# Celthy Univer Liposomal Transfection Reagent

## Product description

Celthy Univer Liposomal Transfection Reagent is a versatile liposome transfection reagent, suitable for DNA, RNA and oligonucleotide transfection, with high transfection efficiency for most eukaryotic cells. Its unique formulation allows for direct addition to the culture medium without serum affecting transfection efficiency, thus reducing serum-related cell damage. There is no need to remove the nucleic acid-Celthy complex or replace with fresh medium after transfection, and it can also be removed after 4~6 hours.

Celthy is supplied in sterile liquid form. Usually, for 24-well plate transfection, about 1.5 μL each time, 1 mL of Celthy can do about 660 transfections; for 6-well plate, about 6 μL each time, 1 mL of Celthy can do about 660 transfections.

## Specifications

| | |
|---|---|
| Catalog Number | C130002S/C130002M |
| Specifications | 0.5 mL/1 mL |

## Properties

| | |
|---|---|
| Form | Liquid |
| Serum Compatible | Yes |
| Cell Type | Established Cell Lines |
| Sample Type | Plasmid DNA, Synthetic siRNA |
| Transfection Technique | Lipid-Based Transfection |

## Storage

Store at 2~8°C, with a shelf life of 1 year. Do not freeze!

## Notes

1. Celthy Univer Liposomal Transfection Reagent requires a high cell plating density, preferably 90%~95%, which helps to reduce the impact of cationic liposome cytotoxicity; if the gene you are studying requires a long time, for example, cell cycle-related genes or cell surface proteins, it is best to choose transfection reagents with lower cell plating density requirements, and liposomal nucleic acid transfection reagents are not suitable.
2. Celthy Univer Liposomal Transfection Reagent can be used for transfection with serum medium, and it is not necessary to change the medium before and after transfection. However, preparation of transfection complexes requires dilution of DNA and transfection reagents in serum-free medium

because serum can affect complex formation. In addition, to test the compatibility of the serum-free medium used with the liposomal nucleic acid transfection reagent, CD293, SFMII, VP-SFM are known to be incompatible.

3. Antibiotics should not be added to the medium during transfection.

4. Using high-purity DNA or RNA helps achieve higher transfection efficiency, as endotoxins in plasmids are detrimental to transfection.

5. Cationic liposomes should be stored at 4°C, and be careful not to open the lid repeatedly for a long time, because it may cause liposome oxidation and affect the transfection efficiency.

6. The DNA concentration and the amount of cationic liposome reagent should be optimized for maximum transfection efficiency for initial use. The ratio of DNA and transfection reagent is usually recommended to be 1:2~1:3, such as seeding 0.5~2×$10^5$ cells in a 24-well plate, using 0.5 μg DNA and 1~1.5 μL of transfection reagent. Optimize the transfection efficiency by adjusting the ratio of DNA/Celthy Univer Liposomal Transfection Reagent to ensure that the cell density is greater than 90%, and the ratio of DNA (μg): Celthy (μL) is 1:0.5~1:5.

7. For research use only.

## Instructions

### 1. Transfection in a 24-well plate

【Note】The amount of transfection reagent used depends on the cell type and other experimental conditions. It is recommended to optimize the optimal amount through gradient settings during initial use.

Adherent cells: One day before transfection (20~24 hours, trypsinize cells and count, and cells were plated (without antibiotics to a density of 90~95% at the time of transfection (0.5~2×$10^5$ cells/well for a 24-well plate.

Suspension cells: On the day of transfection, before preparing DNA complexes, plate cells in 24-well plates at 4~8×$10^5$ cells per 500 μL of growth medium (without antibiotics).

1) Prepare the DNA-Celthy Univer Liposome Transfection Reagent Complex as follows:

a. For each well of cells, dilute 0.5 μg of DNA with 50 μL of serum-free medium (such as OPTI-MEM I medium). Mix well.

b. For each well of cells, dilute 0.6~2.5 μL of Celthy Univer Liposome Transfection Reagent with 50 μL of serum-free medium (such as OPTI-MEM I medium).

Dilute Celthy Univer Liposome Transfection Reagent and incubate at room temperature for 5 mins (mix with the diluted DNA within 30 mins, if the incubation time is too long, the activity will be reduced)

【Note】Even if the Celthy Univer Liposome Transfection Reagent is diluted with OPTI-MEMI, cells can be cultured in DMEM. If DMEM is used as the diluent for the lipid transfection reagent, the DNA must be mixed within 5 minutes of dilution.

2 Mix the diluted DNA and the diluted liposomal nucleic acid transfection reagent (total volume 100 μL, mix gently, and incubate at room temperature (15~25°C for 20 mins to allow the formation of DNA-liposome complexes. The solution may be cloudy at this point, but it will not affect transfection.

【Note】DNA-liposome complexes are stable at room temperature for at least 5 h.

3 Add 100 μL of DNA-Celthy complex directly to each well of the cell culture plate, shake the plate, and mix gently.

【Note】If transfecting under serum-free conditions, use serum-containing normal growth medium for cell plating. Growth medium was removed before complex addition and replaced with 500 μL of serum-free medium.

4 Incubate at 37°C in a 5% $CO_2$ incubator for 24~48 h until transgene expression analysis without removing complexes or changing medium. However, it may be necessary to change the growth medium after 4~6 h without reducing transfection activity.

**Stably transfected cell line:** 24 h after transfection, fresh growth medium was added to the cells at a ratio of 1:10 or higher, and selection medium was added 48 h after transfection.

**Suspension cell line:** After the DNA-Celthy complex was added to the cells, PMA and/or PHA could be added 4 h later if desired. For Jurkat cells, final concentrations of PHA and PMA were 1 μg/mL and 50 ng/mL, respectively, which increased CMV promoter activity and gene expression. For K562 cells, the addition of PMA alone was sufficient to increase promoter activity.

## 2. Adjustment of transfection system

For different cell culture plates, the amount of Celthy Univer Liposome Transfection Reagent, DNA, cells and medium will be different, please refer to the following table (Table 1) for details. For 96-well plate culture, there is no need to plate cells one day in advance, and the complex can be prepared directly in the plate, and then the cell suspension can be added to the complex, which further reduces the transfection time. This modified procedure has been tested with 293-H, 293-F, COS-7L and CHO cells and is slightly less active than the traditional method. Fast steps and efficient transfection of protein-expressing cell lines make liposomal nucleic acid transfection reagents ideal for high-throughput transfection in 96-well plates, such as cDNA library screening and transient protein expression.

**Table 1 Amounts of liposomal nucleic acid transfection reagent, nucleic acid, cells and medium for different culture vessels**

| Culture Vessel | Surf. area per well[1] | Shared Reagents | | DNA Transfection | | RNAi Transfection | |
|---|---|---|---|---|---|---|---|
| | | Vol. of plating medium | Vol. of dilution medium[2] | DNA | Transfection Reagent | RNA | Vol.of plating medium |
| 96-well | 0.3 $cm^2$ | 100 μL | 2×25 μL | 0.1 μg | 96-well | 0.3 $cm^2$ | 100 μL |

| | | | | | | | |
|---|---|---|---|---|---|---|---|
| 24-well | 2 $cm^2$ | 500 µL | 2×50 µL | 0.5 µg | 24-well | 2 $cm^2$ | 500 µL |
| 12-well | 4 $cm^2$ | 1 mL | 2×100 µL | 1 µg | 12-well | 4 $cm^2$ | 1 mL |
| 6-well | 10 $cm^2$ | 2 mL | 2×250 µL | 2~4 µg | 6-well | 10 $cm^2$ | 2 mL |
| 60-mm | 20 $cm^2$ | 5 mL | 2×0.5 mL | 4~8 µg | 60-mm | 20 $cm^2$ | 5 mL |
| 10-cm | 60 $cm^2$ | 15 mL | 2×1.5 mL | 12-24 µg | 10-cm | 60 $cm^2$ | 15 mL |

1) The surface area of cell culture plates provided by different manufacturers may vary.

2) Volume of medium used to dilute DNA or RNAi.

【Note】The usage amount in this table is for reference only, and the specific usage amount needs to be optimized according to the cell type and other experimental conditions. DNA (µg): Celthy (µL) ratio was kept at 1:0.5~1:5 when use.