

TECHNICAL DATA SHEET

MVL5/GMO Transfection Reagent Kit

Storage Conditions:

Store lipid blend at -20°C prior to reconstitution. Following hydration, store lipid blend at 4°C.

Product Description:

The MVL5/GMO transfection reagent is a formulation for transfecting plasmid DNA and siRNA into a wide range of eukaryotic cells. MVL5 and GMO form cationic liposomes, which when complexed with DNA forms a promising nonviral vector for gene delivery and silencing applications^{1,2}. Non-viral vectors are desirable as transfection systems because of their low potential immunogenicity and the ability to transfer very large pieces of nucleic acids². This multivalent cationic lipid vector system exhibits high transfection efficiency, even in the presence of serum². This is advantageous in cell culture applications for its ease of use and its elimination of the potentially adverse effects of serum starvation on the cell cycle². Most importantly, high transfection efficiency in the presence of serum may predict high transfection efficiency *in vivo*². The MVL5/GMO multivalent cationic lipid vector system has been found to outperform other commercially available formulations *in vitro*, particularly in the presence of serum².

Supplied Reagents:

A lipid blend containing MVL5 and glycerol monooleate (GMO) (1:1, mol:mol).

The blend contains the following: 0.6 mg of MVL5 and 0.2 mg of GMO

These quantities are sufficient to produce 1 mL of a 1 mM stock solution of vesicles.

Additional Reagents Required but not Supplied:

Plasmid DNA

Opti-MEM Reduced Serum Medium or DMEM

Eppendorf Tubes

Product

Avanti No.	Description
640009	MVL5/GMO Transfection Reagent Kit

General Protocol:

Preparation of Liposomes:

- To prepare liposomes, hydrate the lipid blend with 1 mL of sterile, high-resistivity water to produce a 1 mM stock solution.
- Close the vial tightly, and incubate the mixture at 37°C for at least 12 hrs.
- Place the vial in a water bath, and sonicate for 10 minutes or until the solution is clear.
- Transfer the solution to a new container (glass vial or 1.5 mL centrifuge tube) by passing the solution through a filter (0.2 μ m pores).
- Store the liposomal solution at 4°C.

Note.

The liposome stock may be re-used for up to four months with sonication prior to each use.

Transfection:

Note. This Protocol is optimized for a 24-well plate format.

- Seed cells to be ~70% confluent at the time of transfection.
 - Dilute the required amount of plasmid DNA to 4 μ g/mL in Opti-MEM or DMEM.
 - Add an appropriate volume of the prepared Liposomal Solution (12 μ L/1 μ g of DNA) to the diluted plasmid DNA.
 - Incubate at room temperature for 20 min.
 - Add 200 μ L of the solution (0.4 μ g DNA) to each well.
 - Following 6 hrs of incubation at 37°C, remove the transfection medium.
 - Wash each well once with phosphate buffered saline.
 - Add fresh culture medium to each well.
 - Harvest cells following an additional 18-48 hr of incubation.

Additional Guidelines:

- Optimum transfection conditions and the amount of DNA per well must be determined empirically for each cell type by the user.
- Prepare sufficient reaction mixture in order to perform duplicate or triplicate analyses.

Component	96-Well	24-Well	12-Well	6-Well
Volume of DNA-Lipid Complex added to each well	100 μ L	200 μ L	400 μ L	1000 μ L
Final DNA per well	0.2 μ g	0.4 μ g	0.8 μ g	2 μ g

General guidelines are applicable to the transfection of plasmid DNA and siRNA.

References:

¹Chan, C-L., Ewert, K.K., Majzoub, R.N., Hwu, Y-K, Liang, K.S., Leal, C., Safinya, C. Optimizing cationic and neutral lipids for efficient gene delivery at high serum content.

The Journal of Gene Medicine, 2014, 16:84-96.

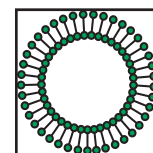
²Leal, C., Ewert, K.K., Shirazi, R.S., Bouxsein, N.F., Safinya, C. Nanogyroids Incorporating Multivalent Lipids: Enhanced Membrane Charge and Pore Forming Ability for Gene Silencing.

Langmuir, 2011, 27(12):7691--7697.

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