

Lipofectamine® 2000 Reagent

Cat. nos.	11668-030	Size:	0.3 mL	Store at 4°C (do not freeze)
	11668-027		0.75 mL	
	11668-019		1.5 mL	
	11668-500		15 mL	

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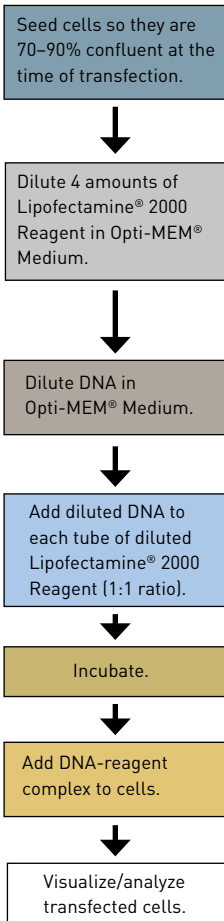
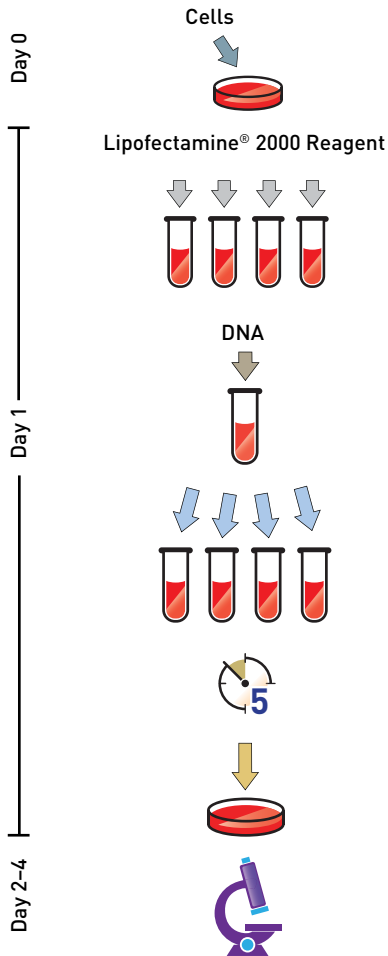
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Description

- Lipofectamine® 2000 Transfection Reagent is a proprietary formulation for transfecting nucleic acids (DNA and RNA) into a wide range of eukaryotic cells.
- Nucleic acid-Lipofectamine® 2000 complexes can be added directly to cells in culture medium, in the presence or absence of serum.
- It is not necessary to remove complexes or change/add medium after transfection.

Important Guidelines for Transfection

- Use Opti-MEM® I Reduced Serum Medium (Cat. no. 31985-062) to dilute Lipofectamine® 2000 Transfection Reagent and nucleic acids.
- The amount of Lipofectamine® 2000 Reagent required for successful transfection varies depending on the cell type and passage number. Start any new transfection by testing the recommended four concentrations of Lipofectamine® 2000 Reagent to determine an optimum amount.
- Prepare high-quality plasmid DNA at 0.5–5 µg/µL in deionized water or TE buffer. A GFP (green fluorescent protein) plasmid can be used to determine transfection efficiency.
- For additional information, and protocols on transfecting suspension cells refer to the manual at www.lifetechnologies.com/manuals.



Plasmid Transfection

Transfect cells according to the following table. Volumes are given on a per well basis. Vary the amount of components according to your tissue culture format. For additional information on scaling your transfection reaction, see page 4.

Component	96-well	24-well	6-well
Adherent cells	1–4 x 10 ⁴	0.5–2 x 10 ⁵	0.25–1 x 10 ⁶
Opti-MEM® Medium	50 µL x 4	50 µL x 4	150 µL x 4
Lipofectamine® 2000 Reagent	2, 3, 4, 5 µL	2, 3, 4, 5 µL	6, 9, 12, 15 µL
Opti-MEM® Medium	250 µL	250 µL	700 µL
DNA (0.5–5 µg/µL)	5 µg	5 µg	14 µg
Diluted DNA	50 µL	50 µL	50 µL
Diluted Lipofectamine® 2000 Reagent	50 µL	50 µL	50 µL
Incubate for 5 minutes at room temperature			
DNA-reagent complex/well	10 µL	50 µL	250 µL
Incubate cells for 1–3 days at 37°C			

The following table shows the amounts of DNA and Lipofectamine® 2000 Reagent used in each transfection reaction.

Amount	96-well	24-well	6-well
DNA/well	100 ng	500 ng	2500 ng
Lipofectamine® 2000 Reagent/well	0.2–0.5 µL	1–2.5 µL	5–12.5 µL

Scaling Up or Down Transfections

Use the multiplication factor in the following table to scale the volumes for your transfection experiment. The factor is based on the relative surface area of a single well from a 24-well plate.

Culture Vessel	Multiplication Factor	Culture Vessel	Multiplication Factor
96-well plate	0.2X	6-well plate	5X
48-well plate	0.5X	60 mm dish	11X
24-well plate	1X	100 mm dish	29X
12-well plate	2X	T75 flask	39X

Co-Transfection of Plasmid DNA and siRNA

Plasmid DNA and siRNA can be transfected at the same time using Lipofectamine[®] 2000 Reagent by adding 40 pmol (~0.6 µg) of siRNA per 1 µg of DNA.

mRNA Transfection

mRNA can be transfected in a 24-well plate by using Lipofectamine[®] 2000 Reagent by adding 0.5–1 µg of mRNA per well.

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