

FectHepG2™ Transfection Reagent

Table 1. Product Package and Storage

Cat No.	Product Name	Amount	Storage
FP317	FectHepG2™ Transfection Reagent	1 mL	4°C: one year

Introduction

FectHepG2™ Transfection Reagent 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.

Feature

- Superior transfection efficiency for HepG2 cells.
- Does not require removal of serum or culture medium.
- Does not require washing or changing of medium after transfection.
- Low cytotoxicity.

Protocols

Important Notes:

1. FectHepG2™ reagent was formulated for DNA transfection ONLY!
2. For high efficiency and lower toxicity, transfect cells at high density. 70~80% confluency is highly recommended.
3. To lower cytotoxicity, transfect cells in presence of serum (10%) and antibiotics.

Procedures for Transfecting HepG2 Cells:

The following procedure is given for transfection in 6-well plates, refer to Table 1 for transfection in other culture formats.

1. Seed cells 18 to 24 hours prior to transfection so that the monolayer cell density reaches to the optimal 70-80% confluency at the time of transfection.
2. For each well, add 1 ml of complete medium with serum and antibiotics freshly 30~60 minutes before transfection.
3. For each well, dilute 1 µg of DNA into 50 µl of serum-free DMEM with High Glucose. Gently pipette up and down or vortex briefly to mix.
4. For each well, dilute 3 µl of FectHepG2™ reagent into 50 µl of serum-free DMEM with High Glucose. Gently pipette up and down 3~4 times to mix.

Note: Never use Opti-MEM to dilute FectHepG2™ reagent and DNA, it contains serum and will disrupt transfection complex.

5. Add the diluted FectHepG2™ reagent **immediately** to the diluted DNA solution all at once. (**Important: do not mix the solutions in the reverse order !**)
6. Immediately pipette up and down 3~4 times or vortex briefly to mix.
7. Incubate for 10~15 minutes at room temperature to allow FectHepG2™/DNA complexes to form.

Note: Never keep the FectHepG2™/DNA complex longer than 20 minutes.

8. Add the 100 µl FectHepG2™/ DNA mixture drop-wise onto the medium in each well and homogenize the mixture by gently swirling the plate.
9. Remove FectHepG2™/DNA complex-containing medium and replace with fresh complete serum/antibiotics containing medium 12~18 hours post transfection.

10. Check transfection efficiency 24 to 48 hours post transfection.

Table 1. Recommended amounts for different culture vessel formats

Culture vessel	Plating medium volume	Dilution medium volume	Plasmid DNA	FectHepG2™ reagent
48-well	300 µl	2 × 15 µl	0.25 µg	0.75 µl
24-well	500 µl	2 × 25 µl	0.5 µg	1.5 µl
12-well	750 µl	2 × 38 µl	0.75 µg	2.25 µl
6-well	1 ml	2 × 50 µl	1.0 µg	3 µl
35-mm dish	1 ml	2 × 50 µl	1.0 µg	3 µl
60-mm dish	2.8 ml	2 × 100 µl	2.5 µg	7.5 µl
10-cm dish	5 ml	2 × 250 µl	5 µg	15 µl
T75 flask	8 ml	2 × 400 µl	9 - 18 µg	27 - 54 µl
250 ml flask	18 ml	2 × 800 µl	25 - 50 µg	75 - 150 µl