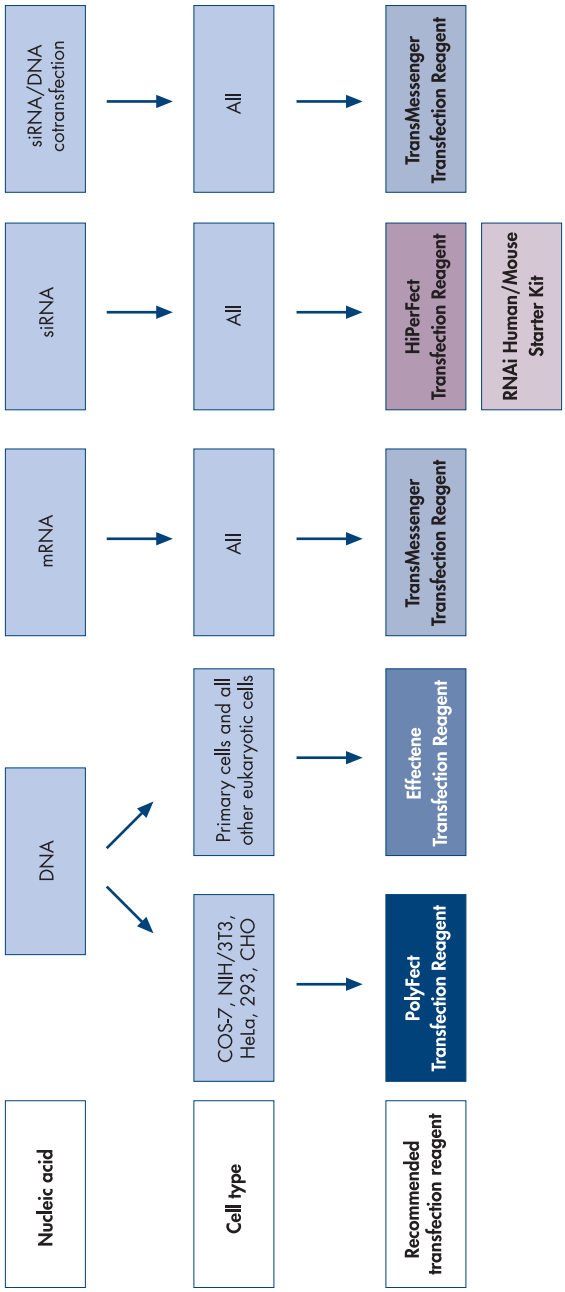
7

Transfection

www.qiagen.com/PG/TransfectionTools

Transfection selection guide

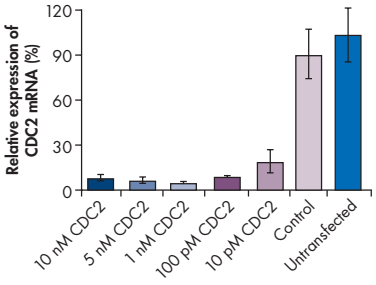


Features of QIAGEN Transfection Reagents

Feature \ Product	PolyFect Transfection Reagent	Effectene Transfection Reagent	HiPerFect Transfection Reagent	RNAi Human/Mouse Starter Kit*	TransMessenger Transfection Reagent
Nucleic acid	DNA	DNA	siRNA	siRNA	mRNA, siRNA/plasmid DNA cotransfection
Benefit for transfection	Fast and easy for standard cell lines	Highly effective for primary and sensitive cells	Low siRNA amounts minimize off-target effects	Easy establishment of RNAi in your lab	Efficient cotransfection
Cell lines	COS-7, NIH/3T3, HeLa, 293, and CHO cells	Eukaryotic cell lines and primary cells	Eukaryotic cell lines, primary cells, suspension cells, and macrophages	Eukaryotic cell lines and primary cells	Eukaryotic cell lines
Transfection in presence of serum	Yes	Yes	Yes	Yes	Yes
Reverse transfection	Yes	Yes	Yes	Yes	Yes
Tested for endotoxins	Yes	Yes	–	–	Yes
Tested for absence of RNases	–	–	Yes	Yes	Yes

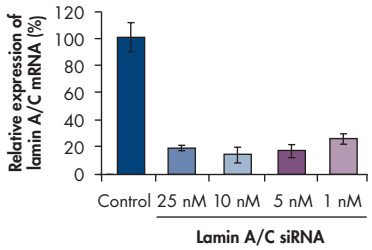
* For more information about the RNAi Human/Mouse Starter Kit, see page 235.

Efficient Transfection of Picomolar siRNA Amounts



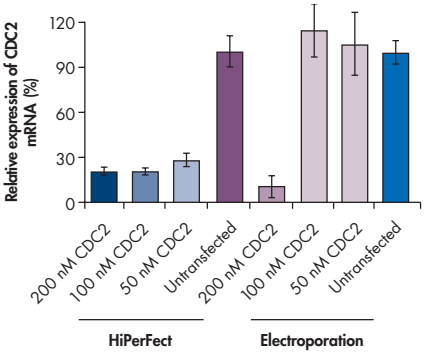
HeLa S3 cells were transfected with a range of amounts of CDC2 siRNA and with a nonsilencing control siRNA (**Control**) using HiPerFect Transfection Reagent (3 µl) in a 24-well plate format. After 48 hours, CDC2 expression was analyzed by quantitative, real-time RT-PCR.

Successful Transfection of Primary Cells



Normal Human Lung Fibroblasts were transfected with siRNA targeted against lamin A/C using HiPerFect Transfection Reagent. Nonsilencing siRNA was also transfected (**Control**). After 48 hours, knockdown was analyzed by quantitative, real-time RT-PCR.

More Effective Transfection of Suspension Cells than Electroporation



Jurkat suspension cells were transfected with various amounts of CDC2 siRNA either using HiPerFect Transfection Reagent or by electroporation. Untransfected cells were also analyzed. After 48 hours, gene expression was analyzed by quantitative, real-time RT-PCR.

HiPerFect Transfection Reagent

For high knockdown using RNAi with reduced off-target effects

- Efficient transfection with minimal risk of off-target effects using low siRNA concentrations
- Effective transfection of primary cells with high cell viability
- Effective transfection of suspension cells and macrophages

Product description

HiPerFect Transfection Reagent is a unique blend of cationic and neutral lipids that enables effective siRNA uptake and efficient release of siRNA inside cells resulting in high gene knockdown even when using low siRNA concentrations.

Applications

HiPerFect Transfection Reagent enables highly efficient siRNA transfection using low siRNA concentrations for applications such as:

- RNA interference studies
- microRNA transfection
- Studies on gene expression and function
- High-throughput reverse transfection for RNAi screening in combination with HP siRNA Sets (pages 225–233)

A list of cell types that have been successfully transfected using HiPerFect Transfection Reagent and experimental details are available at www.qiagen.com/PG/TransfectionTools .

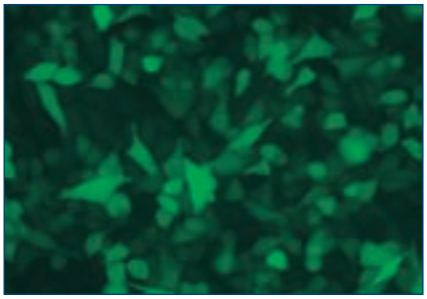
QIAGEN provides a comprehensive range of products and services for RNAi, see Chapter 6.

Product	Contents	Cat. no.
HiPerFect Transfection Reagent (0.5 ml)*	HiPerFect Transfection Reagent for up to 166 transfections in 24-well plates	301704
HiPerFect Transfection Reagent (1 ml)*	HiPerFect Transfection Reagent for up to 333 transfections in 24-well plates	301705
HiPerFect Transfection Reagent (4 x 1 ml)*	HiPerFect Transfection Reagent for up to 1332 transfections in 24-well plates	301707
HiPerFect Transfection Reagent (100 ml)*	HiPerFect Transfection Reagent for transfections in up to 1388 96-well plates	301709
RNAi Human/Mouse Starter Kit (page 235)*	0.5 ml HiPerFect Transfection Reagent, siRNA Suspension Buffer, Nonsilencing Control siRNA (Alexa Fluor® 488 labeled), Hs/Mm_MAPK1 Control siRNA	301799

* Bulk quantities available; please inquire.

For further information: www.qiagen.com/PG/TransfectionTools

Efficent Transfection Using TransMessenger Reagent



Expression of green fluorescent protein (GFP) in HeLa S3 cells. 8 x 10^4 cells were seeded into 48-well plates and transfected 24 hours later with 0.5 µg of an in vitro-transcribed GFP-encoding RNA (transcribed from P7ASP-GFP/Mlu) using 1 µl Enhancer R and 2.5 µl TransMessenger Reagent. Cells were analyzed 24 hours post-transfection by fluorescence microscopy. Approximately 50% of the cells were successfully transfected. (P7ASP-GFP/Mlu kindly provided by J. Bogenberger, Stanford University Blood Center, Palo Alto, CA, USA.)

TransMessenger® Transfection Reagent

For efficient plasmid DNA–siRNA cotransfection or mRNA transfection

- Highly efficient mRNA transfection and plasmid DNA/siRNA cotransfection
- Reproducible, reliable results ensured by stringent quality control
- Efficient transfection of primary neuronal cells (see reference)

Product description

TransMessenger Transfection Reagent is a ready-to-use, lipid-based reagent for RNA transfection or siRNA–plasmid DNA cotransfection of eukaryotic cells.

Applications

Transfection of RNA provides an alternative to DNA transfection and has been used for a number of specific applications including:

- Antisense RNA and RNA interference (RNAi) studies
- Transfection of non-dividing cells
- Direct studies of RNA viruses
- RNA translation studies

Reference

Krichevsky A.M. and Kosik K.S. (2002) RNAi functions in cultured mammalian neurons. Proc. Natl. Acad. Sci. USA **99**, 11926.

Product	Contents	Cat. no.
TransMessenger Transfection Reagent (0.5 ml)	For 60 transfections in 6-well plates or 80 transfections in 12-well plates	301525

For further information: www.qiagen.com/PG/TransfectionTools

PolyFect® Transfection Reagent

For fast and easy DNA transfection of standard cell lines

- Savings of time and effort with fast procedure and easy handling
- Optimized, highly effective transfection using cell-specific protocols
- High cell viability, low cytotoxicity

Product description

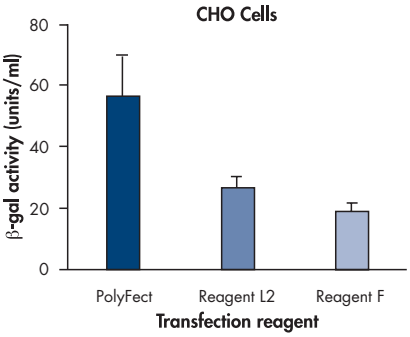
PolyFect Transfection Reagent is based on activated dendrimer technology optimized for the transfection of DNA into COS-7, NIH/3T3, HeLa, 293, and CHO cells.

Applications

PolyFect Transfection Reagent is highly suited for efficient transfection of standard cell lines in:

- Studies on gene expression and function
- Drug discovery and development studies
- High-throughput reverse transfection

High Transfection Efficiencies Using PolyFect Reagent



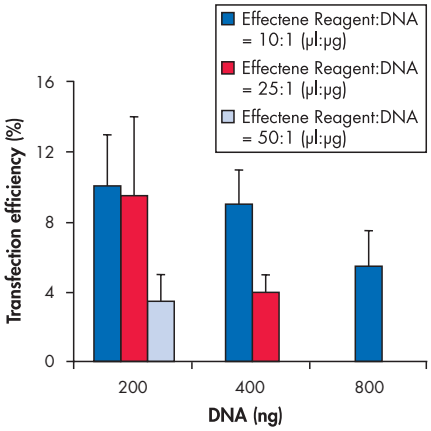
Comparison of transfection efficiencies for PolyFect Reagent and two lipid-based reagents. CHO cells cultured in 6-well plates were transfected with a β-galactosidase reporter plasmid. The appropriate protocol was used for transfection with PolyFect Reagent. Optimized protocols were used for transfection with Reagents L2 and F; these were 10 µl Reagent L2 with 3 µg DNA and 15 µl Reagent F with 2 µg DNA. All transfections were performed in triplicate.

Product	Contents	Cat. no.
PolyFect Transfection Reagent (1 ml)*	For 25–65 transfections in 60 mm dishes or 50–100 transfections in 6-well plates	301105
PolyFect Transfection Reagent (4 x 1 ml)*	For 100–260 transfections in 60 mm dishes or 200–400 transfections in 6-well plates	301107
PolyFect Transfection Reagent (100 ml)*	For 2500–6500 transfections in 60 mm dishes or 5000–10,000 transfections in 6-well plates	301108

* Bulk quantities available; please inquire.

For further information: www.qiagen.com/PG/TransfectionTools

Efficient Transfection of Primary Neuronal Cells Using Effectene Reagent



Primary neuronal cells, cultivated on coverslips in 24-well plates, were transfected with plasmid DNA expressing green fluorescent protein. A range of plasmid DNA to Effectene Transfection Reagent ratios was transfected. Transfection efficiencies are expressed as the percentage of cells that were GFP positive. Highest transfection efficiencies were obtained using 200 ng of DNA with 2–5 µl of Effectene Reagent. (Data kindly provided by J. Meier, I. Strömel, R. Iosub, S. Schmidt, and R. Grantyn, Humboldt University Medical School [Charité], Berlin, Germany.)

Effectene® Transfection Reagent

For DNA transfection of primary cells

- Ideal for primary cells and sensitive cell lines, far lower cytotoxicity and gentler than many alternative reagents
- High transfection efficiency in the presence of serum and with low DNA amounts
- Easy procedure saves effort

Product description

Effectene Transfection Reagent is a non-liposomal lipid reagent for DNA transfection into a broad range of cell lines and primary cells.

Applications

Effectene Transfection Reagent is highly suited for efficient transfection of a wide variety of eukaryotic cells, particularly primary cells, in:

- Studies on gene expression and function
- Drug discovery and development studies
- High-throughput reverse transfection

Product	Contents	Cat. no.
Effectene Transfection Reagent (1 ml)*	1 ml Effectene Reagent, Enhancer, Buffer EC; for 40 transfections in 60 mm dishes or 160 transfections in 12-well plates	301425
Effectene Transfection Reagent (4 x 1 ml)*	4 x 1 ml Effectene Reagent, Enhancer, Buffer EC; for 160 transfections in 60 mm dishes or 640 transfections in 12-well plates	301427

* Bulk quantities available; please inquire.

For further information: www.qiagen.com/PG/TransfectionTools

SuperFect® Transfection Reagent

Activated dendrimer for transfection of a broad range of eukaryotic cell lines with DNA

- Suitable for a broad range of cell lines
- Transfection can be carried out in the presence of serum
- Excellent reproducibility due to precise size and defined shape of activated dendrimers

Product description

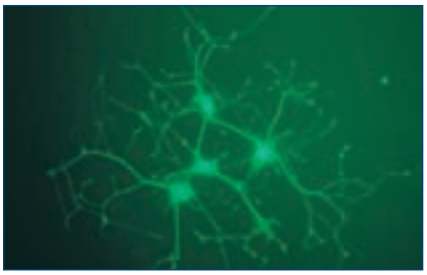
SuperFect Transfection Reagent is based on activated dendrimer technology developed for DNA transfection into a broad range of cell lines.

Applications

SuperFect Transfection Reagent is suited for efficient transfection of a wide variety of eukaryotic cells in:

- Studies on gene expression and function
- Drug discovery
- Development studies

Transfection of Neuronal PC-12 Cells Using SuperFect Reagent



Expression of green fluorescent protein (GFP) in differentiated PC-12 cells 5 days post-transfection. 10^4–10^5 cells previously stimulated with 50 ng/ml NGF were plated per 60 mm dish one day prior to transfection. Transient transfections were performed in 2 ml low-serum growth medium (D-MEM plus 0.05% FBS) using 3 µg of a GFP reporter plasmid and 15 µl SuperFect Reagent. (Data kindly provided by K. Kelly-Spratt, University of Texas Southwestern Medical Center, Dallas, TX, USA.)

Product	Contents	Cat. no.
SuperFect Transfection Reagent (1.2 ml)	For 40 transfections in 60 mm dishes or 160 transfections in 12-well plates	301305
SuperFect Transfection Reagent (4 x 1.2 ml)	For 160 transfections in 60 mm dishes or 640 transfections in 12-well plates	301307

For further information: www.qiagen.com/PG/TransfectionTools