K2 transfection reagent Technical Note

Yue Ma, Zhen Wang and Weilin Jin

School of Life Science and Biotechnology, Shanghai Jiao Tong University, 800 Dong Chuan Road, Minhang, Shanghai, 200240 CHINA

Materials:

Plasmid pEGFP-N1

sterile 24-well culture plates

sterile Eppendorf tubes

DMEM+10% Fetal Bovine Serum

HEK 293T cells

HEK293T cells transfection:

HEK293T cells were grown in a 24-well cell culture plate in DMEM with 10% Fetal Bovine Serum to a confluency of 70-80%, pipet 5µL K2® Multiplier into each well 2h before adding the lipoplex. Prepare the following solutions A and B in 1.5 mL Eppendorf tubes:

Solution A	**Solution B**
30 µL serum-free medium (opti-MEM)	30 µL serum-free medium (opti-MEM)
0.5 µg DNA	2 µL K2® Transfection Reagent

Add solution A into B, mix the solution gently, and incubate the mixture at room temperature for 15min. After incubation, add the complex to the cells, agitate the cell culture plate gently to mix , and the cells were cultured in a CO2 incubator at 37°C. Images were captured between 20-48h after addition of the DNA-lipid complex using Leica DMI 6000B. (Fig 1, upper)

The transfection efficiency with K2 was compared to another commercial transfection reagent FuGENE HD . The transfection of HEK 293T cells with FuGENE HD was performed previously

followed manual. Images were captured 24h and 48h respectively post transfection using Nikon ECLIPSE TS100.

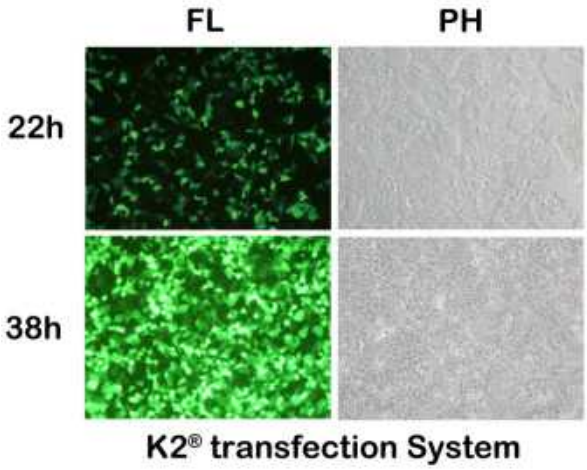
Conclusions:

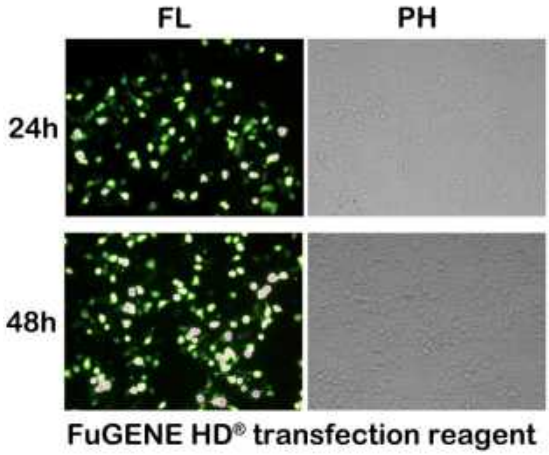
The transfection efficiencies for HEK293T cells can reach more than 90% under the optimized transfection conditions with K2 transfection reagent. The intensity of fluorescence increased over time and at 38 hour the EGFP expression can reach a very high level. Meanwhile, the K2 transfection reagent shows very low toxicity to cells, the cells still stay health 24h post transfection. Moreover, the EGFP expression levels of each cell transfected with K2 are relatively even , comparing to FuGENE HD, which resulting in a high variation of EGFP expression among cells.

In summary, K2 transfection system provieds high efficiency plasmid DNA transfection and relatively even expression with low toxicity.

Fig 1: HEK293T cells were transfected with a plasmid expressing EGFP using the K2 transfection reagent (upper) and FuGENE HD (lower) according to the optimized conditions. FL, fluorescence; PH, phase contrast.

Fig. 1




FL
PH
24h
48h
FuGENE HD® transfection reagent