

LipoTrust™ EX Gene



COSMO BIO CO., LTD.
Inspiration for Life Science

■ Contents

LipoTrust™ EX Gene

■ Amount and Storage

Lyophilized powder containing 1 μmol of cationic lipid suitable for 1 ml To be kept at 2-8 C

■ Description

LipoTrust™ EX Gene Reagent is a proprietary cationic liposome formulation that facilitates highly efficient delivery of **Plasmid DNA** to mammalian cells.

Lyophilized powder is stable so that customers can use this as long as customer keeps as instructed.

■ How to use

Transfection Procedure

A-1. Preparation of liposome suspension

1. Remove the aluminum cap of vial by tool with extreme care and take off the rubber cap.
2. Add 1 mL of **nuclease free** water into the **LipoTrust™ EX Gene** vial and gently shake the vial until no powder is remained. Be kept for 10 minutes before use. Please store this suspension at 2-8°C.

A-2. Transfection

Use this procedure to transfect Plasmid DNA into mammalian cells using **LipoTrust™ EX Gene** per 1 well in a 24-well format. For other formats, see the table in **Recommended Reagent Amounts and Volumes** for appropriate reagent amounts to add.

Tip: To reduce well-to-well variability when transfecting multiple replicates (triplicates), proportionally scale up the reagent volumes to form complexes, then aliquot an equal volume of complexes into each well.

STEP 1. Cell plating

One day before transfection, plate cells and culture them for 1 day in ordinary growth medium without antibiotics per well. Cells should be 60-80% confluent at the time of transfection. (ex. HeLa Cell: 2-4x10⁴ cells/well)

STEP 2. Preparation of Transfection Complex and Application to Cells

1. Add 0.8μg of **Plasmid DNA** in 50 μl of serum free medium previously prepared in a suitable microtube and mix well.
2. Add 2 μl of prepared **LipoTrust™ EX Gene** suspension (**A-1**) into 50μl of serum free medium previously prepared in a suitable microtube and mix well gently.
3. Mix liquid of **STEP 2-1** with that of **STEP 2-2** well and kept for 20 minutes to form **Transfection complex** at room temperature.
* Either liquid can be poured into the other microtube to form **Transfection complex** in this case.
4. Transfer all of this **complex** to well in which cells were plated (**STEP 1**) and mix well with gently rocking the plate back and forth.

STEP 3. Incubation

Incubate the cells at 37°C for 18 to 48 hours.

Incubation time may be varied according to cells or Plasmid DNA so that previous test to examine appropriate conditions is recommended.

Recommended Reagent Amounts and Volumes per 1 well

| STEP | 1 | 2-A) | | 2-B) | |
|------------|--------------------|--|---------------------------|---|----------------------------------|
| Plate/dish | Medium volume/well | Serum-free medium dilution for Plasmid DNA | Plasmid DNA ^{*1} | Serum-free medium dilution for LipoTrust™ EX Gene | LipoTrust™ EX Gene ^{*1} |
| 96 well | 100 μl | 25 μl | 0.2 μg | 25 μl | 0.5 μl |
| 48 well | 200 μl | 50 μl | 0.4 μg | 50 μl | 1 μl |
| 24 well | 500 μl | 50 μl | 0.8 μg | 50 μl | 2 μl |
| 12 well | 1ml | 100 μl | 1.6 μg | 100 μl | 4 μl |
| 6 well | 2.5 ml | 250 μl | 4 μg | 250 μl | 10 μl |
| 10 cm | 15 ml | 1.5 ml | 24 μg | 1.5 ml | 60 μl |

^{*1} Optimum amount of **Oligonucleotides** and **LipoTrust™ EX Gene** may be varied according to kind of cells or conditions. Variation from 1/4 to 4 times of **LipoTrust™ EX Gene** is recommended to obtain optimum transfection efficiency.

■ TIP for Transfection

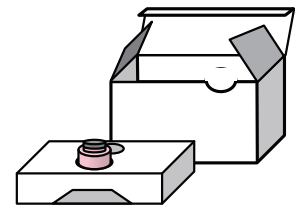
1. Although good transfection efficiency is observed with **LipoTrust™ EX Gene** under serum containing medium (Ex:FEB 10%), use serum free medium at the time of preparation of **Transfection complex**.
2. Use healthy cells
3. Use medium free of antibiotics
4. Use high quality of oligonucleotide
5. Optimum amount of **Plasmid DNA** and **LipoTrust™ EX Gene** may be varied according to kind of cells or conditions. Variation from 1/4 to 4 times of **LipoTrust™ EX Gene** is recommended to obtain optimum transfection efficiency.

This product is distributed for laboratory research use only. Caution: Not for diagnostic, food, cosmetic or household use. The safety and efficacy of this product in diagnostic or other clinical uses has not been established. Never inject human.



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- Inner box can be used as a vial rack.

- Product appearance is subject to changes without notice.