Chemi - Trans™ RNAiMAX Transfection Reagent

（Cat #: T009, T010）

Introduction:

The Chemi - Trans™ RNAiMAX Transfection Reagent is a high efficiency RNA transfection reagent, derived from a type of natural compound and specifically designed for small RNA oligonucleotides (e. g, siRNA and miRNA.). This reagent can provide the robust transfection efficiency in a variety of commonly used and hard-to-transfect mammalian cells. Such as adherent and suspension cells, as well as primary cells.

Advantages:

- ❖ Robust transfection efficiency and low cytotoxicity.
- ❖ Excellent for small RNA transfection.
- ❖ Compatible with serum and antibiotics in culture medium.

Table 1: Product Package & Storage

Cat #	Product Name	Volume	Storage
T009	Chemi -Trans™ RNAiMAX Transfection Reagent	0.75 mL	2 ~ 8 °C,stable for up to 12 months when stored appropriately. (DO NOT FREEZE.)
T010		1.5 mL	

Important Guidelines for Transfection:

(1) For maximum transfection efficiency, using serum-free medium (such as Opti-MEM® Reduced Serum Medium) to dilute siRNA/miRNA mimics or miRNA inhibitor and the Chemi -Trans™ RNAiMAX Transfection Reagent is a must.

(2) While the standard protocols for siRNA transfection being given below, optimization is often needed for maximal transfection efficiency.

(3) It is unnecessary to wash cells and change medium after transfection.

Standard Protocol for siRNA Transfection of Adherent Cells

Step Ⅰ. Cell Seeding:

Cells should be plated 18 to 24 hours prior to transfection so that the monolayer cell density reaches to 50 ~ 80% confluency at the time of transfection. Complete culture medium with serum and antibiotics is freshly added to each well 30 ~ 60 min before transfection.

Step Ⅱ. siRNA Transfection Protocol

Use the following procedure to transfect siRNA into mammalian cells in a 6-well format. For other formats, please refer to ***A Guideline for siRNA transfection (Table 2)***. All amounts and volumes are given on a per well basis. For each transfection sample, prepare complexes as follows:

a. Dilute 2.5 µL (25 pmol) siRNA in 125 µL of serum-free Opti-MEM (or other medium without serum). Vortex to mix gently.

b. Mix Chemi - Trans™ RNAiMAX Reagent gently before use, then dilute 7.5 μL of Chemi - Trans™ RNAiMAX in 125 μL of serum-free Opti-MEM.

c. Add the diluted siRNA solution to the diluted Chemi - Trans™ Reagent all **(1:1 ratio)** at once. mix by pipetting up and down. Incubate for 5 minutes at room temperature to let transfection complex form well.

d. Add the 250 μL of complexes to each well containing cells and medium. Mix gently by rocking the plate back and forth.

e. Incubate cells at 37°C in a CO_2 incubator for 24 to 72 hours. Then, to measure the gene silencing by qRT-PCR and Western Blotting respectively.

Table 2: A Guideline for siRNA transfection per cell culture vessel

Culture Vessel	Growth Medium (mL)	Amount of Plating Cells	Serum - Free Medium (μL)	siRNA (μL)	Chemi - Trans™ RNAiMAX Reagent (μL)
	Volume used per well				
96-well	0.1	1 ~ 4 × 10^4	2 × 5	0.1	0.3
24-well	0.5	0.5 ~ 2 × 10^5	2 × 25	0.5	1.5
12-well	1.0	1 ~ 4 × 10^5	2 × 50	1.0	3.0
6-well	2.0	0.25 ~ 1 × 10^6	2 ×125	2.0	7.5

*[1] We strongly suggest reconstituting siRNA stock solution at 10 μM, so add 2.5 μL siRNA stock solution per well of 6-well plate to make final siRNA as 25 pmol conveniently.

[2] You may perform a rapid 96-well plate transfections by plating cells directly into the transfection complex. Prepare complexes in the plate and directly add cells drop wise at twice the cell density as in the basic protocol in a 100 μL growth medium. Cells will adhere as usual in the presence of complexes.