

DNA transfection of 1C11 cells using « Biontex K2 Transfection system »

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Cell culture

1C11cells were cultured in antibiotic-free High glucose Dubelcco's modified eagle medium (Gibco) containing 10% fetal calf serum in a 10% C02 atmospher. Cells were plated in 12 well plates in 1 ml media 24h before transfection. The transfection was performed when cells reached 80-90% of confluency.

Cell transfection

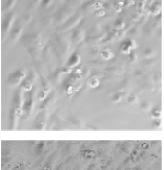
Cells were treated with K2® Multiplier, 2 hours before DNA transfection. For this **K2® Multiplier** was dripped slowly onto the medium and mixed by gently swaying the dishes. **K2® Transfection Reagent** was mixed with Opti-MEM® (life technologies) and left on room temperature during preparation of the DNA. Plasmid-DNA encoding GFP (EGFP-C2) was mixed with Opti-MEM. DNA solution was added to the solution containing the K2® Transfection reagent (not the other way around) and mixed by inverting the tubes, followed by 15 minutes incubation at room temperature. Transfection solution was applied to cells by slow dropwise addition to the medium followed by gently swaying the dishes to achieve mixing. Transfections were incubated at 37C and 10% CO2 for 24 hours. Transfection efficiency was estimated by fluorescence microscopy. **Accurate reagent amounts are display in the table below:**

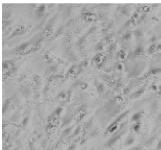
1C11 (r1:3)

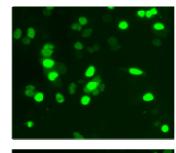
Dish sizes	DMEM	K2 Multiplier	K2 Transfection reagent	OptiMEM/K2 Transfection reagent	DNA [μg]	OptiMEM for DNA	Mix final	Vol
12 well	1 ml	2,5 μΙ	4,2	60µl	1,4 μg	60 μl	120 µl	
12 well	1 ml	5 μΙ	4,2	60µl	1,4 μg	60 μl	120 µl	·

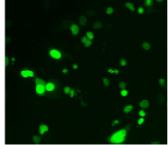
12 well plate

1C11 cells









1:3 /0,3ug 2,5ul K2 multiplier

> 1:3 /0,3ug 5ul K2 multiplier

Conclusions:

From previously tested reagents, the K2 \circledR transfection system is highly efficient in the 1C11 cell line without inducing too much cell death.