TransIT[®]-LT1 Transfection Reagent

Quick Reference Protocol

Instructions for MIR 2300, 2304, 2305, 2306, 2310 Full protocol, SDS and Certificate of Analysis available at mirusbio.com/2300

SPECIFICATIONS

Storage	Store <i>Trans</i> IT [®] -LT1 Transfection Reagent tightly capped at 4°C. <i>Before each use</i> , warm to room temperature and vortex gently.
Product Guarantee	1 year from the date of purchase, when properly stored and handled.

PLASMID DNA TRANSFECTION PROTOCOL



Full protocol and additional documentation available at *mirusbio.com/2300*

Fill in volumes below based on culture vessel used for transfection (Table 1).

- A. Plate cells
 - 1. Plate cells in ____ml complete growth medium (per well).

For adherent cells: Plate cells at a density of $0.8 - 3.0 \times 10^5$ cells/ml.

For suspension cells: Plate cells at a density of $2.5-5.0 \times 10^5$ cells/ml.

2. Culture overnight. Most cell types should be ≥80% confluent on day of transfection.

B. Prepare TransIT®-LT1 Reagent:DNA complexes

- 1. Warm *Trans*IT[®]-LT1 Reagent to room temperature and vortex gently.
- 2. Place ____µl of OptiMEM[®] I Reduced-Serum Medium in a sterile tube.
- 3. Add ____µl plasmid DNA. Mix gently by pipetting.
- 4. Add ____µl of *Trans*IT[®]-LT1 Reagent. Mix gently by pipetting.
- 5. Incubate at room temperature for 15-30 minutes.

C. Distribute complexes to cells

- 1. Add *Trans*IT[®]-LT1:DNA complex mixture drop-wise to different areas of the well.
- 2. Gently rock plate for even distribution of complexes.
- 3. Incubate 24-72 hours.
- 4. Harvest cells and assay as required.

Culture vessel	24-well plate	12-well plate	6-well plate
Surface area	1.9 cm ²	3.8 cm ²	9.6 cm ²
Complete growth medium	0.5 ml	1 ml	2.5 ml
Serum-free medium	50 µl	100 µl	250 µl
DNA (1 µg/µl stock)	0.5 μl	1 µl	2.5 μl
TransIT [®] -LT1 Reagent	1.5 μl	3 μΙ	7.5 μl

Table 1. Recommended starting conditions

Transfection Optimization

Determine the best *Trans*|T[®]-LT1 Reagent:DNA ratio for each cell type. Start with 3 μ of *Trans*|T[®]-LT1 Reagent per 1 μ g of DNA. Vary the concentration of *Tran*|T[®]-LT1 Reagent from 2–6 μ l per 1 μ g DNA to find the optimal ratio.

For additional optimization tips, see <u>full protocol</u>. Cell-type-specific recommendations available at **Reagent Agent:** mirusbio.com/ra

NOTES



Reagent Agent^{*} is an online tool designed to help determine the best solution for nucleic acid delivery based on in-house data, customer feedback and citations.

Learn more at: mirusbio.com/ra

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