

#### Applied Biological Materials Inc

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# DNAfectin™ Plus

Store at 4°C

Cat. No.	Description	Quantity
G2500	DNAfectin™Plus	1.0ml

# Description

**abm**'s DNAfectin<sup>™</sup> Plus is a nanoparticle-based, nonliposomal formulation that enables the efficient transfection of plasmid DNA and short oligonucleotides into a broad range of cells with minimal cytotoxicity. This simple protocol does not require the removal of serum or culture medium, resulting in less variability and low risk of contamination. DNAfectin<sup>™</sup> Plus has been shown to transfect a wide variety of primary, adherent and suspension cell lines with high efficiency.

## **Transfection Protocol**

Use the following conditions as guidelines to transfect mammalian cells in a 6-well or 35mm dish format. For other culture vessels, please refer to Table 1.

- Plating Cells: 18 to 24 hours prior to transfection, seed the cells at a density of 1-3 x 10<sup>5</sup> cells per well in 2.0ml of appropriate growth medium (complete with serum and antibiotics if normally used). Incubate the cells at 37°C in a CO<sub>2</sub> incubator until the cells are 70% to 90% confluent at the time of transfection.
- For each transfection sample, prepare the DNAfectin<sup>™</sup> Plus-DNA complexes as follows:
  - a) Add 2.0µg of DNA into 200µl of serum-free, antibiotic-free medium.
  - b) Warm the DNAfectin™ Plus to room temperature and vortex gently before use.
  - c) Add 6.0µl of the DNAfectin<sup>™</sup> Plus into the DNA solution from step a). Pipette up and down gently several times to mix the solution completely.
  - d) Incubate for 20 minutes at room temperature to form the DNAfectin<sup>™</sup> Plus-DNA complexes. Complexes are stable at room temperature for 3-5 hours.
- 3. Transfer the DNAfectin<sup>™</sup> Plus-DNA solution to the cultured cells drop-by-drop to different areas of the culture dish. Gently rock the culture vessel back-and-forth and side-to-side to evenly distribute the complexes.
- 4. Incubate for 12-16 hours. It is not necessary to change the culture medium after transfection with DNAfectin™ Plus, however, culture medium may be changed between 6-24 hours after transfection for sensitive cell lines.
- 5. Harvest cells and perform downstream analysis.

### **Optimizing Transfection for Specific Cell Lines**

To achieve the maximum transfection efficiency and low cytotoxicity, optimize the transfection conditions by varying cell density along with DNA and DNAfectin<sup>TM</sup> Plus concentrations. Optimal results have been observed when cells are 80-90% confluent and DNA(μg): DNAfectin<sup>TM</sup> Plus (μl) ratios are 1:1 to 1:5.

**Table 1: Reagent Quantities for Different Culture Vessels** 

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Culture Vessel	Volume of plating medium per well	DNA(μg)	DNAfectin™ Plus (μI)	Transfection medium volume
24-well	500µl	0.2-0.4µg	0.6-1.2µl	50µl
12-well	1ml	0.5-0.8µg	1.5-2.5µl	100µl
6-well	2ml	1.0-2.0µg	3-6µl	200µl
35mm	2ml	1.0-2.0µg	3-6µl	200µl
60mm	5ml	3.0-6.0µg	10-20µl	300µl
10cm	10ml	8.0-16.0µg	25-50µl	500µl

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