

Electroporation protocol for HEK293 cells

GTporator®

Transfection protocol

Protocol No. 09/2008-006

Cell line	HEK293
Washing solutions	Phosphate buffered saline (PBS), pH 7.4, GTporator®-M
Cell count	1-3 x 10 ⁶
Electroporation solution	GTporator®-M
Cuvette	2 mm gap width
Volume	80 µl
Temperature	Room temperature
DNA	5 µg in water

Instrument settings

<u>Single amplitude</u>		<u>Radio-frequency</u>	
Voltage	220 V	Voltage	200 V
Capacitance	960 µF	Frequency	40 kHz
Pulse time	25 ms	Pulses	5 x 4 ms
		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 80 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 80%

Transfection rate: 80-95% with variations depending on the quality of DNA.

Results were measured 24 hours after transfection.

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Protean s.r.o., Dolní 2, 370 04 České Budějovice, Czech Republic, Tel.: +420 724 731 831

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E-mail: info@protean.cz, web: www.protean.cz