Polyethylenimine Linear(PEI), MW40000

Product description

Linearized polyethyleneimine PEI (molecular weight 40,000) is a highly charged cationic polymer that readily binds to DNA or other negatively charged biomacromolecules, making it a common and effective cell transfection reagent. In principle, PEI condenses DNA plasmid into positively charged complexes. The complexes can adhere to negatively charged cell surface residues, and then enters the cell through endocytosis. After entering the cell, protonation of the amine leads to an influx of counterions and a decrease in the osmotic potential, causing osmotic swelling and vesicle-releasing complexes into the cytoplasm. Finally, the complex is disassembled, the DNA freely fuse into the nucleus.

As a transient transfection reagent, PEI has the advantages of high efficiency, low cost and relative stability, etc., which has been validated for a wide range of common cell lines including HEK-293, HEK293T, CHO-K1, HepG2 and Hela cell transfection. In HEK293 and CHO cell expression systems, PEI provides excellent transfection results at different sizes (from 96-well plates to 100 L bioreactors).

Composition and storage conditions

Components \ Size	4 mL (2.5 mg/mL)	8 mL (2.5 mg/mL)
PEI-2.5 mg/mL	4 mL	8 mL
2X Transfection Buffer	3 mL	6 mL
For long-term storage, store at -20°C. For frequent use, it can be placed at 4°C to avoid repeated freeze-thaw cycles.		

Experimental operation

Taking the 6-well plate as an example, if the transfection vessel is different, please adjust the dosage proportionally.

1. Cell seeding

   The cell is counted, seeded ((7-8)×10^5 cells each well is appropriate), and cultured in a 5% CO_2 incubator at 37°C for 18-24 h. Transfection is performed at 70-90% cell density.

2. Preparation of DNA-PEI complexes

   1) Dilute 2X Transfection Buffer to 1X with ddH_2O, then dilute 2.5 mg/mL PEI to 1 mg/mL by using 1X Transfection Buffer.

   2) Serum-free culture medium is used to prepare 62.5 μL A and B working solutions respectively,

according to the ratio of DNA plasmid (μg): PEI (μL)=3:8. Taking 3 μg plasmid transfection for example, 3 μg plasmid is added to culture medium for preparing 62.5 μL A solution, and 8 μL PEI (1 mg/mL) is also added to culture medium for preparing 62.5 μL B solution. Note that the amount of DNA plasmid is controlled at 1-5 μg per well.

3) Solution A and B stand for 5 minutes at room temperature, then solution B is added to solution A (note not to reverse the order of addition), mixing gently and standing for 15 minutes to form DNA-PEI transfection complex.

3. Cell Transfection

1) 30-60 min before transfection, replace 2 mL fresh culture medium, then add the above DNA-PEI complex to each well. Gently shake the plate to distribute the transfection solution.

2) Continue the culture for 24 h, the transfected gene expression should be detectable usually.

4. Stable transfection screening (optional)

1) 24 h after transfection, cell is subcultured into fresh culture medium (cell is diluted more than 10 times) and then incubated overnight at 37 °C in a 5% CO_2 incubator.

2) The next day, add screening drug that match transfection resistance genes into medium. Resistant clones can be screened about 1 to 2 weeks, during which time the medium containing screening drug should be changed frequently.

Notes

1. For certain cell types, such as HEK-293, NIH/3T3 and COS cell, seeding two days before transfection significantly increases the expression level of the transferred gene. If this is done, the cell can be reduced appropriately to ensure that the cell density at transfection is still 70-90%.
2. DNA-PEI complexe must be formed in the absence of protein, so DNA and PEI must be diluted with serum-free reagent. Opti-MEM medium is recommended for optimal transfection efficiency.
3. This kit can achieve high transfection efficiency for most cells according to the ratio of DNA and PEI=3:8. User can also adjust according to the situation.
4. This product is for scientific use only.

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