Omnimabs® V siRNA Primary Cell Transfection Reagent

Cat. No.: OM750043

Storage: Omnimabs® V at -20°C, protected from light. Store Trans Enhancer at 2-8°C.

Features

◎Excellent primary cell transfection performance: The positive rate of cell transfection is generally above 90% with extraordinary mRNA knockdown effect, and the Lamin A/C mRNA knockdown efficiency in hepatocytes is above 95%;

◎Extremely Low Cytotoxicity: The cell death rate after transfection is less than 5%. No medium change is needed 4-6 hours after transfection, as this reagent will not increase cell death, and no impact on the accuracy of experimental results;

◎The vast majority of cells can be positively transfected in high efficiency, including most primary cells and ordinary adherent cell lines .

Components

Trans Enhancer	Omnimabs® V
3.4ml	0.1ml
10.2ml	0.3ml
25.5ml	0.75ml
51ml	1.5ml
510ml	15ml

Product overview

Omnimabs® V siRNA primary cell transfection reagent is specifically designed for the efficient transfection of small nucleic acid molecules such as siRNA and miRNA into primary cells. This reagent has excellent primary cell transfection performance. It can efficiently transfect small RNA molecules (200bp or less) such as siRNA, miRNA, and inhibitors into most primary cells, as well as into most ordinary adherent cell lines. It can even efficiently transfect small RNA molecules into some parasite larvae. In particular, the Omnimabs® V siRNA primary cell transfection reagent is formulated with novel biodegradable nanomaterials, exhibiting extremely low cytotoxicity. The viability of primary cells after transfection is almost unaffected, with a cell death rate of only about 5%, which is virtually indistinguishable from that of normally cultured primary cells. Serum has no effect on the transfection efficiency of Omnimabs® V. Therefore, there is no need to use specialized low-serum media, making experiment manipulation more convenient for researchers. In addition, this reagent is equipped with a Trans Enhancer specifically designed for preparing siRNA transfection complexes, eliminating the need for researchers to purchase Opti-MEM separately. This simplifies the purchasing process and significantly reduces the cost of use for researchers.

Protocol:

Use the following procedure to transfect siRNA into cells in a 24-well plate format. For other plate formats, please refer to the table below.

A. Cell seeding: Seed cells one day before transfection, using 500 µl of culture medium per well, so that the cell density is 40-60% at the time of transfection. Try to avoid using antibiotics.

B. Prepare siRNA-Omnimabs® V transfection complex:

1. Dilute 12 pmol siRNA in 25 µl of Trans Enhancer.
2. Dilute 1.5µl Omnimabs® V in 25 µl of Trans Enhancer. Mix gently and incubate at room temperature for 5 minutes. **Note:** Ensure that Step 3 is performed within 15 minutes.
3. After incubating for 5 minutes, mix the siRNA dilution with the Omnimabs® V dilution (total volume 50 µl). Mix gently and incubate at room temperature for 15 minutes.

C. Add complexes to cells in complete growth medium:

1. Add the 50 µl of complexes to each well containing cells and 0.5 ml medium. Mix gently by rocking the plate back and forth for several times.
2. Incubate cells at 37°C for 18-72 hours, then assess gene knockdown efficiency using RT-qPCR. For fam-labeled siRNA transfection, observe transfection positivity via fluorescence microscopy. Observations can be performed approximately 12 hours post-transfection.

Transfection Experiment Optimization:

To improve transfection efficiency, it is advisable to optimize the transfection conditions, particularly when performing the procedure for the first time. For example, for a 24-well culture plate, the amount of siRNA can be adjusted between 6, 12 and 18 pmol (final concentration 10-30 nM) per well, and the amount of Omnimabs® V reagent can be adjusted between 1.0 and 2.0 µl.

Important Guidelines for Transfection:

- Before using the transfection reagent, be sure to gently mix it with a pipette. Avoid using antibiotics during the transfection process, as this can increase cell death.
- Calculate the number of cells to be seeded, ensuring a cell density of approximately 40-60% at transfection. Generally, the seeding cell number for a 24-well plate is around 4 x 10^4 cells per well.
- When using fluorescently labeled siRNA such as Fam-NC for fluorescence observation, the recommended dosage per well of a 24-well plate is 24 pmol, and the dosage should be doubled for 12-well. Before observing fluorescence, it is recommended to aspirate and discard the original culture medium containing fluorescence, wash twice with PBS, and then observe under a fluorescence microscope.
- To minimize procedural steps and reduce well-to-well variability, if the same siRNA is to be added to two or more culture wells, the transfection complexes may be prepared collectively within a single sterile, clean polypropylene (PP) tube. In this pooled preparation, the volumes of siRNA,

Omnimabs® V transfection reagent, and Trans Enhancer should be calculated as the single-well volume multiplied by the total number of wells. Once the transfection complexes have been prepared, equal aliquots are then dispensed into each respective culture well. The specific volume of transfection complex to be added per well is as follows: 12 µL for 96-well plates, 26 µL for 48-well plates, 50 µL for 24-well plates, 100 µL for 12-well plates, and 250 µL for 6-well plates.

Omnimabs® V siRNA Transfection Reagent Formats for Various Cell Culture Vessels

Culture vessel	Surface area/well (cm^2)	Vol. of growth medium (µl)	Vol. of Trans Enhancer (µl)	siRNA Amount (pmol)	Omnimabs ® V (µl)
96-well	0.3	120	2 × 6	3	0.4
48-well	0.8	250	2 × 13	6	0.8
24-well	2	500	2 × 25	12	1.5
12-well	4	1000	2 × 50	24	3
6-well	10	2500	2 × 125	60	7.5

Storage and Transport

Use blue ice for transportation. Store Omnimabs® V at -20 °C, protected from light. Store Trans Enhancer at 2-8°C.

Validity Period

Valid for 1 year under the above storage conditions.

Quality Control

This product has been proven free of biological contamination through quality testing.

Note

This product is intended for research use only.