

## GFP Recombinant Adenovirus

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**CATALOG NUMBER:** ADV-004

**STORAGE:** -80°C

**QUANTITY AND CONCENTRATION:** 50 µl,  $1 \times 10^{11}$  VP/mL in TBS containing 10% Glycerol

### **Background**

Recombinant adenoviruses have tremendous potential in both research and therapeutic applications. There are numerous advantages in using an adenovirus to introduce genetic material into host cells. The permissive host cell range is very wide. The virus has been used to infect many mammalian cell types (both replicative and non-replicative) for high expression of the recombinant protein. Recombinant adenoviruses are especially useful for gene transfer and protein expression in cell lines that have low transfection efficiency with liposome. After entering cells, the virus remains epichromosomal (i.e. does not integrate into the host chromosome so does not activate or inactivate host genes). Recently, recombinant adenoviruses have been used to deliver RNAi into cells.

### **Safety Consideration**

Remember that you will be working with samples containing infectious virus. Follow the recommended NIH guidelines for all materials containing BSL-2 organisms. Always wear gloves, use filtered tips and work under a biosafety hood.

### **Methods**

The appropriate amount of viruses used for infecting cells is critical for the outcome of your experiments. If not enough virus is used, it will not give 100% of infection. If too much virus is used, it will cause cytotoxicity or other undesired effects. The amount of adenovirus cell surface receptors vary greatly among different cell types therefore the optimal concentration differs dramatically between cell types. A range of 10-200 MOI (multiplicity of infection) is used for most cell lines, but up to 1000 MOI may be used for lymphoid cell lines.

Traditionally, Infectivity particles are measured in culture by a plaque-forming unit assay (PFU) that scores the number of viral plaques as a function of dilution. In contrast to the 10-day infection of a classical plaque assay, Cell Biolabs' QuickTiter™ Adenovirus Titer Immunoassay Kit (Cat. #VPK-109) only requires 2-day infection, and there is no agar overlay step. The kit antibody against hexon protein recognizes all serotypes of adenovirus by immunocytochemistry (see Flow Chart).

Seed 293 cells in 24 or 12-well plate for 1 hr



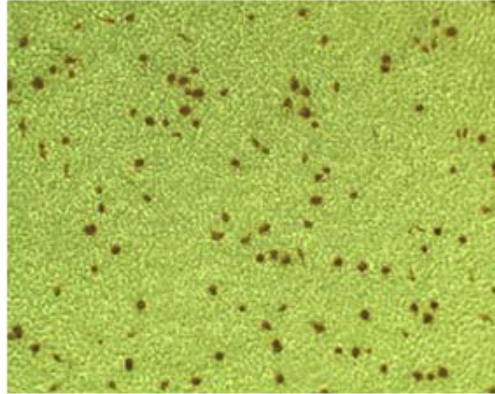
Prepare Adenovirus Serial Dilutions  
and Infect 293 cells for 48 hrs



Anti-Hexon Immunocytochemistry Staining



Count Positive Cells and Calculate Viral Titer



### References

1. Bett AJ, Haddara W, Prevec L and Graham FL. (1994) *Proc Natl Acad Sci U S A.* 91:8802-6.
2. Robbins, P. D., Tahara, H., and Ghivizzani, S. C. (1998) *Trends Biotechnol.* 16, 35-40.
3. Huang, S., Stupack, D., Mathias, P., Wang, Y., and Nemerow, G. (1997) *Proc. Natl. Acad. Sci. U S A.* 94, 8156-8161.
4. Bergelson, J. M., J. A. Cunningham, G. Droguett, E. A. Kurt-Jones, A. Krithivas, J. S. Hong, M. S. Horwitz, R. L. Crowell, and R. W. Finberg. (1997) *Science* 275:1320-1323.

### Recent Product Citations

1. Xu, S. et al. (2016). Activation of mTORC1 in B lymphocytes promotes osteoclast formation via regulation of  $\beta$ -Catenin and RANKL/OPG. *J Bone and Miner Res.* doi:10.1002/jbmr.2800.
2. Kütscher, C. et al. (2016). Overexpression of hypoxia-inducible factor-1 alpha improves vasculogenesis-related functions of endothelial progenitor cells. *Microvasc Res.* **105**:85-92.
3. Lampert, F. M. et al. (2015). Overexpression of Hif-1 $\alpha$  in mesenchymal stem cells affects cell-autonomous angiogenic and osteogenic parameters. *J Cell Biochem.* doi:10.1002/jcb.25361.
4. Jiang, Y. Z. et al. (2014). Distinct roles of HIF1A in endothelial adaptations to physiological and ambient oxygen. *Mol Cell Endocrinol.* **391**:60-67.
5. Haidari, M. et al. (2014). Disruption of endothelial adherens junctions by high glucose is mediated by protein kinase C- $\beta$ -dependent vascular endothelial cadherin tyrosine phosphorylation. *Cardiovasc Diabetol.* **13**:112.
6. 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.
7. 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.
8. Lu, D. et al. (2012). Peroxisome proliferator-activated receptor- coactivator-1 $\alpha$  (PGC-1 $\alpha$ ) enhances engraftment and angiogenesis of mesenchymal stem cells in diabetic hindlimb ischemia. *Diabetes.* **61**: 1153-1159.

9. 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.
10. Perez-Moreno, M. et al. (2008). Loss of p120 catenin and links to mitotic alterations, inflammation, and skin cancer. *Proc. Natl. Acad. Sci. U S A* **105**:15399-15404.
11. Black, S.A. et al. (2008). TGF $\beta$ 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 $\beta$ 1-induced CCN2/CTGF expression. *J. Biol. Chem.* **283**:10835-10847.

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