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Transfection by using K2 system

Cell type:

Human neuroblastoma SH-SY5Y cell line

Culture media:

The SH-SY5Y cells were grown in Ham's F-12 and MEM medium supplemented with 10% fetal bovine serum (FBS), 0.1% nonessential amino acid, 0.1% sodium pyruvate and 1% penicillin-streptomycin at 37°C with 5% CO_2.

Procedures:

The SH-SY5Y cells were plated at 5 x 10^5 cells onto 6 wells culture plates for 24 hours. Then, 20 µl of K2 multiplier was added to the cells with 90-100% confluence in 2 ml of fresh medium for 2 hours. The plasmid DNA containing GFP expression was transfected into the SH-SY5Y cells using K2 reagent following company protocol. The amounts of plasmid DNA and transfection reagents are as follows.

Component	Solution A	Solution B
Plasmid DNA	4 µg	-
Opti-MEM	To 50 µl	34 µl
K2 transfection reagent	-	16 µl
Total	50	50

Solution A was transferred to solution B and then incubates for 15 minutes before adding to the cells. After 24 hours, transfection medium was replaced with fresh medium and the positive clones were identified by green fluorescent of GFP expression under fluorescent microscopy.

Results:

The result from transient transfection showed several positive clones of GFP with efficiency of 70-80%.

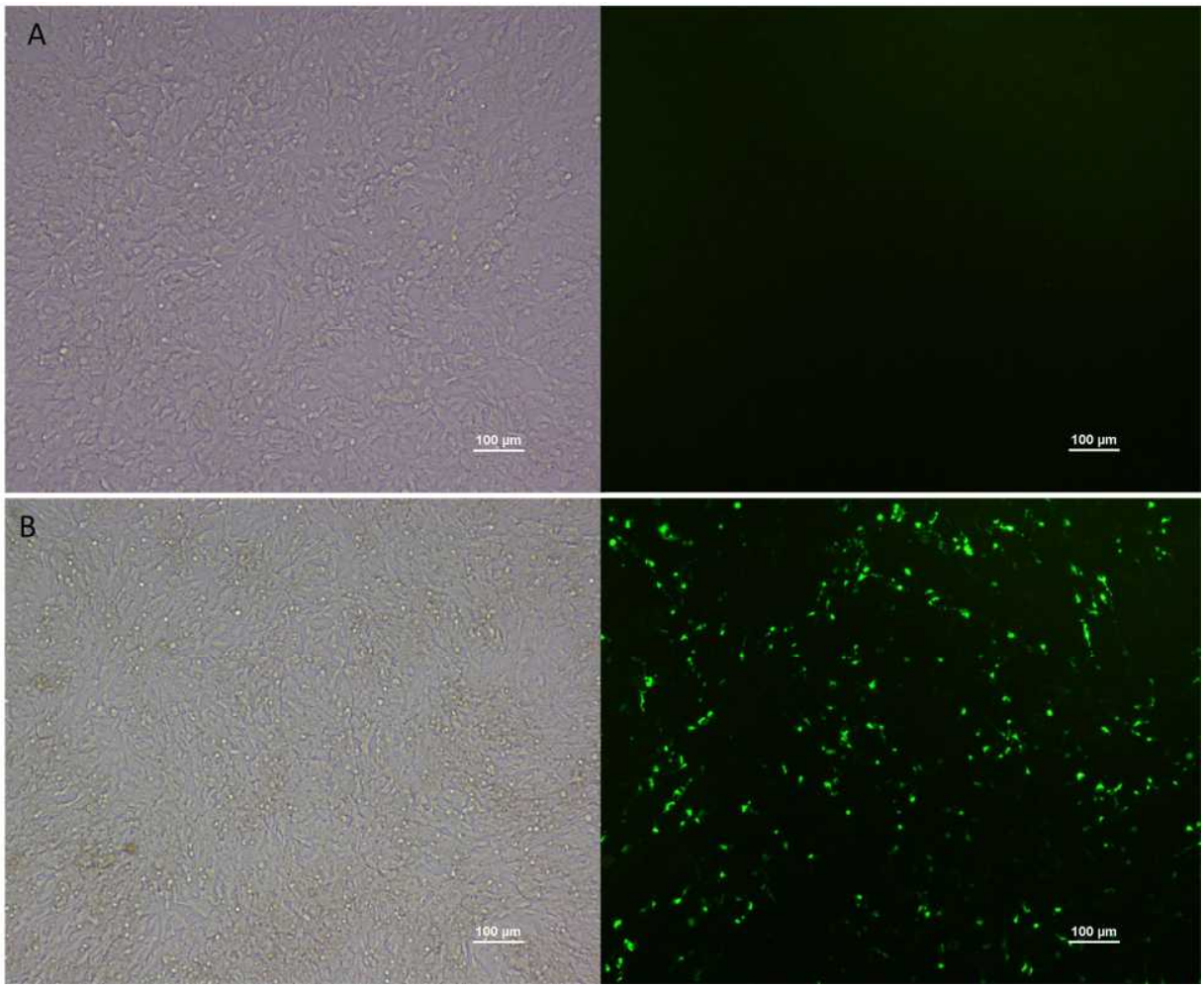

Figure1. The expression of GFP was visualized under fluorescent microscope (10X). Untransfected cells (A). Transfected cells (B).