

## User Bulletin

# Viofectin™ Transfection Reagent

Viofectin™ Transfection Reagent utilizes Viogene's proprietary formulation for the transfection of DNA and RNA into eukaryotic cells. Viofectin™ Transfection Reagent is suitable for many cell types and providing the highly efficiency and low cellular-toxicity.

### Transient transfection of adherent cells

For optimal transfection conditions with Viofectin™ Transfection Reagent, the cells should be 90-95% confluent. Typically, for transfection in 24-well plates,  $5 \times 10^4$  to  $1 \times 10^5$  cells are seeded per well, 24 hours before transfection. For other culture formats, see Table 1.

### Shipping & Storage

Viofectin™ Transfection Reagent is stable for 12 months at 4°C.

Culture Vessel	Number of cells to seed	Vol. of Medium per vessel(ml)	Amount of Plasmid DNA (µg)	Volume of Transfection Reagent (µl)	Total vol. of DNA/reagent complex (µl)
1well/96well	$1 \times 10^4$ - $1.7 \times 10^4$	0.1	0.05~0.2	0.15~0.4	10
1well/24well	$5 \times 10^4$ - $1.0 \times 10^5$	0.5	0.2~1.0	0.6~2.0	30
1well/6well	$2 \times 10^5$ - $4 \times 10^5$	2.0	1.0~3.0	3.0~9.0	120
35mm	$2 \times 10^5$ - $4 \times 10^5$	2.0	1.0~3.0	3.0~9.0	120
60mm	$4 \times 10^5$ - $8 \times 10^5$	5.0	3.0~5.0	6.0~15.0	300
10cm	$1 \times 10^6$ - $6 \times 10^6$	10.0	5.0~10.0	15.0~30.0	600

Table 1: Number of cells to seed before the day of transfection and transfection mix preparation for different cell culture formats.

Culture Vessel	Number of cells to seed	Vol. of Medium per vessel(ml)	Amount of Plasmid DNA (µg)	Volume of Transfection Reagent (µl)	Total vol. of DNA/reagent complex (µl)
1well/96well	$2 \times 10^4$ - $5 \times 10^4$	0.05~0.2	0.15~0.4	0.05~0.2	10
1well/24well	$1 \times 10^5$ - $2 \times 10^5$	0.2~1.0	0.6~2.0	0.2~1.0	30
1well/6well	$2 \times 10^5$ - $5 \times 10^5$	1.0~3.0	3.0~9.0	1.0~3.0	120
35mm	$5 \times 10^5$ - $2 \times 10^6$	1.0~3.0	3.0~9.0	1.0~3.0	120
60mm	$2 \times 10^6$ - $5 \times 10^6$	3.0~5.0	6.0~15.0	3.0~5.0	300
10cm	$5 \times 10^6$ - $1 \times 10^7$	5.0~10.0	15.0~30.0	5.0~10.0	600

Table 2: A Guideline for seeding suspension cells prior to transfection in different culture formats recommended.

## **Transfection procedure**

The following protocol is given for transfection in 24-well plates. Use 1  $\mu$ l of Viofectin™ Transfection Reagent and 1  $\mu$ g of DNA per well as follows. See table 1 or 2 for other culture vessel formats.

1. Preparation of Viofectin™ Working Transfection Reagent: Dilute 1  $\mu$ l Viofectin™ Transfection Reagent with 30  $\mu$ l serum-free medium (without antibiotics), and then mix by gentle pipetting or vortexing for one second. Incubate the working transfection reagent for 5 minutes at room temperature.

Note: avoiding the Viofectin™ Transfection Reagent to contact the plastic tube surface

2. Add the 1  $\mu$ g plasmid DNA into the Viofectin™ Working Transfection Reagent and mix by gentle pipetting

3. Incubate the mixture of DNA and Viofectin™ Transfection Reagent solution at room temperature for 15 minutes. For some cell line the incubation may be up to 30 minutes.

4. Add DNA/Viofectin™ Transfection Reagent complex to cells in each well and mix by gentle shaking plate, Incubate cells at 37°C in CO<sub>2</sub> incubator for 18-48 hrs.

## **Transfection with siRNA**

The following protocol is given for transfection in 24-well plates.

1. Preparation of Viofectin™ Working Transfection Reagent: Dilute 1  $\mu$ l Viofectin™ Transfection Reagent with 30  $\mu$ l serum-free medium (without antibiotics), and then mix by gentle pipetting or vortexing for one second. Incubate the working transfection reagent for 5 minutes at room temperature.

2. Add 100 pmol siRNA into the Viofectin™ Working Transfection Reagent and mix by gentle pipetting

3. Incubate the mixture of siRNA and Viofectin™ Transfection Reagent solution at room temperature for 15 minutes.

4. Add siRNA/ Viofectin™ Transfection Reagent complex to cells in each well and mix by gentle shaking plate, Incubate cells at 37°C in CO<sub>2</sub> incubator for 24-72 hrs.