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PrimeFect™ Transfection Reagent for CHO Cells

Cat. #: M0003-CC Size: 1 ml

Introduction

PrimeFect™ Transfection Reagent for CHO Cells is a biodegradable polymer based transfection reagent that forms a complex with DNA, and transports the complex into a variety of adherent and suspension cell lines. A remarkable feature of the reagent is the rapid and complete degradation of polymer after transfection complex endocytosis, leading to much less cytotoxicity.

Package Information

Components	M0003-CC
PrimeFect™ Transfection Reagent for CHO Cells	1 ml

Storage

At Room temperature

Protocol

1. Cell seeding

On the day of cell seeding, adjust cell density according to your process to reach the exponential growth phase with a viable cell density (VCD) around 2- 2.5× 10^6 cells/ml at the time of transfection.

Cell seeding has to be adjusted according to the culture vessel you are using:

• Shake flask: we recommend seeding 1× 10^6 cells/ml to ensure that cells reach the optimal VCD after 24 hours;

• Other (e.g. culture bags, or bioreactors): the day of transfection will depend on the growth time needed for your cells to reach a VCD of 2-2.5 × 10^6 cells/ml.

2. Preparation of the complexes

The following protocol is given for the transfection of DNA coding for a protein of interest into CHO cells grown in suspension.

The recommended complexation parameters are described in Table 1.

Table 1. Complexation parameters for the transfection of suspension cells.

Parameter	Recommended condition	Range of optimization
DNA amount (per 10^6 cells)	1 µg DNA	0.5 µg–2 µg
Ratio (µg DNA : µl PrimeFect™)	1:2	1:1–1:6
Complexation time	15 min	10 min–20 min

3. Transfection

Follow the procedure below to transfect suspension CHO cells in a 30 ml of cell culture medium. If you wish to transfect the suspension cells in a larger volume, scale up the transfection conditions in proportion to the culture volume.

3.1 On the day of transfection, measure the cell density and determine transfection parameters (DNA amount and PrimeFect™ volume per million cells) according to Table 1. For this example, we assume a VCD of 2× 10^6 cells/ml.

3.2 Dilute 60 µg of DNA in 1.5 ml of serum free DMEM. Mix gently.

3.3 Mix PrimeFect™ gently before use, then dilute 120 µl of PrimeFect™ in 1.5 ml of serum free DMEM. Mix gently.

3.4 Add the 1.5 ml PrimeFect™ solution onto the 1.5 ml DNA solution all at once. Mix immediately the solution, either by briefly vortexing it or inverting the tube few times.

3.5 Incubate the complexes at room temperature for 15 minutes.

3.6 Add the 3 ml PrimeFect™/DNA mix to the cells. Mix gently by rocking the flask back and forth.

3.7 Incubate cells at appropriate temperature, shaking and CO_2 levels (e.g. 37°C, 125 rpm, 8%) and harvest protein when required.

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