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PEI Transfection Reagent

1 Contents

Components	HY-K2014-1 mL	HY-K2014-5 mL	HY-K2014-10 mL
PEI Transfection Reagent	1 mL	1 mL × 5	1 mL × 10

2 Introduction

PEI is a commonly used polymer carrier in gene transfection. It consists of cationic polymers and can introduce nucleic acids into eukaryotic cells. In polymer-based transfection, exogenous DNA forms complexes with cationic polymers that enter host cells by endocytosis.

MCE PEI Transfection Reagent is based on 25 kDa PEI, it is modified by introducing functional genes to enhance the binding ability of DNA, and reduced the cytotoxicity. In addition, the cell uptake and the endosome escape functions of PEI modified was enhanced significantly.

MCE PEI Transfection Reagent is high-efficiency, low-toxicity, strong-stability, and suitable for many cell types, such as HEK-293、HEK-293T、CHO-K1、COS-1、COS-7、NIH/3T3、Sf9、HepG2 and HeLa et, even some hard-to-transfect cells. It can also be applied to large-scale recombinant protein expression and virus production.

3 Flow Diagram of Experiment

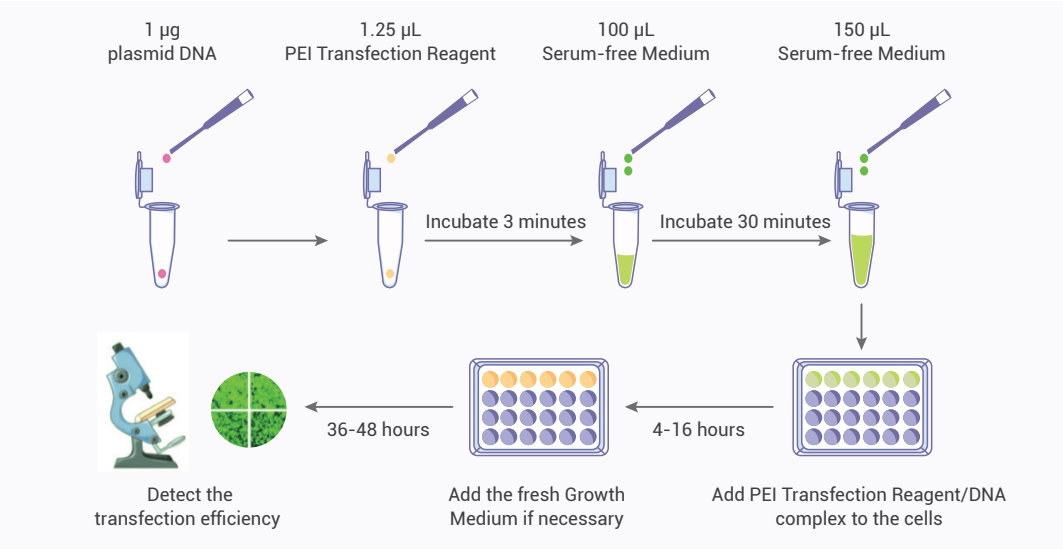

Figure 1. The process of the PEI Transfection Reagent

4 General Protocol

This is the protocol for transfection in the 24-well plate, and the adding volume of other culture devices is shown in Table 1.

1. Prepare cells

1.1 Adherent cells: Plate the cells digested with trypsin one day before transfection (0.3-2 × 10^5 cells plated in a 24-well cluster plate), until the density reaches 60-70% for cell transfection.

1.2 Suspension cells: Plate the cells before transfection and suspend in fresh medium (2-5 × 10^5 cells/500 μL medium).

Note: The viability and general health of cells prior to transfection significantly affect the transfection result. Cells should be at least 90% viable prior to transfection and have had sufficient time to recover from passaging.

2. Prepare PEI Transfection Reagent/ DNA complex

2.1 Add 1 μg plasmid DNA to a 1.5 mL EP tube, and add 1.25 μL PEI Transfection Reagent, incubate for 3 minutes at room temperature.

Note: a) Generally, the ratio of DNA (μg) to PEI Transfection Reagent (μL) is 1:1.25, and the transfection efficiency can be optimized in the range of 1:1 to 1:2 if necessary.

b) Cell type and culture conditions can affect the transfection efficiency. Perform pre-experiment to find out the optimal transfection ratio.

2.2 Add 100 μL Serum-free Medium and mix gently. Incubate for 30 minutes at room temperature.

Note: The complex can be remained stable for 4 hours at room temperature.

2.3 Dilute the complex with 150 μL Serum-free Medium and mix gently.

Table 1 PEI Transfection Reagent: DNA Ratio.

Plate Size	Growth Medium (mL)	DNA (μg)	PEI Transfection Reagent (μL)	Serum-free Medium 1 (μL)	
				For the step of 2.2	For the step of 2.3
96-well	0.1	0.2	0.25	20	30
24-well	0.5	1	1.25	100	150
12-well	1	2	2.5	200	300
6-well	2	2-4	2.5-5	400	600
60 mm	4	3-5	3.75-6.25	800	1200
100 mm	10	5-10	6.25-12.5	2000	3000
125 mL	30-35	30-35	37.5-43.75	6000	9000
500 mL	120-140	120-140	150-175	24000	36000
1000 mL	240-280	240-280	300-350	48000	72000

3. Add the PEI Transfection Reagent/DNA complex to the cells

Remove the medium, add the PEI Transfection Reagent/DNA complex (250 μL) to the cells in the 24-well plate and mix gently. Incubate for 36-48 hours In the incubator. Add the fresh Growth Medium (500/μL per well) after 4-16 hours if necessary.

4. Detection

Detect the transfection efficiency by Western Blot, ELISA, RT-PCR, FCM (Flow Cytometer), reporter gene and other methods, or screening stable cell lines with the addition of screening drugs.

5 Storage

-20°C，1 year.

Avoid repetitive freeze-thaw cycles.

6 Precautions

1. The viability and general health of cells prior to transfection significantly affect the transfection result. Cells should be at least 90% viable prior to transfection and have had sufficient time to recover from passaging.

2. Take high-purity, sterile, contaminant-free nucleic acids for transfection. Endotoxin in plasmids can lead to a significant decrease in transfection efficiency. It is recommended to use MCE Plasmid DNA Maxi Purification Kit (Cat. No.: HY-K1083).

3. Antibiotics may lead to cytotoxicity and reduce transfection efficiency, it's not recommended to add antibiotics.

4. Many factors will affect the transfection efficiency, such as cell type, cell state and density, nucleic acid quality and concentration, and the ratio of transfection reagent to nucleic acid, etc. Perform pre-experiment to find out the best transfection conditions.

5. To improve transfection efficiency, Serum-free Medium is recommended.

6. This product is for R&D use only, not for drug, household, or other uses.

7. For your safety and health, please wear a lab coat and disposable gloves to operate.