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ATGCell

DNAFect® 1.0

For transient and stable transfection of mammalian cells

Catalogue Number: DF01-05, Size: 0.5 ml
Catalogue Number: DF01-10, Size: 1.0 ml
Catalogue Number: DF01-50, Size: 5.0 ml
Catalogue Number: DF01-100, Size: 10 ml

Product Summary

DNAFect® 1.0 is a new generation of lipid-based transfection reagent optimized for DNAs delivery into mammalian cells providing the following advantages:

- High transfection efficiency in many cell types and formats. Comparable with the leading brand Lipofectamine/FuGene/GeneJuice.
- Lower toxicity than the leading brands Lipofectamine/FuGene/GeneJuice.
- There is no need to remove DNA-DNAFect 1.0 complexes or change medium after transfection.
- The transfection complexes can be added directly to cells in culture medium, in the presence or absence of serum.
- Affordability: One ml of DNAFect 1.0 can typically be used for 650 transfections of 24-well plate cultures.

Important Guidelines for Transfection

- We recommend Opti-MEMI serum-reduced medium or any serum-free medium to dilute DNAFect 1.0 and DNA before complexing.
- We recommend not adding antibiotics to media during transfection.
- Maintain the same seeding density between experiments.

Transfection Protocol

Use the following procedure to transfect DNA into mammalian cells in a **24-well format**. For other formats, see **Scaling Up or Down Transfections**. All amounts and volumes are given on a per well basis. Optimization may be required in some cases.

1. One day before transfection, plate 0.5-1.5 x 10^5 cells in 500 µl of growth medium so that cells will be 50-70% confluency at the time of transfection.
2. For each transfection event, prepare complexes as follows:
    a. Dilute DNA in 50 µl of Opti-MEMI medium without serum (or other medium without serum).

   b. Mix DNAFect 1.0 gently before use, then dilutes the appropriate amount of DNAFect 1.0 (see table below) in 50 µl of Opti-MEMI medium. Mix gently and incubate for 5 minutes at room temperature.
   c. After the 5 minute incubation, combine the diluted DNA with diluted DNAFect 1.0 (total volume = 100 µl). Mix gently and incubate for 20 minutes at room temperature. Note: Complexes are stable for at least 4 hours at room temperature.
3. Aspirate off old growth medium, wash once with 1 x PBS, and followed by adding 200 µl Opti-MEM I medium with reduced serum (5% FBS) or other serum-free medium.
4. Add the 100 µl of complexes to each well containing cells and medium. Mix gently by rocking the plate back and forth.
5. Incubate cells at 37°C in a CO_2 incubator for 6-8 hours, or overnight.
6. Add 700 µl of fresh growth medium into each well.
7. Incubate 36-72 hours prior to testing for transgene expression.
8. For stable cell lines: Passage cells at a 1:5 (or higher dilution) into fresh growth medium 24 hours after transfection. Add selective medium (if desired) on the following day.

Scaling Up or Down Transfections

To transfect cells in different cell culture formats, vary the amounts of DNAFect 1.0, DNA, cells, and medium used in proportion to the relative surface area, as shown in the table.

Culture vessel	Surface area per well	Medium		Transfection	
		Planting volume	Dilution volume	DNA	DNAFect 1.0
96-well	0.3 cm^2	40 µl	2 x 10 µl	0.1 µg	0.3 µl
24-well	2 cm^2	200 µl	2 x 50 µl	0.6 µg	1.6 µl
12-well	4 cm^2	400 µl	2 x 100 µl	1.2 µg	3.2 µl
6-well	10 cm^2	1.0 ml	2 x 250 µl	3.0 µg	8.0 µl
60-mm	20 cm^2	2.0 ml	2 x 0.5 ml	6.0 µg	16.0 µl
100-mm	60 cm^2	6.0 ml	2 x 1.5 ml	18.0 µg	48.0 µl

Shipping and Storage

DNAFect 1.0 reagent is shipped at room temperature. Upon receipt, store this reagent at 4°C.