Mergene1000® NIH/3T3 Cell-Specific mRNA Transfection Reagent

Cat. No. : 164425

Size: 100μL/0.5mL,/1mL

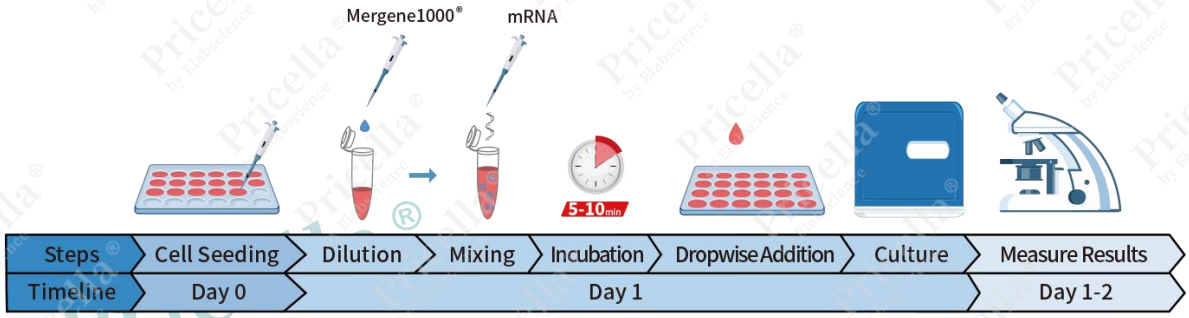
General Information

Product From	Liquid
Product Color	Colorless
Product Packaging	1 tube
Storage	2-8℃
Expiration Date	18 months
Shipping	Ice bag

Background

Mergene1000® NIH/3T3 Cell-Specific mRNA Transfection Reagent is a high-performance mRNA transfection reagent designed for the delivery of mRNA. It can directly deliver mRNA into the cytoplasm for expression, thereby avoiding the limitations of transcriptional regulation and entry into the nucleus. It is specifically formulated for use with NIH/3T3 cells, achieving high transfection efficiency. The reagent is distinguished by its low toxicity, excellent stability, ease of operation, and high reproducibility.

Product Operation Flowchart



Usage Steps

To transfect NIH/3T3 cells, follow the steps outlined below. Useing 24-well plates as an example, mix Mergene1000® NIH/3T3 Cell-Specific mRNA Transfection Reagent (μL) with mRNA (μg) at a ratio of 5:1. This ratio can be adjusted between 3:1 and 7:1 according to the situation. For other sizes of culture plates or dishes, refer to the recommended transfection amounts provided in Table 1.

1. Cell seeding

The day before transfection, add 500 μL DMEM (High glucose) (PM150210) + 10% NCS + 1% P/S (PB180120) medium to each well, inoculate 0.6×10^5 cells/well, and culture the cells for 12 hours. The incubation duration may be adjusted based on the actual conditions of the cells to ensure that the cell confluence reaches 70% to 90% at the time of transfection.

2. Preparation of the transfection complex

(1) Prepare a sterile centrifuge tube, add 50 µL DMEM (High glucose) (PM150210), and then add 1.0 µL of Mergene1000® NIH/3T3 Cell-Specific mRNA Transfection Reagent to the tube containing the medium, and gently blow and mix. Then add 0.2 µg mRNA to the above transfection reagent dilution solution and blow and mix.

Note: The above is the amount of preparation for each well of cells. Please calculate the required volumes based on your specific experimental conditions and requirements.

(2) Allow the above dilution to incubate at room temperature for 5 to 10 minutes.

3. Cell transfection

(1) Add the prepared transfection complex dropwise to the cells and mixed, incubated at 37℃ with 5% CO_2 for culture.

(2) After 12-24 hours of incubation, detect gene expression.

Table 1. Reference dosage of NIH/3T3 cells transfection in different culture vessel

Culture Vessel	Area	Cell Seeding Density	Inoculation Medium	Diluted Final Volume	mRNA Transfection	
					Reagent Amount	mRNA Amount
96-well	0.3 cm^2	1-4×10^4 cells/well	200 µL	10 µL	0.5 µL	0.1 µg
24-well	2.0 cm^2	0.5-1×10^5 cells/well	500 µL	50 µL	1.0 µL	0.2 µg
12-well	4.0 cm^2	1-2×10^5 cells/well	1 mL	100 µL	2.0 µL	0.4 µg
6-well	10.0 cm^2	2.5-5×10^5 cells/well	2 mL	200 µL	5.0 µL	1.0 µg
6 cm	20.0 cm^2	0.5-1×10^6 cells/well	5 mL	0.5 mL	10.0 µL	2.0 µg
10 cm	60.0 cm^2	1.5-3×10^6 cells/well	15 mL	1.0 mL	30.0 µL	6.0 µg

Note: The usage amounts provided in the table are for reference only. The exact amount of mRNA used with Mergene1000® NIH/3T3 Cell-Specific mRNA Transfection Reagent should be optimized according to the cell conditions and other experimental parameters.

Notes

1. The cell inoculation amount and transfection ratio provided above are based on experiments conducted with NIH/3T3 cells and are for reference only. The specific experimental dosage should be adjusted according to the actual conditions.
2. The product is transported with ice bag and can be aliquoted and stored upon use to avoid multiple prolonged openings of the lid.
3. DMEM (High glucose) medium should be prepared separately for the dilution of mRNA and transfection reagents.
4. During transfection, ensure that the degree of cell confluence is not less than 70%, and it is generally maintained at around 70% to 90%. The specific plating density can be adjusted according to the actual conditions of the cells.
5. After transfection, there is no need to remove the transfection complex or replace with fresh culture medium. The actual operation can be based on the cell status, after transfection culture 4-6 hours to choose to change the medium.
6. The use of high purity mRNA is helpful to obtain higher transfection efficiency.
7. The experimental process utilized RNA-free and pyrogen-free materials, such as centrifuge tubes, pipette tips, and buffers.

8. For research use only.
9. For your safety and health, please wear experimental clothes and wear disposable gloves aseptic operation.

Experimental Results Show (For reference only)

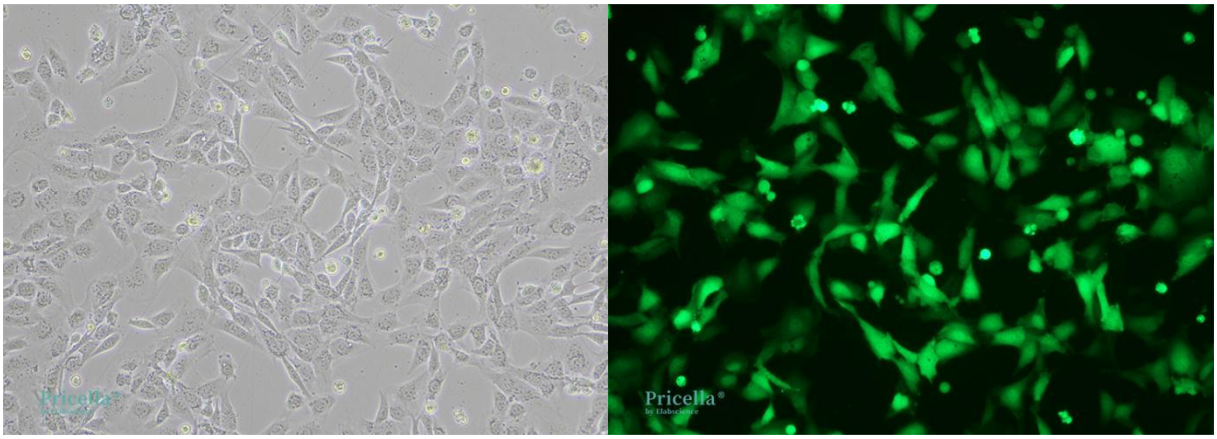


Figure 1. Bright-field and fluorescence images of NIH/3T3 cells transfected with EGFP-mRNA using Mergene1000® NIH/3T3 Cell-Specific mRNA Transfection Reagent.

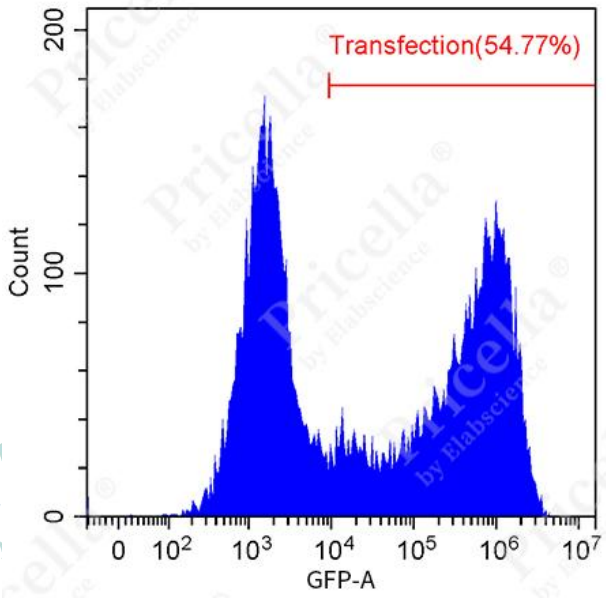


Figure 2. Transfection efficiency of NIH/3T3 cells transfected with EGFP-mRNA using Mergene1000® NIH/3T3 Cell-Specific mRNA Transfection Reagent.