Applied Biological Materials Inc.
Tel: 1-866-757-2414
Email: info@abmGood.com
Website: www.abmGood.com

Lentifectin™

Store at 4°C

Cat. No.	Description	Quantity
G074	Lentifectin™	1.0 mg / 1.0 ml

Description

Lentifectin™ is a transfection reagent specially formulated with multiple cationic polymers for the production of Lentiviral particles *in vitro*. Lentiviral supernatants produced using Lentifectin™ mediated transfection consistently show higher titers than those produced with calcium-phosphate transfection or with other types of lipid transfection reagents.

Shipping and Storage

Upon arrival, Lentifectin™ should be stored at 4°C. Freezing is not recommended.

Transfection Protocol

The following protocol allows the production of recombinant lentiviral particles up to a 10^6 IU/ml titer, and higher titers can be achieved by using a scaled up protocol. We recommend including a negative control (no DNA, no transfection reagents) in your experiment to help you evaluate your results. You will need 1.2 x 10^7 293T cells for each 10 cm dish transfection.

DAY 1:

1. In the afternoon, seed ~1.2 x 10^7 293T cells in a 10 cm dish.

DAY 2: *(Carry out steps 2 - 6 in the morning on the day of transfection)*

2. Check to confirm the cells are 70 - 80% confluent.

3a. For each 10 cm dish, prepare the transfection complex as follows:

Solution A: Dilute 20 µg DNA plasmids (10 µg expression vector and 10 µg of **abm**'s 2nd Generation Packaging Mix (Cat. No. LV003) or 3rd Generation Packaging Mix (Cat. No. LV053) in 1 mL serum-free, antibiotic-free medium.

Solution B: Dilute 80 µL of Lentifectin™ (Cat. No. G074) in 1 mL serum-free, antibiotic-free medium.

3b. Incubate both solutions at room temperature for 5 minutes.

3c. Mix Solutions A and B and incubate at room temperature for 20 minutes. This will create the transfection complex.

4. Add 4.5 mL serum-free medium to the transfection complex.

5. Aspirate medium from the cells to be transfected.

6. Add the complete transfection complex from step 4 to the cells dropwise and incubate at 37°C for 5 - 8 hours. Avoid dislodging the cells by gently adding the mixture against the side wall of the dish.

7. Add 0.65 mL FBS to the 10 cm dish and incubate at 37°C overnight.

Caution: Remember that following the transfection and packaging step, you will be working with infectious virus present in the supernatant collections. Follow the recommended guidelines for working with BL-2 organisms.

DAY 3:

8. Aspirate the transfection medium from the cells and add 10 ml of fresh medium to the cells. Incubate at 37°C for 24 hours.

Note: *Expression of the VSVG glycoprotein causes 293T cells to fuse, resulting in the appearance of large, multinucleated cells known as syncytia. This morphological change is normal and does not affect the production of the lentivirus.*

Proceed with **abm**'s Lentivirus Packaging Protocol for collecting your viral supernatants and performing downstream purification.