User's Manual

ECM® 630 Electroporation System



MA1 45-0051	ECM [®] 630 Electroporator only (110V)
MA1 45-0051int	ECM [®] 630 Electroporator only (220V)



The Electroporation Experts

WEEE/RoHS Compliance Statement

EU Directives WEEE and RoHS

To Our Valued Customers:

We are committed to being a good corporate citizen. As part of that commitment, we strive to maintain an environmentally conscious manufacturing operation. The European Union (EU) has enacted two Directives, the first on product recycling (Waste Electrical and Electronic Equipment, WEEE) and the second limiting the use of certain substances (Restriction on the use of Hazardous Substances, RoHS). Over time, these Directives will be implemented in the national laws of each EU Member State.

Once the final national regulations have been put into place, recycling will be offered for our products which are within the scope of the WEEE Directive. Products falling under the scope of the WEEE Directive available for sale after August 13, 2005 will be identified with a "wheelie bin" symbol.

Two Categories of products covered by the WEEE Directive are currently exempt from the RoHS Directive – Category 8, medical devices (with the exception of implanted or infected products) and Category 9, monitoring and control instruments. Most of our products fall into either Category 8 or 9 and are currently exempt from the RoHS Directive. We will continue to monitor the application of the RoHS Directive to its products and will comply with any changes as they apply.



- Do Not Dispose Product with Municipal Waste
 - Special Collection/Disposal Required

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General Information

Serial Number

The serial number for the ECM[®] 630 is located on the rear of the instrument case. All inquiries concerning these products should refer to the serial numbers on the units.

Calibration

There is no calibration required for the ECM[®] 630.

Warranty

BTX - Harvard Apparatus warranties the ECM* 630 for a period of two years from the date of purchase. At its option, BTX – Harvard Apparatus will repair or replace the unit if it is found to be defective as to workmanship or materials. This warranty does not extend to any instrumentation which has been (a) subjected to misuse, neglect, accident or abuse, (b) repaired or altered by anyone other than BTX - HARVARD APPARATUS without BTX - HARVARD APPARATUS' express and prior approval, (c) used in violation of instructions furnished by BTX - HARVARD APPARATUS. This warranty extends only to the original customer purchaser.

IN NO EVENT SHALL BTX - HARVARD APPARATUS BE LIABLE FOR INCIDENTAL OR CONSEQUENTIAL DAMAGES. Some states do not allow exclusion or limitation of incidental or consequential damages so the above limitation or exclusion may not apply to you. THERE ARE NO IMPLIED WARRANTIES OF MERCHANTABILITY, OR FITNESS FOR A PARTICULAR USE, OR OF ANY OTHER NATURE. Some states do not allow this limitation on an implied warranty, so the above limitation may not apply to you.

Without limiting the generality of the foregoing, BTX - HARVARD APPARATUS shall not be liable for any claims of any kind whatsoever, as to the equipment delivered or for non-delivery of equipment, and whether or not based on negligence.

Warranty is void if the ECM* 630 is changed in any way from its original factory design or if repairs are attempted without written authorization by BTX - HARVARD APPARATUS.

Warranty is void if parts, connections or cell fusion chambers not manufactured by BTX - HARVARD APPARATUS are used with the ECM[®] 630.

General Information (Continued)

If a defect arises within the warranty period, promptly contact BTX – Harvard Apparatus, 84 October Hill Road, Building 7, Holliston, Massachusetts, USA 01746-1388 using our toll free number **1-800-272-2775** (US Only) or **508-893-8999**

(E-mail: **techsupport.btx@harvardapparatus.com**). Goods will not be accepted for return unless an RMA (Returned Materials Authorization) number has been issued by our customer service department. The customer is responsible for shipping charges. Please allow a reasonable period of time for completion of repairs, replacement and return. If the unit is replaced, the replacement unit is covered only for the remainder of the original warranty period dating from the purchase of the original device.

This warranty gives you specific rights, and you may also have other rights, which vary from state to state.

Service

All service under the warranty will be made at the BTX - HARVARD APPARATUS, Holliston, Massachusetts facilities or an authorized service site. Owner will ship instrument prepaid to Holliston, Massachusetts, USA or the service site. BTX - HARVARD APPARATUS will return the instrument after servicing, freight prepaid to owner's address.

Obtaining Service:

Service During Warranty

- 1. Write or call the BTX HARVARD APPARATUS Customer Support Group and describe the nature of the problem.
- Carry out minor adjustments or tests as suggested by BTX - HARVARD APPARATUS.
- If proper performance is not obtained, BTX HARVARD APPARATUS will notify you to ship the instrument, prepaid, to its Service Department. The instrument will be repaired and returned at no charge for all customers in the continental United States.

Customers outside of the continental United States who have purchased our equipment from distributors should contact the distributor. If you have purchased your equipment from us, you should contact us directly. We will repair at no charge, but will not pay for shipment, documentation, etc. These charges will be billed at cost.

Note: Under no condition should the instrument or accessories be returned without prior approval from BTX - HARVARD APPARATUS. An RMA (Returned Materials Authorization) number must be obtained.

General Information (continued)

Out-Of-Warranty Service

Proceed exactly as for Warranty Service, above. If our Service Department can assist you by phone or correspondence, we will be glad to, at no charge.

Repair service will be billed on the basis of labor and materials. A complete statement of time spent and materials used will be supplied. Shipment to BTX - HARVARD APPARATUS should be prepaid. Your bill will include return shipment freight charges.

Disassembly by the user is prohibited. Service should only be carried out by experienced BTX - HARVARD APPARATUS technicians.

Repair Facilities and Parts

BTX - Harvard Apparatus stocks replacement and repair parts. When ordering, please describe parts as completely as possible, preferably using our part numbers. If practical, enclose a sample or drawing. We offer complete reconditioning service.

General Safety Summary

Review the following safety precautions to avoid injury and prevent damage to this product or any products connected to it. To avoid potential hazards, use this product only as specified.

Only qualified personnel should perform service procedures.

To Avoid Fire or Personal Injury

USE PROPER POWER CORD

Use only the power cord specified for this product and certified for the country of use.

CONNECT AND DISCONNECT PROPERLY

Do not connect or disconnect probes or test leads while they are connected to a power source.

GROUND THE PRODUCT

This product is grounded through the grounding conductor of the power cord. To avoid electric shock, the grounding

conductor must be connected to earth ground. Before making connections to the output terminals of the product, ensure that the product is properly grounded.

OBSERVE ALL TERMINAL RATINGS

To avoid fire or shock hazard, observe all ratings and markings on the product. Consult the product manual for further ratings information before making connections to the product.

DO NOT OPERATE WITHOUT COVERS

Do not operate this product with covers or panels removed.

Use Proper Fuse. Use only the fuse type and rating specified for this product.

AVOID EXPOSURE TO CIRCUITRY

Do not touch exposed connections and components when power is present.

DO NOT OPERATE IN LOW IMPEDANCE

Sample: Load or Sample

If the electroporation samples have an impedance of less than 20 \Box the samples may arc and result in sample loss and potential damage to unit.

DO NOT OPERATE WITH SUSPECTED FAILURES

If you suspect there is damage to this product, have it inspected by qualified BTX service personnel.

PROVIDE PROPER VENTILATION

Refer to installation instructions for details on installing the product to ensure proper ventilation.

General Safety Summary (Continued)

DO NOT OPERATE IN WET/DAMP CONDITIONS DO NOT OPERATE IN AN EXPLOSIVE ATMOSPHERE **KEEP PRODUCT SURFACES CLEAN AND DRY** Should you have any safety concerns, immediately contact BTX Technical Services (1-800-272-2775)

Safety Terms and Symbols:

Terms that appear in this manual:



WARNING. Warning statements identify conditions or practices that could result in injury or loss of life.

CAUTION. Caution statements identify conditions or practices that could result in damage to these products or other property.

Symbols that may appear on the products:



Danger High Voltage

Attention Refer to Manual



(Earth)

Terminal



Functional Protective Ground

Electrical & Technical Specifications

Standard Capabilities:

Display	Type: 20-character by 4-line liquid crystal Display. LED backlit
Power Source	
Voltage	100 to 240 Vac, 50 to 60 Hz,
Davier	CAT II
Fusing	2.5 A. T. rating 250 V
Fushig	2.5 A, 1 Tating 250 V
Environmental Characteristic	s:
Intended Use	Indoor use only
Operating Temperature	$10^{\circ}\mathrm{C}$ to + $40^{\circ}\mathrm{C}$
Cooling	Convection through metal case
Relative Humidity	60%
Altitude	2,000 m (operating)
Mechanical Characteristics:	
Footprint	12.5" x 12.25" x 5.5" (W x D x H)
Weight	13.6 lbs (6.2 kg)
Controls	Single rotary encoder with integrated push button

General Specifications

Certifications and Compliances

Overvoltage Category:

CAT III: Products in this Category: Distribution-level mains, fixed installation.

CAT II: Local-level mains, applications, portable equipment.

CAT I: Signal levels in special equipment or parts of equipment, telecommunications, electronics.

Meets requirements of Directive 89/336/EEC for Electromagnetic Compatibility (EC) and Low-Voltage Directive 73/23/EEC for Product Safety.

Compliance was demonstrated to the following specifications as listed in the Official Journal of the European Communities:

EN 50081-1 Emissions

EN 55011 Class B Radiated and Conducted Emissions

EN 55082-1 Immunity

IEC 10004-2 Electrostatic Discharge Immunity

IEC 10004-3 RF Electromagnetic Field Immunity

IEC 10004-4 Electrical Fast Transient/Burst Immunity

Low Voltage Directive 73/23/EEC

CAUTION

FOR RESEARCH USE ONLY NOT FOR CLINICAL USE ON PATIENTS

Introduction

The ECM[®] 630 is a new state of the art exponential decay wave electroporation system designed for in vitro and in vivo electroporation applications. This system incorporates features that make it the most technically advanced exponential decay electroporation system currently available.

ECM[®] 630 features include:

- The generator utilizes the new BTX Power Platform Technology design and novel digital user interface.
- The revolutionary Precision Pulse^{*} System provides the researcher unparalleled power in controlling the time constant.
- With the ability to deliver a maximum of 6000 A in the Low Voltage Mode, the ECM[®] 630 is the most powerful generator in its class.
- Voltage range of 10 V to 500 V with 1 V resolution and 1 $\mu F,$ 25 μF to 3275 μF in 25 μF increments. 25 \Box to 1575 \Box , 25 \Box resolution with "none" setting.
- Voltage range of 50 V to 2500 V with 5 V resolution and either 25 μF or 50 $\mu F.$ 25 \Box to 1575 \Box with 25 \Box resolution.
- Over 200 ECM[®] 600 protocols may be duplicated with this instrument.
- The additional ECM[®] 630 resistor selection "none" will allow researchers to reproduce protocols from competitive systems lacking resistor settings or reporting "unlimited" resistance.
- The ECM* 630 will perform the widest range of electroporation applications among commercially available electroporation generators.

Quick Start

Installation

- 1. Install on a bench or work table.
- **2.** Allow a 1 to 2 inch clearance for proper cooling. It is normal for the instrument to be slightly warmer than its' operating environment.
- 3. Choose an outlet that is readily accessible.

Connecting

- Insert female end of power cord into male power interface on the back panel of the ECM[®] 630.
- 2. Plug male end of power cord into appropriate electrical outlet.
- Insert male banana plugs of the 630 B Safety Stand or alternative electrode device into High Voltage output located on front panel of the ECM* 630.

Initializing

- 1. Push the power switch located on the front panel of the ECM^{\ast} 630.
- **2.** The ECM[®] 630 will go through a series of self-test algorithms to test generator functionality.
- 3. The display will flash:



4. Following this initialization screen, the first time the instrument is initialized, the factory default display will then read:

SET PARA	METERS
VOLTAGE :	02000 LU
RESISTOR :	NONE
CHPHCITUR:	SZYOPH 4

 The first page of the Set Parameter Screen from the last time the ECM[®] 630 was used will be displayed each time the ECM[®] 630 is powered up after this initial start up.



Instrument Controls



Power Switch

- **1.** Electronic Power Switch located on the lower right front panel.
- 2. Press once to initialize the ECM* 630 and once more to turn off.



CAUTION: A stabilization period of 3 seconds is required after connecting to an outlet.

Pulse Switch

- The electronic Pulse Switch on the upper left front panel is activated in the "ready" mode when the Set Parameters screen is displayed.
- Once the start switch is activated, the generator will charge the capacitors to the preset voltage, then automatically deliver the pulse.
- **3.** The maximum charge time is line voltage dependent and will typically be less than 10 seconds.
- A pulse sequence may be aborted by pressing the Pulse switch a second time, before the charging is completed.
- Following the delivery of a pulse, the Pulse switch can be pressed once to leave the feedback mode and return to the "ready" mode.

Parameter Control Knob

The Parameter Control Knob is a rotary encoder controlling both the parameter under control and the value of the parameter under control.

- 1. The display will indicate which parameter is under control by the presence of an arrow to the left of the parameter.
- To select a parameter to adjust, rotate the knob until the arrow is to the left of the desired parameter, then push to select. The arrow will move to the right of the value displayed for that parameter.
- 3. To adjust the value of a parameter under control, rotate the knob clockwise to increase the value and counter clockwise to decrease it. Once values are adjusted, push the knob to lock settings and arrow will return to left of parameters under control.
- In order to move between screens, move the cursor to the bottom of the screen and rotate the knob clockwise to move to the next screen.
- **5.** In order to move to the previous screen, move the cursor to the top of the screen and rotate the knob counterclockwise.

Display

The ECM* 630 Display will show two possible screens, the Set Parameter Screen and the Pulse Delivered screen as well as various Status Messages.

Set Parameter Screen

- 1. The Set Parameters screen consists of two pages showing "Set Parameters" on the first line of each page.
- The Voltage, Resistor (Resistance), and Capacitor (Capacitance) are shown on the first page and the Advanced Features including Save, View and Load are shown on the second page of the Set Parameters screen.
- **3.** The ECM* 630 will beep when toggling from one page to another.

Voltage

- **1.** Voltage indicates the set voltage in volts.
- The low voltage mode range is 10 V to 500 V in 1V increments and the high voltage mode range is 50 V to 2500 V in 5 V increments.
- **3.** The mode is indicated by either LV for low voltage or HV for high voltage after the voltage value.
- 4. A beep is heard when transitioning between LV and HV modes..







Resistor

- **1.** Resistor indicates the set resistance in \Box .
- **2.** The resistance range is 25 □ to 1575 □ in 25 □ increments.
- 3. In the LV mode there is an additional resistor selection of "None". This indicates that there is no timing resistor in the pulsing circuit, thus mimicking settings for competitive electroporators in which there is either no resistor to select, or the resistance is termed "unlimited."

Capacitor

- 1. Capacitor indicates the set capacitance in microfarads (µF).
- In the LV mode the capacitance range is 1μF, 25 to 3275 μF in 25 μF increments.
- 3. In the HV mode the capacitance choice is 25 μ F and 50 μ F.
- **4.** The capacitance and resistance control the time constant. Please refer to Appendix B for explanation and time constant charts.

Advanced Features

- 1. The second Set Parameters screen enables the user to save, view, and load up to three programs. Please note that current parameters will be overwritten once a program is loaded.
- **2.** A default program is used to initialize the system. Program 1 is used to automatically store changes in parameters set each time the system is pulsed.
- **3.** Programs #2 and #3 can be used to set and save experimental parameters for instantaneous use.

Save

- **1.** To save the current set parameters as a program, push the parameter control knob to move the arrow to the left of the program number.
- Rotate the knob to designate the appropriate program number, then push and hold the knob in, releasing after a confirmation screen is displayed.
- The confirmation screen will read "Saved Current Parameters to Set N"(where N is a number from 1 to 3).
- 4. Push the parameter control knob again to return to the Set Parameters screen.

RESISTOR: THE VALUE OF THE TIMING RESISTANCE. TIME CONSTANT Y=RC



View

- **1.** To view the appropriate program parameters, push the parameter control knob to move the arrow to the left of the program number.
- 2. Select the appropriate program number by rotating the parameter control knob, then hold the knob in, releasing after a new screen is displayed.
- **3.** The new screen will display the parameters currently stored under that program number.
- **4.** Push the parameter control knob again to return to the Set Parameters screen.

Load

- To load a saved program, push the parameter control knob to move the arrow to the left of the program number. Please note that in addition to three available programs, there is also the default program as outlined in the "Initializing" section.
- 2. Rotate the knob to change the program number.
- Push and hold the knob in, releasing after a confirmation screen is displayed. The confirmation screen will read "Loaded Set N to Current Parameters" (where N is a number from 1 to 3).
- **4.** Push the parameter control knob again to return to the Set Parameters screen.

On-Line Help

- 1. In the Set Parameter Mode, an On-Line Help function is available. On-Line Help provides a definition for all set parameters and advanced functions.
- **2.** To use On-Line Help, rotate the parameter control knob so that the arrow is to the left of the parameter or feature of interest (not the value of the parameter).
- **3.** Push the parameter control knob in and hold until the definition is displayed.
- 4. Push a second time to return to the Set Parameters screen.

Mode

Low voltage or high voltage range mode (display only, this function is controlled by the voltage parameter)

Voltage

Peak amplitude of the output pulse

Operation: Getting Started

Carefully open the box containing the ECM* 630 Electroporation System. Verify receipt of the following items:

ECM[®] 630 Pulse Generator (1)

Power Cord (1)

Model 630E Electronic Manual (1)

BTX Electroporation Systems may be customized with the addition of various electrodes and accessories. The following items complete a typical system order:

Model 630B Electroporation Safety Stand (1)

Model 660 Cuvette Rack (1)

Model 610 BTX Cuvettes Plus (10)

Model 620 BTX Cuvettes Plus (10)

Model 640 BTX Cuvettes Plus (10)

If you have ordered alternative or different items, please verify their receipt.

Capacitor

The value of the timing capacitance. Time constant t=RC

Resistor

The value of the timing resistance. Time constant t=RC

Save

Save current setup parameters to nonvolatile memory

View

Show the setup parameters stored in nonvolatile memory

Load

Load setup parameters from memory to use

Status Messages

Following the initiation of a pulsing sequence, various status messages are displayed. The following status messages may be observed: Charging, Pulsing, and Pulse Aborted During Charging.

Charging

As soon as the start switch is pressed, the "Charging" status message is displayed. The capacitors in the ECM[®] 630 are charged during the duration of this message.

Pulsing

- Once the capacitors have reached the preset voltage level, they are discharged and the "Pulsing" status message is displayed for the duration of the pulse.
- 2. A click or a beep will be heard during the delivery of the pulse. The sound that is heard is a function of the pulse length. At pulse lengths less than 1 to 2 ms, clicks will likely be heard. At pulse lengths at or above 1 to 2 ms beeps should be heard.
- **3.** Following the pulsing status message, the Pulse Delivered Screen is displayed.
- **4.** Press the encoder or the Pulse switch to return to Set Parameters screen.

Pulsing Aborted During Charging

- **1.** If the start switch is activated a second time prior to delivery of a pulse, the pulsing sequence is aborted and the "Pulsing Aborted" status message is displayed.
- **2.** Press the encoder or the pulse switch once to get back to the Set Parameters screen.



PULSING ABORTED

Pulse Delivered Screen

- The Pulse Delivered Screen is displayed following the delivery of a pulse and is indicated by the display "Pulse Delivered" on the top line of the screen.
- 2. Press the knob to return to the Set Parameters Screen.
- **3.** The Pulse Delivered screen displays the monitored peak Voltage and Time Constant on page 1 and the programmed Resistance and Capacitance on page 2. Please note that a pulse will be delivered and monitored, even if there is no output, so do not use this feature to verify that a pulse was delivered to your sample.
- **4.** Always verify that your sample is connected to the HV output.

Voltage

- 1. Voltage indicates the peak voltage delivered. The voltage delivered is affected by the external load. (Sample)
- Using heavy loads (low resistivity) will result in a slightly lower delivered voltage. This effect is normal and will be more pronounced with small capacitor settings.

Time Constant

Time Constant indicates the exponential decay time constant $1/e\ \text{in}$ ms, ms, or s.

P	ULSE DELIVERED	٦
PEAK	VOLTAGE=0201V	
TIME	CONST =063.1ms	
>	NEXT PAGE	



Operating Basics

Use with Safety Stand 630B and BTX Disposable Electroporation Cuvettes Plus

- **1.** Insert the safety stand banana plugs into the HV Output on the front panel of the ECM[®] 630.
- 2. Adjust the distance between the metal contacts using the black roller for your cuvette. (See: 630 B instruction sheet)
- **3.** Press the green power switch to initialize the ECM[®] 630.
- Rotate parameter control knob to move arrow to voltage and push to select. Rotate knob to adjust voltage. Push again to select that voltage.
- **5.** Rotate parameter control knob to move arrow to Resistor and push to select. Rotate knob to adjust resistance. Push again to select that resistance.
- Rotate parameter control knob to move arrow to Capacitor and push to select. Rotate knob to adjust the capacitance. Push again to select that capacitance.
- 7. Prepare sample, pipette into the appropriate BTX Disposable Cuvettes Plus, place the cuvette in the 630B Safety Stand and secure the safety cover.
- **8.** Press the Start button. The ECM* 630 will charge and then deliver the electroporation pulse, while beeping.
- **9.** Process sample. Do not forget to record appropriate parameters as displayed on the Pulse Delivered screens, for documentation purposes.
- **10.** To return to "ready" mode, press the Pulse button.
- **11.** To abort a pulse before delivery, press the Pulse button during the "charging" mode.

Use with Alternative Electrode Chambers and Applications

The ECM[®] 630 may be used to power all BTX electroporation cuvettes and electrodes. Follow the above instructions in conjunction with instructions provided for the specific electrode. Refer to Appendix A for graphical representation of operating ranges of various chambers and applicators with the ECM[®] 630.

Reproducing ECM® 600 Protocols

The ECM® 630 will reproduce all ECM® 600 protocols:

- 1. Set the voltage as outlined in the ECM[®] 600 protocol.
- Calculate the RC time constant without factoring in the external load by multiplying the ECM* 600 protocol resistance and capacitance values and dividing by 1000 to give a result in msec.



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Operating Basics (Continued)

- Refer to Appendix B. Find the capacitance setting from step 2 and read down the column to find a RC time constant that closely matches. Extrapolate the new ECM[®] 630 resistance setting..
- Set the capacitance from the ECM[®] 600 protocol and the new resistance extrapolated from Appendix B.
- **5.** Follow experimental procedures while optimizing the voltage as outlined in Appendix C.

Reproducing Competitive System Protocols

The ECM[®] 630 is capable of reproducing most exponential competitive system protocols.

BioRad Gene Pulser® and Gene Pulser® II Protocols

For protocols using the Capacitance Extender, Capacitance Extender II, or the Capacitance Extender Plus, use the resistance setting "none", reduce the voltage by 50 V and use the capacitance setting outlined in the protocol. For protocols using the Pulse Controller, Pulse Controller II, or Pulse Controller Plus, use a capacitance setting of 25 μ F, reduce the voltage by 50 V and use the resistance settings outlined in the protocol. Optimization of voltage as outlined in Appendix C is recommended. For additional recommendations, please contact BTX Technical Support.

Other Competitive Exponential Decay Electroporator Protocols

Attempt to identify the resistance and capacitance as well as the voltage outlined in the protocol. Match these as closely to settings allowed with the ECM[®] 630. For low voltage protocols

(V </= 500V), if there is no resistance outlined, use the ECM $^{\circ}$ 630 resistance setting "none". For additional recommendations, please contact BTX Technical Support.

Advanced Operation: Programming

- 1. Set parameters as outlined in Operating Basics.
- **2.** Once presetting of parameters has been completed, rotate the parameter control knob until the save function has been reached.
- Press knob in and release. Now rotate to select which program number the chosen settings (see "Preset Parameters" above) should be saved under. Program #1 is reserved for current parameters in active use and cannot be used for pre-set storage.
- Push and hold knob in, releasing only after a new screen reading "Saved Current Parameters to Set N" is displayed. (N refers to program number designated in step 2)
- 5. Push the parameter control knob again to return to the Set Parameter Screen.
- **6.** From the Set Parameter Screen, use the parameter control knob to rotate until the load function has been reached.
- 7. Push the knob in and release. Now rotate to select the appropriate program number.
- 8. Push and hold knob in, releasing only after a new screen reading "Loaded Set N to Current Parameters" is displayed.
- **9.** Push the parameter control knob again to return to the Set Parameter Screen.

Electroporation

Electroporation is the application of controlled, pulsed electric fields to biological systems. If the biological system contains a lipid bilayer, such as is the case if the system is a suspension of cells or liposomes, the pulsed electric field may overcome the field potential of the lipid bilayer, resulting in a reversible breakdown of the bilayer and a resulting formation of temporal pores in the membrane. The pores formed are of the order of 40 to 120 nm. Most pores reseal within a few seconds, after allowing the transfer of materials into and out of the cells.

During a typical electroporation process, target cells and molecules are mixed together. When an electroporation pulse is delivered, the result is the formation of temporal pores. Before the pores reseal, the target molecules are observed to enter the cells. Upon resealing of the pores, the molecules become incorporated within the cell. The eventual target site depends on the application; for example, molecules can remain in the cytoplasm, interact with the membrane, and move into the nucleus.

Applications for electroporation include permeabilization of virtually all cells to a wide variety of molecules and ions. The most common applications for electroporation are the transformation or transfection of cells with DNA or RNA. Other applications for electroporation include electroactivation, electroinsertion of proteins into cell membranes and electroextraction of molecules from cells. Although electroporation has mainly been used as a research tool, recent work has demonstrated its potential for clinical applications. Some areas being explored include:

- electrochemotherapy which involves electroporation for delivering chemotherapeutic agents directly to tumor cells
- encapsulation of drugs/genes into cells for their use as carrier systems
- · transdermal delivery of drugs/genes
- gene therapy and delivery of drugs/genes with an electroporation catheter.

Electroporation can be characterized by waveform. BTX exponential decay waveform generators, such as the BTX ECM* 399 and ECM* 630 deliver an exponentially decaying pulse. The length of such a discharge waveform is commonly characterized by the time required for the initial voltage to decay to 1/e (roughly 1/3) of the initial value. To achieve a desired pulse length, appropriate resistance and capacitance must be selected on the instrument. Voltage may be directly set on the instrument.



Pore Formation



Electroporation Process



Exponential Decay Wave

Applications

Electroporation

Bacteria and Yeast Electroporation

The most common application is transformation. Field strength and pulse length are critical parameters for reporting, optimization and troubleshooting bacterial and yeast applications.

Mammalian Cell Electroporation

Electroporation has been used successfully to introduce many different molecule types into cells. Most commonly, electroporation is used for the processes of transfection, in which nucleic acid (DNA and RNA), is introduced into cells.

Electroporation can be used to deliver oligonucleotides into cells for gene silencing or anti-sense applications. It can be used to deliver proteins into cells, even large enzymes such as restriction enzymes and antibodies, for various purposes. Peptides have also been electroincorporated. Smaller molecules have been incorporated into cells and liposomes, such as dyes, sugars and dNTP'S.

Electroporation has been used to study cellular activation processes, by electropermeabilizing cells to Ca²⁺, Mg²⁺ and Na⁺. Electroporation is also used to electroinsert proteins into the cell membrane. Finally, electroporation has been used to introduce drugs, such as the chemotherapeutic agent bleomycin, into cancer cells, in vitro and in vivo.

The use of low impedance buffers such as PBS may result in a voltage drop so that the actual peak voltage delivered to samples may be less than the set voltage.

With exponential decay generators, monitoring is necessary to identify the pulse length, or time constant, since this parameter may be very much dependent on the impedance of the sample (sample load).

When using complex and custom electroporation applicators and chambers, the electroporation waveform may be altered and monitoring is again strongly recommended.

Molecules Introduced by Electroporation

- DNA
- RNA
- dNTPS
- Enzymes
- Antibodies
- Other Proteins
- Peptides
- Dyes
- Sugars
- lons
- Other Molecules



High GFP expression in Mouse PE501

Applications (Continued)

Plant Protoplast Electroporation

Electroporation has been used to introduce molecules into plant protoplasts, pollen and most recently, direct transfer into plant tissue (in vivo).

Other Electroporation Applications

- **1.** Transgene incorporation, in which simple transfection of fish embryos has resulted in transgenic zebrafish.
- **2.** Utilization of sperm as biological DNA carriers, in which pulsed fields cause the complexing of DNA to sperm, which then act as carriers upon fertilization.
- **3.** Acrosome enhancement in which an exponential decay pulse enhances the acrosome reaction and facilitates fertilization.
- **4.** Embryonic Stem Cell Chimeras, in which embryonic stem cell transfection, followed by micromanipulation into host blastomere, has resulted in chimeric mice.
- Parthogenesis, in which a repetitive DC pulse stimulates an unfertilized egg to activate and divide as if fertilized, resulting in haploid and diploid embryos.

Appendix A: ECM[®] 630 Electrode Operation Ranges

Figures 1 - 6 display expected operating ranges for the ECM® 630 with various chambers and electrodes. The data lines in the following figures represent the arcing boundaries for the given electrode/chamber model, electroporation media, volume, and voltage/mode. The area at and above each line represents 100% arcing probability. The area beneath each line represents parameters that may lead to the delivery of a full pulse. This data was empirically determined in the BTX Application Laboratory.



Figure 1 Operating Range for the ECM[®] 630 with Model 620 in LV with PBS at various volumes



Figure 2

Operating Range for the ECM[®] 630 with Model 640 in LV with PBS at various volumes



4 mm gap cuvette 20 to 500 V 25 µsec to 70 msec

Appendix A: ECM[®] 630 Electrode Operation Ranges (Continued)









Figure 4 Operating Range for the ECM[®] 630 with PP35 in LV with PBS at various volumes



Figure 5 Operating Range for Model 610/H₂0/HV

Petri Pulser 20 to 400 V 10 µsec to 40 msec

1 mm gap cuvette 50 to 2500 V 680 μsec to 36 msec

Appendix A: ECM[®] 630 Electrode Operation Ranges (Continued)





2 mm gap cuvette 50 to 2500 V 660 µsec to 38 msec

Appendix B: ECM[®] 630 RC Time Constants

SEE PAGES 28 - 38

25 50 75 100 125 150 175 200 225 250 275 25.00 0.025 0.625 1.25 1.875 2.5 3 125 3.75 4 375 5 5.625 6.25 6.875 50.00 0.05 1.25 2.5 3.75 6.25 7.5 8.75 10 11.25 12.5 13.75 75.00 7.5 0.075 1.875 3.75 5.625 9.375 11.25 13.125 16.875 18.75 20.625 15 100.00 0.1 25 75 10 12.5 15 17.5 20 22.5 25 27.5 5 125.00 0.125 3.125 6.25 9.375 12.5 15.625 18.75 21.875 25 28.125 31.25 34.375 150.00 7.5 0.15 3 75 11.25 15 18.75 22.5 26.25 30 33 75 37.5 41.25 26.25 175.00 0.175 4.375 8.75 13.125 17.5 21.875 30.625 35 39.375 43.75 48.125 200.00 0.2 10 15 20 25 30 40 55 5 35 45 50 225.00 0.225 5 625 11 25 16 875 22.5 28 125 33 75 39 375 45 50 625 56 25 61 875 250.00 0.25 6.25 12.5 18.75 25 31.25 37.5 43.75 50 56.25 62.5 68.75 275.00 0 275 6 875 13 75 20.625 27.5 34 375 41.25 48 125 55 61.875 68.75 75 625 Resistor (ohm) 52.5 300.00 0.3 7.5 15 22.5 30 37.5 45 60 67.5 75 82.5 325.00 0.325 8.125 16.25 24.375 32.5 40.625 48.75 56.875 65 73.125 81.25 89.375 350.00 0.35 8.75 17.5 26.25 35 43.75 52.5 61.25 70 78.75 87.5 96.25 103.125 375.00 0.375 18.75 37.5 46.875 84.375 9.375 28.125 56.25 65.625 75 93.75 400.00 90 0.4 10 20 30 40 50 60 70 80 100 110 425.00 0.425 10.625 21.25 31.875 42.5 53.125 63.75 74.375 85 95.625 106.25 116.875 450.00 0.45 11.25 22.5 33.75 45 56.25 67.5 78.75 90 101.25 112.5 123.75 475.00 0.475 47.5 106.875 130.625 11.875 23.75 35.625 59.375 71.25 83.125 95 118.75 500.00 0.5 12.5 37.5 50 62.5 87.5 100 112.5 137.5 25 75 125 525.00 0.525 13.125 26.25 39.375 52.5 65 625 78.75 91.875 105 118,125 131.25 144.375 550.00 0.55 13.75 27.5 41.25 55 68.75 82.5 95 25 110 123 75 137.5 151 25 575.00 0.575 14.375 28.75 43.125 57.5 71.875 86.25 100.625 115 129.375 143.75 158.125 600.00 0.6 15 30 45 60 75 90 105 120 135 150 165 625.00 0.625 15.625 31.25 46.875 62.5 78.125 93.75 109.375 125 140.625 156 25 171.875 650.00 0.65 16.25 32.5 48.75 65 81.25 97.5 113.75 130 146.25 162.5 178.75 675.00 101 25 185 625 0.675 16.875 33 75 50.625 67.5 84 375 118.125 135 151.875 168.75 700.00 0.7 17.5 35 52.5 70 87.5 105 122.5 140 157.5 175 192.5 725.00 0.725 18.125 36.25 54.375 72.5 90.625 108.75 126.875 145 163,125 181.25 199.375 750.00 0.75 18.75 37.5 56.25 75 93.75 112.5 131.25 150 168 75 187.5 206 25 775.00 0.775 19.375 38.75 58.125 77.5 96.875 116.25 135.625 155 174.375 193.75 213.125 800.00 0.8 20 40 60 80 100 120 140 160 180 200 220 825.00 0.825 20.625 41.25 61.875 82.5 103 125 123.75 144 375 165 185 625 206.25 226.875 850.00 0.85 21.25 42.5 63.75 85 106.25 127.5 148.75 170 191.25 212.5 233.75 875.00 0.875 43.75 87.5 196.875 240.625 21.875 65.625 109.375 131.25 153.125 175 218.75 900.00 0.9 22.5 45 67.5 90 112.5 135 157.5 180 202.5 225 247.5 925.00 0.925 23.125 46.25 69.375 92.5 115.625 138.75 161.875 185 208.125 231.25 254.375 950.00 0.95 23 75 47.5 71.25 95 118.75 142.5 166.25 190 213 75 237 5 261 25 975.00 0.975 24.375 48.75 73.125 97.5 121.875 146.25 170.625 195 219.375 243.75 268.125 1000.00 25 50 75 100 125 150 175 200 225 250 275 1025.00 1.025 25.625 51.25 76.875 102.5 128.125 153.75 179.375 205 230.625 256.25 281.875 1050.00 1.05 26.25 52.5 78.75 105 131.25 157.5 183.75 210 236 25 262.5 288.75 1075.00 1 075 26 875 53.75 80,625 107.5 134 375 161.25 188 125 215 241.875 268.75 295 625 1100.00 1.1 27.5 55 82.5 110 137.5 165 192.5 220 247.5 275 302.5 1125.00 253.125 1.125 28.125 56.25 84.375 112.5 140.625 168.75 196.875 225 281.25 309.375 1150.00 1.15 28.75 57.5 86.25 115 143.75 172.5 201.25 230 258.75 287.5 316.25 1175.00 1.175 29.375 58.75 88.125 117.5 146.875 176.25 205.625 235 264.375 293.75 323.125 1200.00 60 90 210 240 270 300 330 12 30 150 180 336.875 1225.00 1.225 30.625 61.25 91.875 122.5 153.125 183.75 214.375 245 275.625 306.25 31.25 281.25 1250.00 1.25 62.5 93.75 125 156.25 187.5 218.75 250 312.5 343.75 1275.00 1.275 31.875 63.75 95.625 127.5 159.375 191.25 223.125 255 286.875 318.75 350.625 1300.00 97.5 357.5 1.3 32.5 65 130 162.5 195 227.5 260 292.5 325 1325.00 1.325 33.125 66.25 99.375 132.5 165.625 198.75 231.875 265 298.125 331.25 364.375 1350.00 1.35 33.75 67.5 101.25 135 168 75 202.5 236 25 270 303 75 337 5 371.25 1375.00 1.375 34.375 68.75 103.125 137.5 171.875 206.25 240.625 275 309.375 343.75 378.125 1400.00 35 105 175 245 280 385 1.4 70 140 210 315 350 1425.00 1.425 35.625 71.25 106.875 142.5 178.125 213.75 249.375 320.625 356.25 391.875 285 1450.00 72.5 217.5 290 398.75 1.45 36.25 108.75 145 181.25 253.75 326.25 362.5 1475.00 1.475 36.875 73.75 110.625 147.5 184.375 221.25 258 125 295 331.875 368.75 405 625 1500.00 1.5 37.5 75 112.5 150 187.5 225 262.5 300 337.5 375 412.5 1525.00 1.525 38.125 76.25 114.375 152.5 190.625 228.75 266.875 305 343.125 381.25 419.375 426.25 1550.00 1.55 38.75 77.5 116.25 155 193.75 232.5 271.25 310 348.75 387.5 1575.00 1.575 78.75 118.125 157.5 196.875 236.25 275.625 315 354.375 393.75 433.125 39.375

Low Voltage Mode RC

300	325	350	375	400	425	450	475	500	525	550	575	600
7.5	8.125	8.75	9.375	10	10.625	11.25	11.875	12.5	13.125	13.75	14.375	15
15	16.25	17.5	18.75	20	21.25	22.5	23.75	25	26.25	27.5	28.75	30
22.5	24.375	26.25	28.125	30	31.875	33.75	35.625	37.5	39.375	41.25	43.125	45
30	32.5	35	37.5	40	42.5	45	47.5	50	52.5	55	57.5	60
37.5	40.625	43.75	46.875	50	53.125	56.25	59.375	62.5	65.625	68.75	71.875	75
45	48.75	52.5	56.25	60	63.75	67.5	71.25	75	78.75	82.5	86.25	90
52.5	56.875	61.25	65.625	70	74.375	78.75	83.125	87.5	91.875	96.25	100.625	105
60	65	70	75	80	85	90	95	100	105	110	115	120
67.5	73.125	78.75	84.375	90	95.625	101.25	106.875	112.5	118.125	123.75	129.375	135
75	81.25	87.5	93.75	100	106.25	112.5	118.75	125	131.25	137.5	143.75	150
82.5	89.375	96.25	103.125	110	116.875	123.75	130.625	137.5	144.375	151.25	158.125	165
90	97.5	105	112.5	120	127.5	135	142.5	150	157.5	165	172.5	180
97.5	105.625	113.75	121.875	130	138.125	146.25	154.375	162.5	170.625	178.75	186.875	195
105	113.75	122.5	131.25	140	148.75	157.5	166.25	175	183.75	192.5	201.25	210
12.5	121.8/5	131.25	140.625	150	159.375	168.75	178.125	187.5	196.875	206.25	215.625	225
120	130	140	150	160	170	180	190	200	210	220	230	240
27.5	138,125	148.75	159.375	170	180.625	191.25	201.875	212.5	223.125	233.75	244.375	255
135	146.25	157.5	165.75	180	191.25	202.5	213.75	225	236.25	247.5	258.75	270
42.5	154.375	106.25	178.125	190	201.8/5	213.75	225.625	237.5	249.375	201.25	2/3.125	285
100	170 605	1/0	107.3	200	212.5	223	237.5	200	202.5	2/3	201.0	300
165	170.025	183.75	190.875	210	223.125	236.25	249.375	202.5	2/3.025	208.75	301.875	315
105 72 F	1/0./0	192.0	200.20	220	233.73	247.3	201.20	2/3	200.75	302.5	310.23	330
190	100.075	201.25	215.625	230	244.375	230.75	273.125	207.0	01.075	310.25	330.025	345
100	202 125	019.75	220	240	200	2/0	200	210.5	200 405	242 75	250 275	300
105	203.125	210.75	234.375	200	203.025	201.20	290.075	312.3	941.95	343.75	979.75	373
12.5	210 375	227.5	243.75	200	286.975	202.0	320.625	937.5	341.20	371.95	398 125	405
2.5	213.375	230.25	203.125	220	200.675	303.75	320.025	357.3	367.5	995	402.5	405
17.5	235 625	259.75	271 875	200	308 125	326.25	344 375	362.5	380.625	308 75	416 875	420
995	943 75	282.5	291.25	300	318 75	337.5	356.95	975	303.75	412.5	431.25	450
125	243.75	271.25	201.25	310	329 375	348 75	368 125	387.5	408 875	428.25	45 625	465
240	260	280	300	320	340	360	380	400	420	440	460	480
47.5	268 125	288 75	309 375	330	350 625	371.25	391.875	412.5	493 125	453 75	474 375	400
255	278 25	297.5	318.75	340	361.25	382.5	403.75	425	446 25	467.5	488.75	510
62.5	284.375	306.25	328.125	350	371.875	393.75	415.625	437.5	459.375	481.25	503.125	525
270	292.5	315	337.5	360	382.5	405	427.5	450	472.5	495	517.5	540
77.5	300.625	323.75	346.875	370	393,125	416.25	439.375	462.5	485.625	508.75	531.875	555
285	308.75	332.5	356.25	380	403.75	427.5	451.25	475	498.75	522.5	546.25	570
292.5	316.875	341.25	365.625	390	414.375	438.75	463.125	487.5	511.875	536.25	560.625	585
300	325	350	375	400	425	450	475	500	525	550	575	600
307.5	333.125	358.75	384.375	410	435.625	461.25	486.875	512.5	538.125	563.75	589.375	615
315	341.25	367.5	393.75	420	446.25	472.5	498.75	525	551.25	577.5	603.75	630
322.5	349.375	376.25	403.125	430	456.875	483.75	510.625	537.5	564.375	591.25	618.125	645
330	357.5	385	412.5	440	467.5	495	522.5	550	577.5	605	632.5	660
37.5	365.625	393.75	421.875	450	478.125	506.25	534.375	562.5	590.625	618.75	646.875	675
345	373.75	402.5	431.25	460	488.75	517.5	546.25	575	603.75	632.5	661.25	690
352.5	381.875	411.25	440.625	470	499.375	528.75	558.125	587.5	616.875	646.25	675.625	705
360	390	420	450	480	510	540	570	600	630	660	690	720
867.5	398.125	428.75	459.375	490	520.625	551.25	581.875	612.5	643.125	673.75	704.375	735
375	406.25	437.5	468.75	500	531.25	562.5	593.75	625	656.25	687.5	718.75	750
382.5	414.375	446.25	478.125	510	541.875	573.75	605.625	637.5	669.375	701.25	733.125	765
390	422.5	455	487.5	520	552.5	585	617.5	650	682.5	715	747.5	780
397.5	430.625	463.75	496.875	530	563.125	596.25	629.375	662.5	695.625	728.75	761.875	795
405	438.75	472.5	506.25	540	573.75	607.5	641.25	675	708.75	742.5	776.25	810
112.5	446.875	481.25	515.625	550	584.375	618.75	653.125	687.5	721.875	756.25	790.625	825
420	455	490	525	560	595	630	665	700	735	770	805	840
127.5	463.125	498.75	534.375	570	605.625	641.25	676.875	712.5	748.125	783.75	819.375	855
435	471.25	507.5	543.75	580	616.25	652.5	688.75	725	761.25	797.5	833.75	870
42.5	479.375	516.25	553.125	590	626.875	663.75	700.625	737.5	774.375	811.25	848.125	885
450	487.5	525	562.5	600	637.5	675	712.5	750	787.5	825	862.5	900
57.5	495.625	533.75	571.875	610	648.125	686.25	724.375	762.5	800.625	838.75	876.875	915
465	503.75	542.5	581.25	620	658.75	697.5	736.25	775	813.75	852.5	891.25	930
472 5	511.975	551 25	590.625	630	669 375	708 75	748 125	787 5	826 875	R66 25	005 695	945

	825	650	875	700	725	750	775	800	825	850	875	900
	15.625	16.25	16.875	17.5	18,125	18.75	19.375	20	20.625	21.25	21.875	22.5
	31.25	32.5	33.75	35	36.25	37.5	38 75	40	41.25	42.5	43.75	45
	46.875	48.75	50.625	52.5	54.375	56.25	58.125	60	61.875	63.75	65.625	67.5
	62.5	00	67.5	70	72.5	75	77.5	80	82.5	85	87.5	90
	78.125	81.25	84.375	87.5	90.625	93.75	96.875	100	103.125	106.25	109.375	112.5
	93.75	97.5	101.25	105	108.75	112.5	116.25	120	123.75	127.5	131.25	135
	109.375	113.73	116.120	122.0	120.070	131.20	130.020	140	144.3/0	148.70	103.120	107.0
	140 625	146.26	151 975	167.6	140	100 76	174 976	190	100	101.06	1/0	202.6
	156.25	169.5	189.75	137.5	181.05	100.75	193.75	200	206.025	010.5	218.75	0.505
-	171 875	178 75	185 625	102.5	100 975	206.25	213 125	200	200 23	293.75	240.625	247.5
E	187.5	195	202.5	210	217.5	200.25	232.5	240	247.5	255	262.5	270
5	203 125	211 25	219 375	227 5	235 625	243 75	251 875	260	268 125	276 25	284 375	292.5
2	218.75	227.5	236.25	245	253.75	262.5	271.25	280	288.75	297.5	306.25	315
ğ	234 375	243.75	253 125	262.5	271.875	281.25	290.625	300	309 375	318.75	328,125	337.5
N.	250	260	270	280	290	300	310	320	330	340	350	360
es	265 625	276.25	286 875	297.5	308,125	318.75	329 375	340	350.625	361.25	371.875	382.5
E	281.25	292.5	303.75	315	326.25	337.5	348.75	360	371.25	382.5	393.75	405
	296.875	308.75	320.625	332.5	344.375	356.25	368.125	380	391.875	403.75	415.625	427.5
	312.5	325	337.5	350	362.5	375	387.5	400	412.5	425	437.5	450
	328.125	341.25	354.375	367.5	380.625	393.75	406.875	420	433.125	446.25	459.375	472.5
	343.75	357.5	371.25	385	398.75	412.5	426 25	440	453.75	467.5	481.25	495
	359 375	373.75	388.125	402.5	416.875	431.25	445.625	460	474.375	488.75	503 125	517.5
	375	390	405	420	435	450	465	480	495	510	525	540
	390.625	406.25	421.875	437.5	453,125	468.75	484.375	500	515.625	531.25	546.875	562.5
	406.25	422.5	438.75	455	471.25	487.5	503.75	520	536.25	552.5	568.75	585
	421.875	438.75	455.625	472.5	489.375	506.25	523.125	540	556.875	573.75	590.625	607.5
	437.5	455	472.5	490	507.5	525	542.5	560	577.5	595	612.5	630
	453 125	471.25	489.375	507.5	525,625	543.75	561.875	580	598.125	616.25	634.375	652.5
	468.75	487.5	506.25	525	543.75	562.5	581.25	600	618 75	637.5	656.25	675
	484.375	503.75	523.125	542.5	561.875	581.25	600.625	620	639.375	658.75	678.125	697.5
	500	520	540	560	580	600	520	640	660	680	700	720
	515.625	536.25	556.875	577.5	598,125	618.75	639.375	660	680.625	701.25	721.875	742.5
	531.25	552.5	573.75	595	616.25	637.5	658.75	680	701.25	722.5	743.75	765
	546.875	558.75	590.625	612.5	634.375	656.25	678.125	700	/21.8/5	743.75	765.625	787.5
	562.5	585	607.5	630	652.5	675	697.5	720	742.5	765	787.5	810
	5/6.125	617.6	024.375	047.5	070.020	710.5	710.875	740	703.125	100.25	001.05	832.5
	093.75	017.3	091.23	600	705 975	712.3	730 23	700	703.75	007.3	65 106	977 6
	805.375	035.75	030.125	700	700.075	751.25	755.025	200	004.075	020.75	030.123	0/7.5
	640 625	666.25	801 975	717.5	743 125	769 75	70/ 375	800	845 625	871.25	906 975	022.5
	656 25	892.5	708.75	736	761.25	787.5	813 75	840	866 25	802.5	918 75	945
	871 875	698.75	725 625	752.5	779 375	806.25	833 125	860	886 875	913.75	940.625	987.5
	687.5	715	742 5	770	797.5	825	852.5	880	907.5	935	982.5	990
	703 125	731.25	759 375	787.5	815 625	843.75	871.875	900	828 125	956.25	984,375	1012.5
	718.75	747.5	776.25	805	833.75	862.5	891.25	920	948.75	977.5	1006.25	1035
	734.375	763.75	793.125	822.5	851.875	881.25	910.625	940	969.375	998.75	1028.125	1057.5
	750	780	810	840	870	900	930	960	990	1020	1050	1080
	765.625	796.25	828.875	857.5	888.125	918.75	949.375	080	1010.625	1041.25	1071.875	1102.5
	781.25	812.5	843.75	875	906.25	937.5	968.75	1000	1031.25	1062.5	1093 75	1125
	796.875	828.75	860.625	892.5	924.375	956.25	988.125	1020	1051.875	1083.75	1115.625	1147.5
	812.5	845	877.5	910	942.5	975	1007.5	1040	1072.5	1105	1137.5	1170
	828.125	861.25	894.375	927.5	960,625	993.75	1026.875	1060	1093.125	1126.25	1159.375	1192.5
	843.75	877.5	911.25	945	978.75	1012.5	1046.25	1080	1113.75	1147.5	1181.25	1215
	859.375	893.75	928.125	962.5	996.875	1031.25	1065.625	1100	1134.375	1168.75	1203.125	1237.5
	875	910	945	980	1015	1050	1085	1120	1155	1190	1225	1260
	890.625	926.25	961.875	997.5	1033.125	1068.75	1104.375	1140	1175.625	1211.25	1246.875	1282.5
	906.25	942.5	978.75	1015	1051.25	1087.5	1123.75	1160	1196.25	1232.5	1268.75	1305
	921.875	958.75	995 625	1032.5	1069.375	1106.25	1143.125	1180	1216.875	1253.75	1290.625	1327.5
	937.5	975	1012.5	1050	1087.5	1125	1162.5	1200	1237.5	1275	1312.5	1350
	953.125	991.25	1029.375	1067.5	1105.625	1143.75	1181.875	1220	1258.125	1296.25	1334.375	1372.5
	968.75	1007.5	1046.25	1085	1123.75	1162.5	1201.25	1240	1278.75	1317.5	1356.25	1395
	100.0 222	11112 10	1/162 1/16	11/11/16	1141 976	5101 26	1 2 1 1 1 1 1 1 1	1.160	1 2010 276	1222 76	1979 196	14176

Low Voltage Mode RC

19920		10000	1.000	1. And the second second		1. S.	1.	AND NO.			CONSIST.		
925	950	975	1000	1025	1050	1075	1100	1125	1150	1175	1200	1225	1250
46.25	47.5	48.75	50	51.25	52.5	53.75	55	56.25	57.5	58.75	60	61.25	62.5
69 375	71.25	73 125	75	76 875	78 75	80.625	82.5	84 375	86.25	88 125	90	91 875	93.75
92.5	95	97.5	100	102.5	105	107.5	110	112.5	115	117.5	120	122.5	125
115.625	118.75	121.875	125	128 125	131.25	134 375	137.5	140.625	143.75	146.875	150	153 125	156.25
138.75	142.5	146.25	150	153 75	157.5	161.25	165	168.75	172.5	176.25	180	183.75	187.5
161.875	166.25	170.625	175	179.375	183.75	188,125	192.5	196.875	201,25	205.625	210	214.375	218.75
185	190	195	200	205	210	215	220	225	230	235	240	245	250
208.125	213.75	219.375	225	230.625	236.25	241.875	247.5	253.125	258.75	264.375	270	275.625	281.25
231.25	237.5	243.75	250	258.25	262.5	268.75	275	281.25	287.5	293.75	300	306.25	312.5
254.375	261.25	268 125	275	281 875	288.75	295.625	302.5	309.375	316.25	323.125	330	336.875	343.75
277.5	285	292.5	300	307.5	315	322.5	330	337.5	345	352.5	360	367.5	375
300.625	308.75	316.875	325	333.125	341.25	349.375	357.5	365.625	373.75	381.875	390	398.125	406.25
323.75	332.5	341.25	350	358.75	367.5	376.25	385	393.75	402.5	411.25	420	428.75	437.5
346.875	356.25	365.625	375	384.375	393.75	403.125	412.5	421.875	431.25	440.625	450	459.375	468.75
370	380	390	400	410	420	430	440	450	460	470	480	490	500
393,125	403,75	414.375	425	435.625	446.25	456.875	467.5	478.125	488.75	499.375	510	520.625	531.25
416.25	427.5	438.75	450	461.25	472.5	483.75	495	506.25	517.5	528.75	540	551.25	562.5
439.375	451.25	463.125	475	486.875	498.75	510.625	522.5	534.375	546.25	558.125	570	581.875	593.75
462.5	475	487.5	500	512.5	525	537.5	550	562.5	575	587.5	600	612.5	625
485.825	498.75	511.875	525	538.125	551.25	564.375	577.5	590.625	603.75	616.875	630	643.125	656.25
508.75	522.5	536,25	550	563.75	577.5	591.25	605	618.75	632.5	646.25	660	673.75	687.5
531.875	546.25	560 625	575	589.375	603.75	618.125	632.5	646.875	661.25	675.625	690	704.375	718.75
555	570	585	600	615	630	645	660	675	690	705	720	735	750
578.125	593.75	609.375	625	640.625	656.25	671.875	687.5	703.125	718.75	734.375	750	765.625	781.25
601.25	617.5	633.75	650	666.25	682.5	698.75	715	731.25	747.5	763.75	780	796.25	812.5
624.375	641.25	658.125	675	691.875	708.75	725,625	742.5	759.375	776.25	793.125	810	826.875	843.75
647.5	665	682.5	700	717.5	735	752.5	770	787.5	805	822.5	840	857.5	875
670.625	688.75	706.875	725	743.125	761.25	779.375	797.5	815.625	833.75	851.875	870	888.125	906.25
693.75	712.5	731.25	750	768.75	787.5	806.25	825	843.75	862.5	881.25	900	918.75	937.5
716.875	736.25	755.625	775	794.375	813.75	833.125	852.5	871.875	891.25	910.625	930	949.375	968.75
740	760	780	800	820	840	860	880	900	920	940	960	980	1000
763.125	783.75	804.375	825	845.625	866.25	886.875	907.5	928.125	948.75	969.375	990	1010.625	1031.25
786.25	807.5	828.75	850	871.25	892.5	913.75	935	956.25	977.5	998.75	1020	1041.25	1062.5
809.375	831.25	853.125	875	896.875	918.75	940.625	962.5	984.375	1006.25	1028.125	1050	1071.875	1093.75
832.5	855	877.5	900	922.5	945	967.5	990	1012.5	1035	1057.5	1080	1102.5	1125
855.625	878.75	901.875	925	948.125	971.25	994.375	1017.5	1040.625	1063.75	1086.875	1110	1133.125	1156.25
878.75	902.5	926.25	950	973.75	997.5	1021.25	1045	1068.75	1092.5	1116.25	1140	1163.75	1187.5
901.875	926.25	950.625	975	999.375	1023.75	1048.125	1072.5	1096.875	1121.25	1145.625	1170	1194.375	1218.75
925	950	975	1000	1025	1050	1075	1100	1125	1150	1175	1200	1225	1250
948.125	973.75	999.375	1025	1050.625	1076.25	1101.875	1127.5	1153.125	1178.75	1204.375	1230	1255.625	1281.25
971.25	997.5	1023.75	1050	1078.25	1102.5	1128.75	1155	1181.25	1207.5	1233.75	1260	1286.25	1312.5
994.375	1021.25	1048.125	1075	1101.875	1128.75	1155.625	1182.5	1209.375	1236.25	1263 125	1290	1316.875	1343.75
1017.5	1045	1072.5	1100	1127.5	1155	1182.5	1210	1237.5	1265	1292.5	1320	1347.5	1375
1040.625	1058.75	1095.875	1125	1153.125	1181.25	1209.375	1237.5	1265.625	1293.75	1321.875	1350	1378.125	1406.25
1063.75	1092.5	1121.25	1150	1178.75	1207.5	1236.25	1265	1293.75	1322.5	1351.25	1380	1408.75	1437.5
1086.875	1116.25	1145.625	1175	1204.375	1233.75	1263.125	1292.5	1321.875	1351.25	1380.625	1410	1439,375	1468.75
1110	1140	1170	1200	1230	1260	1290	1320	1350	1380	1410	1440	14/0	1500
1133.125	1103.75	1194.375	1225	1205.625	1285.25	1316.875	1347.5	1378.125	1408.75	1439.375	1470	1500.625	1531.25
1156.25	1187.5	1218.75	1250	1281.25	1312.5	1343.75	1375	1406.25	1437.5	1468.75	1500	1581.25	1562.5
1179.375	1211.25	1243.125	1275	1306.875	1338.75	1370.625	1402.5	1434.375	1466.25	1498.125	1530	1561.875	1593.75
1202.5	1235	1267.5	1300	1332.5	1365	1397.5	1430	1462.5	1495	1527.5	1560	1592.5	1625
1225,625	1258.75	1291.875	1325	1358.125	1391.25	1424.375	1457.5	149.0 625	1523.75	1000.875	1590	1823,125	1056.25
1071 075	1282.5	1316.25	1350	1303.75	1417.5	1451.25	1485	1518.75	1552.5	1586.25	1620	1053.75	1667.5
12/1.875	1306.25	1340 625	13/5	1408.375	1443.75	14/8.125	1512.5	1040 875	1081.25	1015 625	1650	1054.375	1/18./5
1295	1330	1365	1400	1435	14/0	1505	1540	15/5	1610	1645	1680	1/15	1750
1318.125	1353.75	1389.375	1425	1400.025	1495.25	1531.8/5	1567.5	1003.125	1638.75	10/4.3/5	1710	1745.525	1/81.25
1341.25	1377.5	1413.75	1450	1485.25	1522.5	1538.75	1005	1631.25	1007.5	1703.75	1740	1775.25	1012.5
1304.375	1401.25	1438.125	14/5	1637.5	1548.75	1000.625	1022.5	1009 375	1096.25	1733 125	1000	1806.875	1043.75
1307.5	1449.75	1402.0	1500	1000 100	1001.00	1012.5	1000	1715 695	1769.75	1702.5	1990	1000 105	1000.05
1422.76	1479 5	1511.95	1550	1598.75	1627 5	1866.25	1705	1749.76	1792 5	1921.25	1960	1808.75	1027 5
1400.70	1406.05	1011.20	1530	1814 275	1862.75	1602 105	1700 5	1740.75	1011.05	1260 625	1900	1000.75	1069 75
1400.010	1430.23	1000.020	(3/3	1014.070	1043.13	1030.123	11 36.3	1111.0/3	1011.23	1000.020	1020	1363.013	1200.73

1275 1300 1325 1350 1375 1400 1425 1450 1475 1500 1525 1550 1575 31 875 32.5 33 125 33 75 34 375 35 35 625 36.25 36 875 37.5 38 125 38 75 39 375 65 67.5 70 72.5 75 77.5 63.75 66.25 68.75 71.25 73.75 76.25 78.75 95.625 97.5 99.375 101.25 103.125 105 106.875 108.75 110.625 112.5 114.375 116.25 118,125 132.5 135 127.5 130 137.5 140 142.5 145 147.5 150 152.5 155 157.5 159.375 162.5 165.625 168.75 171.875 175 178.125 181.25 184.375 187.5 190.625 193.75 196.875 191.25 195 198.75 202.5 206.25 210 213.75 217.5 221.25 225 228.75 232.5 236 25 223.125 231.875 236.25 240.625 245 249.375 253.75 258.125 262.5 271.25 227.5 266.875 275.625 260 265 270 280 285 290 295 300 310 315 255 275 305 286.875 292.5 298.125 303.75 309.375 315 320.625 326.25 331.875 337.5 343.125 348.75 354.375 337.5 318.75 325 331.25 343.75 350 356.25 362.5 368.75 375 381.25 387.5 393.75 350.625 357.5 364.375 371.25 378.125 385 391.875 398.75 405.625 412.5 419.375 426.25 433,125 Resistor (ohm) 382 5 300 397.5 405 412 5 420 427 5 435 442 5 450 457 5 465 472 5 414.375 422.5 430.625 438.75 446.875 455 463.125 471.25 479.375 487.5 495.625 503.75 511.875 446.25 455 463.75 472.5 481.25 490 498.75 507.5 516.25 525 533.75 542.5 551.25 478.125 487.5 496.875 506.25 515.625 525 534.375 543.75 553.125 562.5 571.875 581.25 590.625 510 520 530 540 550 560 570 580 590 600 610 620 630 541.875 552 5 563.125 573.75 584.375 595 605.625 616.25 626.875 637.5 648 125 658 75 669.375 573.75 585 596.25 607.5 618.75 630 641.25 652.5 663.75 675 686.25 697.5 708.75 605.625 688.75 736.25 617.5 629.375 641.25 653.125 665 676.875 700.625 712.5 724.375 748.125 675 725 637.5 650 662.5 687.5 700 712.5 737.5 750 762.5 775 787.5 669.375 682.5 695.625 708.75 721.875 735 748.125 761.25 774.375 787.5 800.625 813.75 826.875 701.25 715 728.75 742.5 756.25 770 783.75 797.5 811.25 825 838.75 852.5 866.25 747.5 862.5 733.125 761.875 776.25 790.625 805 819.375 833.75 848.125 876.875 891.25 905.625 765 780 795 810 825 840 855 870 885 900 915 930 945 796.875 812.5 828.125 843.75 859.375 875 890.625 906.25 921.875 937.5 953.125 968.75 984.375 828.75 845 861.25 877.5 893.75 910 926.25 942.5 958.75 975 991.25 1007.5 1023.75 911.25 978.75 1012.5 860.625 877.5 894.375 928.125 945 961.875 995.625 1029.375 1046.25 1063.125 910 945 1015 1050 892.5 927.5 962.5 980 997.5 1032.5 1067.5 1085 1102.5 924.375 942.5 960.625 978.75 996.875 1015 1033.125 1051.25 1069.375 1087.5 1105.625 1123.75 1141.875 956 25 975 993.75 1012.5 1031 25 1050 1068 75 1087.5 1106.25 1125 1143 75 1162 5 1181 25 1007.5 1026.875 1046.25 1123.75 1162.5 1201.25 1220.625 988.125 1065.625 1085 1104.375 1143.125 1181.875 1140 1020 1040 1060 1080 1120 1160 1180 1200 1240 1260 1100 1220 1051.875 1072.5 1093.125 1113.75 1134.375 1155 1175.625 1196.25 1216.875 1237.5 1258.125 1278.75 1299.375 1083.75 1105 1126.25 1147.5 1168.75 1190 1211.25 1232.5 1253.75 1275 1296.25 1317.5 1338.75 1115.625 1137.5 1159.375 1181.25 1203 125 1225 1246.875 1268.75 1290.625 13125 1334.375 1356 25 1378.125 1147.5 1170 1192.5 1215 1237.5 1260 1282.5 1305 1327.5 1350 1372.5 1395 1417.5 1202.5 1179.375 1225.625 1248.75 1271.875 1295 1318.125 1341.25 1364.375 1387.5 1410.625 1433.75 1456.875 1211 25 1235 1258 75 1282 5 1306.25 1330 1353 75 1377 5 1401 25 1425 1449.75 1472 5 1496 25 1243.125 1267.5 1291.875 1316.25 1340.625 1365 1389.375 1413.75 1438.125 1462.5 1486.875 1511.25 1535.625 1475 1575 1275 1300 1325 1350 1375 1400 1425 1450 1500 1550 1525 1511.875 1306.875 1332.5 1358.125 1383 75 1409.375 1435 1460 625 1486.25 1537.5 1563.125 1588 75 1614 375 1338.75 1365 1391.25 1417.5 1443.75 1470 1498.25 1522.5 1548.75 1575 1601.25 1627.5 1653.75 1370.625 1397.5 1424.375 1451.25 1478.125 1505 1531.875 1558.75 1585.625 1612.5 1639.375 1666.25 1693.125 1402.5 1430 1457.5 1485 1512.5 1540 1567.5 1595 1622.5 1650 1677.5 1705 1732.5 1434.375 1462.5 1518.75 1575 1687.5 1743.75 1490.625 1546.875 1603.125 1631.25 1659.375 1715.625 1771.875 1466 25 1495 1523 75 1552 5 1581 25 1610 1638 75 1667.5 1696 25 1725 1753 75 1782 5 1811 25 1498.125 1527.5 1556.875 1586.25 1615.625 1645 1674.375 1703.75 1733.125 1762.5 1791.875 1821.25 1850.625 1560 1620 1680 1740 1770 1800 1860 1530 1590 1650 1710 1830 1890 1898.75 1561.875 1592.5 1623.125 1653.75 1684.375 1715 1745.625 1776.25 1806.875 1837.5 1868.125 1929.375 1593.75 1625 1656 25 1687.5 1718.75 1750 1781.25 1812.5 1843.75 1875 1906.25 1937.5 1968 75 1625 625 1657.5 1689 375 1721 25 1753.125 1785 1816.875 1848 75 1880 625 1912 5 1944 375 1976 25 2008 125 1657.5 1690 1722.5 1755 1787.5 1820 1852.5 1885 1917.5 1950 1982.5 2015 2047.5 1722.5 1855 1987.5 2053.75 1689.375 1755.625 1788.75 1821.875 1888.125 1921.25 1954.375 2020.625 2086.875 1721.25 1755 1788.75 1822.5 1856.25 1890 1923.75 1957.5 1991.25 2025 2058.75 2092.5 2126.25 1753.125 1787.5 1821.875 1856.25 1890.625 1925 1959.375 1993.75 2028.125 2062.5 2096.875 2131.25 2165.625 1785 1820 1855 1890 1960 1995 2030 2065 2100 2135 2205 2137.5 1816.875 1852.5 1888.125 1923.75 1959.375 1995 2030.625 2066.25 2101.875 2173.125 2208.75 2244.375 1848.75 1885 1921.25 1957.5 1993.75 2030 2066.25 2102.5 2138,75 2175 2211.25 2247.5 2283.75 1880 625 1917.5 1954.375 1991 25 2028 125 2065 2101.875 2138.75 2175.625 22125 2249 375 2286.25 2323 125 1912.5 1950 1987.5 2025 2062.5 2100 2137.5 2175 2212.5 2250 2287.5 2325 2362.5 1944.375 1982.5 2020.625 2058.75 2096.875 2135 2173.125 2211.25 2249.375 2287.5 2325.625 2363.75 2401.875 2363.75 1976 25 2015 2053 75 2092 5 2131.25 2170 2208.75 2247 5 2286 25 2325 2402 5 2441 25 2008.125 2047.5 2126.25 2244.375 2283.75 2323.125 2401.875 2441.25 2480.625 2086.875 2165.625 2205 2362.5

Low Voltage Mode RC

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	Capacito	or (u⊢)										
1600	1625	1650	1675	1700	1725	1750	1775	1800	1825	1850	1875	1900
40	40.625	41.25	41.875	42.5	43.125	43.75	44.375	45	45.625	46.25	46.875	47.5
80	81.25	82.5	83.75	85	86.25	87.5	88.75	90	91.25	92.5	93.75	95
120	121.875	123.75	125.625	127.5	129.375	131.25	133.125	135	136.875	138.75	140.625	142.5
160	162.5	165	167.5	170	172.5	175	177.5	180	182.5	185	187.5	190
200	203.125	206.25	209.375	212.5	215.625	218.75	221.875	225	228.125	231.25	234.375	237.5
240	243.75	247.5	251.25	255	258.75	262.5	266.25	270	273.75	277.5	281.25	285
280	284.375	288.75	293.125	297.5	301.875	306.25	310.625	315	319.375	323.75	328.125	332.5
320	325	330	335	340	345	350	355	360	365	370	375	380
360	365.625	371.25	376.875	382.5	388,125	393.75	399.375	405	410.625	416.25	421.875	427.5
400	406.25	412.5	418.75	425	431.25	437.5	443.75	450	456.25	462.5	468.75	475
440	446 875	453.75	460.625	467.5	474 375	481.25	488 125	405	501 875	508.75	515 625	522.5
480	487.5	495	502.5	510	517.5	525	532.5	540	547.5	655	562.5	570
400	607.0	400 E20.05	544 975	552.5	517.5	569 76	570 975	540	E02 125	601.05	600.275	617.5
520	520.125	530.25	544.375	502.5	500.025	505.75 C10.5	570.875	000	000.120	001.25	009.375	017.5
550	508.75	5/7.5	586.25	090	603.75	612.5	621.25	030	638.75	647.5	000.25	740.5
600	609.375	618.75	628.125	037.5	040.875	655.25	000.025	0/5	684.375	693.75	703.125	/12.5
640	650	660	670	680	690	700	/10	/20	730	740	750	760
680	690.625	701.25	711.875	722.5	733.125	743.75	754.375	765	775.625	786.25	796.875	807.5
720	731.25	742.5	753.75	765	776.25	787.5	798.75	810	821.25	832.5	843.75	855
760	771.875	783.75	795.625	807.5	819.375	831.25	843.125	855	866.875	878.75	890.625	902.5
800	812.5	825	837,5	850	862.5	875	887.5	900	912.5	925	937.5	950
840	853.125	866.25	879.375	892.5	905.625	918.75	931.875	945	958.125	971.25	984.375	997.5
880	893.75	907.5	921.25	935	948.75	962.5	976.25	990	1003.75	1017.5	1031.25	1045
920	934.375	948.75	963.125	977.5	991.875	1006.25	1020.625	1035	1049.375	1063.75	1078.125	1092.5
960	975	990	1005	1020	1035	1050	1065	1080	1095	1110	1125	1140
1000	1015.625	1031.25	1046.875	1062.5	1078.125	1093.75	1109.375	1125	1140.625	1156.25	1171.875	1187.5
1040	1056.25	1072.5	1088.75	1105	1121.25	1137.5	1153.75	1170	1186.25	1202.5	1218.75	1235
1080	1096.875	1113.75	1130.625	1147.5	1164.375	1181.25	1198.125	1215	1231.875	1248.75	1265.625	1282.5
1120	1137.5	1155	1172.5	1190	1207.5	1225	1242.5	1260	1277.5	1295	1312.5	1330
1160	1178.125	1196.25	1214.375	1232.5	1250.625	1268.75	1286.875	1305	1323.125	1341.25	1359.375	1377.5
1200	1218 75	1237.5	1256.25	1275	1293 75	1312.5	1331.25	1350	1368 75	1387.5	1406.25	1425
1240	1259 375	1278 75	1298 125	1317.5	1336 875	1356.25	1375 625	1395	1414 375	1493 75	1453 125	1472 5
1000	1200.070	1200	1940	1900	1000.070	1400	1420	1440	1414.070	1400	1600	1690
1000	1940 695	1001 00	1201 075	1402.6	1400 105	1449.75	1464 976	1406	1606 606	1502.05	1546 975	1567.5
1000	1201.025	1400.5	1400 75	1402.5	1423.125	1443.75	1404.375	1400	1555.625	1020.20	1540.875	1007.5
1000	1301.25	1402.5	1420.75	1440	1400.25	1457.5	1506.75	1000	1551.25	1072.0	1090.75	1015
1400	1421.875	1443.75	1465.625	1487.5	1509.375	1531.25	1553.125	1575	1596.875	1618.75	1640.625	1662.5
1440	1462.5	1485	1507.5	1530	1552.5	15/5	1597.5	1620	1642.5	1665	1687.5	1/10
1480	1503.125	1526.25	1549.375	1572.5	1595.625	1618.75	1641.875	1665	1688.125	1711.25	1734.375	1757.5
1520	1543.75	1567.5	1591.25	1615	1638.75	1662.5	1686.25	1710	1733.75	1757.5	1781.25	1805
1560	1584.375	1608.75	1633.125	1657.5	1681.875	1706.25	1730.625	1755	1779.375	1803.75	1828.125	1852.5
1600	1625	1650	1675	1700	1725	1750	1775	1800	1825	1850	1875	1900
1640	1665.625	1691.25	1718.875	1742.5	1768.125	1793.75	1819.375	1845	1870.625	1896.25	1921.875	1947.5
1680	1708.25	1732.5	1758.75	1785	1811.25	1837.5	1863.75	1890	1916.25	1942.5	1968.75	1995
1720	1746.875	1773.75	1800.625	1827.5	1854.375	1881.25	1908.125	1935	1961.875	1988.75	2015.625	2042.5
1760	1787.5	1815	1842.5	1870	1897.5	1925	1952.5	1980	2007.5	2035	2062.5	2090
1800	1828.125	1856.25	1884.375	1912.5	1940.625	1968.75	1996.875	2025	2053.125	2081.25	2109.375	2137.5
1840	1868.75	1897.5	1926.25	1955	1983.75	2012.5	2041.25	2070	2098.75	2127.5	2156.25	2185
1880	1909.375	1938.75	1968.125	1997.5	2026.875	2056.25	2085.625	2115	2144.375	2173.75	2203.125	2232.5
1920	1950	1980	2010	2040	2070	2100	2130	2160	2190	2220	2250	2280
1960	1990.625	2021.25	2051.875	2082.5	2113.125	2143.75	2174.375	2205	2235.625	2265.25	2296.875	2327.5
2000	2031 25	2062.5	2093 75	2125	2156 25	2187.5	2218 75	2250	2281 25	2312.5	2343 75	2375
2040	2071.875	2103.75	2135 625	2167.5	2199 375	2231 25	2263 125	2205	2326 875	2358.75	2390.625	2422 5
2080	2112.5	2145	2177.5	2210	2242 5	2275	2307.5	2340	2372 5	2405	2437 5	2470
2120	2153 105	2186.95	2210 975	2252.5	2285 695	2318 75	2351 875	2395	2418 195	2451 25	2484 375	2517 5
2120	0100.120	2100.20	2219.070	2202.0	0000 75	2010.70	2001.073	0400	9469 75	2401.20	0504.05	2017.0
2100	2180.75	0000 75	0000 105	0007.5	2020.70	0402.05	2090.20	2400	2400.75	0540.75	2001.20	2000
2200	2234.375	2208.75	2303.125	2337.5	23/1.8/5	2400.25	2440.025	2475	2009.375	2043.75	25/8.125	2012.5
2240	2275	2310	2345	2350	2415	2450	2485	2520	2555	2590	2625	2660
2280	2315.625	2351.25	2386.875	2422.5	2458.125	2493.75	2529.375	2565	2600.625	2636.25	2671.875	2707.5
2320	2356.25	2392.5	2428.75	2465	2501.25	2537.5	2573.75	2610	2646.25	2682.5	2718.75	2755
2360	2396.875	2433.75	2470 625	2507.5	2544.375	2581.25	2618.125	2655	2691.875	2728.75	2765.625	2802.5
2400	2437.5	2475	2512.5	2550	2587.5	2625	2662.5	2700	2737.5	2775	2812.5	2850
2440	2478.125	2516.25	2554.375	2592.5	2630.625	2668.75	2706.875	2745	2783.125	2821.25	2859.375	2897.5
2480	2518.75	2557.5	2596.25	2635	2673.75	2712.5	2751.25	2790	2828.75	2867.5	2906.25	2945
2620	9550 375	2508 75	2838 125	2877.5	2716 875	2758 25	2705 825	2835	2874 375	2013 75	2052 125	2002 5

	1925	1950	1975	2000	2025	2050	2075	2100	2125	2150	2175	2200	2225
	48.125	48.75	49.375	50	50.625	51.25	51.875	52.5	53.125	53.75	54.375	55	55.625
	96.25	97.5	98.75	100	101.25	102.5	103.75	105	106.25	107.5	108.75	110	111.25
	144.375	146.25	148.125	150	151.875	153.75	155.625	157.5	159.375	161.25	163.125	165	166.875
	192.5	195	197.5	200	202.5	205	207.5	210	212.5	215	217.5	220	222.5
	240.625	243.75	246.875	250	253.125	256.25	259.375	262.5	265.625	268.75	271.875	275	278.125
	288.75	292.5	296.25	300	303.75	307.5	311.25	315	318.75	322.5	326.25	330	333.75
	336.875	341.25	345.625	350	354.375	358.75	363.125	367.5	371.875	376.25	380.625	385	389.375
	385	390	395	400	405	410	415	420	425	430	435	440	445
	433.125	438.75	444.375	450	455.625	461.25	466.875	472.5	478.125	483.75	489.375	495	500.625
	481.25	487.5	493.75	500	506.25	512.5	518.75	525	531.25	537.5	543.75	550	556.25
~	529.375	536.25	543 125	550	556 875	563 75	570 625	577.5	584.375	591.25	598 125	605	611 875
E	577 5	585	502.5	800	607.5	615	622.5	630	837.5	645	852.5	660	667.5
5	625 625	633.75	641.975	650	658 125	666.95	674 375	682.5	600 625	698 75	706 875	715	723 125
9	673 75	692.5	691.25	700	709.75	717.5	728.25	795	749 75	759.5	761.25	710	779 76
p	701 975	701.05	740 605	750	750 976	769.75	720.20	707.5	706 975	000.00	915 695	006	024 975
st	721.075	731.20	740.025	750	759.375	708.75	//6.125	/5/.5	/90.8/5	000.25	015.025	020	034.370
S.	770	/80	790	800	810	020	830	840	005	005	018	000	890
ř	818.125	828.75	839.375	850	860.625	871.25	881.875	892.5	903.125	913.75	924.375	935	945.625
_	865.25	8//.5	888.75	900	911.25	922.5	933.75	945	956.25	967.5	978.75	990	1001.25
	914.375	926.25	938.125	950	961.875	973.75	985.625	897.5	1009.375	1021.25	1033.125	1045	1056.875
	962.5	975	987.5	1000	1012.5	1025	1037.5	1050	1062.5	1075	1087.5	1100	1112.5
	1010.625	1023.75	1036.875	1050	1063.125	1076.25	1089.375	1102.5	1115.625	1128.75	1141.875	1155	1168.125
	1058.75	1072.5	1086.25	1100	1113.75	1127.5	1141.25	1155	1168.75	1182.5	1196.25	1210	1223.75
	1106.875	1121.25	1135.625	1150	1164.375	1178.75	1193.125	1207.5	1221.875	1236.25	1250.625	1265	1279.375
	1155	1170	1185	1200	1215	1230	1245	1260	1275	1290	1305	1320	1335
	1203.125	1218.75	1234.375	1250	1265.625	1281.25	1296.875	1312.5	1328.125	1343.75	1359.375	1375	1390.625
	1251.25	1267.5	1283.75	1300	1316.25	1332.5	1348.75	1365	1381.25	1397.5	1413.75	1430	1446.25
	1299.375	1316.25	1333.125	1350	1366.875	1383.75	1400.625	1417.5	1434.375	1451.25	1468.125	1485	1501.875
	1347.5	1365	1382.5	1400	1417.5	1435	1452.5	1470	1487.5	1505	1522.5	1540	1557.5
	1395.625	1413.75	1431.875	1450	1468.125	1486.25	1504.375	1522.5	1540.625	1558.75	1576.875	1595	1613.125
	1443.75	1462.5	1481.25	1500	1518.75	1537.5	1556.25	1575	1593.75	1612.5	1631.25	1650	1668.75
	1491.875	1511.25	1530.625	1550	1569.375	1588.75	1608.125	1627.5	1646.875	1666.25	1685.625	1705	1724.375
	1540	1560	1580	1600	1620	1640	1660	1680	1700	1720	1740	1760	1780
	1588.125	1608.75	1629.375	1650	1670.625	1691.25	1711.875	1732.5	1753.125	1773.75	1794.375	1815	1835.625
	1636.25	1657.5	1678.75	1700	1721.25	1742.5	1763.75	1785	1806.25	1827.5	1848.75	1870	1891.25
	1684.375	1706.25	1728 125	1750	1771.875	1793 75	1815.625	1837.5	1859.375	1881.25	1903 125	1925	1946 875
	1732.5	1755	1777.5	1800	1822.5	1845	1867.5	1890	1912.5	1935	1957.5	1980	2002.5
	1780 625	1803 75	1826 875	1850	1873 125	1896.25	1919 375	1942 5	1965 625	1988 75	2011 875	2035	2058 125
	1999 75	1852.5	1976.25	1000	1029 75	1047.5	1071.25	1005	2018 75	20/2 5	2066 25	2000	2113 75
	1876 875	1901 25	1925 625	1950	1974 375	1998 75	2023 125	2047.5	2071 875	2096.25	2120 625	2145	2169 375
	1025	1950	1975	2000	2025	2050	2075	2100	2195	2150	2175	2200	2025
	1079 135	1009 75	2024.275	2000	2025	0101.06	2075	0150.5	0170 105	9909 75	0000.975	2200	0000 806
	1973.123	1890.75	2024.375	2000	2013.025	2101.20	2120.073	2152.5	2170.123	2203.75	2228.373	2235	2200.023
	2021.25	2047.3	2073.75	2100	2120.23	2102.0	21/0./3	2200	2231.23	2237.5	2203.75	2010	2000.20
	2009.3/5	2090.25	2123.123	2150	21/0.0/5	2203.75	2230.020	2201.0	2204.3/5	2311.25	2330.125	2005	2081.0/5
	2117.5	2145	21/2.5	2200	2227.5	2205	2282.0	2310	2337.5	2305	2392.5	2420	2447.5
	2165.625	2193.75	2221.8/3	2250	22/8.125	2306.25	2334.375	2302.5	2390.025	2418.75	2440.875	2475	2503.125
	2213.75	2242.5	2271.25	2300	2328.75	2357.5	2386.25	2415	2443.75	2472.5	2501.25	2530	2558.75
	2261.875	2291.25	2320.625	2350	2379.375	2408.75	2438.125	2467.5	2496.875	2526.25	2555.625	2585	2614.375
	2310	2340	2370	2400	2430	2460	2490	2520	2550	2580	2610	2640	2670
	2358.125	2388.75	2419.375	2450	2480.625	2511.25	2541.875	2572.5	2603.125	2633.75	2664.375	2695	2725.625
	2405.25	2437.5	2468.75	2500	2531.25	2562.5	2593.75	2625	2656.25	2687.5	2718.75	2750	2781.25
	2454.375	2486.25	2518.125	2550	2581.875	2613.75	2645.625	2677.5	2709.375	2741.25	2773.125	2805	2836.875
	2502.5	2535	2567.5	2600	2632.5	2665	2697.5	2730	2762.5	2795	2827.5	2860	2892.5
	2550.625	2583.75	2616.875	2650	2683.125	2716.25	2749.375	2782.5	2815.625	2848.75	2881.875	2915	2948.125
	2598.75	2632.5	2666.25	2700	2733.75	2767.5	2801.25	2835	2868.75	2902.5	2936.25	2970	3003.75
	2646.875	2681.25	2715.625	2750	2784.375	2818.75	2853.125	2887.5	2921.875	2956.25	2990.625	3025	3059.375
	2695	2730	2765	2800	2835	2870	2905	2940	2975	3010	3045	3080	3115
	2743.125	2778.75	2814.375	2850	2885.625	2921.25	2956.875	2992.5	3028.125	3063.75	3099.375	3135	3170.625
	2791.25	2827.5	2863.75	2900	2936.25	2972.5	3008.75	3045	3081.25	3117.5	3153.75	3190	3226.25
	2839.375	2876.25	2913.125	2950	2986.875	3023.75	3060.625	3097.5	3134.375	3171.25	3208.125	3245	3281.875
	2887.5	2925	2962.5	3000	3037.5	3075	3112.5	3150	3187.5	3225	3262.5	3300	3337.5
	2935.625	2973.75	3011.875	3050	3088.125	3126.25	3164.375	3202.5	3240.625	3278.75	3316.875	3355	3393.125
	2983.75	3022.5	3061.25	3100	3138.75	3177.5	3216.25	3255	3293.75	3332.5	3371.25	3410	3448.75
	3031.875	3071.25	3110.625	3150	3189.375	3228 75	3268.125	3307.5	3346.875	3386.25	3425 625	3465	3504 375

Low Voltage Mode RC

	Capacito	or (uF)										
1600	1625	1650	1675	1700	1725	1750	1775	1800	1825	1850	1875	1900
40	40.625	41.25	41.875	42.5	43.125	43.75	44.375	45	45.625	46.25	46.875	47.
80	81.25	82.5	83.75	85	86.25	87.5	88.75	90	91.25	92.5	93.75	9
120	121.875	123.75	125.625	127.5	129.375	131.25	133.125	135	136.875	138.75	140.625	142.
160	162.5	165	167.5	170	172.5	175	177.5	180	182.5	185	187.5	190
200	203.125	206.25	209.375	212.5	215.625	218.75	221.875	225	228.125	231.25	234.375	237.
240	243.75	247.5	251.25	255	258.75	262.5	266.25	270	273.75	277.5	281.25	28
280	284.375	288.75	293.125	297.5	301.875	306.25	310.625	315	319.375	323.75	328.125	332.5
320	325	330	335	340	345	350	355	360	365	370	375	380
360	365.625	371.25	376.875	382.5	388.125	393.75	399.375	405	410.625	416.25	421.875	427.
400	406.25	412.5	418.75	425	431.25	437.5	443.75	450	456.25	462.5	468.75	473
440	446.875	453.75	460.625	467.5	474.375	481.25	488,125	495	501.875	508.75	515.625	522.3
480	487.5	495	502.5	510	517.5	525	532.5	540	547.5	555	562.5	570
520	528 125	536.25	544 375	552.5	560.625	568.75	576.875	585	593,125	601.25	609 375	617.
560	568 75	577.5	586.25	595	603.75	612.5	621.25	630	638.75	647.5	656 25	66
600	609 375	618 75	628 125	837.5	646 875	858.25	665 625	875	684 375	693 75	703 125	712
640	650	680	870	680	690	200	710	720	730	740	750	760
690	690.625	701.25	711.975	722.6	733 135	743.75	764 375	765	775.625	796.95	796.875	907
790	731.95	749.5	753.75	765	778.95	797.5	708.75	P10	891.95	932.5	842 75	007.0
720	731.23	742.0	705.70	007.5	010.23	101.5	780.75	010	021.20	032.3	043.75	000
700	111.0/5	/03./5	795.625	607.5	019.3/5	031.20	043.125	000	000.075	0/0./0	090.025	902.3
008	612.5	825	637.5	850	862.5	8/5	001 075	900	912.5	925	837.5	950
840	853.125	866.25	879.375	892.5	905.625	918.75	931.875	945	958.125	971.25	984.375	997.9
880	893.75	907.5	921.25	935	948.75	962.5	976.25	990	1003.75	1017.5	1031.25	1048
920	934.375	948.75	963.125	977.5	991.875	1006.25	1020.625	1035	1049.375	1063.75	1078.125	1092.3
960	975	990	1005	1020	1035	1050	1065	1080	1095	1110	1125	1140
1000	1015.625	1031.25	1046.875	1062.5	1078.125	1093.75	1109.375	1125	1140.625	1156.25	1171.875	1187.
1040	1056.25	1072.5	1088.75	1105	1121.25	1137.5	1153.75	1170	1186.25	1202.5	1218.75	123
1080	1096.875	1113.75	1130.625	1147.5	1164.375	1181.25	1198.125	1215	1231.875	1248.75	1265.625	1282.
1120	1137.5	1155	1172.5	1190	1207.5	1225	1242.5	1260	1277.5	1295	1312.5	1330
1160	1178.125	1196.25	1214.375	1232.5	1250.625	1268.75	1286.875	1305	1323.125	1341.25	1359.375	1377.
1200	1218.75	1237.5	1256.25	1275	1293.75	1312.5	1331.25	1350	1368.75	1387.5	1406.25	142
1240	1259.375	1278.75	1298.125	1317.5	1336.875	1356.25	1375.625	1395	1414.375	1433.75	1453.125	1472.5
1280	1300	1320	1340	1360	1380	1400	1420	1440	1460	1480	1500	1520
1320	1340.625	1361.25	1381.875	1402.5	1423.125	1443.75	1464.375	1485	1505.625	1526.25	1546.875	1567.5
1360	1381.25	1402.5	1423.75	1445	1466.25	1487.5	1508.75	1530	1551.25	1572.5	1593.75	1613
1400	1421.875	1443.75	1465.625	1487.5	1509.375	1531.25	1553.125	1575	1596.875	1618.75	1640.625	1662.
1440	1462.5	1485	1507.5	1530	1552.5	1575	1597.5	1620	1642.5	1665	1687.5	1710
1480	1503.125	1526.25	1549.375	1572.5	1595.625	1618.75	1641.875	1665	1688.125	1711.25	1734.375	1757.
1520	1543.75	1567.5	1591.25	1615	1638.75	1662.5	1686.25	1710	1733.75	1757.5	1781.25	1803
1560	1584.375	1608.75	1633.125	1657.5	1681.875	1706.25	1730.625	1755	1779.375	1803.75	1828.125	1852.5
1600	1625	1650	1675	1700	1725	1750	1775	1800	1825	1850	1875	1900
1640	1665.625	1691.25	1716.875	1742.5	1768,125	1793.75	1819.375	1845	1870.625	1896.25	1921.875	1947.
1680	1706 25	1732.5	1758.75	1785	1811.25	1837.5	1863.75	1890	1916.25	1942.5	1968.75	1995
1720	1746 875	1773.75	1800 625	1827.5	1854 375	1881.25	1908 125	1935	1961.875	1988 75	2015 625	2042
1760	1787.5	1815	1842.5	1870	1897.5	1925	1952.5	1980	2007 5	2035	2062.5	209/
1800	1828 125	1856.25	1884 375	1912.5	1940 625	1968 75	1996 875	2025	2053 125	2081 25	2109 375	2137
1840	1868 75	1897.5	1926.25	1955	1983 75	2012.5	2041 25	2070	2098 75	2127.5	2156.25	219
1880	1000.75	1038.75	1968 125	1907 6	2026.875	2058.25	2085 625	2115	2144 375	2173.75	2203 125	2002
1020	1050	1090	2010	2040	2020.070	2100	2130	2160	2100	2000	9250	200
1920	1000 000	0001.00	2010	2040	20/0	2100	2130	2160	2190	0000.00	2250	0207
0000	0001.05	2021.25	2001.879	2082.5	2113.125	2143.73	2174.375	2205	0001.020	2200.20	2290.8/5	2327.5
2000	2031.25	2002.5	2093.75	2125	2155.25	2187.5	2218.75	2250	2281.25	2312.5	2343.75	23/3
2040	20/1.8/5	2103.75	2135.625	2167.5	2199,375	2231.25	2263.125	2295	2326.875	2358.75	2390.525	2422.
2080	2112.5	2145	21/7.5	2210	2242.5	22/5	2307.5	2340	23/2.5	2405	2437.5	247
2120	2153.125	2186.25	2219.375	2252.5	2285.625	2318.75	2351.875	2385	2418.125	2451.25	2484.375	2517.
2160	2193.75	2227.5	2261.25	2295	2328.75	2362.5	2396.25	2430	2463.75	2497.5	2531.25	256
2200	2234.375	2268.75	2303 125	2337.5	2371.875	2406.25	2440.625	2475	2509.375	2543.75	2578.125	2612.5
2240	2275	2310	2345	2380	2415	2450	2485	2520	2555	2590	2625	266
2280	2315.625	2351.25	2386.875	2422.5	2458.125	2493.75	2529.375	2565	2600.625	2636.25	2671.875	2707.
2320	2356.25	2392.5	2428.75	2465	2501.25	2537.5	2573.75	2610	2646.25	2682.5	2718.75	275
2360	2396.875	2433.75	2470 625	2507.5	2544.375	2581.25	2618.125	2655	2691.875	2728.75	2765.625	2802.
2400	2437.5	2475	2512.5	2550	2587.5	2625	2662.5	2700	2737.5	2775	2812.5	2850
2440	2478.125	2516.25	2554.375	2592.5	2630.625	2668.75	2708.875	2745	2783.125	2821.25	2859.375	2897.
2480	2518.75	2557.5	2596.25	2635	2873.75	2712.5	2751.25	2790	2828.75	2867.5	2906.25	294
2520	2559.375	2598.75	2638.125	2677.5	2716.875	2758.25	2795.625	2835	2874.375	2913.75	2953.125	2992.5

Low Voltage Mode RC

	2575	2600	2625	2650	2675	2700	2725	2750	2775	2800	2825	2850	2875
	64.375	65	65.625	66.25	66.875	67.5	68.125	68.75	69.375	70	70.625	71.25	71.875
	128.75	130	131.25	132.5	133.75	135	136.25	137.5	138.75	140	141.25	142.5	143.75
	193 125	195	196.875	198 75	200 625	202.5	204 375	206.25	208 125	210	211 875	213.75	215 625
	257.5	260	262.5	265	267.5	270	272.5	275	277.5	280	282.5	285	287.5
	201 975	205	329 125	391.35	224 275	997.6	340.635	2/2 75	346 975	260	353 125	258.25	250 275
	206.05	300	202 75	207.5	401.05	405	409.75	412.5	416.05	420	402.75	497.6	431.35
	300.23	390	393.75	100 75	401.25	405	400.73	412.0	410.23	420	423.15	427.5	401.20
	450.525	455	458.375	403.75	408.125	472.5	4/6.8/5	401.25	485.025	490	494.375	485.75	503,125
	515	520	525	530	535	540	545	550	555	560	565	5/0	5/5
	579.375	585	590.625	596.25	601.875	607.5	613.125	618.75	624.375	630	635.625	641.25	646.875
	643.75	650	656.25	662.5	668.75	675	681.25	687.5	693.75	700	706.25	712.5	718.75
Ê	708.125	715	721.875	728.75	735.625	742.5	749.375	756.25	763.125	770	776.875	783.75	790.625
E	772.5	780	787.5	795	802.5	810	817.5	825	832.5	840	847.5	855	862.5
0	836.875	845	853.125	861.25	869.375	877.5	885.625	893.75	901.875	910	918.125	926.25	934.375
-	901.25	910	918.75	927.5	936.25	945	953.75	962.5	971.25	980	988.75	997.5	1006.25
9	965.625	975	984.375	993.75	1003.125	1012.5	1021.875	1031.25	1040.625	1050	1059.375	1068.75	1078.125
10	1030	1040	1050	1060	1070	1080	1090	1100	1110	1120	1130	1140	1150
ě	1094.375	1105	1115.625	1126.25	1136.875	1147.5	1158.125	1168.75	1179.375	1190	1200.625	1211.25	1221.875
£	1158.75	1170	1181.25	1192.5	1203.75	1215	1226.25	1237.5	1248.75	1260	1271.25	1282.5	1293.75
	1223.125	1235	1246.875	1258 75	1270.625	1282.5	1294.375	1306.25	1318,125	1330	1341.875	1353.75	1365.625
	1287.5	1300	1312.5	1325	1337.5	1350	1362.5	1375	1387.5	1400	1412.5	1425	1437.5
	1951 875	1385	1978 195	1301.95	1404 375	1417.5	1430 625	1443 75	1456 875	1470	1483 125	1408.95	1500 375
	1416.25	1490	1443 75	1457.5	1471.25	1495	1498 75	1512.5	1526.25	1540	1553 75	1587.5	1581 25
	1410.23	1400	1600.975	1609.75	14/1.23	1400	1480.75	1012.0	1520.25	1040	1004.975	1007.0	1001.20
	1400.025	1495	1009.575	1020.70	1036.125	1002.0	1000.075	1001.20	1093.025	1010	1024.373	1000.70	1055.125
	1545	1560	15/5	1590	1605	1620	1635	1000	1005	1580	1695	1/10	1725
	1609.375	1625	1640.625	1656.25	16/1.8/5	1687.5	1703.125	1/18.75	1734.375	1750	1765.625	1/81.25	1/96.875
	16/3.75	1690	1708.25	1/22.5	1/38./5	1/55	1//1.25	1/8/.5	1803.75	1820	1836.25	1852.5	1868.75
	1738.125	1755	1771.875	1788.75	1805.625	1822.5	1839.375	1856.25	1873.125	1890	1906.875	1923.75	1940.625
	1802.5	1820	1837.5	1855	1872.5	1890	1907.5	1925	1942.5	1960	1977.5	1995	2012.5
	1866.875	1885	1903.125	1921.25	1939.375	1957.5	1975.625	1993.75	2011.875	2030	2048.125	2066.25	2084.375
	1931.25	1950	1968.75	1987.5	2006.25	2025	2043.75	2062.5	2081.25	2100	2118.75	2137.5	2156.25
	1995.625	2015	2034.375	2053.75	2073.125	2092.5	2111.875	2131.25	2150.625	2170	2189.375	2208.75	2228.125
	2060	2080	2100	2120	2140	2160	2180	2200	2220	2240	2260	2280	2300
	2124.375	2145	2165.625	2186.25	2206.875	2227.5	2248.125	2268.75	2289.375	2310	2330.625	2351.25	2371.875
	2188.75	2210	2231.25	2252.5	2273.75	2295	2316.25	2337.5	2358.75	2380	2401.25	2422.5	2443.75
	2253.125	2275	2296.875	2318.75	2340.625	2362.5	2384.375	2406.25	2428.125	2450	2471.875	2493.75	2515.625
	2317.5	2340	2362.5	2385	2407.5	2430	2452.5	2475	2497.5	2520	2542.5	2565	2587.5
	2381.875	2405	2428.125	2451.25	2474.375	2497.5	2520.625	2543.75	2566.875	2590	2613.125	2636.25	2659.375
	2446.25	2470	2493.75	2517.5	2541.25	2565	2588.75	2612.5	2636.25	2660	2683.75	2707.5	2731.25
	2510 625	2595	2559 375	2583 75	2608 125	2632 5	2656 875	2681 25	2705 625	2730	2754 375	2778 75	2803 125
	2575	2800	2825	2650	2675	2700	2725	2750	2775	2800	2825	2850	2975
	2690 975	2865	2600.625	2716.25	2741 975	2767.6	2702 125	2019 75	2944 975	2000	2905 625	2001 05	20/6 975
	0209.75	2000	0758.05	0700.5	0000 75	0025	0001 05	0007 5	20112 75	2010	0000 05	2021.20	2040.075
	2/03.75	2730	2/30.25	2102.0	2000.73	2000	2001.23	2007.3	2913.75	2940	2900.23	2892.0	2010.73
	2/00.120	2785	2021.075	2040.75	2075.025	2902.5	2929.375	2800.20	2903.125	3010	3036.675	3003.75	3090.625
	2832.5	2860	2887.5	2915	2942.5	2970	2997.5	3025	3052.5	3080	3107.5	3135	3162.5
	2896.875	2925	2953.125	2981.25	3009.375	3037.5	3065.625	3093.75	3121.875	3150	3178.125	3206.25	3234.375
	2961.25	2990	3018.75	3047.5	3076.25	3105	3133.75	3162.5	3191.25	3220	3248.75	3277.5	3306.25
	3025.625	3055	3084.375	3113.75	3143.125	3172.5	3201.875	3231.25	3260.625	3290	3319.375	3348.75	3378.125
	3090	3120	3150	3180	3210	3240	3270	3300	3330	3360	3390	3420	3450
	3154.375	3185	3215.825	3246.25	3276.875	3307.5	3338.125	3368.75	3399.375	3430	3460.625	3491.25	3521.875
	3218.75	3250	3281.25	3312.5	3343.75	3375	3406.25	3437.5	3468.75	3500	3531.25	3562.5	3593.75
	3283.125	3315	3346.875	3378.75	3410.625	3442.5	3474.375	3506.25	3538.125	3570	3601.875	3633.75	3665.625
	3347.5	3380	3412.5	3445	3477.5	3510	3542.5	3575	3607.5	3640	3672.5	3705	3737.5
	3411.875	3445	3478.125	3511.25	3544.375	3577.5	3610.625	3643.75	3676.875	3710	3743.125	3776.25	3809.375
	3476.25	3510	3543.75	3577.5	3611.25	3645	3678.75	3712.5	3746.25	3780	3813.75	3847.5	3881.25
	3540.625	3575	3609.375	3643.75	3678.125	3712.5	3746.875	3781.25	3815.625	3850	3884.375	3918.75	3953,125
	3605	3640	3675	3710	3745	3780	3815	3850	3885	3920	3955	3990	4025
	3669 375	3705	3740 625	3776.25	3811.875	3847 5	3883 125	3918 75	3054 375	1000	4025.625	4061.25	4096.875
	9709.75	9770	3802.020	3940 5	3870 75	2015	2051 120	3007 6	AU03 2E	1000	4000.020	A199 E	A160.073
	9706.405	3770	3000.25	30042.5	30/0./3	0915	4010 075	050.05	4020.75	4000	4090.25	4102.5	4100.75
	3/98.125	3835	387 1.875	3808.75	3845.025	3962.5	4019.375	4036.25	4093.125	4130	4100.875	4203.75	4240.625
	3862.5	3900	3937.5	3975	4012.5	4050	4087.5	4125	4162.5	4200	4237.5	42/5	4312.5
	3926.875	3965	4003.125	4041.25	4079.375	4117.5	4155.625	4193.75	4231.875	4270	4308.125	4346.25	4384.375
	3991.25	4030	4068.75	4107.5	4146.25	4185	4223.75	4262.5	4301.25	4340	4378.75	4417.5	4456.25
	4055.625	4095	4134.375	4173.75	4213.125	4252.5	4291.875	4331.25	4370.625	4410	4449.375	4488.75	4528.125

2250	2275	2300	2325	2350	2375	2400	2425	2450	2475	2500	2525	2550
56.25	56.875	57.5	58,125	58.75	59.375	60	60.625	61.25	61.875	62.5	63.125	63.75
112.5	113.75	115	116.25	117.5	118.75	120	121.25	122.5	123.75	125	126.25	127.5
168.75	170.625	172.5	174.375	176.25	178.125	180	181.875	183.75	185.625	187.5	189.375	191.25
225	227.5	230	232.5	235	237.5	240	242.5	245	247.5	250	252.5	255
281.25	284.375	287.5	290.625	293.75	296.875	300	303.125	306.25	309.375	312.5	315.625	318.75
337.5	341.25	345	348.75	352.5	356.25	360	363.75	367.5	371.25	375	378.75	382.5
393.75	398.125	402.5	406.875	411.25	415.625	420	424.375	428.75	433.125	437.5	441.875	446.25
450	455	460	465	470	475	480	485	490	495	500	505	510
506.25	511.875	517.5	523.125	528.75	534.375	540	545.625	551.25	556.875	562.5	568.125	573.75
562.5	568.75	575	581.25	587.5	593.75	600	606.25	612.5	618.75	625	631.25	637.5
618.75	625.625	632.5	639.375	646.25	653.125	660	666.875	673.75	680.625	687.5	694.375	701.25
675	682.5	690	697.5	705	712.5	720	727.5	735	742.5	750	757.5	765
731.25	739.375	747.5	755.625	763.75	771.875	780	788.125	796.25	804.375	812.5	820.625	828 75
787.5	796.25	805	81375	822.5	831.25	840	848.75	857.5	866.25	875	883.75	892.5
843.75	853.125	862.5	871.875	881.25	890.625	900	909.375	918.75	928.125	937.5	946.875	956.25
900	910	920	930	940	950	960	970	980	990	1000	1010	1020
956.25	966.875	977.5	988.125	998.75	1009.375	1020	1030.625	1041.25	1051.875	1062.5	10/3.125	1083.75
1012.5	1023.75	1035	1046.25	1057.5	1068.75	1080	1091.25	1102.5	1113.75	1125	1136.25	1147.5
1068.75	1080.625	1092.5	1104.375	1116.25	1128.125	1140	1151.875	1163.75	11/5.625	1187.5	1199.375	1211.25
1125	1137.5	1150	1162.5	11/5	1187.5	1200	1212.5	1225	1237.5	1250	1262.5	1275
1181.25	1194.375	1207.5	1220.625	1233.75	1246.875	1250	12/3.125	1286.25	1299.375	1312.5	1325.625	1338.75
1237.5	1201.20	1200	12/8./5	1292.5	1306.25	1320	1333.75	1347.5	1361.25	13/5	1388.75	1402.5
1293.75	1308.125	1322.5	1335.875	1351.25	1365.625	1380	1394.375	1408.75	1423.125	1437.5	1451.8/5	1466.25
1350	1401 075	1380	1459.405	1410	1425	1440	1455	14/0	1460	1500	1010	1530
1400.20	1421.675	1437.5	1453.125	1406.70	1464.375	1500	1515.625	1031.20	1040.675	1002.0	15/6.125	1093.70
1402.0	14/6./5	1495	1511.25	1027.0	1043.75	1000	10/0.20	1092.0	1008.75	1023	1704 275	1007.0
1010.70	1535.625	1002.0	1007.5	1000.20	1003.125	1020	1030.075	1055.75	10/0.025	1007.0	1704.375	1721.23
1070	1092.0	1010	1027.5	1709.75	1701 975	1740	1759 105	1715	1732.5	1910.6	1000.005	1040 75
1697.5	1706 25	1705	1749.75	1762.6	1721.075	1900	1010 75	1007.5	1056.05	1012.0	1000.025	1040.70
1749.75	1769 195	1782.5	1801.875	1921.25	1940.625	1960	1879 275	1909.75	1010.25	1027.5	1056 975	1076.05
1900	1/03.125	17 52.5	1960	1021.20	1940.025	1600	10/9.3/5	1090.70	1910.120	2000	2020	2040
1856.25	1876 875	1897.5	1018 125	1039.75	1059 375	1680	2000 625	2021.25	20/1 875	2000	2083 125	2103 75
1012.5	1093 75	1057.5	1076.05	19907.5	2019 75	2040	2000.025	2021.20	2041.075	2002.0	2000.120	2103.73
1968 75	1990.625	2012.5	2034 375	2056.25	2078 125	2100	2121.875	2149.75	2165 625	2187.5	2209 375	2231.25
2025	2047.5	20720	2004.010	2115	2137.5	2160	2182.5	22005	2203.025	2250	2203.075	2001.20
2023	2104 375	2127.5	2150 625	2179 75	2196.875	2220	2243 125	2205	2227.5	2312.5	2335 825	2958 75
2137.5	2161 25	2185	2208 75	2232.5	2256 25	2280	2303 75	2327.5	2351.25	2375	2398 75	2422 5
2193 75	2218 125	2242.5	2268 875	2291.25	2315 625	2340	2364 375	2388 75	2413 125	2437.5	2461 875	2486.25
2250	2275	2300	2325	2350	2375	2400	2425	2450	2475	2500	2525	2550
2306.25	2331.875	2357.5	2383 125	2408.75	2434 375	2460	2485.625	2511.25	2536 875	2562.5	2588 125	2613 75
2362.5	2388.75	2415	2441.25	2467.5	2493 75	2520	2546 25	2572 5	2598.75	2625	2851.25	2877.5
2418.75	2445.625	2472.5	2499.375	2526.25	2553 125	2580	2606.875	2633.75	2660.625	2687.5	2714.375	2741.25
2475	2502.5	2530	2557.5	2585	2612.5	2640	2667.5	2695	2722.5	2750	2777.5	2805
2531.25	2559.375	2587.5	2615.625	2643.75	2671.875	2700	2728.125	2756.25	2784.375	2812.5	2840.625	2868.75
2587.5	2616.25	2645	2673.75	2702.5	2731.25	2760	2788.75	2817.5	2846.25	2875	2903.75	2932.5
2643.75	2673.125	2702.5	2731.875	2761.25	2790.625	2820	2849.375	2878.75	2908.125	2937.5	2966.875	2996.25
2700	2730	2760	2790	2820	2850	2880	2910	2940	2970	3000	3030	3060
2756.25	2786.875	2817.5	2848,125	2878.75	2909.375	2940	2970.625	3001.25	3031.875	3062.5	3093,125	3123.75
2812.5	2843.75	2875	2906.25	2937.5	2968.75	3000	3031.25	3062.5	3093.75	3125	3156.25	3187.5
2868.75	2900.625	2932.5	2964.375	2996.25	3028.125	3060	3091.875	3123.75	3155.625	3187.5	3219.375	3251.25
2925	2957.5	2990	3022.5	3055	3087.5	3120	3152.5	3185	3217.5	3250	3282.5	3315
2981.25	3014.375	3047.5	3080.625	3113.75	3146.875	3180	3213.125	3246.25	3279.375	3312.5	3345.625	3378.75
3037.5	3071.25	3105	3138.75	3172.5	3206.25	3240	3273.75	3307.5	3341.25	3375	3408.75	3442.5
3093.75	3128.125	3162.5	3196.875	3231.25	3265.625	3300	3334.375	3368.75	3403.125	3437.5	3471.875	3506.25
3150	3185	3220	3255	3290	3325	3360	3395	3430	3465	3500	3535	3570
3206.25	3241.875	3277.5	3313.125	3348.75	3384.375	3420	3455.625	3491.25	3526.875	3562.5	3598.125	3633.75
3262.5	3298.75	3335	3371.25	3407.5	3443.75	3480	3516.25	3552.5	3588.75	3625	3661.25	3697.5
3318.75	3355.625	3392.5	3429.375	3466.25	3503.125	3540	3576.875	3613.75	3650.625	3687.5	3724.375	3761.25
3375	3412.5	3450	3487.5	3525	3562.5	3600	3637.5	3675	3712.5	3750	3787.5	3825
3431.25	3469.375	3507.5	3545.625	3583.75	3621.875	3660	3698.125	3736.25	3774.375	3812.5	3850.625	3888.75
3487.5	3526.25	3565	3603.75	3642.5	3681.25	3720	3758.75	3797.5	3836.25	3875	3913.75	3952.5
3543 75	2602 126	2822.5	2861 975	2701.05	2740 625	2790	2210.275	2959 75	2000 125	2027 6	2076 975	4016 25

	3225	3250	3275
	80.625	81.25	81.87
	161.25	162.5	163.75
	241.875	243.75	245.62
	322.5	325	327.5
	403.125	406.25	409.37
	483.75	487.5	491.2
	564.375	568.75	573.12
	645	650	655
	725.625	731.25	/35.8/3
_	996 975	812.5	000 60
Ê	967.5	093.75	900.023
R	1048 125	1056.25	1064 37
9	1128.75	1137.5	1146.2
ō	1209.375	1218 75	1228 12
ist	1290	1300	1310
es	1370.625	1381.25	1391.87
Ē	1451.25	1462.5	1473.7
	1531.875	1543.75	1555.62
	1612.5	1625	1637.
	1693.125	1706.25	1719.37
	1773.75	1787.5	1801.2
	1854.375	1868.75	1883.12
	1935	1950	196
	2015.625	2031.25	2046.87
	2096.25	2112.5	2128.7
	2176.875	2193.75	2210.62
	2257.5	2275	2292
	2338.125	2356.25	2374.37
	2418.75	2437.5	2456.2
	2499.375	2518.75	2538.12
	2580	2600	262
	2660.625	2681.25	2701.87
	2741.25	2762.5	2783.7
	2821.875	2843.75	2865.62
	2902.5	2925	2947.
	2903.125	3000.25	3028.37
	3063.75	3067.5	3103.19
	3144.375	3100.75	3193.12
	3305.625	3331.25	3356.87
	3386.25	3412.5	3438.7
	3466.875	3493.75	3520.62
	3547.5	3575	3602
	3628.125	3656.25	3684.37
	3708.75	3737.5	3766.2
	3789.375	3818.75	3848.12
	3870	3900	393
	3950.625	3981.25	4011.87
	4031.25	4062.5	4093.7
	4111.875	4143.75	4175.62
	4192.5	4225	4257.
	4273.125	4306.25	4339.37
	4353.75	4387.5	4421.2
	4434.375	4468.75	4503.12
	4515	4550	458
	4595.625	4631.25	4666.87
	4676.25	4712.5	4748.7
	4756.875	4793.75	4830.62
	4837.5	4875	4912.
	the second se	the second second	1.000
	4918.125	4956.25	4994.37

Capacitor (uF)					
	25.00	50.00			
25.00	0.63	1.25			
50.00	1.25	2.50			
75.00	1.88	3.75			
100.00	2.50	5.00			
125.00	3.13	6.25			
150.00	3.75	7.50			
175.00	4.38	8.75			
200.00	5.00	10.00			
225.00	5.63	11.25			
250.00	6.25	12.50			
275.00	6.88	13.75			
300.00	7.50	15.00			
325.00	8.13	16.25			
350.00	8.75	17.50			
375.00	9.38	18.75			
400.00	10.00	20.00			
425.00	10.63	21.25			
450.00	11.25	22.50			
475.00	11.88	23.75			
500.00	12.50	25.00			
525.00	13.13	26.25			
550.00	13.75	27.50			
575.00	14.38	28.75			
600.00	15.00	30.00			
625.00	15.63	31.25			
650.00	16.25	32.50			
675.00	16.88	33.75			
700.00	17.50	35.00			
725.00	18.13	36.25			
750.00	18.75	37.50			
775.00	19.38	38.75			
805.00	20.00	40.00			
850.00	20.03	41.20			
875.00	21.23	42.50			
900.00	22.50	45.00			
925.00	23.13	46.25			
950.00	23.75	47.50			
975.00	24.38	48.75			
1000.00	25.00	50.00			
1025.00	25.63	51.25			
1050.00	26.25	52.50			
1075.00	26.88	53.75			
1100.00	27.50	55.00			
1125.00	28.13	56.25			
1150.00	28.75	57.50			
1175.00	29.38	58.75			
1200.00	30.00	60.00			
1225.00	30.63	61.25			
1250.00	31.25	62.50			
1275.00	31.88	63.75			
1300.00	32.50	65.00			
1325.00	33.13	66.25			
1350.00	33.75	67.50			
1375.00	34.38	68.75			
1400.00	35.00	70.00			
1425.00	35.63	71.25			
1450.00	36.25	72.50			
1475.00	36.88	73.75			
1500.00	37.50	75.00			
1525.00	38.13	76.25			
1550.00	38.75	77.50			
1575.00	39.38	78.75			

Resistor (ohm)

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Appendix C: Optimization Strategies

General

The success of electro cell manipulation (ECM) lies in selecting appropriate ECM systems capable of delivering the pulses suitable for the cell being electromanipulated. One, or several pulses of the appropriate field strength, pulse length, and wave shape may be required for this purpose.

The key to success with electroporation-based technologies involves a proper combination of biological, physical, chemical, and pulse parameters. In general, cells must be in mid-logarithmic growth for optimal electroporation. Various temperature regimens have been described. It has been shown that a variety of chemical techniques may increase electroporation efficiencies, including addition of EDTA, DMSO, intracellular salts, and serum before or after the pulse. Optimizing protocols abound. Analysis of these optimization regimens has lead to proposals of universal protocols, involving very limited optimization over a narrow range.

Electroporation

- Vary the voltage in order to vary the field strength, keeping other parameters constant. Assay sample for both viability and endpoint. Plot the field strength versus both viability and endpoint and extrapolate the optimal field strength (voltage divided by gap size) and voltage.
- 2. Vary the capacitance/resistance/sample volume at the optimal voltage setting in order to vary the pulse length (time constant) for exponential decay instruments. Directly vary square wave instrument pulse length. Assay sample for both viability and endpoint. Plot the pulse length versus both viability and endpoint and extrapolate the optimal pulse length/parameters.
- **3.** For multiple pulsing systems/protocols, vary the number of pulses at the optimal field strength and pulse length. Assay sample for both viability and endpoint. Plot the number of pulses versus both viability and endpoint, and extrapolate the optimal number of pulses.

Appendix D: Electrical Troubleshooting

Instrument Does Not Power Up

Verify that the power cord is fully inserted in the instrument and in the wall outlet. Verify that the fuse is not blown. Disconnect power cord from the instrument before removing the fuse holder. Replace the fuse, if necessary, with same rated fuse as indicated on back panel.

Unanticipated RC time constants or peak voltage output

Un-anticipated RC time constant or peak output voltage may be a sign that appropriate capacitors are not being selected or are not fully charging. Please remember that the external load (sample) reduces the expected time constant and voltage to various degrees. If you believe there is a problem, contact BTX Technical Support for immediate consultation.

LCD Error Messages

The ECM[®] 630 is constantly monitoring the parameters of some of its internal circuitry. In the case of a malfunction, one of the following messages will appear on the display. Note the instructions on the following page used to confirm the absence of a pulse. In this case, call BTX Technical Support.

EEPROM Failure

The unit has detected a malfunction in its internal memory system. The validity of the data might be compromised. Turning or pressing the knob will bring the Set Parameters screen. Verify carefully every setpoint before pulsing. This verification is performed during power up and every time that data is loaded from memory. Contact BTX Technical Support if this error message is displayed again, after a power up sequence.

Pulsing Aborted Charge Failure

The unit did not charge its selected capacitor bank. Turn or press the encoder knob. Disconnect the load from the HV connector. Select a different capacitor and press the pulse button. Contact BTX[®] Technical Support if a similar message is displayed again.

Pulsing Aborted Charging Timed Out

A charging time limit of 20 seconds is provided for circuit safety. If the capacitors are not charged to the pre-set voltage level after 20 seconds, the "PULSING ABORTED CHARGING TIMED OUT" message is displayed. For assistance with this situation, please contact BTX Technical Support. Press the encoder or the pulse switch once to get back to the Set Parameters screen.

Appendix E: Experimental Troubleshooting

Arcing

Verify electrical component functionality. Verify properties of cell sample (do cells need to be washed? Is the buffer appropriate for application?). Verify properties of transfectant/molecule (Is the DNA well purified?) Try reducing the voltage or increase sample volume until arcing is no longer a problem.

Low (or no) transfection efficiency, or incorporation

Verify physical, biological, and chemical parameters. Verify delivery of the pulse and pulse parameters. Is the voltage correct? Chamber gap? Pulse length or appropriate instrument settings? Number of pulses? If so, follow Optimization Guidelines outlined in Appendix A.

Low viability

Verify physical, biological, and chemical parameters. Are the voltage, chamber gap, pulse length (time constant), pulse number and other instrument settings correct? If so, reduce voltage, pulse length, or number of pulses and re-optimize protocol to improve viability as outlined in Appendix A.

Appendix F: Glossary of Electrical Terms

Amplitude

The instantaneous value of current or voltage in amperes or volts.

Capacitor

A device that stores electric energy in the form of an internal electric field. Energy is delivered when a current flows out of a capacitor. The current normally follows an exponential curve.

Dielectric

A material that has a high resistivity and can store energy in the form of an electric field.

Direct Current (DC)

Current whose amplitude is constant with time. Direct currents are used to form temporary pores in bi-lipid membranes. Cells may fuse when pores in the membranes of two juxtaposed cells reseal after a DC application.

Divergence

The deviation of electric field lines from a parallel homogeneous condition. A highly divergent field has field lines that rapidly change amplitude (or strength) and direction in the area of interest.

Electric Field

The electric potential difference between two points divided by the distance separating those points. Expressed in volt/cm.

Electric Field Force

The mechanical force acting on any electric charge when placed in an electric field.

Exponential Decay

Non linear waveform typical of capacitor charge and discharge currents and voltages. The exponential decay waveform is characterized by its time constant, the time it takes the voltage to decay to 1/e of the peak voltage.

Field Strength

See Electric Field.

Frequency

The number of times an oscillation goes through a complete cycle in one second. The unit is either cycle/sec or (Hz).

Homogenous Electric Field

An electric field where the direction and strength of the field lines are constant.



Appendix G: Glossary of Biological & Technical Terms

Chambers

Electroporation and Electrofusion Chambers are the devices used to hold the cells/molecules to be fused/transfected.

Dielectric Breakdown

The reversible breakdown of lipid bilayer membranes as a result of the application of a DC electroporation pulse. Sufficiently high field strength may increase the membrane potential past a critical point leading to the breakdown of the membrane.

Dielectrophoresis

A consequence of cells being exposed to an inhomogeneous or divergent electric field, resulting in their movement toward electrodes, and subsequent alignment or pearl chain formation.

Electroinsertion

The use of electroporation to insert molecules into lipid bilayer membranes.

Electropermeabilization

The use of electroporation to make cells, protoplasts, or liposomes permeable to ions and small molecules in their extracellular environment.

Electroporation

The application of high electric field pulses of short duration to create temporary pores (holes) in the membranes of cells.

Hydrostatic Pressure

The pressure in liquids at rest.

Lipid Bilayer

An assembly of lipid and protein molecules held together by non-covalent interactions. All biological membranes share this common structure.

Osmotic Pressure

The applied pressure required to prevent the flow of solvents of different concentration across a semipermeable membrane.

Pore

A small, mostly transient, opening in a cell wall caused by the application of a brief high electric field pulse.

Pressure Gradient

The difference in pressure between two points in a medium.



Disposable Electroporation Cuvettes Plus[™]

Appendix G: Glossary of Biological & Technical Terms (Continued)

Protoplasts

The plant cell proper, with the cellulose cell wall removed.

Relaxation Time

The time a system requires to reach equilibrium.

Transfection

The introduction of nucleic acids into animal cells. Stable transfections result in integration of nucleic acids into host chromosomes and the inheritance of associated traits in progeny cells. Transient transfections result in temporary expression of exogenous nucleic acids.

Transformation

The introduction of nucleic acids into microorganisms and plant cells.

Turgor Pressure

The pressure in capillaries.

Appendix H: Electroporation Pulse Generator Compatibility

Certain components of BTX Electroporation are compatible with components of competitive systems.

Please contact BTX Technical Support for details.

Appendix I: Recommended Reading

Eberhard Neumann, Editor, Electroporation and Electrofusion in Cell Biology, Plenum Publishing Corporation, 1989

Michael Kriegler, Gene Transfer and Expression, A Laboratory Manual, Stockton Press, 199

Donald Chang, Editor-in-Chief, Guide to Electroporation and Electrofusion, Academic Press, 1992

Jac A. Nickoloff, Editor, Electroporation Protocols for Microorganisms, in Methods in Molecular Biology, Vol 47, Humana Press, 1995

Jac A. Nickoloff, Editor, Animal Cell Electroporation and Electrofusion Protocols, in Methods in Molecular Biology, Vol 48, Humana Press, 1995

Jac A. Nickoloff, Editor, Plant Cell Electroporation and Electrofusion Protocols, in Methods in Molecular Biology, Vol 55, Humana Press, 1995

For further references regarding specific applications and optimization, please contact BTX Technical Support:

BTX-Division of Harvard Apparatus 84 October Hill Road Hollistion, MA 01746 Phone: 1-508-893-8999 Toll Free: 1-800-272-2775 Fax: 1-508-429-5732 Email: techsupport.btx@harvardapparatus.com Website: www.btxonline.com

Appendix J: Accessories and Replacement Parts

Catalog No.	Model	Description
MA1 45-0001	6300	ECM [®] 630 Electroporation System
MA1 45-0051	630	ECM [®] 630 Electroporator only
MA1 45-0207	630B	Electroporation Safety Stand
MA1 45-0124	610	Disposable Electroporation Cuvettes Plus, 1mm, 50 per bag
MA1 45-0125	620	Disposable Electroporation Cuvettes Plus, 2 mm, 50 per bag
MA1 45-0126	640	Disposable Electroporation Cuvettes Plus, 4 mm, 50 per bag
MA1 45-0400	HT100	Manual 96-Well Plate Handler
MA1 45-0450	HT-P96-2	Disposable 96-Well Plate 2mm gap
MA1 45-0452	HT-P96-4	96-Well Disposable Plate 4mm
MA1 45-0463	HT-P25-P4	25-Well Disposable Plate 4mm pkg 6
MA1 45-0462	HT-P25-4	25-Well Disposable Plate 4mm
MA1 45-0465		25 Well-Adapter HT
MA1 45-0466	HT-P25-2	25-Well Disposable plate 2mm gap
MA1 45-0467	HT-P25-2P	25-Well Disposable Plates 2mm gap pkg 6
MA1 45-0059	Enhancer 3000®	Enhancer 3000 [®] Electroporation Monitoring System

Appendix K: General Care and Cleaning

General Care

Do not store or leave the instrument where the LCD display will be exposed to direct sunlight for long periods of time.



CAUTION

To avoid damage to the instrument, do not expose to sprays, liquids, or solvents.

Cleaning

Inspect the instrument, as often as operating conditions require. To clean the instrument exterior, perform the following steps:

- Remove loose dust on the outside of the instrument with a lint-free cloth. Use care to avoid scratching the clear plastic display filter.
- Use a soft cloth dampened with water to clean the instrument. Use an aqueous solution of 75% isopropyl alcohol for more efficient cleaning.



CAUTION

To avoid damage to the surface of the instrument, do not use any abrasive or chemical cleaning agents. Use caution not to drop or cause any unwarranted physical harm to the instrument during any cleaning operations.

Appendix L: Connecting Electrodes

Cuvettes/Safety Stand

- **1.** Remove the cuvette and the transfer pipette from their sterile packaging.
- **2.** Remove the cuvette cover and fill the cuvette with sample using the transfer pipette and replace cover.
- **3.** Place cuvette on ice for cooling purposes.
- 4. Push the banana plugs of the Safety Stand into the voltage output of the BTX generator. Warning: Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000.
- 5. Open the Safety Stand cover.
- 6. Use the thumb wheel to slide the electrodes open.
- Place a BTX cuvette in between the electrodes with the aluminum of the cuvette coming in contact with the electrodes
- **8.** Secure the cuvette in place by closing the gap with the thumb wheel. The cuvette should fit snugly between the electrodes; however it should be loose enough that it can be pulled out without adjusting the thumb wheel again.
- **9.** Following instructions for the BTX generator. Set the appropriate parameters.
- Deliver the electroporation pulse (s) to the sample.
 Warning: Use proper eye protection during electroporation.
- 11. Remove cuvette cover and extract the cell solution.
- 12. Dispose of cuvette and prepare for the next experiment.

Flat Pack Chambers

- 1. Fill the flat pack chamber with sample using a pipette.
- Push the banana plugs of the Safety Stand into the voltage output of the BTX generator. Warning: Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000.
- 3. Open the Safety Stand cover.
- 4. Use the thumb wheel to slide the electrodes open.





Appendix L: Connecting Electrodes (Continued)

- **5.** Place the Flat Pack Chamber in between the electrodes with the aluminum coming in contact with the electrodes.
- 6. Secure the Flat Pack Chamber in place by closing the gap with the thumb wheel. The Flat Pack Chamber should be snugly placed between the electrodes; however it should be loose enough that it can be pulled out without adjusting the thumb wheel again.
- **7.** Following instructions for the BTX generator, set the appropriate parameters.
- Deliver the electroporation pulse (s) to the sample.
 Warning: Use proper eye protection during electroporation.
- 9. Remove Flat Pack Chamber and extract the cell solution.
- Dispose of Flat Pack Chamber and prepare for the next experiment.

Flat Electrodes

- **1.** Fill the sterile Flat Electrode Chamber using a syringe.
- **2.** Replace the clear plastic tip so the holes in the top align with the holes in the electrode bars.
- **3.** Push the banana plugs at the opposite end of the Flat Electrode (Catalog number45-0217) into the voltage output of the BTX generator. **Warning:** Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000.
- **4.** Following instructions for the BTX generator, set the appropriate parameters.
- Deliver the electroporation pulse (s) to the sample.
 Warning: Use proper eye protection during electroporation.
- **6.** Extract the cell solution. (The chamber can be rested on ice for cooling purposes)
- 7. Clean as appropriate and prepare for the next experiment.



Appendix L: Connecting Electrodes (Continued)

Genetrodes/Genepaddles

- 1. Using the Model 515 Genetrodes holder, loosen the two plastic screws and separate the top half of the positioning plate from the holder. Place the pair of electrodes in the pr determined slots based on the necessary gap size. The electrodes must extend from the holder in the opposite direction of the holder handle. Secure the electrodes by reassembling the holder and tightening the two screws.
- **2.** Attached the micrograbber cable (catalog number 45-0216) to the electrode leads of the Genetrodes/Genepaddles.
- **3.** Push the banana plugs at the opposite end of the micrograbber cable into the voltage output of the BTX generator. **Warning:** Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000.
- 4. Prepare tissue and sample for electroporation.
- **5.** Following instructions for the BTX generator, set the appropriate parameters.
- **6.** Place the Genetrodes/Genepaddles on the sample using a micromanipulator or manually position them.
- Deliver the electroporation pulse (s) to the sample.
 Warning: Use proper eye protection during electroporation.
- **8.** Remove the electrodes carefully, clean as appropriate and prepare for the next experiment.

Tweezertrodes

- **1.** Attach the Model 524 Tweezertrode Cables to the electrode base of the Tweezertrode.
- 2. Push the banana plugs at the opposite end of the tweezertrode cable into the voltage output of the BTX generator. Warning: Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000. Then plug the banana cable into the voltage output of the BTX Generator.
- **3.** Following instructions for the BTX generator, set the appropriate parameters.







Appendix L: Connecting Electrodes (Continued)

- Prepare tissue and sample for electroporation. Grasp the tissue between the Tweezertrode electrodes and measure the interelectrode distance. Adjust generator settings if necessary. Inject the sample into the tissue.
- Deliver the electroporation pulse (s) to the sample.
 Warning: Use proper eye protection during electroporation.
- **6.** Remove the tissue carefully, clean as appropriate and prepare for the next experiment.

2-Needle Array

- 1. Grasping Model 530 or Model 532 2-Needle Array Handle, position the handle over a Model 531 or Model 533 2 Needle Array Assembly and push to secure the 2-needle array to the handle.
- Push the banana plugs at the opposite end of the 2-Needle array handle into the voltage output of the BTX generator.
 Warning: Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000. Then plug the banana cable into the voltage output of the BTX Generator.
- **3.** Following instructions for the BTX generator, set the appropriate parameters.
- **4.** Prepare tissue and sample for electroporation. Apply sample to tissue just before electroporation.
- Remove the safety shield protecting the needles, place into the tissue, and deliver the electroporation pulse(s).
 Warning: Use proper eye protection during electroporation.
- **6.** Discard the 2-needle array and prepare for the next experiment.

Microslides

 Push the banana plugs at the opposite end of the micrograbber cable (catalog number 45-0216) into the voltage output of the BTX generator. Warning: Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000.





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Appendix L: Connecting Electrodes (Continued)

- 2. Attach the Micrograbbers onto the terminal pins of the Meander Chamber slide. Polarity is not important. Tape the cable to the microscope stage to act as a strain relief and to avoid movement of the slide and its wires.
- **3.** Pipette one drop of cell suspension and reagents to the Microslides/Meander Chamber field.
- **4.** Following instructions for the BTX generator and set the appropriate parameters.
- Deliver the electroporation pulse (s) to the sample.
 Warning: Use proper eye protection during electroporation.
- **6.** Remove the microslides/meander fusion chamber carefully and prepare for the next experiment.

Petri Dish Electrode

- Plug the HV cables from the Petri Dish Electrode into the voltage output of the BTX Generator. Warning: Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000.
- 2. Prepare sample for electroporation a 35mm Petri Dish.
- **3.** Place the electrode in the 35mm Petri Dish. Allow it to gently rest on the surface of the dish.
- Following instructions for the BTX generator, set the appropriate parameters. Deliver the electroporation pulse(s) to the sample. Warning: Use proper eye protection during electroporation.
- **5.** Remove the Petri Dish Electrode carefully; clean as appropriate and prepare for next experiment.

Petri Pulser

- Plug the HV cables from the Petri Pulser into the voltage output of the BTX Generator. Warning: Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000.
- Prepare sample for electroporation in 6-well plate or in 35mm Petri Dish.





Appendix L: Connecting Electrodes (Continued)

- Place the electrode in one well of the 6-well plate or 35mm Petri Dish. Allow it to gently rest on the surface of the dish.
- **4.** Following instructions for the BTX generator and set the appropriate parameters.
- Deliver the electroporation pulse (s) to the sample.
 Warning: Use proper eye protection during electroporation.
- Remove the Petri Pulser carefully; clean as appropriate and complete delivery of pulses to all wells if using a 6-well plate.

Caliper Electrodes

- Attach the Caliper Electrodes directly to the voltage output ports of the BTX Generator. Warning: Make sure the Generator is turned off prior to connecting any cables to it. If using the Enhancer 3000 to monitor output, connect the banana plugs into the output ports of the High Voltage Probe. Use the black and red high voltage cables to connect the BTX generator to the input ports of the Enhancer 3000.
- 2. Prepare tissue for electroporation.
- **3.** Following instructions for the BTX generator, set the appropriate parameters.
- 4. Use the Caliper Electrodes to span the target tissue.
- Deliver the electroporation pulse (s) to the sample.
 Warning: Use proper eye protection during electroporation. Do not exceed 500V.
- **6.** Remove the Caliper Electrodes carefully; clean as appropriate and prepare for next experiment.



Appendix M: Plate Handler

Model HT-100 Plate Handler

1. Select a plate size

- a. Select 4mm or 2mm gap plate. For example if currently using 4 mm cuvettes use a 4 mm plate
 - The plate size should be selected according to the electric field desired, the desired volume of the sample, and the capabilities of the electroporator.

2. Load Plate with Cells

- a. Each column should be loaded with the same number of samples if possible.
 - i. Load unused wells in a column with the same sample media at the same volume.
- 3. Connect the Model HT-100 to the color coded banana cable taking care to match the color of the cable to the color ring around the connector, and then plug the banana cable into the voltage output of the electroporator again matching the color polarity.
- **4.** Place plate (HT 96) or the adapter frame containing plate (HT 25) onto the plate handler so the plate matches the nest plate outline on the handler.
- 5. Close HT-100 Plate Handler lid firmly to latch. The front panel latch pops out when securely closed.
 - a. When the lid is closed correctly, the LED for column 1 should flash.
- **6.** Select appropriate column (1 12) to begin electroporation by using the column adjust buttons.
 - a. The unit will default to column 1 when lid is closed.
 - b. Pressing and holding the button allows the unit to rapidly advance through columns after a short pause.

Appendix M: Plate Handler (Continued)

- 7. Configure the electroporator with appropriate settings for voltage, resistance and time capacitance
 - a. Set the electroporator for the total number of pulses per well
 - b. Press pulse on the electroporator to initiate the pulse(s) for that column, the plate handler will beep.
 - c. The column LEDs will flash to indicate the active column. Once a column has been electroporated the LED remains steady until the cover is opened.

NOTE: Once the cover is opened the memory of pulsed columns if wiped out.

d. Press the column select button to switch to the next column.

Plate handlers may be used with both types of HT plates (HT 96 and HT 25). The following will outline the steps needed to configure the unit for the type of plate being used.





The Electroporation Experts

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