

Jurkat Transfection by Electroporation

- 1 Wash 10^7 Jurkat cells in cold serum-free RPMI.
- 2 Resuspend in 180 μ l serum-free RPMI, store on ice in 0.4 cm electroporation cuvette.
- 3 Sterilize DNA: Precipitate 5 pmol (around 10 μ g) plasmid DNA in 100 μ l TE + 10 μ l 3M NaAc by adding 500 μ l EtOH and spinning 10' @ 13000 rpm, 4C.
- 4 Take off sup carefully under the hood and dry pellet briefly. Resuspend in 20 μ l serum-free RPMI by incubation @ 65C.
- 5 Add DNA solution to cells in cuvette and mix.
- 6 Electroporate @ 250 V, 960 μ F.
- 7 Culture cells in 5-10 ml RPMI/10% FCS until analysis 16-48 h later.