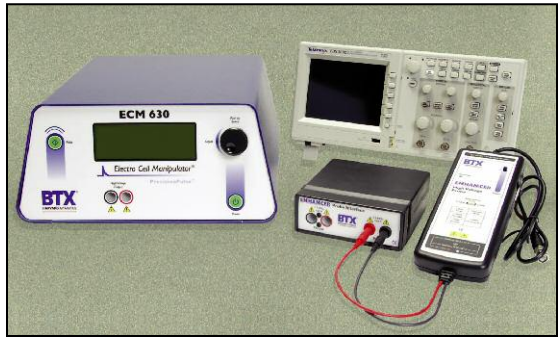
User's Manual

• Enhancer 3000® High Voltage Probe
• Enhancer 3000® High Voltage Interface Box

Including Basic Instructions for Use With
Tektronix®TDS1002B Oscilloscope &
Enhancer Probe Interface & Enhancer High Voltage Probe



45-0057	Enhancer Probe®, Enhancer 3000® Interface Box and Cables
45-0059	Enhancer Probe®, Enhancer 3000® Interface Box, TDS1002B Oscilloscope and Cables (110V)
45-0059INT	Enhancer Probe®, Enhancer 3000® Interface Box, TDS1002B Oscilloscope, and Cables (220V)

BTX®
HARVARD APPARATUS
The Electroporation Experts

Publication 5500-002-REV-C

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.**

Table of Contents

General Information

Serial Number

Serial numbers for the Enhancer 3000®, Tektronix TDS1002B, are 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 Enhancer 3000® Probe or Enhancer 3000® Interface Box. Calibration for the Tektronix TDS1002B Oscilloscope must be arranged through an outside test equipment calibration service. BTX – Harvard Apparatus does not supply calibration services for these oscilloscopes.

Warranty

BTX - Harvard Apparatus warranties the Enhancer 3000® and Enhancer 3000® for a period of one year 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 damage resulting from misuse, neglect or abuse, normal wear and tear, or accident. This warranty extends only to the original customer purchaser.

Failure to use the Enhancer 3000 High Voltage probe to connect a BTX Generator to an external digital oscilloscope for monitoring will result in voiding your warranty; connecting directly to the external monitoring equipment or modified monitoring setup will damage the Generator.

Warranties on the Tektronix, TDS1002B is covered through Tektronix, Inc., P.O. Box 500, Beaverton, OR 97077.

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.

If a defect arises within the one-year warranty period, promptly contact **BTX – Harvard Apparatus, 84 October Hill Road, Holliston, Massachusetts 01746-1388** using out toll free number: **1-800-272-2775.** 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.

General Information (Continued)

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.

Caution

This apparatus is for laboratory research use only and is not for clinical use on patients.

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CAUTION

FOR RESEARCH USE ONLY
NOT FOR CLINICAL
USE ON PATIENTS

General Safety Summary

Please read the following safety precautions to ensure proper use of your Enhancer 3000® High Voltage Probe and Enhancer 3000® Interface Box. To avoid potential hazards and product damage, use these products only as instructed in this manual. If the equipment is used in a manner not specified by the manufacturer, the protection provided by the equipment may be impaired.

To Prevent Hazard or Injury:

OBSERVE ALL TERMINAL RATINGS

To avoid injury or product damage, do not use these products above 2500Vrms CAT II between each input lead and earth or above 5000Vrms CAT II between two inputs. This voltage rating applies to both the 1/100 & 1/1000 settings on the Enhancer 3000®.

GROUND THE ENHANCER 3000®

The Enhancer 3000® is grounded with the shell of the BNC connector through the grounding conductor of the power cord of the measurement equipment.

Before making connections to the input of the Enhancer 3000® Interface Box or Enhancer 3000® High Voltage Probe, make sure that the measurement equipment is properly grounded and that the BNC connector of the Enhancer 3000® is securely connected to the BNC input connector of the measurement equipment.

MAKE PROPER CONNECTIONS

Make sure all connections are made properly and securely.

DO NOT OPERATE WITHOUT COVERS

To avoid electric shock or fire hazard, do not operate these products with the covers removed.

AVOID EXPOSED CIRCUITRY

Do not touch exposed connections when power is present.

General Safety Summary (Continued)

To Prevent Hazard or Injury (Cont'd)

USE PROPER POWER SOURCE

Use four AA cells or 6VDC/200mA or 9VDC/120mA to power the Enhancer 3000®. Do not operate the Enhancer 3000® from a power source that applies more than the voltage specified.

DO NOT OPERATE WITH SUSPECTED FAILURES

If damage is suspected on or to the products, do not operate. Contact qualified service personnel to perform inspection.

DO NOT OPERATE IN EXPLOSIVE ATMOSPHERE

To avoid injury or fire hazard, do not operate these products in an explosive environment.

DO NOT OPERATE IN WET/DAMP CONDITIONS

To avoid electric shock, do not operate these products in wet or damp conditions.

OBSERVE ALL WARNING LABELS ON PRODUCT

Read all labels on the products to ensure proper usage.

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

Protective
(Earth)
Terminal

Functional
Ground
Terminal

Introduction

The BTX Enhancer 3000® High Voltage Probe and the Enhancer 3000® High Voltage Interface Box provide a safe means of monitoring high voltage signals with standard oscilloscopes and other monitoring equipment. For monitoring pulse waveforms, a digital storage oscilloscope is recommended. BTX – Harvard Apparatus recommends use of the Tektronix TDS1002B digital storage oscilloscope or equivalent. If waveform printing or data transfer to a personal computer is required, the Tektronix TDS1002B has the capacity for this utility.

When the Enhancer 3000® monitoring system is used with electroporation / electrofusion generators, actual pulse parameters across the load may be observed. This allows confirmation of pulse amplitudes, pulse widths, pulse intervals, exponential-decay time constants, and AC alignment waveforms.

The Enhancer 3000® monitoring systems consists of the following:

Catalog No. **45-0057**

- Enhancer 3000® High Voltage Probe
- Enhancer 3000® High Voltage Interface Box
- Sheathed Banana Plug Cables, 3 ft. (1 Red, 1 Black)

Catalog No. **45-0059 (110V)**
45-0059INT (220V)

- Enhancer 3000® High Voltage Probe
- Enhancer 3000® High Voltage Interface Box
- Sheathed Banana Plug Cables, 3 ft. (1 Red, 1 Black)
- Tektronix TDS1002B Digital Storage Oscilloscope

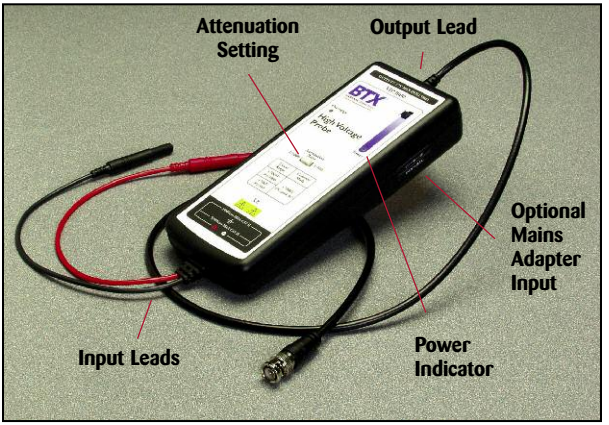
Note: *Low voltage X1/X10 probes are not included with the TDS1002B Oscilloscope supplied in these systems.*

45-0057

45-0059 (110V)
45-0059INT (220V)

Introduction (Continued)

Enhancer 3000® Probe Features



Enhancer 3000® Indicators

Power Indicator – Lights red when power is applied and unit is switched on.

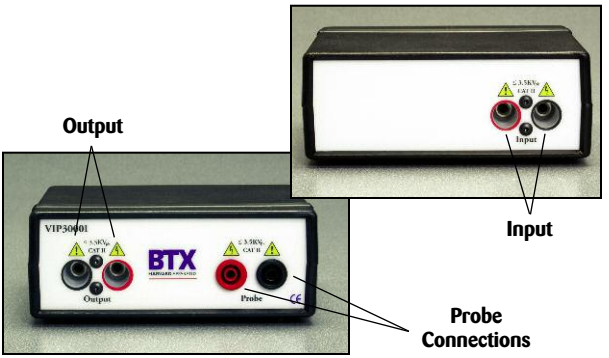
Low Battery Indication – Power indicator flashes red when batteries are low.

Overrange Indicator – Lights red if the voltage of the input signal exceeds the linear operating range of the probe. When this happens, the signal on the probe output does not accurately represent the signal on the probe input.

Enhancer 3000® Attenuation Settings

1/100 & 1/1000 Selectable Attenuation

Enhancer 3000® Interface Box Features



Installation

Initial Setup

1. Remove equipment from packing material.
2. Visually inspect equipment to verify that no damage has occurred during shipping and that all equipment purchased has been received.
3. If the Tektronix TDS1002B Oscilloscope was purchased, connect the three wire, molded power plug shipped with the unit to the power entry jack on the side of the unit. Use only an approved AC line cord with a molded IEC320/C13 connector certified for country of use. Only connect to grounded power receptacles to ensure proper grounding; do not use adapter plugs.

WARNING. *Only use approved line cords to ensure proper grounding.*

4. The Tektronix TDS1002B Oscilloscope as supplied through BTX – Harvard Apparatus does not include low voltage X1/X10 oscilloscope probes. The sections in the Tektronix TDS1002B User Manual pertaining to these probes are not applicable for this test system.
5. Read all of the user manuals to become familiar with all features and functions of the equipment.

Installation (Continued)

Installing Batteries in the Enhancer 3000®

Remove the battery compartment cover at the rear side of the unit. Install four AA cells following the polarity indication markings. Replace the battery compartment cover.

AC Adaptor Charge Available

Plug charger adaptor to the Voltage Interface Probe connector and to wall outlet.

Connecting an Optional Mains Adapter to the Enhancer 3000®

If battery powered operation is not desired, an appropriate mains adapter may be connected to the power jack on the side of the unit. The supplied voltage must be less than 12VDC and greater than 4.4VDC, otherwise damage to the probe could result. Polarity is '+' inside and '-' outside. If the polarity connection is incorrect, the unit will not power up. A built-in circuit protects the probe from damage in this situation.

CAUTION. *Only use mains adapters which meet the required specifications.*

Location Requirements

- A sturdy, level, clean, non-flammable, dry surface
- Adequate power supply
- Room temperature 0˚C to +40˚C (32˚F to 104˚F)
- Relative Humidity of 25 to 85%
- A non-explosive atmosphere

System Connections

WARNING. *Follow all safety precautions listed in the equipment's user manuals.*

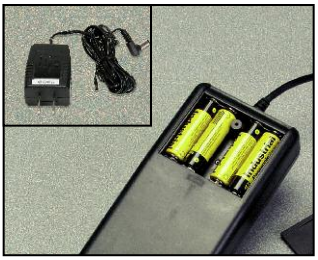
Connecting Enhancer 3000® to an Oscilloscope

Connect the BNC connector on the output lead to one of the oscilloscope's vertical input BNC connectors. Only use an approved line cord with the oscilloscope to ensure that the oscilloscope and the Enhancer 3000® are properly grounded.

WARNING. *Only use approved line cords to ensure proper grounding.*

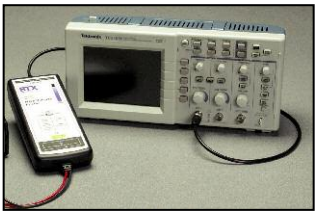
Connecting Enhancer 3000® to Enhancer 3000®

Connect the sheathed banana plug input cables of the Enhancer 3000® Probe to the probe output jacks of the Enhancer 3000®. Be sure to match the red/black color-coding of the cables to the connectors.



Installing Batteries in the Enhancer 3000®

Connecting an Optional Mains Adapter to the Enhancer 3000®



Connecting Enhancer 3000® to an Oscilloscope

Connecting Enhancer 3000® to Enhancer 3000®

Installation (Continued)

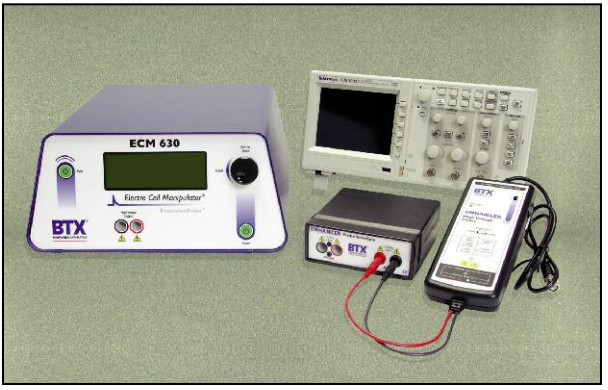
Connecting Electroporation/Electrofusion Generators to Enhancer 3000®

Using the sheathed banana plug cables provided, connect the Electroporation / Electrofusion Generator output to the Enhancer 3000® input. Be sure to match the red/black color-coding of the cables to the connectors.

WARNING. *Only use approved cables for high voltage connections. Replace any frayed or cut cables to prevent arcing to personnel or equipment. Turn off electroporation / electrofusion generators before connecting / disconnecting cables.*

Connecting Loads to Enhancer 3000®

Connect the sheathed banana plug cables provided with the load (i.e. safety stand, plate handler, electrodes, etc.) to the Enhancer 3000® output. Be sure to match the red/black color-coding of the cables to the connectors.



Operation: Getting Started

Ensure all connections are securely made as per the installation instructions.

Applying Power

Turn on power to oscilloscope, Enhancer 3000®, and Electroporation / Electrofusion Generator.

Electroporation / Electrofusion Generator Settings

Set experiment parameters on your Electroporation / Electrofusion Generator.

NOTE: Refer to your Electroporation / Electrofusion Generator user's manual for detailed setup information.

Enhancer 3000® Probe Attenuation Setting

Set the Enhancer 3000® attenuator selector switch as follows:

Use 1/100 for Generator LV Mode (500V or less)

Use 1/1000 for Generator HV Mode (505V and higher)

Basic Oscilloscope Settings

NOTE: Refer to Oscilloscope User's Manual for detailed operation instructions.

The most important oscilloscope controls and settings to understand are Vertical Scale and Position (Volts per division), Horizontal Scale and Position (Seconds per division), Trigger Level, Trigger Position and Trigger Mode. It is also important to set the probe attenuation at the oscilloscope to match the attenuation setting of the Enhancer 3000®.

DEFINITIONS

Division: The oscilloscope screen is marked with a grid. Each line in the horizontal and vertical direction represents a division.

Vertical Scale: The vertical scale (volts per division) determines how many volts each horizontal line on the oscilloscope display represents. For example, if the vertical scale is set for 25V/DIV and the signal goes up two lines, then the signal represents a 50V signal.

Horizontal Scale (Time Base): The horizontal scale (seconds per division) defines how much time each vertical line on the oscilloscope display represents. For example, if your pulse traverses 5 divisions and your horizontal scale is set for 200ms/div, then the pulse is 1 second in length.

Operation: Getting Started (Continued)

Basic Oscilloscope Settings (Cont'd)

DEFINITIONS (Cont'd)

Trigger Level: The Trigger Level defines the amplitude of input signal required to cause the oscilloscope to take a sample in time of the input from the probe. If the trigger level is set too high, say 100V when trying to measure a 75V pulse, then the oscilloscope will not trigger and the waveform will not be output on the screen. If the trigger level is set too low, say around 1V or 2V for the same 75V pulse, then random circuit noise can cause false triggers. Trigger Level should generally be set to 50% of Voltage set-point.

Trigger Position: Trigger Position determines where on the time scale the trigger event will be placed. It is important to keep the trigger position on screen while acquiring waveforms. It is necessary to check trigger position anytime the horizontal scale (time base) setting is adjusted.

Trigger Mode: Trigger Mode defines how the oscilloscope treats triggers. If it is set to Auto, the oscilloscope displays the input in real time until a trigger event occurs. It then holds the waveform for a moment until returning back to real time display. If it is set to Normal, then the oscilloscope refreshes the display every trigger event. If it is set to Single, the oscilloscope will only display one trigger event and then requires the user to reset the oscilloscope to get a fresh trigger. Trigger Mode also encompasses the rising edge or falling edge setting. If set to rising edge then the trigger event happens when the signal crosses the trigger level while going from a lower voltage to a higher voltage.

Probe Attenuation Setting: For the TDS1002B oscilloscope, to change (or check) the probe attenuation setting, press the VERTICAL MENU button (of the input channel you are using), and then press the menu selection next to Probe until the correct setting is displayed. The attenuation setting should be set to match the probe attenuation setting (i.e. if the Enhancer 3000® is set to 1/100, set the oscilloscope to 100X).

Operation: Getting Started (Continued)

Functional Checks

Functional Check using Exponential Decay Wave Generators

1. Verify or set the Enhancer 3000® attenuation to 1/100 and verify or set the oscilloscope probe attenuation to 100X. Set DC voltage at about 200V and capacitance / resistance to values that will lead to a pulse with a time constant of approximately 50 msec. For example, resistance set to 1000 ohms and capacitance set to 50 microfarads. Adjust oscilloscope settings as directed in oscilloscope manual.
2. Adjust both Vertical and Horizontal positions to zero.
3. Set Vertical Scale to 50V/div.
4. Adjust Horizontal Scale to 25msec/div.
5. Set the trigger level to 100V, rising edge, normal mode. Make sure trigger is set for the proper input channel.
6. Follow the generator instructions for the initiation of the pulse. Verify that the oscilloscope has recorded the pulse and is displaying it on the screen.

Functional Check using Square Wave Generators

1. Verify or set the Enhancer 3000® attenuation to 1/100 and verify or set the oscilloscope probe attenuation to 100X. Set DC voltage at about 200V and a pulse length of 50 msec. Adjust oscilloscope settings as directed in oscilloscope manual.
2. Adjust both Vertical and Horizontal Positions to zero.
3. Set Vertical Scale to 50V/div.
4. Adjust Horizontal Scale to 25msec/div.
5. Set the Trigger Level to 100V, rising edge, normal mode. Make sure trigger is set for the proper input channel.
6. Follow the generator instructions for the initiation of the pulse. Verify that the oscilloscope has recorded the pulse and is displaying it on the screen.

Operation: Waveform Monitoring

Monitoring an Exponential Decay Wave Pulse

1. Verify all equipment is properly connected and powered on.
2. Choose appropriate Vertical Scale (Volts/Div) setting on the oscilloscope. Generally the volts per division should be one third to one quarter the voltage set-point on the generator. Next chose appropriate Horizontal Scale (Sec/Div) setting. This should be about one quarter the length of the pulse. Set the trigger level to one half the voltage setpoint on the generator. Trigger settings should be on rising edge and normal.
3. Send one pulse and adjust Vertical Scale (Volts/Div) and Horizontal Scale (Sec/Div) as well as Vertical and Horizontal Position to display the waveform on oscilloscope. Proceed with monitoring of experiment.

Monitoring a Square Wave Pulse

1. Verify all equipment is properly connected and powered on.
2. Choose appropriate Vertical Scale (Volts/Div) setting on the oscilloscope. Generally the volts per division should be one third to one quarter the voltage set-point on the generator. Next chose appropriate Horizontal Scale (Sec/Div) setting. This should be about one quarter the length of the pulse. Set the trigger level to one half the voltage setpoint on the generator. Trigger settings should be on rising edge and normal.
3. Send one pulse and adjust Vertical Scale (Volts/Div) and Horizontal Scale (Sec/Div) as well as Vertical and Horizontal Position to display the waveform on oscilloscope. Proceed with monitoring of experiment.

Operation: Waveform Monitoring (Continued)

Monitoring Alternating Current - Voltage and Frequency

1. Verify all equipment is properly connected and powered on.
2. For the pulse to be monitored, the voltage must be greater than the trigger level position indicated on the screen. To increase or decrease the trigger level, adjust the trigger position with the Trigger Level Knob. Change the voltage resolution by adjusting the Vertical Scale (Volts/Div) setting as needed. Change the pulse length resolution by adjusting the Horizontal Scale (Sec/Div) as needed. Change the baseline position by adjusting the Vertical Position as needed.
3. Deliver a pulse and visualize the pulse profile. Make adjustments as desired. These settings will be saved for the next experiment.

Note: For fusion experiments utilizing both AC alignment and DC pulsing, use the above instructions to monitor the AC alignment and calculate the appropriate AC parameters. Follow the Monitoring a Square Wave Pulse instructions to monitor the DC pulse and calculate the appropriate DC parameters.

Monitoring Guidelines for BTX Generators

ECM® 2001

Follow instructions in the previous section for AC alignment and DC square wave pulse monitoring.

ECM® 830 ELECTROPORATOR, SQUAREWAVE

Follow the instructions in the previous section for square wave pulse monitoring.

ECM® 630, ECM® 399

Follow the instructions in the previous section for exponential decay wave pulse monitoring.

Operation: Waveform Monitoring (Continued)

Measurement of Key Parameters

ELECTROPORATION - DC PARAMETERS

Note about Automatic Oscilloscope Measurements: After monitoring a pulse, some oscilloscopes will display various measurements. Some combinations of electrode and buffers will allow some overshooting on the rising edge of a pulse. The automatic measurement will then measure the maximum voltage achieved during the overshoot. A manual measurement of the voltage using the cursor menu would then be indicated.

Note: The following directions are generic and reflect Tektronix TDS Series controls. Please refer to your oscilloscope owner's manual for specific cursor controls.

MANUAL PEAK VOLTAGE MEASUREMENT

1. Stabilize the display by pressing the Run/Stop key.
2. Press the Cursor key.
3. Change the cursor type to voltage.
4. Change the source to reflect the input channel used.
5. Move Cursor 1 to the 0-volt baseline by turning the Cursor 1 Adjust knob.
6. Move Cursor 2 to the top of the waveform by turning the Cursor 2 Adjust knob. The voltage measurement is indicated in the window labeled "Delta".
7. The display can be re-enabled by pressing the Run/Stop key.

FIELD STRENGTH CALCULATION

1. Obtain the value of the peak output voltage applied.
2. The field strength is obtained using the following equation: $E = Vp/d$

Where E is the Field Strength in kV/cm or V/cm, Vp is the voltage in kV or V, and d is the chamber gap in cm.

Operation: Waveform Monitoring (Continued)

Measurement of Key Parameters (Cont'd)

PULSE LENGTH MEASUREMENT

1. Stabilize the display by pressing the Run/Stop key.
2. Press the Cursor key.
3. Change the cursor type to time.
4. Change the source to reflect the input channel used.
5. Move the left most vertical bar (Cursor 1) to the beginning of the pulse by turning the Cursor Adjust knob.
6. Move the right most vertical bar (Cursor 2) to the end of a square pulse or to the point where an exponential decay curve reaches 1/e (0.37) of the peak voltage. Rotating Cursor 2 Adjustment knob moves the right cursor.
7. The time measurement is indicated in the window labeled "Delta".
8. The display may be re-enabled by pressing the Run/Stop key.

Electrofusion-AC/RF Parameters

AUTOMATIC AC/RF AMPLITUDE MEASUREMENT

1. Enter the measurement menu by pressing the Measure key.
2. Change the source to reflect the input channel used.
3. Change the measurement type to Peak to Peak for voltage measurement.
4. Change the measurement type to frequency for frequency measurement.

MANUAL AC/RF AMPLITUDE MEASUREMENT

1. Stabilize the display by pressing the Run/Stop key.
2. Press the Cursor key.
3. Change the cursor type to voltage.
4. Change the source to reflect the input channel used.
5. Move Cursor 1 to the bottom peaks of the AC waveform by turning the Cursor 1 Adjustment knob.
6. Move Cursor 2 to the top peaks of the AC waveform by turning the Cursor 2 Adjustment knob. The Peak-to-Peak voltage measurement is indicated in the window labeled "Delta".

Operation: Waveform Monitoring (Continued)

Electrofusion-AC/RF Parameters (Cont'd)

AC/RF DURATION MEASUREMENT

1. Stabilize the display by pressing the Run/Stop key.
2. Adjust pulse width knob so that an entire AC wave is displayed.
3. Press the Cursor key.
4. Change the cursor type to time.
5. Change the source to reflect the input channel used.
6. Move Cursor 1 to the beginning of the AC pulse by turning the Cursor Adjustment knob.
7. Move Cursor 2 to the end of the AC pulse by turning the Cursor Adjustment knob. The AC/RF duration measurement is indicated in the window labeled "Delta." The unit for pulse duration is in seconds.
8. Note: The maximum length of time the display will hold is 50 seconds. If the protocol uses a longer AC length then it can not be measured with this equipment.

FREQUENCY MEASUREMENT

1. Stabilize the display by pressing the Run/Stop key.
2. Press the Cursor key.
3. Change the cursor type to time.
4. Change the source to reflect the input channel used.
5. Move Cursor 1 to the middle of one positive peak by turning the Cursor 1 Adjustment knob.
6. Move Cursor 2 to the middle of the next positive peak by turning the Cursor 2 Adjustment knob. The frequency of the AC waveform is indicated in the window labeled "Delta". The unit for frequency is Hertz (Hz).

Operation: Waveform Monitoring (Continued)

Electrofusion-AC/RF Parameters (Cont'd)

AC TO DC RAMP TIME MEASUREMENT

1. Adjust the Horizontal Scale to about 250µs/div.
2. Stabilize the display by pressing the Run/Stop key.
3. Press the Cursor key.
4. Change cursor type to time.
5. Change the source to reflect the input channel used.
6. Readjust the Horizontal Scale to 25µs.
7. Move Cursor 1 to the end of the AC pulse by turning the Cursor Adjustment knob.
8. Move Cursor 2 to the beginning of the square pulse by turning the Cursor 2 adjustment knob. The Ramp Time measurement is indicated in the middle window labeled Delta.

Basic Waveform Printing

1. Turn off the oscilloscope and printer.
2. Connect the oscilloscope to the printer using the appropriate cable.
3. Turn on the oscilloscope and printer.
4. Push UTILITY > Options > Hard Copy Setup.
5. Change the settings to match those of your printer.
6. Push the PRINT or HARDCOPY button. The printer should begin printing a copy of the oscilloscope screen after a few seconds.

General Cleaning and Maintenance

Enhancer 3000® & Enhancer 3000®

To clean exterior surfaces, use a soft, lint-free cloth to remove loose dust. For more efficient cleaning, use a soft cloth dampened with water or an aqueous solution of 75% isopropyl alcohol. Do not immerse the Enhancer 3000® or Enhancer 3000®. Avoid using abrasive cleaners. Avoid using chemicals containing benzene or similar solvents.

The Enhancer 3000® requires no maintenance. If the Enhancer 3000® Probe does not work properly, contact BTX - Harvard Apparatus for appropriate instructions.

Tektronix TDS1002B Oscilloscope

Refer to the Tektronix user's manuals for cleaning and maintenance instructions.

Appendix A: Specifications

Enhancer 3000® High Voltage Probe Specifications

Attenuation	1:100 / 1:1000
Bandwidth	DC to 70 MHz (-3 dB)
Accuracy	±2%
Input Impedance	10 M Ω// 10 pF each side to ground
Input Voltage:	
Differential Range[1]	±700 V (DC+Peak AC) or 700 Vrms @ 1/100 ±7000 V (DC+Peak AC) or 5000 Vrms @ 1/1000
Common Mode Range[1]	±7000 V (DC+Peak AC) or 2500 Vrms @ 1/100 & 1/1000
Absolute Max. Voltage[1]	±7000 V (DC+Peak AC) or 2500 Vrms @ 1/100 & 1/1000 in common mode ±7000 V (DC+Peak AC) or 5000 Vrms @ 1/100 & 1/1000 in differential mode
Output Voltage:	
Max. Amplitude	±7 V (into 50k ohm load)
Offset (typical)	< ±5 mV
Noise (typical)	0.9 mVrms
Source Impedance (typical)	50 Ω
CMRR (typical)	-80 dB @ 60 Hz, -50 dB @ 1 MHz
Ambient Operating Temperature	-10˚ to 40˚C
Ambient Storage Temperature	-30˚ to 70˚C
Ambient Operating Humidity	25 to 85% RH
Ambient Storage Humidity	25 to 85% RH
Pollution Degree	2 (Not to be operated in conductive pollutants atmosphere)
Power Requirements:	
Standard	4 AA cells or 6 VDC/200 mA mains adapter[2] or regulated 9 VDC/120 mA mains adapter[2]
Length of BNC Cable	88.9 cm (35 in)
Length of Input Leads	30.48 cm (12 in)
Weight	454 g (1 lb)
Dimensions, L x W x H	20.3 x 8.3 x 3.8 cm (8 x 3.25 x 1.5 in)

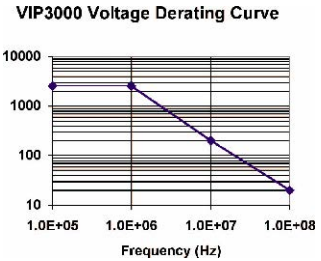
Notes:

1. *Voltage limit is the lesser of the DC + Peak AC and RMS values.*
2. *a. The supplied voltage must be less than 12 V and greater than 4.4 V, otherwise the probe could be damaged or will not operate properly.*
   b. Polarity is "+" inside and "-" outside. For wrong polarity, a built-in circuit protects the probe; no danger or damage will occur.
   c. When the voltage of the batteries become too low, the power indicator will flicker.

Appendix A: Specifications (Continued)

Enhancer 3000® Derating Curve

The derating curve of the absolute maximum input voltage in common mode is shown to the right.



Enhancer 3000® High Voltage Interface Box Specifications:

Input, Output, Probe Connections:

Type	4 mm Banana Jack
Voltage	2500 Vrms, 3500 Vpk CAT II
Current (Max. Continuous)	15 A
Current (Peak)	1000 A for 100 µs
Dimensions. L x W x H	13.5 x 13.5 x 5.8 cm (5.3 x 5.3 x 2.3 in)
Weight	330 g (0.73 lbs)

High Voltage Cable Specifications

Type	Retractable Sheath Banana Plug
Length	91.44 cm (3 ft)
Wire Gauge	18 AWG
Insulation	PVC
Voltage Rating	1000 Vrms IEC 1010-2-31, Category III
Current	10 A
Operating Temperature	+55° C (+131° F) Max.

Enhancer 3000® & Enhancer 3000® Safety Compliance

The Enhancer 3000® HV Probe and Enhancer 3000® HV interface Box are in compliance with the following specifications as listed in the Official Journal of the European Communities:

LOW VOLTAGE DIRECTIVE 73/23/EEC, AS AMENDED BY 93/68/EEC:

EN61010-1/A2:1995
Safety requirements for electrical equipment for measurement, control, and laboratory use.

EN61010-2-031:1994
Particular requirements for hand-held probe assemblies for electrical measurement and test equipment.

EC Declaration of Conformity–Low Voltage

Appendix B: Accessories and Replacement Parts

Catalog No.	Description
45-0057	Enhancer 3000® with Interface Box and Cables
45-0059	Enhancer 3000SC® with Interface Box, Oscilloscope, Communications Module and Cables, (110 V)
45-0059INTPP	Enhancer 3000SC® with Interface Box, Oscilloscope, Communications Module and Cables, (220 V)
45-0062	Enhancer 3000® High Voltage Interface Box
45-0063	Enhancer 3000® High Voltage Probe
45-0064	Oscilloscope, Tektronix TDS1002B w/o 1X/10X Low Voltage Probes
45-0066	Cable Set, Patch Cord, Banana Plug, Red/Black, 3 ft. (Consists of qty. 1 each red and black cables)

45-0062

45-0063

45-0064

45-0066

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 and electrofusion efficiencies, including addition of EDTA, “intracellular salts”, and serum prior to 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.

Optimizing with the Enhancer 3000®

Use the Enhancer 3000® and an oscilloscope to assist in protocol optimization through their monitoring, display, and calculation capabilities.

ELECTROPORATION

1. Vary the voltage in order to vary the field strength kv/cm, 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 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 kv/cm 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 C: Optimization Strategies (Continued)

ELECTROFUSION

AC Alignment

1. Set AC amplitude and frequency. Initiate alignment manually, monitoring the process visually on an inverted microscope. When alignment is optimal, discontinue alignment and initiate DC fusion. Use oscilloscope and the Enhancer 3000® to visually verify the AC, AC amplitude, and AC frequency.
2. If AC amplitude is unknown, initiate alignment manually, and begin increasing the amplitude from 0 V, until optimal cell motion is observed. When alignment is optimal, discontinue alignment and initiate DC fusion. Use oscilloscope and the Enhancer 3000® to visually verify the AC, calculate amplitude, and verify frequency.
3. Record duration of AC alignment for further experiments in an automatic mode. Use oscilloscope and the Enhancer 3000 to verify the AC duration.

DC Fusion

1. Vary the voltage in order to vary the field strength, keeping other parameters constant. Assay sample for both viability and fusion yields. Plot the field strength versus both viability and fusion yields, and extrapolate the optimal field strength and voltage. Use oscilloscope and the Enhancer 3000® to verify voltage and field strength.
2. Vary the pulse length at the optimal voltage. Assay sample for both viability and fusion yield. Plot the pulse length versus both viability and fusion yields, and extrapolate the optimal pulse length. Use oscilloscope and the Enhancer 3000 to verify pulse length.
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 fusion yields. Plot the number of pulses versus both viability and fusion yields, and extrapolate the optimal number of pulses. Use oscilloscope and the Enhancer 3000® to verify field strength, pulse length, and number of pulses.

Appendix D: Experimental Troubleshooting

1. **Arcing:** Verify electrical component functionality. Verify properties of cell sample (do cells need to be washed? Verify properties of transfectant/molecule (Is the buffer appropriate for application?). Try reducing the voltage until arcing is no longer a problem. Use oscilloscope and the Enhancer 3000® to verify pulse profile, actual peak voltage, and pulse length during an arc.
2. **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 correct? Pulse length or appropriate instrument settings correct? Number of pulses correct? If so, follow Optimization Guidelines outlined in Appendix C. Use oscilloscope and the Enhancer 3000® to verify pulse delivery, peak voltage, field strength, and pulse length.
3. **Low viability:** 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, reduce voltage, pulse length, or number of pulses and re-optimize protocol to improve viability as outlined in Appendix C. Use oscilloscope and the Enhancer 3000 to verify pulse delivery, peak voltage, field strength, pulse length, and number of pulses.
4. **No/negligible alignment:** Verify physical, biological, and chemical parameters. Verify frequency and amplitude. Re-optimize amplitude and duration as outlined in Appendix C. Use oscilloscope and the Enhancer 3000® to verify AC, frequency, and amplitude.
5. **Low fusion yield:** Verify physical, biological, and chemical parameters. Troubleshoot alignment as outlined in 4. Verify delivery of DC fusion pulse, voltage, field strength, and pulse length. Re-optimize the DC fusion parameters as outlined in Appendix C. Use oscilloscope and the Enhancer 3000 to verify DC pulse, voltage, field strength, pulse length, and number of pulses.