# TransIT®-Keratinocyte Transfection Reagent

Quick Reference Protocol

Instructions for MIR 2800, 2804, 2805, 2806, 2810
Full protocol, SDS and Certificate of Analysis available at mirusbio.com/2800



### **SPECIFICATIONS**

Storage	Store <i>Trans</i> IT®-Keratinocyte 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**



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

#### A. Plate cells

- Plate cells in \_\_\_ml complete growth medium (per well).
- 2. Culture overnight. Most cell types should be ≥80% confluent on day of transfection.

## B. Prepare TransIT®-Keratinocyte Reagent:DNA complexes

- 1. Warm *Trans*IT®-Keratinocyte 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®-Keratinocyte Reagent. Mix gently by pipetting.
- 5. Incubate at room temperature for 15-30 minutes.

### C. Distribute complexes to cells

- 1. Add *Trans*IT®-Keratinocyte:DNA complex mixture drop-wise to different areas of the
- 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 <sup>2</sup>	3.8 cm <sup>2</sup>	9.6 cm <sup>2</sup>
Complete growth medium	0.5 ml	1 ml	2.5 ml
Serum-free medium	50 μΙ	100 μΙ	250 μΙ
DNA (1 μg/μl stock)	0.5 μΙ	1 μΙ	2.5 μΙ
TransIT®-Keratinocyte Reagent	1.5 μΙ	3 μΙ	7.5 µl

### **▶** Transfection Optimization

Determine the best *Trans*IT\*-Keratinocyte Reagent:DNA ratio for each cell type. Start with 3 µl of *Trans*IT\*-Keratinocyte Reagent per 1 µg of DNA. Vary the concentration of *Trans*IT\*-Keratinocyte Reagent from 2–6 µl per 1 µg DNA to find the optimal ratio.

For additional optimization tips, see full protocol.



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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