

Electroporation protocol for T-cells

GTporator®

Transfection protocol

Protocol No. 09/2008-001

Cell line	T-cells (cell lines and primary cells)
Washing solutions	Phosphate buffered saline (PBS), pH 7.4, GTporator®-M
Cell count	1 x 10 ⁶
Electroporation solution	GTporator®-M
Cuvette	2 mm gap width
Volume	100 µl
Temperature	Room temperature
DNA	5 µg in water

Instrument settings

<u>Single amplitude</u>	<u>Radio-frequency</u>	<u>Transf. into nucleus</u>
Voltage 220 V	Voltage 220 V	Program C009
Capacitance 1500 µF	Frequency 40 kHz	Volume 120 µl
Pulse time 34 ms	Pulses 5 x 4 ms	Vector 2.5-5 µg
	Interval 1 s	
	Modulation 100%	

1. Wash the cells once in 1 ml PBS, and once in 300 µl of GTporator® solution
2. Resuspend the cells in 100 µl GTporator® solution with 5 µg of plasmid DNA
3. Transfer into a 2 mm electroporation cuvette
4. Electroporate
5. Immediately add pre-warmed culture medium with serum, transfer the cells into cell culture flask and place in incubator
6. Test the gene expression 24 h after electroporation

Notes:

- The overall incubation time in the GTporator® solution must not exceed 20 minutes to guarantee a successful electroporation!
- The best results are obtained with DNA purified by anion-exchange chromatography.
- DNA should be dissolved in sterile water, using the TE buffer slightly lowers the survival rate.

Typical results:

Survival rate in control cells 50%

Transfection rate: 15-25% with variations depending on the quality of DNA.

Results were measured 24 hours after transfection.

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