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Targeting Systems Introduces :

Targefect-293

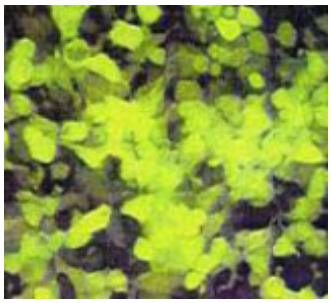
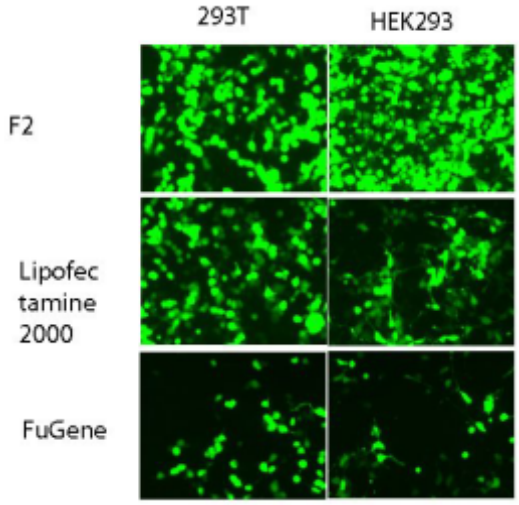


Figure 1 : HEK 293 cells transfected with Targefect-293
Transfection efficiency was approximately 95% (Data courtesy of Dr. Peter Ordentlich, Dr. Ronald M Evans' lab, Howard Hughes Medical Institute, Gene Expression Laboratory, The Salk Institute of Biological Studies, La Jolla, CA)



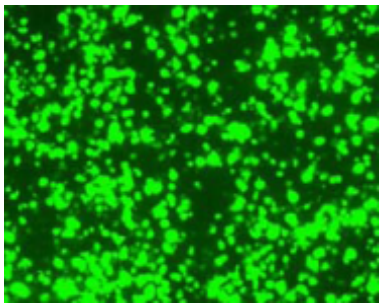


Figure 2 : A comparison of transfection efficiency of Targefect- 293 with lipofectamine 2000 and Fugene 6.
LowerPanel : Transfection of BAC DNA (bacterial artificial chromosomes) into HEK-293 cells using Targefect -293 Data Courtesy of Dr Fuchun Zhou and Dr S. Gao, Univ. of Texas Health Sc Ctr at San Antonio, TX.

Features

- Targefect-293 Transfection Reagent is specifically developed to achieve the highest transfection efficiency in HEK 293 cells.
- This reag ent efficiently transfects HEK-293 cells efficiently with low toxicity.
- Achieve higher transfection efficiencies and save time by transfecting in the presence of serum and eliminating media changes.
- The reagents are supplied with an optimized protocol, and a ready-to-use single vial.
- Targefect-293 is quality control tested on a standard ATCC HEK 293 cell line.

Description

Targefect-293 Transfection Reagent is a modification of the Targefect reagents to achieve the highest transfection efficiency in HEK 293 cells. The levels of transgene expression obtained using this reagent are much higher than those obtained with the Targefect F-1 reagent cited in the publication list shown below. Transfections using the Targefect-293 reagent can be performed in the presence of serumcontaining media, with no media change.

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Transfection Protocol

Set up cells to be transfected so that they are about 70-80% confluent at the time of the experiment.

Prepare transfection complexes as follows :

Note : It is important to use high glucose DMEM (DMEM containing 4500 mg/liter glucose for complex formation D o not vortex the Targefect-293 reagent. Thaw the reagent if it arrives frozen and gently mix it by inverting the tube several times and then store at 4º C. Do not freeze this reagent.

Tube #	DMEM	Plasmid DNA	Targefect-293
1	0.6 ml	6 µg	12 µl
2	0.6 ml	6 ug	18 ul
3	0.6 ml	10 ug	12 ul

Note : Condition 3 is recommended for large DNA fragments/plasmids.

Add DMEM 1 first, then add DNA, mix well by flicking the tube about 12 times to create a vortexing action.
Add Targefect-293 next, mix well again by flicking the tube. Incubate the tubes at 37º C for 20 minutes to form the transfection complexes. Add 250 µl of transfection complex to 2 ml of completemedia per well of a 6-well dish. Swirl the dish to mix transfection complexes with complete media and incubate at 37º C overnight. Assay at 24-48 hrs after transfection
Recommended volumes of transfection complex for performing transfection in different size dishes:

Culture Vessel	Volume of plating medium	DNA (µg) in transfection complex volume (µl)	Targefect-293 in transfection complex volume (µl)
96well	100 µl	0.2 µg in 25 µl	0.5 µl in 25 µl
24well	500 µl	0.8 µg in 50 µl	1.5 µl in 50 µl
12well	1 ml	1.6 µg in 150 µl	3.5 µl in 150 µl
35mm	2 ml	3.0 µg in 250 µ	6 µl in 250 µl
6well	2 ml	3.0µg in 250 µl	6 µl in250 µl
60mm	5 ml	6.0 µg in 0.5 ml	12 µl in 0.5ml
10cm	15 ml	18 µg in 1.5 ml	36 µl in 1.5 ml

Note: The above conditions are standardized using media with 5% fetal bovine serum.

Product No.	Quantity	T.S. list Price
293-01	1ml	$170

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Selected publications using Targefect for transfecting HEK293 cells

1) Downes M, Ordentlich P, Kao H-Y, Jacqueline GA Alvarez and Evans RM. (2000) Identification of a nuclear domain with deacetylase activity. Proc. Natl.. Acad. Sci., USA, 97 (19): 10330-10335

2) Kao H-Y, Downes M, Ordentlich P and Evans RM (2000) Isolation of a novel histone deacetylase reveals that class 1 and class II deacetylases promote SMRT-mediated repressio. Genes and Development. 14: 55-66

3) Radu V. Stan, Eugene Tkachenko, and Ingrid R. Niesman PV1 Is a Key Structural Component for the Formation of the Stomatal and Fenestral Diaphragms Mol. Biol. Cell, Aug 2004; 15: 3615 - 3630.

4) Marc Tini, Arndt Benecke, Soo-Joong Um, Joseph Torchia, Ronald M. Evans, and Pierre Chambon (2002) Association of CBP/p300 Acetylase and Thymine DNA Glycosylase Links DNA Repair and Transcription. Molecular Cell 9: 265-277.

5) Hung-Ying Kao, André Verdel, Chih-Cheng Tsai, Cynthia Simon, Henry Juguilon, and Saadi Khochbin (2001) Mechanism for Nucleocytoplasmic Shuttling of Histone Deacetylase . J. Biol. Chem., Dec 2001; 276: 47496 -47507.

6) Yanhong Shi, Michelle Hon, and Ronald M. Evans (2002) The peroxisome proliferator-activated receptor , an integrator of transcriptional repression and nuclear receptor signaling. PNAS , 99:2613-2618.

7) Hung-Ying Kao, Chih-Hao Lee, Andrei Komarov, Chris C. Han, and Ronald M. Evans (2001) Isolation and Characterization of Mammalian HDAC10, a Novel Histone Deacetylase. J. Biol. Chem., Dec 2001; 277: 187 - 193.

8) Yanhong Shi, Michael Downes, Wen Xie, Hung-Ying Kao2, Peter Ordentlich,1 Chih-Cheng Tsai, Michelle Hon, and Ronald M. Evans Sharp, an inducible cofactor that integrates nuclear receptor repression and activation. Genes and Development Vol. 15, No. 9, pp. 1140-1151, May 1, 2001

9) Radu V. Stan. Multiple PV1 dimers reside in the same stomatal or fenestral diaphragm Am J Physiol Heart Circ Physiol, Apr. 2004; 286: 1347 - 1353.