targetingsystems

1453 N. Cuyamaca St, El Cajon, CA 92020
Phone: (866)-620 4018, (619)-562-1518
Fax: (619)-562-1326 • info@targetingsystems.net
www.targetingsystems.com

Transfection Protocl For Transfecting Cells In The Absence Of Serum (Old Protocol)

Transfection of cells in absence of serum

Cells incubated with transfection complexes in the absence of serum. Please vortex F-1 reagent at full speed for 30 seconds once or twice just before use. Do not vortex the f-2 reagent or Virofect. Conditions recommended most are in bold.

Tube #	DMEM(high glucose)	DNA	Targefect	Enhancer Reagent
1	1 ml	1.5 µg	2 µl F-2	-
2	**1 ml**	**2 µg**	**4 µl F-2**	
3	1 ml	2 µg	8 µl F-2	
4	**1 ml**	**2 µg**	**2.5 µl F-1**	
5	**1 ml**	**2 µg**	**5 µl F-1**	
4	**1 ml**	**5 µg**	**5 µl F-1**	**15 µl Peptide enhancer**
5	**1 ml**	**2 µg**	**5 µl F-1**	**15 µl Virofect**
6	**1 ml**	**2 µg**	**6 µl F-2**	**15 µl Virofect**

Note: Conditions 4, 5, 6 are for transfecting cells with Targefect plus enhancer reagents. Contact our tech support for details.

Add DMEM first, mix well, then add targefect, mix well, Add enhancer last and mix well again. Incubate at 37 °C to form transfection complexes.
Add 1ml of transfection complex per well of a 6-well dish, Add 0.3 ml of transfection complex per well of a 12-well dish or 0.15 ml of transfection complex per well of a 24 well dish (make sure cells are covered well or else add 0.2 ml complex per well. Incubate cells with transfection complexes at 37 °C for 3 hrs. Add complete media with serum, add 0.5 ml complete media per well of a 24 well dish, 1 ml per well of a 12-well dish, 2 ml per well of a 6-well dish. Mix gently and incubate overnight. Replace media the next day. Assay at 30-48hrs post transfection. We recommend using media 199 with 10-20% serum as the complete media.

Note: The peptide enhancer of Vriofect is recommended for enhancement of gene transfer into certain difficult to transfect cell types. For details contact tech support at 1-866-620-4018, 1-619-562-1518 or email us at targsys@aol.com

targetingsystems

1453 N. Cuyamaca St, El Cajon, CA 92020
Phone: (866)-620 4018, (619)-562-1518
Fax: (619)-562-1326 • info@targetingsystems.net
www.targetingsystems.com

General Transfection Protocol (Preferred Protocol)

Transfection of cells with Targefect F-1 or F-2 in presence of serum (highly recommended protocol)

In this protocol the cells incubated with transfection complexes in presence of serum. **Complexes between DNA and targefect are still formed in DMEM or OptiMEM1 without serum**.

Tube #	DMEM(high glucose)	DNA	Targefect	Enhancer Reagent
1	0.5 ml	6 µg	12 µl F-2	
2	0.5 ml	6 µg	12 µl F-2	25 µl Virofect
3	0.5 ml	6 µg	12 µl F-1	
4	0.5 ml	6 µg	12 µl F-1	25 µl Virofect
5	0.5 ml	10 µg	20 µl F-1	
6	0.5 ml	10 µg	20 µl F-2	

Note: Use of Virofect enhancer is recommended for difficult to transfect cell-types which have adenoviral receptors on the cell surface.

Add DMEM first, mix well, then add targefect, mix well, add enhancer last and mix well again. Incubate at 37 °C to form transfection complexes. Add 70 µl of transfection complex to 0.25 ml of complete media per well of a 24 well dish

Add 50 µl of transfection complex to 0.5 ml of complete media per well of a 24-well dish.
Add 125 µl of transfection complex to 1 ml of complete media per well of a 12 well dish.
Add 250 µl of transfection complex to 2ml of complete media per well of a 6-well dish.

Swirl the dish to mix transfection complexes with complete media. Assay at 30-48 hrs post transfection

NOTE: We recommend adding serum to cell culture media (final concentration of 5-10% FCS). If the cells require growth in serum free media you can keep the cells in cell culture media with 10% FCS for only a 3 hr time period after addition of transfection complexes and then replace with complete media without serum.