MaxFection™ 8600 *in vitro* Transfection Reagent

MF8600-015 for ～200 in vitro transfections
Store at -20 °C

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Description

MaxFection™ 8600 belongs to an efficient new class of non-viral gene delivery based on biodegradable cationic polymer.

Principle

The remarkable transfection efficiency of MaxFection™ 8600 is due to its intracellular degradation property, which may maximize gene unloading into nucleus as well as minimize cytotoxicity of cationic residues.

Reagent Supplied

1) 10×MaxFection™ 8600 concentrated solution (0.45mL, =4.5mL 1X reagent)
2) Buffer solution (>2mL)

Formulation

MaxFection™ 8600 is sterile apyrogenic solution of cationic polymer in an acidic pH 5.0 buffer.

Considerations for Transfection with MaxFection™ 8600

1. DNA quality

High quality DNA is very important for successful transfection.
DNA should be sterile and free of any contaminant such as endotoxins.
The OD_{260}/OD_{280} ratio should be at 1.6 or greater.

2. Cell density at transfection

The recommended cell density (confluency) for most cell types is 50-70% for transfection. The cells should not be confluent or at stationary phase prior to transfection.

3. Transfection incubation time

Detection of transgene expression should be performed within 24-72 hours. An optimal post-transfection incubation time can be determined using a reporter gene (such as Luciferase, galactosidase or Green Fluorescent Protein).

4. Choice of promoter

High transfection efficiency depends not only on cell line type but promoter under which the gene of interest is expressed. MaxFection™ 8600 may be applied for cytomegalovirus (CMV) promoter, one of the best known for high transgene expression in a wide variety of cell lines, and others such as ORF promoter.

5. MaxFection™ 8600/DNA equivalent

• The required amount of MaxFection™ 8600 solution depends on the amount of DNA and the number of equivalents needed.
• **Initially, we recommend** to use 1 μg of DNA and 15 μl of 1× MaxFection™ reagent per well of 24-well plates (*see* Table 1).
• **Subsequent optimization** may further increase the transfection efficiency in your transfection test depending on cell line type and the gene plasmid.

• The DNA quantity can range from 0.5 to 10 μg for 100,000 cells; likewise MaxFection™ 8600/DNA ratio can range from 15 to 40uL 1X .

6. Transfection in the presence of serum

MaxFection ™ 8600-mediated high transfection efficiency is not seriously affected by 10% serum.

7. Protocol for transfection of adherent cells in a 24-well plate.

Quantities and volumes should be scaled up according to the number of cells or wells to be transfected. (*See* Table 1 for scale-up ratios).

1) Prepare DNA working solution: Dilute 1μg of DNA with *Buffer solution* in a 1.5 mL of EP tube to give 10 μL of DNA working solution;

2) Prepare 1×MaxFection™ 8600 solution: Dilute 3 μL of 10×MaxFection™ 8600 concentrated solution with 27 μL of *Buffer solution*; You may scale up as needed.

3) Add 30 μL of 1×MaxFection™ 8600 solution into 20 μL of DNA working solution and vortex-mix (or pipette) the solution **immediately** for 5 sec;

4) Incubate for 30 minutes at room temperature;

5) Add 25 μl of the MaxFection™ 8600/DNA mixture to each well;

6) Gently rock the plate back and forth and from side to side to achieve even distribution of the complexes;

7) Incubate at 37°C for 1-4 hour transfection.

8) For serum-free transfection, replace serum-free culture medium with complete culture medium

9) Monitor transient transfection after 24-72 h post-transfection

Notes. *1. Initially, we recommend the use of 1 μg DNA and 15* μL *1×MaxFection™ 8600 per well of 24-well plate (see Table 1) in* ***serum-free transfection for 1-4 h and 24-72 h post-transfection in complete medium****.*

2. Subsequent optimization may increase the equivalent number from 20-100 depending on the cell line and the gene expressed.

Table 1. Scale-up equivalent numbers were used according to the surface area of the tissue culture plate

Culture Plate	Growth Area (cm^2/well)	Cell seeding density	Recommended DNA amount (μg)	optimal amount of DNA (μg)*	μL of 1×MaxFection™ 8600 (μL)
96	0.3	$0.5\text{-}1\times10^4$	0.3-1	0.5	7.5
48	0.7	$1\text{-}3\times10^4$	0.5-1	0.5	7.5
24	2	$2\text{-}6\times10^4$	1-1.5	1	15
12	4	$0.4\text{-}1.2\times10^5$	1-3	1.5	22.5
6	9.5	$1\text{-}2\times10^5$	2-4	2	30

Actual values depend on the cell line type

Cell lines successfully transfected with MaxFection™ 8600 include:

Permanently growing cell lines		
HeLa-	Human	Cervix epitheloid carcinoma cells
MCF-7	Human	Breast adenocarcinoma cells
PC-3	Human	Prostate adenocarcinoma cells
SKOV-3	Human	Ovarian carcinomacells
HepG2	Human	Hepatoma cells
A549	Human	Type II pneumocytes
NIH 3T3	Mouse	Embrionic fibroblasts
COS-7	Monkey	African green monkey kidney cells