

Mergene1000[®] NIH/3T3 Cell-Specific siRNA Transfection Reagent

Cat. No. : 164440

Size: 100 μ L / 0.5mL / 1mL

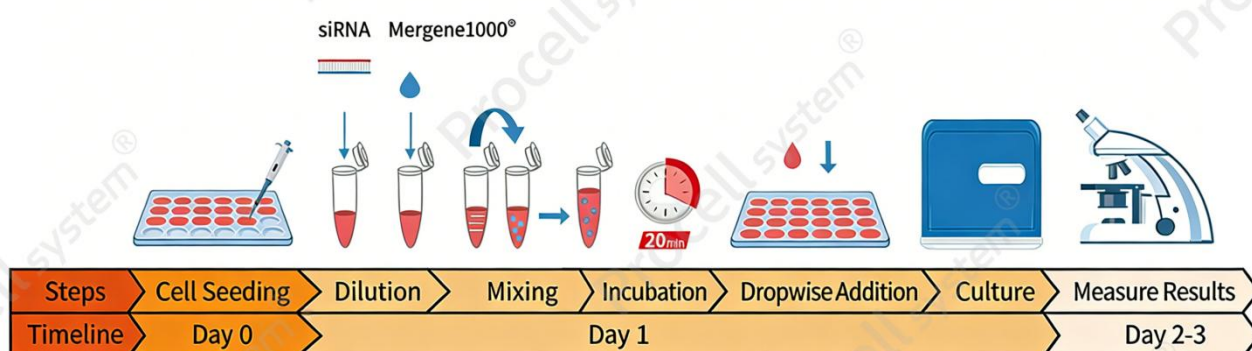
General Information

Product From	Liquid
Product Color	Colorless
Product Packaging	1 tube
Storage	2-8 $^{\circ}$ C [®]
Expiration Date	18 months
Shipping	Ice bag

Background

Mergene1000[®] NIH/3T3 Cell-Specific siRNA Transfection Reagent is a new and stable siRNA[®] transfection reagent with efficient RNA compression ability. It can quickly and efficiently transfect RNA into cells, while forming a stable complex to effectively protect RNA from nuclease degradation. 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. Using 24-well plates as an example, mix Mergene1000[®] NIH/3T3 Cell-Specific siRNA Transfection Reagent (μ L) with siRNA (pmol) at a ratio of 1:15. This ratio can be adjusted between 1:10 and 1:30 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 μ L DMEM (High glucose) (PM150210) + 10% FBS + 1% P/S (PB180120) medium to each well, inoculate 0.5×10^5 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 50% to 70% at the time of transfection.

2. Preparation of the transfection complex

- (1) Prepare two sterile centrifuge tubes. In one tube, add 15 pmol siRNA and DMEM (High glucose) medium (PM150210) to a final volume of 10 μ L, and gently mix by pipetting. In the other tube, add 1.0 μ L of Mergene1000® NIH/3T3 Cell-Specific siRNA Transfection Reagent and 9.0 μ L of DMEM (High glucose) medium (PM150210) to a final volume of 10 μ L, 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) Mix the above two equal volume diluents, gently mix by pipetting, and incubate for 20 minutes at room temperature.

3. Cell transfection

- (1) Add the prepared 20 μ L transfection complex dropwise to the cells and mixed, incubated at 37°C with 5% CO₂ for culture.
- (2) After 18-48 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	siRNA Transfection	
					Reagent Amount	siRNA Amount
96-well	0.3 cm ²	1-4 × 10 ⁴ cells/well	200 μ L	2 × 5 μ L	0.2 μ L	3 pmol
24-well	2.0 cm ²	0.5-1 × 10 ⁵ cells/well	500 μ L	2 × 10 μ L	1.0 μ L	15 pmol
12-well	4.0 cm ²	1-2 × 10 ⁵ cells/well	1 mL	2 × 20 μ L	2.0 μ L	30 pmol
6-well	10.0 cm ²	2.5-5 × 10 ⁵ cells/well	2 mL	2 × 50 μ L	4.0 μ L	60 pmol
6 cm	20.0 cm ²	0.5-1 × 10 ⁶ cells/well	5 mL	2 × 0.1 mL	8.0 μ L	120 pmol
10 cm	60.0 cm ²	1.5-3 × 10 ⁶ cells/well	15 mL	2 × 0.3 mL	24.0 μ L	360 pmol

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

Notes

1. This product is only used for scientific research or further research, not for diagnosis and treatment.
2. 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.
3. The product is transported with ice bag and stored at 2-8°C. It can be aliquoted and stored upon use to avoid multiple prolonged openings of the lid.
4. DMEM (High glucose) medium should be prepared separately for the dilution of siRNA and transfection reagents.
5. During transfection, ensure that the degree of cell confluence is not less than 50%, and it is generally maintained at around 50% to 70%. The specific plating density can be adjusted according to the actual conditions of the cells.
6. 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.

- The use of high purity siRNA is helpful to obtain higher transfection efficiency. Before transfection, ensure that siRNA gene silencing expression does not affect cell viability.
- The experimental process utilized RNA-free and pyrogen-free materials, such as centrifuge tubes, pipette tips, and buffers.
- For in vitro transfection only.
- For your safety and health, please wear experimental clothes and wear disposable gloves aseptic operation.

Experimental Results Show (For reference only)

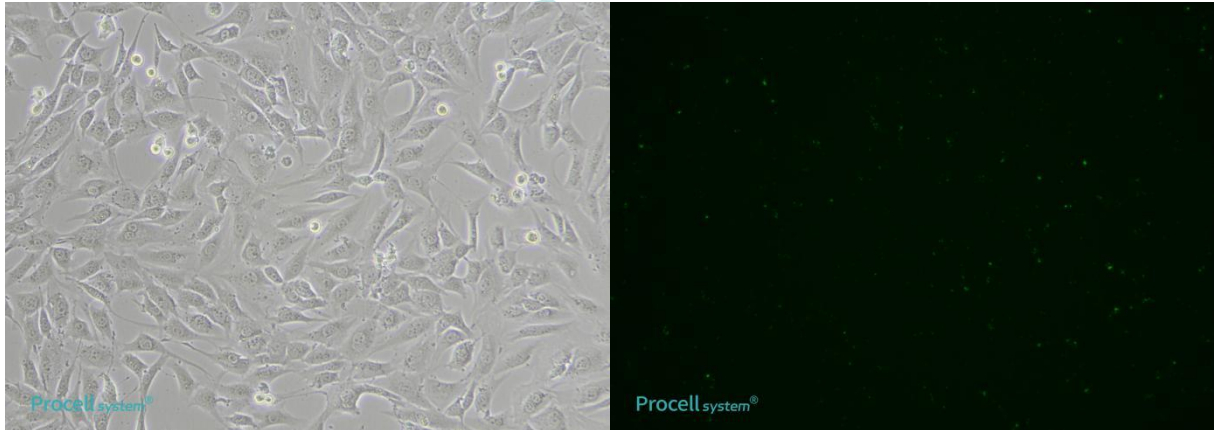


Figure 1. Bright-field and fluorescence images of NIH/3T3 cells transfected with FAM-siRNA using Mergene1000[®] NIH/3T3 Cell-Specific siRNA Transfection Reagent.

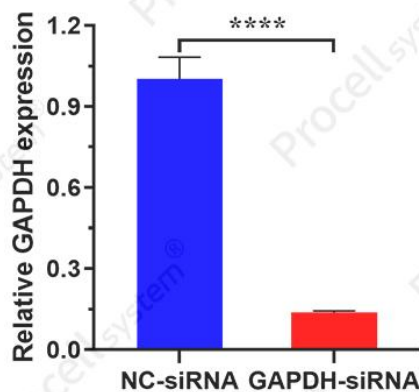


Figure 2. NIH/3T3 cells were transfected with NC-siRNA and GAPDH-siRNA using Mergene1000[®] NIH/3T3 cell-specific siRNA transfection reagent, and the relative GAPDH expression was detected by qPCR.