

Bio-Rad Labs Optimization Strategy with the RF Module

Function (Settings)	Range	Optimization Strategy
Total volts	5 - 400 v	Vary voltage in 0.2 cm cuvette for field strength of 0.7-1.4 kV/cm (160-280V setting)
% Mod (Percent modulation)	0 -100%	100% (decrease to 0% for square wave if no killing)
RF Freq. (Radio frequency)	5 -50 kHz	40-50 kHz
Burst Dur. (Burst duration)	0.1-100 msec	2 msec
No. Brsts (Number of bursts)	1 - 25	5-10 (repeat 1 -2 times)
Interv. (Burst interval)	0.1 - 10 sec	100 msec
cuvette size	.2cm	try .4cm if cell death high, .1cm if no killing

Example Cell Types

Reported by Dr. Donald Chang, Hong Kong University, 8/96 developed with the Gene Pulser II/ RF module system:

Ptk-2 cells (loaded with a plasmid DNA containing a fusion gene of calmodulin and GFP, i.e. the "green fluorescent protein")

Conditions: 1.0 - 1.3 kV/cm, 20 or 30 KHz, 1X10 pulses or 3X5 pulses

Rat hippocampus neurons (loaded with FITC dextran)

Conditions: 0.8 - 1.4 kV/cm, 40 KHz, 1X5 pulses

Neuroblastoma cells (loaded with a [Ca²⁺] indicator, Calcium Green-1 conjugated with 10 kD dextran)

Conditions: 0.8 - 1.2 kV/cm, 40 KHz, 2X5 pulses

Hela cells (loaded with a plasmid DNA containing a fusion gene of calmodulin and the green fluorescent protein)

Conditions: 0.8 - 1.2 kV/cm, 30 KHz, 2X10 pulses

In all of the above experiments, the pulse width was always set at 2 ms, the interval between pulses was 100 ms.

Electroporation Buffer should be Phosphate Buffered Sucrose