Cat # SL100468 Store at 4 °C

LipoJet™ In Vitro DNA and siRNA Transfection Kit (Ver. II)

----- A General Protocol for Transfecting Mammalian Cell

- ☐ 100 µl
- ☐ 500 µl
- ☐ 1000 µl

SignaGen® Laboratories
9601 Medical Center Drive
Rockville MD 20850
FAX. 301-560-4919
TEL. 301-330-5966
Toll Free. 1-(866)-918-6812
Email: info@signagen.com
Web: www.signagen.com

This product is for laboratory research ONLY and not for diagnostic use

Introduction:

Based on our innovative and proprietary lipid-conjugation technology, LipoJet™ Transfection Kit, formulated from novel fluorinated cationic lipids, exhibits significant difference from other lipids transfection reagents in the market. LipoJet™ Transfection Kit is the most powerful yet very gentle gene delivery tool for a variety of applications including plasmid DNA and/or siRNA for most of mammalian cell types. Compared with leading products in the market, LipoJet™ is more cost-effective and always provides higher transfection efficiency with less cytotoxicity.

Contents Per Kit:

1. 1 x 1.0 ml of LipoJet™ DNA In Vitro Transfection Reagent
2. 1 x 8.0 ml of LipoJet™ Transfection Buffer (5x)

Important Guidelines for Transfection:

- LipoJet™ reagent was formulated for DNA and siRNA transfection. The following standard protocol is given for DNA/siRNA co-transfection to mammalian cells. For a protocol of DNA and siRNA transfections, please email us at info@signagen.com
- For better efficiency, choosing LipoJet™ Transfection Buffer (1x) is a must.
- To lower cytotoxicity, transfect cells in presence of serum (10%) and antibiotics.

A General Protocol for DNA/siRNA Co-transfection.

Step I. Preparation of Working Solution of LipoJet™ Transfection Buffer (1x)

LipoJet™ Transfection Buffer (5x) is provided as 5x concentrated stock solution. To make working solution, dilute one part of the stock solution with 4 parts of ddH_2O. The LipoJet™ Transfection Buffer (1x) working solution is stable at RT for 24 months.

Note: Always keep LipoJet™ Transfection Buffer (5x) at RT. If refrigerated, white precipitates may appear. It won't affect the transfection efficiency. After dilution with 4 parts of ddH_2O to make LipoJet™ Transfection Buffer (1x) working solution, the white precipitates will disappear.

Step II. Cell Seeding:

Cells should be plated 18 to 24 hours prior to transfection so that the monolayer cell density reaches to the optimal 60~70% confluency at the time of transfection. Complete culture medium with serum and antibiotics is freshly added to each well 30~60 minutes before transfection.

Note: High serum levels (>5%) with antibiotics do NOT have inhibitory effect on transfection efficiency. We recommend using complete serum/antibiotics-containing medium to grow the cells during transfection.

Step III. Preparation of LipoJet™ -DNA/siRNA Complex and Transfection Procedures:

For DNA/siRNA co-transfection, we recommend using 1.0 µg DNA and 10 to 50 nM siRNA per well in a 6-well plate. The following protocol is given per well of 6-well plate. For other culture formats, please refer to **Table 1**.

- For each well of 6-well plate, dilute 1 µg of DNA and 20 to 100 pmoles siRNA into 200 µl of LipoJet™ Transfection Buffer (1x) prepared from **Step I**. Mix by vortexing.
- Add 4 µl of LipoJet™ reagent, vortex briefly to mix.
- Incubate for ~10 min at RT to allow LipoJet™/DNA/siRNA complex to form.

  Note: Never keep the LipoJet™/DNA/isRNA complex longer than 20 min.
- Add the LipoJet™/DNA/siRNA transfection mix to the cells in serum containing medium drop wisely.
- Swirl plate gently to homogenize.
- Check transfection efficiency 24 to 72 hours post transfection. 48~72 hours usually give better efficiency.

Table 1. Recommended Amounts for Different Culture Vessel Formats

Culture Dish	Culture Medium (ml)	Plasmid DNA (µg)	siRNA Final 10~50 nM (pmoles)	LipoJet™ Transfection Buffer (1x) (µL)	LipoJet™ Reagent (µL)
24-well	0.25	0.25	2.5~12.5	25	1~1.5
12-well	0.5	0.5	5~25	50	2~3
6-well	2	1.0	20~100	200	4 ~6
35 mm dish	2	1.0	20~100	200	4 ~6
60 mm dish	4	2.0	40~200	400	8 ~ 12
10 cm/T75	10	5	100~500	800	20 ~ 30
15 cm/T175	20	10	200~1000	1600	40 ~ 60

Storage: LipoJet™ Reagent is stable for up to 12 months at +4 °C after receipt