*Trans*IT®-2020 Transfection Reagent

Quick Reference Protocol

Instructions for MIR 5400, 5404, 5405, 5406, 5410

Full protocol, SDS and Certificate of Analysis available at mirusbio.com/5400

SPECIFICATIONS

Storage	Store *Trans*IT®-2020 Reagent tightly capped at -20°C. ***Before each use***, 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/5400***

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 × 10^5 cells/ml.
   For suspension cells: Plate cells at a density of 2.5-5.0 × 10^5 cells/ml.
2. Culture overnight. Most cell types should be approximately 80% confluent at the time of transfection.

B. Prepare *Trans*IT®-2020 Reagent:DNA complexes

1. Warm *Trans*IT®-2020 to room temperature and vortex gently.
2. Place ___µl of Opti-MEM® I Reduced-Serum Medium in a sterile tube.
3. Add ___µl plasmid DNA. Mix gently by pipetting.
4. Add ___µl of *Trans*IT®-2020 Reagent. Mix gently by pipetting.
5. Incubate at room temperature for 15-30 minutes.

C. Distribute complexes to cells

1. Add *Trans*IT®-2020: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.

Table 1. Recommended starting conditions

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
***Trans*IT®-2020 Reagent**	1.5 µl	3 µl	7.5 µl

Transfection Optimization

Determine the best *Trans*IT®-2020 Reagent:DNA ratio for each cell type. Start with 3 µl of *Trans*IT®-2020 Reagent per 1 µg of DNA. Vary the concentration of *Trans*IT®-2020 Reagent from 1-4 µl per 1 µg DNA to find the optimal ratio.

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

NOTES

Reagent Agent®

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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