BTXpress High Performance Electroporation Solution & Kits

Introduction

BTXpress High Performance Electroporation Solution and Kits provide increased transfection efficiencies and cell viabilities during electroporation of DNA. This buffer can used with a broad range of cells including those considered "hard to transfect". The BTXpress Electroporation solution is compatible with all BTX generator systems in cuvette applications.

Specifications

Storage	Store the BTXpress High Performance Electroporation Solution at 4°C. All other materials can be stored at room temperature.
Stability	6 months from the date of purchase, when properly stored and handled.
Number of Electroporations	One ml of BTXpress High Performance Solution is sufficient for 4 electroporations in 0.4 ml cuvettes or 10 electroporations in 0.2 cm cuvettes.

Materials

MATERIALS SUPPLIED

BTXpress High Performance Electroporation Solution Kits include BTX plus cuvettes with transfer pipettes. These cuvettes may also be purchased separately.

Kit Part Numbers	**2mm Gap** Cuvette includes transfer pipette (Blue Cap)	**4mm Gap** Cuvettes include transfer pipettes (Yellow cap)	**BTXpress Electroporation Solution**
45-0800	5	5	2 mls
45-0803	50	NA	5.0 mls
45-0804	NA	20	5.0 mls
45-0806	100	NA	10.0 mls
45-0807	NA	40	10.0 mls
Reagent Only Part Numbers	**No Cuvettes**	**No Cuvettes**	**BTXpress Electroporation Solution**
45-0802	NA	NA	5.0 mls
45-0805	NA	NA	10.0 mls

MATERIALS REQUIRED, BUT NOT SUPPLIED

Before You Start

IMPORTANT TIPS FOR OPTIMAL ELECTROPORATIONS

Optimize electroporation conditions for each cell type to ensure successful results. The suggestions below generally yield highly efficient electroporations. Table 1 presents recommended starting pulse conditions for DNA electroporation into select hard to transfect cells.

- **DNA Purity.** Use highly purified, sterile, and contaminant-free DNA. Endotoxin-free DNA (bacterial lipopolysaccharide-free) is optimal. Do not use nucleic acid that has been purified using ethanol precipitation. High salt concentrations used in ethanol precipitation methods can affect electroporation.
- **DNA Concentration.** Use DNA stocks that range from 1 to 5 mg/ml. Use of stocks with higher concentrations may lead to non-uniform mixing with cells. Use of stocks with lower concentrations may dilute the electroporation mix too much.
- **Avoid storing the cells in BTXpress Electroporation Solution.** Incubation of the cells in BTXpress Electroporation Solution at room temperature for more than 15 minutes may be harmful to the cells.
- **Split cells regularly.** Maintain cells such that they are actively growing. Split the cell culture one day before electroporation as needed. This step may not be required for slow-growing or suspension cells.
- **Cell passage number.** Use of very low or very high passage cells may affect experimental results. Use cells of similar passage number for experimental reproducibility.
- **Post-electroporation incubation time.** Determine the optimal incubation time post-electroporation for each cell type. Test a range of incubation times. The optimal incubation time is generally 12 - 48 hours, but will vary depending on the goal of the experiment and the electroporated DNA
- **Optimized electroporation for other cell types.** For cells other than those listed in Table 1, more optimization will be required. If using a Square wave system the general pulse conditions for most cells fall with in a range of 120-200 Volts. Pulse length ranges can vary depending on cell type and range between 5-20msec for a single pulse using a 2mm gap cuvette. If using a 4mm gap cuvette the voltage ranges are 170-300V and pulse lengths of 5-25msec for a single pulse.
- For Exponential decay wave systems, voltage ranges for common cell lines are in the range of 200-300 V and a capacitance range of 800-1000uF with a resistance setting of “none” when using a 4mm gap cuvettes. For 2mm gap cuvettes the ranges are 80-160V and 400-950uF with resistance set to “none”.

Procedure

The procedure below describes how to perform electroporations using the BTXpress High Performance Electroporation Solution. This protocol can be followed using either a BTX square wave or exponential decay wave electroporator.

Table 1

Recommended electroporation conditions for select cell types using Square Wave Systems

Cell Type	Cuvette Size	Cell Density (X x 106 cells/ml)	Voltage	Number of Pulses	Pulse Length (milliseconds)
MEF	2 mm	5	170	1	15 msec
	4 mm	5	240	1	15-20 msec
Jurkat E6-1	2 mm	10	180	1	15 msec
	4 mm	10	290	1	15-20 msec
Niks (Kerotinocytes)	2 mm	3	180	1	15 msec
	4 mm	3	300	1	15-20 msec

Table 2

Recommended electroporation conditions for select cell types using Exponential Decay Wave Systems

Cell Type	Cuvette Size	Cell Density (X x 106 cells/ml)	Voltage	Capacitance (µF)	Resistance (ohms)
MEF	2 mm	5	150	950	None
	4 mm	5	230	950	None
Niks (Kerotinocytes)	2 mm	3	170	950	None
	4 mm	3	280	950	None
Jurkat E6-1	2 mm	10	150	950	None
	4 mm	10	260	950	None
SK-N-MC	2 mm	5	90	950	None
	4 mm	5	240	950	None
K562	2 mm	10	130	950	None
	4 mm	10	250	950	None
HL-60	2 mm	10	110	950	None
	4 mm	10	275	950	None
RAW 264.7	2 mm	5	140	950	None
	4 mm	5	260	950	None
THP-1	2 mm	10	125	950	None
	4 mm	10	250	950	None
HEK-293	2 mm	5	160	950	None
	4 mm	5	250	950	None
PC-12	2 mm	3	130	950	None
	4 mm	3	240	950	None

A. Preparation of cells 1 day prior to electroporation

1. Maintain the cells in appropriate complete growth medium and cell density as recommended by ATCC. Appropriate cell density and growth conditions can also be determined empirically.
2. If necessary, divide cultured cells approximately 18 – 24 hours prior to electroporation so that the cells reach optimal cell density at time of electroporation. For suspension cells, optimal cell densities are approximately 1 – 2 million cells/ml. For adherent cells, optimal cell densities are approximately 70 – 80% confluent. Users may also determine their own best cell densities.
3. Incubate the cells overnight.

B. Prepare for electroporation

1. Warm all required solutions to room temperature for 15 – 30 minutes before use.
2. Harvest cells for electroporation. Count cells to determine the total number of cells per milliliter.
3. Determine the total volume of BTXpress High Performance Electroporation Solution required to perform all the desired electroporations: Multiply the number of electroporations by 0.1 ml (for 0.2 cm cuvettes) OR by 0.25 ml (for 0.4 cm cuvettes), and ad 10% more to account for pipetting errors.
4. Determine the volume of cells from step B-2 required for all electroporations according to the formula:

Electroporation Cell Density Required cells/ml

$$\text{Cell volume required (ml)} = \frac{\text{Electroporation cell density/ml}}{\text{Harvested cell density/ml}} \times \text{Total Volume of BTXpress from step 3}$$

$$\frac{*5\times10^6 \text{ c/ml}}{6\times10^6 \text{ c/ml}} \times 0.3\text{ml} = 0.25 \text{ mls}$$

** This is the number of cells per ml needed for each electroporation.*

** Refer to Table 1.*

5. Pipette the volume of cells determined in step B-4 into a new tube and centrifuge at 1000 x g for 5 minutes. Aspirate the supernatant.
6. During the centrifugation, add complete growth medium to a culture plate to accept cells after electroporation.
7. Resuspend the cells from step B-5 in BTXpress High Performance Electroporation Solution, using the volume determined in step B-3.

Do NOT allow cells to incubate in BTXpress Solution for more than 15 minutes.

C. Perform electroporation

1. Add DNA to the cells in BTXpress High Performance Electroporation Solution Solution. Use 20 µg DNA per ml of cells. Mix gently but thoroughly. Do not create bubbles.
2. Aliquot 100 µl DNA/cell mix to each 0.2 cm cuvette OR add 250 µl DNA/cell mix to each 0.4 cm cuvette.
3. Electroporate the cells at room temperature. Refer to Table 1 for appropriate pulse conditions, or determine them experimentally.
4. Immediately after performing each electroporation, transfer the electroporated cells into the culture vessel prepared in step B-6. If desired, use a micropipette to transfer an exact amount (e.g. 100 µl) from each 0.4 cm cuvette. For 0.2 cm cuvettes, we recommend adding an additional 100 µl of complete media using a micropette. Then use a transfer pipette to gently mix and transfer the entire contents to the culture plate.

   NOTE: *Transfer the cells immediately following electroporation to a culture vessel containing warm complete culture media.*

Incubation of the cells for more than 15 minutes may prove harmful to the cells.

5. Incubate the electroporated cells in complete culture medium in appropriate growth conditions (e.g. 37°C, 5% CO2 in a humidified incubator) for 12 – 72 hours or as required.
6. Harvest cells and perform a reporter assay or other assay as required.

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