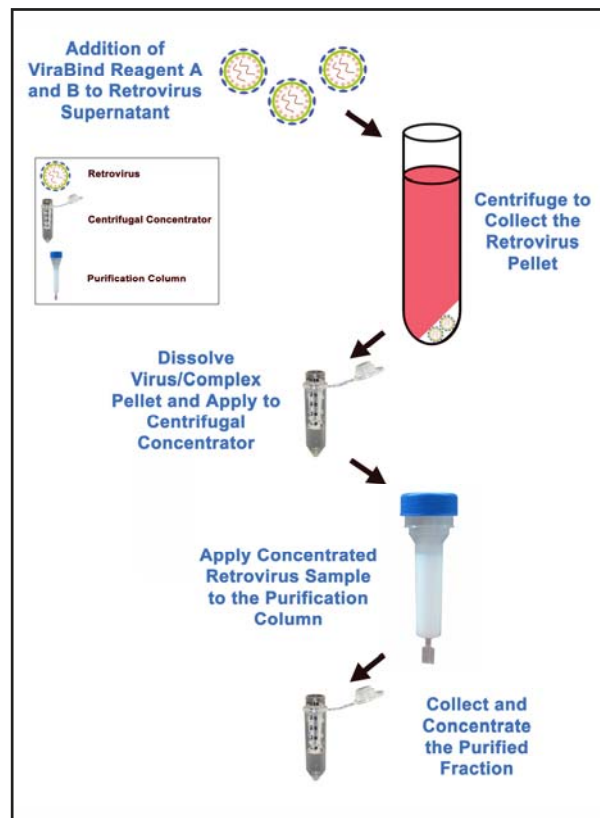
Gutova, M. et al (2008). Urokinase plasminogen activator and urokinase plasminogen activator receptor mediate human stem cell tropism to malignant solid tumors. *Stem Cells* **26**:1406-1413. (RTV-501, RTV-502)

ViraBind™ Retrovirus Concentration & Purification Kits

Ultracentrifugation methods used for lentiviral supernatants are tedious and time-consuming and usually only partially purify your virus. Alternatively, filter-based methods have low sample capacity and do not substantially concentrate the virus.

ViraBind™ Retrovirus Concentration and Purification Kits produce purified lentivirus with extremely high titer without the need for ultracentrifugation. The virus is pelleted from solution using our proprietary isolation technology, then resuspended in a smaller volume. The resuspended retrovirus is then applied to either a purification column or a dialysis device for purification. If desired, the purified virus may be further concentrated using a centrifugal concentrator.

- **Fast:** Obtain purified virus in about 4-6 hours
- **High Titer:** Concentrate 500-fold to 10^9 - 10^{10} TU/ml, sufficient for *in vivo* studies
- **High Yield:** Recover >60%
- **High Throughput:** Process greater volumes per prep than filter-based purification methods



Retrovirus Concentration and Purification Procedure.

Selection Guide for Retrovirus Concentration & Purification Kits

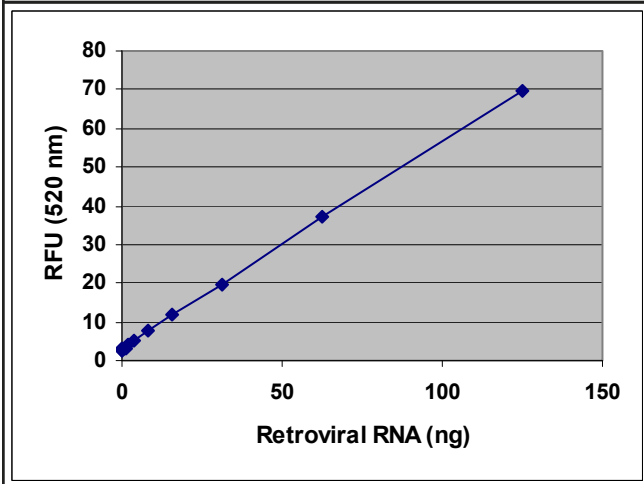
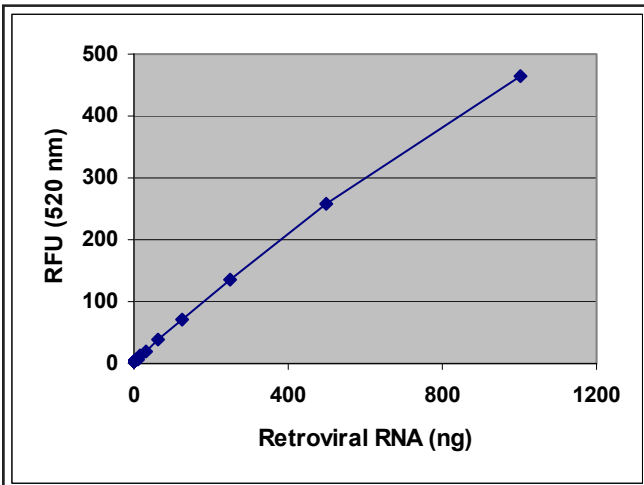
	ViraBind™ Retrovirus Concentration and Purification Kit	ViraBind™ PLUS Retrovirus Concentration and Purification Kit	ViraBind™ PLUS Retrovirus Concentration and Purification Mega Kit
Purification Method	Proprietary Reagent Cocktail + Purification Column	Proprietary Reagent Cocktail + Dialysis	Proprietary Reagent Cocktail + Dialysis
Total Time	6-8 hours	10-24 hours	10-24 hours
Capacity per Prep (Supernatant)	100 mL	50 mL	500 mL

Product Name	Size	Catalog Number
ViraBind™ Retrovirus Concentration and Purification Kit (100 ml/prep)	2 Preps	VPK-130
	5 Preps	VPK-131
	25 Preps	VPK-131-5
ViraBind™ PLUS Retrovirus Concentration and Purification Kit (50 ml/prep)	2 Preps	VPK-135
ViraBind™ PLUS Retrovirus Concentration and Purification Mega Kit (500 ml/prep)	2 Preps	VPK-136
	10 Preps	VPK-136-5

QuickTiter™ Retrovirus Rapid Quantitation Kit

This kit specifically measures the viral nucleic acid content of purified virus or unpurified viral supernatant. This method is ideal for a quick measurement of viral titer, either before or after purification of your retrovirus.

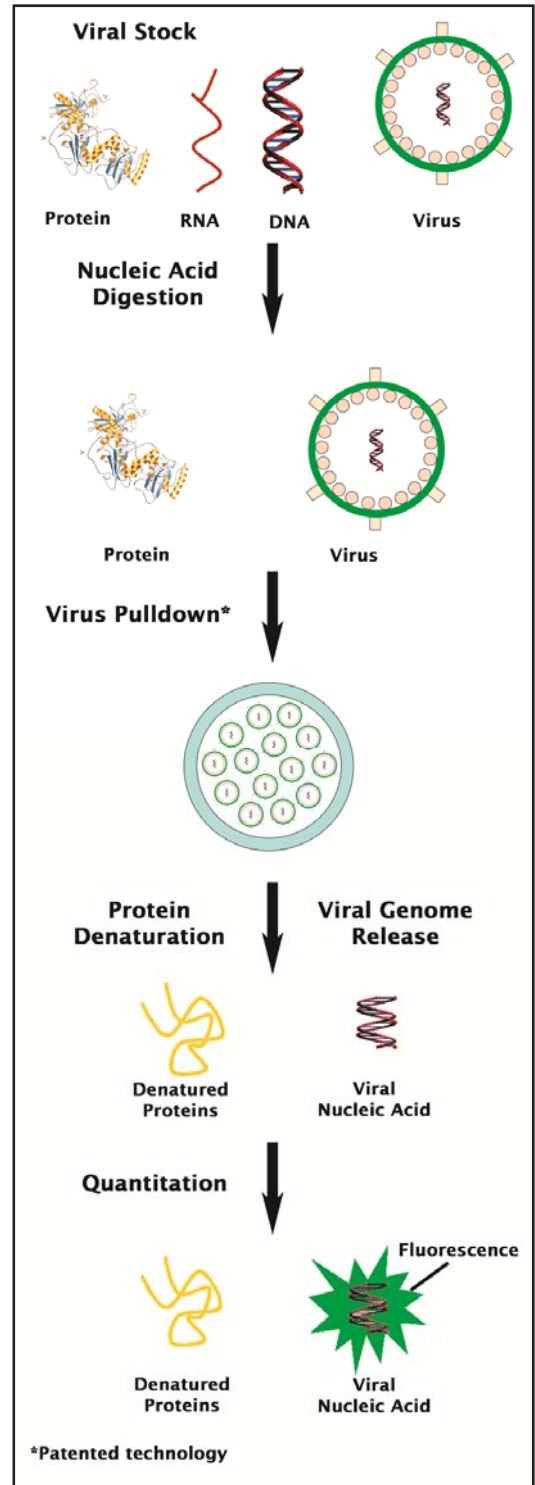
- **Ultra-fast Results:** 45-60 minute procedure
- **Convenient:** Titer may be measured before purification step
- **Sensitive:** Limit of detection = 1.5×10^9 VP/mL from 2 mL of retroviral supernatant



Retrovirus RNA Standard Curve. The QuickTiter™ Retrovirus RNA Standard was diluted according to the assay protocol. Fluorescence was measured on a SpectraMax Gemini XS Fluorometer (Molecular Devices) with a 485 / 538 nm filter set and a 530 nm cutoff.

Recent Product Citation

Ito, T. et al. (2012). Stem cell factor programs the mast cell activation phenotype. *J. Immunol.* **188**:5428-5437.



Assay Procedure for the QuickTiter™ Retrovirus Quantitation Kit.

Product Name	Detection	Size	Catalog Number
QuickTiter™ Retrovirus Quantitation Kit	Fluorometric	20 Assays	VPK-120

ViraDuctin™ Retrovirus Transduction Kit

The efficiency of retrovirus transduction can be low compared to other viruses. The rate at which retroviral vectors bind to cells is controlled mostly by diffusion. Additionally, the presence of transduction inhibitors such as proteoglycans and glycosaminoglycans in retroviral supernatants can lead to poor gene transfer. Additives such as Polybrene® can boost transduction efficiencies, but they do not eliminate these transduction inhibitors.

Our ViraDuctin™ Retrovirus Transduction Kit provides superior transduction efficiencies even when compared to transductions in the presence of Polybrene®. A proprietary reagent cocktail forms a super-complex with the retrovirus which is pelleted away from the supernatant, removing detrimental transduction inhibitors that decrease infection efficiency.

- **More Robust:** Removes harmful transduction inhibitors from retroviral supernatant
- **Higher Transduction Efficiencies:** Compared to infections in the presence of Polybrene or no additives
- **Versatile:** Particularly useful for nonpermissive cells including primary cells and stem cells, but may boost transduction rates in a wide variety of cells

Recent Product Citations

1. Gandhi, M. et al. (2012). Homologous chromosomes make contact at the sites of double-strand breaks in genes in somatic G0/G1-phase human cells. *PNAS* **109**:9454-9459.
2. Miyoshi, N. et al. (2010). Defined factors induce reprogramming of gastrointestinal cancer cells. *PNAS* **107**:40-45.

Product Name	Size*	Catalog Number
ViraDuctin™ Retrovirus Transduction Kit	40 Transductions	RV-200
	200 Transductions	RV-201

*Number of transductions shown is based on use in a 24-well plate. This product may also be used with 96-well, 12-well or 6-well plates, as well as 60mm or 100mm dishes. See product insert for specific details.

Polybrene is a registered trademark of Abbott Laboratories.



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