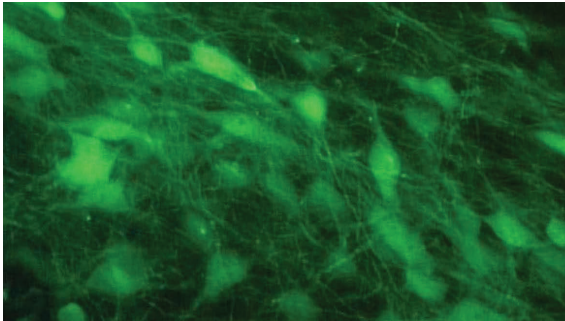
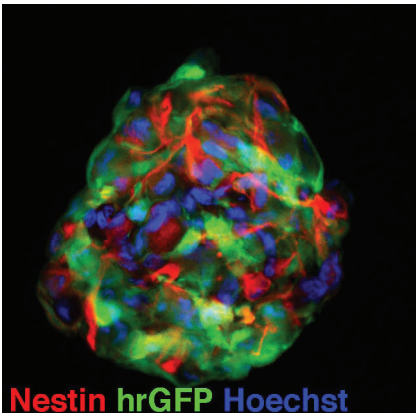


The VVF is located in the new wing of the Children's Memorial Research Center in a suite of rooms designed for BL2+ work. The suite consists of a gowning/distribution room, 3 culture rooms and a central equipment area totaling approximately 800ft². Specific areas within this suite are dedicated to the production of recombinant adeno-associated virus (rAAV) and HIV-based lentivirus and have access limited to the vector technicians.



Dopamine neurons in rat brain transduced with rAAV2 harboring green fluorescent protein. (Courtesy of Tamas Virag, Martha C. Bohn lab)



Rat bone marrow-derived neuroprogenitors transduced with a lentivirus harboring gene fluorescent protein. (Courtesy of Aleksandra Glavaski, Martha C. Bohn Lab)

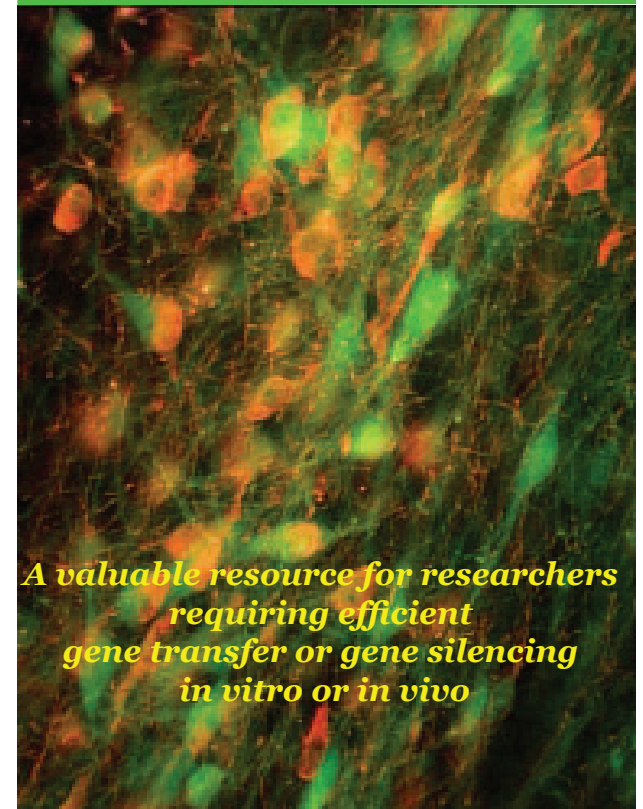


Children's Memorial Research Center
Children's Memorial Hospital
Feinberg School of Medicine
Northwestern University

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Children's Viral Vector Facility (VVF)



*A valuable resource for researchers
requiring efficient
gene transfer or gene silencing
in vitro or in vivo*



rAAV Vectors:

Adeno-associated viruses (AAV) are non-pathogenic human single strand DNA parvoviruses. Recombinant rAAV vectors are devoid of viral genes and can hold up to a 4.5kb expression cassette between the inverted terminal repeats (ITRs). Recent advances in technology for packaging rAAV in the absence of helper virus permits these vectors to be produced at high titers of 10^{12-14} vector genomes (vg)/ml. rAAV vectors are able to transduce dividing and non-dividing cells and are excellent for *in vivo* studies. The VVF offers serotype 2 rAAV.

Lentiviral Vectors (LV):

LV vectors belong to the family retrovirae. The most commonly used lentivirus for gene delivery is based on HIV-1. These LV vectors have a cloning capacity of 6-7kb. LV vectors produced in the VVF are pseudotyped with the vesicular stomatitis virus glycoprotein (VSV-G) to broaden cellular tropism. VSV-G LV vectors are particularly useful for transducing non-dividing neurons and have a high tropism for stem cells and other cultured cells. LV vectors are also useful for generating transgenic rats and for gene silencing studies. LV vectors produced by the VVF are packaged with a four plasmid system and have a titer of 10^{7-9} transducing units (tu)/ml. Gene silencing LV vectors are directly produced from the Northwestern gene silencing library of shuttle plasmids: <http://www.biochem.northwestern.edu/hta/RNAi.htm>

Services of the VVF

1. High titer, high quality, helper-free viral vectors for laboratory research on a subsidized fee-for-service, no-strings attached basis.
 - rAAV2 (recombinant adeno-associated viral vector serotype 2)
 - rLV (recombinant HIV-based lentivirus pseudotyped with the VSV-G envelope)
2. Consultation on viral vector design, including:
 - Virus type
 - Promoter
 - Expression cassette design
 - Virus use in vitro and in vivo
 - Biosafety requirements
 - Letters of support for grant applications
3. AAV or LV DNA backbones for cloning shuttle plasmids. (The VVF does not clone shuttle plasmids)
4. Small aliquots of rAAV and rLV viruses with a cell marker gene such as green fluorescent protein from the VVF's shuttle plasmid library.
5. Other shuttle plasmids from the VVF's library are available. Users are requested to consider depositing additional shuttle plasmids into the library.

Note that an active IBC approval number is required for requesting a vector.

Costs

Our costs to prepare shuttle plasmid DNA and generate viruses represent a pass through cost to cover supplies and does not include technicians' salaries.

2009 Prices for Northwestern University Researchers

rAAV (~700-750 μ l of $\sim 10^{12-13}$ vg/ml): \$1250
rAAV (20 μ l aliquot from VVF repository): \$150

rLV (~700-750 μ l of $\sim 10^{8-9}$ tu/ml): \$1250
rLV (~200 μ l of $\sim 10^{7-8}$ tu/ml): \$500
rLV (~80 μ l of $\sim 10^{7-8}$ tu/ml): \$250*
rLV (20 μ l aliquot from repository): \$150

*recommended for in vitro screening of silencing vectors (vg=vector genomes; tu=transducing units)

Maxi prep of shuttle plasmid for small viral preps: \$200
Double CsCl purified shuttle plasmid for large viral preps: \$500

All other Affiliations: full cost (call for quote)

Contact Information

Additional information and vector request forms can be found at:
<http://www.childrensmrc.org/vector/>

Addition Resources
<http://www.biochem.northwestern.edu/hta/RNAi.htm>
<http://www.addgene.org>

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