

## Thermo Scientific DharmaFECT Transfection Reagents



Maintain cell viability with reagents specially formulated to deliver siRNA



Achieve robust siRNA uptake for dependable gene silencing



Expand your RNAi applications with an optimized co-transfection reagent



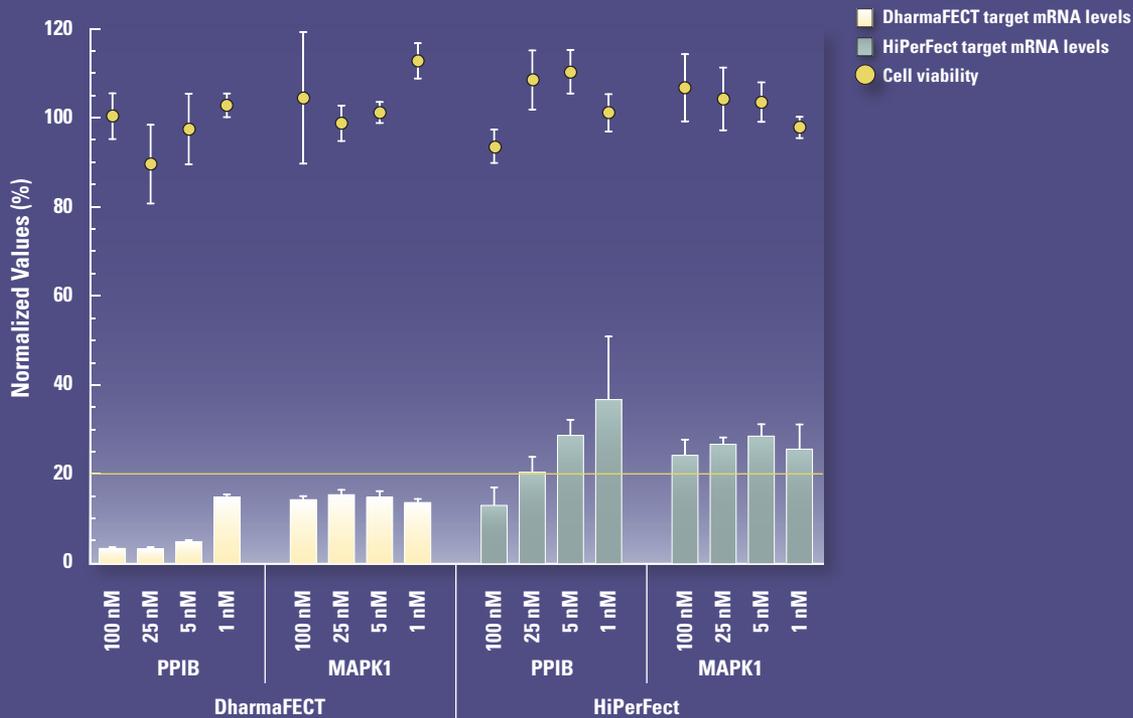
## Thermo Scientific DharmaFECT transfection reagents deliver

Efficient siRNA delivery is critical for successful gene silencing. Even highly potent siRNA may appear non-functional without effective delivery. DharmaFECT® transfection reagents provide efficient and reliable siRNA transfection at low siRNA concentrations with low cellular toxicity.

- Specialized formulations for successful delivery of siRNA to more cell lines
- Effective delivery for target silencing at low siRNA concentrations
- Low toxicity even at a broad range of experimental conditions

# efficient delivery

*DharmaFECT provides highly efficient delivery at low siRNA concentrations*



**DharmaFECT achieved >80% silencing at all siRNA concentrations, while HiPerFect (Qiagen) only achieved this level with PPIB at 100 nM.**

HeLa cells were transfected with SMARTpool™ reagents targeting Cyclophilin B (PPIB) or MAPK1 at concentrations of 1, 5, 25, and 100 nM.

mRNA expression (bars) was determined by branched DNA assay (Panomics Quantigene Reagent System) and cell viability (data points) was determined by alamarBlue (Biosource International).



## Multiple formulations ensure siRNA transfection success

No single transfection reagent effectively delivers siRNA to the hundreds of different cell lines being used in RNAi experiments. Why compromise delivery efficiency and cell viability with a "one-size-fits-all" transfection reagent?

DharmaFECT is the only transfection reagent offered in four distinct siRNA-specific formulations to ensure optimal siRNA delivery in a wide range of cell lines.

- No single lipid reagent outperformed the panel of DharmaFECT formulations in a benchmark study of twenty common cell lines
- Independent third-party optimization of siRNA transfection demonstrated DharmaFECT formulations outperforming four competitor lipid reagents in 9 of 10 cell lines<sup>1</sup>

# multiple formulations

### DharmaFECT formulations deliver siRNA effectively to more cell lines

#### Optimal lipid for siRNA transfection in a 384-well format<sup>1</sup>

Adapted from Borawski et al.

| Cell Line  | Lipid Reagent      |
|------------|--------------------|
| HeLa       | DharmaFECT 4       |
| SKOV3      | DharmaFECT 4       |
| A549       | DharmaFECT 2       |
| HCT116     | DharmaFECT 1       |
| Huh7       | DharmaFECT 3       |
| A673       | Lipofectamine 2000 |
| XMD5       | DharmaFECT 3       |
| HEK293     | DharmaFECT 1       |
| UMR        | DharmaFECT 1       |
| MDA-MB-231 | DharmaFECT 4       |

Lipids tested were DharmaFECT 1-4, HiPerFect (Diagen), TransIT-TKO (Mirus) Lipofectamine 2000 and Oligofectamine (Invitrogen).

<sup>1</sup> Borawski, J., et al. "Optimization Procedure for small interfering RNA Transfection in a 384-well format." *J. Biomolecular Screening* 12 (2007); 546-559.

| Cell Line | Successful Transfection |                     |         |         |         |
|-----------|-------------------------|---------------------|---------|---------|---------|
|           | DharmaFECT formulation  | Other Manufacturers |         |         |         |
|           |                         | Lipid A             | Lipid B | Lipid C | Lipid D |
| A549      | ✓                       |                     |         |         | ✓       |
| BxPC3     | ✓                       |                     |         | ✓       | ✓       |
| DU 145    | ✓                       | ✓                   | ✓       | ✓       | ✓       |
| HEK293    | ✓                       |                     |         |         | ✓       |
| HeLa      | ✓                       | ✓                   | ✓       | ✓       | ✓       |
| HeLa S3   | ✓                       |                     |         | ✓       | ✓       |
| Hep G2    | ✓                       |                     |         |         | ✓       |
| HT - 29   | ✓                       |                     | ✓       | ✓       | ✓       |
| LNCaP     | ✓                       |                     | ✓       |         |         |
| MCF7      | ✓                       |                     |         |         |         |
| PC - 3    | ✓                       |                     |         |         |         |
| H1299     | ✓                       | ✓                   |         | ✓       |         |
| H1080     | ✓                       | ✓                   |         | ✓       |         |
| CHO K1    | ✓                       |                     | ✓       |         | ✓       |
| A7R5      | ✓                       |                     |         |         |         |
| 3T3 L1    | ✓                       |                     |         |         | ✓       |
| NKR-49F   | ✓                       | ✓                   |         | ✓       | ✓       |
| RAT - 2   | ✓                       |                     |         |         |         |
| H9C2      | ✓                       | ✓                   |         |         |         |
| AR42J     | ✓                       |                     |         |         |         |
| COS 7     | ✓                       |                     |         |         |         |

DharmaFECT formulations deliver siRNA effectively to more cell lines than lipids from other manufacturers.

Optimization studies with DharmaFECT 1, 2, 3, 4 and four other lipid transfection reagents were assessed for effective siRNA delivery (>80% knockdown) and low toxicity (>80% viability). Checkmarks indicate successful transfection.



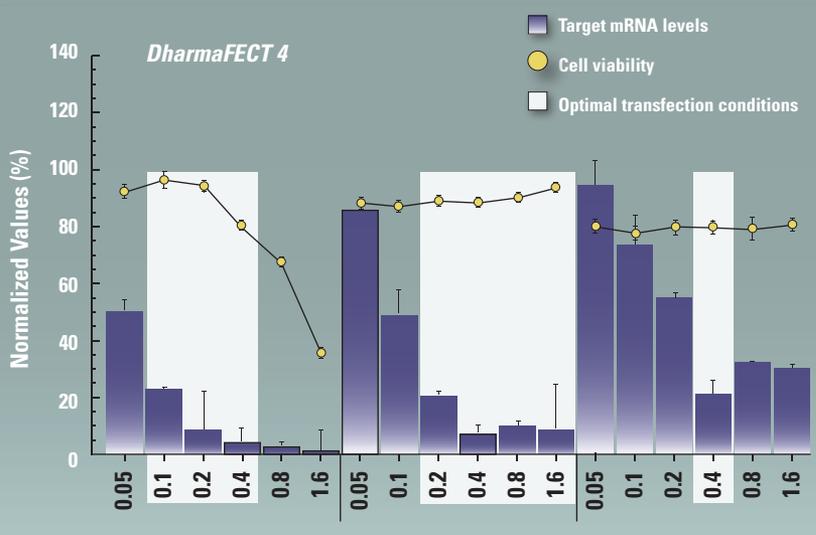
## Effective siRNA delivery for reliable silencing

Experimental reproducibility is essential to RNAi studies, yet poor siRNA delivery or cellular toxicity may confuse the interpretation of results.

DharmaFECT siRNA transfection reagents ensure effective delivery across a broad range of cell densities and lipid volumes with minimal effect on cell viability.

- Fast and easy optimization producing reliable and reproducible RNAi experiments
- A broad window of optimal conditions provides experimental flexibility
- Well-suited for high-throughput RNAi screening

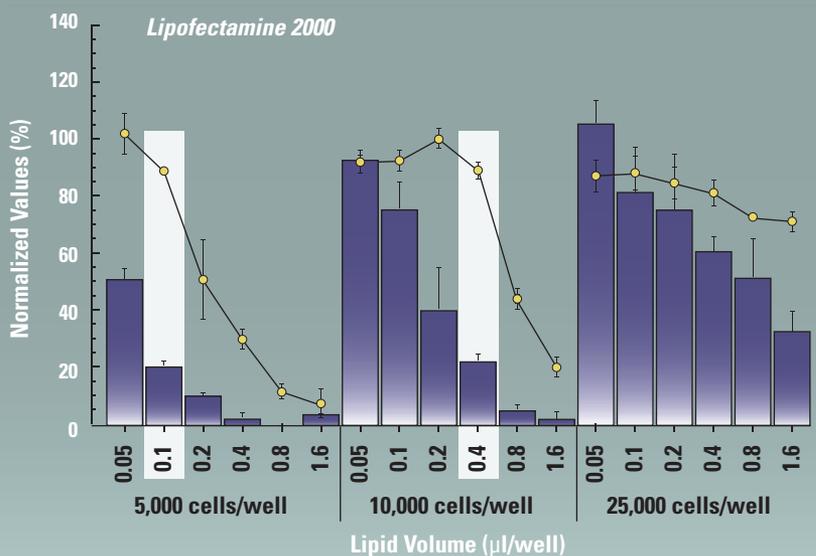
# reliable and reproducible



**DharmaFECT siRNA transfection reagents are effective across a wider range of conditions**

*DharmaFECT reagents are effective across a broader range of experimental conditions when compared to Lipofectamine 2000 (Invitrogen).*

*Several cell densities and lipid volumes were investigated to determine optimal transfection conditions, shown by the shaded boxes. Three cell densities of HepG2 cells were transfected with GAPD siRNA (100 nM) using a range of volumes (0.05 - 1.6 μl/well) of Lipofectamine 2000 and DharmaFECT 4 transfection reagents.*



*mRNA levels (bars) were assessed by branched DNA assay (Panomics Quantigene Reagent System) and cell viability (data points) was determined by alamarBlue (Biosource International).*

## DharmaFECT Duo reagent for RNAi molecules and plasmid delivery

Co-transfection of siRNA or miRNA reagents with a plasmid reporter system is increasingly common, yet few transfection reagents on the market will effectively deliver both molecules.

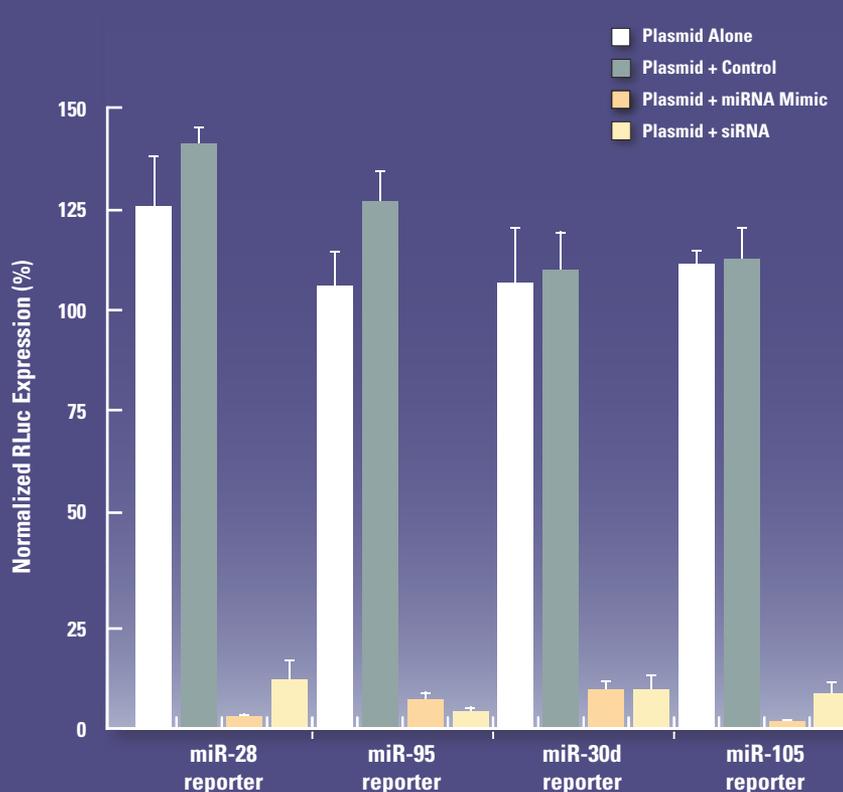
In co-transfection experiments optimized for cell viability, DharmaFECT Duo reagent resulted in both reliable plasmid expression and potent siRNA-mediated knockdown.

DharmaFECT Duo transfection reagent is specially formulated to provide:

- High co-transfection efficiency of plasmid and siRNA or miRNA reagents
- Efficient siRNA target knockdown plus plasmid expression
- Reliable delivery for reporter-based miRNA regulation experiments

# co-transfect with confidence

*DharmaFECT Duo is highly effective in co-transfection of plasmid with siRNA or miRNA mimics*



**DharmaFECT Duo transfection reagent achieves reliable co-transfection results for a variety of experimental goals. The graph demonstrates specific modulation of the Renilla luciferase (Rluc) gene:**

- Rluc expression is similar for Plasmid Alone and Plasmid + Control
- Down-regulation of Rluc as a result of miRNA mimic (plasmid + miRNA mimic)
- Greater than 85% knockdown of Rluc expression by siRNA (Plasmid + siRNA)

The psiCHECK-2 vector (100ng/well; Promega) with cloned microRNA recognition sites for miR-28, miR-95, miR-30d and miR-105, respectively, were complexed alone or with RNAi reagents (10 nM) using DharmaFECT Duo (0.2uL/well) in MCF-7 cells at 10,000 cells/well (96-well plate). The RNAi reagents were Thermo Scientific Dharmacon miRIDIAN Mimics (Negative Control, miR-28, miR-95, miR-30d and miR-105, respectively) or siRNA (Renilla luciferase pool). Firefly and Renilla luciferase expression was assessed at 48 hours using the Dual-Glo Luciferase Assay System (Promega) and normalized to identically treated psiCHECK-2 empty vector.



## Find the DharmaFECT formulation that's right for you

DharmaFECT 1 formulation is the most broadly-applicable lipid for effective siRNA delivery across cell lines. In a number of cases, another DharmaFECT formulation gave even better results – emphasizing the value of optimized reagents.

- Choose DharmaFECT formulation 1, 2, 3 or 4 for optimal siRNA transfection or DharmaFECT Duo reagent for effective plasmid and siRNA co-transfection
- Review validated recommendations to help you select the appropriate DharmaFECT formulation for your research (see table)

*Cell line-specific DharmaFECT formulation recommendations (96-well format)*

| Cell line     | Cell type                             | Recommended DharmaFECT formulation | DharmaFECT volume/well (μL) | Plating density/well | Additional Successful DharmaFECT formulations | Validated DharmaFECT Duo protocol available |
|---------------|---------------------------------------|------------------------------------|-----------------------------|----------------------|---|---|
| <b>Human</b>  |                                       |                                    |                             |                      |   |   |
| A549          | Lung carcinoma                        | 1                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 2, 3, 4                                       |   |
| BxPC3         | Pancreas adenocarcinoma               | 2                                  | 0.2                         | 5 x 10 <sup>3</sup>  | 1, 3, 4                                       |   |
| DU 145        | Prostate carcinoma                    | 1                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 2, 3, 4                                       |   |
| HEK293        | Kidney transformed embryonic cells    | 1                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 2, 4  |   |
| HeLa          | Cervical epithelial adenocarcinoma    | 1                                  | 0.2                         | 5 x 10 <sup>3</sup>  | 2, 3, 4                                       | X   |
| HeLa S3       | Cervical epithelial adenocarcinoma    | 4                                  | 0.4                         | 5 x 10 <sup>3</sup>  | 1, 2, 3                                       |   |
| Hep G2        | Hepatocellular carcinoma              | 4                                  | 0.4                         | 1 x 10 <sup>4</sup>  | 1, 2  | X   |
| H1299         | Lung carcinoma                        | 2                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 4   |   |
| HT-1080       | Fibrosarcoma                          | 4                                  | 0.2                         | 5 x 10 <sup>3</sup>  | 1, 2, 3                                       |   |
| HT-29         | Colorectal carcinoma                  | 1                                  | 0.2                         | 5 x 10 <sup>3</sup>  | 2, 3, 4                                       |   |
| MCF-7         | Breast adenocarcinoma                 | 1                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 2, 4  | X   |
| MCF-10a       | Breast adenocarcinoma                 | 1                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 2   | X   |
| MDA-MB-453    | Breast adenocarcinoma                 | 2                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 1, 3, 4                                       |   |
| hMSC          | Mesenchymal stem cells                | 1                                  | 0.4                         | 5 x 10 <sup>3</sup>  | 2, 3, 4                                       |   |
| PC3           | Prostate carcinoma                    | 2                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 3   |   |
| SK-BR3        | Breast adenocarcinoma                 | 2                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 1, 3, 4                                       |   |
| 786-O         | Kidney adenocarcinoma                 | 1                                  | 0.4                         | 5 x 10 <sup>3</sup>  | 2   |   |
| HCT-116       | Colorectal carcinoma                  | 2                                  | 0.1                         | 5 x 10 <sup>3</sup>  | 4   |   |
| MDA-MB-231    | Breast adenocarcinoma                 | 4                                  | 0.1                         | 5 x 10 <sup>3</sup>  | 1   |   |
| MDA-MB-157    | Breast epithelial medullary carcinoma | 1                                  | 0.5                         | 5 x 10 <sup>3</sup>  | 4   |   |
| Huh7          | Hepatoma                              | 4                                  | 0.05                        | 5 x 10 <sup>3</sup>  | 1, 2  |   |
| SK-OV-3       | Ovarian adenocarcinoma                | 3                                  | 0.4                         | 1 x 10 <sup>4</sup>  | 1, 2, 4                                       |   |
| DLD-1         | Colorectal adenocarcinoma             | 2                                  | 0.4                         | 5 x 10 <sup>3</sup>  | 1, 3  |   |
| OVCAR 3       | Ovarian adenocarcinoma                | 1                                  | 0.1                         | 5 x 10 <sup>3</sup>  | 2, 3, 4                                       |   |
| HUVEC         | Human umbilical vein endothelial      | 4                                  | 0.2                         | 2 x 10 <sup>4</sup>  | 1, 2  |   |
| U-87 MG       | Brain glioblastoma                    | 1                                  | 0.1                         | 5 x 10 <sup>3</sup>  | 2, 3, 4                                       |   |
| JEG-3         | Placenta, choriocarcinoma, epithelial | 3                                  | 0.2                         | 1 x 10 <sup>4</sup>  | -   |   |
| LNCaP         | Prostate carcinoma                    | 2                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 1, 3  |   |
| ARPE19        | Retinal pigment epithelial            | 4                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 1, 2  |   |
| Saos-2        | Bone, osteosarcoma, epithelial        | 1                                  | 0.05                        | 1 x 10 <sup>4</sup>  | 2, 3, 4                                       |   |
| <b>Rodent</b> |                                       |                                    |                             |                      |   |   |
| A7R5          | Rat aortic smooth muscle              | 2                                  | 0.1                         | 5 x 10 <sup>3</sup>  | 1   |   |
| C2C12         | Mouse myoblasts                       | 1                                  | 0.2                         | 5 x 10 <sup>3</sup>  | 2, 3, 4                                       |   |
| CHO K1        | Chinese hamster ovary                 | 1                                  | 0.4                         | 1 x 10 <sup>4</sup>  | 2   |   |
| ES - D3       | Mouse embryonic stem cells            | 1                                  | 0.2                         | 2 x 10 <sup>3</sup>  | 2   | X   |
| ES - E14TG2a  | Mouse embryonic stem cells            | 1                                  | 0.2                         | 2 x 10 <sup>3</sup>  | 2   |   |
| H9C2          | Rat heart myoblasts                   | 1                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 2, 3, 4                                       | X   |
| J774          | Mouse macrophage                      | 4                                  | 0.2                         | 1 x 10 <sup>4</sup>  | -   |   |
| NIH / 3T3     | Mouse embryonic fibroblast            | 1                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 3   | X   |
| NRK - 49F     | Rat kidney fibroblast                 | 2                                  | 0.2                         | 1 x 10 <sup>4</sup>  | 1, 4  |   |
| RAT2          | Rat fibroblast                        | 1                                  | 0.2                         | 2 x 10 <sup>4</sup>  | 2   |   |
| 3T3 L1        | Mouse embryonic fibroblast            | 1                                  | 0.2                         | 5 x 10 <sup>3</sup>  | 3   |   |
| <b>Other</b>  |                                       |                                    |                             |                      |   |   |
| COS 7         | African green monkey kidney           | 2                                  | 0.4                         | 5 x 10 <sup>3</sup>  | 1, 3, 4                                       |   |

*All conditions listed resulted in control gene silencing of 85 % or better and viability of 80 % or better in a 96-well format. X indicates cell lines where DharmaFECT Duo transfection reagent successfully delivered plasmid and siRNA with greater than 70 % viability. Conditions should always be re-evaluated in the context of assay-specific requirements for cell density.*



## Ordering DharmaFECT transfection reagents

Our online resources for over 100 cell lines will help you determine the right DharmaFECT formulation for your research.

- Optimized, validated protocols available for over 25 cell lines
- Customer-provided recommendations for the optimal DharmaFECT formulation in over 80 cell lines
- DharmaFECT Set of 4 (one tube of each formulation) available for optimization of novel cell types

easy online ordering

### DharmaFECT products

| Product                   | Size                                   | Catalog #. |           |
|---------------------------|--|------------|-----------|
| <b>siRNA Transfection</b> |  |            |           |
| DharmaFECT 1              | 0.2 mL                                 | T-2001-01  |           |
|                           | 0.75 mL                                | T-2001-02  |           |
|                           | 1.5 mL                                 | T-2001-03  |           |
|                           | 5 x 1.5 mL                             | T-2001-04  |           |
|                           | 2 x 10 mL                              | T-2001-07A |           |
| DharmaFECT 2              | 0.2 mL                                 | T-2002-01  |           |
|                           | 0.75 mL                                | T-2002-02  |           |
|                           | 1.5 mL                                 | T-2002-03  |           |
|                           | 5 x 1.5 mL                             | T-2002-04  |           |
| DharmaFECT 3              | 2 x 10 mL                              | T-2002-07A |           |
|                           | 0.2 mL                                 | T-2003-01  |           |
|                           | 0.2 mL                                 | T-2003-02  |           |
|                           | 0.2 mL                                 | T-2003-03  |           |
|                           | 5 x 1.5 mL                             | T-2003-04  |           |
| DharmaFECT 4              | 2 x 10 mL                              | T-2003-07A |           |
|                           | 0.2 mL                                 | T-2004-01  |           |
|                           | 0.2 mL                                 | T-2004-02  |           |
|                           | 0.2 mL                                 | T-2004-03  |           |
|                           | 5 x 1.5 mL                             | T-2004-04  |           |
| DharmaFECT Set of 4       | 2 x 10 mL                              | T-2004-07A |           |
|                           | 0.2 mL x 4                             | T-2005-01  |           |
|                           | 0.75 mL x 4                            | T-2005-02  |           |
| DharmaFECT Set of 4       | 1.5 mL x 4                             | T-2005-03  |           |
|                           | <b>siRNA / plasmid co-transfection</b> |            |           |
|                           | DharmaFECT Duo                         | 0.2 mL     | T-2010-01 |
| 0.75 mL                   |  | T-2010-02  |           |
| 1.5 mL                    |  | T-2010-03  |           |
| 5 x 1.5 mL                |  | T-2010-04  |           |

### General ordering guidelines

| Plate type | Package size (ml) | Approximate number of transfections per package |
|------------|-------------------|---|
| 96 well    | 0.02              | 200 - 1000                                      |
|            | 0.75              | 750 - 3750                                      |
|            | 1.50              | 1500 - 7500                                     |
| 24 well    | 0.02              | 200   |
|            | 0.75              | 750   |
|            | 1.50              | 1500  |
| 12 well    | 0.20              | 100   |
|            | 0.75              | 375   |
|            | 1.50              | 750   |
| 6 well     | 0.20              | 50  |
|            | 0.75              | 187   |
|            | 1.50              | 375   |

complete, validated protocols available online at  
[www.thermo.com/dharmaconprotocols](http://www.thermo.com/dharmaconprotocols)

# Thermo Scientific DharmaFECT Transfection Reagents



## Contact Information

### North America

2650 Crescent Dr., Suite 100  
Lafayette, CO 80026  
Toll free: 800 235 9880  
Tel: 303 604 9499  
Fax: 303 604 3286

US web: [www.thermo.com/dharmacon](http://www.thermo.com/dharmacon)  
US e-mail: [dharmacon.lab@thermofisher.com](mailto:dharmacon.lab@thermofisher.com)

### Other Countries:

Technical support: 00800 73724648

### Belgium

Tel: 0800 80543  
Fax: 0800 80178  
Technical support: 053 85 71 92

### France

Tel: 08009 14294  
Fax: 08009 11885  
Technical support: 0800 50 84 97

### Germany

Tel: 0800 1830746  
Fax: 0800 1810366  
Technical support: 0228 2427409

### The Netherlands

Tel: 08000 223464  
Fax: 08000 223291  
Technical support: 076 50 31 880

### United Kingdom

Tel: 08009 171501  
Fax: 01372 840545  
Technical support: 0800 252 185

### Switzerland

Tel: 08005 63505  
Fax: 08008 38669  
Technical support: 0800 56 31 40

US Literature # 00034-04-L-04-U

Euro Literature # PB 2008 14

© 2009 Thermo Fisher Scientific Inc. All rights reserved. QuantiGene is a trademark of Bayer. alamarBlue is a trademark of Accumed International, Inc. HiPerfect is a trademark of QIAGEN Inc. Lipofectamine and Oligofectamine are trademarks of Invitrogen Corporation. TransIT-TKO and TransIT are trademarks of Mirus Bio Corporation. FuGENE is a trademark of Fugent, L.L.C. siPORT is a trademark of Ambion, Inc. psiCHECK and Dual-Glo is a trademark of Promega. All other trademarks are the property of Thermo Fisher Scientific Inc. and its subsidiaries.

**Thermo**  
SCIENTIFIC