

Super⁺ siRNA Transfection Reagent OM642675

Product Description

OmnimAbs Super⁺ siRNA transfection reagents are a new generation of animal-origin free lipid transfection reagents specially designed for transfecting RNAi duplexes (Super⁺ siRNA) into eukaryotic cells. OmnimAbs and OmnimAbs PM are made from different lipids and perform excellent in most cell lines with some differences. We recommend use OmnimAbs for cell line and OmnimAbs PM for difficult and primary cell application.

Features

- High transfection efficiencies with maximum knockdown.
- Minimal cytotoxicity to reduce non-specific effects and cellular stress.
- Allow achieving high levels of knockdown despite differences in cell density and other variations.
- Strong resistance to serum interference, even in the presence of serum, it can have good transfection effect.

Experimental data

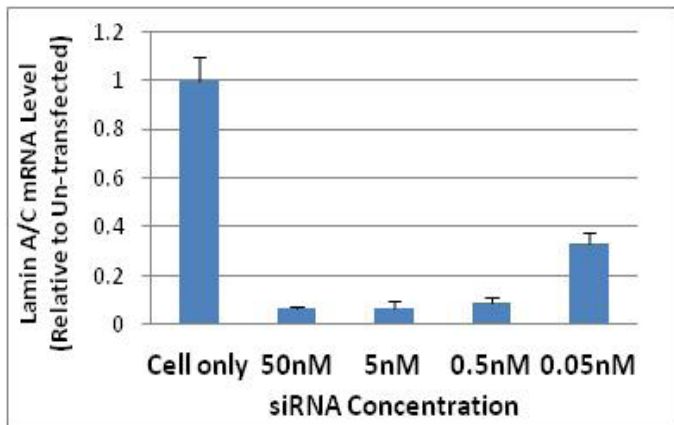


Fig. 1. OmnimAbs Super⁺ siRNA transfection reagent offers high level of gene knockdown with low concentration of Super⁺ siRNA. Lamin A/C Super⁺ siRNA-OmnimAbs transfection complexes were prepared in 96-well plates, and then A549 cells were added to each well.

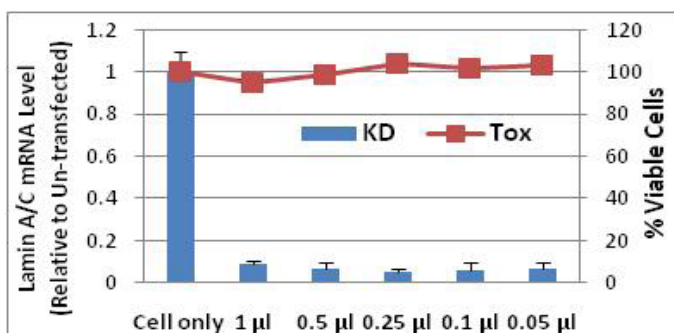


Fig. 2 OmnimAbs Super⁺ siRNA transfection reagent offers high level of gene knockdown with low toxicity. A549 cell lines were transfected with Lamin A/C Super⁺ siRNA (final concentration: 10nM) using OmnimAbs Super⁺ siRNA transfection reagent. OmnimAbs concentrations added to the cells exceeding transfection concentration of 20 times, the cells still do not produce significant toxicity, which makes these product suitable to optimization. Lamin A/C knockdown was measured by qRT-PCR.

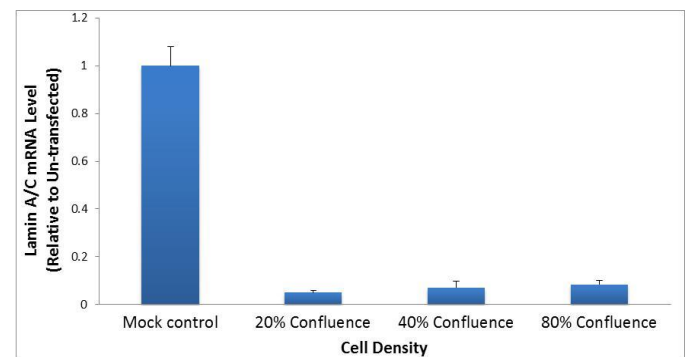


Fig. 3. OmnimAbs transfection reagent offers consistent high level of gene knockdown despite differences in cell density. A549 cells at different cell density were transfected with Lamin Super⁺ siRNA (final concentration: 10nM) using OmnimAbs.

Super+ siRNA Transfection Reagent Protocol

Product Description

OmnimAbs Super+ siRNA transfection reagents are a new generation of animal-origin free lipid transfection reagents specially designed for transfecting RNAi duplexes (Super+ siRNA) into eukaryotic cells. OmnimAbs and OmnimAbs PM are made from different lipids and perform excellent in most cell lines with some differences. We recommend use OmnimAbs for cell line and OmnimAbs PM for difficult and primary cell application. Super+ siRNA-OmnimAbs 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 but complexes may be removed after 4-6 hours.

Advantages of OmnimAbs Super+ siRNA Transfection Reagent

- High transfection efficiencies with maximum knockdown.
- Minimal cytotoxicity to reduce non-specific effects and cellular stress.
- Allow achieving high levels of knockdown despite differences in cell density and other variations.
- Strong resistance to serum interference, even in the presence of serum, it can have good transfection effect.

Storage, Stability, and Special Handling

OmnimAbs Super+ siRNA transfection reagent is stable for shipping at 2 - 10°C and long-term storage at -20°C. Trans Enhancer needs to be stored at 2 - 8°C. OmnimAbs Super+ siRNA transfection reagent is intended for research use only.

Important Guidelines for Transfection

- Do not add antibiotics to media during transfection as this may cause cell death.
- Use 20 nM Super+ siRNA as a starting point, although Super+ siRNA concentration can be much lower than 20 nM.
- Forward transfection and reverse transfection protocols can be used for most cell lines.

Protocol: Forward Transfection

Use the following procedure to transfect Super+ siRNA into cells in a 24-well format. For additional formats, see Scaling Up or Down Transfections. Optimize transfections as described in Optimizing Transfections, especially if transfecting a cell line for the first time. All amounts and volumes are given on a per well basis.

A. Plate cells

1. One day before transfection, plate cells in 500 µl of growth medium without antibiotics so that cells will be 30-50% confluent at the time of transfection.

B. Prepare Super+ siRNA-OmnimAbs complex

1. Dilute 12 pmol Super+ siRNA in 25 µl of Trans Enhancer.
2. Dilute 1.5 µl OmnimAbs in 25 µl of Trans Enhancer. Mix gently and incubate for 5 minutes at room temperature. **Note:** Proceed to Step 3 within 15 minutes.
3. After the 5 minute incubation, combine the diluted Super+ siRNA with the diluted OmnimAbs (total volume = 50 µl). Mix gently and incubate for 15 minutes at room temperature.

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 in a CO₂ incubator for 18-72 hours prior to testing for knockdown of gene expression. Medium may be changed after 4-6 hours, if needed, but not necessary. **Note:** The optimal incubation time for gene silencing analysis depends on the cell type, the gene targeted, and the method of analysis. This can be determined by performing a time-course experiment.

Protocol: Reverse Transfection

Use this procedure to reversely transfect Super+ siRNA into cells in a 24-well format (for other formats, see Scaling Up or Down Transfections). In reverse transfections, the complexes are prepared inside the wells, after which cells and medium are added. Reverse transfection is faster to perform than forward transfection and is the method of choice for high-throughput transfection. All amounts and volumes are given on a per well basis.

A. Prepare Super+ siRNA-OmnimAbs complex

1. Dilute 12 pmol Super+ siRNA in 25 µl of Trans Enhancer without serum.
2. Dilute 1.5 µl OmnimAbs in 25 µl of Trans Enhancer without serum. Mix gently and incubate for 5 minutes at room temperature. **Note:** Proceed to Step 3 within 15 minutes.
3. After the 5 minute incubation, combine the diluted Super+ siRNA with the diluted OmnimAbs (total volume = 50 µl). Mix gently and incubate for 15 minutes at room temperature.

B. Seed appropriate number of cells in 500 µl of an appropriate culture medium into the well on top of the Super+ siRNA-OmnimAbs complexes to give 30-50% confluence 24 hours after plating.

C. Incubate the cells with the transfection complexes under their normal growth conditions and monitor gene silencing after an appropriate time (e.g., 18-72 h after transfection, depending on experimental setup).

Optimizing Transfection: To achieve optimal transfection efficiency for each new cell line, it may be necessary to evaluate a number of parameters related to your transfection conditions, such as varying amount of Super+ siRNA and OmnimAbs reagent. Testing 6, 12, 18 pmol Super+ siRNA (final concentration 10 - 30 nM) and 1.0 - 2.0 µl OmnimAbs for 24-well format should be sufficient to achieve desired results.

Scaling Up or Down Transfections: To transfect cells in different tissue culture vessels, vary the amounts of OmnimAbs, Super+ siRNA, cells, and medium used in proportion to the surface area, as shown in the table.

OmnimAbs Transfection Reagent Formats for Various Cell Culture Vessels

Culture vessel	Surface area per well	Vol. of growth medium	Vol. of Trans Enhancer	Super+ siRNA Amount (pmol)	OmnimAbs (µl)
96-well	0.3 cm ²	100 µl	2 x 6 µl	3	0.4
48-well	0.8 cm ²	250 µl	2 x 13 µl	6	0.8
24-well	2 cm ²	500 µl	2 x 25 µl	12	1.5
12-well	4 cm ²	1 ml	2 x 50 µl	24	3.0
6-well	10 cm ²	2.5 ml	2 x 125 µl	60	7.5