Transfectamine™ 5000 Transfection Reagent

Catalog number: 60020, 60021, 60022
Unit size: 0.5 mL, 1 mL, 5 mL

Component	Storage	Amount		
		Cat No. 60020	Cat No. 60021	Cat No. 60022
Transfectamine™ 5000 Transfection Reagent	Freeze (<-15 °C), Minimize light exposure	0.5 mL	1 mL	5 mL

OVERVIEW

Transfectamine™ 5000 Transfection Reagent is a powerful and versatile transfection reagent for the introduction of nucleic acids into eukaryotic cells, or more specifically, into animal cells. It can effectively transfect a variety of payloads into a variety of adherent and suspension cell lines. It can be used for plasmid DNA transfection as well as siRNA- and shRNA-based gene knockdown experiments and gene expression studies. It offers consistently high transfection efficiency in a wide variety of adherent and suspension cell lines, including difficult-to-transfect cells. The low toxicity of Transfectamine™ 5000 also allowed higher viability of transfected cells. Transfectamine™ 5000 is easier to use compare to most other transfection reagents and does not require special medium.

AT A GLANCE

Protocol summary

1. Prepare cells for transfection
2. Prepare Transfectamine™ 5000-DNA mixture
3. Add Transfectamine™ 5000-DNA mixture to cell culture
4. Culture overnight
5. Analyze transfection efficiency with appropriate method

Important Thaw component at room temperature before starting the experiment.

PREPARATION OF WORKING SOLUTION

1. Mix 2.5 ug of DNA with 200 uL of serum-free medium.
2. Add 7.5 uL of Transfectamine™ 5000 to Step 1.
3. Mix well and incubate at room temperature for 20 minutes.

Note Ratio of Transfectamine™ 5000 and DNA need to be optimized for different cell line, in general: Transfectamine™ 5000 Transfection Reagent (uL) to DNA (ug) Ratio = 3 - 5 uL to 1ug

Sample protocol detail for 6-well and 10 cm plate

Component	6 well plate (per well)	10 cm plate
Fresh culture medium	2 mL	6 mL
Plasmid	~2.5 ug	7.5~10 ug
Serum-free medium	200 uL	600 uL
Transfectamine™ 5000 Transfection Reagent	~7.5 uL	~22.5 uL

SAMPLE EXPERIMENTAL PROTOCOL

Preparation of Cell Culture

1. Culture cells to ~ 90% confluency at time of transfection.

2. Replace with fresh growth medium before transfection. For example, replace with 2 mL of medium per well for 6-well plates and 6 mL of medium for 10 cm plates.

Transtection Protocol

1. Add Transfectamine™ 5000 -DNA mixture to culture plate and culture overnight.

Note Recombinant protein can start to be detected as early as 16 hours post transfection. Maximal expression level may be observed 72~96 hours post transfection.

EXAMPLE DATA ANALYSIS AND FIGURES

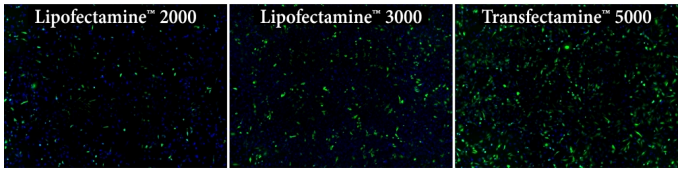

Figure 1. Transfection efficiency comparison in HeLa cells using Transfectamine™ 5000, Lipofectamine 2000 and Lipofectamine 3000 reagents. Each reagent was used to transfect HeLa cells in a 96-well format, and GFP expression was analyzed 24 hours post-transfection. Transfectamine™ 5000 transfection reagent provided higher GFP transfection efficiency compared to Lipofectamine 2000 and Lipofectamine 3000 reagents.

DISCLAIMER