

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

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

- ☐ 100 µl
- ☐ 500 µl
- ☐ 1000 µl



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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 x 1.0 ml of LipoJet™ DNA In Vitro Transfection Reagent
- 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](mailto: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<sub>2</sub>O. 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<sub>2</sub>O 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/siRNA 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