

Special MATra Protocols

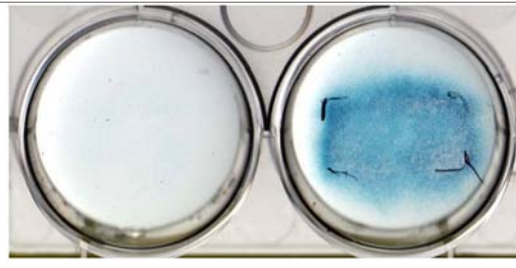
Viral Infection – MA Lipofection Enhancer Protocol

Viral infection is highly cell surface receptor-dependent. For instance, adenoviruses are dependent on cells to express CAR (Coxsackie's and adenovirus receptor) and HIV on cells to express CD4.

Unfortunately, many important and interesting target tissues for fundamental research and gene therapy are non-permissive to viral gene delivery (tumor tissues and apical surface of lung epithelium may express variable, little or none of the required receptors).

- The association of viral vectors with **MA Lipofection Enhancer** is sufficient to force infection of non-permissive cells as shown with adenovirus.
- Magnet Assisted Transfection also increases retroviral infectious capacity.

NIH 3T3 cells (lacking the coxsackie and adenovirus receptor, CAR) were transfected with a recombinant adenovirus (coding for LacZ) mixed with MA Lipofection Enhancer in the presence and in the absence of permanent magnets positioned under the culture plates.



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1. Cells should be plated in the same manner as required for standard viral gene delivery. For example, the confluency can be high for adenoviral vectors but must be low for retroviral vectors, which require cell division for infection. Cells must be plated the day prior transfection.
2. Provide a suitable amount (see examples below) of **MA Lipofection Enhancer** in a tube large enough to contain the volume of virus preparation added in step 3.
3. Add your virus preparation (e.g. retroviral supernatant or purified adenovirus diluted in HBS, PBS or cell culture medium) to the tube(s) containing **MA Lipofection Enhancer** and mix immediately by pipetting or gentle vortexing. Thereafter, incubate 15 minutes at room temperature.
4. The ratios virus / **MA Lipofection Enhancer** should be adjusted according to the viral titers and cell types used. For optimization, we suggest as a starting point to use 1.5 μL , 3 μL , 6 μL and 12 μL of **MA Lipofection Enhancer** with a fixed quantity of virus preparation / supernatant.
5. Add the mixture prepared in step 3 to the cells in duplicate or triplicate.
6. Place the cell culture plate upon the **Universal Magnet Plate** and incubate under standard cell culture conditions for 10-20 minutes.
7. Remove the **Universal Magnet Plate** plate. Optionally perform a medium change.
8. Cultivate the cells under standard conditions until evaluation of transgene expression.
9. Depending on the viral vector type, the quantity of virus and the cell types used, this protocol would have to be adjusted.

Cell Type	Virus	MA Lipofection Enhancer
K562	Adenovirus (200 MOI)	6 μL
Human PBL	Adenovirus (500 MOI)	3 – 6 μL
NIH-3T3	Adenovirus (200 MOI)	3 – 6 μL
NIH-3T3	Retrovirus ($1-5 \times 10^3$ X gal CFU/ml)	3 – 6 μL