# **TransIT-PRO® Transfection Reagent**

**Quick Reference Protocol** 

Instructions for MIR 5720, 5730, 5740 and 5750
Full protocol, SDS and Certificate of Analysis available at mirusbio.com/5740



## **SPECIFICATIONS**

Storage	Store <i>Trans</i> IT-PRO® Reagent tightly capped at -20°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/5740

#### Fill in volumes below based on total culture volume (Table 1).

### A. Maintenance of cells

- Passage suspension CHO or 293 cells 18-24 hours prior to transfection to ensure that cells are
  actively dividing at the time of transfection. DO NOT proceed with transfection if cells are not
  doubling every 24 hours or are < 95% viable by trypan blue exclusion.</li>
- 2. Incubate cells overnight at appropriate temperature and CO<sub>2</sub> levels (e.g. 37°C, 5-8% CO<sub>2</sub>, shaking).

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

- 1. Seed cells at a density of  $2 \times 10^6$  cells/ml immediately before transfection. DO NOT proceed with transfections if cells are not doubling normally or are not at high viability.
- 2. Warm TransIT-PRO® Reagent to room temperature and vortex gently.
- 3. Place \_\_\_ml of serum-free medium (e.g. Opti-MEM® or Opti-PRO<sup>TM</sup> SFM)\* in a sterile tube.
- 4. Add µg plasmid DNA. Mix gently by pipetting.
- 5. Add µl of *Trans*IT-PRO® Reagent. Mix gently by pipetting.
- 6. Incubate complexes at room temperature for recommended amount of time: For complexes that will be added to suspension 293 cells, incubate for 15-20 minutes. For complexes that will be added to suspension CHO cells, incubate for 5-10 minutes.

## C. Distribute complexes to cells

- 1. Add TransIT-PRO® Reagent: DNA complexes to cultured cells.
- Incubate cells for 2-14 days depending on cell type, culture temperature, nature of the protein, and detection method. For further optimization information, please see the full protocol.
- 3. Harvest cells and/or supernatant and assay as required.

Table 1. Volume scaling worksheet for DNA transfections with TransIT-PRO® Transfection Reagent.

Starting conditions per milliliter of complete growth medium							
	Per 1 ml		Total culture volume		Reagent quantities		
Serum-free Complex Medium	0.1 ml	×	ml	=	ml		
Plasmid DNA (1 μg/μl stock)	1 μΙ	×	ml	=	µІ		
TransIT-PRO® Reagent	1 μΙ	×	ml	=	µІ		

## **▶** Transfection Optimization

Determine the best TransIT-PRO® Reagent:DNA ratio for each cell type. Start with 1  $\mu$ I of TransIT-PRO® Reagent per 1  $\mu$ g of DNA. Vary the concentration of TransIT-PRO® Reagent from 0.5-2  $\mu$ I per 1  $\mu$ g DNA to find the optimal ratio. TransIT-PRO® Transfection Reagent is a key component of the CHOgro® Expression System (MIR 6260) and the CHOgro® High Yield Expression System (MIR 6270), which are optimized platforms for transient, high titer protein production in suspension CHO derived cells.

For additional optimization tips, see full protocol.

<sup>\*</sup>When using TransIT-PRO® Reagent with the CHOgro® Expression System, form transfection complexes in CHOgro® Complex Formation Solution.



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

©1996-2025 All rights reserved. Mirus Bio LLC. All trademarks are the property of their respective owners. For terms and conditions, visit www.mirusbio.com

Rev.A 042721