IPA® / DOUBLE IPA®

Integrated Patch Amplifier

ELECTROPHYSIOLOGY
PATCH-CLAMP SYSTEM

WITH

SutterPatch® SOFTWARE

Operation Manual

SUTTER INSTRUMENT

One Digital Drive
Novato, CA 94949 USA

Voice: +1 415-883-0128
Fax: +1 415-883-0572
Web: www.sutter.com
Email: info@sutter.com

The IPA and Double IPA systems have two international safety certifications:

1) The CE mark is for compliance to health, safety and environmental protection standards for products sold within the European Economic Area:

2) The RoHS (Restriction of Hazardous Substances) Directive 2002/95/EC restricts the use of hazardous substances for electronic equipment sold within the European Union:

DISCLAIMER

The **IPA** system consists of one electronic amplifier with integrated digitizer and one headstage. The **Double IPA** system consists of one electronic amplifier with integrated digitizer and two headstages. All references to an IPA system also include a Double IPA system, unless otherwise noted. The purpose of the system is for the stimulation and measurement of cellular preparations. No other use is recommended.

This instrument is designed for use in a laboratory environment. It is not intended for, nor should it be used in human experimentation or applied to humans in any way. This is not a medical device.

Do not open or attempt to repair the instrument.

Do not allow an unauthorized and/or untrained operative to use this instrument.

Any misuse will be the sole responsibility of the user/owner, and Sutter Instrument Company assumes no implied or inferred liability for direct or consequential damages from this instrument if it is operated or used in any way other than for which it is designed.

SAFETY WARNINGS AND PRECAUTIONS

Electrical

- **Operate the IPA system using 100 – 240 VAC, 50 - 60 Hz line voltage. This instrument is designed for use in a laboratory environment that has low electromagnetic noise and mechanical vibration. Surge suppression is recommended at all times.**

Fuse Replacement: Replace only with the same type and rating:

Line Voltage: 100 – 240 VAC	
Fuse Rating	Manufacturer Examples
	RoHS Compliant (Lead Free)
T2.0, 250V	Bussmann: GMC-2-R, S506-2A Littelfuse: 239.002.P

IPA & DIPA Fuses

Type: 5 x 20 mm glass tube, Medium Time Delay (Slow Blow), RoHS compliant.
Rating: T2.0A 250V (Time Delay, 2 Amps, 250 Volts)
Examples: Bussmann: GMC-2-R, S506-2A
Littelfuse: 239.002.P

- Avoiding Electrical Shock and Fire-related Injury
- Always use the grounded power cord provided to connect the system's power adapter to a grounded/earthed mains outlet. This is required to protect you from injury in the event that an electrical hazard occurs.
- Do not disassemble the system. Refer servicing to qualified personnel.
- To prevent fire or shock hazard, do not expose the unit to rain or moisture.

Operational

Failure to comply with any of the following precautions may damage this instrument.

- This instrument is designed for operation in a laboratory environment (Pollution Degree I) that is free from mechanical vibrations, electrical noise and transients.
- Operate this instrument only according to the instructions included in this manual.
- Do not operate this instrument near flammable materials. The use of any hazardous materials with this instrument is not recommended and, if undertaken, is done so at the users' own risk.
- Do not operate if there is any obvious damage to any part of the instrument.

Other

- Retain the original packaging for future transport of the instrument.
- Sutter Instrument Company reserves the right to change specifications without prior notice.
- Use of this instrument is for research purposes only.

Handling Micropipettes

⚠ **Failure to comply with any of the following precautions may result in injury to the users of this instrument as well as those working in the general area near the instrument.**

- **The micropipettes used with this instrument are very sharp and relatively fragile. Avoid contact with micropipette tips to prevent accidentally impaling yourself.**

- **Always dispose of micropipettes by placing them into a well-marked, spill-proof "sharps" container.**

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1. INTRODUCTION

Welcome to the newest breakthroughs in patch-clamp technology! Our passion is creating the finest available electrophysiology research instrumentation. With over two centuries of combined experience from across the patch-clamp industry, our expert team has designed new microelectrode amplifier-recording systems that are powerful enough to satisfy experienced patch-clampers, yet easy-to-use for recent entrants.

Sutter Instrument Company is a leading manufacturer of innovative precision instrumentation in the neuroscience field. We have a worldwide reputation for the highest quality and performance of pipette pullers, micromanipulators, light sources and wavelength switchers. We are proud to apply this same commitment of excellence to the next generation of patch-clamp instrumentation.

1.1 Overview

Advanced Design

The IPA® Integrated Patch Amplifier is the world's first fully integrated microelectrode patch-clamp system, which facilitates and streamlines your experimental setup. All of the electronics (amplifier and digitizer) used in stimulating, compensating and recording from cells are integrated by design into a single printed circuit board (the Double IPA adds a second PCB).

The accompanying SutterPatch® software brings the controls and displays for full-featured data acquisition, data analysis, and graphics/layout together into a single, unified program, including a software control panel for direct access to all of the IPA amplifier functions.

The SutterPatch software was developed in the powerful Igor Pro system environment. Igor Pro, by WaveMetrics, Inc., is a data collection, management and analysis platform with a rich set of built-in functions and routines for scientific programs.

From concept to production, from hardware to software, these fully integrated systems provide leading-edge patch-clamp systems that are affordable, and easy-to-setup and use.

1.2 Software Highlights

- Full-featured electrophysiology package
- Single program for data acquisition, analysis and hardware control
- Complex experimental automation
- Publication-quality graphics

Convenient:	All SutterPatch software is run by a single application. No need to launch multiple programs or to move data between programs.

Comprehensive: All data recordings, analyses, graphs, layouts, configurations and controls are saved in a single experiment file. This ensures that data are kept together with their complete contexts.

Automation: Automate your experiment using a rich set of data acquisition, data analysis, and amplifier controls. Create complex "Paradigms" that can respond to changing conditions via conditional steps and loops.

Note: "IPA" figures and examples in this manual may be from either an IPA or Double IPA system.

1.3 Experiment Structure

Experiment:

An Experiment is the highest-level structure in the SutterPatch world. An Experiment file can encompass all SutterPatch activity for the entire day, such as instructions (Paradigms), data acquisition parameters (Routines), recorded data (Series), execution settings, history, and comments. During reanalysis, data can be included from multiple experiments. Typically, one Experiment is created for each cell or preparation recorded from.

Paradigm:

A Paradigm is a sequence of control instructions used in an Experiment. Every Experiment contains at least one Paradigm, whether pre-planned by the user or automatically created by the system.

A loaded Paradigm "pool" file can contain multiple Paradigms for rapid access and execution. Such "planned" Paradigms can contain simple sequences, or sophisticated control structures, using a rich set of operations, such as conditional "If-then" decisions, nested loops, user-defined variables, hardware commands, and data acquisition Routines.

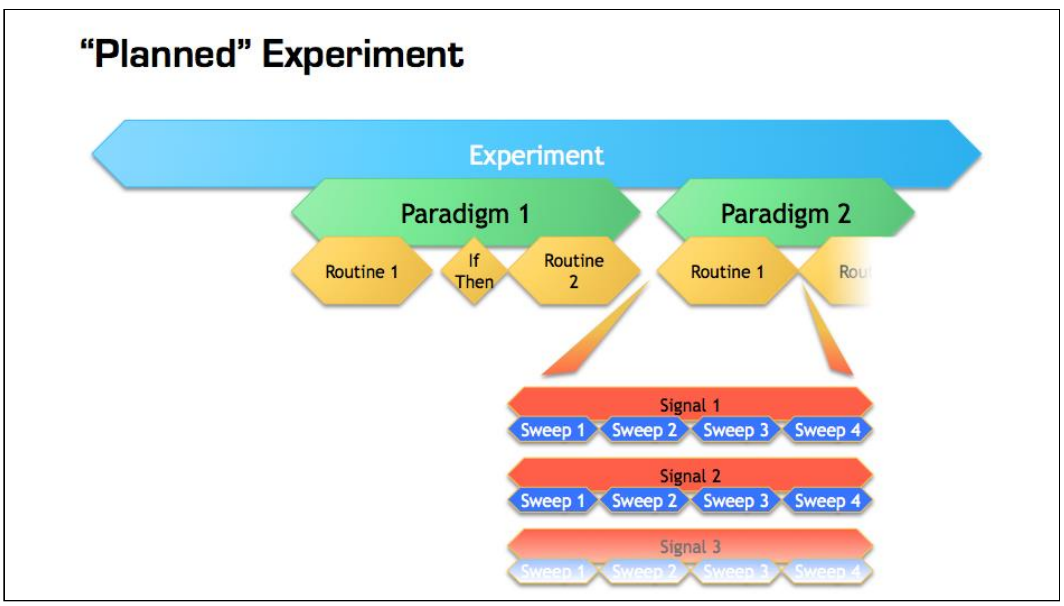

Figure 1-1. Data Structure - Planned Paradigms

An Experiment with two "planned" Paradigms running Routines.

However, if a Routine is manually run in the Scope window, an “auto-triggered” Paradigm is created as a container. This default Paradigm ensures that each Series is associated with a Paradigm in the context of an Experiment. If an auto-triggered Paradigm is already the active Paradigm, it is used for subsequent manually-run Routines.

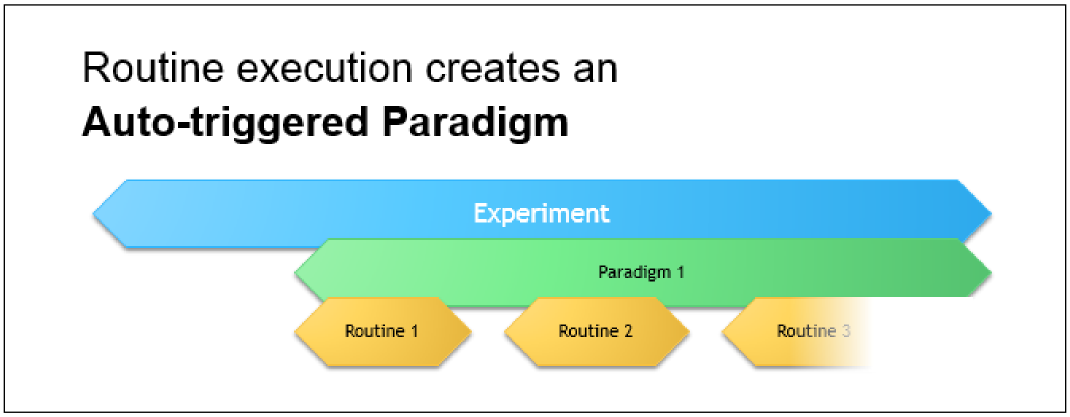

Figure 1-2. Data Structure - Auto-triggered Paradigms

An Experiment with manually run Routines using an “auto-triggered” Paradigm.

A Paradigm’s “data” includes all data points, variable values, and metadata tags from the course of a Paradigm. Altogether, this allows reconstruction of the exact course of an experiment. While a Paradigm could be compared to an itinerary, the Paradigm data correspond to the route a journey actually took. If conditional control is used in a Paradigm, e.g., for the number of loop cycles or a decision in an “If-then” step, these actions are recorded in the Paradigm data, even though they are not predetermined. (See the Data Browser.)

Routine Parameters:

A Routine is the set of data acquisition and online data analysis parameters that control input and output channel timing, triggering, command waveforms, display and real-time analysis.

A loaded Routine “pool” file can contain multiple Routines for rapid access and execution.

Series (Routine Data):

Recording Routine data creates a Series composed of all sweeps of data from all input signals. Multiple runs of a Routine create multiple Series of data. All Series are automatically stored in the current Experiment file.

Channel:

A Channel corresponds to a physical output (D/A) or a physical or virtual input (A/D) of the IPA system.

Analog input channels are used to record data, and are displayed in their own panes in the Scope window. There are two dedicated internal analog input channels ‘(Current’ and ‘Voltage’) for each attached headstage. General-purpose Auxiliary analog input channels (‘Aux-

IN') allow recording from external instruments. Virtual input channels allow creative processing of the physical input channels.

Analog output channels are used to send electrical stimuli, such as analog command waveforms to the preparation. There is a dedicated, internally configured, analog output channel ('StimOUT)' for each attached headstage. General-purpose Auxiliary analog output channels ('AuxOUT') can send output signals to external instruments.

Digital Output bits are also referred to as digital output channels ('DigOUT').

All 'Aux' and 'DigOUT' channels are available via the included BNC "octopus" breakout cable or the optional Patch Panel rack mount panel.

Signal:

Named analog input and output channels are referred to as Signals. A Signal is either the scaled representation of a physical channel, or the virtual result of a computation.

Sweep:

A Sweep is the sum off all data points from all Signals, acquired for a single period from time zero, for a fixed duration. In SutterPatch Software, the Sweep Duration is determined by the duration of the command waveform.

Trace:

A Trace is a Sweep applied to a single Signal. Therefore, a Sweep can be described as the collection of Traces across all Signals.

Segment:

A Segment exists as a user-defined section of the command waveform. Each Segment has a waveform type, an amplitude, and a duration. A command waveform can be composed of up to 50 Segments.

SutterPatch Metadata

Metadata are additional information associated with stored data. These can include information about the preparation (cell, tissue, animal), instrumentation (hardware, software), environmental parameters (temperature, atmospheric composition, etc.), stimuli (chemical compounds, light, acoustic, etc.) and other parameters. Metadata information is associated with the running of Paradigms and Routines, and their resulting data.

Metadata are dynamically recorded with a timestamp during an experiment. Information that can be determined by the system, such as the connected hardware, SutterPatch version, user Login Name, or the change of a digital output level, are automatically recorded without user intervention. In addition, the user can enter values for a large number of user-defined Metadata parameters, such as identifiers for the experimental animal or cell, the animal species, age and genotype, information about the recording solutions, and the electrodes or stimuli applied during the experiment. SutterPatch currently keeps track of ~ 600 Metadata parameters.

Terminology Comparison:

A table of equivalent terms to other electrophysiology software packages:

SutterPatch	PATCHMASTER	pCLAMP
Experiment	Compound Data	N/A
Paradigm	Protocol	Sequencing Keys
Routine	PGF Sequence	Protocol
Series	Series	Trial
Sweep	Sweep	Sweep
Signal	Signal	Signal
Trace	Trace	Trace
Segment	Segment	Epoch

Table 1-1. Software Terminology

2. INSTALLATION

2.1 Computer Requirements

Minimum Configuration

Operating System:	Windows:	Version 10 (64-bit versions) Most language packs are compatible. (listed in OS: Control Panel > System)
	macOS:	Version 10.11 (El Capitan) to 10.15 (Catalina) (listed in OS: Apple > About this Mac)
	Virtual machines and OS emulators, such as Parallels and VMWare Fusion, are not supported.	
CPU (Central Processing Unit):	Dual-core i5	
RAM (Random Access Memory):	3 GB	
Hard Disk (Free Space):	500 GB The drive should be configured as the primary system drive.	
Display Resolution:	XGA (1024 x 768)	
Computer Ports:	(1) USB 2.0 High Speed port Computer USB add-in card adapters are not recommended, as compatibility can be problematic. External USB hubs are not supported.	

Recommended Configuration (for Bandwidths > 50 kHz)

Operating System:	< same >
CPU:	< same >
RAM:	8 GB
Hard Disk (Free Space):	SSD (Solid State Drive) 500 GB or greater The hard drive should be configured as the primary system drive.

Display Resolution:	Full HD (1920 x 1080)
	High resolution (> 96-DPI) displays, such as Retina, 4K, 5K, Quad-HD and Ultra-HD, are not supported.
Computer Ports :	< same >

2.2 SutterPatch System Environment

The SutterPatch software runs in the Igor Pro 8 system environment. Igor Pro is widely used by scientists to acquire and analyze data, and to create publication-quality presentation graphics.

Igor Pro Features

- High-speed data display
- Large data set handling
- Waveform arithmetic
- Extensive set of built-in data analyses
- Image display and processing
- High-quality presentation graphics
- Graphical and command-line user interfaces
- Automation
- Extensibility via C and C++ modules
- Extensive online Help and PDF manual

SutterPatch 2.1 requires a 64-bit version of Igor Pro to run. The SutterPatch 2.1 full installer is recommended, as it automatically installs Igor Pro 8.0.4 (64-bit English version) and SutterPatch 2.1, while the SutterPatch 2.1 Updater only updates SutterPatch. Igor Pro 8.0.4 or higher is necessary to run the SutterPatch 2.1 XOPs

Igor Pro 8.0.4 has a 30-day trial period where it is fully functional and fully supports SutterPatch 2.1. After 30-days, if the Igor Pro 8 license has not been activated, Igor Pro 8 runs in a demo mode with limited functionality that does not support the SutterPatch application.

Note: Japanese versions of Igor Pro are not supported by SutterPatch.

Although Igor Pro 8.0.4 (64-bit) is strongly recommended for full support, SutterPatch 2.1 can still be run in Igor Pro 7 (64-bit), but only if the newly introduced 2 features (long names) are avoided.

For third-party applications that need a 32-bit version of Igor Pro 8:

Windows:	A 32-bit version of Igor Pro 8 is available in the installer.
macOS:	There is no 32-bit version of Igor Pro 8; run Igor Pro 7.

2.3 Mounting Instructions

Rack Mounting: The IPA amplifier is ready for mounting in a standard 19" wide equipment rack in a 1U (DIPA: 2U) space. A rack mount hardware kit consisting of hex screws, washers and cage nuts is included.

Benchtop Usage: Attach the four included stick-on feet to the bottom of the IPA amplifier.

2.4 Electrical Connections

Typical AC Power: 60 Hz, 120 V

50 Hz, 240V

The IPA amplifier can also run on power from 100 to 250 VAC - no switches need be set.

The AC power should be as clean as possible:

- At a minimum, a surge protector should be used to protect against high-voltage spikes; if lightning strikes are a concern, it should be rated > 1000 joules and > 40 kA.
- If you experience brownouts or voltage sags, a switching power supply (SPS) can be used to supply clean power to your instruments.
- To protect against power interruptions, use a universal power supply (UPS) for uninterrupted clean power.

2.5 Install Hardware

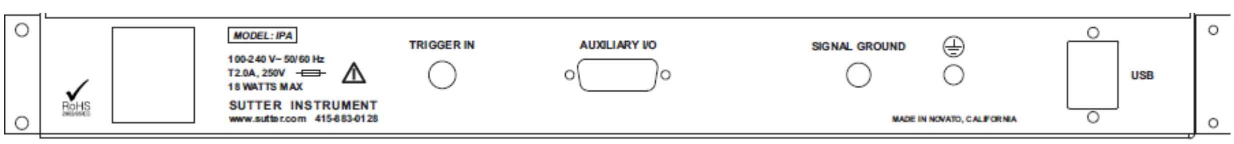

Figure 2-1. Rear of IPA Cabinet

1. Plug the female end of the power cord into the IPA rear-panel power receptacle.
2. Plug the male end of the included power cord into a grounded electrical mains outlet.

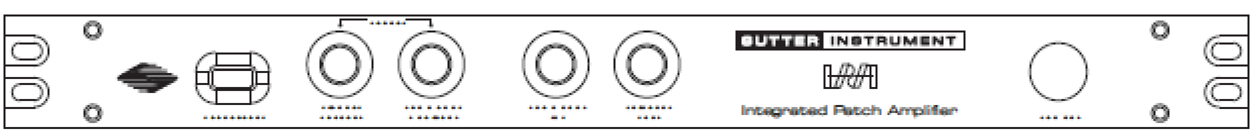

Figure 2-2. Front of IPA Cabinet

3. Set the IPA power button OFF (unlit position).

WARNING! Hot-swapping of headstages should be avoided – components can be damaged. Turn off the IPA system power before handling headstages.

4. Plug the IPA headstage into the HEADSTAGE port on the front of the IPA amplifier - the amplifier and headstage serial numbers should match.

   For a Double IPA system, attach the lower serial-numbered headstage to HEADSTAGE 1, and the higher serial-numbered headstage to HEADSTAGE 2, as each headstage is individually tuned to its channel.

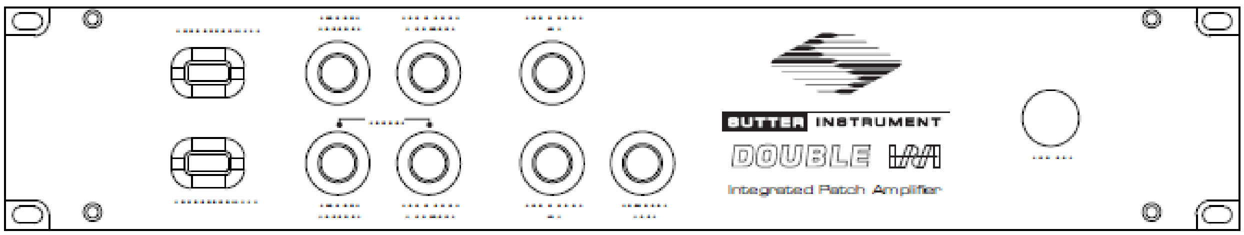

Figure 2-3. Front of Double IPA Cabinet

5. Plug the included I/O “octopus” breakout cable, or the optional Patch Panel cable, into the AUXILIARY I/O port on the back of the IPA amplifier.
6. Connect the supplied USB 2.0 cable to your computer’s USB 2.0 port, and to the IPA amplifier’s rear-panel USB port.
7. Connect the included electrode holder(s) to the headstage(s). See the Holder section for holder assembly instructions.

2.6 Install Software

Power on the computer.

A: It is strongly recommended to use the latest SutterPatch software version available.

   Download the latest version of SutterPatch installer software from https://www.sutter.com/AMPLIFIERS/SutterPatch.html and choose the “Download” tab. Navigate your file browser to the download file and run it.

B: If internet access is not available, attach the included USB flash drive to your computer USB port, and navigate to the flash drive.

   1. Install the software for ‘All Users’ by double-clicking on:
      - Windows: sutterpatch_win_full
      - Macintosh: sutterpatch_mac_full
   2. Follow the installer prompts:
      - If an existing version of Igor Pro 7 is found, it is recommended to replace it with Igor Pro 8. Make sure to keep a backup copy of all user files and parameters you may have placed into the program folder and its sub-folders.

- However, older versions of Igor Pro can also be kept, as different versions of Igor Pro can coexist on the same computer.
- If an existing version of Igor Pro 8 is found, SutterPatch sample files are over-written.
- The new install uses the Preferences from prior SutterPatch installations.

3. Upon completion, the installer will report a successful installation. The following files and folders are installed:

• IPA QuickStart Guide (PDF)	(Includes Igor Pro 8 Serial Number and Key.)
• Release Notes (PDF)	
• SutterPatch manual (PDF)	
• SutterPatch2 / SutterPatch folder	(Includes sample Routine Pool, Paradigm Pool, and Experiment data files).
• SutterPatch2 / SP_Code folder	
• SutterPatch2 / SP_Driver folder	(XOP)

4. Launch Igor Pro 8 and activate its license as instructed. You will need to enter the Igor Pro 8 Serial Number and Activation Key found in your IPA Quick Start Guide.
5. "Eject" the flash drive - wait for the "Safe to Remove Hardware" prompt, and unplug it from the computer.

2.7 Test System

2.7.1 Install Model Cell

1. Attach the model cell to Headstage 1 and tighten the screw collar.
2. Plug the supplied 1 mm grounding wire into the gold sockets on the headstage and model cell.
3. If the headstage is not inside a Faraday cage, completely surround the model cell/headstage assembly with alternative electromagnetic shielding, such as aluminum foil, and connect the shielding material to the headstage ground - a short wire with a metal alligator clip on each end makes a convenient shield-ground connector.

2.7.2 Startup

1. Power-on the IPA amplifier by pressing the silver POWER button on its front – it lights up as blue. (It can take a few seconds for the USB connection to be established.)

2. Launch the Igor Pro (SutterPatch) application by clicking on the 'Igor Pro 8' icon.

   A SutterPatch splash screen displays while opening files.

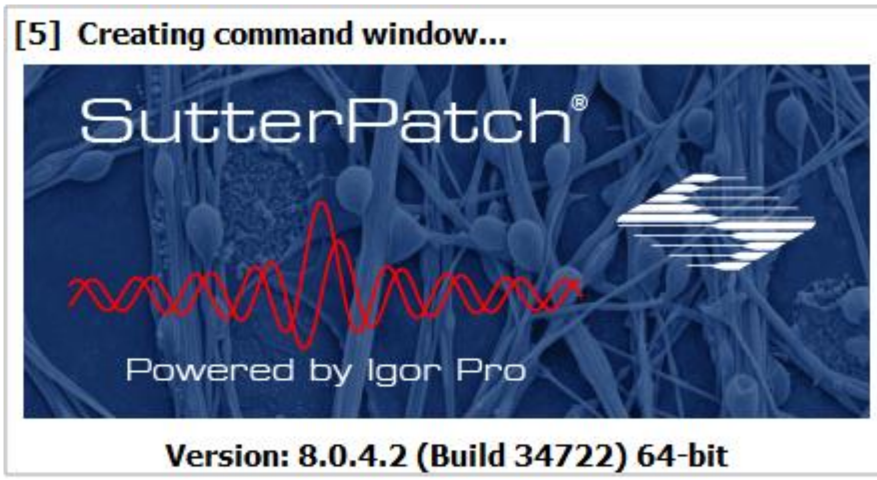

Figure 2-4. Splash Screen

Then the Welcome to SutterPatch "start" window displays.

Figure 2-5. Welcome Screen

3. Click on the 'Start' button in the 'Welcome to SutterPatch' window, and the application begins compiling. This process may take several seconds.

4. Specify the Experiment file name and storage location when prompted.

5. If the IPA amplifier is OFF or disconnected from the computer, this dialog displays:

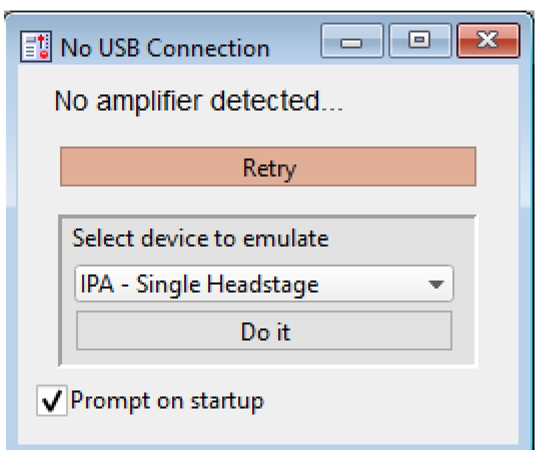

Figure 2-6. Emulation Modes

   a. Turn on and/or reconnect the amplifier, and then click 'Retry', or

   b. Click on 'Select device to emulate' and select 'IPA' or 'DIPA'.

Note: If amplifier hardware is not attached, and 'Prompt on startup' is disabled, the program will automatically start up in the last known emulation state.

6. The Dashboard panel is displayed.

Figure 2-7. Dashboard.

Click on the 'Acquire Data' icon, and a second level of the Dashboard is displayed.

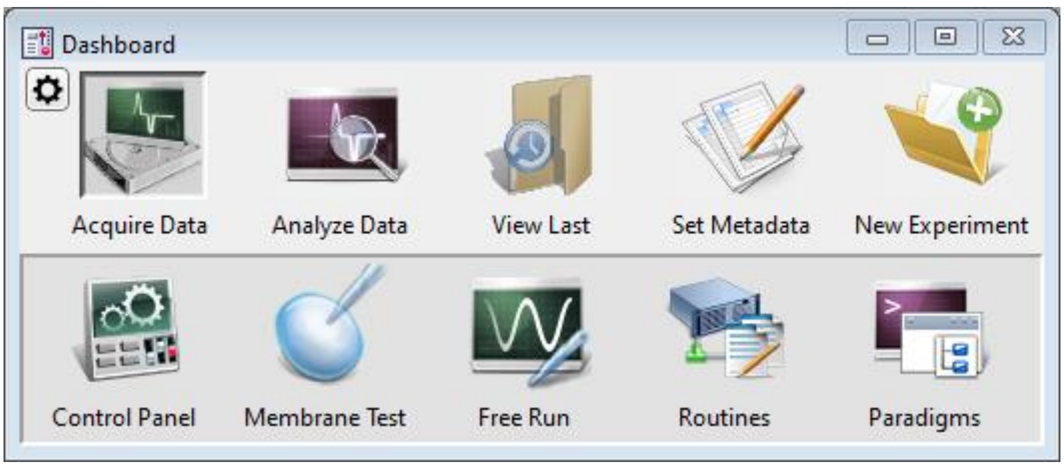

Figure 2-8. Acquisition Dashboard.

7. Click on the 'Control Panel' icon and the Amplifier Control Panel is displayed.

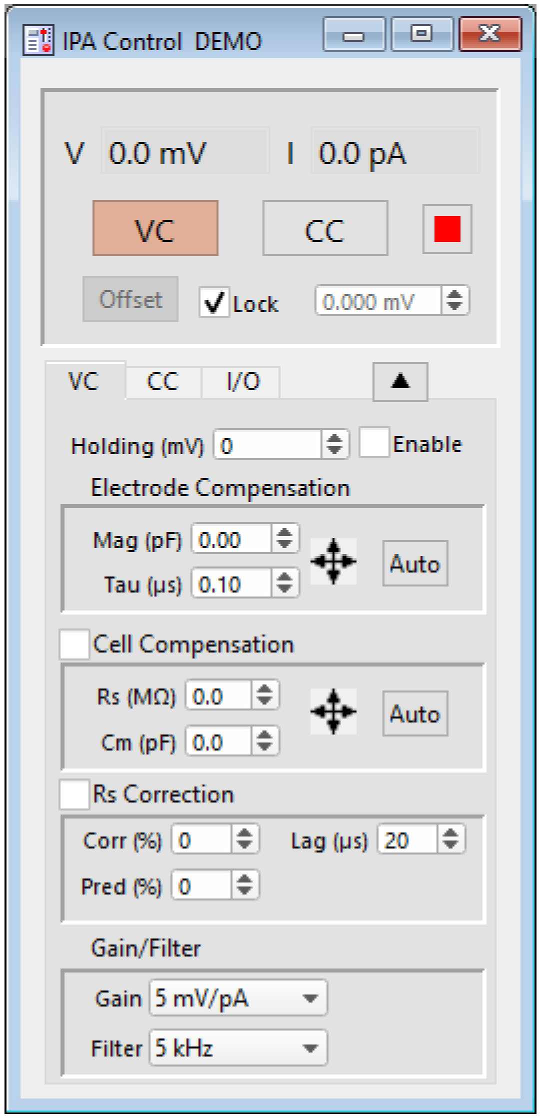

Figure 2-9. Amplifier Control Panel

a. If "Control DEMO" is displayed in the Amplifier Control Panel title bar, you are running in a hardware emulation mode - to run the physical instrument, ensure that the amplifier is powered on and its USB cable is connected, choose "New Experiment" from the Dashboard, and select "IPA".

b. For the next step, make sure that the Amplifier Control Panel is in voltage-clamp mode – the VC button at the top of the Amplifier Control Panel should be highlighted in red.

2.7.3 Run a Membrane Test

The Membrane Test is useful for a quick check of the IPA system functionality. It tests the three basic steps necessary for recording in a whole-cell configuration.

1. Go to the Dashboard window and click on the 'Acquire Data' icon.

Figure 2-10. Dashboard

2. Click on the 'Membrane Test' icon.

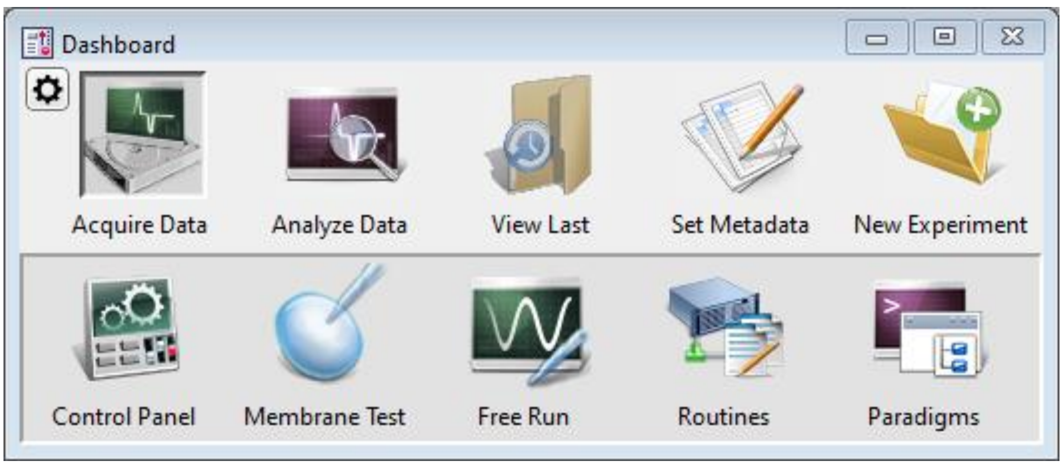

Figure 2-11. Dashboard – Acquisition

The following test values assume a 5 kHz filter…

3. Test the BATH mode:

   This mode simulates placing an electrode into the bath solution and sending a voltage pulse through the solution.

   a. Set the Model Cell switch: Bath

   b. Click on the Membrane Test 'Bath' button.

   c. Verify readings: Pipette Resistance: ~10 MΩ

4. Test the SEAL mode:

This mode simulates an electrode making contact onto a cell and forming a high-resistance gigaohm seal with the membrane.

a. Set the Model Cell switch: Seal

b. Click on the Membrane Test 'Seal' button.

c. Verify reading: Seal Resistance: ~1 GΩ to 1 TΩ

5. Test the CELL mode:

This mode simulates an electrode breaking into a cell and achieving a successful whole-cell patch.

a. Set the Model Cell switch: Cell

b. Click on the Membrane Test 'Cell' button.

c. Verify readings:

	Series Resistance:	~10 MΩ
	Membrane Resistance:	~500 MΩ
	Membrane Capacitance:	~28 pF

6. For a dual-headstage system:

a. Move the model cell, ground wires and shielding to Headstage 2.

b. Set the Scope window to 'Headstage 2'.

c. Repeat steps 3 – 5.

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3. HARDWARE OPERATION

3.1 IPA Front Panel

The front panel of the IPA system is used for the headstage and external I/O connections, and a power button.

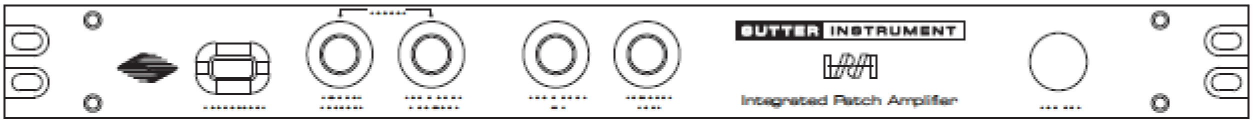


Figure 3-1. Front of IPA Cabinet

The front panel from left to right:

HEADSTAGE:	HDMI-style Type A		For IPA headstage.
SCOPE-SIGNAL OUTPUT:	BNC		A scaled analog output signal of the headstage response signal.
• VC mode:	mV/pA		Variable gain (Amplifier Control Panel)
	Example:		Membrane Test
	Model Cell:		BATH position (10 MΩ)
	Amplitude	=	10 mV
	Gain	=	5 mV/pA
	Current response	=	10 mV/10 MΩ = 1 nA (V/R = I)
	Scaled output voltage	=	I * Gain
			1 nA * 5 mV/pA
		=	5 V
• CC mode:	mV/mV		Variable gain (Amplifier Control Panel)
	Example:		Current Clamp
	Gain	=	100 mV/mV
	Response signal	=	10 mV
	Scaled output voltage	=	V * Gain
		=	10 mV * 100 mV/mV
		=	1 V

SCOPE-COMMAND MONITOR:	BNC		A scaled analog output of the headstage Stimulus signal (StimOUT).
• VC mode:	10 mV/mV		Constant gain
	Example:		Voltage Clamp
	Command voltage:		10 mV
	Scaled output voltage:		V * Gain
		=	10 mV * 10 mV/mV
		=	100 mV
• CC mode:	0.5 mV/pA		Constant gain
	Example:		Current Clamp
	Command current	=	200 pA
	Scaled output voltage	=	I * Gain
		=	200 pA * 0.5mV/pA
		=	100 mV
COMMAND IN:	BNC		A scaled analog input that adds an external signal to the headstage Stimulus signal (StimOUT).
• VC mode:	10 mV/mV		Constant gain
	<u>Example</u>:		Voltage Clamp
	External Command	=	5 mV
	Stimulus signal	=	20 mV
	Total stimulation	=	(V * Gain) + StimOUT
		=	(5 mV* 10 mV/mV) + 20 mV
		=	70 mV
• CC mode:	2 pA/mV		Constant gain
	Example:		Current Clamp
	External Command	=	1 mV
	Stimulus signal	=	5 pA
	Total stimulation :		(V * Gain) + StimOUT
		=	(1 mV * 2 pA/mV) + 5 pA
		=	7 pA

TRIGGER OUT:	BNC	Digital Trigger pulse output. Automatically sent at the start of continuous acquisition or each triggered sweep (including Membrane Test.
POWER:	Button	Turn power to unit On / Off. Lights up blue when 'On'

3.2 Double IPA Front Panel

The front panel of the Double IPA (vs. IPA) system adds support for a second headstage in an upper row of connectors:

HEADSTAGE 2

SCOPE-SIGNAL OUTPUT

SCOPE-COMMAND MONITOR

COMMAND IN

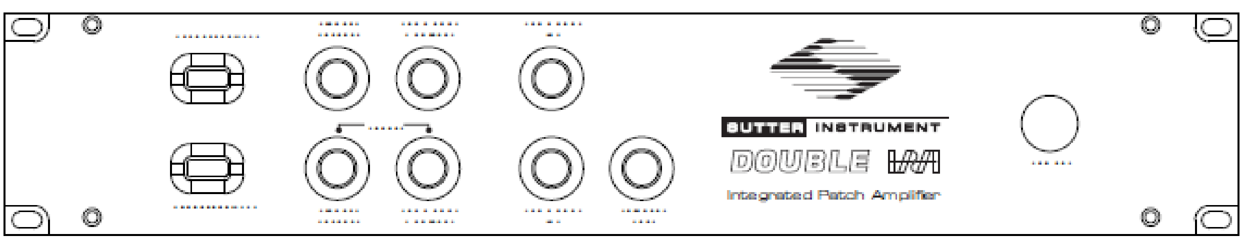


Figure 3-2. Front of Double IPA Cabinet

3.3 IPA Rear Panel

The rear panel of the IPA system is used for grounding, USB connection, and signal I/O. The rear panel of the Double IPA amplifier is essentially the same as for the IPA amplifier.

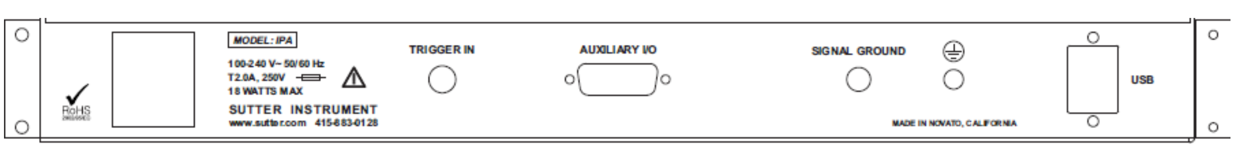


Figure 3-3. Rear of IPA Cabinet

[Unlabeled]:	Power-entry receptacle	For AC power cord

TRIGGER IN:	BNC	Digital input trigger.
AUXILIARY I/O:	DA-15 D-sub connector	External analog input and output channels, digital output channels, signal ground. See Appendix E for pin definitions.
SIGNAL GROUND:	4 mm Banana socket	Low-voltage grounding.
EARTH GROUND:	4 mm Banana socket	Instrument grounding.
[Unlabeled]:	USB Type B receptacle	USB 2.0 computer communication.

3.4 Grounding

Proper grounding is essential for the integrity of an electrophysiology laboratory setup. It greatly affects the "noise" within your system, and hence the quality of your data recordings. Low noise levels are especially needed for miniature post-synaptic recordings. While AC (mains) line-noise can be software-filtered out of a data signal, it is much more desirable to have a well-grounded electromagnetically clean hardware environment to start with.

For a properly grounded laboratory, an electrical connection is needed from your laboratory's electrical system to an "earth" ground. If your building's electrical grid does not provide an good earth ground, you can create your own earth ground by making use of the building's plumbing system, or by inserting a heavy metal bar deep into the earth.

The equipment in a rig should all be grounded to a single point to avoid ground loops. Installing a bus bar to the earth ground also helps to prevent ground loops. Consider standardizing your setups by using a GP-17 Ground Point on each rig.

"Signal" ground is a sensitive ground for very low voltages:

- BNC shields: Hard-wired to signal ground (single-ended).
- Bath ground electrode: Connect to the headstage signal ground jack.
- Shielding (Faraday cage): Connect to the rear panel SIGNAL GROUND socket.

However, due to the complexity of grounding factors, you may need to test various strategies for the best grounding configuration for your system. For example, when multiple headstages are used, one or both headstages might need to be grounded.

A grounded power cable is provided with this instrument.

3.5 Headstage

The IPA headstage supports both voltage- and (true) current-clamp in the same headstage.

Feedback resistor:	500 MΩ	
Whole-cell capacitance compensation:	0 – 100 pF	
Current-clamp rise time:	17.5 μs	(100 MΩ load, 20 kHz filter)

The headstage noise, as measured with an 8-pole Bessel filter:

Bandwidth	Open-Circuit Noise (RMS)
0.1 – 1 kHz	< 0.25 pA
0.1 – 5 kHz	< 0.75 pA
0.1 – 10 kHz	< 1.4 pA

Measuring "open-circuit", i.e., with no attachments so the headstage input is exposed to the air, provides a fairly consistent baseline for such headstage noise measurements. Conversely, measuring noise with an electrode in the bath generates the worst noise conditions.

A 1 mm gold pin signal-ground socket is on the back of the headstage.

The IPA headstage cable length can be increased with a 6-foot HDMI (non-powered) extension cable.

Note: DIPA headstages are serialized, whereby the lower-numbered headstage is matched to the HEADSTAGE 1 port, and the higher-numbered headstage is matched to the HEADSTAGE 2 port.

WARNING! Hot-swapping of headstages should be avoided – components can be damaged. Turn off the IPA system power before handling headstages.

3.6 Holder

A "holder" attaches a microelectrode (pipette) to a headstage. It provides mechanical stability for the pipette, low-noise for the electrical circuit, and chemical inertness from its physical components. Our pipette holders accept electrode glass in the range of 1.0 – 1.7 mm OD (Outer Diameter) using sized-by-color silicone gaskets.

The standard pipette holder included with the IPA amplifier is composed of low-noise polycarbonate and Teflon. A suction tube projects at a right-angle from the middle of the barrel.

An ultra-low-noise quartz pipette holder is optionally available. While polycarbonate is a proven material for patch pipette holders, it undergoes significant thermal expansion. Uneven warming can lead to motion of the pipette tip, and is often incorrectly perceived as drift in the micromanipulator. Quartz has a significantly lower thermal expansion coefficient and virtually eliminates thermal drift.

WARNING! Quartz is fragile, and can crack or shatter on impact. Treat your quartz pipette holder with the same care as with an optical component.

3.6.1 Assembly

The holder is assembled from 8 parts incorporated into a main barrel:

End Cap – Gasket – Silver Wire – Barrel – Tubing – Gold Pin – Pin Cap - Lockdown Ring

Note: The silver wire must be chlorided before use. See "Chloriding Silver Wire".

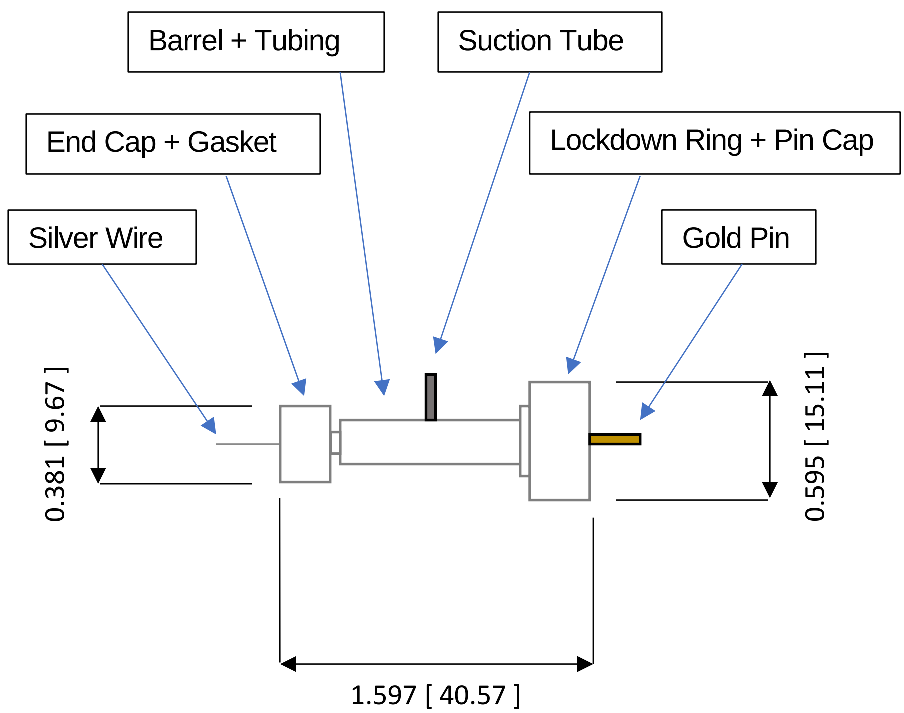


Figure 3-4. Electrode Holder

Figure dimensions are in "inches [mm]".

1. Cut the silver wire to size: the depth it extends into the pipette plus half the length of the barrel.
2. Thread the silver wire through the barrel.
3. Cut a small piece of clear tubing sized to fill the tiny "end-cup" in the pin-side of the barrel – the end with the narrower shaft.
4. Thread the small piece of tubing over the silver wire into the end-cup.
5. Crimp the end of the silver wire just slightly over the end of the tubing.
6. Slide the lockdown ring over the tubing-side of the barrel, with the ring's threads facing outwards.
7. Insert the gold pin into the recessed end of the pin cap - push it through the pin hole until it stops.
8. Screw the pin cap onto the barrel. Pressure from the compressed snippet of tubing ensures electrical contact between the silver wire and the gold pin.

9. Find a silicone gasket with an ID (inner diameter) just greater than your pipette OD (outer diameter):

Gasket ID	Color
1.1 mm	Clear
1.2 mm	Green
1.5 mm	Orange-Red
1.75 mm	Blue

10. Thread the gasket onto the silver wire on the end-cap side - the side with the narrower shaft.
11. Thread the end cap onto the silver wire and loosely tighten until it makes contact with the gasket.
12. Carefully thread a solution-filled micropipette onto the silver wire and into the gasket, and then push it into the barrel until it reaches the back end of the bore in the middle of the barrel.
13. Tighten the end cap onto the barrel.
14. Attach the holder to a headstage with the lockdown ring.

Assembly tips

1. The silver wire should be kept straight – do not bend or twist it.
2. Ensure good contact between the silver wire and the gold pin.

   Check for proper tubing height and wire-crimping length – avoid excess or insufficient amounts.

   For the most stable configuration, solder the silver wire to the end of the gold pin. Apply only a small bead of solder to the top of the pin in the very middle - avoid any excess solder that might interfere with the parts properly mating, as excess solder can result in air or solution leaks.
3. Fire-polish glass electrodes on both ends to prevent scratching the silver wire or the holder barrel.
4. The rubber gasket will wear out over time and need replacing.

3.6.2 Chloriding Silver Wire

The silver wire should be chlorided before first-time use, and then re-chlorinated monthly, or as needed.

Chemical Method

1. If needed, use a razor blade or fine sandpaper to rub off any insulation.
2. Optionally clean the silver (Ag) wire with ETOH (ethanol) to remove finger oils.

3. Immerse the silver wire in common household bleach (sodium hypochlorite) in glassware for 5 – 30 minutes until it turns purple-gray in color.
4. Remove the chlorided silver wire and rinse in distilled water.
5. Dry for storage.

Electrochemical Method

1. If needed, use a razor blade or fine sandpaper to rub off any insulation.
2. Optionally clean the silver (Ag) wire with ETOH (ethanol) to remove finger oils.
3. Connect a silver wire to each pole (positive & negative) of a household battery (1.5 V – 9 V).
4. Immerse the two silver wires in a solution of KCL (3 M) in glassware for 5 – 10 minutes. The wires should not touch each other. Bubbling around the silver wire indicates electroplating is occurring.

   Alternatively, use HCL (1M) with a 2 hour immersion time.
5. The charging polarity for the wires should be reversed a few times during the process.
6. A fully chlorided silver wire should be purple-gray in color. Remove the chlorided silver wires and rinse in deionized water.
7. Dry for storage.

Re-Chloriding Silver Wire

1. Pass the used silver wire through a flame - the wire should become bright silver in color.

   Alternatively, use a razor blade or fine sandpaper to scrape off any existing chloride.
2. Chloride the wire as described above.

3.6.3 Holder Maintenance

Holders must be properly maintained for good noise performance.

Storage:

1. Holders should be clean and dry.
2. Store in a container with desiccant.

Before 1st time use:

1. Disassemble the holder.
2. Rinse the polycarbonate parts in 70% ethanol.
3. Blot dry.
4. Store in a container with desiccant overnight.

After daily use:

1. Rinse holders with distilled water. For more thorough cleaning, wash with ethanol.
2. Blot dry.

Caution! Washing with soapy water can leave a film.

Continual cleaning with ethanol can degrade the polycarbonate parts.

Do not clean with methanol or strong organic solvents such as acetone.

Weekly Cleaning:

1. At least once per week, disassemble holder.
2. Clean the polycarbonate parts with 10 – 20 s sonication in distilled water.
3. Blot dry.
4. Store in a container with desiccant overnight.

3.7 IPA Amplifier Control Panel

This software interface controls the IPA amplifier settings. It replaces all physical knobs, dials and meters, such as found on manually-controlled amplifiers.

Most of these settings can also be programmatically controlled in a Paradigm.

Most editable numeric fields can also be adjusted via a control panel with three slider bars (for 3 significant digits) by right-clicking on the field.

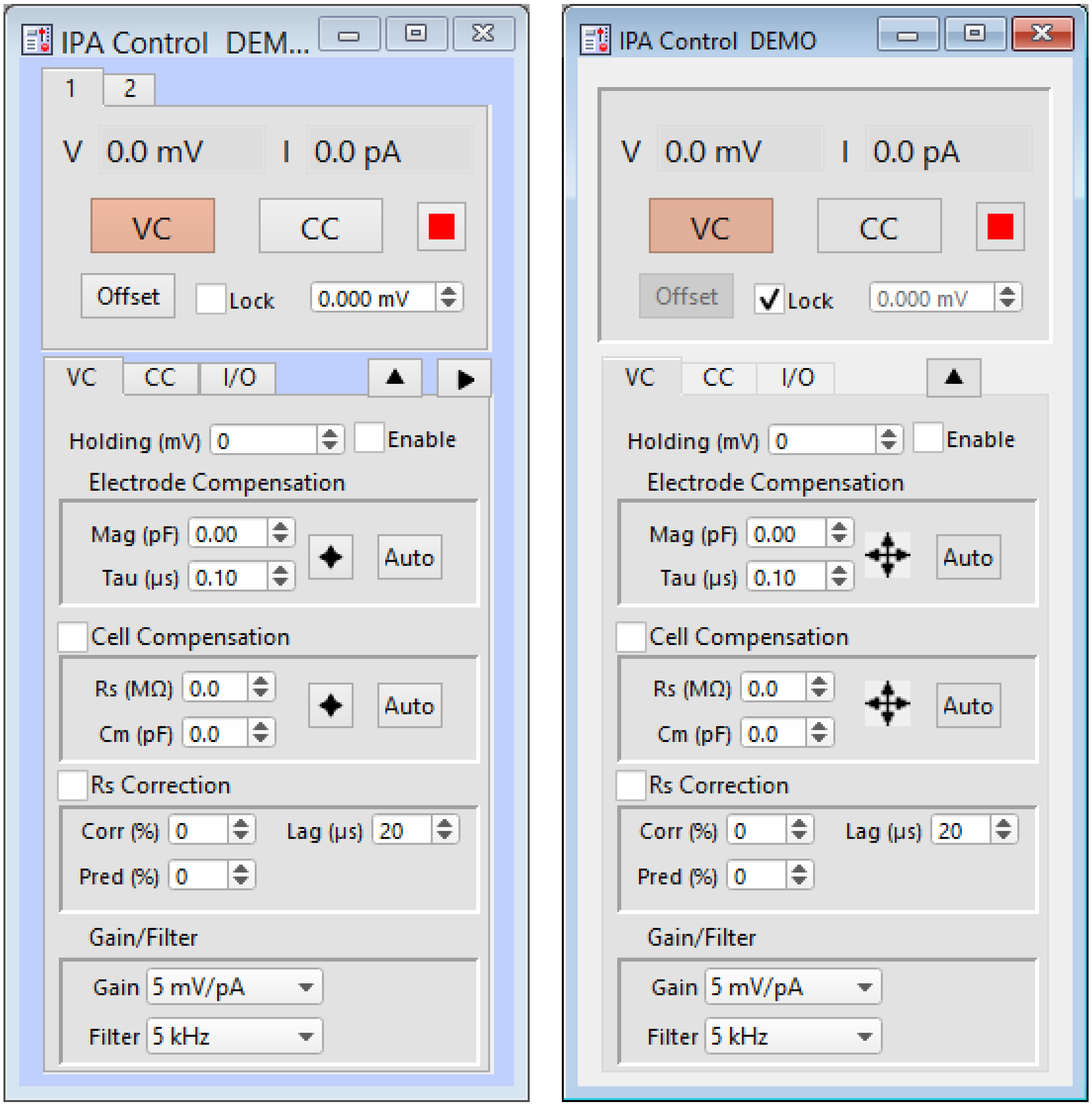


Figure 3-5. Amplifier Control Panels

General Controls

1 - 4 Headstage # tabs

When only a single IPA amplifier is being used, headstage selection tabs are not displayed.

When multiple headstages or amplifiers (IPA and/or Double IPA) are used, multiple headstage tabs will display. Each headstage maintains its own settings.

Clicking on a headstage tab will open its last-used “VC” or “CC” active mode settings. This will also blink the power light of the attached amplifier, which is useful in identifying which headstages are associated with which amplifiers.

Note: If a headstage is unattached while an IPA or Double IPA amplifier is in use, its input channel will be at “ground”. For older IPA amplifiers, the channel might appear as saturated.

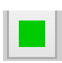
V ‘V’ meter: Displays the Voltage input channel level.
I ‘I’ meter: Displays the Current input channel level.

VC VC button: Switch the IPA amplifier from Current Clamp to Voltage Clamp mode. The button for the active mode is highlighted in red.

A “gentle” switch is used to protect cells against transition spikes that could degrade the electrode seal and integrity of the recording. The amount of voltage to hold the current steady is automatically injected into the cell during the transition (< 100 ms), before stepping to the new voltage command level.

CC CC button: Switch the IPA amplifier from Voltage Clamp to Current Clamp mode. The button for the active mode is highlighted in red.

A “gentle” switch is used to protect cells against transition spikes that could degrade the electrode seal and integrity of the recording. The amount of current to hold the voltage steady is automatically injected into the cell during the transition (< 100 ms), before stepping to the new current command level.



Reset USB button: Shift-click to re-establish the USB connection to the amplifier.

- All USB channels are reset.
- A green button indicates that a stable USB connection to the amplifier has been established.
- A red button indicates that there is no USB connection to the amplifier, or that SutterPatch is in Demo (hardware emulation) mode.
- When multiple amplifiers are connected, if any one instrument loses its connection, the button turns red.
- It can take several seconds for the USB connection to be re-established.
- If the amplifier is attached in demo mode, a USB reset will disable demo mode and run the hardware “live.

Right-click menu Click almost anywhere in the Amplifier Control Panel to access:

- Reset USB Click to re-establish the USB connection to a Sutter amplifier.

- All USB channels are reset.
- It can take several seconds for the USB connection to be re-established.
- If the amplifier is attached in demo mode, a USB reset will disable demo mode and run the hardware "live.

- Reset Amplifier Controls — Reset all hardware settings of all attached Sutter IPA/DIPA amplifiers and their headstages to factory defaults.

Offset — Click on the Offset button to apply a compensatory potential to automatically zero the current signal. The auto Offset value is an approximation that might need further adjustment.

[±250.000 mV] — The Offset value is to the right of the Auto Offset button, and is independent of the holding potential.

- Numeric values can be directly typed into the numeric field, or
- For fine adjustments, use the up / down spinners to increase or decrease the setting by ~0.015 mV.

  The Offset spinner step size (~0.015 mV) is based on the 16-bit resolution of a 1 V DAC. The actual spinner step size resolution is 0.01525878 mV.
- For moderate adjustments, increase the spinner increment by 10x (~0.15 mV) by holding down the Shift key and then clicking on the spinners.
- For fastest operation, select the offset field, hover the cursor over the numeric field or spinners, and hold down the Shift key while simultaneously scrolling up or down with the mouse wheel.

Note: The IPA Demo mode auto Offset value is fixed at -0.200 mV.

This Offset counteracts any inherent hardware circuitry offsets, as well as electrode-in-the-solution offsets. Adjust the offset in voltage-clamp mode when the electrode is initially placed into the bath solution. This adjusts the voltage output so that the electrode current signal is zero, providing a reference baseline for measurements.

Lock Once an electrode offset has been applied, use the 'Lock' check box to prevent accidental changes to the Offset.

Voltage Clamp Controls

VC The 'VC' tab displays amplifier Voltage Clamp controls:

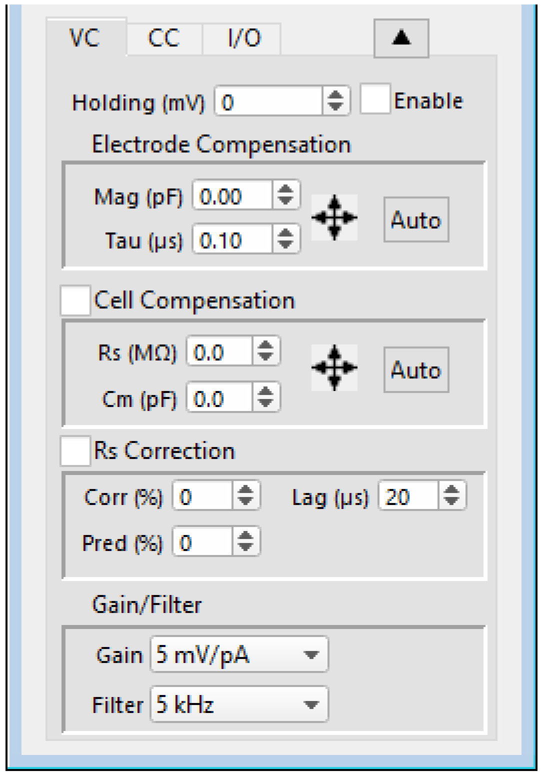


Figure 3-6. VC Control

Holding (mV): [±1000]

After achieving a seal, the holding voltage is typically set to the cell's equilibrium or "resting" membrane potential (typically -60 to -80 mV for neurons). This control is active during acquisition - changes are applied to the next sweep.

- Numeric values can be directly typed into the numeric field, or
- For fine adjustments, use the up / down spinners to increase or decrease the setting by 1 mV.
- For moderate adjustments, increase the spinner increment to 10 mV by holding down the Shift key and then clicking on the spinners.
- For fastest operation, select the offset field, hover the cursor

over the numeric field or spinners, and hold down the Shift key while simultaneously scrolling up or down with the mouse wheel.

Whenever you enable or leave the Amplifier Control Panel, a tag is written to the Log window with the Holding changes.

Electrode Compensation: Electrode capacitance compensation.

Mag (pF): [0.00 – 25.00] Magnitude
[shared with CC mode]

Tau (μs): [0.10 – 4.50]

Opens a 2-D slider panel for simultaneous tuning of both parameters.

Note: When dragging with a mouse, slow down when approaching panel boundaries, else undershoot or overshoot of the values can occur.

Auto Automatically sets approximate values.

After making an on-cell gigaohm seal, large microelectrode capacitance spikes are visible. To remove the transients, click the ‘Auto’ button. Then autoscale the signal, and if needed, adjust the ‘Mag’ and ‘Tau’ controls individually or use the slider panel. For a square pulse command (such as a Membrane Test ‘Seal command), the goal is to eliminate the edge-effect spike transients.

Disable during Lock-In operations.

Note: The IPA Compensation controls do not affect demo data.

Note: Disable ‘Cell Compensation’ and ‘Rs Correction’ when the amplifier is running a Membrane Test in Cell mode.

Cell Compensation Whole-cell capacitance compensation.

Rs (MΩ): [0 – 100.0] Series Resistance

Cm (pF): [0 – 100.0] Membrane Capacitance

Opens a 2-D slider panel for simultaneous tuning of both parameters.

Note: When dragging with a mouse, slow down when approaching panel boundaries, else undershoot or overshoot of the values can occur.

Auto — Automatically sets approximate values using small “gentle” steps to avoid hyperpolarization.

After breaking into a cell, i.e., going “whole cell”, additional large capacitive transients are now generated by the entire membrane of the cell.

To remove the transients, click the ‘Auto’ button. Then zoom in on the signal, and if needed, adjust the ‘Rs’ and ‘Cm’ controls separately or use the slider panel until the signal is adequately compensated. For a square pulse command, the goal is to eliminate the edge-effect transients with minimal distortion of the response signal.

The IPA system is optimized for real-world measurements from real electrodes, so when used with the model cell, the compensation might need several more ‘Auto’ adjustments to compensate the model cell capacitance.

Warning! Disable ‘Cell Compensation’ and ‘Rs Correction’ when the amplifier is running a Membrane Test in Cell mode, or results will not be valid.

Note: The IPA Compensation controls do not affect demo data.

Rs Correction — Whole-cell Series Resistance compensation.

“Rs Correction” is used to correct command potential voltage drops, to minimize rise-time delays and slow decay phases in the current response, and to reduce unwanted filtering effects, caused by Series resistance.

Corr (%):	[0 – 100]	Correction
Pred (%):	[0 – 99]	Prediction
Lag (μs):	[20 – 200]	(RC filter component)

$$\text{Lag} = 1 / (2 * \pi * \text{Bandwidth})$$

Rs Correction requires that the Electrode and Cell Compensations are first applied.

Set the Prediction (Pred) to “supercharge” the command potential. Small transients should become visible at the start and end of the current response.

Next, increase the Correction (Corr) current injected into the membrane to sharpen the rise time. As the Corr setting is increased, the current response transients also increase in size. Avoid overshooting, as if the correction is set too high, internal feedback can lead to oscillation of the circuit, i.e., “ringing” and loss of a patch.

Reduce oscillation of the circuit by adjusting the ‘Lag’ setting - larger values increase the stability of the circuit, but also increase the rise time.

Remove the Prediction/Correction transients in the signal by reducing the Cell Compensation ‘Rs’ setting until a minimum value is found. Then adjust the Cell Compensation ‘Rs’ setting again until the best result is achieved, or try over again with lower Prediction/Correction settings.

Note: Disable ‘Cell Compensation’ and ‘Rs Correction’ when the amplifier is in ‘Capacitive’ mode, or when running a Membrane Test in Cell mode, or results will not be valid.

Note: The IPA Correction controls do not affect demo data.

Gain

Input Gain

mV/pA	Signal Range
0.5	± 20 nA
1	± 10 nA
2.5	± 4 nA
5	± 2 nA
10	± 1 nA
25	± 400 pA

New voltage-clamp gain settings are applied to headstage Current input signals when new Scope windows are created.

Filter

Input Filter (low-pass 4-pole Bessel)

kHz	Data Point Interval
0.5	(2 ms)
1	(1 ms)
2	(500 µs)
5	(200 µs)
10	(100 µs)
20	(50 µs)

This low-pass filter is applied to the active headstage input signals, and serves to reduce the headstage input sampling rate (as set in Routine Editor / Acquisition & Routine Parameters.)

Filter settings are shared between the voltage- and current-clamp modes. New filter settings are applied when new Scope acquisition windows are created.

Tip: For experiments where the shape of the response is of interest (such as minis, etc.), an input filter rate of 10 kHz is commonly used.

Note: The Demo mode does not apply filtering - demo data uses the sampling rate timing.

Current Clamp Controls

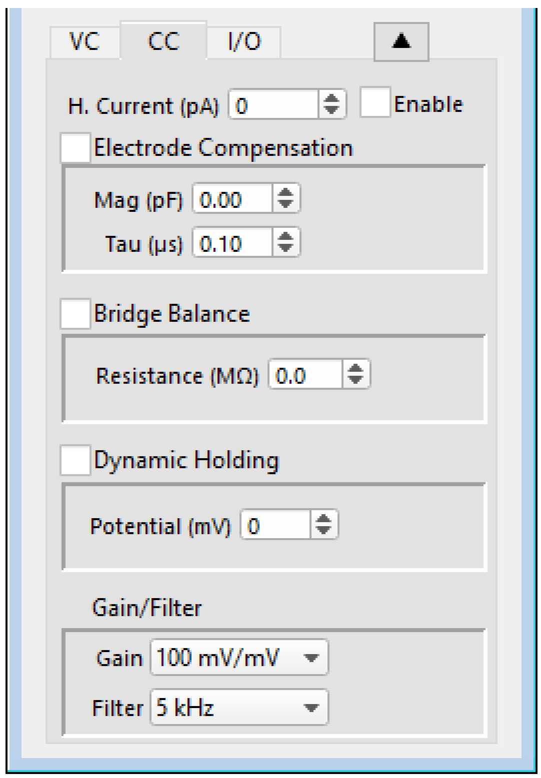


Figure 3-7. CC Control

CC The 'CC' tab displays the IPA Current Clamp controls:

H. Current (pA): [±1000]

A Holding Current can be preset here. Use the 'Enable' checkbox to activate it.

Electrode Compensation Electrode Capacitance Compensation

Mag (pF): [0.00 – 25.00] Magnitude

[shared with VC mode]

To remove microelectrode capacitance-charging transients, and reduce their filtering effects (which increases signal amplitudes and rise-times), click the 'Auto' button. Then autoscale the signal, and if needed, adjust the 'Magnitude' control.

CC mode Electrode Compensation is also known as "Capacitance Neutralization".

Tip: If you consistently lose cells when switching into CC mode, consider adjusting the CC mode electrode compensation value in Set Preferences / Hardware / Stability Control.

Note: The IPA Compensation controls do not affect demo data.

Bridge Balance

Resistance (MΩ): [0 – 200.0] [shared with VC mode 'Cell Compensation']

Bridge Balance is an adjustment to remove voltage-drop effects due to the electrode Series resistance, when current is flowing into the preparation. Voltage readings from the cell during current flow (injection) are corrected.

As a superior method of calculation, instead of a current-clamp 'Auto' bridge balance button, the 'Cell Compensation' Series resistance value from voltage-clamp mode is carried over. Start in VC mode and use Auto 'Cell Compensation'. When you switch into CC mode, the VC mode 'Cell Compensation' value is copied into the Bridge Balance 'Magnitude' field.

To fine-tune the value, run the Membrane Test and zoom in on the initial rising phase. With Bridge Balance disabled, there is a DC shift visible at the beginning of the rising signal. Enable Bridge Balance and adjust until the DC shift disappears.

However, after adjustment, there may be a small glitch at the beginning of the rise, due to electrode and headstage capacitance that doesn't entirely go away. (Further adjustments to the Electrode Compensation may need to be made.) In some cases, it can be difficult to determine the exact Bridge Balance value, but as long as the Series resistance is significantly smaller than the cell resistance, the errors are very small.

Note: If you are simply recording voltage (I=0) without any current injection, then Bridge Balance can be ignored or left disabled.

Dynamic Holding

Potential (mV): [±1000]

Maintain the membrane holding potential at a set target level without it drifting over time.

The tracking time constant (tau) is ~18 ms, equating to a rise time of 40 ms, or a bandwidth of 8.75 Hz.

Note: When Dynamic Holding is enabled, the Holding Current is automatically disabled, as the current output is dynamically adjusted by the system.

Gain

Input Gain

mV/mV	Signal Range
10	±1 V
20	± 500 mV
50	± 200 mV
100	± 100 mV
200	± 50 mV
500	± 20 mV

New current-clamp gain settings are applied to headstage Voltage input signals when new Scope windows are created.

Filter

Input Filter (low-pass 4-pole Bessel)

kHz	Data Point Interval
0.5	(2 ms)
1	(1 ms)
2	(500 µs)
5	(200 µs)
10	(100 µs)
20	(50 µs)

This low-pass filter is applied to the active headstage input signals, and serves to reduce the headstage input sampling rate (as set in Routine Editor / Acquisition & Routine Parameters.)

Filter settings are shared between the voltage- and current-clamp modes. New filter settings are applied when new Scope acquisition windows are created.

Tip: For experiments where the shape of the response is of interest (such as action potentials, etc.), an input filter rate of 10 kHz is commonly used.

Note: The Demo mode does not apply filtering - demo data uses the sampling rate timing.

I/O Controls

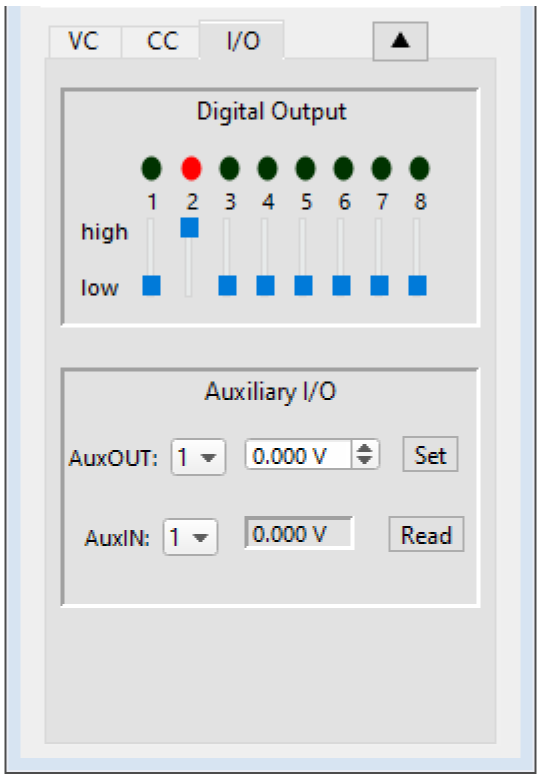


Figure 3-8. I/O Control

I/O The 'I/O' tab contains the Digital Output and Auxiliary I/O controls. For a Double IPA system, the I/O tab is only visible when Headstage 1 is selected.

Digital Output

This section controls the holding bit pattern generated by the Digital Outputs of the IPA amplifier. Eight TTL-compatible digital channels are displayed. Switch between digital states by clicking on a channel slider 'high' or 'low' end, or by clicking and dragging the slider button. When a channel state changes, its dot also changes color:

• On	Red dot	high	(+3.3 V)
• Off	Black dot	low	(0 V)

Note: When multiple Sutter amplifiers are connected, the Digital Outputs are only active for the main amplifier.

Auxiliary I/O

General purpose "auxiliary" analog input/output channels are available.

AuxOUT 1 & 2: [±10 V]

Select an auxiliary analog output channel from the drop-down list. Directly edit its voltage level, or use the spinners to change the value in 0.100 V increments, and then click the 'Set' button to apply the value.

Note: Demo-mode output channels are reset to zero when selected.

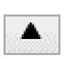
Tip: When the IPA system is used as a data acquisition system for external instruments, the auxiliary outputs can be used as holding levels.

Set — Click the 'Set' button to apply the value to the selected output channel.

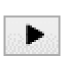
AuxIN 1 – 4: [±10 V] (Read Only)
Select an auxiliary analog input channel from the drop-down list.

Read — Click the 'Read' button to display a single-point reading of the selected input channel.

Tip: This is useful for monitoring slow-changing parameters, such as temperature.



The vertical Show / Hide button displays / hides the tabbed controls below this button.



The horizontal Show / Hide button displays / hides the Double IPA input monitor, which displays the real-time Voltage and Current input channel values for both headstages.

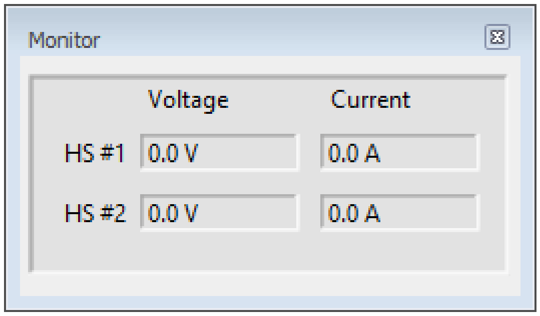


Figure 3-9. Headstage Input Monitor

3.8 Lock-In Adjustments

The "lock-in" system is used to detect very small changes in membrane capacitance measurements, such as for single-vesicle fusion and retrieval measurements. Our system uses a digital implementation of phase-sensitive detection (PSD) to make its measurements.

1) To perform lock-in analysis, a reference waveform needs to be supplied to the headstage. A suitable sine wave needs to be generated via the Routine Editor / Waveform Editor / Sine / Sine Wave Cycles / For Lockin. (Also see Membrane Test.)

2) The lock-in response signal is processed via a virtual input channel. Measurements, such as membrane conductance, are reported. Calculations are made using 'conductance' (1 / resistance) instead of 'resistance'.

Setup in the Routine Editor / Input Channels / Virtual#:

Math Type	LockIn
Current Channel	Select a (source) input channel with a current (amperage) signal.
Trace Kind	Select the LockIn measurement to perform. The selected 'Trace Kind' is automatically set as the Virtual Channel label.
CM	Computed membrane capacitance.
GM	Computed membrane conductance.
GS	Computed series conductance.
DC	DC component of measured signal.
RealY	Real number part of the lock-in response signal.
ImagY	Imaginary number part of the lock-in response signal.
Cycles to Average	[1 – 1000]

Cycles to Skip [1 – 1000]

V-reversal [±1000 mV]

When using a calculated stimulus trace, enter the reversal potential for the ion under study, such as for (Na+) sodium spikes or (K+) potassium tail currents.

3) The lock-in phase detection can be manually tuned via the menu item SutterPatch / Hardware Control / Lock-In Adjustments. Phase adjustments are made to optimize the headstage signals, and should be done in voltage-clamp mode.

Enable Manual Adjustments Adjustments can be made using direct field editing, spinners, or a field right-click slider panel.

Absolute Values

Phase Delay Adjustment Apply a phase delay to the calculations.

[±1.00 µs]

Reset Reset to '0.00 s'

Attenuation Adjustment Apply a gain to the calculations.

[0.001 – 9.999]

Reset Reset to '1.000'

List Results Display results in the Command window.

The lock-in computation is quite stable - its calibration values do not change day-to-day. However, lock-in measurements can be affected by experimental conditions, including the amplifier itself. In particular, the electrode compensation has a large influence on the results, and needs to be properly set. Due to its sensitivity, run the electrode compensation on a pulse before running LockIn adjustments.

Start with the Phase Delay Adjustment, and then follow with the Attenuation Adjustment.

The sensitivity of the SutterPatch software lock-in results is as good as for a hardware lock-in amplifier.

When making absolute capacitance measurements, you can improve the consistency of the measurements by adjusting the lock-in phase adjustment to a known reference capacitance, such as with the model cell.

The SutterPatch lock-in calculations are based on the Lindau/Neher method of time-resolved capacitance measurements in single cells. (See the Algorithms appendix for the reference.)

Note: Demo mode is not designed to respond to lock-in phase and attenuation adjustments.

3.9 System Integration

The IPA systems can be integrated with other suitable laboratory equipment.

3.9.1 Using Peripheral Equipment

The IPA system can control peripheral equipment, such as:

- Cameras
- Light sources
- Pulse generators
- Solution changers
- Wavelength switchers

Auxiliary analog output signals can be used to control other instruments within a range of ±10 V. Digital outputs use TTL-compatible voltage signals. Analog and digital holding levels are set in the Amplifier Control Panel

The digital command output can be formatted as either an 8-bit "word" or a single "bit", as selected in the Routine Editor / Output Channels & Waveform section. Actual command output patterns are configured in the Routine Editor / Output Channels & Waveform / Waveform Editor.

Note: The Analog and Digital controls in the Amplifier Control Panel provide a way to quickly and easily test the behavior and operation of peripherals, without the need to create or modify Routines.

3.9.2 Using Multiple Sutter Amplifiers

Any combination of two IPA or Double IPA amplifiers can be connected and run simultaneously by the SutterPatch program.

One amplifier is designated the "main" amplifier, and provides a clock signal to control the timing of the "secondary" amplifier. To setup, install the amplifiers, and then connect the main amplifier front panel TRIGGER OUT BNC to the secondary amplifier rear panel TRIGGER IN BNC.

When more than one IPA headstage is attached, each headstage has its own numbered tab [1 – 4] in the Amplifier Control Panel. Up to 16 analog input channels can be configured, using a mix of headstage, auxiliary and virtual input channels from either amplifier. However, Digital Outputs (8) are only available from the "main" amplifier.

When a computer is powered on, its USB ports and attached hardware are detected and "enumerated", i.e., receive a sequence number. The Amplifier Control Panel displays each attached

Sutter headstage in a numbered tab based on its enumerated sequence. You will need to determine the actual headstage sequence during installation, but these numbers should not change after installation, unless the attached equipment is changed or the USB ports are reconfigured.

Note: Only one amplifier can be used for holding levels.

Demo mode does not support multiple-amplifier configurations.

3.9.3 Using Non-Sutter Amplifiers

An IPA or Double IPA system can also be operated as a stand-alone data acquisition system interfacing to non-Sutter amplifiers. You continue to control the Sutter digitizer via the Amplifier Control Panel and SutterPatch software.

The IPA digitizer interfaces to external amplifiers via the rear panel Auxiliary I/O Cable or optional Patch Panel BNCs:

AuxOUT1 & 2	These two auxiliary analog output channels can be used to send stimulus waveforms to external instruments, such as non-Sutter microelectrode amplifiers.
AuxIN1 – 4	These four auxiliary analog input channels can be used to digitize signals from external instruments, such as non-Sutter microelectrode amplifiers.
DigOUT1 – 8	Digital output patterns can be sent via eight digital output channels to a variety of peripheral equipment.

Auxiliary analog and digital holding levels are set in the Amplifier Control Panel I/O tab.

Note: Sutter amplifier output levels into Sutter systems attenuate by < 0.2%.
HEKA amplifier output levels into Sutter systems attenuate by 0.%.
Axon Instruments amplifier output levels into Sutter systems attenuate by 5%.

3.9.4 Using Non-Sutter Data Acquisition Systems

The IPA or Double IPA system can also be operated as a stand-alone amplifier using non-Sutter data acquisition systems, while the Sutter amplifier continues to be controlled via the Amplifier Control Panel.

IPA System Front Panel Connections

BNC	Channel
COMMAND IN	Stimulus to the preparation.

	Combines the analog input signal from an external source with the stimulus (command waveform) sent to the preparation. The Command In external signal is summed with the IPA internal StimOUT output signal, and is then sent to the IPA headstage.
SCOPE - SIGNAL OUTPUT	Data from the preparation. This BNC supplies the response from the IPA preparation. The current or voltage response from the headstage is directly available from this analog BNC output, and can be connected to an external data acquisition system for digitization and recording.
SCOPE - COMMAND MONITOR	Data from the Stimulus signal. This BNC allows you to monitor the stimulus channel. The analog stimulus delivered to the IPA headstage (voltage or current) is directly available on this BNC, and can be connected to an external data acquisition system for digitization and recording.

3.10 IPA Maintenance

This unit should require minimal maintenance when operated according to specifications.

3.10.1 Inspection

Periodically inspect all cables and connections to make sure that all cables are sound and that all connections are firm and evenly seated.

Warning! Turn off the IPA power before plugging / unplugging headstages.

3.10.2 Cleaning

Routine cleaning of the IPA system is required to prevent excessive dust accumulations. Wipe all exterior surfaces with a dry, soft, cotton cloth.

3.10.3 Calibration

The IPA amplifiers do not support user calibration.

4. SOFTWARE OPERATION

4.1 Acquisition

SutterPatch acquisition operations.

4.1.1 Acquisition Control

The interactive acquisition controls for both Routines and Paradigms are grouped into this control panel.

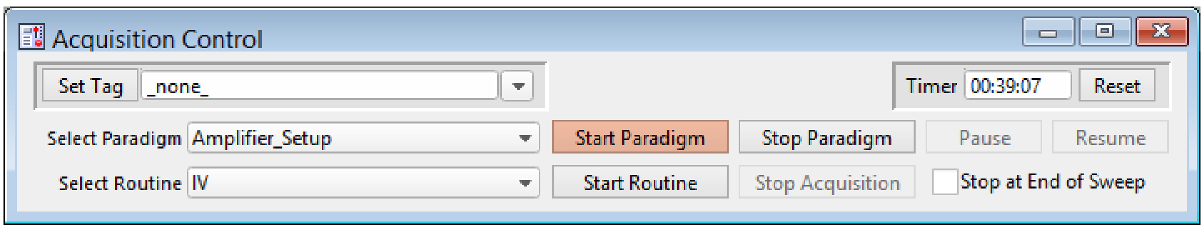


Figure 4-1. Acquisition Control

Set Tag

Click the Set Tag button to create a time-stamped text comment in the Paradigm metadata.

When run during acquisition, the comment tag is also written to the Routine metadata, and when the data is opened in a reanalysis Scope window, a black vertical cursor displays at that time point.

Enter the comment text into the field, or select text from a drop-down list of recently used entries.

Note: In emulation mode, tag timing is not accurate, and tags are not set in the first sweep.

Timer

A running clock displays the time in “hh:mm:ss” since the last timer reset, or since a new experiment established a USB connection or emulation mode.

Reset

Reset the Timer to 00:00:00.

Select Paradigm

The active Paradigm is selected from the loaded ‘Paradigm Pool’ list. See the Paradigm Editor to load a different

	Paradigm Pool.
Start Paradigm	Manually run the selected Paradigm - any existing acquisition in the Scope window is stopped, and a "planned" (user named) Paradigm is created.
Stop Paradigm	Terminate the current Paradigm. The next Paradigm to run starts a new “planned” named Paradigm. Otherwise, the next Routine to run starts a new date/time-stamped “auto-triggered” Paradigm.
Pause	Pause a running Paradigm.
Resume	Resume running the active Paradigm.
Select Routine	The active Routine is selected from the loaded ‘Routine Pool’ list. See the Routine Editor to load a different Routine Pool.
Start Routine	Manually run the selected Routine - start recording and displaying data in the Scope window. When you click the ‘Start’ button, the Scope is cleared, and data recording starts after ~300 ms. When acquisition is running, the Scope window updates every 200 ms. If the Sweep Start-to-Start time is ≥ 5 s, the “Time to next sweep: # s” is reported below the Start / Stop buttons. If Metadata prompts are configured for Routines or Paradigms, the Confirm Metadata Settings dialog displays just before recording begins. If measurement graphs are enabled, a docked “child” Analysis window opens and plots sweep-by-sweep measurements. If no prior auto-triggered Paradigm is running, a new date/time-stamped Paradigm is created.
Stop Acquisition	Stop recording Routine data. When you click the ‘Stop Acquisition’ button, data acquisition is halted for the Series.
Stop at End of Sweep	When ‘Stop at End of Sweep’ is enabled, and you click the ‘Stop Acquisition’ button in the middle of a sweep, the partial sweep completes before data acquisition is halted; otherwise, when disabled, the partial sweep is discarded when acquisition stops.

4.1.2 Acquisition Measurements & Graphs

Make real-time changes to the online measurements and graphs, even during data acquisition, with this dialog.

To access this dialog, click on the Scope (Acquisition) window button 'Measurements / Edit Measurements'.

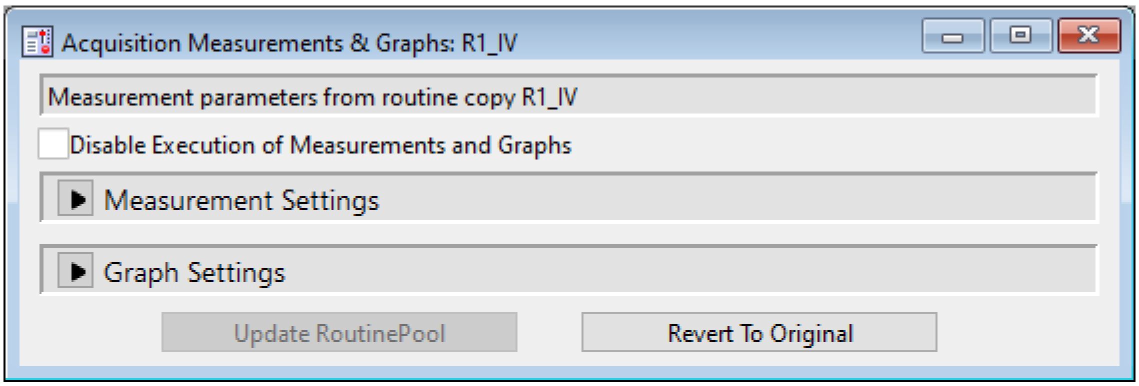


Figure 4-2. Acquisition Measurements & Graphs

This dialog is the same as in the Routine Editor / Real Time Measurements & Graphs dialog, with two extra buttons:

Update Routine Pool	Save your edits to the Routine.
Revert to Original	Discard any edits.

4.1.3 Camera Module

The SP_Camera window displays:

Camera	Select a camera name from those attached to the computer.
Start	View live video. This data is not stored.
Capture	Take a single picture. If live video is running, this will take a picture while live video continues to run. The image time-stamp is reported in the Log window.
Last Capture	A thumbnail of the last picture taken in the Experiment is displayed.

[video screen]

All pictures are stored in the current Experiment. To review pictures, go to:

Data Navigator	Select a Paradigm or Routine. Any associated images are listed in the Preview pane. Click on an image name to display the image.
Data Browser	Navigate to the Data / Images folder. Right-click on an image name, and select 'New Image'.

Tip: For dark-room experiments, the window background color can be adjusted by the operating system.

Windows:	In the Windows Control Panel / Appearance / Personalization window, scroll down and select the High Contrast Black theme, or use the Magnifier tool with option 'Turn on color inversion' enabled.
macOS:	Press 'Control-Option-Command-8' to set the System Preferences / Accessibility / Display / Invert Display colors option, or open its menu with 'Command-Option-5'.

Full-camera drivers have been successfully tested for the following camera models:

Sentech drivers:

STC-MC33USBVGA

STC-MCS231U3V

STC-MB83USBVGA

STC-MBCM401U3V

STC-MBCM401U3V-NIR

STC-HD203DV

Photometrics PVCAM drivers:

Photometrics:	Prime 95B
	Prime 95B 25mm
Qimaging:	Electro

4.1.4 Free Run

The Free Run (Scope) window simulates a one-channel oscilloscope, and is a quick method of viewing repetitive data.

It operates similarly to the Scope acquisition window, with unsupported controls removed or disabled.

Settings Window

Signal Parameters

Channel: Select an input channel to monitor:

Current1 - 2

Voltage1 - 2

AuxIN1 - 4

The default channel is 'Current1' in VC mode, and 'Voltage1' in CC mode.

Duration: The duration of the data sweep.

[100, 200 ms, 0.5 , 1 , 2 s]

Sampling rate: [5, 10, 20, 50 kHz]

4.1.5 Membrane Test

The Membrane Test is primarily used to monitor seal formation and cell health in a voltage-clamp whole-cell patch-clamp configuration.

However, current-clamp mode operation is also supported, if 'Electrode Compensation' (capacitance neutralization) is perfectly set.

Scope (Membrane Test)

This Scope (Membrane Test) window operates similarly to the Scope (acquisition) window. The default Scope window top pane displays the Current signal from the active Sutter headstage, and the pane beneath it displays the corresponding Voltage command signal.

Differences to the Scope window for the Membrane Test:

- Unsupported buttons and controls were removed (Persistence Display, Signal Layout, Sweeps Counter).

- During an Experiment, the Autoscale button state persists between windows.
- The Membrane Test opens in a "running" state.
- Closing the Membrane Test Settings panel halts acquisition.

Membrane Test Settings

Configure all Membrane Test parameters in the Membrane Test Settings panel. This panel automatically opens when the Membrane Test is opened or started.

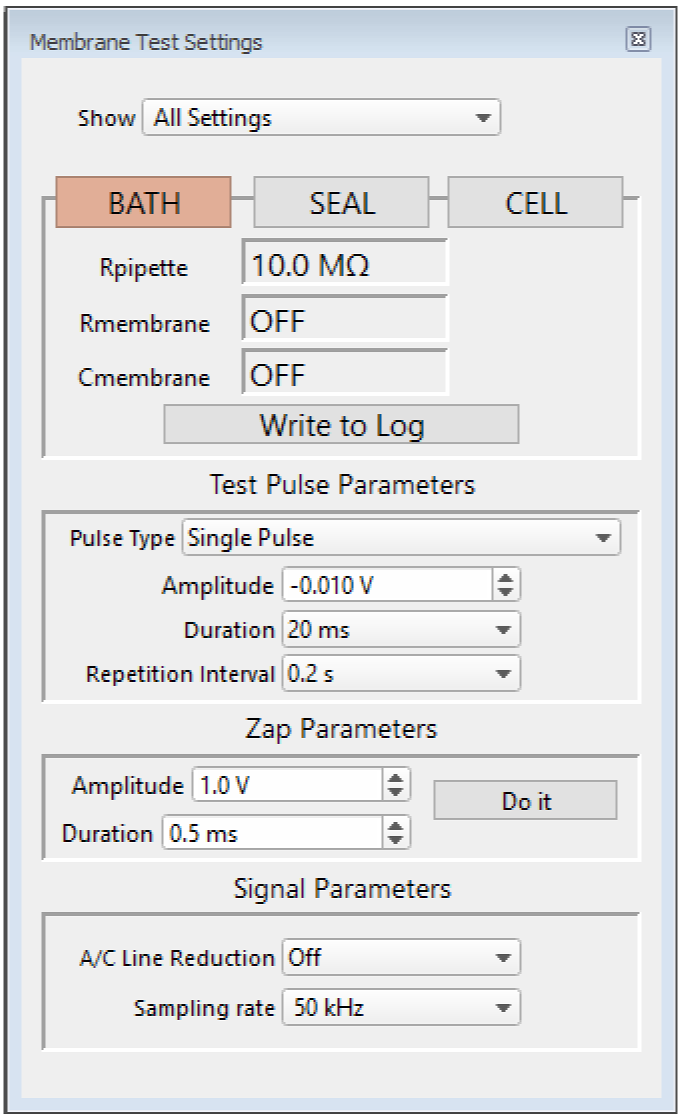


Figure 4-3. Membrane Test Settings

Show	Monitor Only
	Monitor + Test Pulse
	All Settings

 Open the Membrane Test 'Preferences' settings dialog.

Membrane Test Preferences

- Second Membrane Test channel
  - MT analysis on second channel
- ZAP controls
- Sound controls
- Line Frequency Reduction options
  - A/C Line Frequency: 50 Hz / 60 Hz
- Enable Holding with Seal mode selection

  When the Membrane Test 'Seal' mode is selected, the control panel 'V- or I-holding' level is also enabled.

Running Average Trace	The average of the last "N" traces, as set in 'Measure Parameters'.
Line Thickness	[0.25, 0.50, 0.75, 1 – 10]
Color	Open a color selection panel.
Average Traces	The last "N" traces used in the running average.
Line Thickness	[0.25, 0.50, 0.75, 1 – 10]
Color	Open a color selection panel.
Capture Trace	Set which trace is used by the "push pin" snapshot control in the Signal Parameters section.
Source	• Signal: Use the raw Membrane Test trace. • Average: Use the running average trace.
Line Thickness	[0.25, 0.50, 0.75, 1 – 10]
Color	Open a color selection panel.

The Membrane Test Analysis panel contains the three basic steps to form a whole-cell seal:

1) Bath — With a new pipette in the bath solution, a low-resistance square pulse is visible. The pipette resistance should be very low if the tip is not clogged.

For whole-cell patch clamping of dissociated/cultured cells, typical pipette resistances are 1 – 5 MΩ. For brain slice recordings, pipette resistances up to 20 MΩ or higher are used.

2) Seal — When an on-cell patch is formed between the pipette and the cell, voltage transition spikes are visible. The seal resistance increases as the seal forms. The goal is to achieve a "gigaseal" with a resistance above 1 GΩ.

This configuration is used for single-channel recordings, as well as inside-out and outside-out patches

3) Cell — After breaking through the cell membrane and creating a whole-cell patch, membrane resistance and capacitance measures are calculated from the resulting capacitance spikes.

The Membrane Test calculations are displayed in real-time numeric fields:

Bath	Rpipette (MΩ)	Pipette Resistance meter [Model cell = ~10 MΩ]
Seal	Rseal (MΩ)	Seal Resistance meter [Model cell = ~1 GΩ to 1 TΩ] (open circuit)
Cell	Rseries (MΩ)	Series Resistance meter [Model cell = ~10 MΩ]
	Rmembrane (MΩ)	Membrane Resistance meter [Model cell = ~500 MΩ]
	Cmembrane (pF)	Membrane Capacitance meter [Model cell = ~28 pF]

Warning! When in Cell mode, disable the Amplifier Control Panel 'Cell Compensation' and 'Rs Correction', so that whole-cell capacitance spikes are generated for the calculation of these values. Otherwise, these fields are reported as "OFF".

Tip: 'Series Resistance' and 'Access Resistance' are equivalent terms. It is helpful to monitor the health of the cell via the Series Resistance, as if it increases by more than 5%, the electrode tip might be clogged.

Note: Demo values for Cell mode can vary from the model cell values.

Reported values are dependent upon experimental variables and settings, such as cell and pipette size, solution conductivity, test pulse duration, etc.

Write to Log Click the 'Write to Log' button to write the last acquired measurements for that mode to the Log window. Valid measurements are logged for the active headstage.

Test Pulse Parameters

Pulse Type

- Single Pulse [1, 2, 5, 10, 20, 50, 100, Other: ±999.00 mV]

  A single monopolar square pulse.
- Double Pulse A symmetrical bipolar (biphasic) square pulse.
- Triangle A train of 5 symmetrical bipolar triangular pulses.

  [dPatch only]
- Sine A train of 5 sine wave pulses.

  [dPatch only]
- RMS Noise No pulse – the holding level is output.

Amplitude VC mode: [±1.00 V] A pulse amplitude is required. Any value less than ±0.1 mV (absolute) is reset to ±0.10 mV.

CC mode: [±2000.00 pA] A pulse amplitude is required. Any value less than ±0.1 pA (absolute) is reset to ±0.10 pA.

Amplitude is relative to the 'Holding' level in the Amplifier Control Panel.

Duration [10, 20, 50, 100, 200 ms]

Set long enough for the signal to reach its asymptote, or measurements can be incorrect.

Repetition Interval [0.1, 0.2, 0.5, 1.0, 2.0, 5.0, 10 s]

Sweep start-to-start interval.

Zap Parameters

After a gigaohm patch has been achieved, use Zap in the Seal mode as an alternative to suction in creating a whole-cell patch.

Amplitude [0.1 - 1.0 V]

Set the amplitude of the square wave zap pulse.

Duration	[0.1 – 2.0 ms]
Do It	Click the 'Do It' button to send a single square wave voltage pulse from the headstage to the preparation to disrupt the cell membrane.

Signal Parameters

Channel	Headstage 1
	Headstage 2
A/C Line Reduction	Off
	50 Hz
	60 Hz
Sampling rate	10 kHz
	20 kHz
	50 kHz

4.1.6 Paradigm Editor

The Paradigm Editor is an advanced feature that opens up a world of complex experimental control via Paradigms and Paradigm Pools. A rich set of operators and actions are available to control and/or automate data acquisition and analysis.

The Paradigm Editor allows you to create "Planned Paradigms", which offer almost unlimited flexibility in creating and/or automating your patch-clamp experiments, such as running Routines and directly controlling amplifier settings.

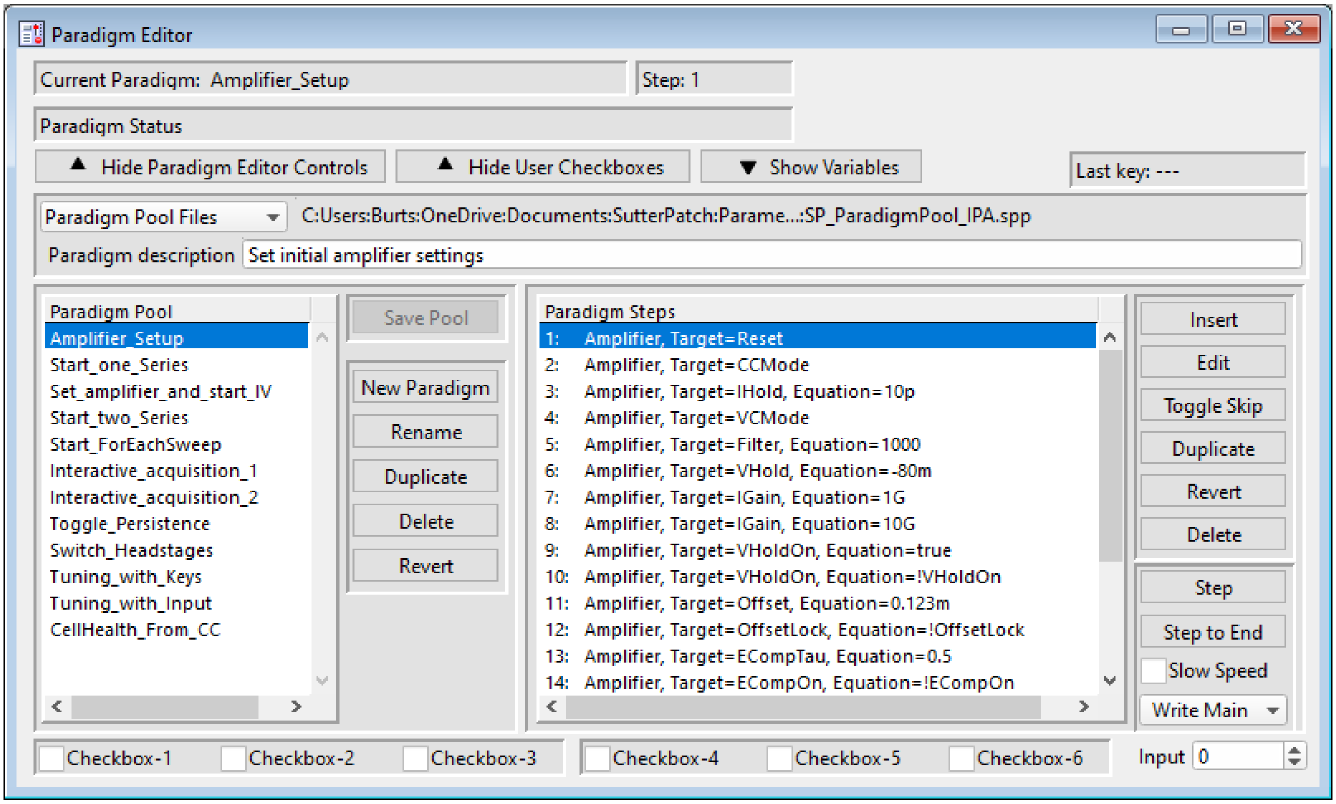


Figure 4-4. Paradigm Editor

Loaded Paradigms display on the left, while loaded Paradigm Steps display on the right. A bottom section can display interactive checkbox controls and/or variables.

Controls

Start/Stop, Set Tag and Reset Timer controls are located in the Acquisition Control panel.

Current Paradigm: The name of the currently loaded Paradigm.

Step: The highlighted Paradigm Step.

Paradigm Status: Status information about Paradigm execution.

Show/Hide Paradigm Editor Controls

The Paradigm Editor controls (and checkboxes) for the Paradigm Pool and Paradigm Steps can be displayed or hidden.

Show/Hide User Checkboxes

Checkbox controls are displayed at the bottom of the Paradigm Editor controls, for use in conditional Paradigm step execution. This display is dependent upon Show Editor Controls.

Show/Hide Variables: A Variables table can be displayed at the bottom of the Paradigm Editor. These paradigm variables can be utilized in any equation.

Variable names can be edited to any label, but they are only informational, and are not supported in equations.

Last key: The last key (or key combination) pressed on the keyboard is displayed here, such as used in Shortcuts or the 'If" and 'ElseIf" Paradigm steps. (See sample Paradigm 'Tuning with Keys'.)

Note: Function and Control (Ctrl/Cmd) Shortcut key combinations are not displayed.

Paradigm Pool Files These operations affect the entire "Paradigm Pool".

New Paradigm Pool: Create a new blank Paradigm Pool and optionally copy Paradigms into it from the existing Paradigm Pool.

The suggested name is auto-incremented from the previously loaded Paradigm Pool name.

Load Paradigm Pool: Load the Paradigms of a previously saved Paradigm Pool file into the Paradigm Pool.

Revert to Last Saved: Undo any unsaved changes to the Paradigm Pool.

Save Paradigm Pool: Save the Paradigm Pool using its existing file name and path.

Save Paradigm Pool As: Save the Paradigm Pool to a new file, and switch to the new file. The default file name is the same as the original file name.

Save Paradigm Pool Copy: Save the Paradigm Pool to a new file, but do not switch to the new file. The default file name has 'Copy of' prepended to it.

Merge Paradigm Pools: Insert the Paradigms from a previously saved Paradigm Pool file into the loaded Paradigm Pool.

[The file path and file name of the loaded Paradigm Pool file are displayed.]

Paradigm description: A user description of the active Paradigm.

Paradigm Pool A column of paradigm names from the loaded Paradigm Pool.

- Click on a Paradigm name to highlight it as the active Paradigm and display its steps.

- Double-click on a Paradigm name to start execution of the Paradigm and display its steps.
- Click-and-drag a Paradigm name to change its position in the column.
- To select multiple Paradigms, use a Shift-click mouse drag, or individually Shift-click the Paradigm names. Multiple Paradigms can thus be deleted, or saved to a new Paradigm Pool.

Save Pool — Save the Paradigm Pool using its existing file name.

New Paradigm — Create a new blank Paradigm in the Paradigm Pool.

Rename — Rename the selected Paradigm.

- Valid characters are A-Z, a-z, 0-9, and "_".
- Special characters are not allowed.
- Spaces are replaced by an underscore.
- The name cannot start with a number – such entries will have an 'X' prepended to the name.

Duplicate — Add a copy of the selected Paradigm to the Paradigm Pool. The Paradigm name number is appended or incremented.

Delete — Remove the selected paradigm from the Paradigm Pool.

Revert — Select a paradigm and click the 'Revert' button. All editable steps are reset to their originally loaded values, as long as the Paradigm Pool has not been saved.

Paradigm Steps — A column of instructions from the active paradigm is displayed. These instructions are sequentially run by the paradigm.

- Click on a paradigm step to highlight it as the active step.
- Double-click on a paradigm step to view or edit its settings.
- Click-and-drag a paradigm step to change its position in the column.

Note: Step values are usually in SI standard units, i.e. "Volts" and "Amperes".

Step Buttons

Insert Inserts a new command Step into the Paradigm Steps column:

Amplifier

Each Sweep

Routine

Analysis

Camera

Clear Key

Execute

Export

Front Window

Hide Window

Reset Timer

Scope Operation

Set Axis

Set Checkbox

Set Metadata

Set Solution

Set Tag

Update Inputs

Set Variable

Set Write Steps

Sound

View Last

Write to Log

	Alert
	Beep
	Comment
	Wait
	Pause

	Flow Control
	Break
	Chain
	For Loop
	Jump
	Label

	Condition
	If
	ElseIf
	Else
	(See details in Insertable Steps list below.)
Edit	If a highlighted Step is configurable, clicking the Edit button (or double-clicking the step) will open it in the Paradigm Steps Editor for configuration. Also, if a highlighted Step's text is partially hidden, use the Edit button to view the entire entry.
Toggle Skip	Mark a step so it is not executed. A semicolon is prepended to the Step number to "comment out" the instructions, and a Skip status is appended to the Step text.

Example: A ‘Beep’ command in Paradigm step #2:

; 2 Beep, Skip=true

The leading semicolon ";" prevents this step from being executed by the instruction queue, and the ‘Skip’ status is displayed.

Duplicate	Insert a copy of the selected step.
Revert	Select a Step to be reverted, and click the Revert button. Editable fields are reset to their originally loaded values, as long as another Paradigm has not been loaded.
Delete	Delete the selected step. For multi-line steps, optionally delete the entire group.
Step	Execute the selected step, then move to the next step. Executing a single step does not terminate a running Paradigm, even if it is the last step in the Paradigm. Note: A ‘For’ loop is processed as a single step.
Step to End	Execute the selected step and all following steps as fast as the system allows.
Slow Speed	Execute ‘Step to End’ at ~1 second per step.

<u>Write to Log</u>

- Write None — Write No Steps
- Write Main — Write Main Steps

  Action-oriented steps are recorded in the Paradigm metadata (visible in the Metadata ‘By Event’ view):

  Amplifier

  Break

  Camera

  Chain

  Execute

  For Each Sweep

  Reset Timer

Routine

Set Checkbox

Set Solution

Set Variable

Wait

- Write All — Write All Steps

  Log the main steps and additional steps into the Paradigm metadata (visible in the Metadata 'By Event' view.)

<u>Insertable Steps</u>

<u>**Amplifier**</u>

Control the IPA amplifier hardware.

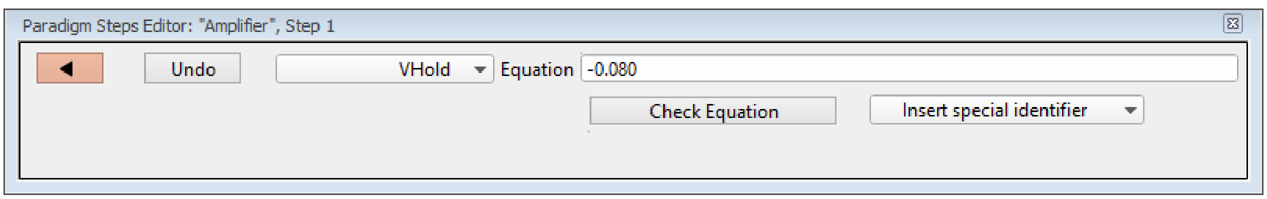


Figure 4-5. Step: Amplifier

Default Setting: *Amplifier, Target=VHold, Equation=-0.080*

 Close the 'Paradigm Steps Editor'.

Undo — Remove any unsaved edits to this step.

[drop-down list] — Amplifier settings:

SelectProbe — (select active probe)

[1 - 4]

Most Paradigm Step commands apply to the "active" probe, the Sutter headstage presently controlled by the Amplifier Control Panel. Select the target headstage.

For a single headstage system, the active probe is always headstage number "1".

CCMode	(amplifier current clamp)
	Place the amplifier into Current-Clamp mode.
VCMode	(amplifier voltage clamp)
	Place the amplifier into Voltage-Clamp mode.
Hold	(IHold in CC-mode, VHold in VC-mode)
	[±0.000,000,020 A (±20 nA), or ±1.000 V (±1000 mV)]
	Set the active headstage holding level.
IHold	(amplifer holding current, A)
	[± 0.000,000,020 A (±20 nA)]
	Set the active headstage holding level in Current-Clamp mode.
IHoldOn	(amplifier holding current On)
	Enable the active headstage holding level in Current-Clamp mode.
VHold	(amplifier holding voltage, V)
	[±1.000 V (±1000 mV)]
VHoldOn	(amplifier holding voltage On)
	Enable the active headstage holding level in Voltage-Clamp mode
IGain	(amplifier current gain, V/A)
	Set the gain for the active 'Current' input channel using standard unit numbers (V/A) or scientific notation (1 mV/pA = "1e9".). The value is converted to a preset Gain level:

- 0.5 mV/pA
- 1 mV/pA
- 2.5 mV/pA
- 5 mV/pA
- 10 mV/pA
- 25 mV/pA

To help reduce signal saturation from too high a gain, a 90% threshold promotes the equation value to the next higher Gain setting.

VGain (amplifier voltage gain, V/V)

Set the gain for the active 'Voltage' input channel using standard unit numbers (V) or scientific notation (1 mV = "1e3"). The value is converted to a preset Gain setting:

- 10 mV/mV
- 20 mV/mV
- 50 mV/mV
- 100 mV/mV
- 200 mV/mV
- 500 mV/mV

To help reduce signal saturation from too high a gain, a 90% threshold promotes the equation value to the next higher Gain setting.

Filter (amplifier input filter, Hz)

Apply a preset filter level to the input channels:

- 500 (500 Hz)
- 1000 (1 kHz)
- 2000 (2 kHz)
- 5000 (5 kHz)
- 10000 (10 kHz)
- 20000 (20 kHz)

To help prevent over filtering, a 10% threshold promotes the equation value to the next higher filter level.

Offset	(amplifier pipette offset, V)
	[±0.5 (±500 mV)]
OffsetLock	(amplifier pipette offset lock On)
	[1 = On, 0 = Off]
VTrack	(amplifier tracking potential, V)
	[±1 (±1000 mV)]
	Set a value and enable slow CC Tracking
VTrackOn	(amplifier tracking potential On)
	[1 = On, 0 = Off]
	Enable slow CC Tracking
ECompMag	(amplifier electrode compensation magnitude, F)
ECompPhase	(amplifier electrode compensation phase, fraction)
ECompOn	(amplifier electrode compensation phase On)
	1 = On, 0 = Off]
CmComp	(amplifier cell compensation Cm, F)
	Set a cell capacitance value and enable cell capacitance compensation
RsComp	(amplifier cell compensation Rs, Ohm)
	Set a series resistance value and enable cell capacitance compensation
RsCompOn	(amplifier cell compensation On)
	[1 = On, 0 = Off]
RsCorr	(amplifier Rs correction, fraction)

[0.00 – 1.00] Converted to a percentile

RsLag (amplifier Rs correction lag, s)

RsCorrOn (amplifier Rs correction On)

[1 = On, 0 = Off]

Bridge (amplifier bridge balance, Ohm)

Reset (reset amplifier controls)

AutoEComp (amplifier auto electrode compensation)

AutoCellComp (amplifier auto cell compensation)

AuxOUT1 (Auxiliary Output-1, V)

AuxOUT2 (Auxiliary Output-2, V)

DigOUTWord (Digital Output Word)

DigOUT1 (Digital Output-1)

DigOUT2 (Digital Output-2)

DigOUT3 (Digital Output-3)

DigOUT4 (Digital Output-4)

DigOUT5 (Digital Output-5)

DigOUT6 (Digital Output-6)

DigOUT7 (Digital Output-7)

DigOUT8 (Digital Output-8)

LockInAdjustOn (set LockIn adjustments On)

LockInPhaseAdj (set LockIn phase delay adjustment)

LockInAttenAdj (set LockIn attenuation adjustment)

[Equation field] A free-form text field. This field is evaluated and its value passed to the “target” function.

[Errors are reported under this field.]

Note: Values in the Equation field are rounded to whole numbers.

While Amplifier steps are configured in standard units (Amperes, Volts), the Amplifier Control Panel displays values in scaled units.

Check Equation — Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports “Syntax is ok.”

Insert special identifier — Acquisition, amplifier and reference settings are available for use in equations. (See list below.)

Each Sweep

Control the Paradigm operations on a “per sweep” basis of a Routine. Commands to be executed are inserted between the “EachSweep, Target” line and the “ForEachEnd” line.

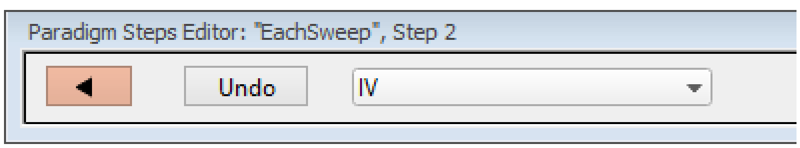


Figure 4-6. Step: Each Sweep

Default Setting: *ForEachSweep*
EachSweep, Target=untitled
ForEachEnd

 Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

[drop-down list] — Select a Routine name from the loaded Routine Pool.

Edit routine ‘<name>’ — Open for editing in the Routine Editor.

For example, when tracking whole-cell Rs values in real-time measurement graphs, update the Cell Compensation sweep-by-sweep, by inserting an Amplifier step (with its AutoCellComp command selected) within an EachSweep loop

Note: When using ‘Each Sweep’ to record data, the minimum sweep start-to-start time is +200 ms. For faster execution times, use the ‘Routine’ step.

Routine

Run acquisition.

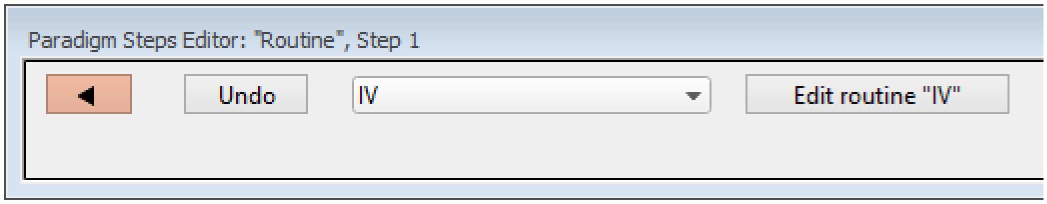


Figure 4-7. Step: Routine

Default Setting: *Routine, Target=untitled*

 Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

[drop-down list] — Select an acquisition type, or a Routine to record a data Series.

Membrane Test — Repeats [1 – 999]

Indefinitely — Repeat an “infinite” number of times.

Write to Metadata

Configuration

- No change
- Bath
- Seal
- Cell

Free Run

Duration — [100 ms – 999.9 s / inf]

Indefinitely — Acquire for an “infinite” duration.

Add Channel — Clear

List of input channels

[selected channels]

List of Routine names from Routine Pool

[selected Routines]

Edit routine “<name>” Open for editing in the Routine Editor.

Note: The time from starting this command to recording data is +300 ms.

“Single-stepping” this command (when no Paradigm is running) will create an auto-triggered Paradigm.

Analysis

Save an analysis to the Analysis Editor, or combine it with prior analyses.

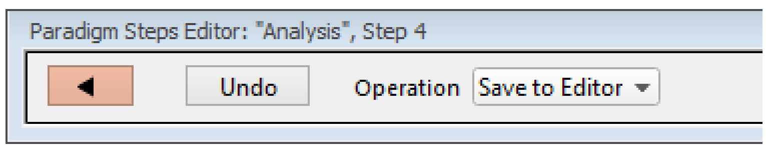


Figure 4-8. Step: Analysis

Default Setting: *Analysis, Operation=Save to Editor*

 Close the ‘Paradigm Steps Editor’.

Undo Remove any unsaved edits to this step.

[Operations]

- Save to Editor Save the latest analysis
- Append to Last Append to the prior analysis
- Average with Last Average with the prior analysis
- Show Table Display analysis as numeric table
- Show Graph (1 – 8) Display analysis as visual graph

Camera

Take a single picture and/or run a live video preview. A Camera window is opened behind the Paradigm Editor and Scope windows.

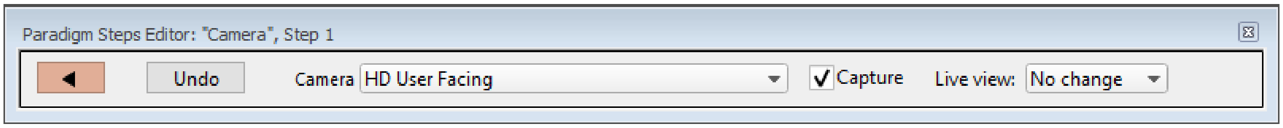


Figure 4-9. Step: Camera

Default Setting: *Camera, Camera=_Camera_Name, Capture =true*

 Close the 'Paradigm Steps Editor'.

Undo Remove any unsaved edits to this step.

Camera Select a camera on the computer system.

Capture Take a picture when executed.

Live view: Configure the state of the live view:

- No Change Keep last settings
- Stop Stop live view
- Start Start live view

Clear Key

Clear the 'Last key' field in the Paradigm Editor, which holds the last-pressed keyboard key since the start of the Paradigm.

Default Setting: *ClearKey*

Execute

Extend the functionality of SutterPatch by running an Igor command.

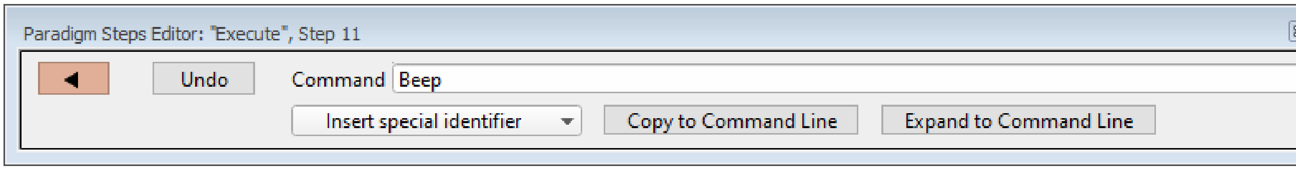


Figure 4-10. Step: Execute

Default Setting: *Execute, Command=Beep*


Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

Command — [ ]

Run any Igor command accepted by the Command window, including user-created Functions.

Note: Igor syntax usually requires that open/close parentheses “( )” be appended to the end of a command. However, exceptions include the “beep” and “print” commands, for which no parentheses are used.

Insert Special Identifier

Special references can also be used within commands:

- p[1..16] (n’th Paradigm variable)
- s[series-count, sweep-count, trace-count, routine name]

  (trace of specified series)

  Reference an input trace (data wave) via counts of Series #, Sweep #, Trace # (Scope position), and the Routine name.

  The “current” item is the “active” trace in the Scope window, and has a count value of zero.

If a “count” number is non-zero, it is used as an offset from the current count value of zero. Any fractions in count numbers are truncated to integers.

If the routine name is left blank, the current routine name is used.

Ex: s[0,0,0,]

The current series, current sweep, current trace, of the current routine.

- t[#] (trace of current sweep of current series)

  Reference the input trace (data wave) in Scope position “n” for the last sweep of the current Series.

- eq[equation] (result of the given equation)

Copy to Command Line: Append the Command text to the Command window’s Command line.

Expand to Command Line: Append the Command entry to the Command window’s command line, after processing it to be compatible with Command window execution, i.e., any variables are replaced by their values.

Example 1: Reset the Timer.

Set the Execute ‘Command’ to:

Paradigm_ResetTimer()

Note the open and close parentheses at the end.

Example 2: Create an FFT graph of your data.

The Paradigm Steps:

1. ForEachSweep

2. EachSweep, Target=*YourRoutineName*

3. Execute, Command=FFT/OUT=3/DEST=Voltage1_FFT t[2]

4. If, Left=sweep, Operation="=", Right=1

5. Execute, Command=Display Voltage1_FFT

6. EndIf

7. Execute, Command=SetAxis Bottom 0,60

8. ResetTimer

9. ForEachEnd

In Step 2: Replace "*YourRoutineName*" with your own Routine name, or use the sample "IV" Routine.

In Step 3: The Igor 'FFT' command is run, and "t[2]" retrieves the Scope's second input trace.

Export

Export data graphs into a Layout window.

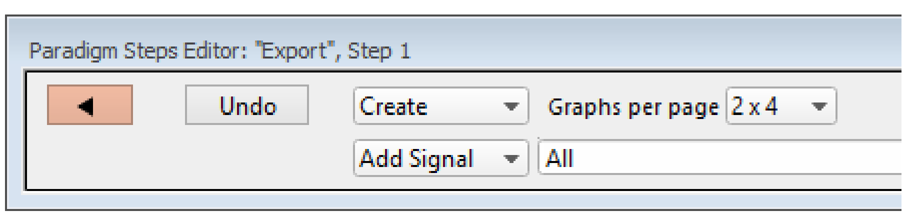


Figure 4-11. Step: Export

Default Setting: *Export, Signal=Layout*

 Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

[drop-down list]

- Append — Append graphs to an open Layout window, or into a new window.
- Create — Create a new Layout window.

Graphs per page — Set the graph layout configuration for new Layout windows:

- 1 — Graph fills entire page
- 2 — Graphs stacked
- 3 — Graphs stacked
- 2 x 2 — matrix
- 2 x 4 — matrix

[drop-down list] — Select signals to be exported from a list of default names.

- Clear — Clear the signal field, set it to ‘off’.
- All — Select all entries.
- All Signals — Select all input signals.
- < List of input signals >
- All Analyses — Select all Analysis graphs
- < List of Analysis graphs >
- [List of selected signals] — User-edited names can be directly entered into the signal field.

Note: The sequence of signals is not used for positioning in the Layout window – signal positioning is based on their Scope window sequence.

Front Window

Set the specified window as the front window.

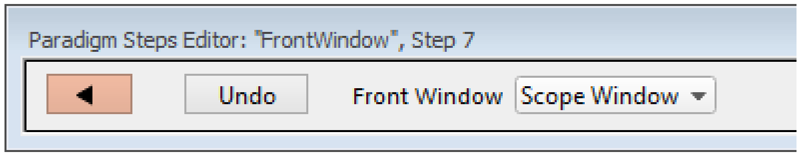


Figure 4-12. Step: Front Window

Default Setting: Front Window, Target=Scope Window

 Close the 'Paradigm Steps Editor'.

Undo Remove any unsaved edits to this step.

Front Window
- Analysis Editor
- Camera Window
- Control Panel
- Dashboard
- Data Navigator
- Equation Editor
- Log Window
- Paradigm Editor
- Routine Editor
- Scope Window
- Shortcut Editor
- Solution Editor
- Template Editor

Hide Window

Hide the specified window.

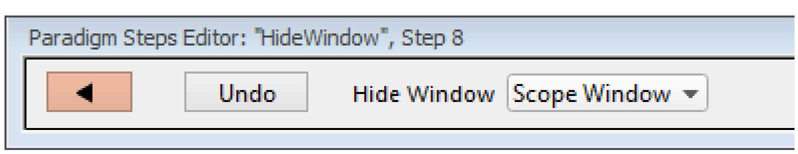


Figure 4-13. Step: Hide Window

Default Setting: HideWindow,Target=Scope Window

	Close the 'Paradigm Steps Editor'.
Undo	Remove any unsaved edits to this step.
Hide Window	Analysis Editor Camera Window Control Panel Dashboard Data Navigator Equation Editor Log Window Paradigm Editor Routine Editor Scope Window Shortcut Editor Solution Editor Template Editor

Reset Timer

Reset the Paradigm Editor Timer to 00:00:00.

Default Setting: *ResetTimer*

Scope Operation

Control which Scope window signals are displayed, and how the sweep display operates.

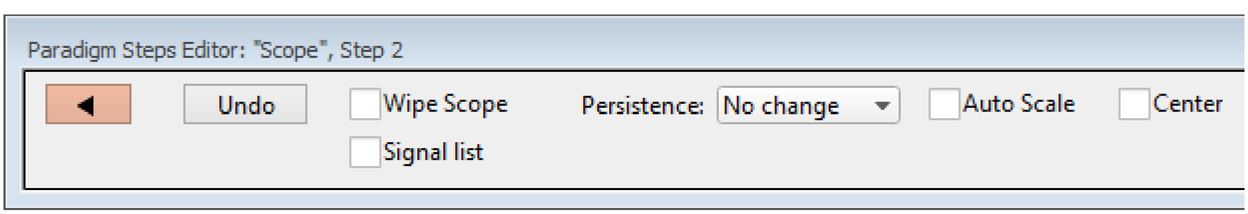


Figure 4-14. Step: Scope Operation

Default Setting: *S*cope, Wipe=false


Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

Wipe Scope — Clear the Scope window of all sweeps, except the last one.

Signal list: — Enable to display a list of input signals

Add Signal

Clear — Clear the signal list.

All Signals — Select all signals.

[List of all possible input signals]

[List of selected input signals]

You can directly edit the list. User-defined signal labels can also be used.

Persistence:

No change

On

Off

Autoscale — A one-time Autoscale of the Y-axes of all selected signals to their incoming data, i.e., to their visible sweeps data limits, and resets the X-axes to the full sweep duration.

Center — Center the active signal so the mean of the Y-axis data is vertically centered in the signal pane. Only the Y-axis offset is automatically adjusted, not the scaling; the X-axis is unaffected.

Set Axis

Modify the axis scaling of a signal.

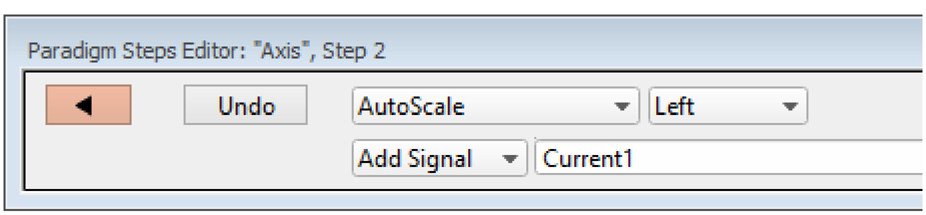


Figure 4-15. Step: Set Axis

Default Setting: *Axis, Axis=Autoscale, Kind=Left, Target=Current1*

 Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

[drop-down list] — Select the operation to perform.

- Autoscale — Match the axis range to the data range.
- Autoscale from Zero — Display from zero to the largest value.
- Full scale — Display the full range of the axis.
- Use last — Keep using the last-used settings.

[drop-down list] — Select the axis orientation

- Left — Apply to the Y-axes on the signal list.
- Bottom — Apply to the X-axes for all signals.

Add Signal — Manage the signal list.

- Clear — Clear the signal list.
- All Signals — Select all signals.
- [List of all input signals]

[list of selected signals] — Select an input signal to modify from a list and/or directly enter user-defined signal labels.

Set Checkbox

Set Checkbox uses simple “on / off” toggles. Checkbox status can be read by ‘If” and ‘ElseIf’ steps to make “yes/no” decisions and control the execution path of the Paradigm. If the equation evaluates to a non-zero value, the checkbox is enabled, i.e., ”on”.

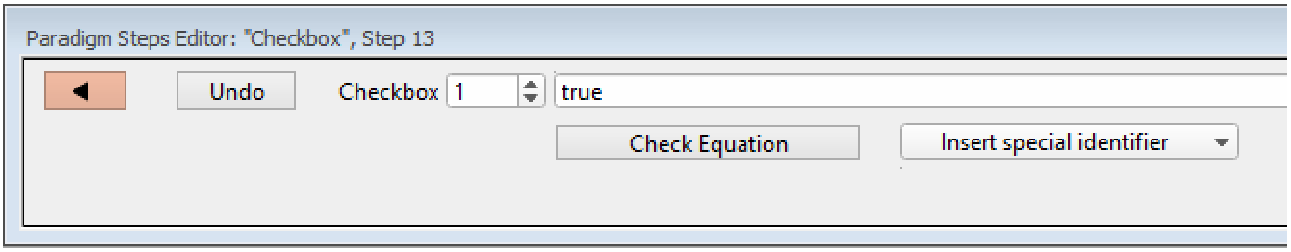


Figure 4-16. Step: Set Checkbox

Default Setting: *Checkbox, Count=1, Equation=true*

 Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

Checkbox — Checkboxes [1 – 3] are local: they are cleared whenever a Paradigm is started.

Checkboxes [4 – 6] are global: they are not automatically cleared, so their status persists across all Paradigms in the Experiment.

[Equation field] — A free-form text field, evaluated to a value, and applied to the Checkbox.

[Errors are reported under this field.]

Check Equation — Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is ok.”

- The constant “True” evaluates to ‘1.000’.
- The constant “False” evaluates to ‘0.0000’.

Insert special identifier — Acquisition, amplifier and reference settings are available for use in equations. (See list below.)

Set Mark

The ‘Set Mark’ step marks (or unmarks) a sweep for later processing.

Use within a conditional paradigm step to mark or unmark a sweep, based upon experimental conditions.

For example, when used within a paradigm ‘If’ step, if the leak current is too high, unmark the sweep, else mark the sweep. This is an easy way to process just the sweeps that have a reasonable leak current.

Figure 4-17. Step: Set Mark

Default Setting: *SetMark, Value=Set*

Close the 'Paradigm Steps Editor'.

Undo — Remove any unsaved edits to this step.

Mark

- Set
- Clear

Marking / unmarking a sweep marks / unmarks that sweep in all signals in the same Series. Marked sweeps are loaded into the Data Navigator as "marked".

Marks are used by the Data Navigator 'Available actions':

Action Potential Analysis

Synaptic Event Analysis

Average Selected Sweeps

Display Signal/Sweep

Export Data

Set Metadata

Define Metadata parameter values to apply.

Go to Preferences > Metadata to set the detail level.

Default Setting: *Metadata, Value=*

The "Set Metadata Paradigm Step Value" dialog opens.

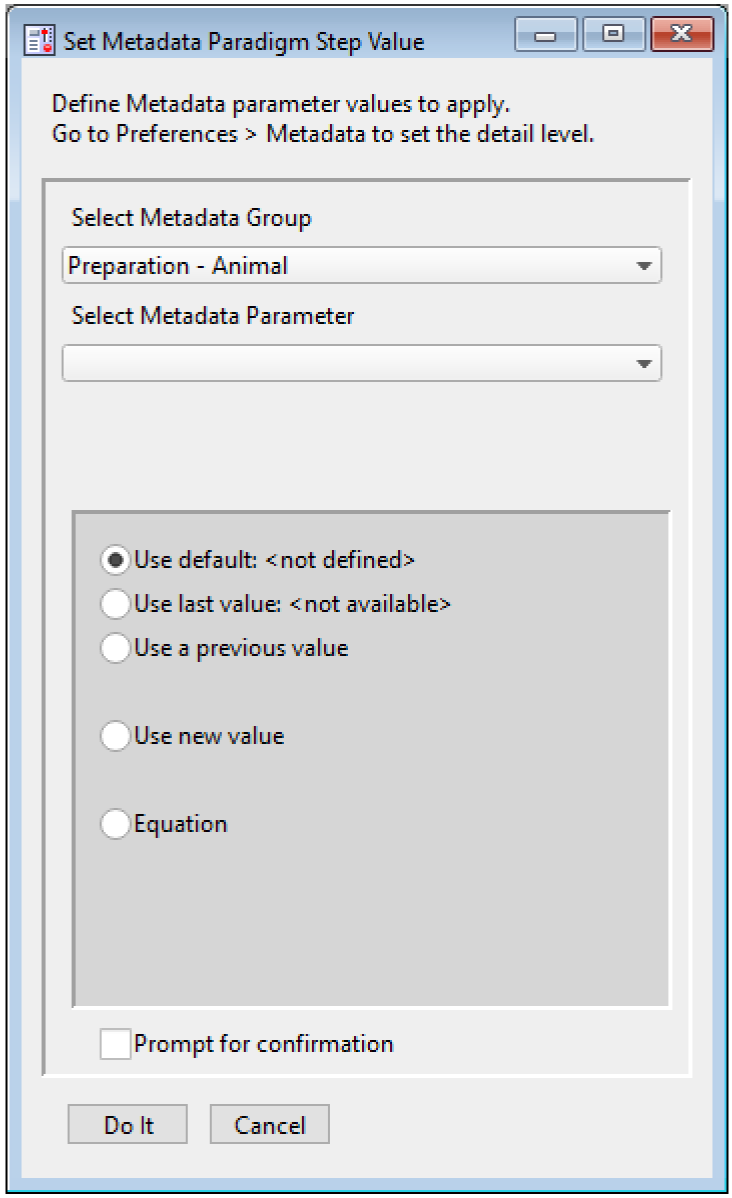


Figure 4-18. Step: Set Metadata

Select Metadata Group

• Operator	(Full detail level)
• Preparation – Animal	(Basic detail level)
• Preparation – Tissue	(Basic detail level)
• Preparation – Cell	(Basic detail level)
• Experiment	(Basic detail level)
• Electrode	(Extended detail level)
• Recording Solutions	(Extended detail level)
• Paradigm	(Full detail level)

- Cell Health / Quality Control (Full detail level)
- Series (= Routine Data) (Full detail level)
- Stimulus (Basic detail level)

Select Metadata Parameter	Entries depend on the selected Group.
[Metadata Parameter Info]	
• Use default:	
• Use last value:	
• Use a previous value	
• Use new value	
• Equation	
[<Define Equation>]	
Check	Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is ok.”
Special identifier	Special functions for use in equations. (See list below.)
Prompt for confirmation	Display a metadata prompt before acquisition.

.

Set Solution

A “solution” command is used to turn solution valves ‘on’ or ‘off’ in perfusion systems. A predefined digital pattern or analog level can be automatically output with this step. Solution settings are configured and numbered in the Solution Editor.

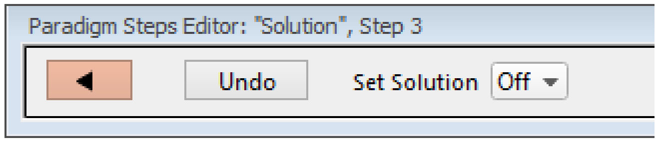


Figure 4-19. Step: Set Solution

Default Setting: *Solution, Target=Off*

Close the 'Paradigm Steps Editor'.

Undo — Remove any unsaved edits to this step.

Set Solution — Select a solution number to activate its valve. The number of available solutions depends on the Solution Editor configuration.

[1 – 24]

Set Tag

A comment tag is automatically written to the Paradigm metadata with this step. Enter the comment into the 'Tag text' field.

When run during acquisition, the comment tag is also written to the Routine metadata, and when the data is opened in a reanalysis Scope window, a black vertical cursor displays at that time point.

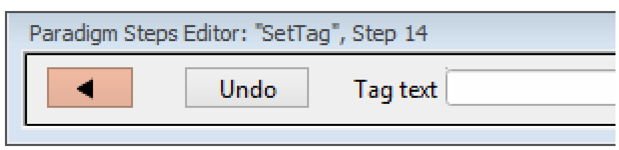


Figure 4-20. Step: Set Tag

Default Setting: *SetTag, Text=*

Close the 'Paradigm Steps Editor'.

Undo — Remove any unsaved edits to this step.

Tag text — Enter the comment text.

Note: The comment text for this Paradigm step is maintained separately from the manually triggered 'Set Tag' button text.

[1 – 24]

Update Inputs

Takes a single “live” reading from the Auxiliary Inputs.

Use for slowly changing variables, such as temperature.

Set Variable

Variables allow flexible control of any operation using equations.

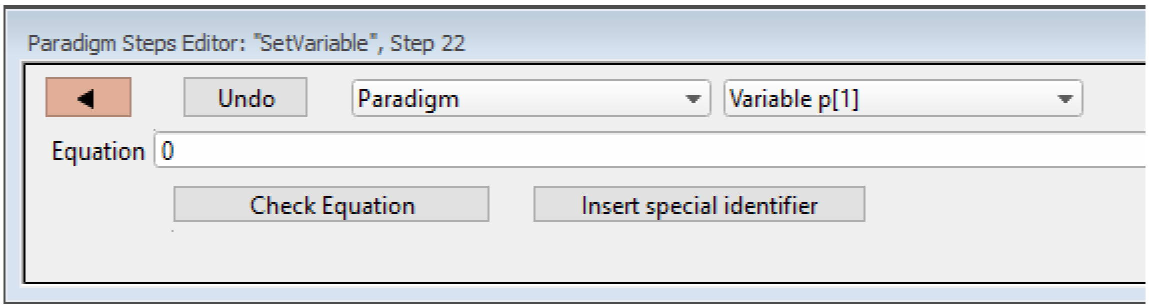


Figure 4-21. Step: Set Variable

Default Setting: *SetVariable, Target=Paradigm, Count=1, Equation=p[1]*

 Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

[list of targets]

- Paradigm — Set the value of a Paradigm Variable_p[#]

  [1 – 16, All Variables]

  When ’All Variables’ is selected, if varying values are desired, enter their values into the Equation field as a comma-separated list; simple equations (those without internal commas) can also be used in place of a value.) If there are more variables than list values, the “extra” variables are unused and unchanged. If a list value is blank, the corresponding variable is unchanged.

- Paradigm_Input — Set the value of the Paradigm Editor ‘Input’ control.

- [Routine Names]

  Select a Routine and set the value of its Variable_r[#]

  [1 – 16, All Variables]

  When ’All Variables’ is selected, if varying values are desired, enter their values into the Equation field as a comma-separated list; simple equations (those without internal commas) can also be used instead of a value.) . If there are more variables than list values, the “extra” variables are unchanged. If a list value is blank, the corresponding variable is unchanged.

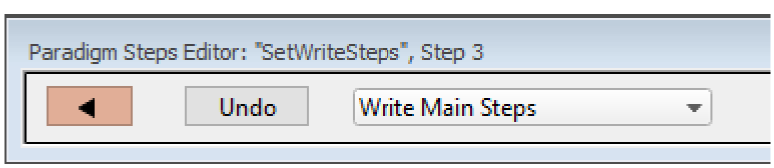
Equation	Evaluates to a value to set variables or the Paradigm Editor ‘Input’ control. You can likewise set the value of a variable by inserting special identifiers; for example, ‘Input’ reads the ‘Input’ control. (See sample Paradigm ‘Tuning_with_Input’.)
Check Equation	Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is ok.”

Insert special identifier

Acquisition, amplifier and reference settings are available for use in equations. (See list below.)

Set Write Steps

Configure the level of logging Paradigm metadata.



Default Setting: *SetWriteSteps, Value=Main*

Close the ‘Paradigm Steps Editor’.

Undo Remove any unsaved edits to this step.

[Drop-down list]

- Write No Steps
- Write Main Steps

Action-oriented steps are recorded in the Paradigm metadata (visible in the Metadata 'By Event' view):

Amplifier

Break

Camera

Chain

Execute

For Each Sweep

Reset Timer

Routine

Set Checkbox

Set Solution

Set Variable

Wait

- Write All Steps

Log the main steps and additional steps into the Paradigm metadata (visible in the Metadata 'By Event' view.)

Sound

Output a note from the computer speaker.

The frequency can be defined by a fixed value or an equation.

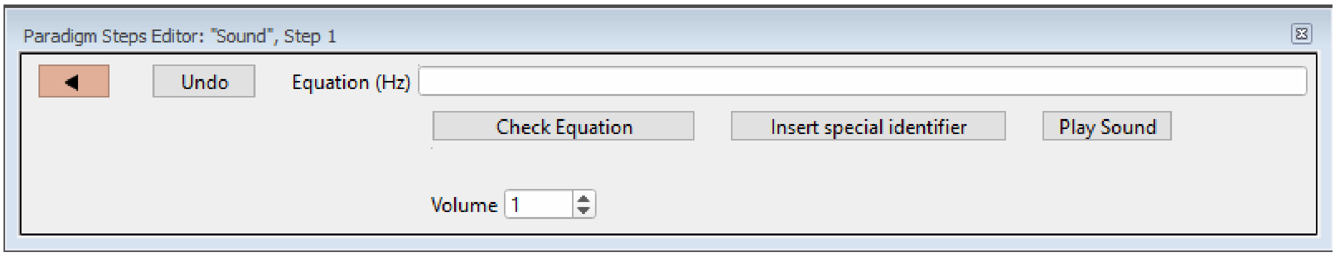


Figure 4-22. Step: Sound

Default Setting: *Sound, Equation=, Volume=1*

Close the 'Paradigm Steps Editor'.

Undo

Remove any unsaved edits to this step.

Equation (Hz)

[250 – 8000]

Specify as an equation or fixed value.

The sound output has a linear frequency response range within its limits.

< 250 Hz:	two clicks
> 8 kHz:	8 kHz tone

Check Equation

Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is ok."

Insert special identifier

Acquisition, amplifier and reference settings are available for use in equations.
(See list below)

Play Sound

Test the sound output.

Volume

[0.1 – 1.0]

Use the spinners for 10% increments, or directly edit the field.

Output is via the standard sound output that Igor uses:

- Windows: Built-in speakers, or a computer sound card with external speakers

  Note: Lower frequency tones are attenuated in volume on lower-quality speakers

- macOS: Built-in speakers

This paradigm step can also be utilized as an Igor programming command. For instance, using an equation, one could listen to the membrane resistance of the cell under investigation

Example: Output a note.

Enter this equation in the Command window command line:

SutterPatch#Paradigm_PlaySound(400, 1)

View Last

Display the last recording in a Scope (analysis) window.

Write to Log

Enter text to be written to the Log window.

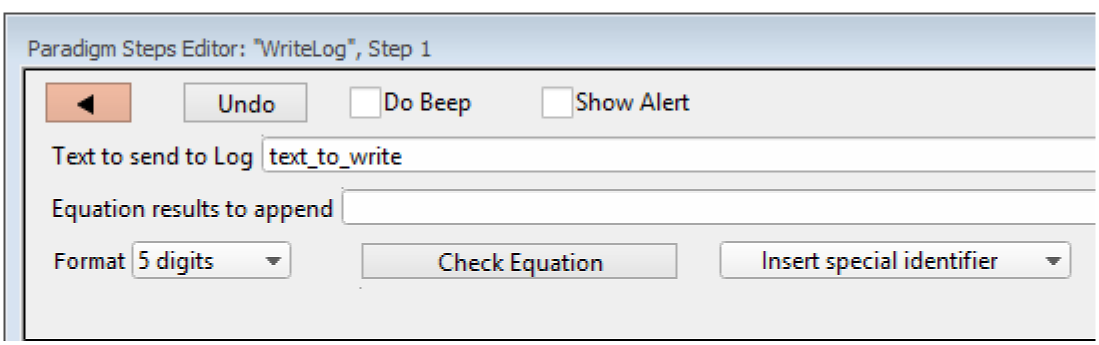


Figure 4-23. Step: Write to Log

Default Setting: *WriteLog, Alert=true, Text=text_to_write, Equation=, DoBeep*

Close the 'Paradigm Steps Editor'.

Undo	Remove any unsaved edits to this step.
Do Beep	Generate a beep before writing.
Show Alert	Display and/or edit the Alert text, then write it to the Log window.

Text to send to log [ ]

Equation result to append [ ]

Multiple equations in a comma-separated list can be evaluated.

Format	Time Date 1 – 12 digits
Check Equation	Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is ok."

Insert special identifier

Acquisition, amplifier and reference settings are available for use in equations.
(See list below.)

Run-time dialog: Write to log

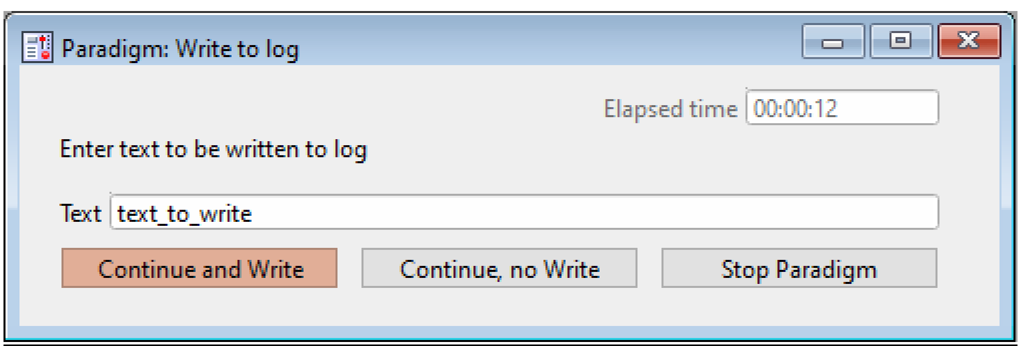


Figure 4-24. Write to Log Run-Time Window

Elapsed time	A time counter for the Alert.
Text	Edit the text message.
Continue and Write	Write to the Log window.
Continue, no Write	Do not write to the Log window.

Stop Paradigm | Stop the Paradigm.

Alert

Display an “Alert” dialog box that pauses Paradigm execution until manually dismissed.

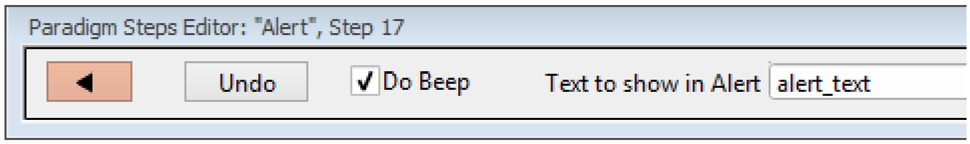


Figure 4-25. Step: Alert

Default Setting:	*Alert, Text=alert_text, DoBeep=true*
	Close the ‘Paradigm Steps Editor’.
Undo	Remove any unsaved edits to this step.
Do Beep	Sound a “beep” from the computer
Text to show in Alert	Enter a message to the user.

Beep

Generate a “beep” sound from the computer speaker.

Default Setting: *Beep*

Comment

A text message can be displayed in a floating window.

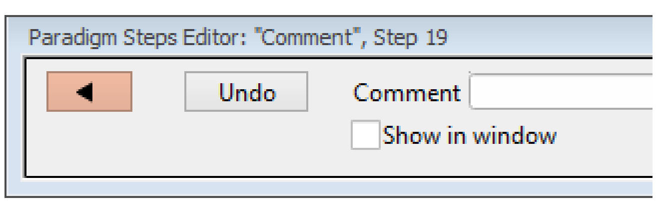


Figure 4-26. Step: Comment

Default Setting: *Comment, Text=*

	Close the ‘Paradigm Steps Editor’.
Undo	Remove any unsaved edits to this step.
Comment	Enter the comment text.
	To display multiple lines of text (up to 3), use “\r” as a line separator. Enter up to 40 characters per line, with a maximum of 100 characters per Comment.

Note: Text characters are from the ANSI character set.

Show in window	A ‘Paradigm Comment’ window is displayed with the comment text, and closes when the paradigm ends.

Wait

Temporarily pause execution of the Paradigm for a defined duration.

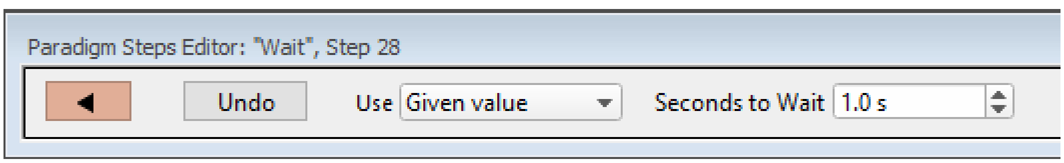


Figure 4-27. Step: Wait

Default Setting:	*Wait, Time=1*
	Close the ‘Paradigm Steps Editor’.
Undo	Remove any unsaved edits to this step.
Use	[Given value, Variable p[1] – p[16]]
Seconds to Wait	[displays for “Given value”]
	Click the spinners for 0.1 s increments, or type in a value. The precision of the wait time is 5 ms.

Pause

Pause execution of the Paradigm until the Resume button is manually clicked.

Break

Use a Break step to stop the execution of a Paradigm, or to interrupt For Loop and For Each Sweep loops.

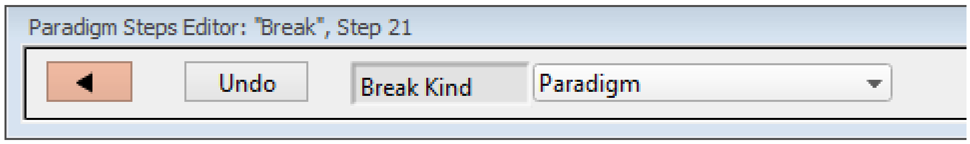


Figure 4-28. Step: Break

Default Setting: *Break, Kind=Paradigm*

 Close the ‘Paradigm Steps Editor’.

Undo: Remove any unsaved edits to this step.

Break Kind: Paradigm
ForLoop

Chain

Use to link step execution to another Paradigm.

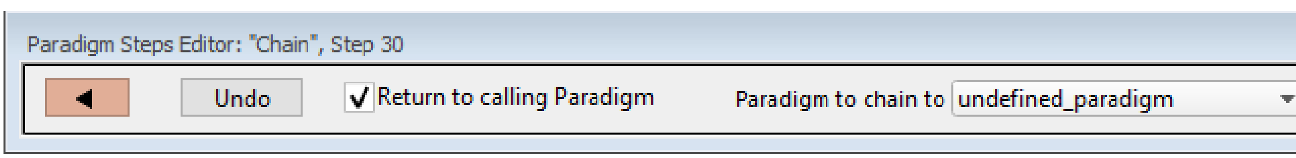


Figure 4-29. Step: Chain

Default Setting: *Chain, Target=undefined_Paradigm, Return=true*

 Close the ‘Paradigm Steps Editor’.

Undo: Remove any unsaved edits to this step.

Return to calling Paradigm:
Once execution of the target Paradigm has completed, return execution to this Paradigm.

Paradigm to chain to: Paradigm execution will shift to the selected Paradigm.

For multiple Chains (or recursive calls), you can link a maximum of eight Paradigms.

For Loop

Use a standard programming "For loop" to repeat a set of steps.

Note: A 'For' loop is processed as one step

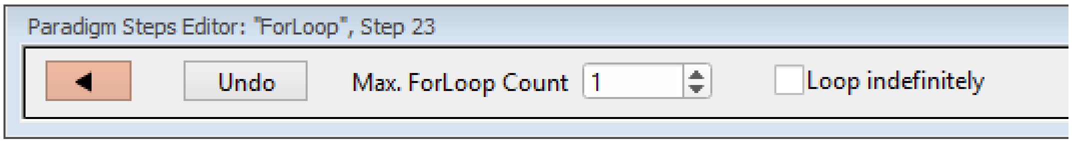


Figure 4-30. Step: For Loop

Default Setting:	*ForLoop, Max=1* *ForEnd*
	Close the 'Paradigm Steps Editor'.
Undo	Remove any unsaved edits to this step.
Max. ForLoop Count	Number of loop cycles to run
Loop Indefinitely	Sets 'Max. ForLoop Count' to 'inf'

Jump

Shift the Paradigm sequence to an arbitrary step. When executed, a jump occurs to the step after the target Label.

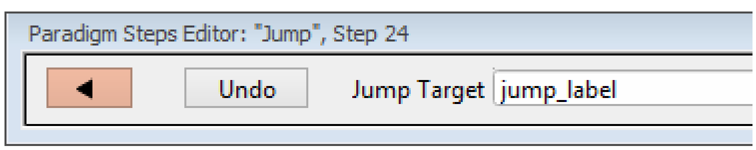


Figure 4-31. Step: Jump

Default Setting:	*Jump, Target=jump_label*
	Close the 'Paradigm Steps Editor'.
Undo	Remove any unsaved edits to this step.
Jump Target	Enter the Label of the step to jump to.

Label

Create a Label for a Jump step.

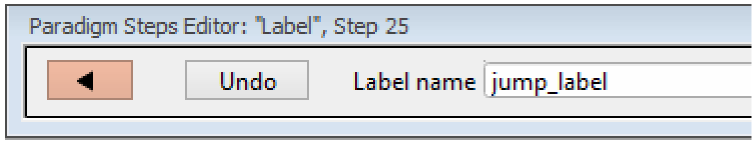


Figure 4-32. Step: Label

Default Setting: *Label, Target=jump_label*

 Close the 'Paradigm Steps Editor'.

Undo Remove any unsaved edits to this step.

Label name Assign a name to the Label.

If

This step allows conditional Paradigm flow control between multiple choices.

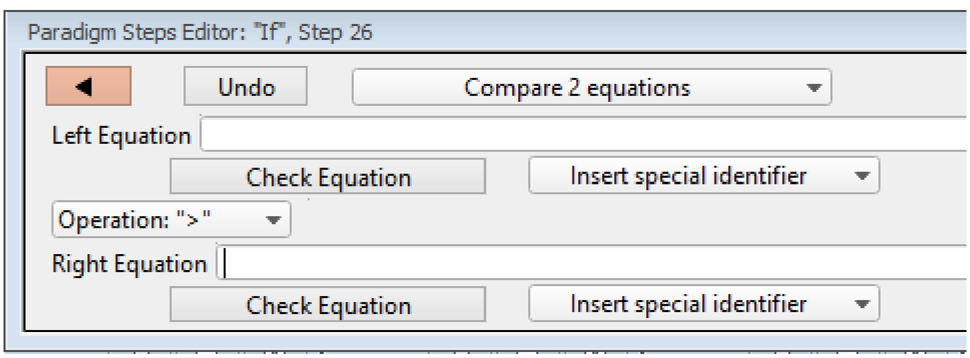


Figure 4-33. Step: If

Default Setting: *If, Left=, Operation='>', Right= EndIf*

 Close the 'Paradigm Steps Editor'.

Undo Remove any unsaved edits to this step.

[drop-down list] Operation selection.

- Compare 2 equations

  Left Equation Evaluated to a value.

Check Equation	Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is ok.”
Insert special identifier	Acquisition, amplifier and reference settings are available for use in equations. (See_list_below.)
Operation:	Comparison operators.
>	Greater than
>=	Greater than or equal to
=	Equal to
!=	Not equal to
<=	Less than or equal to
<	Less than
Note:	Be careful when comparing two floating-point numbers for equality, as minor variations in resolution can affect the outcome.
Right Equation	Evaluated to a value.
Check Equation	Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is ok.”
Insert special identifier	Acquisition, amplifier and reference settings are available for use in equations. (See_list_below.)

- Check for key pressed — “Last key” typed on keyboard.

  The “Last key” field is cleared at the start of a Paradgim.

Key to check for	Enter a text key, or insert a “special” key.
Insert special key	Use a “non-text” key.

  - Space
  - Return

  - Esc

- Check checkbox status — Select a checkbox to monitor for “on/off” status.

  Checkboxes are displayed at the bottom of the Paradigm Editor window.

  Checkbox

[1 – 3]	Paradigm-level “local” checkboxes are cleared at the start of a Paradigm.
[4 – 6]	Experiment-level “global” checkboxes persist across Paradigms for the entire Experiment.

- Answer of yes-no-alert

Do Beep	Your computer beeps once when the alert displays.	
Alert Text [ ]	Enter your alert question text.	

  Run-time dialog

Elapsed Time	A time counter for the alert.	
Yes	‘Yes’ button	(value = 1)
No	‘No’ button	(value = 0)
Stop Paradigm	Manually abort the Paradigm.	

ElseIf

Allow conditional Paradigm flow control between multiple choices.

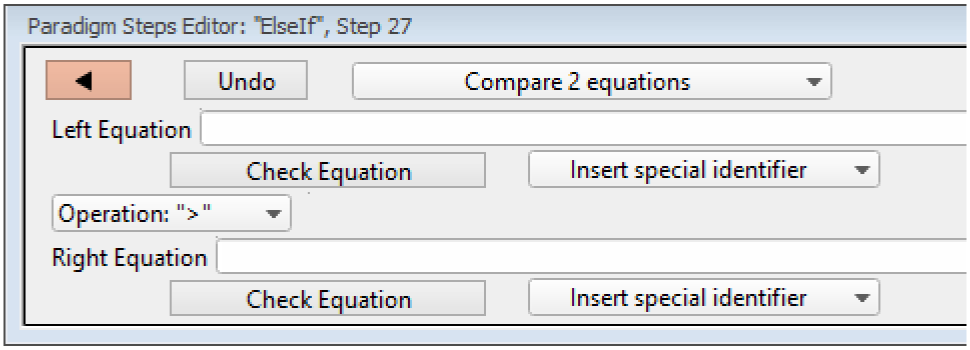


Figure 4-34. Step: Else If

Default Setting: *ElseIf, Left=, Operation=’>’, Right=*

 Close the ‘Paradigm Steps Editor’.

Undo — Remove any unsaved edits to this step.

[drop-down list] — Operation selection.

- Compare 2 equations

  Left Equation — Evaluated to a value.

  Check Equation — Check the equation syntax (for sweep #1). The equation is evaluated, and if valid, it reports "Syntax is ok.”

  Insert special identifier — Acquisition, amplifier and reference settings are available for use in equations. (See list below.)

  Operation — Comparison operators.

>	Greater than
>=	Greater than or equal to
=	Equal to
!=	Not equal to
<=	Less than or equal to
<	Less than

Note: Be careful when comparing two floating-point numbers for equality, as minor variations in resolution can affect the outcome.

Right Equation	Evaluated to a value.
Check Equation	Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is ok."
Insert special identifier	Acquisition, amplifier and reference settings are available for use in equations. (See list below.)

- Check for key pressed — "Last key" typed on the keyboard.

  The "Last key" field is cleared at the start of a Paradigm.

  Key to check for — Enter a text key, or insert a "special" key.

  Insert special key — Use a "non-text" key.
  - Space
  - Return
  - Esc

- Check checkbox status — Select a checkbox to monitor for "on / off" status.

  Checkboxes are displayed at the bottom of the Paradigm Editor window, and are cleared at the start of a Paradigm.

  Checkbox

[1 – 3]	Paradigm-level "local" checkboxes ..
[4 – 6]	Experiment-level "global" checkboxes persist across Paradigms for the entire Experiment.

- Answer of yes-no alert

Do Beep	Your computer beeps once when the alert displays.
Alert Text [ ]	Enter your alert question text.

Run-time dialog

Elapsed Time	A timer of how long the Alert has been displayed.	
Yes	'Yes' button	(value = 1)
No	'No' button	(value = 0)
Stop Paradigm	Manually abort the Paradigm.	

Else

This step allows Paradigm flow control to continue to the next step if the previous condition fails.

Default Setting: *Else*

Checkboxes

Checkboxes are useful for quick conditional control of Paradigm steps. They are visible at the bottom of the Paradigm Editor window.

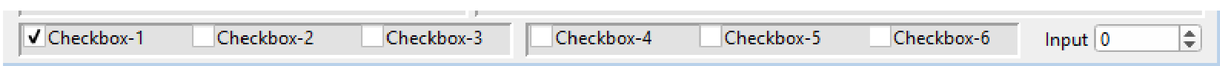


Figure 4-35. Checkboxes

[1 – 3] These "local" checkboxes are cleared when a Paradigm starts. They provide Paradigm-specific controls that are only valid for the current Paradigm session.

[4 – 6] These "global" checkboxes are cleared when an Experiment starts. They can be used across all Paradigm Pools for the entire Experiment.

Input

Routine and paradigm variables can be set to this value. Manually enter a value, or set via the paradigm step 'Set Variable'.

[-1.00 – 1.00] This value is restricted to ±1.00 to enable scrolling through a defined range of values. It can be rescaled when used in an equation.

Paradigm Variables Pane The Paradigm Variables table displays at the bottom of the Paradigm Editor.

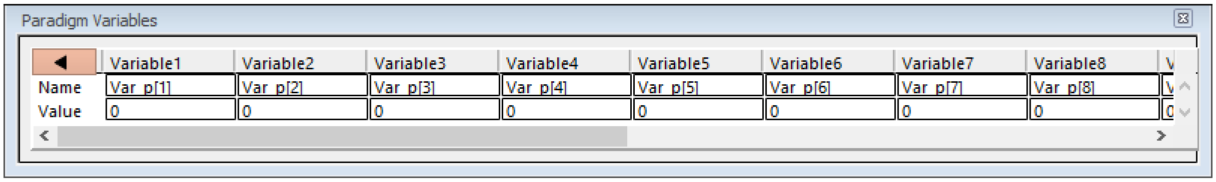


Figure 4-36. Paradigm Variables

These variable can be used in any equation, or in the paradigm step Execute, and persist across experiments. The table can be directly edited during non-acquisition, or set via the paradigm step Set Variable.

	'Close' button	Closes the Variables table.
	Variable[1 – 16]	16 columns of Paradigm Variables.
Name:	Var_p[1 – 16]	Paradigm Variable names can be edited to any text.
	Note:	These names are for display only, and are notr supported in equations.
Value:	[ ]	Numeric values can be manually entered, or programmatically set via the paradigm step 'Set Variable'.

<u>Special identifiers</u> The following acquisition, amplifier and reference settings are available for use in equations.

<u>Timing</u>

Time	(present date-time, s)
Timer	(timer time, s)
ParadigmTime	(time at start of paradigm, s)
RoutineTime	(time at start of routine, s)

Paradigm Parameters

Loop — (active paradigm ForLoop count)

Sweep — (active paradigm EachSweep count)

Sweep count of the active sweep in the Scope window.

LastSweep — (total number of sweeps in active Routine)

During acquisition, this is set according to the routine parameters. Once acquisition terminates, this is replaced by the count of the last acquired sweep, i.e., the last sweep in the stored series.

Processing can occur before or after the last sweep of a series.

Example: Use in a 'ForEachSweep' loop Routine, to compare an 'If' step equation to the sweep number.

```
ForEachSweep
   EachSweep, Target=IV
   If, Left=sweep, Operation= '=', Right=LastSweep- 1
     Alert, Text=LAST SWEEP, DoBeep=true
   EndIf
ForEachEnd
```

AqStopped — (last acquisition was stopped)

1= the last acquisition was stopped
0 = the last acquisition completed

Stimulant — (last applied stimulant concentration)

From the Solution Editor 'Concentration' setting, for a solution configured as a 'Chemical Stimulant'.

Input — (Input variable on paradigm window)

Hold[1..4] — (holding of n'th output channel)

p[1..16] — (n'th paradigm variable)

r[1..16] — (n'th routine stimulus variable)

Analysis Results

m[1..16] (n’th analysis measurement value)

gx[1..16] (n’th analysis graph x value)

The X-value of the last data point in the latest version of gaph[#]

gy[1..16] (n’th analysis graph y value)

The Y-value of the last data point in the latest version of graph[#]

Signal Readings

AuxIN[1..4] (auxiliary input, V)

A single-point reading, such as from a slowly changing temperature probe

Imon (amplifier current reading, A)

Vmon (amplifier voltage reading, V)

Mean[name or count,start,width]
(mean of given input signal)

‘name’ = signal name

‘count’ = window-signal position

‘start’ = time of start, s (of measurement range)

‘width’ = duration, s (of measurement range)

Headstage

ActiveProbe (active probe)

[1 – 4]

The “active” probe number is the Sutter headstage presently controlled by the Amplifier Control Panel.

For a single headstage system, the active probe is always headstage number "1".

NumProbes (number of probes)

The number of IPA headstages attached to the system.

IPA Settings

CCMode	(amplifier current clamp)
VCMode	(amplifier voltage clamp)
Hold	(IHold in CC-mode, VHold in VC-mode) [in Routines] [±0.000,020 A (±20,000 pA), or ±1.000 V (±1000 mV)]
IHold	(amplifer holding current, A) [± 0.000,020 A (±20 nA)]
IHoldOn	(amplifier holding current On)
VHold	(amplifier holding voltage, V) [±1.000 V (±1000 mV)]
VHoldOn	(amplifier holding voltage On)
IGain	(amplifier current gain, V/A) Read the gain of the active voltage-clamp 'Current' input channel.
VGain	(amplifier voltage gain, V/V) V/V evaluates to mV/mV. Read the gain of the active current-clamp 'Voltage' input channel.
Filter	(amplifier input filter in VC- and CC-mode, Hz) Read the low-pass filter of the input channels.
IFilter	(amplifier input filter in VC-mode, Hz) Read the low-pass filter of the 'Current' input channels.
VFilter	(amplifier input filter in CC-mode, Hz) Read the low-pass filter of the 'Voltage' input

	channels.
Offset	(amplifier pipette offset in VC-mode, V)
OffsetLock	(amplifier pipette offset lock On in VC-mode)

IPA Compensation

ECompMag	(amplifier electrode compensation magnitude, F)
ECompTau	(amplifier electrode compensation tau, s)
ECompOn	(amplifier electrode compensation On in CC-mode)
CmComp	(amplifier cell compensation Cm, F)
RsComp	(amplifier cell compensation Rs, Ohm)
RsCompOn	(amplifier cell compensation Rs On)
Bridge	(amplifier bridge balance, Ohm)
BridgeOn	(amplifier bridge balance On)

IPA Correction

RsCorr	(amplifier Rs correction, fraction)
RsPred	(amplifier Rs prediction, fraction)
RsLag	(amplifier Rs correction lag, s)
RsCorrOn	(amplifier Rs correction On)

Membrane Test

Relectr[1..2]	(electrode/seal/access resistance, Ohm)
	Value from last Membrane Test.
Rmemb[1..2]	(membrane resistance (cell mode), Ohm)
	Value from last Membrane Test.
Cmemb[1..2]	(membrane capacitance (cell mode), F)
	Value from last Membrane Test.
RMSNoise[1..2]	(membrane test RMS noise, A)

Value from last Membrane Test.

Lock-In

LockInPhaseAdj	(Lock-In phase delay adjustment, s)
LockInAttenAdj	(Lock-In attenuation adjustment)

4.1.7 Reanalysis Measurements & Graphs

Make real-time changes to the offline measurements and graphs with this dialog.

To access this dialog, click on the Scope (Analysis) window button 'Measurements / Edit Measurements'.

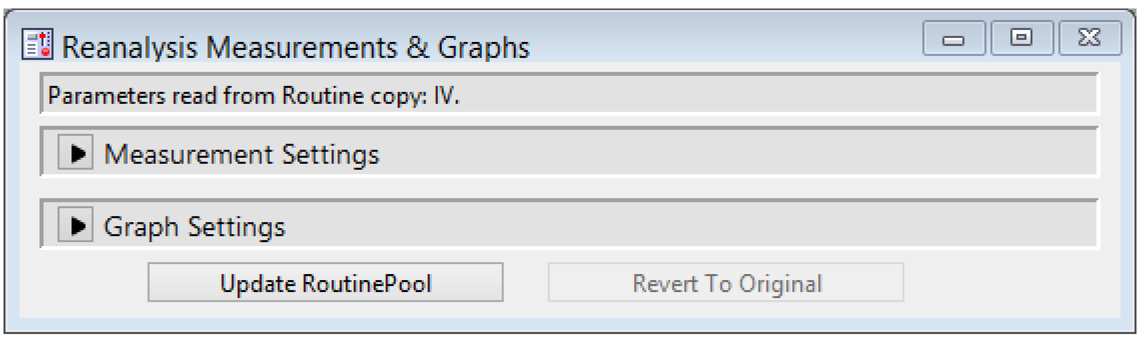


Figure 4-37. Reanalysis Measurements & Graphs

This dialog is the same as in the Routine Editor / Real Time Measurements & Graphs dialog, with an extra field:

Measurement parameters from: The Routine name and date/time stamp.

4.1.8 Routine Editor

Routines contain the settings that are in effect during data acquisition. The Routine Editor allows you to define acquisition parameters, set input and output channels, and to create stimulus waveforms and online analyses. The Routine Editor is the central place to create and manage saved Routine Pools and data acquisition settings.

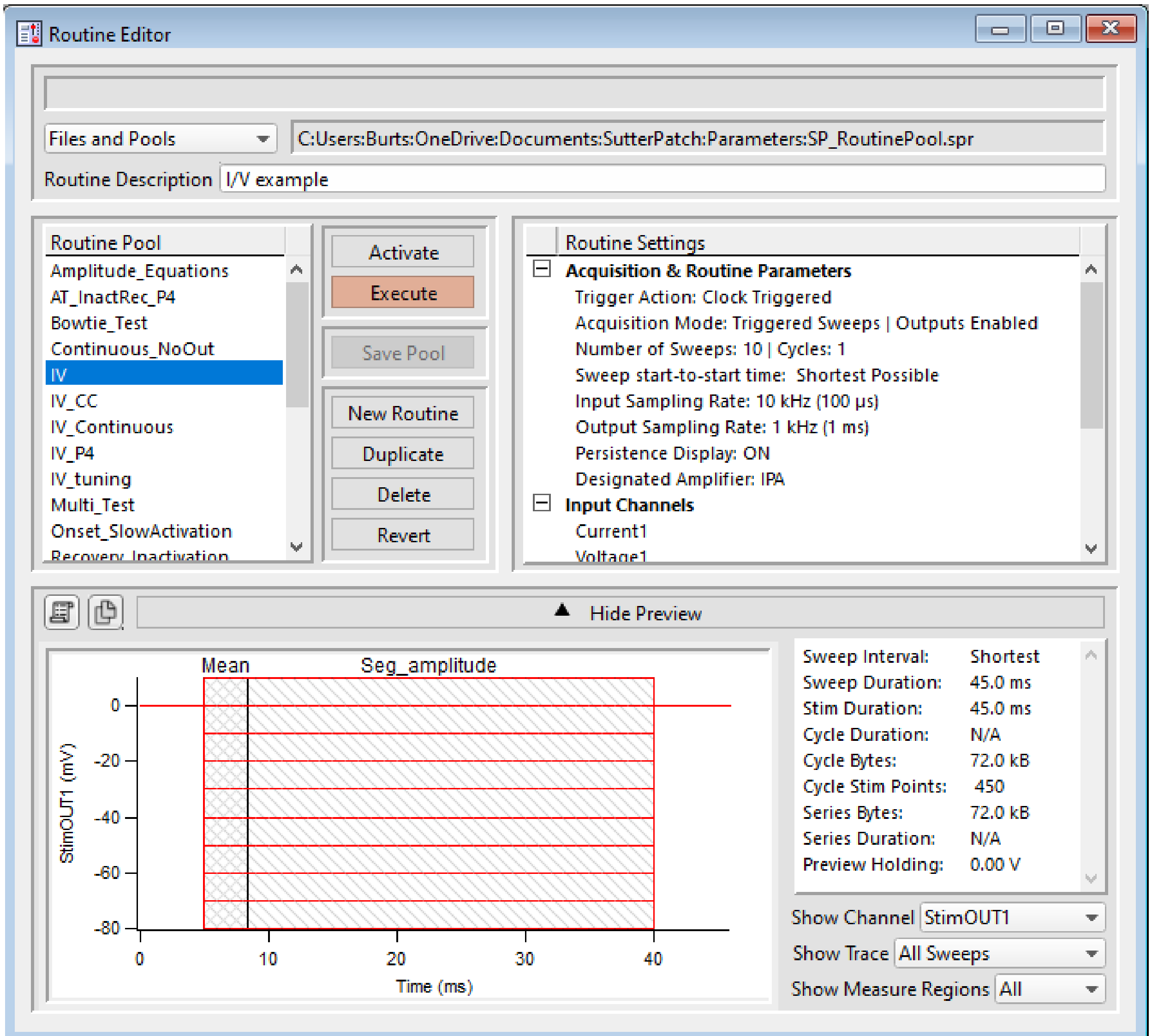


Figure 4-38. Routine Editor

The Routine Editor is structured to hold one or more Routines within its Routine Pool. The Routine Pool thus provides easy access to the set of Routines used in an experiment.

Tip: SutterPatch comes with a Sample Routine Pool that contains a collection of frequently used experimental scenarios. Rather than creating a new Routine, it might be easier to Duplicate a sample Routine and modify it until it meets your particular needs.

<u>Status Field</u>	Notifications on edits and Routine names are displayed here.	
<u>Files and Pools</u>	[drop-down list …]	
	Most recently used list of the last 5 Routine Pool files	
	Load Routine Pool	Load the Routines of a previously saved Routine Pool file into the Routine Pool
	New Default Routine Pool	Create a new blank Routine Pool.
	New Routine Pool	Create a new Routine Pool either with a blank default Routine, or populated with Routines from the currently loaded Routine Pool.
	Get Default Sample Routine Pool	Load the Routines from the default Dendrite sample Routine Pool file SP_RoutinePool.spr.
	Revert to Last Saved	Undo any unsaved changes to the Routine Pool
	Save Routine Pool	Save the Routine Pool using its existing file name and path.
	Save Routine Pool As	Save the Routine Pool to a new file, and switch to the new file. The default file name has an increment number appended to the original file name.
	Save Routine Pool Copy	Save the Routine Pool to a new file, but do not switch to the new file. The default file name has 'Copy of' prepended to the original file name.
	Merge Routine Pools	Insert the Routines from a previously saved Routine Pool file into the loaded Routine Pool.
	Merge PatchMaster PGF File	Insert the "Routines" (PGF Sequences) from a PatchMaster PGF file into the loaded Routine Pool.
	Convert Routine Pool	Convert the loaded Routines to be compatible with the attached amplifier or emulation mode. The original file is overwritten.
	Send Last Used List to History	Copy the pathname of the last used Routine and paste it into the Command window history.

Table 4-1. Routine Files and Pools

New Routine Pool dialog:

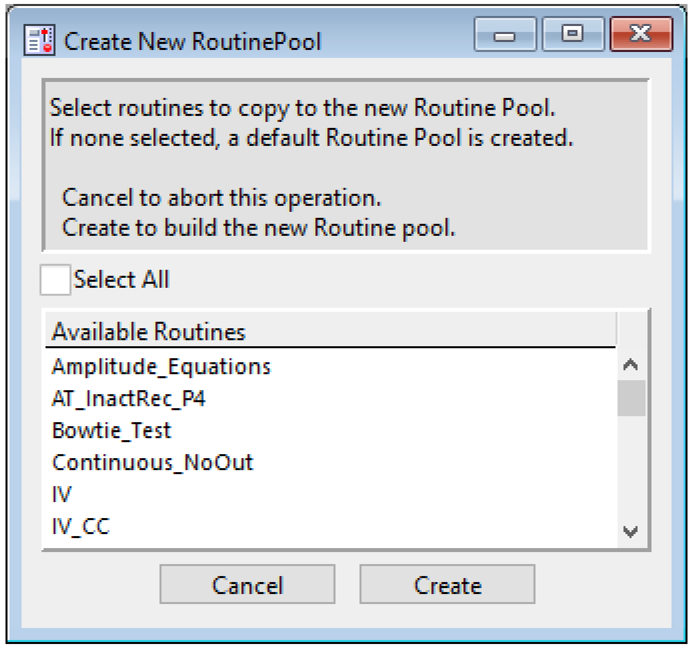


Figure 4-39. New Routine Pool

Create a new Routine Pool populated with a "blank" Routine, or select Routines from the loaded pool to populate the new pool.

[File Path field]

The file path of the current Routine Pool displays on the right of the 'Files and Pools' list. If the Routine Pool has not been previously saved, this field is blank.

Routine Description

A Routine Description text comment can be edited and saved with the Routine.

Routine Pool

The Routine Pool section lists the names of all currently loaded Routines. Selecting a Routine name loads it into the Routine Settings section for editing and activation. As the Routine Pool contents are held in memory, the switching times between Routines are very fast.

Double click a Routine name to edit it.

- Allowable characters are A-Z, a-z, 0-9, and "_".
- Special characters and spaces are not allowed.
- The maximum length of a Routine name is 22 characters.

To select multiple Routines, use a Shift-click mouse drag, or individually Shift-click the Routine names. Multiple Routines can thus be deleted, or saved to a new Routine Pool.

Note: When a Routine is selected, if it was created for a different system (i.e., IPA vs. Double IPA), and the Preferences / Hardware / Routine Conversion is set to 'Prompt user', then a Routine Conversion dialog displays.

Activate	Open or refresh the Scope window with the latest Routine settings, but do not start acquisition. This button is renamed to "In Progress" during a recording.
Execute	Open or refresh the Scope window and immediately start recording. The latest Routine settings are applied to the Scope window. This button is renamed to "Convert" if a routine designed for a different amplifier type is selected.
Save Pool	Save the Routine Pool using its existing file name.
New Routine	Add a blank Routine to the Routine Pool, and open it for editing. The default Routine name is "untitled" with an increment number appended.
Duplicate	Add a copy of the selected Routine to the Routine Pool. The Routine name number is appended or incremented.
Delete	Remove the selected Routine from the Routine Pool.
Revert	Discard any unsaved changes to the selected Routine.

Table 4-2. Routine Editor Buttons

Waveform Preview

The stimulus waveform is graphically displayed at the bottom of the Routine Editor.

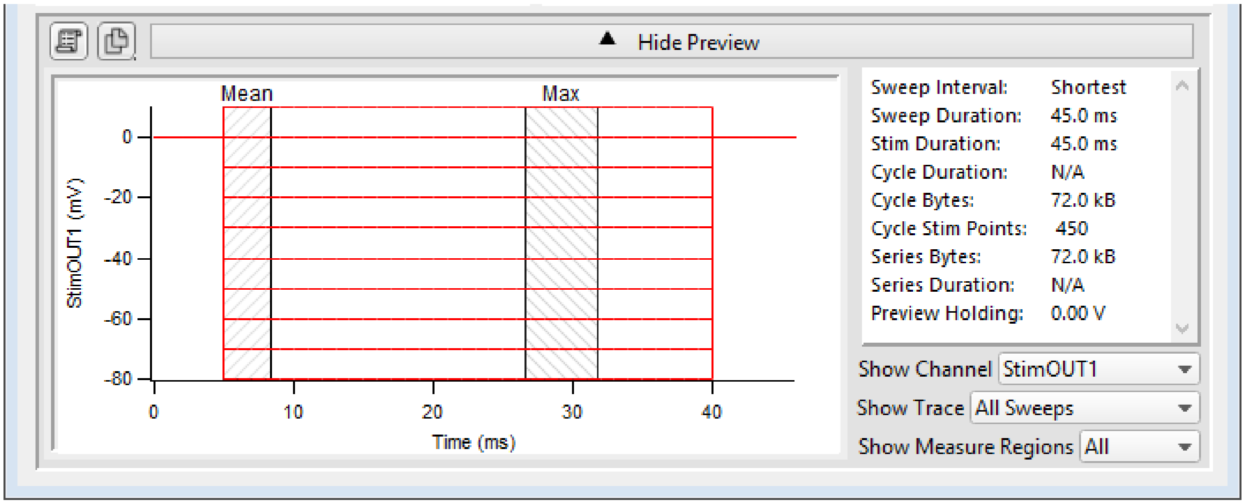


Figure 4-40. Waveform Preview Pane

The waveform preview and its settings are updated live to reflect changes in the Waveform Editor and Amplifier Control Panel.

Measurement regions can be manually repositioned in the Preview pane. Click and hold a measurement region to highlight it in black, then drag it to a new position, and release. This also updates its Measurement Settings / Region Timing 'Start/End Time' settings.

To change the region's duration, click-drag the region's right-edge cursor; its Region Timing 'End Time' setting is updated, while its 'Start Time' is unchanged.

The preview for the digital output 'DigOUTWord' sets its Y-axis to ' Digital State (Word)', and displays the decimal value of the selected bits.

Note: A "Cityscape" display mode is used, i.e. plotting with straight horizontal and vertical lines connecting the preview sample points (vs. smooth interpolated transitions).

	Copy to Layout	Copy the stimulus graph into a new Layout window, or append to an existing Layout page.
	Copy to Clipboard	Copy the stimulus graph to the system clipboard.
Show / Hide Preview		Expand or collapse the Preview pane.

X- and Y-axis Control

- Hover the mouse cursor over an axis line until the cursor turns into a double-headed arrow, then scroll up or down to contract/expand the axis.
- In the preview pane, click and drag the mouse cursor to surround a region of interest with a bounding box (the "marquee"). Right-click in the box and select one of the expand/shrink options.

Some key settings and display controls are listed on the right of the Preview pane.

Units are in ‘s’, or if < 1 s, then in ‘ms’.

Sweep Interval:	The interval of time between the starts of consecutive triggered sweeps (Sweep Start-to-Start Time) in the active Routine. When set to ‘Shortest’, this equals the longest Sweep Duration + 200 ms.
Sweep Duration:	The amount of time in a sweep during which signal recording occurs with the active Routine. • Outputs enabled The sweep duration is based upon the longest stimulus waveform duration set in Output Channels & Waveform / Waveform Editor. • Outputs disabled The sweep duration is based upon the longest duration set in Input Channels / Edit Signals / Waveform Editor.
Stim Duration:	The maximum amount of time during which output stimulation occurs in a sweep. Set in Output Channels & Waveform / Waveform Editor / Duration.
Cycle Duration:	The amount of time for a cycle. Set in Acquisition & Routine Parameters.
Cycle Bytes:	The number of bytes of data in a cycle.
Cycle Stim Points:	The number of points in which output stimulation occurs in a cycle.
Series Bytes:	The number of bytes of data in the Series.
Series Duration:	The amount of time for the Series.
Preview Holding:	The holding level in the Amplifier Control Panel.
Show Channel:	A list of output channels to preview.
Show Trace:	Select how to display autoscaled sweep traces in the preview pane.

- Time Course — Display all traces in continuous linear time.
- All Sweeps — Display all traces overlaid from time zero.

  (not available for 'Show Channel: All Channels')
- Sweep # — Display a trace from a single sweep.

Show Measure Regions: A list of measurement regions to preview.

- None — No regions displayed.
- All — All regions displayed.
- m[#] — Select a single region to display.

Routine Settings

The Routine Settings are split into 5 main sections. Click on a section header or item to open its sub-window.

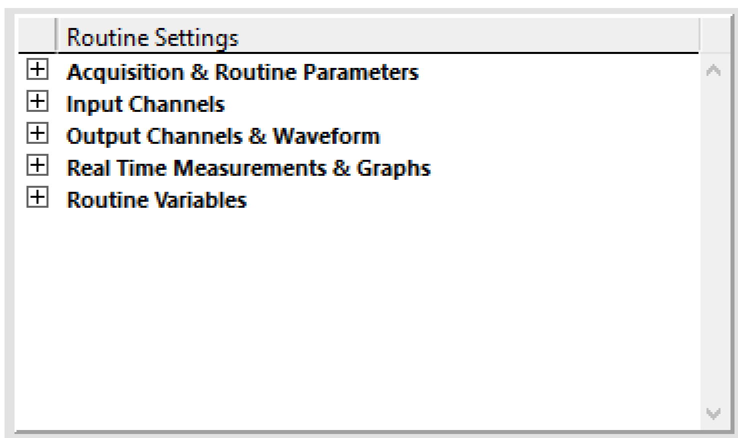


Figure 4-41. Routine Settings

Routine Editor: Acquisition & Routine Parameters

Acquisition timing parameters are controlled in this section, such as sweep duration and sampling rates. The settings in this section are shared by all input and output channels.

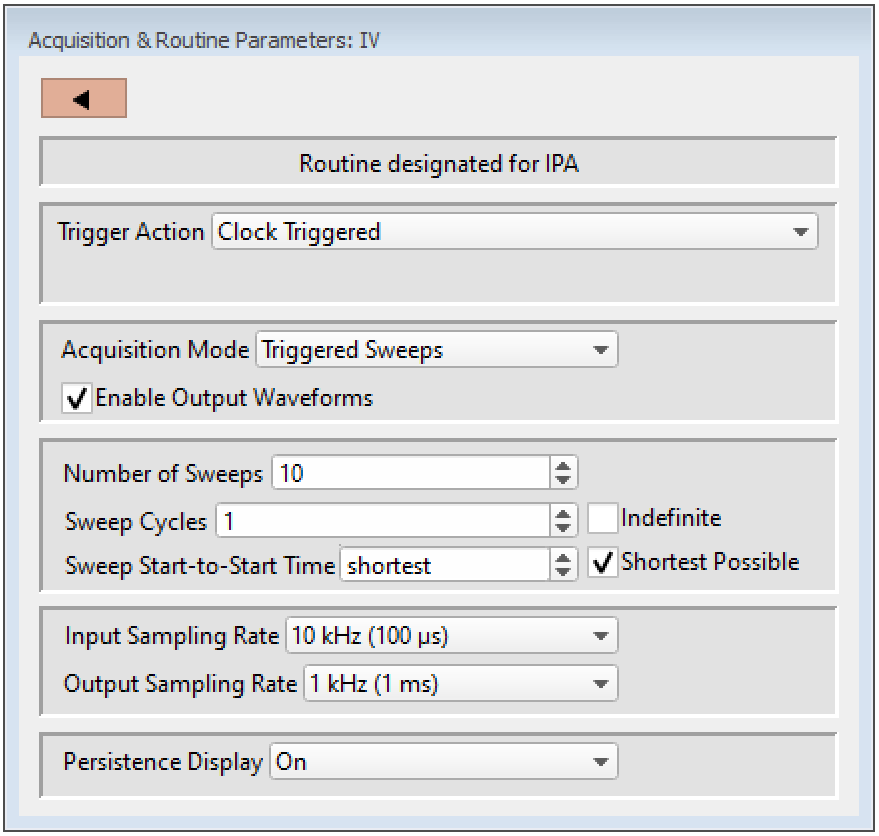


Figure 4-42. Acquisition & Routine Parameters

Trigger Action

Control how and when recordings occur.

- Clock Triggered: Start a recording timed by the SutterPatch program. Hardware trigger inputs are ignored.
- Externally Triggered Sweep:

  Use an external signal from other laboratory equipment to trigger the recording of each sweep in a Series.

  Connect the external trigger to the 'Trigger In' BNC on the rear panel of the IPA amplifier.

  However, if a Routine is run within a Paradigm 'Each Sweep' step, then the hardware trigger is substituted by a software trigger generated by the Paradigm.

- Externally Triggered Series:

  Use an external signal from other laboratory equipment to trigger the start of a Series, then operate like Clock Triggered. A command waveform is only generated for the first cycle.

  Connect the trigger signal to the 'Trigger In' BNC on the rear panel of the IPA amplifier.

  However, if a Routine is run within a Paradigm 'Each Sweep' step, then the hardware trigger is substituted by a software trigger generated by the Paradigm.

- Event Triggered: Use an amplitude event in an input signal to trigger data acquisition for a set length of time.

  This functionality is useful to reduce extraneous data when infrequent events occur during long recordings, or to align random events within sweeps.

  The Acquisition Mode is set to 'Continuous Sweeps', to continuously monitor the signal for a trigger event.

  Event Triggered Settings: Click on the 'Event Triggered' field to open this dialog.

Input Channel To Scan	Trigger on this channel. The unit of the input channel is used for the trigger threshold.	
Pre-Trigger Duration	[0 – 0.06 s]	
Trigger Threshold	[±20.000 nA]	current input
	[±0.75 V]	voltage input
Trigger Polarity	Rising Edge	
	Trailing Edge	
Minimum Trigger Duration	[0.0001 – 0.045 s]	

Acquisition Mode

- Triggered Sweeps: Each sweep is started by a software trigger from a Routine or Paradigm, or by an external hardware trigger.

To allow for system delays, there is a short gap (~200 ms) between sweeps. The resolution of the Sweep Start-to-Start time is 1 ms.

- Continuous Sweeps: Uninterrupted data without time gaps between sweeps are recorded when the ‘Sweep Start-to-Start Time’ is set to the ‘Sweep Duration’.

  Data are displayed as sweeps, not as a continuous “rolling” display.

  This option does not support:

  - Pausing of sweeps during recording.
  - Paradigm step ‘For Each Sweep’.
  - Very short sweeps.

  Note: The IPA demo mode display of continuous sweeps includes artificial gaps between them. Recording with hardware attached does not have any gaps

Enable Output Waveforms

Output channel waveforms can be optionally disabled.

When outputs are disabled, you can control the sweep and segment durations (for analysis measurements) via the Input Channels ‘Edit Signal’ button.

Number of Sweeps [1 – 65000]

The number of sweeps to record.

Sweep Cycles [0, 1 – 65000]

The number of times to automatically repeat the entire set of sweeps recorded by a single Series.

A value of zero is replaced by “indefinite”, and the Indefinite checkbox is automatically enabled.

Indefinite The number of Sweep Cycles is undefined - sweeps continue to record until acquisition is stopped.

Indef Post Stim [for 'Continuous Sweeps' with Output Waveforms.]

After the designated number of Sweep Cycles is reached, the Output Waveform is disabled while Sweeps continue to record until acquisition is manually stopped.

The largest file size that SutterPatch can record is 2.5 G samples.

Sweep Start-to-Start Time ['s', or if < 1 s, then in 'ms']

The time from the start of recording a sweep to the start of the next sweep.

Shortest Possible [for Triggered Sweeps]

Accommodates the longest waveform duration in the Series + overhead processing time (100 ms).

Sweep Duration [for Continuous Sweeps]

The sweep duration is the longest waveform duration in the Series.

[Outputs enabled] As set in Output Channels & Waveform / Waveform Editor.

[Outputs disabled] As set in Input Channels / Edit Signals / Waveform Editor.

Note: In demo mode, the sweep start-to-start times can vary during acquisition, especially on slower computers.

Input Sampling Rate: Applies to all input channels.

Sampling Rate	Sample Interval
100 Hz	(10 ms)
200 Hz	(5 ms)
400 Hz	(2.5 ms)
800 Hz	(2 ms)
1 kHz	(1 ms)
2 kHz	(500 µs)
4 kHz	(250 µs)
5 kHz	(200 µs)

8 kHz	(125 µs)
10 kHz	(100 µs)
20 kHz	(50 µs)
25 kHz	(40 µs)
40 kHz	(25 µs)
50 kHz	(20 µs)
100 kHz	(10 µs)
200 kHz	(5 µs)

According to the Nyquist sampling theorem, the input *sampling* rate should oversample the input *filter* rate (in the Amplifier Control Panel) at a minimum of 2x,

Also, if the input filter rate is greater than the input sampling rate, the filter rate is ignored.

Note: IPA amplifiers record both stimulus and response signals via physical analog channels, so all recorded signals are precisely in sync, with no timing delays between them.

Output Sampling Rate: Applies to all output channels.

Sampling Rate	Sample Interval
100 Hz	(10 ms)
200 Hz	(5 ms)
400 Hz	(2.5 ms)
800 Hz	(2 ms)
1 kHz	(1 ms)
2 kHz	(500 µs)
4 kHz	(250 µs)
5 kHz	(200 µs)
8 kHz	(125 µs)
10 kHz	(100 µs)

20 kHz	(50 µs)
25 kHz	(40 µs)
40 kHz	(25 µs)
50 kHz	(20 µs)
100 kHz	(10 µs)

Note: New Routines use a 1 kHz default output channel sampling rate, as command waveforms usually do not require high-resolution time changes - increase the sampling rate as needed for more complex waveforms.

Persistence Display

For a recording, display each new sweep without erasing any previous sweeps.

- Off — Set the Scope window into non-persistence display mode.
- On — Set the Scope window into persistence display mode.
- Keep current setting — Do not change the Scope window's prior setting.

Routine Editor: Input Channels

Configure the input channels.

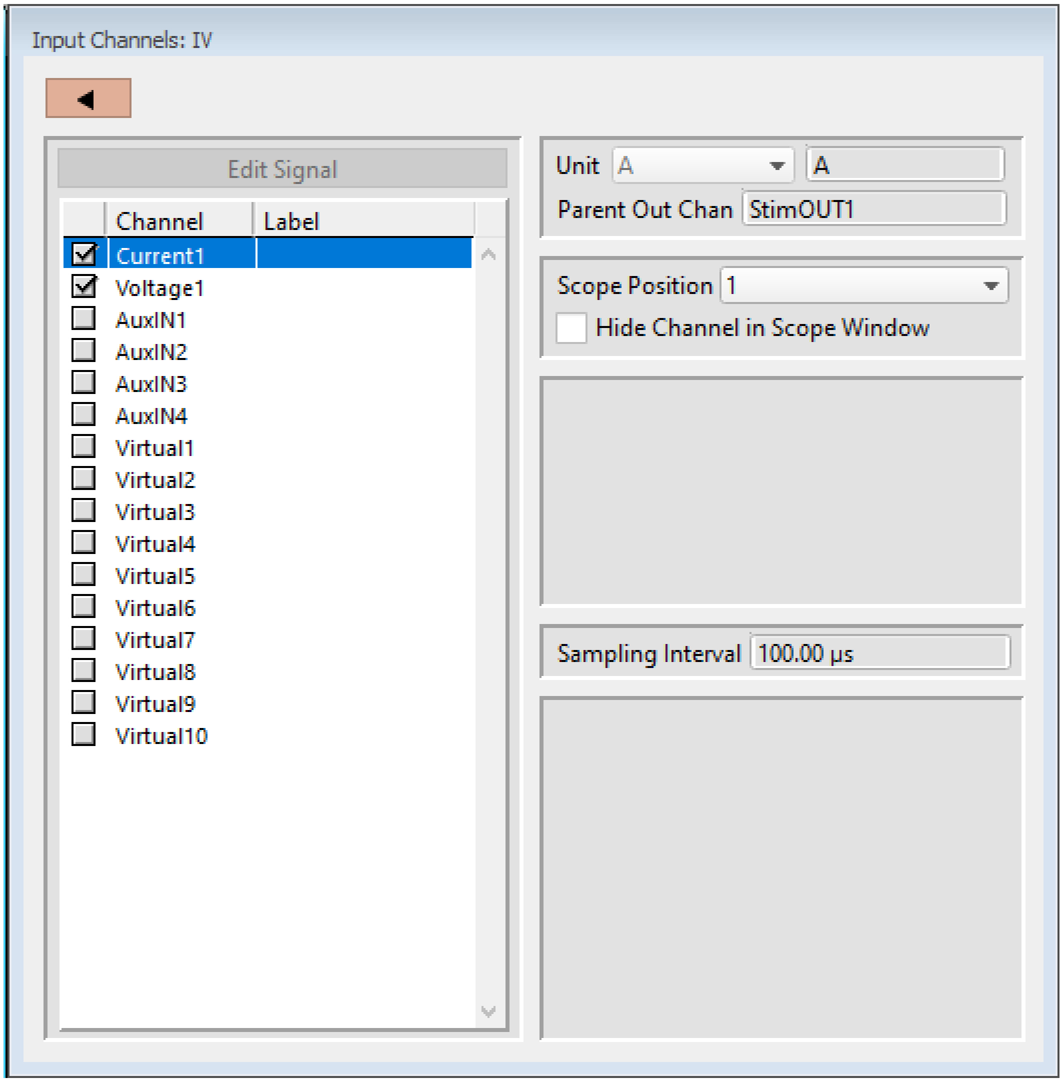


Figure 4-43. Input Channels

Edit Signal [for disabled Output Waveforms]

When Output Waveforms are disabled in the Acquisition Parameters section, segment timing can still be controlled via a modified Waveform Editor. This is a "timing only" version of the Waveform Editor, which controls the duration of segments to facilitate segment-based measurements.

Channel

Enable up to 16 Input Channels for recording data:

Current#	Analog input current channels hardwired from the IPA headstage.
Voltage#	Analog input voltage channels hardwired from the IPA headstage.
AuxIN[1 – 4]	Four auxiliary analog input channels allow you to directly digitize and record input signals from connected non-Sutter external equipment.
Note:	In Emulation mode, the AuxIN channels display a ±20 mV sine wave.
Virtual[1 – 10]	Ten virtual channels are available. Virtual channel data are mathematically transformed data from another input channel, or are entirely computed from an equation.

Label

A user-editable signal name for a channel.

These labels are used in:

- Routine Settings overview for Input and Output Channels
- Parent Out Chan'
- Virtual channel Math Equations and Source Channels
- Scope window signal panes
- Data Navigator Preview pane
- Metadata Input Signal Name

To rename an Input Channel, first enable it, then double-click it and enter the new name. If the label is used by another channel, an underscore and increment number are appended to the new label.

When a virtual input channel is enabled, a default 'Math Type' label is automatically generated for it.

Unit

The base unit of measurement.

• 'Current' Channels	Fixed at 'A' for current.
• 'Voltage' Channels	Fixed at 'V' for voltage.
• 'AuxIN' Channels	Default is 'V', but is editable.

- 'Virtual' Channels — Same as its Source channel units; is only editable for virtual Math Type 'Equation'.

Note: The resolution of the signal units are automatically set.

Parent Out Chan — The "Parent Output Channel" shows which output channel is associated with which input channel. The ouput channel timing is also used for measurements with 'Cursors Relative to Segments'.

Auxiliary Input channels allow you to change the 'Parent Out Chan'.

However, Virtual channels cannot directly select Parent Output channels. Instead, they utilize their Source channel's Parent channels.

Scope Position — The input channel panes can be repositioned in the Scope window.

Hide Channel in Scope Window

The selected input channel is hidden in the Scope window.

Scaling — [for AuxIN and Virtual Input channels]

Equation — Enter a fixed value or equation for the input channel scaling factor of the external signal.

Factor — [read-only field]

The input channel scaling factor (converted from an equation).

Note: The IPA digitizer uses a high-resolution 16-bit ADC with 64-bit data, so data resolution is not an issue when scaling input signals.

Offset (V) — Apply an amplitude offset to the input signal (after any scaling).

Tip: To use 'mV' units, enter: '#m' or '#e-3'

To use 'pA' units, enter: '#p' or '#e-12'

Scaled Offset — [read-only field]

Raw offset values are converted to input units.

Sampling Interval — [read-only field]

The duration of a single data sample.

Virtual Input Channels

Virtual input channels allow you to perform a variety of mathematical transformations on input signals in real time. When a virtual input channel is enabled, its configuration fields are ungrayed.

Math Type — Apply a data transformation to a virtual input channel.

- Baseline Subtract
- Bessel Filter
- Differentiate
- Downsample
- Equation
- Integrate
- Leak
- Line Frequency
- LockIn
- Smooth
- Stimulus
- Sweep Average
- Sweep Subtract

- BaselineSubtract — Subtract a fixed value from all data points in an input trace. This is useful for adjusting for an offset, or resetting a baseline.

  Source Channel — Select an input channel to process.

  Baseline From — Select how to calculate the subtraction value.

  - Value — Subtract a fixed value.

    Value — Spinner adjusts in 1 pA or 1 mV increments.

  - Trace — Subtract the average of the entire input trace.

  - Sweep Time — Subtract the average of the data between the Start Time and End Time.

    Start Time — Set the starting time of the data to be averaged.

    End Time — Set the ending time of the data to be averaged.

- Segment #s — Subtract the average of a Segment.

  Start Ratio — Set the starting time of the data to be averaged, as a ratio relative to the starting time of the Segment duration.

  Start Time — [derived value]

  End Ratio — Set the ending time of the data to be averaged, as a ratio relative to the ending time of the Segment duration.

  End Time — [derived value]

- BesselFilter — A frequency-domain filter with excellent response characteristics for preserving the shape of a biological signal.

  Source Channel — Select an input channel to filter.

  Filter Bandwidth — Select a frequency range.

  - LowPass — Allow signal frequencies less than the cutoff frequency, and block all higher frequencies, such as high frequency noise.

  - HighPass — Allow signal frequencies greater than the cutoff frequency, and block all lower frequencies.

  Filter Order — [1, 2, 4, 8]

  Number of “poles” in the filter. A higher number provides a sharper (more accurate) response, but consumes more processing time and system resources.

  Cutoff Frequency — [0.01 Hz to < ½ the sampling rate]

  Restrict frequencies from this boundary point onwards.

- Downsample — Apply downsampling to an input signal, i.e., reduce the sampling rate of the signal data.

  Source Channel — Select an input channel to downsample.

Source Sampling Interval [read-only field (μs)]

Reduction Factor [2 – 100]

Only whole numbers are used; non-whole numbers are rounded up or down.

Sampling Rate [read-only field (Hz)]

New sampling rate of the reduced signal.

- Differentiate Apply differentiation to an input signal. The instantaneous rate of change in the signal is displayed.

  Source Channel Select an input channel to differentiate.

- Equation Specify an equation to process an input signal.

  Source Channel Select an input channel to process.

  Equation [ ] Click field to access the 'Specify math equation' editor.

  Note: The full equation is always visible as a tool tip, by hovering the mouse cursor over the 'Math Equation' field.

  Specify math equation for virtual signal

  [<equation>] A free-form text field.

  Errors are reported under this field.

  Check Equation Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports that the syntax is "ok".

  Insert special identifier Special references can also be used within commands.

  - p[#] n'th paradigm variable

  - s[series-count, sweep-count, trace-count, routine name]

    Access an arbitrary input trace (data wave) via counts of Series #, Sweep #, Trace # (Scope position), and the Routine name.

    The "current" item is the "active" trace in the Scope window, and has a count value of zero.

If a “count” number is non-zero, it is used as an offset from the current count value of zero. Any fractions in count numbers are truncated to integers.

If the routine name is left blank, the current routine name is used.

Ex: s[0,0,0,]

The current series, current sweep, current trace, of the current routine.

- t[#] n’th input trace

  Access the input trace (data wave) in Scope position “n” for the last sweep of the current Series. This numbering can differ from the Scope Position "n", if signals are re-arranged or hidden.

  Tip: You can duplicate a trace by using this.

Undo All changes in the equation editing session are discarded.

(See the Equation Editor for more details.)

- Integrate Display the integral of the data signal. This is equivalent to the signed area under a curve.

  Source Channel Select an input channel to integrate.

- Leak Remove leakage current from the data signal. This is the small passive current when the cell is in a resting state.

  This feature is only enabled when the Routine includes an output channel with P/N Leak Pulse enabled.

  Source Channel Select an input channel to process.

  Show Leak Displays the accumulated leak currents after the subtracted data in a sweep.

  Leak Zero Segment Identify a segment with no active cellular response to the command signal.

When set to zero, the field is set to 'OFF'. To re-display the numeric spinners, enter a non-zero number into the field.

Note: The mean of the second half of the specified segment is used to compute an averaged leak current, which is then used to correct the P/N leak average. This option reduces the influence of a constant leak-current, which is otherwise included in the leak current of the main signal.

- LineFreq Remove AC line frequency noise (hum) from the data signal.

Source Channel	Select an input channel to process.	
Line Frequency	50 Hz	Most of rest of world.
	60 Hz	Canada, (Carribean), Central America, (Japan), Mexico, (South America), South Korea, Taiwan, USA.

Alternating current (AC) power contains 50 or 60 Hz oscillations that can cause sinusoidal line-frequency noise in recorded signals. This FFT-based filter reduces such noise by > 90% over 6 harmonics. The adjusted signal is displayed in real time.

- LockIn Measure cell characteristics (such as membrane capacitance) with high signal-to-noise sensitivity, using a dual-phase software lock-in amplifier.

Note: This feature is only enabled when the Routine includes an output channel with a waveform set to 'Sine / Sine Wave Cycles / For LockIn'.

Calculations are made using 'conductance' instead of 'resistance'.

Current Channel — Select a (source) input channel with a "current: signal.

Trace Kind — Select the LockIn measurement to display.

The selected 'Trace Kind' is automatically set as the Virtual Channel label.

CM — Computed membrane

	capacitance.
GM	Computed membrane conductance.
GS	Computed series conductance.
DC	DC component of measured signal.
RealY	Real number part of the lock-in response signal.
ImagY	Imaginary number part of the lock-in response signal.

Cycles to Average [1 – 1000]

Cycles to Skip [1 – 1000]

V-reversal [±1000 mV]

When using a calculated stimulus trace, enter the reversal potential for the ion under study, such as for (Na+) sodium spikes or (K+) potassium tail currents.

Note: See the SutterPatch Algorithms Appendix for the math used in the LockIn computation.

- Smooth Smooth the data with a "moving average" noise-reduction filter.

  Source Channel Select an input channel to smooth.

  Smoothing Type:

  - Gausian A standard filter with excellent 10 – 90% rise-time response.

    Smooth Operations [1 – 32767]

    # of smoothing operations to perform.

  - Boxcar A fast time-domain filter with excellent 0 – 100% rise-time response.

    Smooth Repetitions [1 – 32767]

    # of smoothing repetitions to perform.

    Boxcar Window Points [1 – 99]

    # of points in boxcar sliding window.

Note: For best performance, only odd values are used.

- Stimulus Replicate the command Waveform.

  Source Channel — Select an input channel – the waveform from its 'Parent Out Chan' is used.

- SweepAverage Average the input traces.

  Source Channel — Select an input channel to average.

  Average Type — Cumulative: Average all processed sweeps together

  Run Average: Average the last "N" sweeps

  Number of Sweeps

- SweepSubtract Subtract a sweep from the input trace.

  Source Channel — Select an input channel to process.

  Source Channel — Select an input channel to process.

  Reference Sweep — Select a sweep to be subtracted from all other sweeps. If the sweep does not yet exist, no subtraction occurs.

Routine Editor: Output Channels & Waveform

Configure the output channels and command waveform.

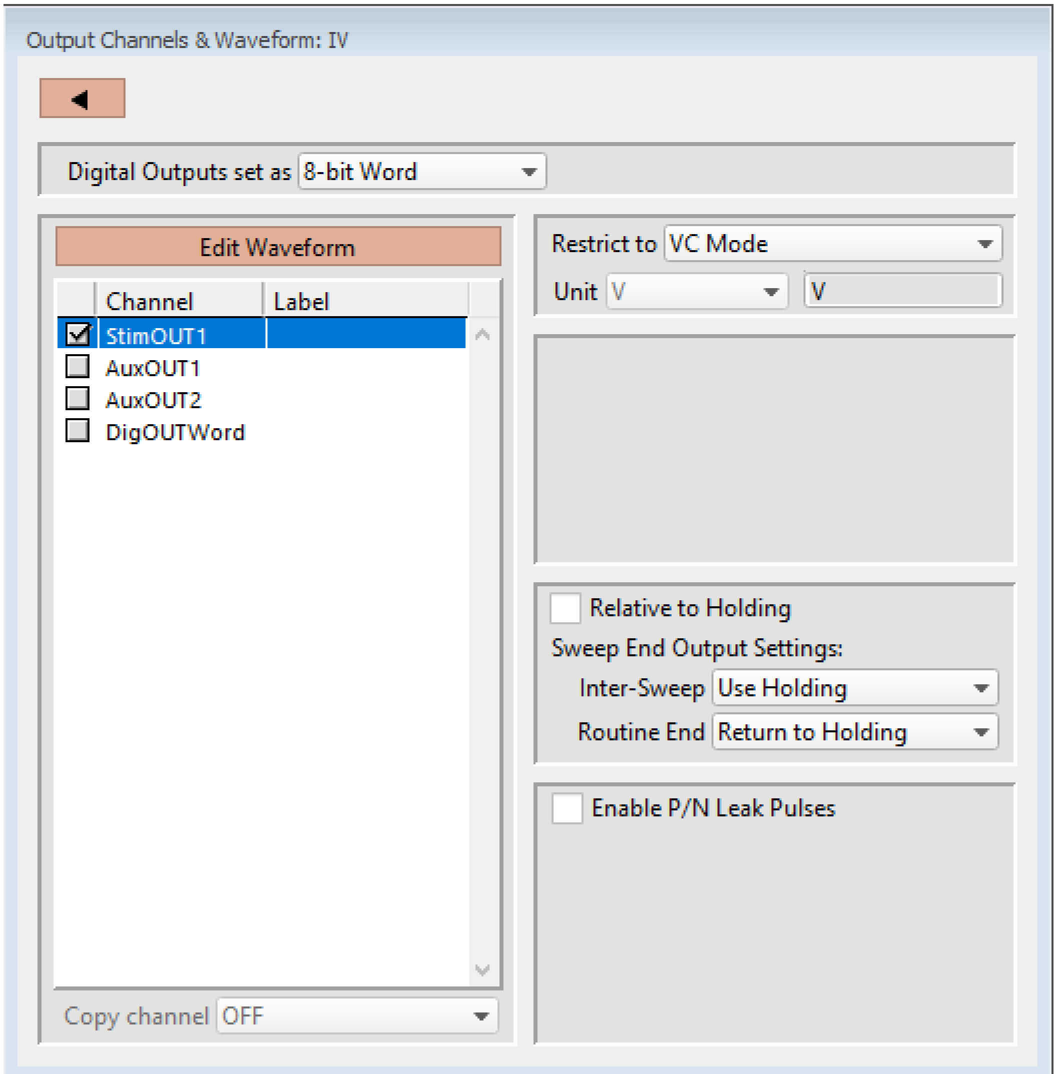


Figure 4-44. Output Channels & Waveform

[HW Status field] Hardware information on the selected channel is displayed.

Digital Outputs set as

Eight digital output channels (bits) can be set individually or as a group.

- Individual bits — Each DigOUT(1 – 8) bit is individually set in its own Waveform Editor table.

  The waveform preview uses the bit's binary word value for its Y-axis value, i.e. if bit 3 is 'HIGH', it has a "word" value of 4.

- 8-bit Word — The 8-bit digital output pattern is controlled by a single decimal number (0 - 250), which is also the waveform preview amplitude value.

  The waveform preview uses the binary bit pattern word value for its Y-axis value, i.e., if bits '1' and '3' are 'HIGH', the 8-bit word has a value of 5.

Edit Waveform

Click the Edit Waveform button to access the Waveform Editor table and create a stimulus waveform. Or alternatively, double-click on an Output Channel name. (See the Waveform Editor section below.)

Channel

Click on the Output Channel checkboxes to enable analog and digital output channels in the Routine. Click on a channel name to highlight and select it – the channel output parameters are displayed for configuration.

The default StimOUT channnels are hardwired to the IPA headstage.

The AuxOUT auxiliary analog output channels) can be used to send stimulus waveforms to external instruments.

The digital outputs are available as either a single 8-bit "word", or as 8 individual 1-bit channels, as set in the Acquisition & Parameters section.

To rename any of the Output Channels, double-click on the respective name and enter a new one.

Tip: If a signal is connected to the front panel 'COMMAND IN' BNC, that signal is summed with the StimOUT waveform that is sent to the headstage.

Note: For StimOUT channels, the actual DAC output signal is passed through a 20 kHz low-pass filter before entering the headstage.

Label

A user-defined signal name for the channel.

These are used in:

- 'Copy Channel'
- Waveform Preview pane 'Show Channel'
- Metadata: Output Signal Name

To rename an Output Channel, first enable it, then double-click it and enter the new name. If the same label is reused for another channel, an underscore and increment number will be appended to the new label.

Copy channel

Copies one channel's waveform to another channel, for output channels of the same type (i.e., "Stim", "Aux", or Digital). If a channel is enabled, then highlighting another or blank channel of the same type ungrays the 'Copy channel' field, and changes it from 'OFF' to 'None', with a drop-down list of available channels to copy from.

Restrict to

Ensures that the matching headstage is in the proper VC/CC mode, else the Routine cannot be activated or executed.

- VC Mode — The Amplifier Control Panel matching headstage must be in VC mode to run the routine.

  The default setting for new routines is 'VC Mode'. This prevents CC mode pA (10^{-12} A) current outputs from being accidentally overscaled by VC mode routines using mV (10^{-3} V) voltage outputs

- CC Mode — The Amplfier Control Panel matching headstage must be in CC mode to run the routine.

Note: The IPA amplifier can be switched into any mode (VC or CC) while a recording is in progress. However, it is your own responsibility to correctly interpret data with mixed recording modes.

Unit

Enter the base unit of measurement. The signal unit resolution is automatically adjusted.

- StimOUT Channels

  Fixed at 'V' for voltage clamp and "other" experiment types; fixed at 'A' for current clamp experiments.

- AuxOUT Channels

  Can be edited to any setting.

Scaling [for AuxOUT channels]

Offset — Apply an offset to the Auxiliary output channel (after any scaling.)

Factor — Apply a scaling factor as a fixed value or an equation.

(See the Equation Editor for more details.)

Relative to Holding

If 'Relative to Holding' is enabled, the headstage output signal is the command waveform summed with the Holding level in the Amplifier Control Panel. If the holding level is set to '0', this setting has no effect.

For the Auxiliary channels, the command waveform is summed with the Amplifier Control Panel I/O Auxiliary Output settings.

For digital channels, the command waveform is relative to the Amplifier Control Panel I/O Digital Output settings.

Note: When enabled, this setting is updated live by holding level changes in the Amplifier Control Panel.

Sweep End Output Settings

Control how the amplifier output levels (including I/O Auxiliary and Digital Output levels) are handled when the system is not acquiring data.

Inter-Sweep

This is the time between sweeps - after a sweep ends, but before the next sweep starts.

- Use Waveform Value — Set the output signals to their last values in the command waveform, at the end of a sweep.

  Use to avoid generating a short (potentially disruptive) glitch in your preparation, caused by returning to holding levels at the end of a sweep.

- Use Holding — Set the output signals to the Amplifier Control Panel "holding" levels, at the end of a sweep.

  This ensures that your cells are kept in a resting state as much as possible, and that each output sweep starts from the same holding level.

Routine End

This is the time after the Routine ends, until the next Routine starts.

- Use Waveform Value — Set the output signals to their last values in the command waveform, at the end of a Routine.

  Use to avoid generating a short (potentially disruptive) glitch in your preparation, caused by returning to the holding levels at the end of a Routine.

- Return to Holding — Set the output signals to the Amplifier Control Panel "holding" levels, at the end of a Routine.

  This ensures that your cells are kept in a resting state as much as possible.

Note: In demo mode, after changing the holding levels, if 'Relative to Holding' is enabled, then the 'Return to Holding' levels are updated immediately, otherwise they are only updated when a Routine is activated.

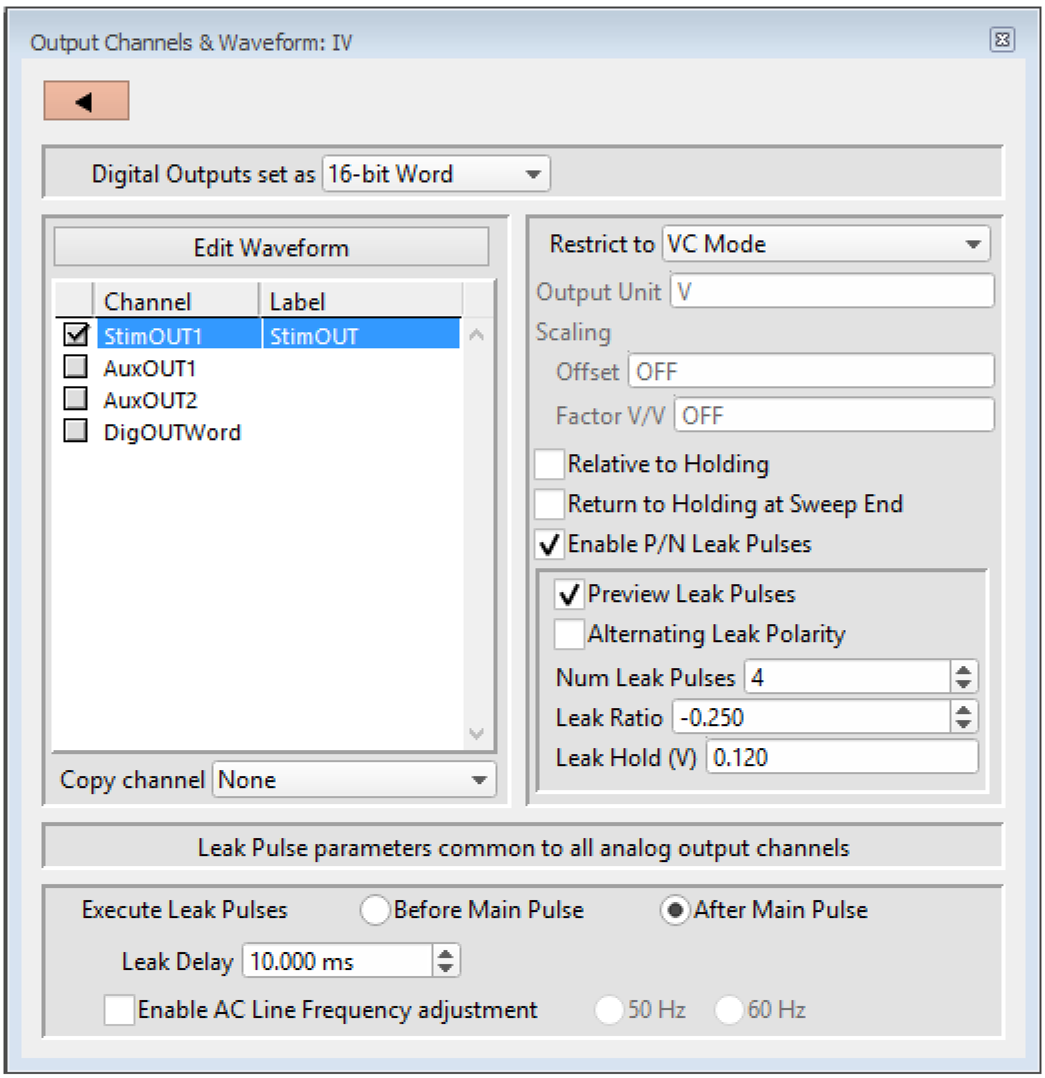


Figure 4-45. Output Channels & Waveform with Leak

Enable P/N Leak Pulses

[The P/N Leak Pulses section is only displayed if this field is enabled.]

Online P/N leak subtraction automates the removal of leakage currents from the data.

Endogenous leak currents can flow, even while a cell is in its resting state, from conditions such as an imperfect or leaky seal, or via existing ion channels, and affect response amplitudes. If endogenous leak conductance is an issue with your cell type, and/or high temporal resolution is required along with a need to reduce capacitive transients (e.g., with voltage-gated sodium currents), click 'Enable P/N Leak Pulses' and configure its settings below.

A "leak pulse" is a replica of the stimulus waveform, and is used to record a fraction of the leakage current. In this technique, leak pulses are generated, and the responses are averaged, scaled, and subtracted from the main response to remove the effects of leakage.

Note: The sub-pulses are stored as part of the sweep. This ensures that if any events occur during the sub-pulses or between the sub- and main pulses and causes unexpected or hard-to-interpret effects, the full original recording condition can be examined.

Preview Leak Pulses

Display the leak subtraction pulses in the Routine Editor Waveform Preview panel. A leak subtraction pulse is a scaled copy of the main stimulus waveform.

Alternating Leak Polarity

You can reduce directional bias in the leak conductance by alternating the polarity of the leak subtraction pulses on a sweep-by-sweep basis, as long as no ion channels are activated.

Num Leak Pulses

Set the number of leak pulses used to average out noise and leak conductance. Adjust this number in accordance with the amount of noise in the signal. With the high precision of modern 16-bit digitizers, this number can sometimes be reduced to less than 4 leak sub-pulses.

Note: As each leak pulse replicates the stimulus waveform, larger numbers of leak pulses is not recommended, as this can greatly increase the total duration of a sweep during acquisition, and the noise in the sub- and main pulses can add up and actually increase.

The default setting of '4' Leak Pulses, when used with the default Leak Ratio (-0.250) operates equivalently to pCLAMP's default P/N setting (4 subsweeps for P/4).

Leak Ratio

Set the leak subtraction pulse size relative to the main waveform pulse, using a ratio between +1 and -1. The setting should be low enough that no electrically-gated ion channels are activated. For instance, a Leak Ratio setting of 0.25 will generate leak pulses at ¼ the amplitude of the main stimulus waveform, while a Leak Ratio of 0.2 will generate leak pulses at 1/5 the main pulse amplitude.

Note: The program scales the leak subtraction pulses based upon the Leak Ratio setting, not the number of Leak Pulses. This means that the Leak Ratio can be set independently from Num Leak Pulses, instead of those settings being interdependent.

Tip: As an alternate way to avoid electrical activation of ion channels, use a negative ratio to reverse the polarity of the leak pulses relative to the main pulse.

Leak Hold

The leak pulses holding level can be set differently from the Routine main holding level, for flexibility in finding a suitable leak pulse voltage range. The scaled waveform amplitudes are measured relative to the Leak Hold level, but are subtracted relative to the IPA holding level.

Set to a fixed value, or enter as an equation.

(See the Equation Editor section for more details.)

Check Equations	Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports that the syntax is “ok”.
Insert special identifier	Acquisition, amplifier and reference settings for use in equations. (See list in Equation Editor.)
Undo	All changes in the equation editing session are discarded.

Leak Pulse parameters common to all D/A channels

Execute Leak Pulses

Leak pulses can be set to run before or after the main waveform pulse.

- Before Main Pulse — Sub-pulses are output relative to the Leak Hold level. After the sub-pulses complete, the signal goes to the IPA Holding level for the duration of the Leak Delay before the main pulse.

- After Main Pulse — After the main pulse completes, the signal goes to the Leak Hold level for the duration of the Leak Delay setting, and then outputs sub-pulses relative to the Leak Hold level.

Leak Delay [0 – 1,000.000 s]

If a settling time is needed between the leak pulses and the main waveform pulse, Leak Delay will insert a time delay between the execution of the leak pulses and the main pulse. Provide enough time to avoid interference of the leak pulses with any active currents or inactivation of ion channels.

When leak pulses occur before the main pulse, Leak Delay uses the amplifier's Holding level; when leak pulses occur after the main pulse, Leak Delay uses the Leak Pulses 'Leak Hold' level.

Enable A/C Line Frequency adjustment

The effect of AC line-frequency noise (hum) can be automatically reduced during P/N leak subtraction recording:

- 50 Hz — Enable the reduction of 50 cycle AC line noise.
- 60 Hz — Enable the reduction of 60 cycle AC line noise.

This Line Frequency adjustment automatically calculates the proper interpulse interval for the P/N sub-pulses, so that they are counter-phased to the line frequency of the output signal, which reduces hum without filtering the signal.

Routine Editor: Waveform Editor

Click the 'Edit Waveform' button to open the Waveform Editor and design a command waveform for the selected output channel.

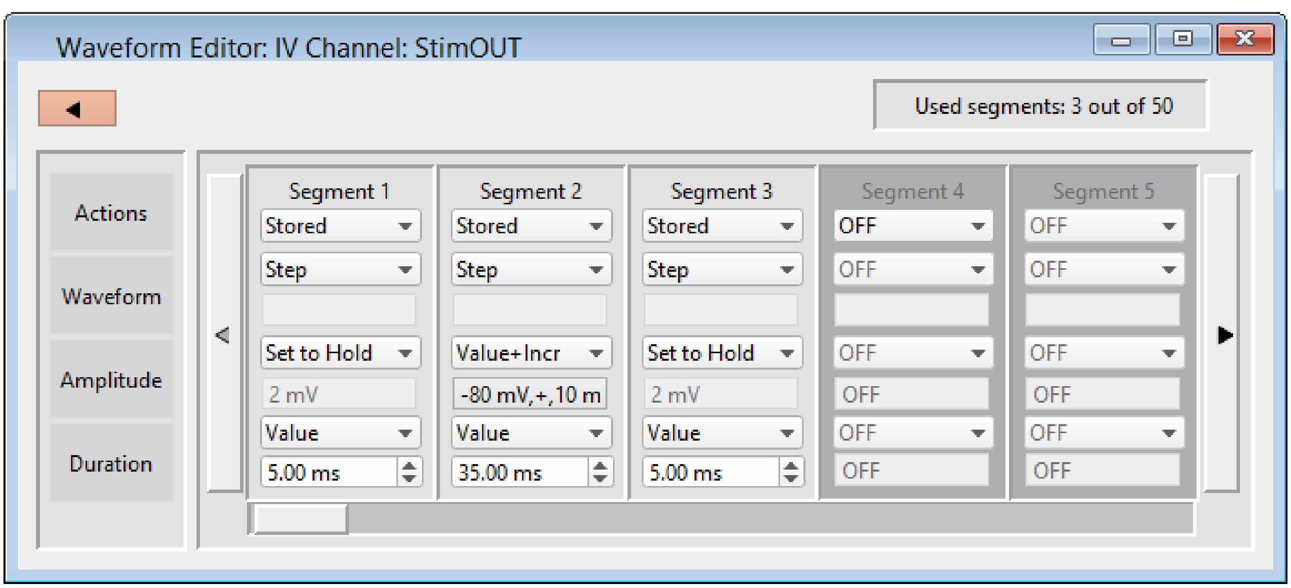


Figure 4-46. Waveform Editor

Close button  Use this button to close the Waveform Editor window.

Used segments: [# out of 50] Up to 50 contiguous segments can be configured in a waveform.

Actions

OFF — Unused segments are labeled as 'OFF'.

Tip: A segment with a Duration of '0' ms is equivalent to 'OFF'. This is a convenient way to skip a segment instead of deleting it.

Stored — Enable a segment for stimulation and recording.

Not In Leak — If P/N LeakPulses are enabled, this will optionally exclude the segment from being generated in the P/N Leak Subtraction output wave.

This is useful for inactivation or recovery studies, when commands do not change for long periods of time.

Insert — Insert a default Segment into the current position, and increment the position of the following Segments, i.e. move them to the right.

Copy — To copy a segment, click the segment's Actions list and select 'Copy'. A copy is inserted as the next segment.

To copy multiple segments, select the segments to be copied. Then, for the segment to be inserted before, click its Actions list, select 'Copy', and enter the number of times to copy the segments - the selected segments are inserted before the "Copy" segment.

Delete — To remove a segment, select its 'Delete' Action.

Note: If there is only one segment, it cannot be deleted - there is always at least one segment enabled.

To remove multiple segments, select the desired segments. Then, click any segment's Actions list and select 'Delete'. All selected segments are deleted.

Tip: To select multiple segments, in Windows use Ctrl-click, or in macOS use Command ⌘-click, to highlight each segment, or use Shift-click to highlight a range of segments.

	Any following segments shift their Segment #'s down by the number of deleted segments.		
Waveform	Select the waveform shape. For Waveform types Sine / Chirp / Squarewave / Template / Triangle, a 'Parameters' field displays below this field, to allow quick access to their parameters.		
Step	The waveform amplitude rapidly jumps from a pre-existing level to the new level within one sample point, and stays at the new level for the duration of the segment. The resulting waveform shape looks like a step. The first segment typically consists of a Step waveform set to the holding level amplitude (Set to Hold).		
Ramp	The waveform amplitude goes from the previous amplitude to the new amplitude as a smooth straight line - the sloping line looks like a ramp.		
Tip:	To create a sawtooth pattern, follow a ramp segment with a zero duration Step segment. This sharply resets the ramp amplitude to the baseline level – the next ramp will start from the baseline level again.		
Sine	The waveform is a sinusoidal wave.		
	Sine Wave Cycles	Multiple	One or more cycles
		Single	One cycle, where the Cycle Duration is equal to the Segment Duration.
		For LockIn	For sensitive capacitance measurements. A corresponding virtual input LockIn channel also needs to be enabled. For multiple Sine LockIn segments in a Routine

waveform, use the same sine wave Amplitude and Cycle Duration for each Segment.

Amplitude	Amplitude of the first peak from the sine wave baseline.
Tip:	To offset a sine wave from the default baseline (0 units), set the segment Amplitude value, or enable Routine Editor / Output Channel 'Relative to Holding'. For LockIn measurements, the larger the sine wave amplitude, the better the signal-to-noise ration for the measurements - just be sure to avoid the activation range of voltage-gated ion channels
Cycle Duration	One cycle length (ms) [For LockIn only] A preset list of frequencies are available: 2.0 ms (500 Hz) 1.0 ms (1 kHz)
Segment Duration	Sine wave duration (ms)

Squarewave — The waveform generates a train of rectangular pulses.

Base Amplitude Increment	Increment the baseline amplitude for each succesive pulse. [±1.00 V, ±20.000 nA]
Step1 Amplitude	Amplitude of first pulse. Value [±1.00 V, ±20.000 nA] Var_r[1 – 16]
Step1 Width	Duration of first pulse (ms).

		Value [ ] Var_r[1 – 16]	
	Step2 Amplitude	Amplitude of second pulse. Value [±1.00 V, ±20.000 nA] Var_r[1 – 16]	
	Step2 Width	Duration of second pulse (ms) Value [ ] Var_r[1 – 16]	
	Segment Duration	Square wave duration (ms)	
Chirp	This waveform generates a sinusoidal wave that changes its frequency over time.		
	Chirp Type	Linear	A linear change in frequency.
		Geometric	A geometric change in frequency.
		Note:	For a geometric chirp, a minimal frequency spread is enforced: the End Frequency has to be at least double the Start Frequency, or half or less than the Start Frequency.
	Amplitude	[±1000.00 mV, ±20000.00 pA]	
	Start Frequency	[1 – 50000 Hz]	
	End Frequency	[1 – 50000 Hz]	
	Segment Duration	Chirp wave duration (ms)	
Template	Assign an arbitrary waveform to a segment.		

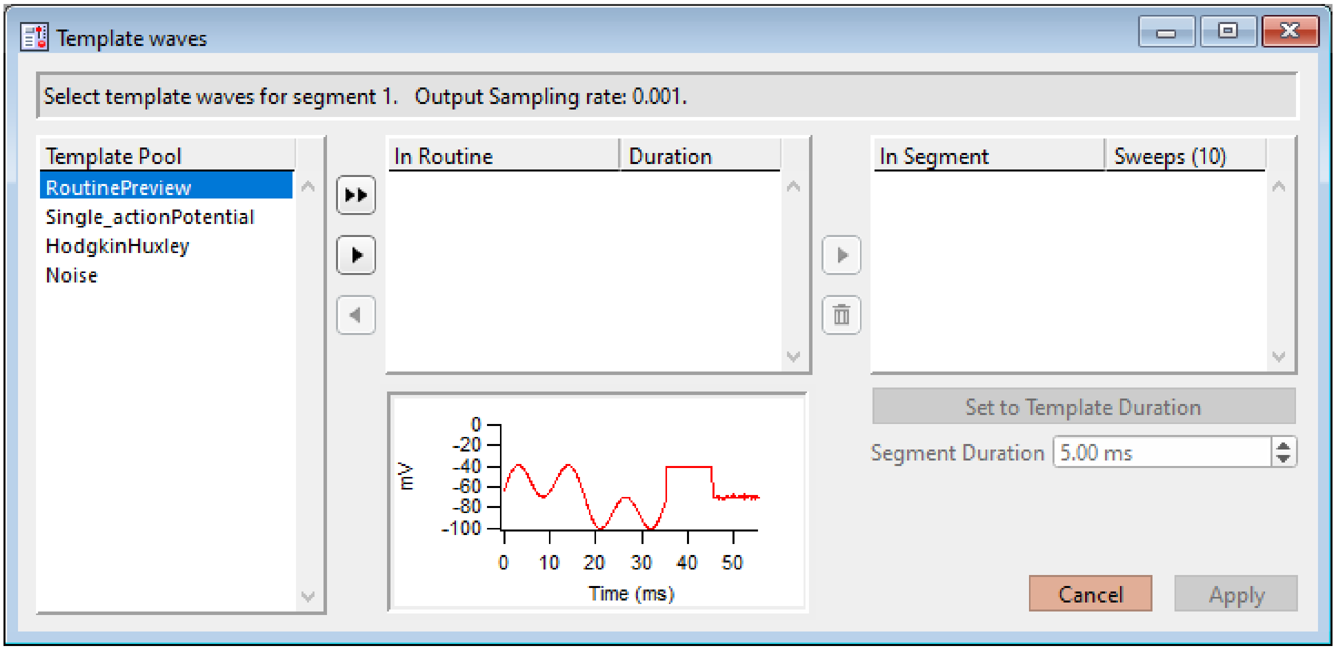


Figure 4-47. Template Waves

[Status field] The Routine segment # and output sampling rate are displayed

If the template sampling rate does not match a Routine sampling rate, the template data are interpolated to match the Routine sampling rate.

Template Pool Templates loaded in the Template Editor Pool plus extracted templates.

Copy the selected template wave from the Template Pool into a Routine and Segment.

Copy the selected template wave from the Template Pool into a Routine.

In Routine Templates stored in the Routine.

Each output channel can have a maximum of 16 template waves stored in its routine. The same template can be used in multiple segments. Each segment can have one or multiple templates assigned to it.

Probably the most used case will simply be a single template paired with a single segment, but the possibilities are endless.

Note: To avoid unnecessary increase in the sizes of routines and Routine Pool files, only include templates that are actually used by segments.

Duration (ms) The duration of the template trace.

Copy the selected template from a Routine to a Segment.

Delete the template wave from a Segment or an unused Routine.

In Segment Each segment can have one or multiple templates assigned to it.

If only one template is listed, then for any number of sweeps, the segment output wave will be the same for all sweeps.

If more than one template is assigned to a segment, and the number of sweeps is greater than 1, then an output wave is created as:

1st sweep with the 1st template on the list.
2nd sweep with the 2nd template.
Etc.

If there are more sweeps than templates, then the templates are reused (round robin) for each subsequent sweep.

Sweeps Number of sweeps in the Routine.

Set to Template Duration

Set the segment duration to match the template (sweep) duration.

Segment Duration The Segment Duration can be manually adjusted here.

When typing in a value, if no unit type is entered, the unit type defaults to seconds (s). If you enter a number followed by an 'm' or 'ms', the unit type is milliseconds (ms).

Triangle The waveform generates a train of triangular pulses.

Base Amplitude Increment

Increment the baseline amplitude for each succesive pulse.

Peak Amplitude — Amplitude of the triangle pulse.

Ramp1 Width — Duration of the initial phase.

Ramp2 Width — Duration of the secondary phase.

Segment Duration — Duration of the triangle train.

Membrane Test — The waveform applies a predefined negative pulse step (-5 mV) with a 50% duty cycle for Routine-based measurements.

Use with the Real Time Measurements 'Analysis Functions':

- MT Series Resistance
- MT Membrane Capacitance
- MT Membrane Resistance

Warning! For valid results, 'Cell Compensation' and Rs Correction should be disabled in the Amplifier Control Panel.

Amplitude (*analog*) Set the waveform amplitude for a Segment.

For the Chirp, Sine, Squarewave and Triangle waveforms, this is used as a baseline offset.

For Auxiliary output channels, when the Output Channel / Scaling Factor is not "1", i.e., when scaling is applied to the signal, then a non-editable scaled output field is also displayed below the amplitude value field.

Set to Hold — Use the Amplifier Control Panel holding level for the Segment amplitude.

Tip: Record an initial baseline in Segment 1 to help intepret your data (or set in the last segment.)

For voltage-clamp experiments, records the leak current along with the actual holding voltage.

For current-clamp experiments, records the actual cell potential along with the actual holding current.

Avoid using the last Segment for this, as post-stimulation data can be recorded, such as from tail currents.

Value — Use a single number for the segment amplitude.

Value List — Set an arbitrary segment amplitude for each sweep.

[Sweep | Value]	For each sweep, enter a number.
Fill Remaining List	Copy the active value to all remaining sweeps in the list.
Segment Duration	Adjust the duration of the Segment.
Number of Sweeps	Adjust the number of sweeps in the Routine.

Value+Increment — Increment the segment amplitude for each sweep.

Start Value	• Holding • Value [±1000.00 mV, ± 20000 pA]
Increment Value	[±1000.00 mV, ± 20000 pA]
Segment Duration	Adjust the duration of the Segment.
Number of Sweeps	Adjust the number of sweeps in the Routine.

Equation — Specify the segment amplitude as an equation.

[Equation field]	A free-form text field. Errors are reported under this field.
Check Equations	Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports that the syntax is “ok”.
Insert special identifier	Acquisition, amplifier and reference settings are available for use in equations. (See list in Equation Editor.)
Undo	All changes in the equation

editing session are discarded.

(See the Equation Editor for more details.)

Note: Computing an equation for an output wave consumes significant computing power, as every data point needs to be computed by the CPU. While a slight update delay in such operations is expected, for computers with marginal computing power, the "beach ball" icon displays while the computer is unresponsive and busy processing.

Var_r[1 – 16] Variable labels are displayed if the Routine Variables table is enabled.

Amplitude *(digital)* Digital settings are displayed if digital outputs are enabled.

Bit Set the digital level for an individual bit.

- LOW = O
- HIGH = 1

Bit Word Values are the decimal number of an 8-bit word (0 – 255), also displayed as a bit pattern.

Duration Set the segment duration.

Value Use a single number for the segment duration. [0 – 12 ks]

When typing in a value, if no unit type is entered, the unit type defaults to seconds (s). If you enter a number followed by an 'm' or 'ms', the unit type is milliseconds (ms).

Value List Set an arbitrary segment duration for each sweep from a list of numbers.

[Sweep | Value] For each sweep, enter a number. [0 – 12 ks]

(Blank lines are removed.)

Fill Remaining List Copy the active value to all remaining sweeps in the list.

Number of Sweeps Adjust the number of sweeps in the Routine.

Value+Increment	Increment the segment duration for each sweep.	
	Start Value	[ms]
	Increment Value	[ms]
	Number of Sweeps	Adjust the number of sweeps in the Routine.

Equation

Specify segment duration as an equation

[Equation field] A free-form text field.

[Errors are reported under this field.]

Check Equation — Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports that the syntax is "ok".

Insert special identifier — Acquisition, amplifier and reference settings are available for use in equations. (See list in Equation Editor.)

Undo — All changes in the equation editing session are discarded

(See the Equation Editor for more details.)

Var_r[1 – 16] — Variable labels are displayed if the Routine Variables table is enabled.

Segment Controls

To copy or delete multiple Segments, click on the background area of used Segment(s) - the background color turns gold, and enables the Actions items: Copy and Delete.

Standard mouse behaviour is used to select multiple Segments:

• Individual Segments:	Windows:	Ctrl-click
	macOS:	command (⌘)-click
• Range of Segments	Both:	Shift-click

To step through the Segments, click on the left or right Segment arrow buttons.

To scroll through the Segments, use the slider at the bottom of the Waveform Editor.

Routine Editor: Real Time Measurements & Graphs

Online analyses are configured in the Real Time Measurements & Graphs dialog. Measurement regions display in the Scope window, and their associated analyses are plotted in an Analysis sub-window during acquisition.

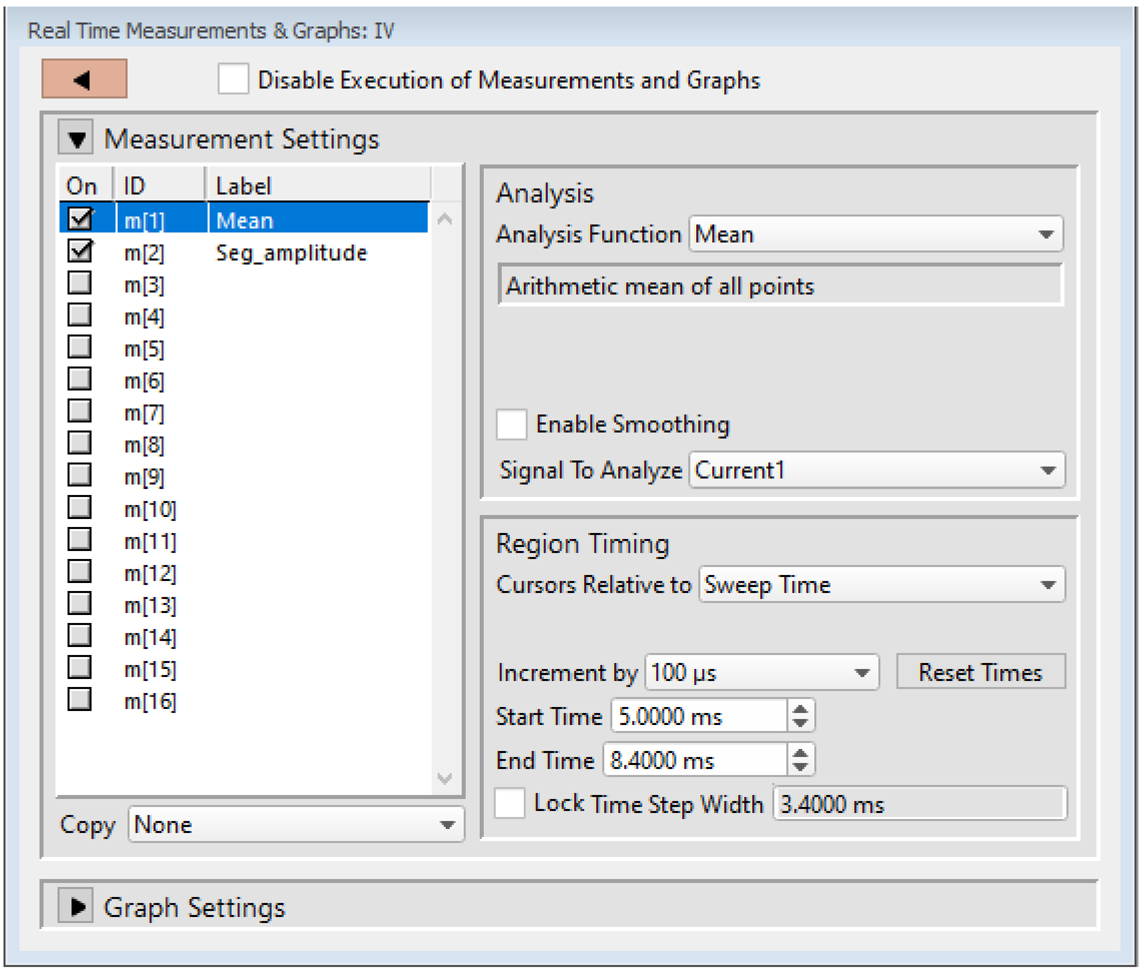


Figure 4-48. Real-Time Measurement Settings

Disable Execution of Measurements and Graphs

Block all measurements and analyses with one click.

Measurement Settings

On Enable an analysis to run.

ID	Measurement regions are identified with an ID number: m[#] [1 – 16]
Label	These measurement labels display in the Waveform Preview and Scope windows, and can be used in equations. A Label name is automatically generated from the Analysis Function; double-click to manually edit.
Copy	Copy to the selected Measurement another Measurement's settings.

Clear All Measurements

All measurements and their settings are cleared.

Analysis

Analysis Function

Choose from 23 predefined Analysis statistics for each measurement:

Absolute area	Negative area values are converted to positive and summed with the positive area values.
Absolute peak	Largest absolute value.
Area	Signed area - negative values negate positive values.
Decay time	10 - 90% decay (fall) time, "peak to end".
Decay Tau	Time constant of 'Decay time'.
Frequency	Number of threshold crossings per second (Hz).
Max Slope	Maximum slope of simple linear regression fit.
Max value	Value of largest sample.
Mean	Arithmetic mean of the samples.
Min value	Value of smallest sample.
MT Series Resistance	Membrane test 'Rseries' value of a patch.
MT Membrane Capacitance	Membrane test Cmembrane' value of a whole-cell patch.
MT Membrane Resistance	Membrane test 'Rmembrane' value of a whole-cell patch.
Rise Tau	Time constant of 'Rise time'.
Rise time	0 - 100% rise time, "start to peak".
RMS noise	Root-Mean-Square noise.
Segment amplitude	Maximum amplitude of the selected segment.
Segment duration	Duration of the selected segment.

Slope	Slope of simple linear regression fit.
Std deviation	Standard deviation of the samples: $\sqrt{(\text{variance})}$
Time of absolute peak	Time from sweep start to largest absolute value.
Time of max	Time from sweep start to largest sample.
Time of min	Time from sweep start to minimum sample.
Time to threshold	Time from sweep start to first threshold crossing.
Variance	Variance of the samples.
Weighted tau	Weighted time constant. (Area / Peak with end-of-measurement y0 subtracted.)

These analyses can be directly plotted, or used in more complex equations. (See the Equation Editor section for more details.)

[Status field] A short description of the selected Analysis.

Threshold

This amplitude level needs to be crossed by the data to trigger measurements for:

- Rise/Decay time
- Rise/Decay Tau
- Frequency
- Time to threshold

Polarity

The direction of a Threshold crossing.

- Positive — Positive direction threshold crossing.
- Negative — Negative direction threshold crossing.
- Largest Change — Use the polarity direction of the largest change for Rise and Decay analyses.

Enable Smoothing

[2 – 200]

Set the number of Gaussian smoothing operations per measurement.

Smoothen noisy data to reduce the effects of high-frequency noise on measurements by averaging the data sample points with an unweighted sliding average.

Note: Smoothing is not applied to the analyses ‘Segment duration’ and ‘Segment amplitude’. These are fixed values not subject to modification.

Signal to Analyze

For each enabled Analysis measurement, select the signal to be measured from the list of Input Channels. A measurement is only made on one input channel, but it can be used in multiple graphs.

<u>Region Timing</u>

Cursors Relative to Set the measurement boundaries with left / right cursors.

Note: Cursor Start times cannot be greater than their End times.

- Sweep Time — Set relative to the start time of a sweep (time zero).

  Increment by: [ ]

  The 'Start Time' or 'End Time' spinners increment by this amount.

  The listed time values depend upon the input filter bandwidth.

  Reset Times — Set the cursor Start / End Time to the beginning and ending of the sweep.

  Start time — Set the left cursor start time (s).

  End time — Set the right cursor start time (s).

  Lock Time step width — Fix the width of the measurement region.

  The measurement width is maintained at a constant value when the cursor 'Start Time' is updated.

  [ ] The width of the cursors in seconds.

  The minimum width size is 2 sample points.

- Segment Time — Set the time range as a ratio of the Segment duration.

  Uses the segment timing from the input signal's "Parent Output Channel".

  Out Channel — [Output Channel list] [Segment #]

  Increment by:
  [0.001, 0.002, 0.005, 0.01, 0.02, 0.05, 0.1, 0.2, 0.5]

Increment the Start / End Ratios by a relative amount.

Reset Ratios [0.0000] / [1.0000]

Reset the Start / End Ratios to span the entire segment.

Tip: If unwanted segment boundary issues occur, where measurements occur in a neighboring segment, increase the Start Ratio or decrease the End Ratio until the issue is resolved.

Start Ratio [0 = beginning of Segment]

Set the left cursor as a ratio of the Segment duration

Time [0.0000 s] Cursor start-time read-only field.

End Ratio [1 = end of Segment]

Set the right cursor as a ratio of the Segment duration.

Time [1.0000 s] Cursor end-time read-only field.

If the Start / End Ratios extend past the boundary of a Segment, and the measurement is switched to a beginning or ending Segment, the Start / End Ratios are reset to '0' and '1' respectively.

Cursor Time Width The width of the measurement region is reported.

Warning! The various input and output sampling rates and time durations might not exactly match up with the measurement region. Please check for Segment boundary issues when performing measurements.

Graph Settings

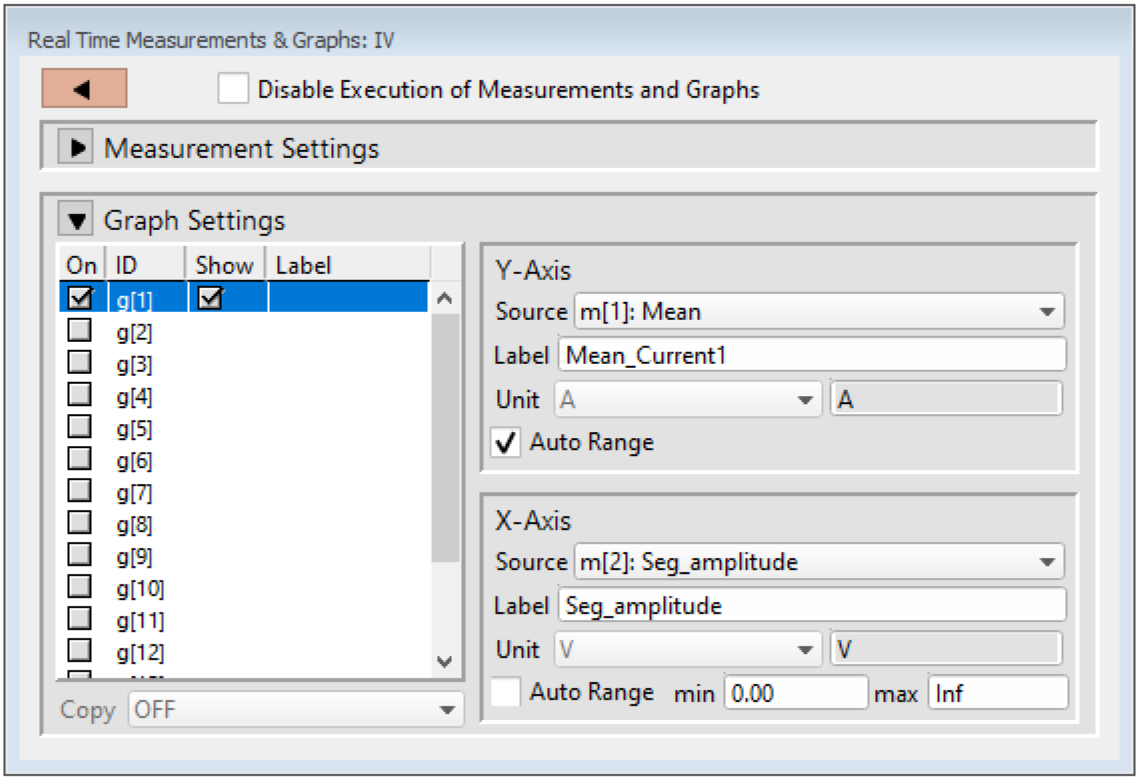


Figure 4-49. Real Time Graph Settings

On	Enable a graph to configure its settings.
ID	Graphs have a default ID (identification): g[1] - g[8]
Show	Display this graph in an Analysis window during acquisition and analysis.
	Note: If the Y-Axis 'Equation' field is blank, the graph will also be blank.
Label	Double-click to manually enter a graph label for the Analysis.
Copy	Use to transfer graph settings to a new graph. Highlight a disabled Graph ID, (but do not enable it), then select from the drop-down list of enabled graphs.
Y-Axis	Select a measurement ID to plot against the graph Y-Axis, or select a Y-Equation for a customized Y-axis plot.

Source	Select a Measurement, or enter an equation (see the Equation Editor for more details.)
Label	Enter a customized name for the graph Y-Axis.
Equation	Click to edit with the Equation Editor (see the Equation Editor for more details.)

For example, to monitor cell compensation, use the Special ID 'RsComp'. Then track cell changes sweep-by-sweep by running a planned Paradigm with a Routine step inside a 'For Each' step.

If running from a planned Paradigm, see the 'For Each' step example to track cell changes sweep-by-sweep.

Unit — Select a standard unit from the drop-down list, or enter a custom unit type.

Note: Standard unit solutions, such as 'pA' or 'mV', are automatically calculated and displayed in the graph.

Auto Range — Restrict the graph Y-axis range.

- Y-min — Lower limit of the Y-axis.
- Y-max — Upper limit of the Y-axis.

X-Axis — Display the graph X-axis with a standard Time base, or select a measurement ID to plot against the X-axis, or use an X-Equation for a customized X-axis plot.

Label — Enter a customized name for the X-Axis.

Equation — Click to edit with the Equation Editor.

(See the Equation Editor for details.)

Unit — Select a standard unit from the drop-down list, or enter a custom unit type.

Note: Standard unit resolutions, such as 'pA' or 'mV', are automatically calculated and displayed in the graph.

Auto Range — Restrict the graph X-axis range.

- X-min — Lower limit of the X-axis.
- X-max — Upper limit of the X-axis.

Routine Editor: Routine Variables

Up to 16 Routine Variables can be configured for stimulus control. These variables allow interactive manual control of the waveform amplitude and duration, or automatically via Paradigms.

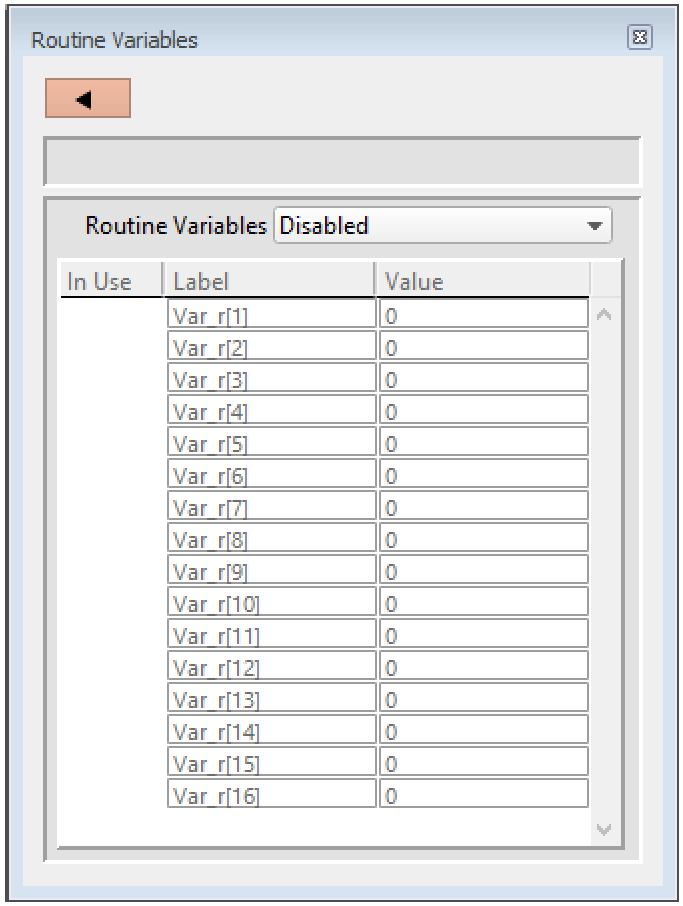


Figure 4-50. Routine Variables

Routine Variables

- Disabled
- Enabled — Once Routine Variables are enabled, they become visible in the Waveform Editor Amplitude and Duration lists.

In Use — A checkmark means this Routine Variable is in use by the Waveform Editor.

Label — Var_r[1 – 16] — Edit the default variable name if desired.

Value — Numeric values can be manually entered or set by a Paradigm.

Note: If Routine Variables are enabled and then disabled, the Waveform Editor converts its 'Var_r' settings to 'Value' settings using the last enabled values.

4.1.9 Scope (Acquisition)

The Scope (acquisition) window is used for viewing and recording digitized time-series data, displayed as a smooth interpolated line.

Note: Only one Scope window can be open at a time. For example, if a Scope window is open for data acquisition, then opening the Membrane Test, Free-Run, or data for reanalysis, will close the acquisition Scope window, and re-open it as the new type of Scope window.

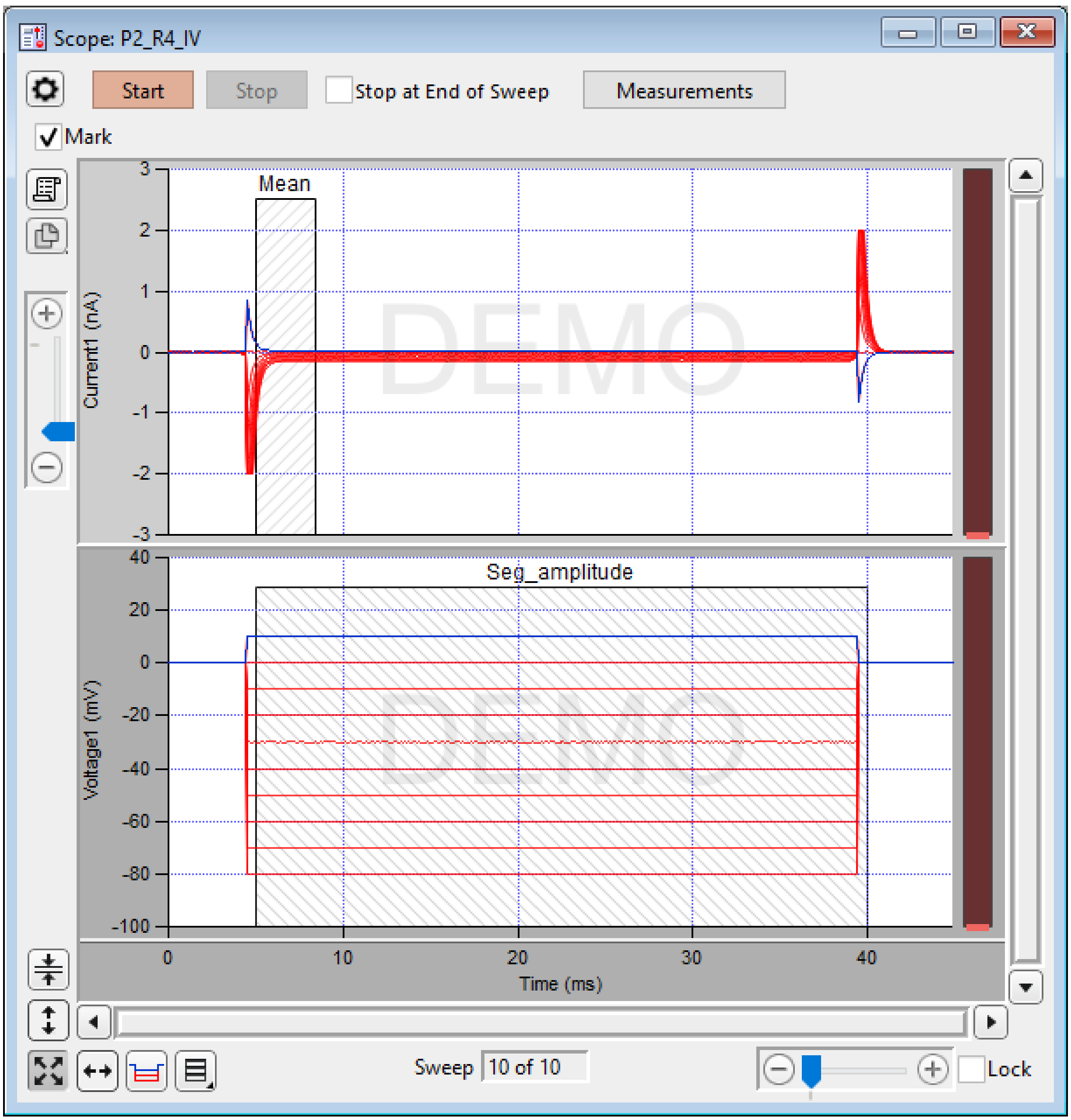


Figure 4-51 Scope (Acquisition) Window

The Scope window is titled with the active Paradigm sequence number + Routine name.

Controls

Signals The central area of the Scope window graphically displays input data in up to 16 separate signal panes. Click on a signal pane to make it "active" - the Y-axis border area displays in a lighter color, and the Y-axis controls (magnify, scroll) apply to it. Non-active panes display with a darker Y-axis border area.

If multiple signals are displayed stacked on top of each other, you can vertically resize the panes by clicking and dragging them with a resizing cursor. Position the mouse cursor over a pane separator (the horizontal area between panes) to change it to the resizing cursor (a horizontal line with a vertical double-headed arrow.)

Note: Two additional data points are appended to the sweep data to support post-sweep holding levels and segment boundary rounding issues.

Signal Display Mode

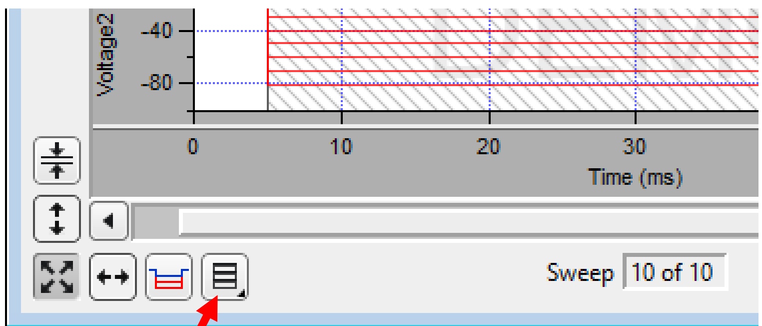


Figure 4-52. Signal Layout

Signal pane arrangements are set by the 'Select signal layout' button  in the lower left corner of the Scope window. Select how the input signals are graphically arranged:

- Stack Vertical column of signals
- Single Only the active signal
- [m x n] Tiled array of signals with 'm' rows and 'n' columns

Cursors Measurement cursors are visible as light gray vertical bars in the signal panes. Each measurement region is bounded by a start-time cursor (the left edge) and an end-time cursor (the right edge).

To move a measurement region, click and drag it with the mouse - the region briefly turns dark when selected.

To resize a measurement region, click and drag an end-time cursor (the right edge of a region.)

Magnification and Scrolling

Signals can be magnified/unmagnified using several X- and Y-axis display controls in the Scope window. Any magnification applied to the signals persists during acquisition.

- Magnification Combo 

  Click on the "+" and "-" buttons to magnify / unmagnify by steps, or click and drag the slider to smoothly zoom / unzoom the active signal. The Y-axis magnification only controls the active pane; the X-axis magnification controls all signals.

- Lock

  Enable to retain the X-axis scaling and position for the next activated or created Scope window (Acquisition, Analysis, Free Run, Membrane Test).

  However, any changes to the X-axis duration (rescaling or autoscaling) or position (scrolling) disables the 'Lock' option.

- Axis Zoom

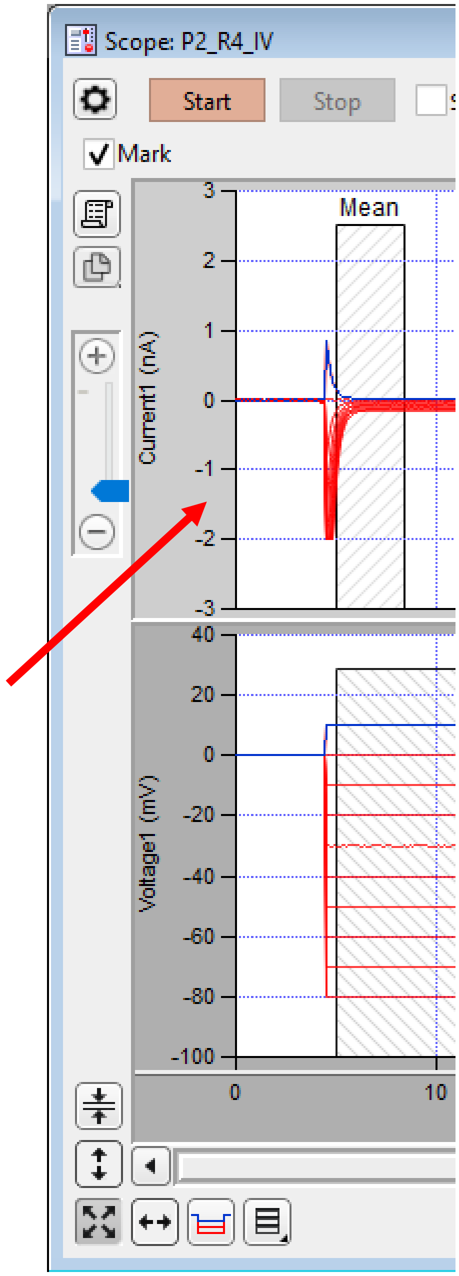


Figure 4-53. Axis Magnification

When the mouse is moved into the X- or Y-axis areas, the cursor changes to a double-headed arrow. As you click and drag the mouse cursor, a dark bar displays in the axis showing the magnification area; or scroll the mouse wheel up / down to expand / shrink the X-axes or the active Y-axis

- Area Zoom:

  Any area of interest in a signal pane can be graphically selected and expanded.

  1. Move the mouse cursor into a signal pane - it changes into a large "+".

2. Click and drag a bounding box around the desired data. (This box is also referred to as a "marquee".)
3. Right-click in the marquee and select the desired action.

Expand	Applies to all signals.
Horiz Expand	Applies to all signals.
Vert Expand	Applies to active signal.
Shrink	Applies to all signals.
Horiz Shrink	Applies to all signals.
Vert Shrink	Applies to active signal.

- Axis Scroll Bars

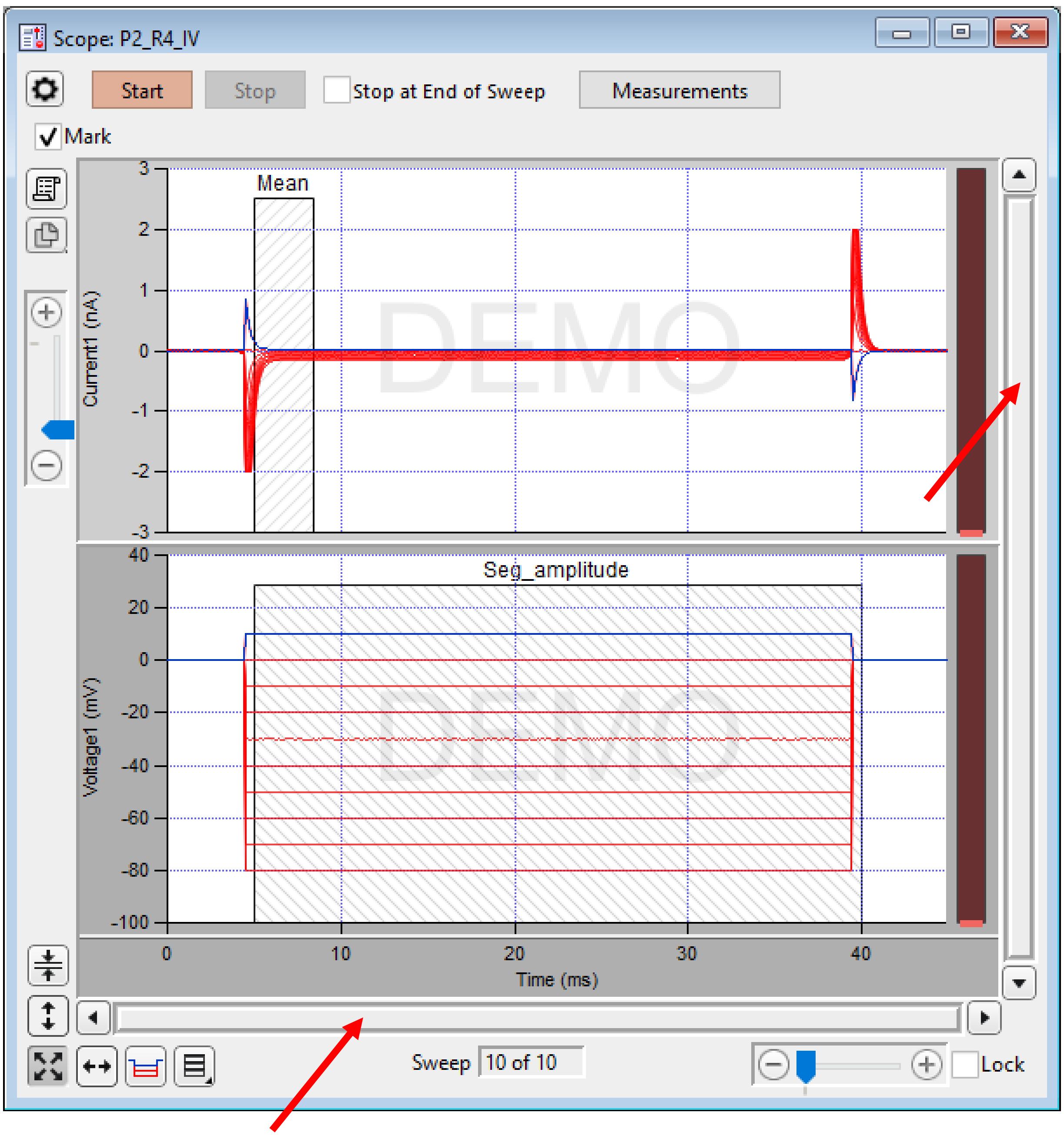


Figure 4-54. Axis Scroll Bars

The X-axis scroll bar is directly underneath the X-axis, while the Y-axis scroll bar is on the far right-edge of the Scope window. Click and drag the scroll bar slider buttons, or use their directional buttons to move the displayed signals in the desired direction. (The size of the X- and Y-axis scroll bar slider buttons reflects the amount of signal magnification.) The Y-axis scroll bar controls the active signal pane; the X-axis scroll bar controls all panes.

- Center:

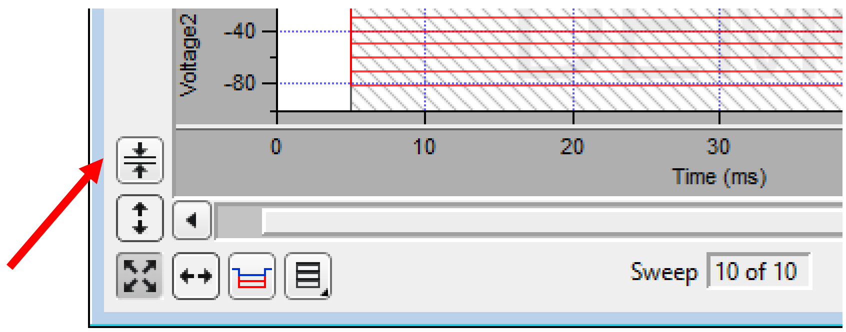


Figure 4-55. Center Button

Center the active signal. Vertically off-center or off-screen data is repositioned so for the active sweep, the mean of the Y-axis data is vertically centered in the signal pane. The Y-axis offset is adjusted, while the Y-axis scaling is kept unchanged; the X-axis remains unchanged.

To center all signals, shift-click the button.

- Autoscale Axes

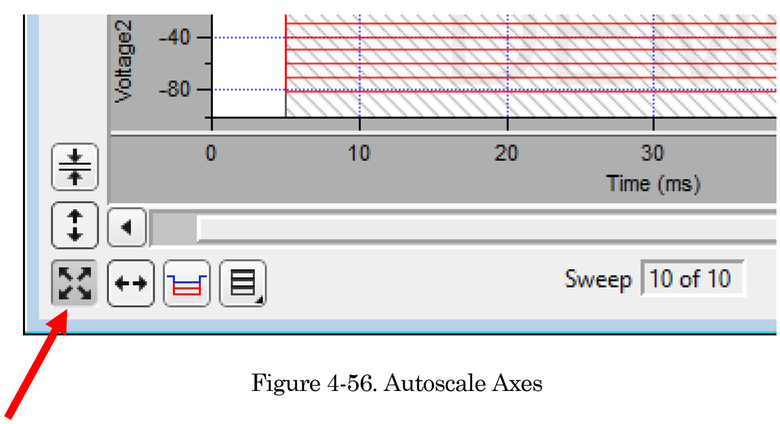


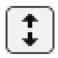
Figure 4-56. Autoscale Axes

 Autoscale.

Click to atoscale once the Y-axes of all visible signals to their visible sweeps data limits, and to reset all X-axes to their full sweep duration.

To include the zero amplitude in the Y-ranges, configure “Continuous autoscale from zero” in the Set Preferences / Scope Window / Acquisition / Y axis initial settings.

To continuously autoscale the Y-axes of all visible signals during acquisition, in Windows right-click (or Shift-click or Ctrl-click) the button, or in macOS Control-click the button; also all X-axes are reset to their full sweep duration. The Autoscale button remains depressed in this state.



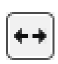
Y-Autoscale.

Click to autoscale once the Y-axis of the active signal. To include the zero amplitude in the Y-range, configure “Continuous autoscale from zero” in the Set Preferences / Scope Window / Acquisition / Y axis initial settings.

To autoscale the Y-axes of all visible signals, in Windows right-click (or Shift-click or Ctrl-click) the button, or in macOS Control-click the button.

Tip: To invert the Y-axis of the active signal, such as for data with reversed polarity from an outside-out patch, right-click in the Y-axis of the signal and select Axis Properties / Axis Range. Either reverse the Manual Range Settings / Minimum and Maximum values, or disable the Manual Range and enable the Autoscale Settings / Reverse axis.

- X-Scale.



Full scale all X-axes.

All signals X-axes are reset to their full-scale setting.

- Amplitude Meters

Amplitude meters are displayed on the right border of signal panes (excluding virtual channels). They provide visual feedback on the integrity of your data recordings, similarly to how audiometers monitor audio signals.

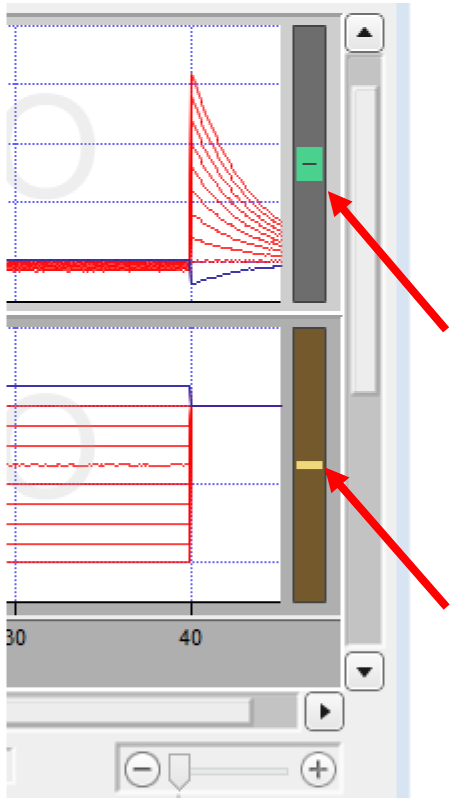


Figure 4-57. Amplitude Meters

For the Triggered Sweeps acquisition mode, each displayed signal has its own Y-axis amplitude meter on the inner-right side of the associated Scope pane.

For the Continuous Sweep acquisition mode, or if acquisition has not yet started, these meters are completely black.

The height of the colored meter bars represents a signal's data range vs. the full recording range of the IPA digitizer. The color of the meter bar corresponds to the data "health":

- Green: Good — Signal within appropriate range

  When the recorded data are within acceptable amplitude limits, the amplitude meter is green.

- Yellow: Caution — Signal approaching limits

  (within 10% of range limits)

  If too little hardware gain is applied, and there is a danger that saturation will occur, as the data are near the upper limit, the ampli-

			tude meter is yellow, as a warning sign to decrease your hardware gain. If too little hardware gain is applied, and the recorded signal has insufficient amplitude resolution, the amplitude meter will be a thin yellow line. When this occurs, the digitizer is not utilizing enough bits to accurately represent the data, and your signal of interest might be contaminated with noise. In this case, consider increasing your hardware gain.
•	Red:	Danger	Signal too large or small (within 1% of, or at range limits) When an amplitude meter is displayed in red, it indicates that the data might have gone out of range and be invalid. If too much hardware gain is applied, the recorded signal will be in danger of saturating, i.e. your data will exceed the amplitude limits of the digitizer. If the data saturates, those data points are substituted with the maximum amplitude of their input channel. If too little hardware gain is applied, the signal is too small for accurate measurement, and the hardware gain needs to be increased.

Other Buttons

	Scope Settings: Set all marks in sweeps of active series Clear all marks in sweeps of active series ------ Begin with all marks set (in sweeps of active series)

	Begin with all marks cleared (in sweeps of active series)
Start	Start recording and displaying digitized analog data in the Scope window input channels. When you click the ‘Start’ button, the Scope is cleared, the Control Panel Offset is locked, and data recording starts (after ~300 ms). When acquisition is running, the Scope window updates every 200 ms. If the Sweep Start-to-Start time is ≥ 5 s, the “Time to next sweep: # s” is reported below the Start / Stop buttons. Metadata prompts are set for Routines or Paradigms, the Confirm Metadata Settings dialog displays just before recording occurs. If measurement graphs are enabled, a docked “child” Analysis window opens and plots sweep-by-sweep measurements. If no paradigm is running, an “Auto-triggered Paradigm” is generated and assigned a Paradigm name with the current Date/Time.
Stop	Stop recording data immediately. If in the middle of a sweep, any partial sweep in progress is not saved with the data. If external triggering is configured, after clicking ‘Stop’, click the ‘Do Trigger’ button, and then ‘Stop’ again.
☐ Stop at End of Sweep	If the ‘Stop at End of Sweep’ checkbox is enabled, then the current sweep will complete before data acquisition is stopped, and the last recorded sweep will be a complete sweep of data. The message ‘Waiting to stop’ displays below the Stop / Start buttons, until the last sweep completes and acquisition stops. If no sweep is in progress, acquisition stops at the end of the next sweep to be recorded.
Measurements	Show Cursors: Display measurement cursors in the Scope window. Hide Cursors: Do not display cursors in the Scope window. Button displays as “Measurements(H)”. Lock Cursors: Prevent cursors from being moved or altered. Button displays as “Measurements(L)”. — No Measurements or Graphs Analyze with Current Measurements

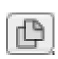
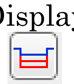
	Analyze with Original Routine Measurements Analyze with Routine Last Executed Measurements Load Default Measurements Save as Default Measurements Edit Measurements: Open a special Reanalysis Measurements & Graphs dialog, where all edits apply instantly to the measurements and graphs, even during acquisition. These changes override the loaded routine for quick interactive control.
Mark	Enable to "mark" the current (or upcoming) sweep. See the Data Navigator to process marked sweeps.
Do Trigger	This green button appears when acquisition is started for a Routine configured with an external trigger, and provides a manual trigger option. The message "Waiting for trigger…" also displays.
Copy to Layout 	Copy all visible Scope signals and analyses into a new Layout window, or append to an existing Layout page.
Copy to Clipboard 	Copy the active signal graph to the system clipboard or, if the 'Shift' key is pressed, the complete Scope window.
Persistence Display 	Enabled: Data from all acquired sweeps of the Series are displayed, i.e., multiple sweeps are displayed. Disabled: Previous sweeps are cleared, and the newest acquired sweep is displayed.
Sweep # of #	The active sweep number vs. the total number of configured sweeps is reported. If multiple cycles are set, the active sweep cycle number is inserted between them.

Table 4-3. Other Scope Buttons

<u>Right-click Menus</u>

X-Axis	Autoscale All Axes	Scale all signals Y-axes to their data, and set all signals X-axes to their full-scale range.
	Full-scale X Axis	Set all signals X-axes to their full-scale range.

Axis Properties	Modify the axes style and components.
Y-Axis	Autoscale All Axes Scale all signals Y-axes to their data, and set all signals X-axes to their full-scale range.
Continuous Autoscale Axis	Continuously scale the signal's Y-axis to its data.
Autoscale Axis	Scale the signal's Y-axis to its data.
Full scale Y Axis	Set the signal's Y-axis to its full-scale range.
Axis Properties	Modify the axis style and components.
Hide Signal <name>	Hide the selected signal in the Scope.
Show Signal <name> Only	Show the selected signal in the Scope, hide all other signals.
Stack All Signals	Display all signals in a single column.

Main window area

To display the limited data modification menu, right-click in the blank area in a signal pane.

Note: If you click too close to the signal data, the full data modification menu displays instead. If you are having this issue, click near a horizontal or vertical edge of the signal pane.

Autoscale All Axes	Scale all signals Y-axes to their data, and set all signals X-axes to their full-scale range.
Add Annotation	Add a floating text-box label to the signal pane.
	To edit or delete an annotation, double-click on it.
Parametric Plot	Open a separate graph window of X vs. Y input signals.
Y-signal	Select an input signal for the Y-axis.
X-signal	Select an input signal for the X-axis.
Plot	Update the plot using the new parameters.
Copy to Layout	Copy the Parametric Plot graph into a new Layout window, or append to an existing Layout page.

Copy to Clipboard — Copy the Parametric Plot graph to the system clipboard, or if the 'Shift' key is pressed, the complete Scope window.

Time Range — The time range of the data to be plotted.

- Full Trace — Use the entire trace for the time range.
- Sweep Time — Set relative to the start time of a sweep (time zero).
  - Start Time — Set the starting time.

    Once the Start Time is within 2 ms of the End Time, further Start Time increments will increase the End Time by the same amount.
  - End Time — Set the ending time.

    Once the End Time is within 2 ms of the Start Time, the End Time cannot be decremented.
- Segment Time — Set the time range as a ratio of the Segment duration.
  - Segment — Select the Segment number.
  - Start Ratio — [0 = beginning of Segment]

    Set the starting time ratio.
  - End Ratio — [1 = end of Segment]

    Set the ending time ratio.

Export Graphics — Copy the signal and open in a separate window.

Saves to Windows / Graph Macros.

Toggle Cursor Info — Show/Hide the Cursor Info pane to measure X-Y data points or set a fitting range. (See the Signal Data section below.)

Colors — Adjust the colors used by the active signal pane:

- graph background — The background of the pane.
- all axes — The X- and Y-axis areas.

all grids	The grid lines in the pane.
all tick labels	The tick labels in the X- and Y-axis areas.
all axis labels	The axis labels in the X- and Y-axis areas.

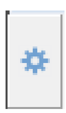
Hide Signal '<name>'	Hide the selected signal in the Scope.
Show Signal '<name>' Only	Show the selected signal,in the Scope, and hide all other signals.
Stack All Signals	Display all signals in a stacked signal layout.

Signal data

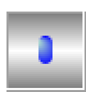
To display the full data modification menu displays with numerous options to modify sweeps and data points, such as marker symbols and lines.

The extra SutterPatch entries include most of those from the right-click main window menu (see above), such as:

Toggle Cursor Info	Show/Hide the Cursor Info pane to manually measure X-Y data values, or to set a fitting range.

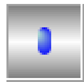


Options menu

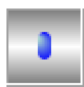
-  One Mover Moves All — Draggable cursor mover tool moves all cursors together with a single control.
- All Styles — Change the cursor symbol style.
- Show Cursor Pairs — Display up to 5 sets of cursor symbol pairs.

Cursor A

	Cursor symbol for data point 'A'.
A:	Symbol letter (beginning cursor of the pair).
R1_A_IV	(Default) wave name.

	Draggable cursor mover tool for the cursor pair.
pnt:	Data point number (starting from zero).
X:	X-axis value of data point ‘A’
Y:	Y-axis value of data point ‘A’
ΔY	Difference of the cursor pair Y values.

<u>Cursor B</u>

⊠	Cursor symbol for data point ‘B’.
B:	Symbol letter (ending cursor of the pair).
R1_A_IV	(Default) wave name.
	Draggable cursor mover tool for the cursor pair.
pnt:	Data point number (starting from zero).
X:	X-axis value of data point ‘B’
Y:	Y-axis value of data point ‘B’
ΔX	Difference of the cursor pair X values.

<u>Cursor Instructions</u>

1. Click on symbol ‘A’ to enable it.
2. Manually drag the highlighted symbol onto a data point in the Scope, or enter the data point number in the ‘pnt’ field.
3. Click on symbol ‘B’ to enable it.
4. Manually drag the highlighted symbol onto a data point in the Scope, or enter the data point number in the ‘pnt’ field.

   X- and Y-measurements are displayed for the cursor pair data points.
5. Fitting can also be applied to the cursor pair data. Right-click on the data, and select ‘Quick Fit’ for a list of built-in Igor fitting functions.

   The fit is displayed in the graph, and the fitting information is written to the Command window.

Marquee — Click and drag the mouse to surround a region of interest:

Expand	Set the signal's Y-axis range from the marquee vertical data limits, and set all signals X-axes ranges from the marquee horizontal data limits.
Horiz Expand	Set all signals X-axes ranges from the marquee horizontal data limits.
Vert Expand	Set the signal's Y-axis range from the marquee vertical data limits.
Shrink	Move the signal's Y-axis current limits to the position of the marquee vertical data limits, and move all signals X-axes current limits to the position of the marquee horizontal data limits.
Horiz Shrink	Move all signals X-axes current limits to the position of the marquee horizontal data limits.
Vert Shrink	Move the signal's Y-axis current limits to the position of the marquee vertical data limits.
Extract Template	Copy the last sweep to the Template Editor.

4.1.10 Solution Editor

Control perfusion systems with named solution control settings.

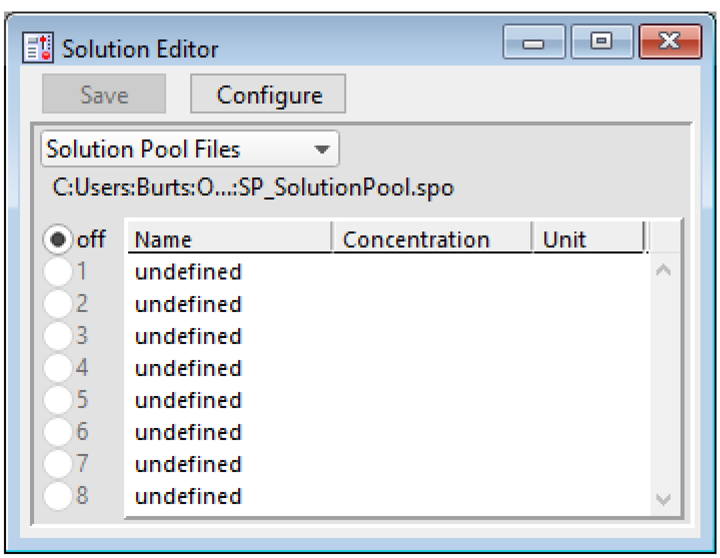


Figure 4-58. Solution Editor

You can create a named list of Solutions control "valves" to manage your commands to physical valves and solution changers.

Save	Save Solution Editor changes to the current Solution Pool file (*.spo). This button becomes active once a Solution is selected for editing.
Configure	Open the Configure Solutions dialog to categorize solution types and configure output channels.

Solution Pool Files   A Solution Pool file (*.spo) can contain multiple defined Solutions.

New Solution Pool	Create a blank Solution Pool.
Load Solution Pool	Load the Solutions of a previously saved Solution Pool file into the Solution Pool.
Revert to Last Saved	Undo any unsaved changes to the Solution Pool.
Save Solution Pool	Save the Solution Pool using its existing file name and path.
Save Solution Pool As	Save the Solution Pool to a new file, and switch to the new file.
Save Solution Pool Copy	Save the Solution Pool to a new file, but do not switch to the new file.

Note: Default file names are auto-incremented from the previously loaded Solution Pool name.

The file path and file name of the loaded Solution Pool file is displayed.

[Off, 1 – #]	Manually select a "valve" radio button to activate its corresponding Solution configuration. A radio button is available for selection (ungrayed) when its name is changed from 'undefined'. The number of radio buttons is set by the number of "Configured" solutions with outputs. Only one "valve" can be active at a time.
Name	Double-click on a field to edit it; click-and-drag to move it up or down in the table.
Concentration	Enter a concentration value for the solution.

Tip: You can access the concentration value from the last-used 'Test Compound' solution valve in SutterPatch fields that accept the Special Identifier 'Solutions'.

Unit	The unit type of the concentration.

Configure Hardware Controls

 Close Dialog button.

[# Solutions] Set the number of Solutions to configure. Changing this number creates a new solution pool.

[4, 8, 12, 16, 20, 24]

Loading other Solution Pool files allows a virtually unlimited number of Solutions to be accessed in an Experiment.

Description A text note for the Solution.

[Solution Type list] A list of predefined solution types.

- Initial Condition
- Washout
- Control
- Test Compound
- Not a Solution

[Output Channel] Select a physical output channel and set its value.

• No Output		
• AuxOUT1	[±10.000 V]	Analog output voltage.
• AuxOUT2	[±10.000 V]	Analog output voltage.
• DigOUT Word	[0 – 255]	Decimal value of an 8-bit digital word.
• DigOUT1 – 8		A single digital bit is set “high”.

4.1.11 Template Editor

Templates allow any data waveform or portion of an existing data wave to be incorporated into a command waveform. The Template Editor can manage and manipulate such templates.

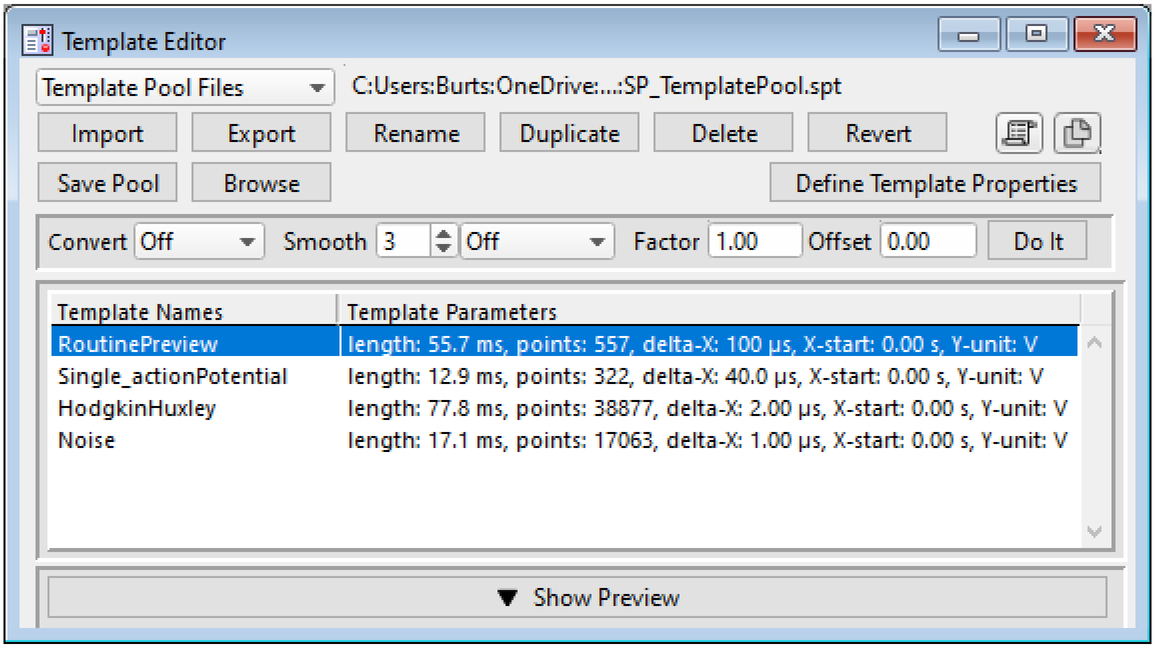


Figure 4-59. Template Editor

Template Pool Files A Template Pool file (*.spt) can contain multiple defined Templates.

New Template Pool	Create a blank Template Pool
Load Template Pool	Load the Templates of a previously saved Template Pool
Revert to Last Saved	Undo any unsaved changes to the Template Pool
Save Template Pool	Save the Template Pool using its existing file name and path
Save Template Pool As	Save the Template Pool to a new file, and switch to the new file. The default file name is the same as the original file name.
Save Template Pool Copy	Save the Template Pool to a new file, but do not switch to the new file. The default file name increments.
Merge Template Pools	Insert the Template from a previously saved Template Pool file into the loaded Template Pool

The file path and file name of the loaded Template Pool file is displayed.

Import

Select a template file (*.ibw).

Alternatively, in a Scope window or preview pane, click and drag the mouse to surround a region of interest with a bounding box (the “marquee”). Right-click in the box and select ‘Extract Template’. A template with the signal name is added to the template list.

An extracted template is composed of a single sweep:

- Scope window (Acquisition): last sweep
- Scope window (Analysis): selected sweep
- Preview pane: last or selected sweep

Note: ‘Extract Template’ is not implemented for graphs or the Data Navigator preview pane. Also, it is only valid with monotonically increasing or decreasing X-axes.

Export

Export the selected template to a 1-D Igor wave file (*.ibw).

To export a portion of a sweep, select the region of interest with the mouse, and use the marquee ‘Extract Template’ right-click command. The new wave can now be exported.

Rename

Edit the name of the selected template. Allowable characters are A-Z, a-z, 0-9, and “_”. Special characters are not allowed; spaces are replaced by an underscore.

Duplicate

Add a copy of the selected template to the list. The new template name’s number is appended or incremented.

Delete

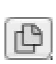
Remove the selected template from the list.

Revert

Discard any unsaved changes to the selected template.

 Copy to Layout

Copy the selected template graph into a new Layout window, or append to an existing Layout page.

 Copy to Clipboard

Copy the selected template graph to the system clipboard.

Save Pool

Save the template pool using its existing file name.

Browse	Create a template from the Experiment data in the Data Browser.
Define Template Properties	Update a data wave's X- and Y-axis parameters to be compatible with SutterPatch templates.
Enter X-increment	The data point time interval is changed, which also adjusts the length of the trace.
Enter X-start	The X-axis starting time for the data.
Enter Y-unit	The Y-axis base unit (enclose in double quotes.)
Convert	The data is interpolated to match the new sampling rate. While the number of samples is updated, the length of the trace is unchanged.
Smooth	Apply smoothing to the template. • Off • Boxcar — A fast time-domain filter with excellent 0 – 100% rise-time response. • Gaussian — A standard filter with excellent 10 – 90% rise-time response.
Factor	Adjust the template scaling factor. Values are displayed with SI unit prefixes.
Offset	Adjust the template offset. Values are displayed with SI unit prefixes.
Do It	Apply the adjustments to the template parameters.
Template Names	A list of the loaded templates.

Click on a Template entry to make it the active one.

Double-click on a Template Name to rename it.

Click-and-drag a Template entry to reposition it in the list.

Template Parameters	Parameter settings description.
Show/Hide Preview	Display / Hide a preview pane with the selected template.

The preview pane X- and Y-axes can be controlled in two ways:

- Hover the mouse over an axis line until the cursor turns into a double-headed arrow, then scroll up or down to contract / expand the axis.
- In the preview, click and drag the mouse to surround the region of interest with a bounding box (the "marquee"). Right-click in the box and select one of the expand / shrink options.

To measure X-Y data points or set a fitting range, select 'Toggle Cursor Info' from the right-click menu. (See 'Right-Click Menus' for Scope windows.)

4.2 Data Analysis

Online data analysis is configured in the Routine Editor with measurement settings. These same settings are also available during offline data reanalysis.

For complete flexibility in controlling how analyses are performed, use a Paradigm Execute step to run virtually any SutterPatch command, Igor analysis or user-defined function.

General analyses include:

- Analysis Editor
- Equation Editor
- Igor Analyses
- Metadata Review
- Paradigm Editor
- Routine Editor
- Scope / Reanalysis Measurements

Specialized analyses are available via the reanalysis Scope or the Data Navigator:

- Action Potential Analysis
- Single-Channel Analysis
- Synaptic Analysis

4.2.1 Action Potential Analysis

Action potentials (APs) are analyzed with this offline dialog. Access via the Scope (analysis) window ‘Measurements’ button, or the Data Navigator (signal) ‘Available actions’ menu.

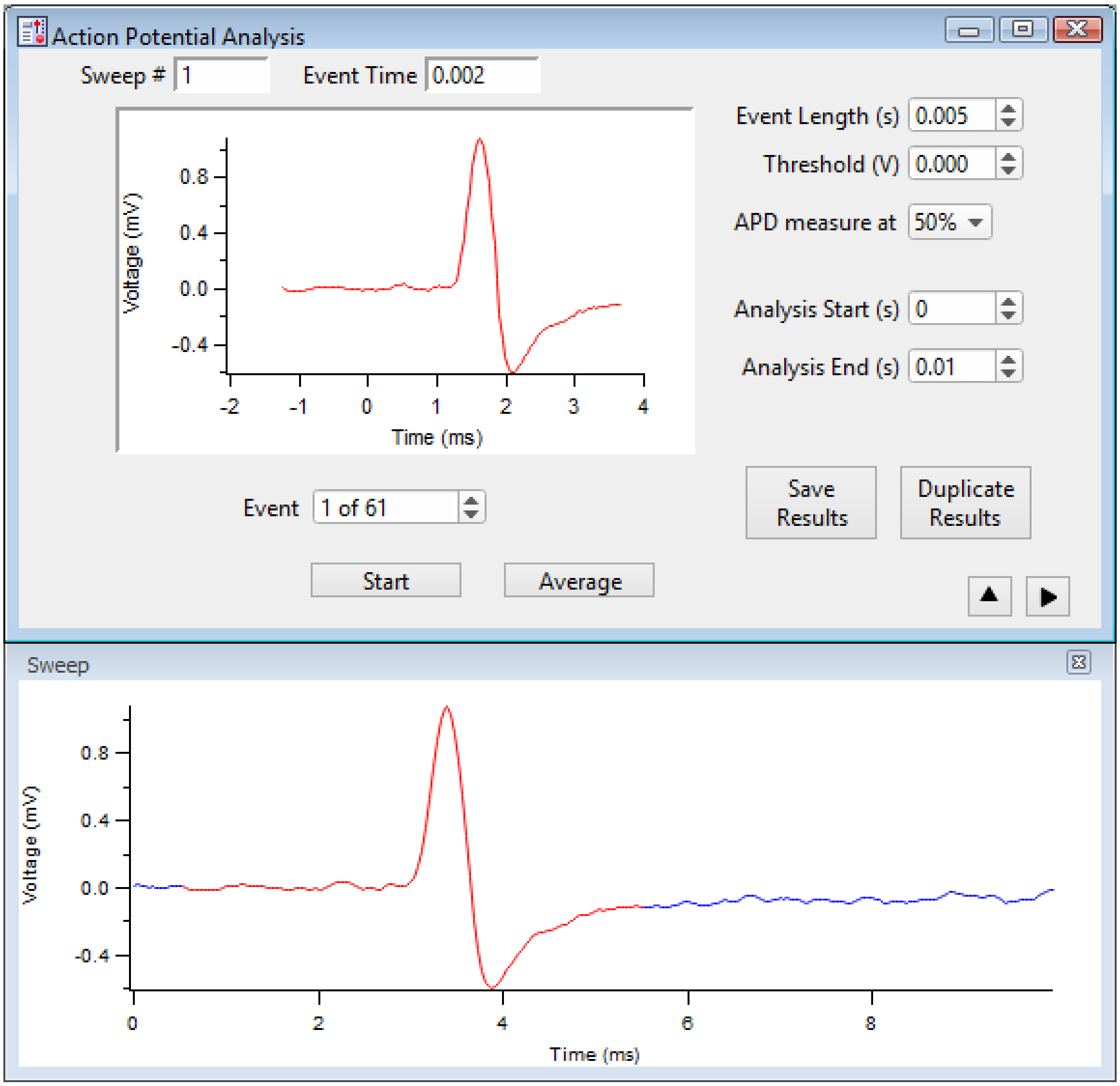


Figure 4-60. Action Potential Analysis

Sweep # — The sweep number of the selected action potential Event.

When set to ‘0’, this indicates that averaged Results measurements are being displayed.

Select sweeps for processing by “marking” them in the Data Navigator tree (or during Scope acquisition or reanalysis.)

Event Time — Time point when the potential of the selected Event crosses the threshold.

[Event pane]	A graph of the selected Event, with the X-axis zero point reset to the Threshold point. To measure X-Y data points or set a fitting range, select 'Toggle Cursor Info' from the right-click menu. (See 'Right-Click Menus' for Scope windows.)
Event [# of #]	[Event number vs. total Events] Cycle through the analyzed Events in the Event pane, with the Event displays in red in the 'Sweep' pane.
or	
Sweep [#]	Cycle through the unanalyzed sweeps in the 'Sweep' pane.
[Sweep pane]	The graph of a selected sweep, or a sweep containing a selected Event (red). To measure X-Y data points or set a fitting range, select 'Toggle Cursor Info' from the right-click menu. (See 'Right-Click Menus' for Scope windows.)
Start	Run the Action Potential Analysis and display the Results pane.
Average	Display the averaged Event in the Event pane, and its measurements in the Results panel. The 'Sweep #' is set to '0'.
Event Length (s)	The Event duration in the Event pane; displays in red in the Sweep pane.
Threshold (V)	[±0.1000] This voltage level needs to be reached or exceeded for analysis of an Event to be triggered.
APD measure at:	Set the Action Potential Duration percentile. Measures the duration of an Event at a percentile of the Event's repolarization amplitude. <u>[20 / 30 / 40 / 50 / 60 / 70 / 80 / 90 / 100 %]</u>
Analysis Start (s)	[0+] Set the start time of the Sweep data to be analyzed.
Analysis End (s)	Set the end time of the Sweep data to be analyzed.
Save Results	Results are displayed in a table and a Layout window; prior results are overwritten.

A 'Results' table of all Events automatically opens:

[ ]	Row number, one row per Event.
Sweep Number	Sweep number the Event is in.
Event Time (s)	Time point of the Event start.
Threshold (V)	Amplitude of the Event threshold.
Threshold Time (s)	Time point of the "trigger" threshold time.
Peak (V)	Amplitude of the Event peak.
Peak Time (s)	Time point of the Event peak.
AP Duration (s)	Duration of the action potential at the AP repolarization percentile.
AHP (V)	Peak amplitude of the 'After Hyper-Polarization' phase.
AHP Time (s)	Time point of 'After Hyper-Polarization; the Event re-crosses the threshold amplitude at this time.

An 'Action Potential Analysis Results' report automatically opens in a special Layout window:

Signal Pathname:	The Igor Pro experiment pathname for the analyzed signal.
Analysis Prefix:	The prefix for the signal's analysis objects in the Igor Pro Data:Analysis' folder.
Total time analyzed =	Includes the Start / End times for all analyzed sweeps. [s]
Number of events detected =	Total number of Events found.
Event Frequency =	The average frequency of the found Events. [Hz]
All Sweeps analyzed or Sweeps analyzed:	Every sweep was analyzed. [list of analyzed sweep #s]
[Event graph]	A graph of the averaged Event. [V vs. s]

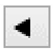
[Phase plot] A graph of the phase plot for visual inspection of the derivatives.

[dV/dt (V/s) vs. V]

Duplicate Results Results are copied to a new Results table and a new Layout window.

 Show/Hide the Sweep pane (below).

Display a graph of the sweep containing the selected Event.

 Show/Hide the Results pane (on the right).

Display the Action Potential Measurements results.

Results pane Measurement results are displayed for the Event selected in the main window.

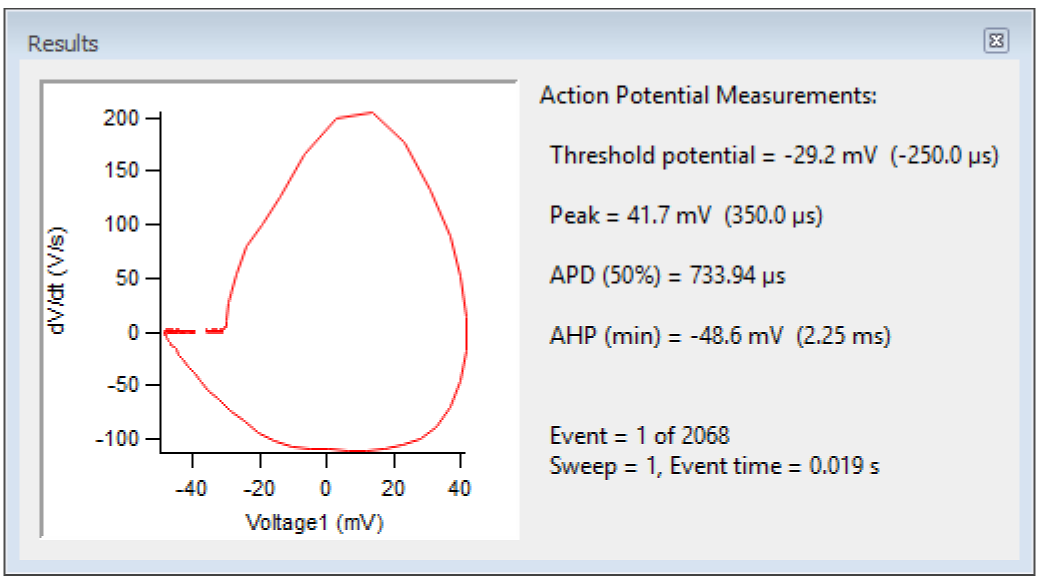


Figure 4-61. Action Potential Measurements

Phase plot A graph of the phase plot for visual inspection of the derivatives.

[dV/dt (V/s) vs. V] Y-axis vs. X-axis.

To measure X-Y data points or set a fitting range, select 'Toggle Cursor Info' from the right-click menu. (See 'Right-Click Menus' for Scope windows.)

Threshold potential = [V (s)]

Event starting amplitude (time from Threshold setting).

The actual biological start of the selected Event - the Event's actual starting amplitude, and its timepoint relative to the 'Threshold' setting timepoint.

See the Algorithms appendix for more information.

Peak = [V (s)]

The largest amplitude excursion of the Event (and the timepoint of the 'Threshold' setting)

Two measures of an Event's peak amplitude are reported, voltage and time. Time is reported relative to the 'Threshold' timepoint.

APD (%) = [%, s]

Action Potential Duration of the Event at (n %) of amplitude repolarization.

AHP (min) = [V (s)]

The largest amplitude excursion of the "After HyperPolarization" phase of the Event (time from 'Threshold' setting)

Two measures of the AHP amplitude are reported, voltage and time. AHP is when the action potential repolarization phase drops to its lowest point below the resting membrane potential, i.e. during the hyperpolarized refractory period of the cell.

Event = The analyzed Event (of the total number of Events) found in the data.

Sweep = The sweep number of the analyzed Event.

Event time = The sweep time of the start of the analyzed Event.

Events found = The number of averaged Events.

Event frequency = The average frequency of the found Events.

4.2.2 Analysis Editor

View and manipulate the data in your Experiment's various analyses and graphs.

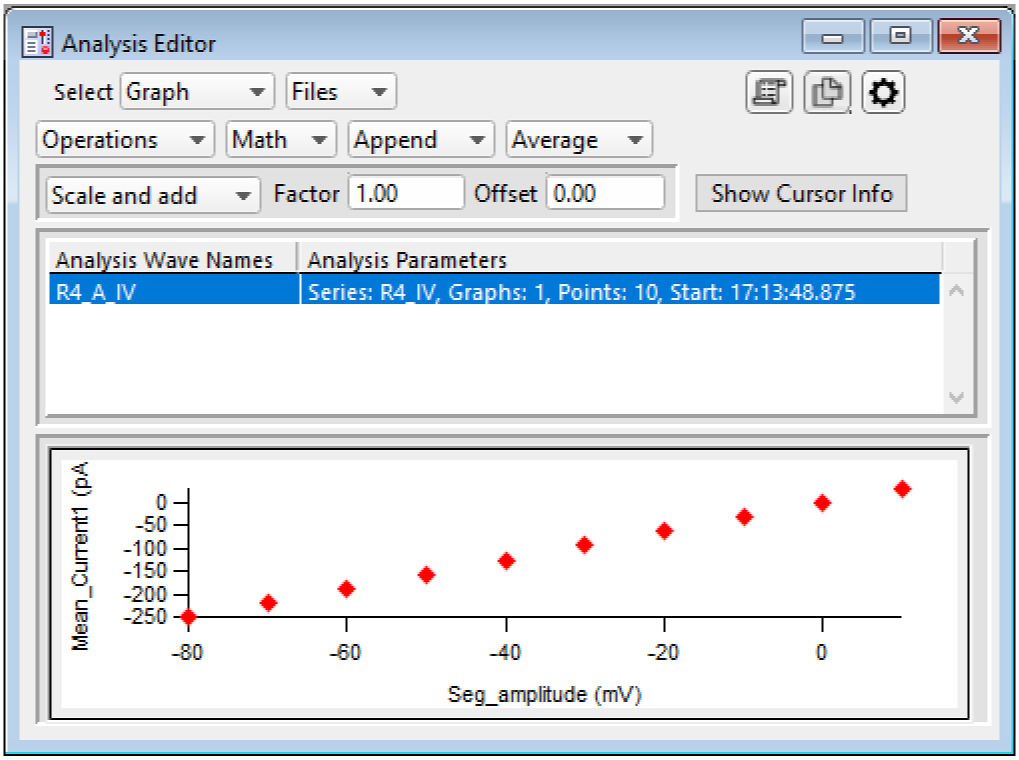


Figure 4-62. Analysis Editor

Select — Choose how to view the data:

Table — View a spreadsheet of the Analysis data

Column 1: Row number.

Column 2: X-data for the first graph.

Column 3: Y-data for the first graph.

Column m – n:

Pairs of X- and Y-data columns repeat for each additional graph.

The first graph (X-Y-pair) that uses X-axis time units populates its X-data column with time values; subsequent graphs that use X-axis time units have blank X-data columns, as they use the first time-column created.

Note: The first data point is in row 0, so the last data point is in row [N – 1].

Row N is a blank row that contains grayed-out cells. It is used to manually add extra rows of data to the table. Once a number is entered into one of these cells, the blank (gray) row is automatically moved to the row beneath it.

Warning! Editing the table will permanently alter its data.

Graph-[1 – 8] — Select an Analysis graph for the selected data wave. The graph number refers to its original Analysis window position.

Files — Import or export an analysis graph file.

Export Graphs	Save the entire graph as a multi-dimensional Igor Binary Wave file (*.ibw).
Export Graph X-column	Save the X-column data, including labels, as a one-dimensional Igor Binary Wave file (*.ibw).
Export Graph Y-column	Save the Y-column data, including labels, as a one-dimensional Igor Binary Wave file (*.ibw).
Import Graphs	Open and display a saved graph.

Note: Import of one-dimensional Igor Binary Wave files (*.ibw) is not supported.

Copy to Layout — Copy the selected analysis graph or table into a new Layout window, or append to an existing Layout page.

Copy to Clipboard — Copy the selected analysis graph or table to the system clipboard.

Options

Show Axes Color	Display a background color for the axes.
Show Grid	Display X & Y grid lines in the graph.
Show Error Bars	Display SEM error bars for averaged data.
Show Markers	Display data points with marker symbols.
Show Lines	Display a line between data points. (Toggle between 'markers' and 'lines', or both.)
Cell Separator: TAB	Use tab separators when exporting a table (e.g. to Excel).
Cell Separator: Comma	Use comma separators when exporting a table (e.g. to MS-Works).
Include Column Labels	Column labels appear on the first line of an exported table.

Operations	Duplicate	Insert a copy below the highlighted item.
	Delete Graph or Table	Delete the entire analysis wave.
	Delete Single Graph	Delete the selected graph.
	Note:	If an analysis cannot be deleted, it likely exists in another Graph window or Layout page - first close the other analysis instance via menu items Windows / Graphs, or Windows / Layouts, or Windows / Layout Macros.
Math	Normalize: zero to maximum	Rescale the data whereby the largest absolute point is set to 1.0 (or -1.0), while the zero point is maintained in, or relative to, the data range.
	Normalize: minimum to maximum	Rescale the data whereby the smallest point is set to zero and the largest point is set to 1.0.
	Invert	Reverse the Y-axis sign of the data points.

Append — Select an analysis wave for appending with the loaded wave. Time-course data are plotted relative to the loaded analysis wave's "time zero".

Average — Select an analysis wave to be averaged with the loaded wave. A weighted average is performed, i.e., the number of data sets is accounted for when averaging in new data.

Two new entries are inserted into the wave list after the loaded wave:

1. The averaged wave.
2. The SEM (Standard Error of Means) data points wave.

If Options / Show Error Bars is enabled, the SEM data are used to display error bars in the corresponding averaged data graph.

(See Appendix F: SutterPatch Algorithms for the SEM algorithm.)

Scale and add — Use to combine available analysis waves, with optional scaling and offset applied.

When the Factor is '1.00' and the Offset is '0.00', this operation will simply add the selected wave to the displayed wave.

Factor — Set a scaling factor for a data wave that will be added to the displayed data.

Values are displayed with SI unit prefixes.

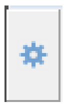
To subtract a data wave, change the Factor to a negative number.

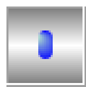
Offset — Set an offset for a data wave that will be added to the displayed data.

Values are displayed with SI unit prefixes.

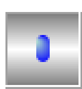
Show/Hide Cursor Info

Open the Cursor Info pane to manually measure X-Y data values, or to set a fitting range.

 Options menu

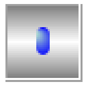
-  One Mover Moves All — Draggable cursor mover tool – move all cursors together with a single control
- All Styles — Change the cursor symbol style.
- Show Cursor Pairs — Display up to 5 sets of cursor symbol pairs.

Cursor A

	Cursor symbol for data point ‘A’.
A:	Symbol letter (beginning cursor of the pair).
R1_A_IV	(Default) wave name.
	Draggable cursor mover tool for the cursor pair.
pnt:	Data point number (starting from zero).
X:	X-axis value of data point ‘A’
Y:	Y-axis value of data point ‘A’
ΔY	Difference of the cursor pair Y values.

Cursor B

	Cursor symbol for data point ‘B’.
B:	Symbol letter (ending cursor of the pair).
R1_A_IV	(Default) wave name.

 Draggable cursor mover tool for the cursor pair.

pnt: Data point number (starting from zero).

X: X-axis value of data point ‘B’

Y: Y-axis value of data point ‘B’

ΔX Difference of the cursor pair X values.

<u>Cursor Instructions</u>

1. Click on symbol A to enable it.
2. Manually drag the highlighted symbol onto a data point in the graph, or enter the data point number in the ‘pnt’ field.
3. Click on symbol B to enable it.
4. Manually drag the highlighted symbol onto a data point in the graph, or enter the data point number in the ‘pnt’ field.

   X- and Y-measurements are displayed for the cursor pair data.
5. Fitting can also be applied to the cursor pair data. Right-click in the graph, and select ‘Quick Fit’ for a list of built-in Igor fitting functions.

   The fit is displayed in the graph, and the fitting information is written to the Command window.

Analysis Wave Names — Loaded analyses available for manipulation.

Analysis Parameters

- Series: R#_ — Name of the Series.
- Graphs: # — Number of graphs in the analysis wave.
- Points: # — Number of data points in the graph.
- Start: # — Start time of analysis wave.
- Average: # — Number of graphs averaged or appended.

Tip: If the Analysis Parameters text is not fully visible, increase the width of the Analysis Editor window.

[Graph & Table pane] — Data point markers are plotted, or a numeric table is displayed.

X- and Y-axes can be magnified to be larger or smaller. Place the mouse cursor in the axis ticks region, then scroll the mouse wheel up or down. The axis ticks region does not include the tick label (numbers) area.

The marquee tool is also supported in the Graph pane. Click and drag a bounding box around the region of interest, then right-click in it for magnification options.

To measure X-Y data points or set a fitting range, select 'ToggleCursor Info' from the right-click context menu. (See 'Show/Hide Cursor Info' above.)

4.2.3 Analysis Window

Scope measurements are plotted in an Analysis window docked on the right side of the Scope window. An Analysis window can be resized or closed, but not undocked from the Scope window.

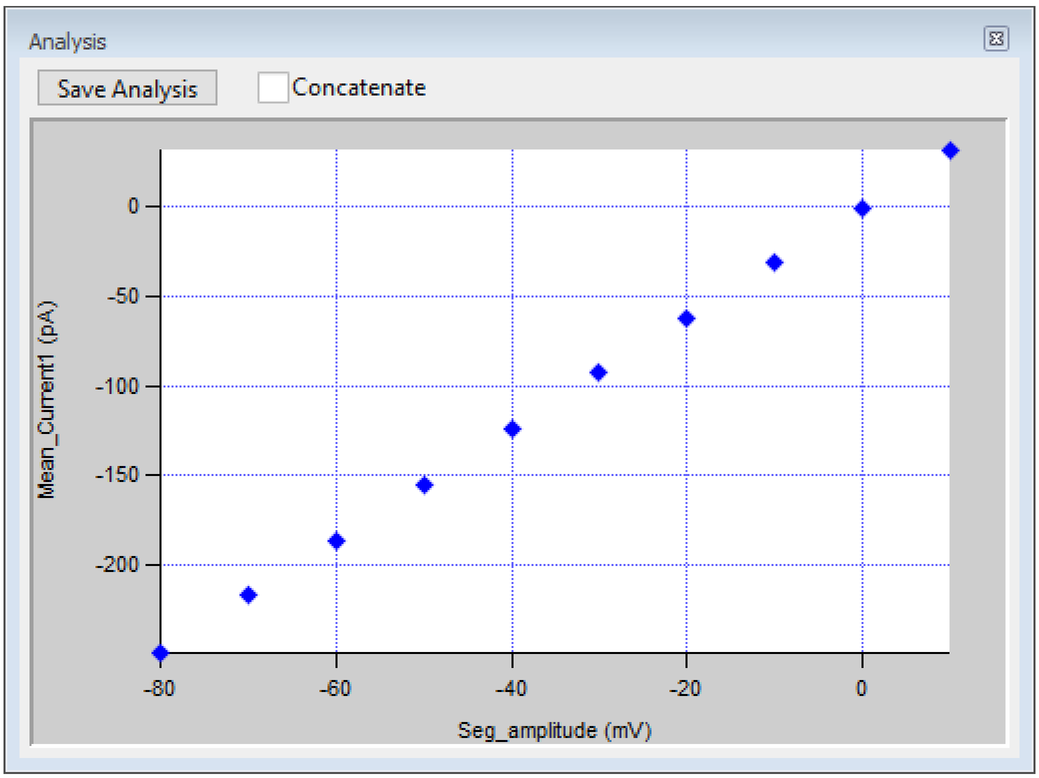


Figure 4-63. Analysis Window

Online measurements are plotted during data acquisition in real-time. Online analysis measurements are configured in the Routine Editor 'Real Time Measurements & Graphs' section. A separate pane is created in the Analysis window for each enabled Measurement graph.

Data can be selected for offline review or analysis via the Data Navigator (or 'View Last' Dashboard button.) When stored data are rerun for analysis, the data displays in a "reanalysis" Scope window, and the measurements are graphed in the accompanying Analysis window. The last measurements applied to the data are automatically used to reanalyze the data.

Save Analysis This button saves the displayed analyses with the Experiment.

Saved analyses are viewable in the Analysis Editor (or also the Data / Data Browser 'Data: Analysis' folder.

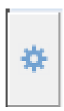
Concatenate — Append new measurements to the existing measurements in the graph. [when docked to the acquisition Scope]

Clear — Erase all measurements from the graph display.

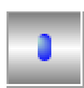
[Graph panes] — The graph pane X and Y-axes can be magnified to be larger or smaller. Place the mouse cursor in the axis ticks region (do not include the tick labels or numbers), then scroll the mouse wheel up or down.

The marquee tool is also supported in the Graph pane. Click and drag a bounding box around the region of interest, then right-click in it for magnification options.

To manually measure X-Y data values, or to set a fitting range, right-click in the graph to display a data modification menu and select 'Toggle Cursor Info'. The 'Cursor Info' pane displays:



'Options' menu

-  One Mover Moves All

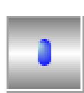
  Draggable cursor mover tool - move all cursors together with a single control.
- All Styles — Change the cursor symbol style.
- Show Cursor Pairs — Display up to 5 sets of cursor symbol pairs.

Cursor A

 Cursor symbol for data point 'A'.

A: Symbol letter (beginning cursor of the A/B pair).

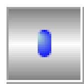
R1_A_IV (Default) wave name.

 Draggable cursor mover tool for the cursor pair.

pnt: Data point number (starting from zero).

X:	X-axis value of data point 'A'
Y:	Y-axis value of data point 'A'
ΔY	Difference of the cursor pair 'Y' values.

Cursor B

	Cursor symbol for data point 'B'.
B:	Symbol letter (ending cursor of the A/B pair).
R1_A_IV	(Default) wave name.
	Draggable cursor mover tool for the cursor pair.
pnt:	Data point number (starting from zero).
X:	X-axis value of data point 'B'
Y:	Y-axis value of data point 'B'
ΔX	Difference of the cursor pair 'X' values.

Cursor Instructions

1. Click on symbol A to enable it.
2. Manually drag the highlighted symbol onto a data point in the graph, or enter the data point number in the 'pnt' field.
3. Click on symbol B to enable it.
4. Manually drag the highlighted symbol onto a data point in the graph, or enter the data point number in the 'pnt' field.

   X- and Y-measurements are displayed for the cursor pair data.
5. Fitting can also be applied to the cursor pair data. Right-click in the graph and select 'Quick Fit' for a list of built-in Igor fitting functions.

   The fit is displayed in the graph, and the fitting information is written to the Command windows.

4.2.4 Data Browser

The Data Browser can be used to access and display all of the Experiment's data objects, such as data waves, analysis graphs, layouts, images, metadata, Paradigms and Routines. Access it from the Data menu.

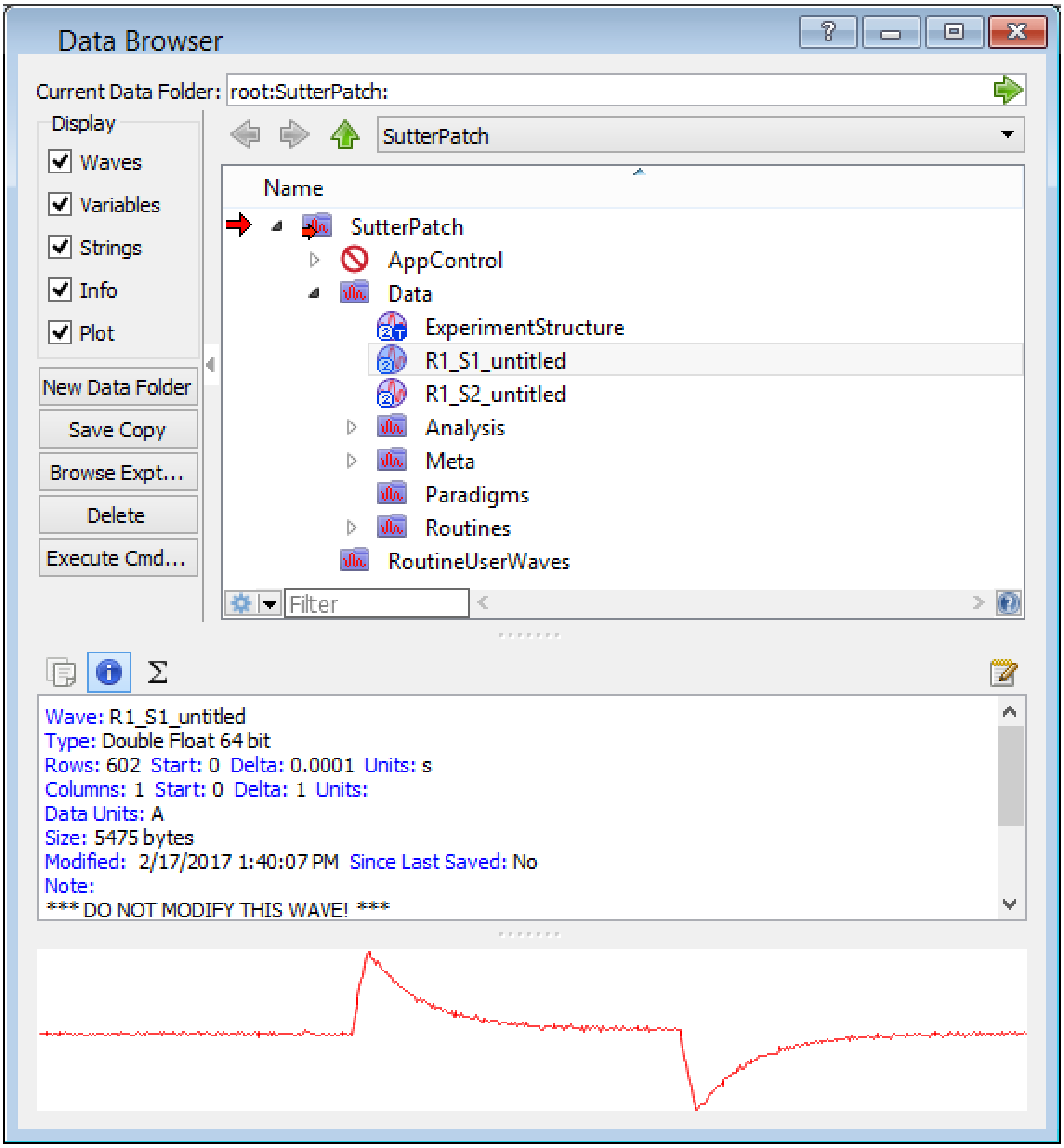


Figure 4-64. Data Browser

Objects are displayed in a tree structure, using a path "root" of 'SutterPatch'.

Warning! If this window is kept open during data acquisition, the Experiment can unexpectedly terminate!

Also, for the macOS, opening and closing windows can take a long time.

The ‘Data’ subfolder contains additional subfolders, followed by recorded data waves, arranged per Signal.

ExperimentStructure The sequence of Paradigms and Routines.

<Names of data Series…>

Analysis This folder contains data measurements, including results from fits, and Event tables.

- Wave names that include “_M_” contain the status of sweep marks. A marked sweep has a value of ‘1’, while an unmarked sweep has a value of ‘0’.
- Wave names that include “_A_” contain analysis measurements.
- Wave names that include “_df_” contain the differentiated average action potential (phase plot) waveform.

Images This folder contains stored images that display in the preview pane.

Meta This folder contains a table of general system metadata parameters (unformatted).

Routines This folder contains limited information on the used Routines.

Right-click Menu

Display Display the first sweep of the data in a visual graph.

Edit Display the Analysis data in a numerical table.

SutterPatch signal data are stored in two-dimensional data waves, with one column per trace, and one row per sample point.

Warning! Editing data here permanently alters the raw data. Modify at your own risk!

Copy Full Path Copy the object’s path to the clipboard. This is in relation to an internal (hidden) Igor data folder, not the computer’s file system. This path can be used by advanced Igor users in user functions and executable commands.

4.2.5 Data Navigator

The Data Navigator window organizes and displays all levels of data for the current Experiment.

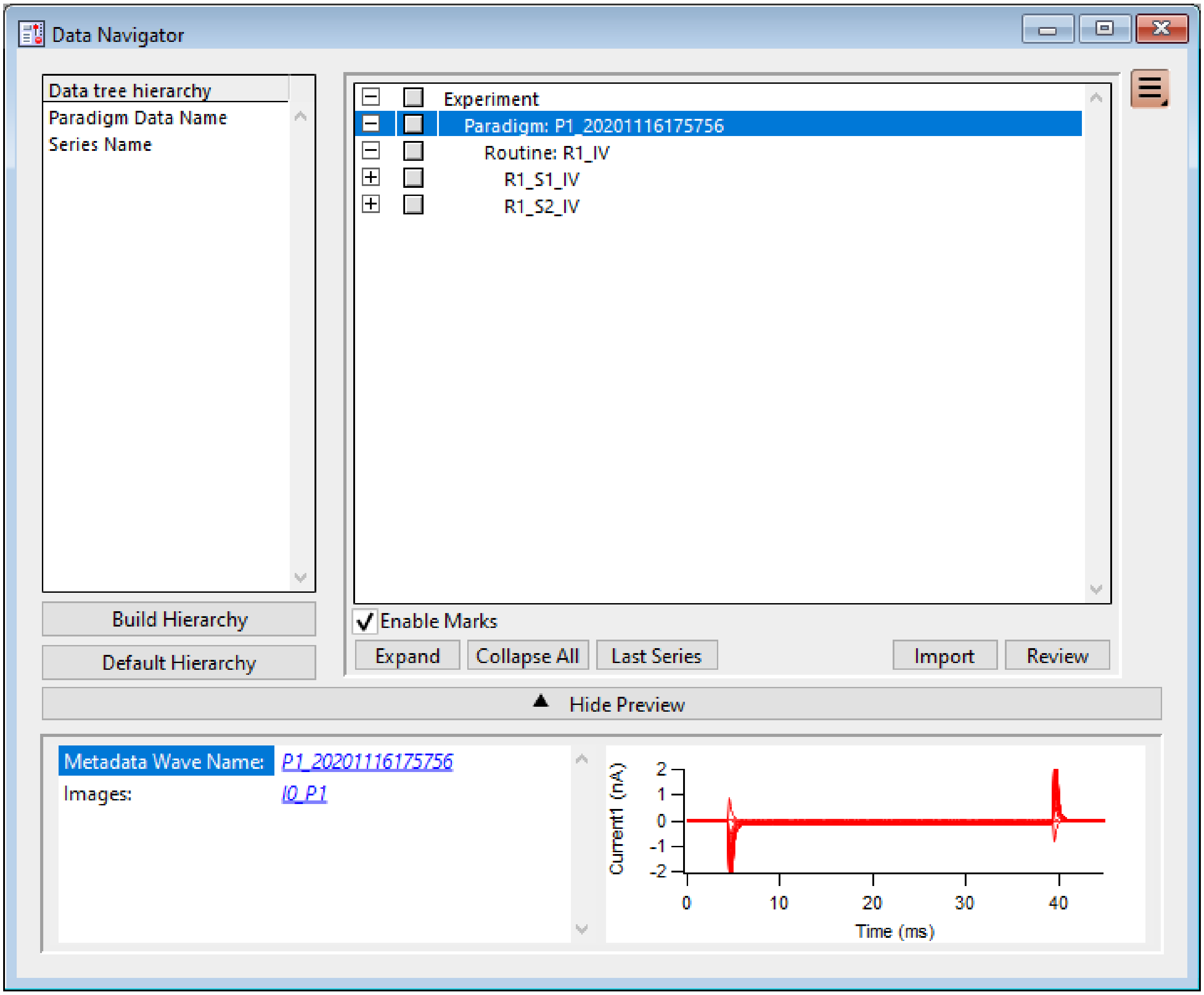


Figure 4-65. Data Navigator

Data tree hierarchy — The hierarchy of sorting levels is displayed in this pane.

Build Hierarchy — Re-organize the data tree using custom settings.

Select parameter group — Organize by metadata parameters:

[Parameter availability depends on the selected ‘Set Preferences / Metadata’ detail level.]

All Categories

Frequently Used

Experiment Hierarchy

Tag

Operator

Preparation - Animal

Preparation - Tissue

Preparation - Cell

Experiment

Amplifier

Instrumentation and Software

Electrode

Recording Solutions

Paradigm

Cell Health / Quality Control

Series (= Routine Data)

Data Acquisition Settings

Imaging

Stimulus

Available parameter	Click on a specific parameter from the selected group.
▶	Click on the “copy” button to insert the selected parameter above the highlighted level in the Hierarchy pane.
Hierarchy	The Hierarchy pane displays the new sorting hierarchy for the current Experiment.
Del ▲ ▼	The sorting levels can be re-organized by selecting an entry and clicking on the Up/Down keys to reposition it, or using the ‘Del’ key to remove it.
Do It	Click on the ‘Do It’ button to apply these changes to the data tree.

Default Hierarchy — Restore the default experimental hierarchy:
Paradigm > Routine (> Signal > Sweeps)

Data Tree Window

The current Experiment's data are arranged in the data tree (down to the Sweep level.)

Enable Marks

Allow multiple items to be marked for processing by the Available Actions menu:

- Action Potential Analysis (sweeps)
- Synaptic Event Analysis (sweeps)
- Average Selected Sweeps
- Display (signals)
- Export Data (all levels)

Sweeps "marked" during acquisition are loaded into the Data Navigator as "marked".

Marking / unmarking sweeps in a signal marks / unmarks those sweeps in all signals in the same Series.

Mark a range: highlight a marked starting selection, shift-click on the ending selection.

Clear a range: highlight an unmarked starting selection, shift-click on the ending selection.

Expand

All nodes of the data tree are expanded down to the Signal level.

To expand a node to the next lower level, with a mouse click on a "+" node, or double-click the node name. With a keyboard, use the up/down arrow keys to select a "+" node and press the space key.

Collapse All

All nodes of the data tree are collapsed up to the Paradigm level.

To collapse the lower levels of a node, with a mouse click on a "-" node, or double-click the node name. With a keyboard, use the up/down arrow keys to select a "-" node and press the space key.

Last Series

The last Routine's first signal is highlighted in the data tree and displayed in the Preview pane.

Import

Select a previously saved SutterPatch experiment to incorporate into the current experiment.

The same data set (experiment) cannot be imported twice.

When a Series name already exists in the Data Navigator, imported Series are renamed to avoid conflicts.

Review

Paradigm: Its signals are displayed in a Scope window as continuous data.

Note: This button changes to the Analyze button when a Routine, signal or sweep is selected.

Analyze

Routine, Signal, Sweep:

The Routine's signals are displayed in the Scope (analysis) window and Measurements analysis is run.

Note: This button changes to the Review button when a Paradigm is selected or marked.

Show Preview / Hide Preview

The displayed Preview pane items are based upon the selected hierarchy level.

Note: The Data Navigator Preview pane does not support mouse operations.

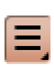
<u>Experiment</u>

Experiment Name:	The experiment file name.
HDF5 File Name:	For optional HDF5 files.
Paradigms:	Total number of Paradigms in the Experiment.
Routines	Total number of Routines in the Experiment.
Total data wave bytes:	Combined size of all data waves in the Experiment.

<u>Paradigm</u>

Metadata Wave Name:	Click to display the Paradigm's metadata.

	Images:	Open any saved images.
	[Preview sub-pane]	Displays the first signal of the first Routine.
<u>Routine</u>		
	Metadata Wave Name:	Click to display the Routine metadata.
	Signals:	Number of signals in the Routine.
	Sweeps:	Number of sweeps in the Routine.
	Routine Data Name:	Click to display the named Routine parameters.
	Analysis:	Click an analysis name to open it into the Analysis Editor.
	Images:	Open any saved images.
	[Preview sub-pane]	Displays the first signal of the selected Routine.
<u>Signal</u>		
	[Preview pane]	Displays the selected Signal.
<u>Sweep</u>		
	[Preview pane]	Displays the selected Sweep.

 Available Actions button

A menu lists various actions for the marked and/or highlighted data levels. These actions are also available via a right-click on the selected data level.

<u>Experiment</u>

Copy Signal Data Paths	Copy the Series internal Igor path to the system clipboard. (root:SutterPatch:Data:Series_name)

Export Data (See Preferences)	Export all marked data in the Experiment to file(s). Uses the 'Set Preferences / Data Export' options.
Mark All Sweeps	Mark all sweeps in the Experiment.
Unmark All Sweeps	Unmark all sweeps in the Experiment.

<u>Paradigm</u>

Review Paradigm Data	Display all Series (with all signals and sweeps) in the Paradigm. Each signal displays in a pane in a Paradigm Review window.
Note:	This action is not supported with the HDF5 file preference "Keep only one Sweep in Memory".
View Metadata	Display the Paradigm metadata in the Metadata Review sub-window, docked to the right of the Data Navigator window.
Copy Signal Data Paths	Copy the Series internal Igor path to the system clipboard. (root:SutterPatch:Data:Series_name)
Export Data (See Preferences)	Export the marked Paradigm data to file(s). Uses the 'Set Preferences / Data Export' options.

Discard Paradigm Data	Remove the marked Paradigms and their data from the Experiment. If the last Paradigm is discarded, when acquiring another Paradigm in the same

Experiment, the new Paradigm name will be incremented past the discarded Paradigm name.

Mark All Sweeps	All sweeps in the Paradigm are marked.
Unmark All Sweeps	Marks are removed from all sweeps in the Paradigm.

<u>Routine</u>

Analyze Routine Data	Display the marked sweeps of all signals in the highlighted Series in a Scope (analysis) window.
Review Routine Data	Display all signals (with all sweeps) from the highlighted Series. Each signal displays in a pane in a Routine Review window.
Note:	This action is not supported with the HDF5 file preference "Keep only one Sweep in Memory".
View Metadata	Display the highlighted Series' metadata in the Metadata Review sub-window docked to the right of the Data Navigator window.
View Routine Settings	Display the highlighted Series' settings and preview in the Routine Settings window.
Copy Signal Data Paths	Copy the Series internal Igor path to the system clipboard. (root:SutterPatch:Data:Series_name)
Show in Data Browser	Open Igor's Data Browser window to examine the highlighted Series' data waves.
Export Data (See Preferences)	Export the marked Series to file(s).

	Uses the 'Set Preferences / Data Export' options.

Discard Routine Data	Remove the highlighted Series data from the Experiment. If the last Series is discarded, when acquiring another Series in the same Experiment, the new Series name will be incremented past the discarded Series name.

Mark All Sweeps	All sweeps in the Series are marked.
Unmark All Sweeps	Marks are removed from all sweeps in the Series.

<u>Signal</u>

Action Potential Analysis	Analyze action potentials from the marked sweeps of the highlighted signal.
Single Channel Analysis	Analyze single-channel Events in the highlighted signal.
Synaptic Event Analysis	Analyze synaptic events (EPSPs, minis, etc.) from the marked sweeps of the highlighted signal
------------	
Analyze Routine Data	Display all signals in the Series, and their marked sweeps, in a Scope (analysis) window.
Edit Signal	Display all sweeps of the highlighted signal as numeric columns in an editable table.
Display Signal	Highlighted signal: display the marked sweeps in the signal in a graph window. Marked signal: display all sweeps in the signal in a graph window.

Average Marked Sweeps	Average the marked sweeps in the highlighted signal and display in the Analysis Editor.
View Metadata	Display the highlighted signal's metadata in a Metadata Review sub-window docked to the right of the Data Navigator window.
View Routine Settings	Display the Series parameters in the Routine Settings window.
Copy Signal Data Path	Copy the Series internal Igor path to the system clipboard. (root:SutterPatch:Data:Series_name)
Show in Data Browser	Open Igor's Data Browser window to examine the highlighted signal's data waves.
Export Data (See Preferences)	Highlighted signal: Export the marked sweeps in the signal. Marked signal: Export all sweeps in the signal. Uses the 'Set Preferences / Data Export' options.

Mark All Sweeps	All sweeps in the Series are marked.
Unmark All Sweeps	Marks are removed from all sweeps in the Series.

<u>Sweep</u>

Extract Sweep	Create a graph of the highlighted sweep in the Analysis Editor.
Display Sweep	Display the highlighted sweep in a graph window.

Export Data (See Preferences)	
	Export the highlighted sweep. (To export a portion of a sweep, use the Template Editor.) Uses the ‘Set Preferences / Data Export’ options.

Mark All Sweeps	All sweeps in the Series are marked.
Unmark All Sweeps	Marks are removed from all sweeps in the Series.

4.2.6 Data Table

The Data Table provides direct access to the sample points in a data Series, using a spreadsheet-style presentation.

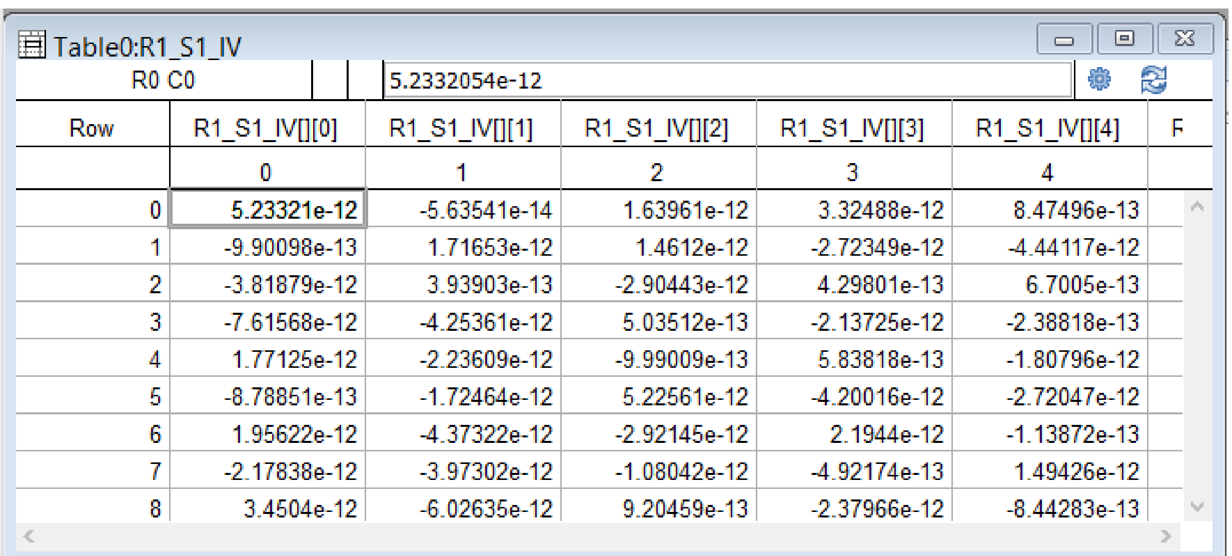


Table0:R1_S1_IV

R0 C0 5.2332054e-12

Row	R1_S1_IV[][0]	R1_S1_IV[][1]	R1_S1_IV[][2]	R1_S1_IV[][3]	R1_S1_IV[][4]
	0	1	2	3	4
0	5.23321e-12	-5.63541e-14	1.63961e-12	3.32488e-12	8.47496e-13
1	-9.90098e-13	1.71653e-12	1.4612e-12	-2.72349e-12	-4.44117e-12
2	-3.81879e-12	3.93903e-13	-2.90443e-12	4.29801e-13	6.7005e-13
3	-7.61568e-12	-4.25361e-12	5.03512e-13	-2.13725e-12	-2.38818e-13
4	1.77125e-12	-2.23609e-12	-9.99009e-13	5.83818e-13	-1.80796e-12
5	-8.78851e-13	-1.72464e-12	5.22561e-12	-4.20016e-12	-2.72047e-12
6	1.95622e-12	-4.37322e-12	-2.92145e-12	2.1944e-12	-1.13872e-13
7	-2.17838e-12	-3.97302e-12	-1.08042e-12	-4.92174e-13	1.49426e-12
8	3.4504e-12	-6.02635e-12	9.20459e-13	-2.37966e-12	-8.44283e-13

Figure 4-66. Data Table

Warning! Editing data permanently alters the raw data. Modify at your own risk!

Data Tables are accessed from the Data Navigator by highlighting a data Series, and selecting its Action menu ‘Edit’ command, or by right-clicking on it and selecting the ‘Edit’ command. From the Data / Data Browser, select a Series from the Data folder, then right-click the menu item ‘Display’.

To allow adding data to the table, the last row of data in the table is followed by a final row of blank (gray) cells. Manually entering data into the final blank row causes a new last row of data to be created in the table, followed by a new final blank row.

4.2.7 Edit Virtual Signals

The Scope (Analysis) window Measurements button provides access to the 'Edit Virtual Signals' dialog. Use it when applying different analysis scenarios to recorded data with "pseudo" input signals, in conjunction with the 'Reanalysis Measurements & Graphs' dialogs.

Virtual input channels allow you to perform a variety of mathematical transformations on input signals. To enable a virtual signal, highlight a signal name. When a virtual input channel is enabled, its configuration fields are ungrayed. Changes to the highlighted signal are saved when you click the 'Do It' button, and changes in unhighlighted signals are discarded.

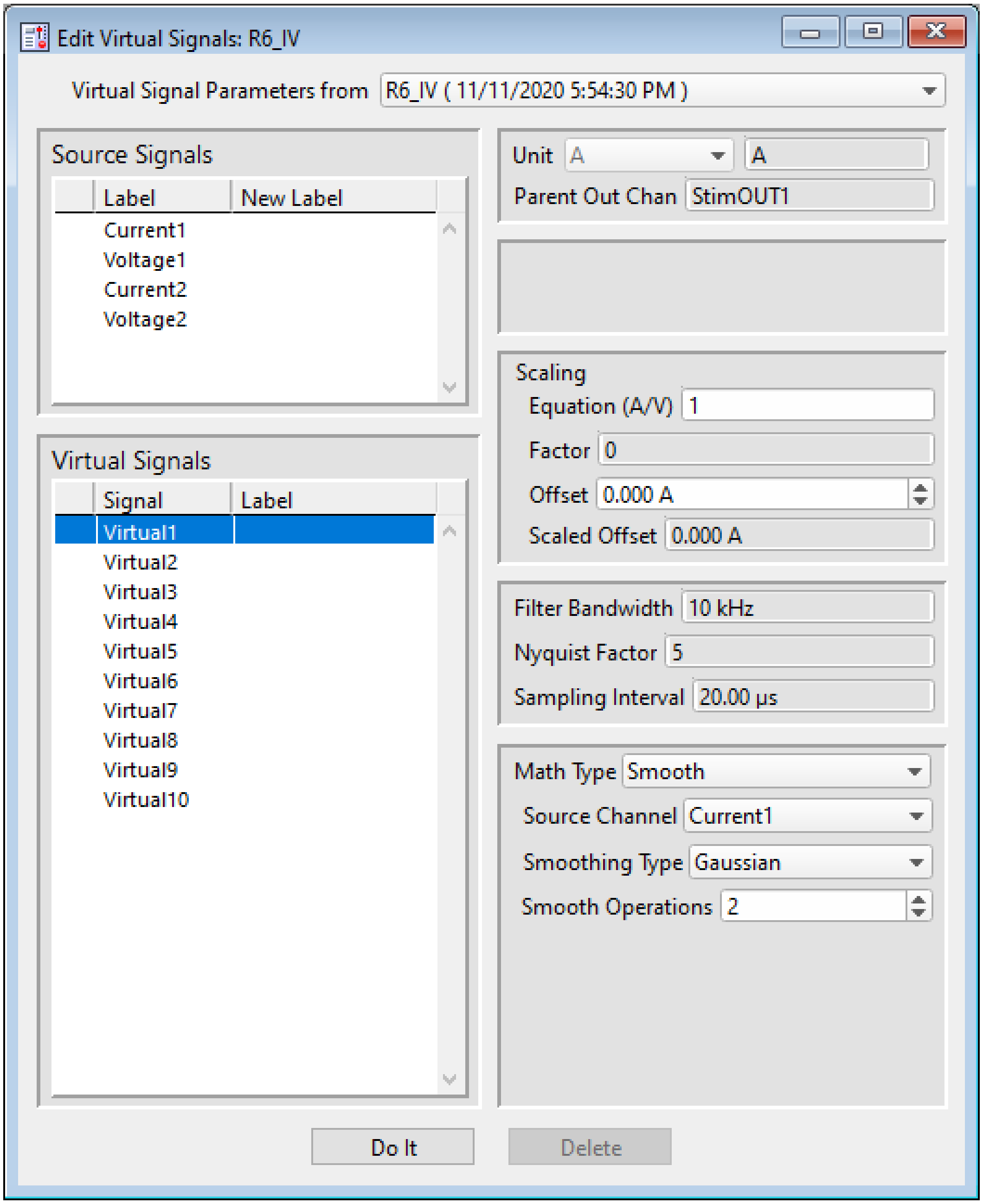


Figure 4-67. Edit Virtual Signals

Virtual Signal Parameters from [<Series name> (date/time stamp)]

Overwrite with Original Routine

<u>Source Signals</u>

Label

New Label

Virtual Signals

Signal

Label

Unit — The base unit of measurement from its Source signal. The resolution of the unit is automatically adjusted in the signal.

Parent Out Chan

Scaling

Equation — Use a value or an equation for the scaling ratio.

Factor — Apply scaling to interpret the input signal data. Specify as a numeric value or an equation.

Note: The dPatch system acquires data with a high-resolution 16-bit ADC into 64-bit data words, so data resolution is not an issue when scaling input signals.

Offset — Apply an amplitude offset to the input signal (after any scaling).

For “mV” units, append with ‘m’ or ‘e-3’.

For “pA” units, append with ‘p’ or ‘e-12’.

Example: 5 picoamps using engineering notation: 5p
or in equivalent scientific E-notation: 5e-12

Scaled Offset

Filter Bandwidth

Nyquist Factor

Sampling Interval

Math Type — Apply a data transformation to a virtual input signal:

- BaselineSubtract

  Subtract a fixed value from all data points in an input trace. This is useful for adjusting for an offset, or resetting a baseline.

  Source Channel — Select an input channel to process.

Baseline From — Select how to calculate the subtraction value.

- Value — Subtract a fixed value.
  - Value — Spinner adjusts in 1 pA or 1 mV increments.
- Trace — Subtract the average of the entire input trace.
- Sweep Time — Subtract the average of the data between the Start Time and End Time.
  - Start Time — Set the starting time of the data to be averaged.
  - End Time — Set the ending time of the data to be averaged.
- Segment #s — Subtract the average of a Segment from the input trace.
  - Start Ratio — Set the starting time of the data to be averaged, as a ratio relative to the starting time of the Segment duration.
  - Start Time — [derived value]
  - End Ratio — Set the ending time of the data to be averaged, as a ratio relative to the ending time of the Segment duration.
  - End Time — [derived value]

- BesselFilter — A frequency-domain filter with excellent response characteristics for preserving the shape of a biological signal.

Source Channel — Select an input channel to filter.

Filter Bandwidth — Select a frequency range.

- LowPass — Allow signal frequencies less than the cutoff frequency, and block all higher frequencies, such as high frequency noise.
- HighPass — Allow signal frequencies greater than the cutoff frequency, and block all lower

frequencies.

Filter Order [1, 2, 4, 8]

Number of “poles” in the filter

Cutoff Frequency (Hz) [100 to < ½ the sampling rate]

Restrict frequencies from this boundary point onwards.

Integrator Reset [for Capacitive Mode]

- Ignore — Capacitive-mode transients are displayed in the data.
- Blank — The data during capacitive transients are made invisible by replacing those data points with NaNs (Not a Number).

  Blank Duration [10 us – 1 s]
- Mask — The data during capacitive transients are replaced by the last data value before the transient discharge, simulating a sample-and-hold operation.

  Mask Duration [10 us – 1 s]

  The default value of 500 µs should be sufficient to encompass the reset transient duration.

- Differentiate — Apply differentiation to an input signal. The instantaneous rate of change in the signal is displayed.

  Source Channel — Select an input channel to differentiate

- Equation — Specify an equation to process an input signal.

  Source Channel — Select an input channel to process.

  Equation [ ] — Click field to access the ‘Specify math equation’ editor.

  Note: The full equation is always visible as a tool tip, by hovering the mouse cursor over the ‘Math Equation’ field.

Specify math equation for virtual signal

[<equation>] A free-form text field

Errors are reported under this field.

Check Equation Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports "Syntax is "ok".

Insert special identifier

Special references can also be used within commands:

- p[#] n'th Paradigm variable.

- s[series-count, sweep-count, trace-count, routine name]

  Access an arbitrary input trace (data wave) via counts of Series #, Sweep #, Trace # (Scope position), and the Routine name.

  The "current" item is the "active" trace in the Scope window, and has a count value of zero.

  If a "count" number is non-zero, it is used as an offset from the current count value of zero. Any fractions in count numbers are truncated to integers.

  If the routine name is left blank, the current routine name is used.

  Ex: s[0,0,0,]

  The current series, current sweep, current trace, of the current routine.

- t[#] Access the input trace (data wave) in Scope position "n" for the last sweep of the current Series.

  This numbering can differ from the Scope Position "n" if signals are re-arranged or hidden

Undo	All changes in the equation editing session are discarded.

(See the Equation Editor for more details.)

- Integrate — Display the integral of the data signal. This is equivalent to the signed area under a curve.

Source Channel	Select an input channel to integrate.

- Leak — Remove leakage current from the data signal. This is the small passive current when the cell is in a resting state.

  This feature is only enabled when the Routine includes an output channel with P/N Leak Pulse enabled.

Source Channel	Select an input signal to process.
Show Leak	Display the accumulated leak currents after the subtracted data in a sweep. Display the average of the leak sub-pulses.
Leak Zero Segment	Identify a segment with no active cellular response to the command signal. When set to zero, the field is set to 'OFF'. To re-display the numeric spinners, enter a non-zero number into the field.
Note:	The mean of the second half of the specified segment is used to compute an averaged leak current, which is then used to correct the P/N leak average. This option reduces the influence of a constant leak-current, which is otherwise included in the current of the main signal.

- LineFreq — Remove AC line frequency noise (hum) from the data signal.

Source Channel	Select an input channel for noise reduction.	
Line Frequency	50 Hz	Most of rest of world.
	60 Hz	Canada, (Carribean), Central America, (Japan), Mexico, (South America), South Korea, Taiwan, USA.

Alternating current (AC) power contains 50 or 60 Hz oscillations that can cause sinusoidal line-frequency noise in recorded signals. This FFT-based filter reduces such noise by > 90% over 6 harmonics. The adjusted signal is displayed in real time.

- LockIn — Measure cell characteristics (such as membrane capacitance) with high signal-to-noise sensitivity, using a dual-phase software lock-in amplifier.

Note: This feature is only enabled when the Routine includes an output channel with a waveform Segment set to 'Sine / Sine Wave Cycles / For LockIn'.

Calculations are made using 'conductance' (1 / resistance) insead of 'resistance'.

Current Channel — Select a (source) input channel with a "current" signal.

Trace Kind — Select the LockIn measurement to display.

The selected 'Trace Kind' is automatically set as the Virtual Channel label.

CM — Computed membrane capacitance.

GM — Computed membrane conductance.

GS — Computed series conductance.

DC — DC component of measured signal.

RealY — Real number part of the lock-in response signal.

ImagY — Imaginary number part of the lock-in response signal.

Cycles to Average — [1 – 1000]

Cycles to Skip — [1 – 1000]

V-reversal — [±1000 mV]

When using a calculated stimulus trace, enter the reversal potential for the ion under study, such as for (Na+) sodium spikes or (K+) potassium tail currents.

Note: See the SutterPatch Algorithms appendix for the math used in the LockIn computation.

- Smooth — Smooth the data with a “moving average” noise-reduction filter.

  Source Channel — Select an input channel to smooth.

  Smoothing Type

  - Gausian — A standard filter with excellent 10 – 90% rise-time response

    Smooth Operations [1 – 32767]

    # of smoothing operations to perform.

  - Boxcar — A fast time-domain filter with excellent 0 – 100% rise-time response

    Smooth Repetitions [1 – 32767]

    # of smoothing repetitions to perform.

    Boxcar Window Points [1 – 99]

    # of points in boxcar sliding window.

    Note: For best performance, only odd values are used.

  Integrator Reset [for Capacitive Mode]

  - Ignore — Capacitive-mode transients are displayed in the data.
  - Blank — The data during capacitive transients are made invisible by replacing those data points with NaNs (Not a Number).

    Blank Duration [10 us – 1 s]

  - Mask — The data during capacitive transients are replaced by the last data value before the transient discharge, simulating a sample-and-hold operation.

Mask Duration [10 us – 1 s]

The default value of 500 µs should be sufficient to encompass the reset transient duration.

- Stimulus Replicate the command waveform.

  Source Channel Select an input channel – the waveform from its Parent Out Chan is used.

- SweepAverage Average the input traces.

  Source Channel Select an input channel to average.

  Average Type Cumulative

  Average all processed sweeps together.

  RunAverage

  Average the last "N" sweeps.

  Number of Sweeps

- SweepSubtract Subtract a sweep from the input trace.

  Source Channel Select an input channel to process.

  Reference Sweep Select a sweep to be subtracted from all other sweeps. If the sweep does not yet exist, no subtraction occurs.

4.2.8 Equation Editor

The Equation Editor manages simple or complex expressions that evaluate to a value. Such math equations can be used to create stimulus waveforms, or for data analysis.

Access the Equation Editor from the SutterPatch menu.

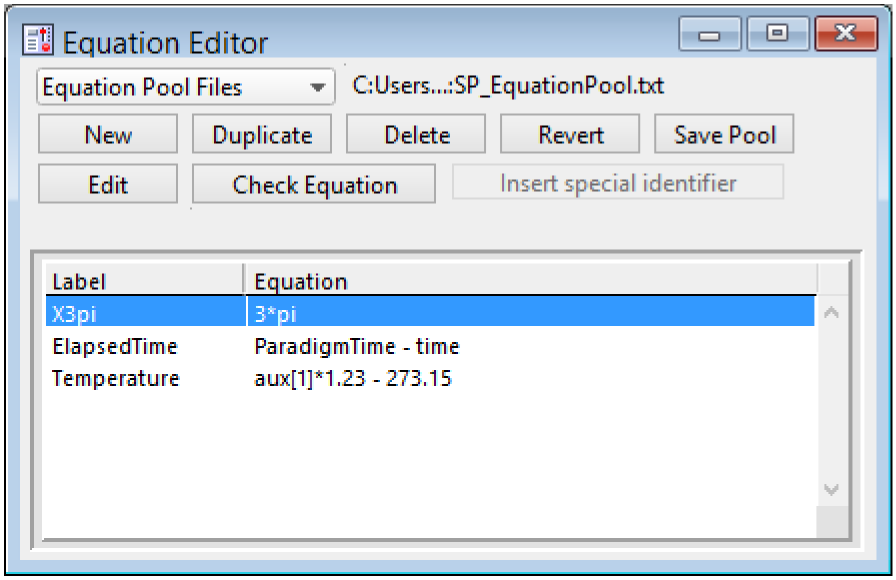


Figure 4-68. Equation Editor

Equation Pool Files — Equations are created and saved in an Equation Pool file.

New Equation Pool	Create a blank Equation Pool file.
Load Equation Pool	Load the Equations of a previously saved Equation Pool file into the Equation Pool.
Revert to Last Saved	Undo any unsaved changes to the Equation Pool.
Save Equation Pool	Save the Equation Pool using its existing file name and path.
Save Equation Pool As…	Save the Equation Pool to a new file, and switch to the new file. The default file name is the original file name.
Save Equation Pool Copy…	Save the Equation Pool to a new file, but do not switch to the new file. The default file name has 'Copy of' prepended to it.
Merge Equation Pools	Insert the Equation from a previously saved Equation Pool file into the loaded Equation Pool.

Note: Equation Pool files are simple text files (*.txt) that can be directly edited.

New — Create a blank Equation.

Duplicate — Add a copy of the selected Equation to the Equation Pool.

Delete — Remove the selected Equation from the Equation Pool.

Revert — Select an Equation and click the Revert button. All editable steps are reset to their last saved settings.

Save Pool — Save the Equation Pool using its existing file name.

Edit — Make edits to the 'Equation' field.

Check Equation — Check the equation syntax. The equation is evaluated for sweep #1, and if valid, it reports that the syntax is "ok".

Insert special identifier
Acquisition, amplifier and reference settings are available for use in equations. (See list below.)

Table

Label	Column of editable equation names, for quick usage in place of the equation.
Equation	Column of equations in free-form text fields.

Special Identifiers

Timing

Time	(present date-time, s)
Timer	(timer time, s)
ParadigmTime	(time at start of paradigm, s)
RoutineTime	(time at start of routine, s)

Paradigm Parameters

Loop	(active paradigm ForLoop count)
Sweep	(active paradigm EachSweep count)
LastSweep	(active paradigm sweep count of last sweep) Processing can occur before or after the last sweep of a series.

Example: In a Paradigm 'If' step, compare 'sweep' numbers in a ForEachSweep loop.

```
ForEachSweep
   EachSweep, Target=IV
   If, Left=sweep, Operation= '=', Right=LastSweep-1
      Alert, Text=LastSweep, DoBeep=true
   EndIf
ForEachEnd
```

AqStopped (last acquisition was stopped)

The last Routine-Series did not complete by itself.

Stimulant (last applied stimulant concentration)

From the Solution Editor 'Concentration' setting, for solutions configured as a 'Chemical Stimulant'.

Input (Input variable on paradigm window)

Hold[1..4] (holding of n'th output channel)

p[1..16] (n'th paradigm variable)

r[1..16] (n'th routine stimulus variable)

<u>Analysis Results</u>

m[1..16] (n'th analysis measurement value)

gx[1..16] (n'th analysis graph x value)

gy[1..16] (n'th analysis graph y value)

<u>Signal Readings</u>

AuxIN[1..4] (reading of auxiliary input, V)

A single-point voltage reading from an Auxiliary Input channel, such as from a slowly changing temperature probe.

Note: This usage does not require setting up a Routine Input Channel.

Imon (amplifier current reading, A)

In the Amplifier Control Panel (pA).

Vmon (amplifier voltage reading, V)

In the Amplifier Control Panel (mV).

Mean[name or count,start,width] (mean of given input signal)

'name' = signal name

'count' = window-signal position

'start' = time of start, s (of measurement range)

'width' = duration, s (of measurement range)

Headstage

ActiveProbe (active probe)

[1 – 4]

The "active" probe number is the Sutter headstage presently controlled by the Amplifier Control Panel.

For a single headstage system, the active probe is always headstage number "1".

NumProbes (number of probes)

[1 – 4]

The number of IPA headstages attached to the system.

IPA Settings

CCMode (amplifier current clamp)

VCMode (amplifier voltage clamp)

Hold (IHold in CC-mode, VHold in VC-mode)

[±0.000,000,020 A (±20,000 pA), or ±1.000 V (±1000 mV)]

IHold (amplifer holding current, A)

[± 0.000,000,020 (±20,000 pA)]

IHoldOn (amplifier holding current On)

VHold (amplifier holding voltage, V)

[±1.000 V (±1000 mV)]

VHoldOn	(amplifier holding voltage On)
IGain	(amplifier current gain, V/A)
	The gain of the active voltage-clamp ‘Current’ input channel.
VGain	(amplifier voltage gain, V/V)
	The gain of the active current-clamp ‘Voltage’ input channel.
Filter	(amplifier input filter in VC- and CC-mode, Hz)
	Read the low-pass filter of the input channels.
IFilter	(amplifier input filter in VC-mode, Hz)
	Read the low-pass filter of the ‘Current’ input channel in VC-mode.
VFilter	(amplifier input filter in CC-mode, Hz)
	Read the low-pass filter of the ‘Voltage’ input channel in CC-mode.
Offset	(amplifier pipette offset in VC-mode, V)
OffsetLock	(amplifier pipette offset lock On in VC-mode)

IPA Compensation

ECompMag	(amplifier electrode compensation magnitude, F)
ECompTau	(amplifier electrode compensation tau, s)
ECompOn	(amplifier electrode compensation On in CC-mode)
CmComp	(amplifier cell compensation Cm, F)
RsComp	(amplifier cell compensation Rs, Ohm)
RsCompOn	(amplifier cell compensation Rs On)
Bridge	(amplifier bridge balance, Ohm)
BridgeOn	(amplifier bridge balance On)

IPA Correction

RsCorr	(amplifier Rs correction, fraction)
RsPred	(amplifier Rs prediction, fraction)
RsLag	(amplifier Rs correction lag, s)
RsCorrOn	(amplifier Rs correction On)

Membrane Test

Relectr[1..2]	(electrode/seal/access resistance, Ohm)
	Value from last Membrane Test.
Rmemb[1..2]	(membrane resistance (cell mode), Ohm)
	Value from last Membrane Test.
Cmemb[1..2]	(membrane capacitance (cell mode), F)
	Value from last Membrane Test.
RMSNoise[1..2]	(membrane test RMS noise, A)
	Value from last Membrane Test.

Lock-In

LockInPhaseAdj	(Lock-In phase delay adjustment, s)
LockInAttenAdj	(Lock-In attenuation adjustment)

Other identifiers are forwarded to Igor Pro's 'Execute' command.

Equation Extras

Constants

true	1
false	0
ON	1
OFF	0

The following constants have 27-digit precision:

e	2.71...	(Euler's number)
pi	3.14...	(π)

Parsing and Operators

Equation parsing is executed from left to right, processing the highest precedence level operators first, except for comparison and bitwise operators, which associate from right to left.

Precedence	Operation Type	Operator
8	Comment	;
7	Exponentiation, Arithmetic operations: Left Shift, Right Shift	^, <<, >>
6	Negation (logical) operations: Unary Negation, Logical Negation	-, !
5	Multiplication, Division, Remainder	*, /, %
4	Addition, Subtraction	+, -
3	Bitwise operations: And, Or, Nor, Xor	&, |, nor, %^
2	Comparison operations: Greater Than, Greater Than or Equal, Less Than, Less Than or Equal, Equal To, Not Equal To	>, >=, <, <=, !=
1	Logical operations: And, Or, Conditional	&&, ||, ? :
0	All other operations	Round, trunc, ceil, floor, exp, sqrt, ln, log, sin, cos, tan, asin, acos, atan, abs, rad, deg, noise, random

Table 4-4. Equation Parser

For expressions using Comparison and Logical operators, it is recommended to use parentheses to explicitly define the order of execution.

There are also some differences in operator processing between the SutterPatch equation parser and the Igor Pro command parser:

Comment:

SutterPatch	All characters to the right of a semi-colon are ignored
Igor Pro:	A semi-colon separates multiple commands on the same command line. An Igor comment uses a double slash: //

The Conditional operator “? : ” is a shortcut for an if-else-endif expression. It evaluates as:

<expression> ? <True> : <False>

If the <expression> operand evaluates as non-zero, the <True> numeric operand is evaluated.

If the <expression> evaluates as zero, the <False> numeric operand is evaluated.

For complex expressions, only the real portion is evaluated.

Note: The " : " is a colon with 2 blank spaces around it.

An arithmetic left shift (<<) is the same as a bitwise left shift, whereby the least significant bit is padded with a zero. However, while an arithmetic right shift (>>) fills the most significant bit with its original value, thus preserving the sign, a bitwise right shift pads the most significant bit with a zero. A bitwise right shift can be constructed from existing operators.

Example: Shift # right by “n” bits

/ 2^n

Syntax

All equations use the same syntax as Igor Pro, with a few additions:

- Three kinds of brackets [ ], { }, ( ), can be used equivalently to improve the clarity of nested expressions.
- Numeric values can be written in scientific E-notation using exponents:

  5e-12 (5 picoamps)

  or in equivalent engineering notation using unit prefixes:

  5p (5 picoamps)

Prefix	Exponent	Prefix Name		Prefix	Exponent	Prefix Name
k	10^{3}	Kilo		**m**	10^{-3}	milli
M	10^{6}	Mega		**µ (or u)**	10^{-6}	micro
G	10^{9}	Giga		**n**	10^{-9}	nano
T	10^{12}	Tera		**p**	10^{-12}	pico
P	10^{15}	Peta		**f**	10^{-15}	femto
E	10^{18}	Exa		**a**	10^{-18}	atto
Z	10^{21}	Zetta		**z**	10^{-21}	zepto
Y	10^{24}	Yotta		**y**	10^{-24}	yokto

Table 4-5. Engineering Notation

- Insert an equation from the Equation Editor Pool into an Equation field by entering “#” followed by the label of the equation, e.g. “#MyLabel(5)”. This passes the argument “5” to the equation labeled “MyLabel” for evaluation.

Example: Using an LED light source

To stimulate in increments of light intensity, use an equation to transform light intensity values in Routine variables into actual stimulus values with amplitudes in volts.

Build an equation in the equation pool as follows:

equation = ln(r[1]) * 2.55 + 3

The natural log of the Routine Variable r[1] is multiplied by 2.55 and added to 3.

label = power_to_volts

In the Waveform Editor, set a Segment Amplitude field to ‘Equation’, and enter the equation as “#power_to_volts”.

A simplified version of the Equation Editor allows Equations (and equation labels) to be used in the following areas:

Paradigm Editor

Steps: Amplifier, Checkbox, Set Variable, Sound, Write Log, If, Else If

Routine Editor

Input Channels: Virtual Channels: Math Type: Equation

Output Channels: P/N Leak Pulses: Leak Hold, Waveform Editor: Amplitude, Duration

Measurements: Time to Threshold, X-Axis, Y-Axis

Note: Computing an equation for a data wave consumes significant computing power, as every data point needs to be computed by the CPU. While a slight update delay in such operations is expected, for computers with marginal computing power, the “beach ball” icon displays while the computer is unresponsive and busy processing.

4.2.9 Igor Analyses

Additional mathematical operations are located in the Analysis main menu. These built-in Igor analyses are documented in the Igor Pro Help:

- Curve Fitting — Create your own fitting equation
- Quick Fit — Use a pre-defined equation:

  line
  poly [3 – 10]
  poly_XOffset [3 – 10]
  gauss
  Ior
  Voight
  exp_XOffset
  dblexp_XOffset
  exp
  dblexp
  dblexp_peak
  sin
  HillEquation
  Sigmoid
  Power
  LogNormal
  poly2D [1 – 10]

Gauss2D

FitBetweenCursors

Weight from Error Bar Wave

Textbox Preferences

Example: Perform a fit on a section of a sweep:

1. In the Data Navigator, select the sweep, and use the 'Extract Sweep' command on it.
2. In the Analysis Editor, click the 'To Layout' button.
3. In the Layout window, right-click for the "Show Window" command.
4. With the Graph window active, select the main menu item Graph > Show Info.
5. In the Graph window cursor pane, drag cursors 'A' and 'B' onto the data.
6. Select main menu Analysis / Quick Fit and the fit of your choice.

Built-in Igor analyses are documented in the Igor Pro Help:

- Transforms
    - Fourier Transforms
    - Periodogram
    - Lomb Periodogram
    - MultiTaperPSD
    - Discrete Wavelet Transform
    - Continuous Wavelet Transform
    - Wagner Transform
    - Short-Time Fourier Transform
- Convolve
- Correlate
- Differentiate
- Integrate
- Smooth
- Interpolate
- Filter
- Resample

- Sort
- Histogram
- Compose Expression
- Packages
    - Average Waves
    - Batch Curve Fitting
    - Function Grapher
    - Global Fit
    - Igor Filter Design Laboratory
    - Median XY Smoothing
    - MultiPeak Fitting
    - Percentiles and Box Plot
    - Wave Arithmetic

4.2.10 Layout Window

The Layout window is used to prepare your data for publication. Scope window input signals analysis graphs and other objects can be exported to a Layout window for graphical arrangement and editing.
A default Layout window is automatically created when SutterPatch is launched - display it with the menu command SutterPatch / Layout Page / Show Layout. Only one Layout window exists at a time. If no Layout window exists, it can be manually created via a ‘Layout’ button , such as found in Scope windows. The Layout window can also be created by running a Paradigm ‘Export’ step.

Note: A new Layout window is sometimes created hidden behind other windows.

Clicking a Layout button appends its associated items into an existing Layout page (or a new Layout window.) When a Scope window is exported to the Layout window, each signal and analysis graph is appended as an individual object.

The default configuration of “2 x 4” (‘column’ x ‘row’) objects per page can be changed in Preferences / Export Graphics or the Paradigm ‘Export’ step, and is applied when a new Layout window is created:

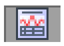
- 1 single pane
- 2 2 stacked panes
- 3 3 stacked panes
- 2 x 2 matrix

- 2 x 3 matrix
- 2 x 4 matrix

Once a Layout window page is filled, additional objects are automatically appended into additional Layout pages.

A toolbar displays in the upper-left edge of the Layout – the top two buttons reconfigure the toolbar buttons:

:

-  Operate Selection tools and object insertion mode.
-  Draw Mode Drawing tools mode.

When a Layout window is the active window, the main menu "Layout" command also displays to customize the layout. The 'Append to Layout' command allows you to append additional objects into the Layout window.

4.2.11 Metadata Review

"Metadata" parameters describe the system environment, attached Sutter instrumentation, Paradigm and Routine acquisition settings, and tag information.

Metadata parameters can be retrieved several different ways:

Data Navigator

- Select a Paradigm or Routine, and click its 'Metadata Wave Name' in the Preview pane.
- Select a Paradigm or Routine, then click the 'Available actions' button and select View Metadata.
- Right-click a Paradigm or Routine and select View Metadata.

Once the Metadata Review window is open, new Paradigm or Routine selections also update the Metadata Review.

Scope (analysis)

For Routines from the Dashboard 'View Last' button or the Data Navigator 'Analyze' button, use the 'View Metadata' button in their Scope window.

To display metadata from two different Paradigms, display the first Paradigm in a 'Review Paradigm Data' window and click the 'View Metadata' button. Then select the second Paradigm metadata as listed above.

To display metadata from two different Routines, display the first Routine in a 'Review Routine Data' window or Scope (analysis) window and click the 'View Metadata' button. Then select the second Routine metadata as listed above.

Paradigm Data [ ] Displays the name of the Paradigm.

Routine Data [ ] Displays the name of the Routine.

Metadata parameters from all detail levels are displayed (see Set Preferences / Metadata).

- By Event — Events are grouped by [time-stamp] [Event #] [Event type]. Highlighted fields are editable.

- By Parameter — Parameters are grouped into major categories. (see below)

  Routine parameters that are set by their higher-level Paradigm are labeled as 'Prior value', as those time stamps are set at the time of the Paradigm, not the Routine; prior non-Routine tags are also labeled as 'Prior value'.

Expand All — All settings entries are displayed.

Two columns of information are presented (parameter name and value). If the first column's text does not fully display, either increase the width of the window, or adjust the indentation of the second column – drag it when the mouse cursor turns into a double-headed arrow

Collapse All — All settings entries are hidden, collapsed up to the Event or Parameter level.

'By Parameter' Metadata Categories / Parameters

<u>Tag</u>
Tag Number
Tag Creation Timestamp
Timer Time at Tag Creation
Tag Signals
Tag Source Event

<u>Operator</u>
Login Name

<u>Experiment</u>
Experiment Timestamp

<u>Amplifier</u>
Amplifier Sequence Number
Amplifier Manufacturer
Amplifier Model

Amplifier Firmware Version
Amplifier Serial Number
Amplifier Channel
Number of Available Headstages
Headstage Sequence Number
Headstage Model

<u>Instrumentation and Software</u>
Interface Sequence Number
Interface Manufacturer
Interface Model
Interface Firmware Version
Interface Serial Number
Interface Input Channel (physical)
Interface Out. Ch. (physical or logical)
Interface Signal Type
Interface Number of Digital Outputs
Computer Name
Physical Computer Memory
Operating System Platform
Operating System
Software Environment
Software Environment Version
Software Environment Build
Software Environment Kind
Software Environment Serial Number
Data Acquisition Software
Data Acquisition Software Version
Data Acquisition Software Build
Data Acquisition XOP Version

<u>Paradigm</u>
Paradigm Data Sequence Number
Paradigm Data Base Name
Paradigm Name
Paradigm Description
Paradigm Data Start Timestamp

<u>Series (= Routine Data)</u>
Series Sequence Number
Series Base Name
Routine Name
Routine Acquisition Mode
Routine Description
Routine Data Start Timestamp
Routn. Completed / Terminated Early
Number of Input Signals

<u>Sweep</u>
Sweep Number

<u>Data Acquisition Settings</u>

Active Headstage	
Recording Mode	
Current Gain	
Voltage Gain	
Headstage Gain	
Headstage Feedback Mode	[dPatch only]
Filter Cutoff Frequency	
Filter Type	
Input Offset Voltage	[VC mode, ‘0’ in Demo mode]
Input Offset Lock On/Off	[VC mode]
Input Liquid Junction Potential	
Subtract Pip. Offset in Current Clamp	[dPatch]
Input Signal Name	
Input Signal Units	
Input Scaling Factor	[for AuxIN channels]
Input Full-scale Minimum	
Input Full-scale Maximum	
Input Sampling Rate	
Auxiliary Input Signal Offset	[for AuxIN only]
Virtual Signal Scaling Offset	[for Virtual Input channels]
Virtual Signal Math Type	[for Virtual Input channels]
Virtual Signal Equation	[for Virtual Input channels]
Virtual Signal Source Channel	[for Virtual Input channels]
Virtual Signal Source Signal Name	[for Virtual Input channels]
Virtual Signal Subtract Baseline Type	[for Virtual BaselineSubtract]
Virtual Signal Subtract Baseline Start	[for Virtual BaselineSubtract]
Virtual Signal Subtract Baseline End	[for Virtual BaselineSubtract]
Virtual Signal Filter Type	[for Virtual BesselFilter]
Virtual Signal Filter Order	[for Virtual BesselFilter]
Virtual Signal Filter Cutoff Frequency	[for Virtual BesselFilter]
Virtual Signal Integrator Reset Strategy	[for Virtual BesselFilter & Smoothing, Capacitive mode]
Virtual Signal Integrator Reset Duration	[for Virtual BesselFilter & Smoothing, Capacitive mode]
Virtual Signal Leak Display On/Off	[for Virtual Leak]
Virtual Signal Leak Zero Segment	[for Virtual Leak]
Virtual Signal Line Frequency Base	[for Virtual LineFreq]
Virtual Signal LockIn Trace Kind	[for Virtual LockIn]
Virtual Signal LockIn Cycles to Average	[for Virtual LockIn]
Virtual Signal LockIn Cycles to Skip	[for Virtual LockIn]
Virtual Signal LockIn Reversal Potential	[for Virtual LockIn]
Virtual Signal Smoothing Algorithm	[for Virtual Smoothing]
Virtual Signal Smoothing Factor	[for Virtual Smoothing]
Virtual Signal Sweeps Processed	[for Virtual SweepAverage]
Virtual Signal Reference Sweep	[for Virtual SweepSubtract]
Electrode Fast Magnitude	[VC mode]
Electrode Fast Time Constant	[VC mode]
Whole-cell Compensation On/Off	[VC mode]
WC Comp – Series Resistance	[VC mode, if WC Comp On]
WC Comp – Membrane Capacitance	[VC mode, if WC Comp On]

Series Resistance Correction On/Off	[VC mode]
Series Resistance Prediction Value	[VC mode, if Rs Correction On]
Series Resistance Correction Value	[VC mode, if Rs Correction On]
Series Resistance Corr. Lag Time	[VC mode, if Rs Correction On]
Capacitance Neutralization On/Off	[CC mode]
Capacitance Neutralization Mag.	[CC mode, if Cap Neut On]
Capacitance Neutralization Tau	[CC mode, if Cap Neut On]
Bridge Balance On/Off	[CC mode]
Bridge Balance Resistance	[CC mode, if Bridge Balance On]
Current Clamp Dynamic Hold On/Off	[CC mode]
Current Clamp Dyn. Hold Potential	[CC mode, if Dynamic Hold On]
Current Clamp Dynamic Hold Speed	[CC mode, if Dynamic Hold On]
CC Dynamic Hold On for Acquisition	[CC mode, if Dynamic Hold On]
Command Signal Name 1	
Command Signal Units 1	
Command Full-scale Minimum 1	
Command Full-scale Maximum 1	
Command Sampling Rate 1	
Command Holding Enabled 1	
Command Holding Value 1	[‘0’ if Holding disabled]
Auxiliary Output Signal Name 1	[if AuxOUT1 enabled]
Auxiliary Output Scaling Factor 1	[if AuxOUT1 enabled]
Auxiliary Output Offset 1	[if AuxOUT1 enabled]
Auxiliary Output Holding Value 1	[‘0’ in demo mode]
Command Signal Name 2	[if StimOUT 1 & 2 enabled]
Command Signal Units 2	[if StimOUT 1 & 2 enabled]
Command Full-scale Minimum 2	[if StimOUT 1 & 2 enabled]
Command Full-scale Maximum 2	[if StimOUT 1 & 2 enabled]
Command Sampling Rate 2	[if StimOUT 1 & 2 enabled]
Command Holding Enabled 2	[if StimOUT 1 & 2 enabled]
Command Holding Value 2	[‘0’ if Holding disabled]
Auxiliary Output Signal Name 2	[if AuxOUT2 enabled]
Auxiliary Output Scaling Factor 2	[if AuxOUT2 enabled]
Auxiliary Output Offset 2	[if AuxOUT2 enabled]
Auxiliary Output Holding Value 2	[‘0’ in demo mode]
Digital Holding Pattern (1 → N)	[1 – 8]

4.2.12 Paradigm Review

'Paradigm Review' displays data from all Series within the Paradigm, in a Time Course or Concatenated view in a modified Scope window. (For more information on the window controls, see the Scope (Analysis) section below.)

This view also displays the tags that occur between Series.

To display the Series_Signal_Routine name of the selected data at the bottom of the window, click on the data.

To open a Series into a Scope (Analysis) window, right-click on the Series data and select Analyze <Series Name>' from the menu list.

4.2.13 Reanalysis Measurements & Graphs

The Scope (Analysis) window Measurements button provides access to the 'Reanalysis Measurements & Graphs' dialog. Use it to apply different analysis scenarios to recorded data, by configuring measurements on input channels and analysis graphs.

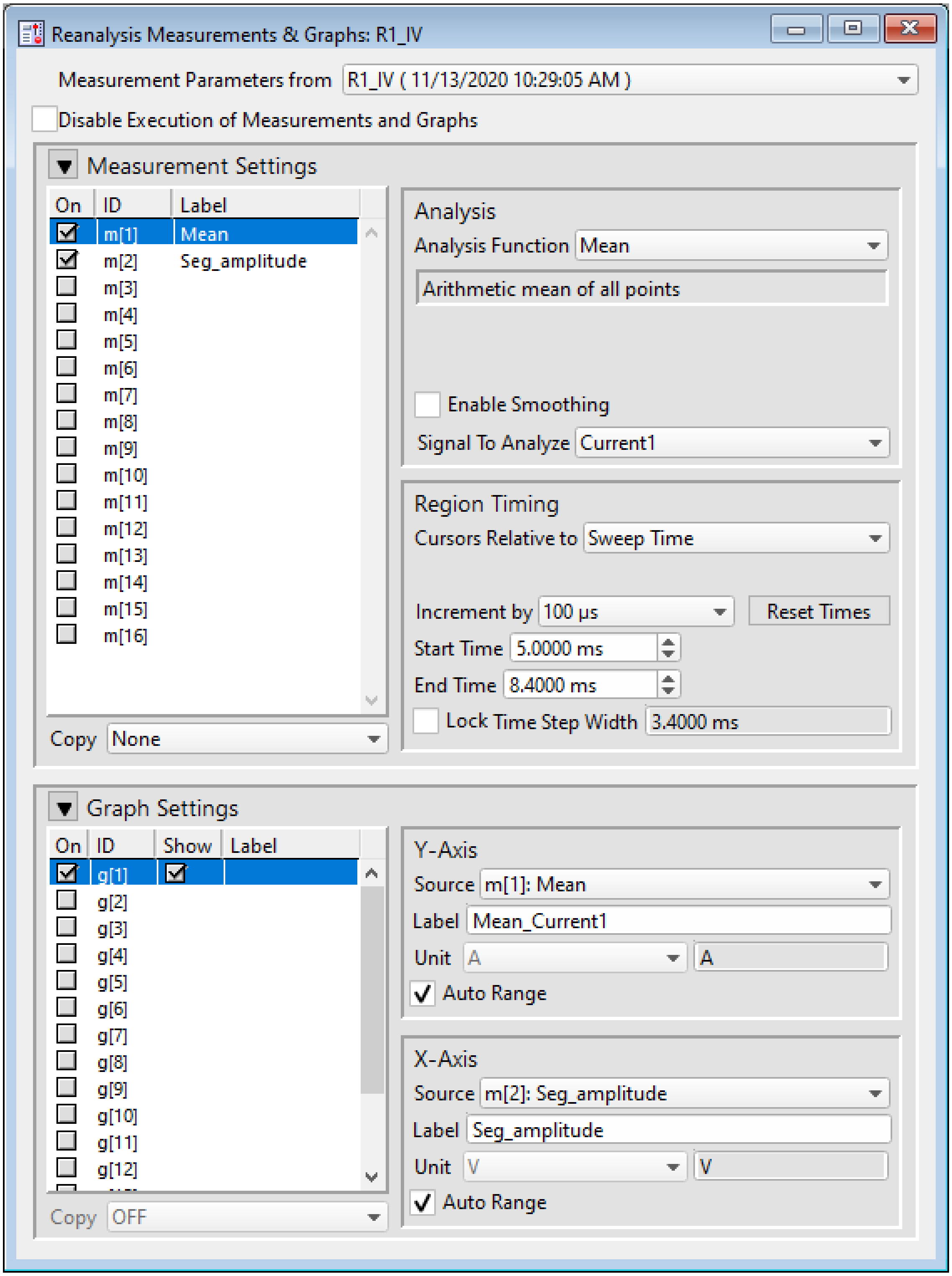


Figure 4-69. Reanalysis Measurements & Graphs

This dialog operates similarly to the Routine Editor: Real Time Measurements & Graphs dialog, with the addition of optional configurations:

Measurement parameters from	Select which Routine settings to record with.

- Current Settings (newly modified settings)
- Last Executed (last used version)
- Original Routine (original loaded version)

Analysis Examples

Example 1: Plot the mean of the data (using sample routine IV)

1. Set measurement m[5] to the 'Mean' analysis and select signal Current1.
2. Enable graph [g5].
3. From the graph's Y-Axis list, select m[5]. The Equation field displays:

   m[5]

4. Set 'X-Axis' to 'time' .
5. Run the analysis.
6. An Analysis window displays a graph of the mean vs. time.

Example 2: Plot the difference between two measurements

1. Set measurement m[5] to the 'Mean' analysis and select signal Current1.
2. Set measurement m[6] to the 'Mean' analysis, using the same signal.
3. Adjust the m[6] cursors Start/End times so they do not overlap with the m[5] cursors.
4. Enable graph [g6].
5. For the graph's Y-Axis, select 'Y-Equation' and enter the equation as:

   m[5] – m[6]

6. Set the X-Axis to 'time'.
7. Run the analysis.
8. An Analysis window displays a graph of the difference vs. time.

4.2.14 Routine Data Review

'Routine Data Review' displays the selected Series data in a Time Course or Concatenated view in a modified Scope window.

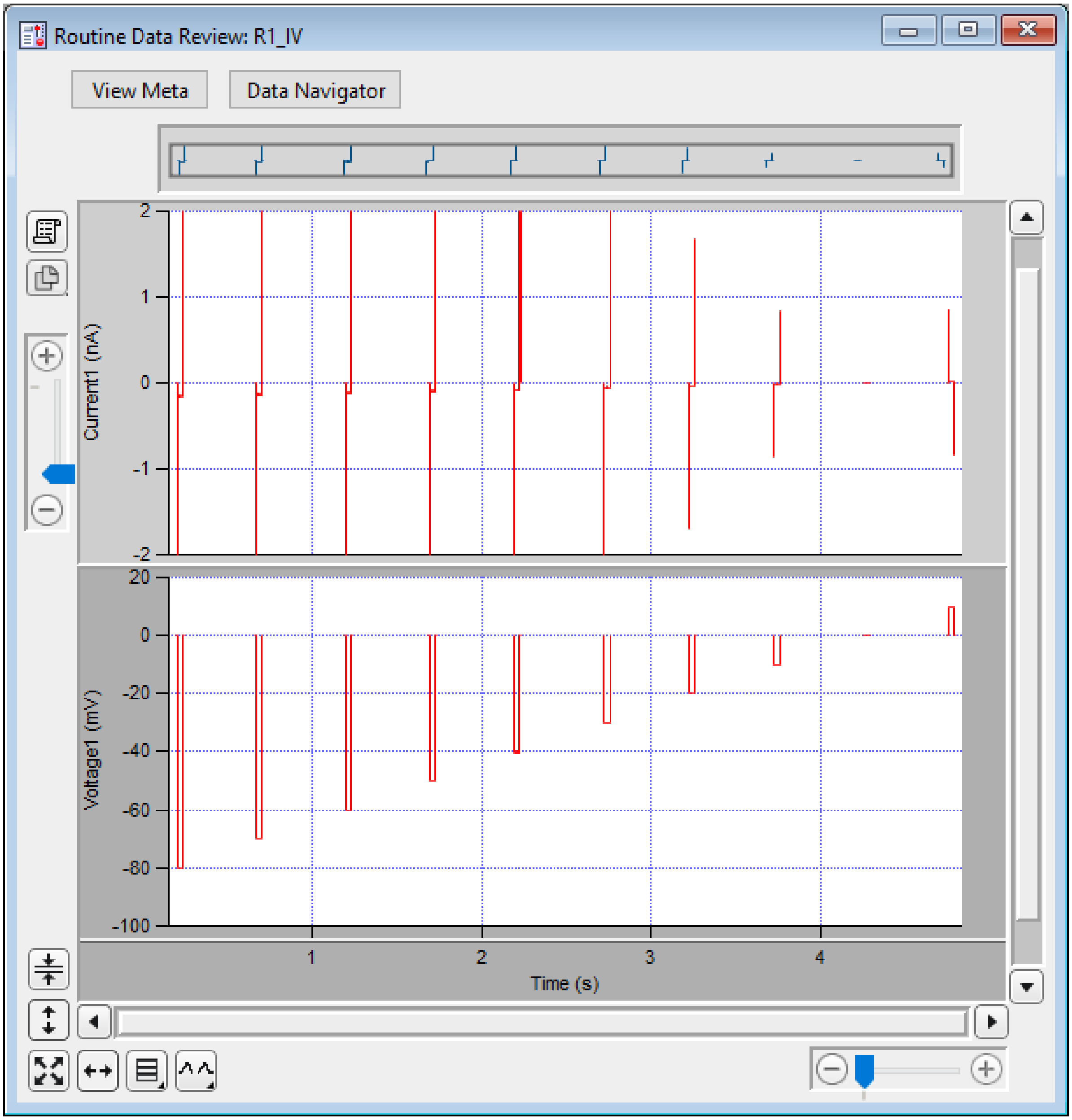


Figure 4-70. Routine Data Review

For more information on the window controls, see the Scope (Analysis) section below.

Open this window from the Data Navigator by highlighting a Series, and selecting the 'Review Routine Data' command from a right-click menu or the 'Available actions' button.

To display a signal name at the bottom of the window, click on the signal.

To open a Series in the Routine Review into a Scope (Analysis) window, right-click in a signal, and select 'Analyze <Series Name>' from the menu list.

4.2.15 Routine Settings

The Routine Settings window reports the same settings as would be seen in the Routine Editor / Routine Settings, however the preview pane does not support interactive dragging of measurement regions.

Open this window from the Data Navigator by highlighting a Series, and selecting the 'View Routine Settings' command from a right-click menu or the 'Available actions' button, or by selecting 'Routine Data Name' in the Data Navigator preview pane.

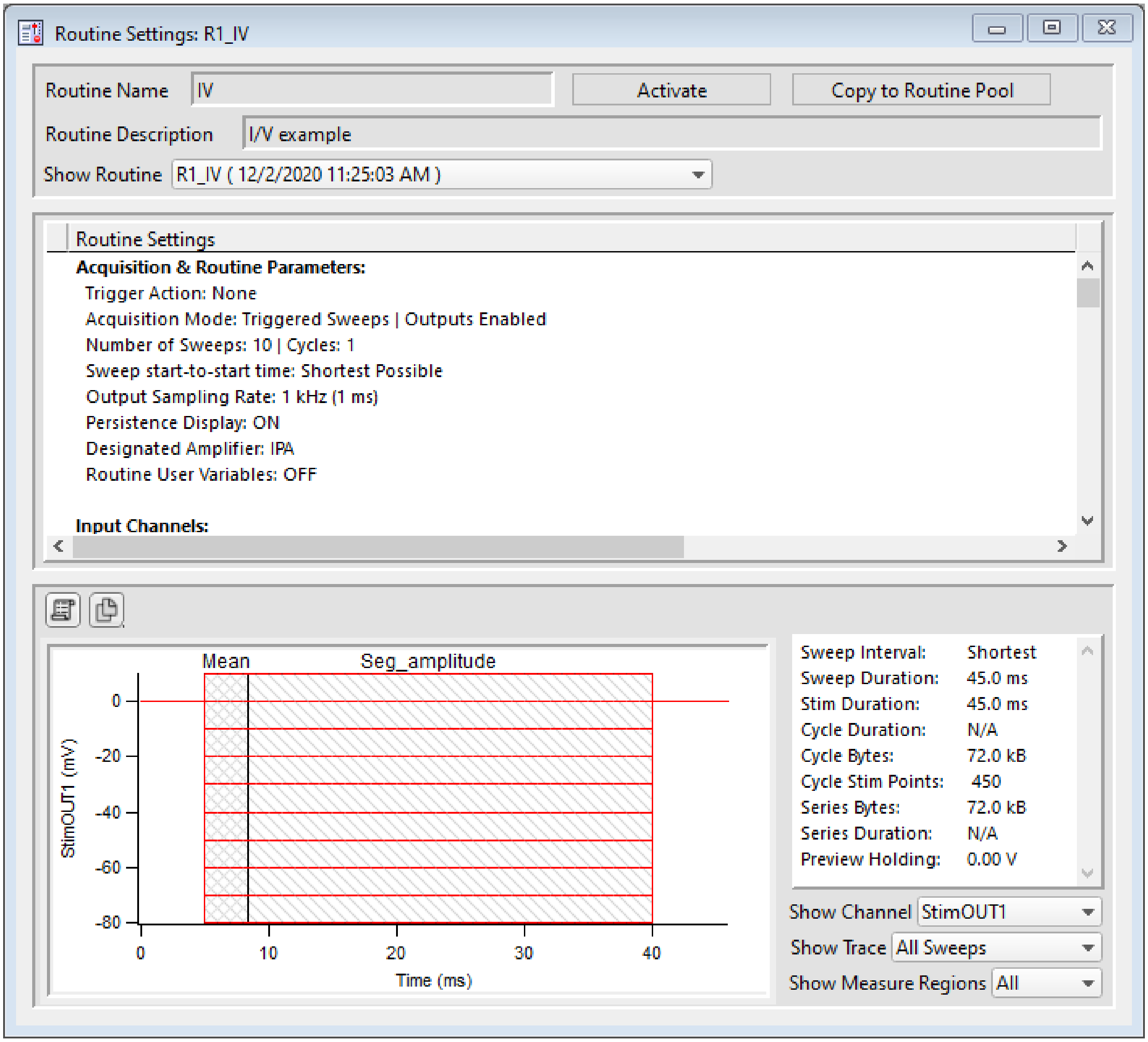


Figure 4-71. Routine Settings

Routine Name	Displays the Routine name.
Activate	Opens the Scope window loaded with these settings.
Copy to Routine Pool	Adds this routine to the loaded Routine Pool.
Routine Description	Displays the Routine description.
Show Routine Copy	
Routine Settings	Listing of all settings from all sections of the Routine.
Preview panel	Display of the stimulus waveform.

Copy to Layout	Copy the visible stimulus waveforms into a new Layout window, or append to an existing Layout page.
Copy to Clipboard	Copy the visible stimulus waveforms to the system clipboard.

Some key acquisition settings and display controls are listed on the right of the Preview pane:

Show Channel	Select the output signal to display.
Show Trace	Select the output trace to display.
Show Measure Regions	Select the measurement region to display.

4.2.16 Scope (Analysis)

This analysis version of the Scope window is used to display and reanalyze stored data.

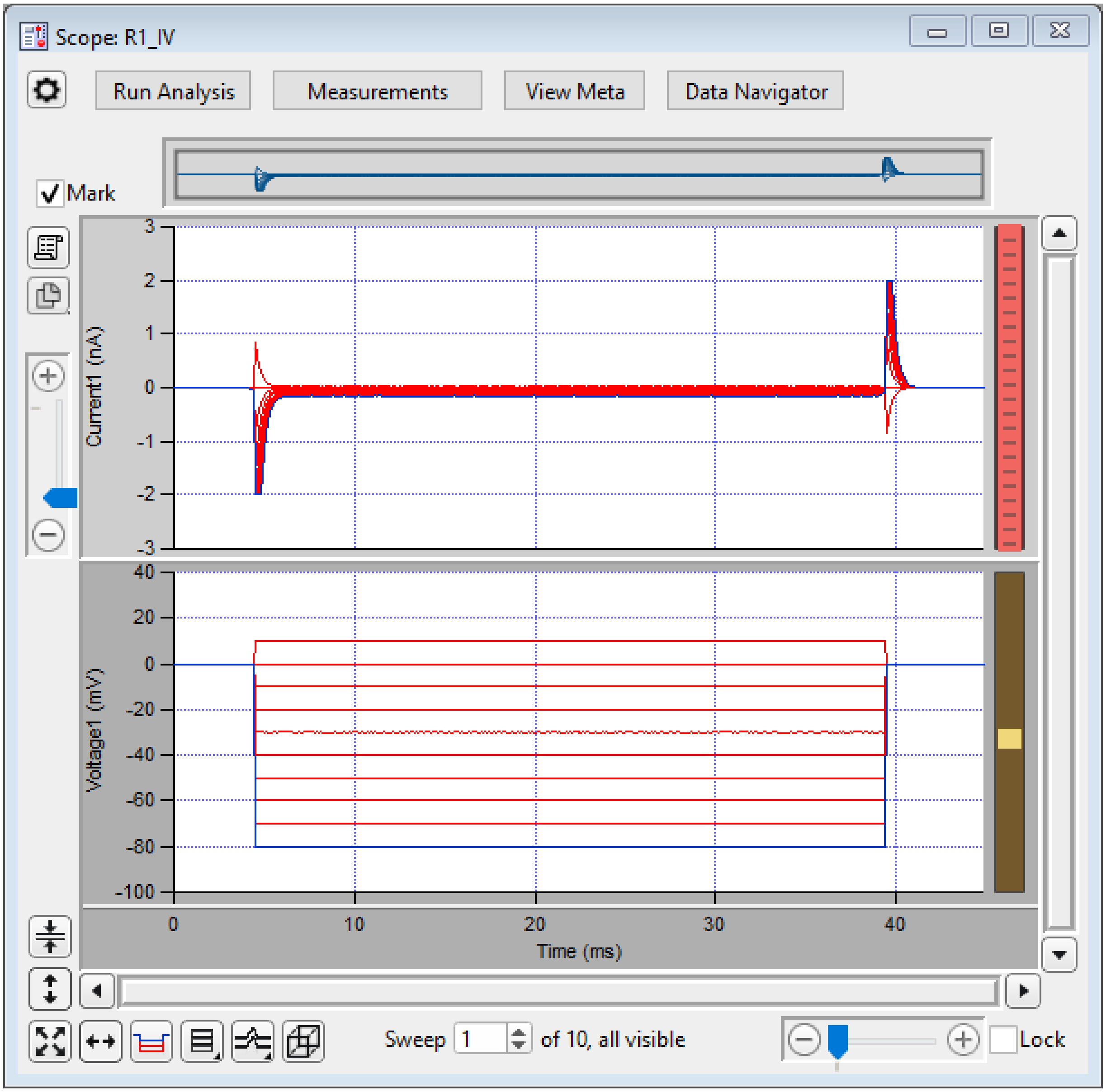


Figure 4-72. Scope (Analysis) Window

Both physical and virtual channels can be displayed here. Many of the controls are the same as in the Scope (acquisition) window, however additional window controls are also included.

<u>Navigation pane</u>

The Navigation pane appears at the top of the Scope window. It displays an overview of the active signal's full-scale data, with a gray box surrounding the magnification area.

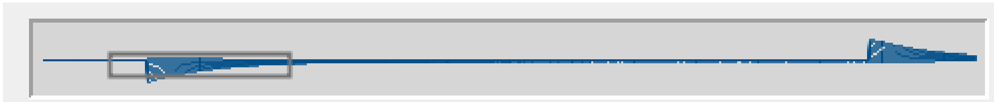
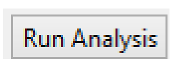
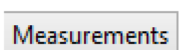


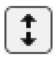
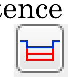
Figure 4-73. Navigation Pane

The Navigation pane "magnification" box can be used to scroll through the active signal's data. Place the mouse cursor over the magnification box and it changes into a 'hand' icon; click and drag the magnification box to scroll through the data.

Buttons

	Scope Settings: Set all marks in sweep of active series Clear all marks in sweep of active series ------ Begin with all marks set (in sweeps of active series) Begin with all marks cleared (in sweeps of active series)
	Run the defined analysis for the data series, and graph the results in the Analysis window. To stop a long-running analysis, click on the 'Abort' button in the bottom right corner of the main screen.
	Show Cursors: Display measurement cursors in the Scope window. Hide Cursors: Do not display cursors in the Scope window. Button state displays as "Measurements(H)". Lock Cursors Prevent cursors from being moved or altered. Button displays as "Measurements(L)". --- No Measurements or Graphs Analyze with Current Measurements Analyze with Original Routine Measurements Analyze with Routine Last Executed Measurements --- Load Default Measurements Save as Default Measurements --- Edit Measurements Open a special Reanalysis Measurements &

	Graphs dialog, where all changes apply instantly and interactively to the measurements and graphs, even during analysis. These edits (temporarily) override the loaded routine for fast response. Edit Virtual Signals — Open the virtual input signals dialog for editing, Action Potential Analysis Analyze action potentials. Synaptic Event Analysis Analyze synaptic events (EPSPs, mEPSPs, etc.) Single Channel Analysis Analyze a single ion channel.
View Meta	Display any extra information (metadata) associated with the displayed data Series, such as the operator, preparation details, solution information, etc.
Data Navigator	Open a Data Navigator window with all of your Experiment data and metadata available in a tree structure.
Mark	Enable to highlight the active sweep. The Data Navigator can process marked sweeps as a group.
Copy to Layout 	Copy all visible Scope signals and analyses into a new Layout window, or append to an existing Layout page.
Copy to Clipboard 	Copy the active signal graph to the system clipboard or, if the 'Shift' key is pressed, the complete Scope window.
Center 	Center the mean of the data in the selected signal pane. The Y-axis offset is automatically adjusted, while the Y-axis scaling is unchanged.
Autoscale	Click to autoscale the Y-axes of all visible signals to their visible sweeps data limits, and to reset all X-axes to their full sweep duration.

	To include the zero amplitude in the Y-ranges, configure “Autoscale from zero” in the Set Preferences / Scope Window / Reanalysis / Y axis initial settings To autoscale the Y-axes of all (including hidden) signals, in Windows right-click / Shift-click / Ctrl-click the button, or in macOS ctrl-click the button; also all X-axes are reset to their full sweep duration.	
Y-Autoscale 	Click to autoscale the Y-axis of the selected signal to its visible sweeps data limits. To include the zero amplitude in the Y-range, configure “Autoscale from zero” in the Set Preferences / Scope Window / Reanalysis / Y axis initial settings. To autoscale the Y-axes of all visible signals, in Windows right-click (or Shift-click or Ctrl-click) the button, or in macOS Control-click the button. Tip: To invert the Y-axis of the selected signal, such as for data with reversed polarity from an outside-out patch, right-click in the Y-axis of the signal and select Axis Properties / Axis Range. Either reverse the Manual Range Settings / Minimum and Maximum values, or disable the Manual Range and enable the Autoscale Settings / Reverse axis.	
X-Scale 	Full-scale all X-axes. All signals X-axes are reset to their full-scale setting.	
Persistence Display 	Display all sweeps (per Preferences settings). Otherwise, when disabled, only one sweep is displayed at a time.	
Signal Display 	Graphically arrange the input signals. Stack: A vertical column of signals Single: Only the active signal m x n: A tiled array of signals with ‘m’ rows and ‘n’ columns	
Sweeps Display	This button has 3 modes:	
	Sweeps 	Each trace starts from time zero to the duration of the waveform.
	Time Course 	Display sweeps in time sequence on a single time axis. Portions without data are left blank (such as the time between triggered sweeps.) Note: Emulation mode has a minimum 0.5 s interval between sweeps, both triggered and continuous. If the sweep duration is less than 0.5 s, the time between sweeps will be padded with

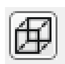
		“blank” time.
	Concatenated 	Display sweeps similarly to the Time Course mode, but any blank portions are replaced by a vertical line.
	The ‘Show 3D view of current signal’ button brings up a separate 3D display window attached to the right of the Analysis window. The Sweep data are color-coded for amplitude, and their 3D graph can be rotated in any direction.	
Sweep #:	The ‘Sweep #’ display at the bottom of the Scope window indicates the ‘active sweep’ number, the total number of sweeps in the Series, and either “all” or the total number of visible sweeps (per Preferences).	

Table 4-6. Scope Window Buttons

<u>Right-click Menus</u>

Right-click on the indicated regions to display a context-sensitive menu.

X- and Y-Axes — Same as the acquisition Scope.

Main window area — To display a limited data modification menu, right-click in the blank area in a signal pane.

Note: If you click too close to the signal data, the full data modification menu displays instead. If you are having this issue, click near to a horizontal or vertical edge of the signal pane.

Same as the acquisition Scope, plus these additional items:

- 3D View
- Zero Baselines
- Show All Sweeps (with triggered sweeps)
- Select Sweeps (with triggered sweeps)

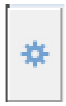
Signal data — To display the full data modification menu, right-click on or near the data,

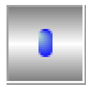
Same as the acquisition Scope, plus these additional items:

- Hide Sweep_# (with triggered sweeps)
- Show Sweep_# Only (with triggered sweeps)

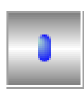
Also, to manually measure X-Y data values, or to set a fitting range, open a Cursor Info pane:

- Toggle Cursor Info   Show/Hide the Cursor Info pane.

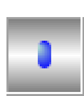
 Options menu

-  One Mover Moves All — Draggable cursor mover tool moves all cursors together with a single control.
- All Styles — Change the cursor symbol style.
- Show Cursor Pairs — Display up to 5 sets of cursor symbol pairs.

Cursor A

	Cursor symbol for data point ‘A’.
A:	Symbol letter (beginning cursor of the pair).
R1_A_IV	(Default) wave name.
	Draggable cursor mover tool for the cursor pair.
pnt:	Data point number (starting from zero).
X:	X-axis value of data point ‘A’
Y:	Y-axis value of data point ‘A’
ΔY	Difference of the cursor pair Y values.

Cursor B

	Cursor symbol for data point ‘B’.
B:	Symbol letter (ending cursor of the pair).
R1_A_IV	(Default) wave name.
	Draggable cursor mover tool for the cursor pair.

pnt: Data point number (starting from zero).

X: X-axis value of data point ‘B’

Y: Y-axis value of data point ‘B’

ΔX Difference of the cursor pair X values.

Cursor Instructions

1. Click on symbol ‘A’ to enable it.
2. Manually drag the highlighted symbol onto a data point in the Scope, or enter the data point number in the ‘pnt’ field.
3. Click on symbol ‘B’ to enable it.
4. Manually drag the highlighted symbol onto a data point in the Scope, or enter the data point number in the ‘pnt’ field.

   X- and Y-measurements are displayed for the cursor pair data points.
5. Fitting can also be applied to the cursor pair data. Right-click on the data, and select ‘Quick Fit’ for a list of built-in Igor fitting functions.

   The fit is displayed in the graph, and the fitting information is written to the Command window.

Channel Timing Delays

IPA amplifiers record both headstage stimulus and response signals via physical analog channels, so all recorded signals are precisely in sync, with no timing delays between them.

4.2.17 3D View Window

The Scope (Analysis) 3D View window creates a 3D representation of your data, color-coded to show amplitude variations.

The axis definition in 3D View is based on the change of a waveform over the course of successive sweeps. In a two-dimensional display, the X-axis represents the Sweep Time, while the Amplitude is plotted on the vertical Y-axis. For consistency, the vertical axis in the SutterPatch 3D view is also defined as the Y-axis. In the default orientation of the 3D View, the Z axis, on which the Sweep Number is plotted, points backward and to the right.

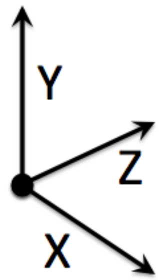


Figure 4-74. 3D Axes Definition

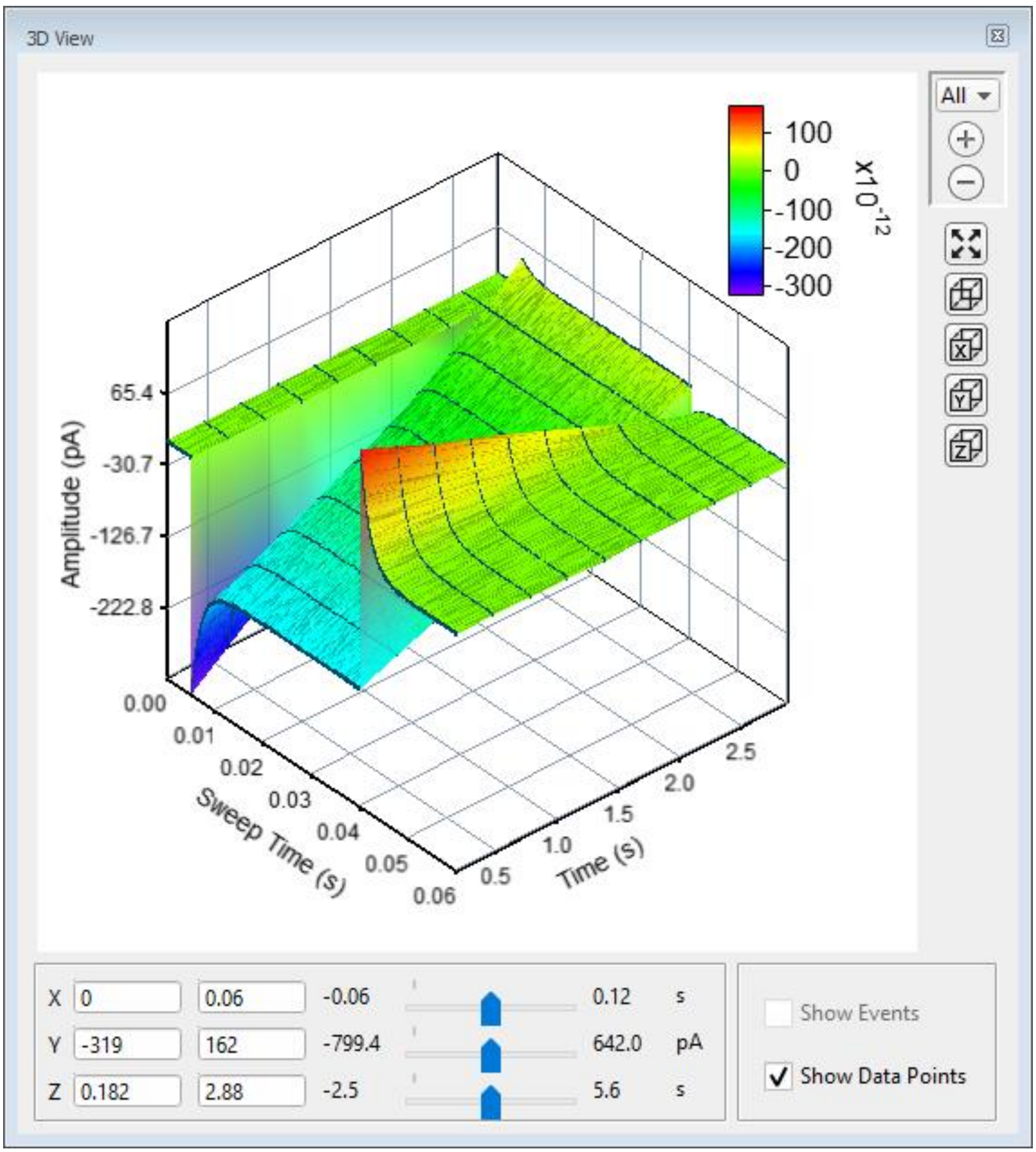


Figure 4-75. 3D View

A "heat map" bar illustrates the color measurement units.

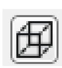
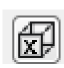
Magnification buttons are located in the upper right corner of the window for the selected axis:

- All (All 3 axes)
- X (Sweep Time)
- Y (Amplitude)
- Z (Time)

	Zoom in	(Magnify)
	Zoom out	(Unmagnify)
	Autoscale	(Set to the data limits)

X, Y and Z axis limits can be set in the bottom section of the 3D View window. Their delta value is preserved when using the scroll bars to update the visual graph (and the numeric axes limits.)

The 3D graph viewing angle can be changed with a set of 3D buttons:

	= Default View	(X, Y & Z axes display)
	X = Right View	(Y & Z axes display)
	Y = Top View	(Z & X axes display)
	Z = Front View	(X & Y axes display)

Alternatively, you can rotate the display in any direction by simply clicking and dragging the 3D graph. If you release the mouse button while dragging, the 3D display will rotate in the direction of the mouse drag.

Show Events Display tagged events in the 3D graph.

Show Data Points Display data points as surface dots in the 3D graph.

4.2.18 Set Metadata

A variety of optional experimental parameters (preparation, electrode, etc.) can be associated with an Experiment, Paradigm, or Routine as user-configurable "metadata".

Predefine the Metadata parameter values here.

Show Summary   An overview of the user-defined metadata parameters.

Metadata Summary dialog

This list summarizes all Metadata parameters (from all Metadata detail levels) for which values have been defined. Double-click a line to show and edit the details for a parameter.

Metadata Parameter	Parameter name.
Current Value	Parameter value.
Increment Enabled	If enabled, double-click to review details.
Prompt before	Display metadata prompts before running:
▪ Expt	Experiment
▪ Pdgm	Paradigm
▪ Routn	Routine

Metadata Group   Listed per the SutterPatch > Set Preferences > 'Metadata detail level'.

Basic Level 1   (The default metadata groups.)

Preparation – Animal
Preparation – Tissue
Preparation – Cell
Experiment
Stimulus

Extended Level 2   (Includes two more metadata groups.)

Electrode
Recording Solution

Full Level 3   (Includes all metadata groups.)

Operator
Paradigm
Cell Health / Quality Control

Series (= Routine Data)

Metadata Parameter — The available metadata parameters are displayed by detail level, as set in SutterPatch > Set Preferences > Metadata.

[Parameter description]

<u>Configuration choices for the selected parameter</u>

- Do not write this parameter — This parameter is not stored.

  If this parameter was previously written in this Experiment, then its Previous Value is displayed.
- Use last value — The parameter used in the previous acquisition is written.
- Use a previous value — Select from a drop-down list of the previous 20 metadata values used for acquisition in the experiment.
- Use new value — Enter a new value for the metadata parameter.
- Increment — Numerically increment the value:

  By
  - Experiment — At the start of each Experiment.
  - Paradigm — At the start of each Paradigm.
  - Routine — At the start of each Routine.

  Prefix — Enter text to be prepended to the value

  Start value — The initial value (including decimals and negative numbers).

  Increment: — Select an arithmetic operator [+, -, *, /]

  [ ] — Enter the incremental amount.

  Suffix — Enter text to be appended to the value.

Prompt for confirmation (before):

- Experiment — At the start of an Experiment.
- Paradigm — At the start of a named Paradigm (i.e., pre-planned, not auto-triggered by a Routine).
- Routine — At the start of a Routine.

<u>Confirm Metadata Settings for</u> — This dialog displays whenever an Experiment, Paradigm or Routine is started with prompts enabled.

Write	Enable to write the selected metadata parameter with the Experiment, Paradigm or Routine.
Metadata Parameter	The name of the selected metadata parameter.
Next Value	The metadata value to write.
Update	Enable so that edits made to 'Value' will update the 'last value' for the next prompt. This field is automatically disabled after each execution.
Prompt	Disable to remove the metadata parameter from those listed in the Confirm Metadata Settings dialog. The 'Confirm Metadata Settings' dialog only displays if a metadata parameter has been enabled for 'Prompt'.

BASIC	EXTENDED	FULL	GROUP / Parameters	NOTES
		F	**OPERATOR /**	
		F	Full Operator Name	
B	E	F	**PREPARATION - ANIMAL /**	
B	E	F	Animal Identifier	
B	E	F	Animal Species	Binomial species name
	E	F	Animal Strain	Strain, breed or variety characterizing the animal
	E	F	Animal Genotype	
B	E	F	Animal Age	
B	E	F	Animal Age Units	Ex.: h, d, m
B	E	F	Animal Sex / Gender	Ex.: 1: F, 2: M, 3: Undetermined
B	E	F	Animal Weight	
B	E	F	Animal Weight Units	
B	E	F	Animal Circadian Time or Phase	
	E	F	Animal Preparation Date	ISO Date, Format: YYYY-MM-DD
	E	F	Animal Preparation Time	Time of Day, Format: hh:mm[:ss.000]
	E	F	Animal User Parameter 1 Name	
	E	F	Animal User Parameter 1	
	E	F	Animal User Parameter 2 Name	
	E	F	Animal User Parameter 2	
	E	F	Animal User Parameter 3 Name	
	E	F	Animal User Parameter 3	
	E	F	Animal User Parameter 4 Name	
	E	F	Animal User Parameter 4	

	E	F	Animal User Parameter 5 Name	
	E	F	Animal User Parameter 5	
B	E	F	**PREPARATION - TISSUE /**	
B	E	F	Tissue Preparation Identifier	
B	E	F	Organ	
	E	F	Organ Region	
	E	F	Preparation Method	
	E	F	Tissue Preparation Date	ISO Date, Format: YYY-MM-DD
	E	F	Tissue Preparation Time	Time of Day, Format: hh:mm[:ss.000]
	E	F	Tissue Incubation Duration	
	E	F	Tissue Incubation Duration Units	
	E	F	Tissue Incubation Temperature	
	E	F	Tissue Incubation Temperature Units	
	E	F	Tissue Incubation Solution	
	E	F	Tissue User Parameter 1 Name	
	E	F	Tissue User Parameter 1	
	E	F	Tissue User Parameter 2 Name	
	E	F	Tissue User Parameter 2	
	E	F	Tissue User Parameter 3 Name	
	E	F	Tissue User Parameter 3	
	E	F	Tissue User Parameter 4 Name	
	E	F	Tissue User Parameter 4	
	E	F	Tissue User Parameter 5 Name	
	E	F	Tissue User Parameter 5	
B	E	F	**PREPARATION - CELL /**	
B	E	F	Cell Preparation Identifier	
	E	F	Acutely Dissociated Cells	
	E	F	Cell Line	
	E	F	Slice Preparation	
	E	F	Whole-organ Preparation	
	E	F	In-situ Recording	
	E	F	Stem Cell Preparation	
	E	F	User-defined Preparation	
B	E	F	Cell Type	
B	E	F	Cell Identifier	
B	E	F	Cell Preparation Date	ISO Date, Format: YYY-MM-DD
B	E	F	Cell Preparation Time	Time of Day, Format: hh:mm[:ss.000]
	E	F	Cell Dissociation Solution	
	E	F	Cell Preparation Dissociation Temperature	
	E	F	Cell Prep. Dissociation Temperature Units	
B	E	F	Cell Preparation Incubation Duration	

B	E	F	Cell Prep. Incubation Duration Units	
B	E	F	Cell Preparation Incubation Temperature	
B	E	F	Cell Prep. Incubation Temperature Units	
B	E	F	Cell Preparation Incubation Solution	
B	E	F	Ion Channel	
	E	F	Cell Fluorescent Marker	
	E	F	Cell Diameter	
	E	F	Cell User Parameter 1 Name	
	E	F	Cell User Parameter 1	
	E	F	Cell User Parameter 2 Name	
	E	F	Cell User Parameter 2	
	E	F	Cell User Parameter 3 Name	
	E	F	Cell User Parameter 3	
	E	F	Cell User Parameter 4 Name	
	E	F	Cell User Parameter 4	
	E	F	Cell User Parameter 5 Name	
	E	F	Cell User Parameter 5	
B	E	F	**EXPERIMENT /**	
		F	Experiment Category 1 Name	
		F	Experiment Category 1	
		F	Experiment Category 2 Name	
		F	Experiment Category 2	
		F	Experiment Category 3 Name	
		F	Experiment Category 3	
		F	Experiment Category 4 Name	
		F	Experiment Category 4	
		F	Experiment Category 5 Name	
		F	Experiment Category 5	
B	E	F	Experiment User Parameter 1 Name	
B	E	F	Experiment User Parameter 1	
B	E	F	Experiment User Parameter 2 Name	
B	E	F	Experiment User Parameter 2	
B	E	F	Experiment User Parameter 3 Name	
B	E	F	Experiment User Parameter 3	
B	E	F	Experiment User Parameter 4 Name	
B	E	F	Experiment User Parameter 4	
B	E	F	Experiment User Parameter 5 Name	
B	E	F	Experiment User Parameter 5	
		F	Experiment Category Parameter 1 Name	
		F	Experiment Category Parameter 1	
		F	Experiment Category Parameter 2 Name	
		F	Experiment Category Parameter 2	
		F	Experiment Category Parameter 3 Name	

		F	Experiment Category Parameter 3	
		F	Experiment Category Parameter 4 Name	
		F	Experiment Category Parameter 4	
		F	Experiment Category Parameter 5 Name	
		F	Experiment Category Parameter 5	
	E	F	**ELECTRODE /**	
	E	F	Electrode Identifier	
	E	F	Electrode Glass Manufacturer	
	E	F	Electrode Glass Item Number	
		F	Electrode Glass Lot Number	
		F	Electrode Glass Material	
		F	Electrode Glass Item Outer Diameter	
		F	Electrode Glass Item Inner Diameter	
		F	Filamented Glass	
	E	F	Electrode Glass Ramp Test Value	
	E	F	Pipette Puller Manufacturer	
	E	F	Pipette Puller Model	
		F	Pipette Puller Serial Number	
		F	Puller Filament Type	
		F	Puller Filament Item Number	
		F	Pull Program Number	
		F	Pull Program Parameters	
		F	Pull Program Air Mode	
		F	Pull Program Air Pressure	
		F	Puller Heat-on Enabled	
		F	Puller Heat-on Time	
		F	Electrode Fire-polished	
		F	Electrode Coated	
		F	Electrode Coating Material	
		F	Electrode Beveled	
		F	Electrode Bevel Angle	
	E	F	Electrode User Parameter 1 Name	
	E	F	Electrode User Parameter 1	
	E	F	Electrode User Parameter 2 Name	
	E	F	Electrode User Parameter 2	
	E	F	Electrode User Parameter 3 Name	
	E	F	Electrode User Parameter 3	
	E	F	Electrode User Parameter 4 Name	
	E	F	Electrode User Parameter 4	
	E	F	Electrode User Parameter 5 Name	
	E	F	Electrode User Parameter 5	
	E	F	RECORDING SOLUTIONS	
	E	F	Solution Pair Identifier	

	E	F	Solution Pair Name	
	E	F	Bath Solution Identifier	
	E	F	Bath Solution Name	
		F	Bath Solution Batch	
		F	Bath Solution Composition	
		F	Bath Solution Preparation Date	
		F	Bath Solution Preparation Time	
		F	Bath Solution pH	
		F	Bath Solution pH Adjustment Agent	
		F	Bath Solution Osmolarity	
		F	Bath Solution Osmolarity Adj. Agent	
	E	F	Pipette Solution Identifier	
	E	F	Pipette Solution Name	
		F	Pipette Solution Batch	
		F	Pipette Solution Composition	
		F	Pipette Solution Preparation Date	
		F	Pipette Solution Preparation Time	
	E	F	Pipette Solution pH	
		F	Pipette Solution pH Adjustment Agent	
	E	F	Pipette Solution Osmolarity	
		F	Pipette Solution Osmolarity Adj. Agent	
		F	Liquid Junction Potential, computed	
		F	Liquid Junction Potential, measured	
		F	Solution User Parameter 1 Name	
		F	Solution User Parameter 1	
		F	Solution User Parameter 2 Name	
		F	Solution User Parameter 2	
		F	Solution User Parameter 3 Name	
		F	Solution User Parameter 3	
		F	Solution User Parameter 4 Name	
		F	Solution User Parameter 4	
		F	Solution User Parameter 5 Name	
		F	Solution User Parameter 5	
		F	**PARADIGM /**	
		F	Bath Temperature	
		F	Bath Temperature Units	
		F	Ambient Temperature	
		F	Ambient Temperature Units	
		F	Atmospheric Composition	
		F	Atmospheric Pressure	
		F	Atmospheric Pressure Units	
		F	Atmospheric Humidity	
		F	Paradigm User Comment	

		F	Paradigm User Parameter 1 Name	
		F	Paradigm User Parameter 1	
		F	Paradigm User Parameter 2 Name	
		F	Paradigm User Parameter 2	
		F	Paradigm User Parameter 3 Name	
		F	Paradigm User Parameter 3	
		F	Paradigm User Parameter 4 Name	
		F	Paradigm User Parameter 4	
		F	Paradigm User Parameter 5 Name	
		F	Paradigm User Parameter 5	
		F	**CELL HEALTH / QUALITY CONTROL /**	
		F	Cell Health User Parameter 1 Name	
		F	Cell Health User Parameter 1	
		F	Cell Health User Parameter 2 Name	
		F	Cell Health User Parameter 2	
		F	Cell Health User Parameter 3 Name	
		F	Cell Health User Parameter 3	
		F	Cell Health User Parameter 4 Name	
		F	Cell Health User Parameter 4	
		F	Cell Health User Parameter 5 Name	
		F	Cell Health User Parameter 5	
B	E	F	**STIMULUS /**	
	E	F	Key Stimulus	
	E	F	Stimulus Duration	
	E	F	Compound Group	
	E	F	Compound Group Index	
B	E	F	Compound Identifier	
B	E	F	Compound Name	
B	E	F	Compound Concentration	
B	E	F	Compound Concentration Units	
	E	F	Compound Batch	
	E	F	Compound Lot	
	E	F	Compound Salt Code	
	E	F	Compound Solution	
	E	F	Compound Vehicle / Solubility Enhancer	
	E	F	Compound Vehicle Concentration	
	E	F	Compound Vehicle Concentration Units	
	E	F	Compound Reservoir Identifier	
	E	F	Application Tip Identifier	
	E	F	Compound Plate Identifier	
	E	F	Compound Plate Row	
	E	F	Compound Plate Column	
	E	F	Chem. Stimulus User Parameter 1 Name	

	E	F	Chem. Stimulus User Parameter 1	
	E	F	Chem. Stimulus User Parameter 2 Name	
	E	F	Chem. Stimulus User Parameter 2	
	E	F	Chem. Stimulus User Parameter 3 Name	
	E	F	Chem. Stimulus User Parameter 3	
	E	F	Chem. Stimulus User Parameter 4 Name	
	E	F	Chem. Stimulus User Parameter 4	
	E	F	Chem. Stimulus User Parameter 5 Name	
	E	F	Chem. Stimulus User Parameter 5	
B	E	F	Light Stimulus Wavelength	
B	E	F	Light Stimulus Intensity	
B	E	F	Light Stimulus Intensity Units	
	E	F	Light Stimulus User Parameter 1 Name	
	E	F	Light Stimulus User Parameter 1	
	E	F	Light Stimulus User Parameter 2 Name	
	E	F	Light Stimulus User Parameter 2	
	E	F	Light Stimulus User Parameter 3 Name	
	E	F	Light Stimulus User Parameter 3	
	E	F	Light Stimulus User Parameter 4 Name	
	E	F	Light Stimulus User Parameter 4	
	E	F	Light Stimulus User Parameter 5 Name	
	E	F	Light Stimulus User Parameter 5	
B	E	F	Mechanical Stimulus Intensity	
B	E	F	Mechanical Stimulus Intensity Units	
	E	F	Mechanical Stimulus User Parameter 1 Name	
	E	F	Mechanical Stimulus User Parameter 1	
	E	F	Mechanical Stimulus User Parameter 2 Name	
	E	F	Mechanical Stimulus User Parameter 2	
	E	F	Mechanical Stimulus User Parameter 3 Name	
	E	F	Mechanical Stimulus User Parameter 3	
	E	F	Mechanical Stimulus User Parameter 4 Name	
	E	F	Mechanical Stimulus User Parameter 4	
	E	F	Mechanical Stimulus User Parameter 5 Name	
	E	F	Mechanical Stimulus User Parameter 5	
B	E	F	Acoustic Stimulus Frequency	
B	E	F	Acoustic Stimulus Intensity	
B	E	F	Acoustic Stimulus Intensity Units	
	E	F	Acoust. Stimulus User Parameter 1 Name	
	E	F	Acoust. Stimulus User Parameter 1	
	E	F	Acoust. Stimulus User Parameter 2 Name	

	E	F	Acoust. Stimulus User Parameter 2	
	E	F	Acoust. Stimulus User Parameter 3 Name	
	E	F	Acoust. Stimulus User Parameter 3	
	E	F	Acoust. Stimulus User Parameter 4 Name	
	E	F	Acoust. Stimulus User Parameter 4	
	E	F	Acoust. Stimulus User Parameter 5 Name	
	E	F	Acoust. Stimulus User Parameter 5	
B	E	F	Thermal Stimulus Temperature	
B	E	F	Thermal Stimulus Temperature Units	°C, °F or K
	E	F	Thermal Stimulus User Parameter 1 Name	
	E	F	Thermal Stimulus User Parameter 1	
	E	F	Thermal Stimulus User Parameter 2 Name	
	E	F	Thermal Stimulus User Parameter 2	
	E	F	Thermal Stimulus User Parameter 3 Name	
	E	F	Thermal Stimulus User Parameter 3	
	E	F	Thermal Stimulus User Parameter 4 Name	
	E	F	Thermal Stimulus User Parameter 4	
	E	F	Thermal Stimulus User Parameter 5 Name	
	E	F	Thermal Stimulus User Parameter 5	
B	E	F	Electrical Stimulus Frequency	The frequency of an external electrical stimulus
B	E	F	Electrical Stimulus Intensity	The intensity of an external electrical stimulus
B	E	F	Electrical Stimulus Intensity Units	The intensity units of an external electrical stimulus
	E	F	Electrical Stimulus User Parameter 1 Name	
	E	F	Electrical Stimulus User Parameter 1	
	E	F	Electrical Stimulus User Parameter 2 Name	
	E	F	Electrical Stimulus User Parameter 2	
	E	F	Electrical Stimulus User Parameter 3 Name	
	E	F	Electrical Stimulus User Parameter 3	
	E	F	Electrical Stimulus User Parameter 4 Name	
	E	F	Electrical Stimulus User Parameter 4	
	E	F	Electrical Stimulus User Parameter 5 Name	
	E	F	Electrical Stimulus User Parameter 5	
	E	F	Other Stimulus User Parameter 1 Name	
	E	F	Other Stimulus User Parameter 1	
	E	F	Other Stimulus User Parameter 2 Name	
	E	F	Other Stimulus User Parameter 2	
	E	F	Other Stimulus User Parameter 3 Name	
	E	F	Other Stimulus User Parameter 3	
	E	F	Other Stimulus User Parameter 4 Name	
	E	F	Other Stimulus User Parameter 4	
	E	F	Other Stimulus User Parameter 5 Name	

	E	F	Other Stimulus User Parameter 5	
		F	**SERIES (= ROUTINE DATA) /**	
		F	Routine User Comment	

Table 4-7. Metadata Parameters

4.2.19 Single Channel Analysis

Perform analysis of low-noise currents from single ion channels. When single channel analysis is activated, a Single Channel Analysis control panel is opened, and the Scope window active signal is overlaid with the transition levels of a single-channel opening or closing event.

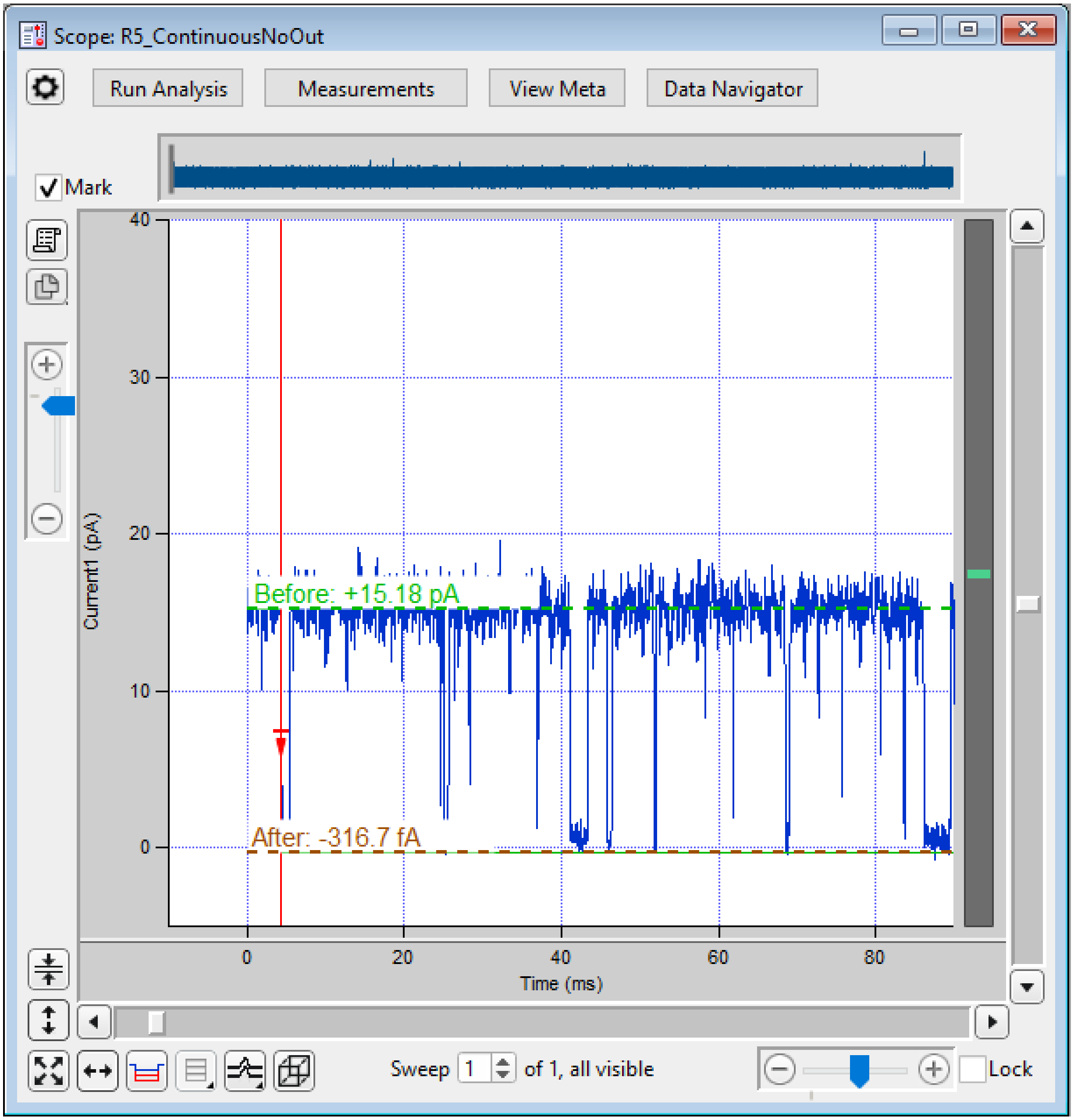


Figure 4-76. Single-Channel Scope

Access single-channel analysis via the Scope (analysis) window 'Measurements' button, or the Data Navigator (signal) 'Available actions' menu, or the main menu SutterPatch > Available Analysis Modules > Single Channel Analysis.

Note: Single-channel analysis only operates in the Scope 'Sweeps' display mode; the Concatenated and Time Course display modes are not supported.

Scope window Levels

- A dashed green "Before" line displays the amplitude of the previous event, i.e., the level before the transition point. Manually adjust by dragging with the mouse.

- A dashed brown "**After**" line displays the amplitude of the selected event, i.e., the level after the transition point. Manually adjust by dragging with the mouse.

- A solid green line displays the idealized trace of the found events.

  At times, the dashed amplitude lines might superimpose onto the idealized trace.

- A vertical red line displays at the transition point between the two levels, with a red arrow indicating the direction of the transition.

When the analysis is first started, amplitude levels for the first found (open or closed) channel are displayed. Event levels are automatically overlaid onto the raw data in the Scope window, based upon the settings in the Single Channel Analysis control panel / Current Transition Controls section. If the initial levels are incorrect, a couple basic settings need to be adjusted

1. Determine the starting amplitude of the data before the first transition.

   Zoom in on the Scope data, so that the open and closed state amplitudes are well visualized. Or run the 'Plots and Tables' 'Current Amplitude Histogram' to find the amplitude peaks in the binned data.

2. Set the 'Current Transition Controls' estimated 'Amplitude' value for the first Event transition in the data. (Use negative numbers for negative-going transitions.)

3. Set the Start Level number for the first transition (0 = baseline, 1+ = open states).

4. Click on the 'Find target transition' section 'Clear All' button.

5. The Scope window resets the "Before" and "**After**" transition levels to proper values.

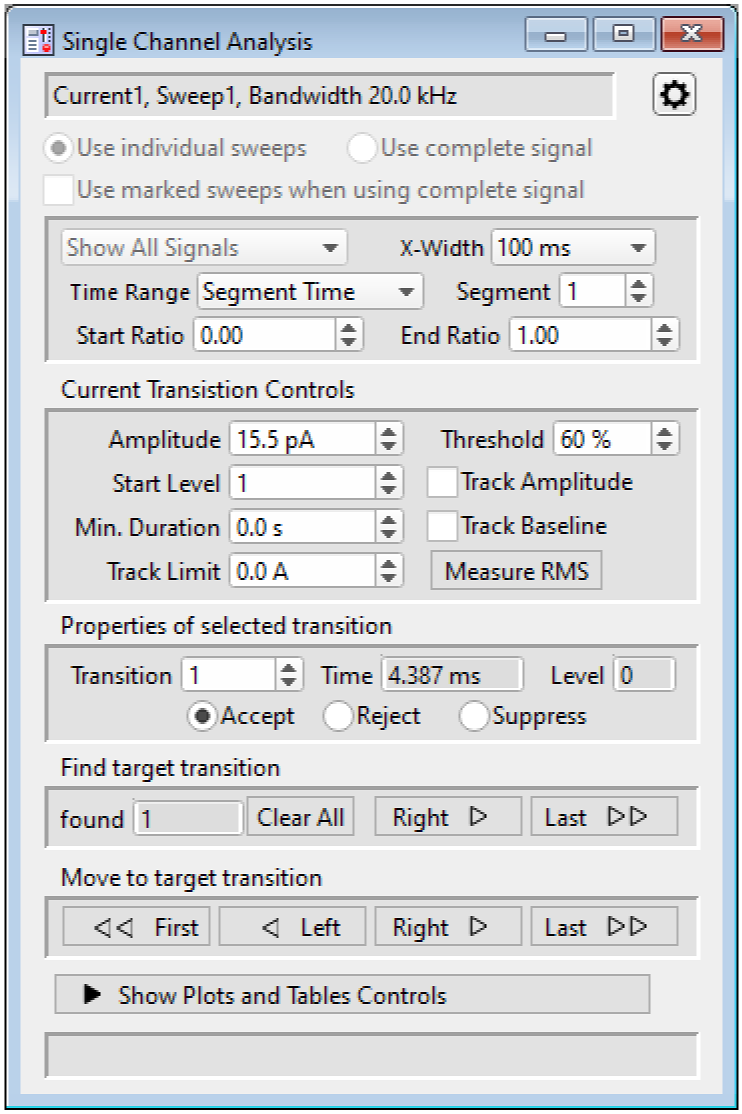


Figure 4-77. Single Channel Analysis

[Descriptive information]

- Use individual sweeps — Perform analysis on a single sweep at a time.
- Use complete signal — Perform analysis on all sweeps in the active signal.

[ ] Use marked sweeps when using complete signal

Perform analysis on a group of marked sweeps.

 Single Channel Analysis Preferences

Dock to Scope

Baseline Average Duration = x.x ms

Calculate the averaged baseline amplitude with the duration of the last baseline data (closed state) before the transition to an open state.

Mean Amplitude Duration = x.x ms

Calculate the mean amplitude using up to the duration of the open state data at that level, after a transition to that level.

Cell Separator: Tab

Cell Separator: Comma

Invalid values: Use NaN

Invalid values: Use empty string

Digits in table entries:

Digits = 3, 4, 5, 7, 9, 11, 13, 15

Signal Controls

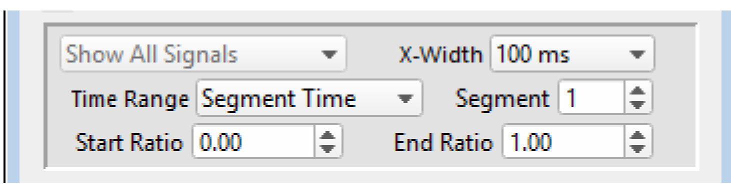


Figure 4-78. Signal Controls

Control the display of single channel data in the Scope window:

Show All Signals

X-Width: [Sweep / 1 s / 500, 200 ,100, 50, 20, 10, 5, 2, 1 ms]

The Scope window X-axis duration is reset to this value whenever a 'Find' or 'Move' transition operation is performed.

Time Range	Full Trace		
	Sweep Time	Start Time (s)	
		End Time (s)	
	Segment Time	Start Ratio	Relative to the start of a segment.
		End Ratio	Relative to the end of a segment.

Current Transition Controls

Setup the basic event level parameters.

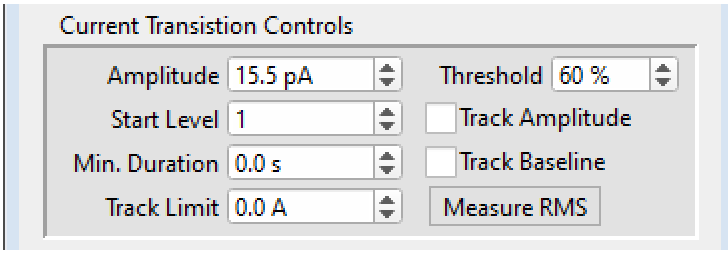


Figure 4-79. Current Transition Controls

Amplitude — Set to the expected transition size of the initial ion-channel level in the data.

For downwards-opening (negative) channels, use a negatively-signed value.

Start Level — The starting state of the ion-channel data:

Level 0	=	Closed State
Level 1	=	First Open State
Level 2	=	Second Open State
Etc.		

If there are multiple levels in the data, the program will try to automatically detect them. However, overlapping Events are treated as a single Event of the highest level.

Min. Duration — The minimum duration for a "found" event.

Track Limit — The maximum amount that an event Amplitude or Baseline level can change while being automatically tracked.

For baseline tracking, the absolute value must be > 0. Set manually, or set to 3 * RMS via the 'Measure RMS' button.

Threshold — [50 – 90%]

The percentile of the Amplitude value (open state) that needs to be reached by the raw data to "find" a transition.

Track Amplitude — Automatically adjust the predicted transition Amplitude of the next found transition, based on the prior data.

Track Baseline — Automatically adjust the baseline amplitude (Level 0) based on the prior data.

To use, the 'Track Limit' absolute value must be > 0.

Measure RMS — To measure the RMS (Root-Mean-Square) noise in the signal, adjust the signal trace in the Scope window such that it shows a stretch of current without any event activity (i.e., all channels are closed), then click on the 'Measure RMS' button.

The RMS value is displayed at the bottom of the dialog, and a 3 x RMS value populates the Track Limit field.

<u>Properties of selected transition</u>

View or alter how a transition is processed by the analysis.

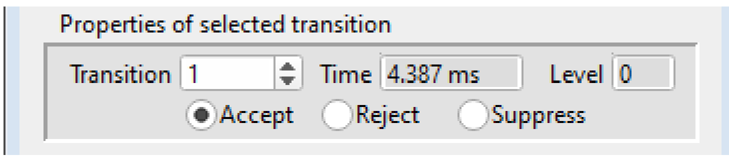


Figure 4-80. Properties of Selected Transition

Transition — The count number of the selected (active) transition.

Time — The start time of the selected transition.

Level — The level number of the selected transition.

Note: If other unexpected levels are detected "below the baseline", they are assigned a negative number. Negative levels might be detected from noise, biological artifacts, or a mischaracterized initial level that should be reset.

<u>Status</u> — <u>The operational status of the selected transition.</u>

- Accept — The selected transition is included in the idealized trace and all Plots. This is the default status.
- Reject — The selected transition is not included in Plots or the idealized trace.
- Suppress — The selected transition is not included in Plots or the idealized trace.

  Any events contiguous to a suppressed transition will not be included in Plots.

Find target transition

Find an event transition based on the 'Current Transition Controls', and process the transition based on the 'Properties of selected transition'. An event is defined by a sharp transition at the start of the event, for the duration of the transition level.

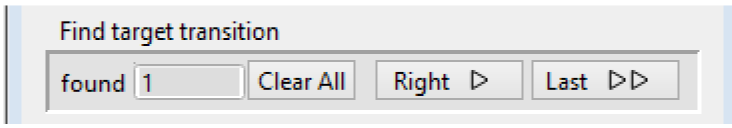


Figure 4-81. Find Target Transition

found	The total number of found transitions (including rejected and suppressed transitions).
Clear All	Reanalyze the data - reset the number of found transitions to zero, and move to the first found transition.
Right >	Find, move to, and process the next transition.
Last >>	Find and process all subsequent transitions, and move to the last transition.

Note: Multiple open levels are handled in a simplistic fashion. It is assumed that there is only one channel open, and that it is open for the same state throughout the duration of the Event.

Example:

Level 1 openings:	The Event duration is from the transition to the Level 1 amplitude, to the next transition to a different Level amplitude.
Level 2 openings:	The Event duration is from the transition to the Level 2 amplitude, to the next transition to a different Level amplitude.

Etc.

Move to target transition

Among the 'found' (processed) transitions, move to a "neighbor" transition, or jump to the beginning or ending transition.

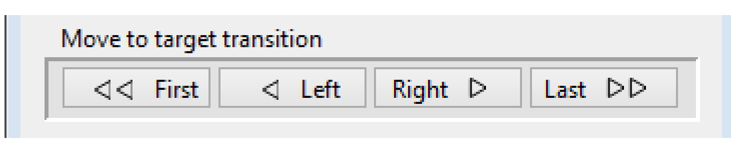


Figure 4-82. Move to Target Transition

<< First	Move to the first found event.
< Left	Move to the prior found event.

Right >	Move to the next found event.
Last >>	Move to the last found event.

Show/Hide Plots and Tables Controls

This button opens/closes the 'Tables and Plots' dialog, docked on the right of this dialog.

[   ] Status bar for the total number of events in a particular Plot.

Single Channels: Plots and Tables

Note: All plots and histograms support measuring X-Y data points or setting a fitting range via the 'Toggle Cursor Info' entry on their right-click menus. (See 'Right-Click Menus' for Scope windows.)

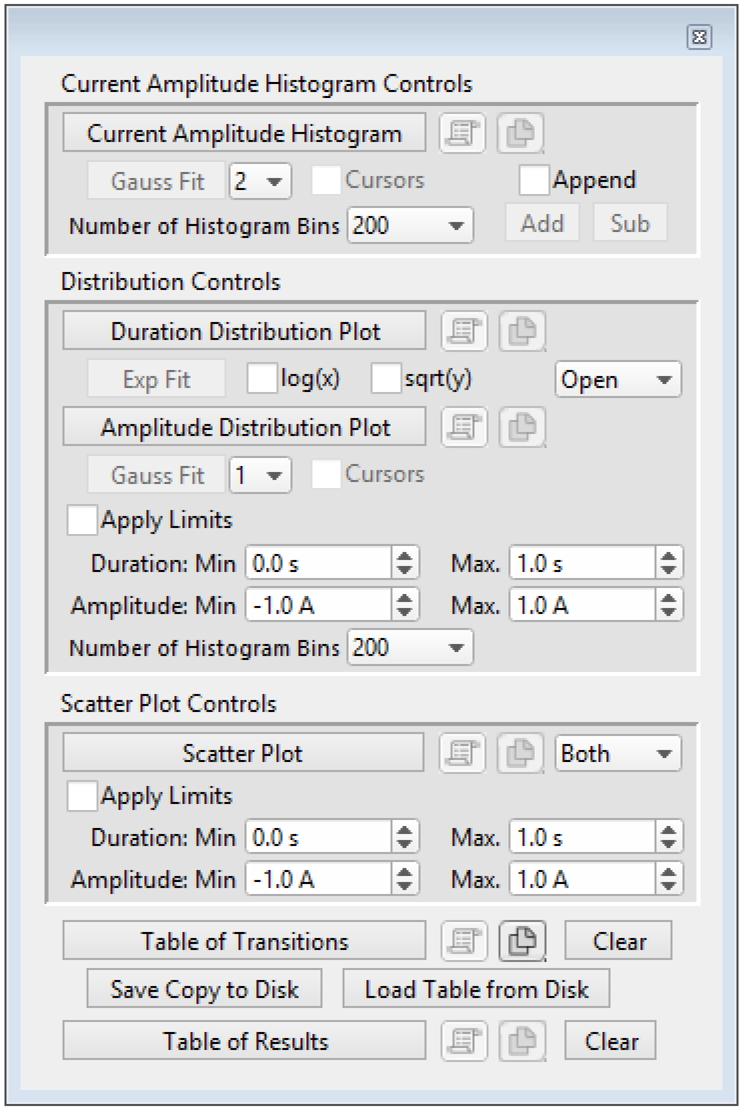


Figure 4-83. Plots and Tables Controls

Current Amplitude Histogram Controls

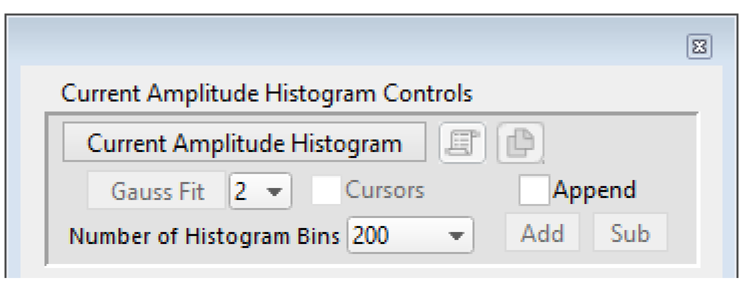


Figure 4-84. Current Amplitude Histogram Controls

A current amplitude histogram is often the first analysis performed on an uncharacterized channel, whereby all data points are binned by amplitude. It is used to determine:

- The quality of the recording.
- The number of levels in the open state.
- The first estimate of the open state amplitude(s).
- The first estimate of the baseline closed state.
- The frequency of openings.

Current Amplitude Histogram	Click to create a histogram plot of the raw data. No prior settings are needed to run this.
Copy to Layout Page	Copy the 'Current Amplitude Histogram' into a new Layout window, or append to an existing Layout page.
Copy to Clipboard	Copy the 'Current Amplitude Histogram' to the system clipboard. With the 'Shift' key pressed: as a graph, otherwise as a table.
Gauss Fit	Click button to perform a Gaussian fit on the histogram. SutterPatch will automatically find and fit up to the three largest peaks. You can the fitting ranges as needed with manual cursors. To fit additional smaller distributions, reposition the cursors and click the Append button. See the 'Table of Results' for the fitting components.
[1, 2, 3]	Select the number of peaks (levels) to fit. When more than one peak is selected, the graph reports amplitudes (in relation to the closed state peak) and P(open) and P(closed) values.
Cursors	Display fitting cursors in the plot window. To reposition cursors, first create the histogram, and then enable 'Cursors' – this will open a Cursor Bar at the bottom of the plot window. Next, drag the cursor's symbol (labeled "A", "B", etc.) from the cursor bar to the new position (near to the X-axis) on the plot.
Append	Modify the 'Current Amplitude Histogram'.
Add	Add the current data to the existing 'Current Amplitude Histogram'.

Sub — Subtract the current data from the existing ‘Current Amplitude Histogram’.

Number of Histogram Bins — [4000, 2000, 1000, 500, 200, 100, 50]

Distribution Controls

Create histogram plots of the found events.

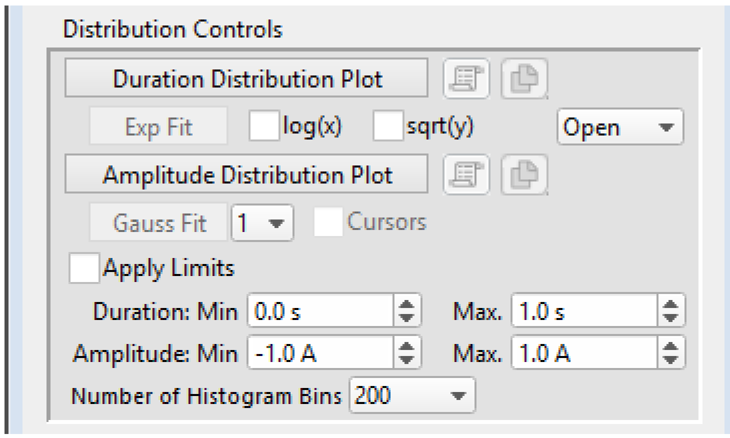


Figure 4-85. Distribution Controls

Duration Distribution Plot — Click to create a duration (dwell-time) histogram plot of the found Events.

 Copy to Layout — Copy the ‘Duration Distribution Plot’ into a new Layout window, or append to an existing Layout page.

 Copy to Clipboard — Copy the ‘Duration Distribution Plot’ to the system clipboard.

Exp Fit — Apply an exponential fit to the data.

log(x) — Set the X-axis to a log scale.

sqrt(y) — Use the square-root of the Y-axis data.

Open / Closed — Select open or closed state data for the Distribution plots.

Amplitude Distribution Plot — Click to create an amplitude histogram plot of the selected state’s found Events.

The Amplitude Distribution Plot bins “transition deltas”, which measures the *directional change* in amplitude for each transition (not the raw amplitude).

For example, an opening transition to 15 pA bins on the X-axis at 15 pA, while a following closing transition back to 0 pA bins on the X-axis at -15 pA, i.e., the delta of the transition's Before and After amplitudes.

The histogram bins plot as colored lines:

Open = **red**

Closed= **blue**

Copy to Layout — Copy the 'Amplitude Distribution Plot' into a new Layout window, or append to an existing Layout page.

Copy to Clipboard — Copy the 'Amplitude Distribution Plot' to the system clipboard.

Gauss Fit [1, 2, 3] Select the number of components in the fit.

Cursors — When an Amplitude Distribution Plot exists, you can enable draggable fitting cursors in the graph.

To position a cursor, drag its cursor symbol (labeled "A", "B", etc.) from the cursor bar onto the data at the desired position.

Apply Limits

Duration: Min [0.0 – 1.0 s]

Max. [0.0 – 1.0 s]

Amplitude: Min [-1.0 – 1.0 A]

Max. [-1.0 – 1.0 A]

Number of Histogram Bins [1000, 500, 200, 100, 50, 20]

Scatter Plot Controls

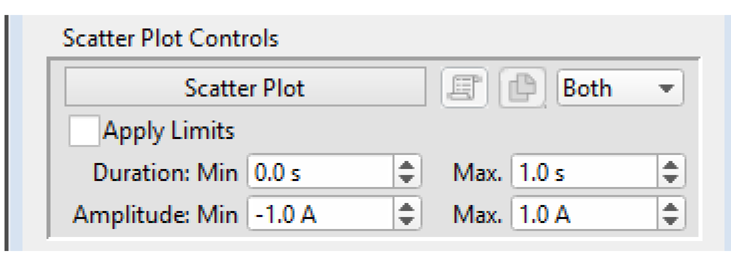


Figure 4-86. Scatter Plot Controls

The scatter plot uses "transition deltas" for event amplitudes, which plot on the Y-axis as the *directional change* in amplitude for each transition; the X-axis plots the duration of the event.

For example, an opening transition to 15 pA will plot on the Y-axis at 15 pA, while a following closing transition back to 0 pA plots on the Y-axis at -15 pA, i.e., the delta of the transition's Before and After amplitudes.

Events are plotted as colored symbols:

Open = **red**

Closed= **blue**

Selected event (transition) = green

Copy to Layout — Copy the 'Scatter Plot' into a new Layout window, or append to an existing Layout page.

Copy to Clipboard — Copy the 'Scatter Plot' to the system clipboard.

Open / Closed / Both — Select which states are plotted.

Apply Limits — Apply data limits to the Events used in "scatter" plots.

Duration: Min [0.0 – 1.0 s]

Max. [0.0 – 1.0 s]

Amplitude: Min [-1.0 – 1.0 A]

Max. [-1.0 – 1.0 A]

<u>Table Controls</u>

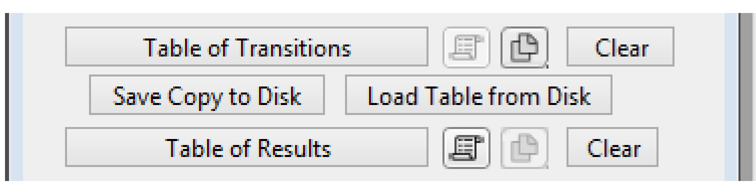


Figure 4-87. Table Controls

Table of Transitions — A listing of all Event transitions.

Header Row: Column names.

Column Header row: Event numbers.

Row 0: Status — The operational status of the selected transition.

1	=	Accepted	Include the selected transition in the idealized trace and all Plots. This is the default status
2	=	Rejected	Do not include the selected transition in the idealized trace or Plots.
3	=	Suppressed	Do not include the selected transitions in the idealized trace or Plots. Any Events contiguous to a suppressed transition will not be included in Plots.

Row 1: Time — Time of the start of the Event, i.e., the transition point.

Row 2: Level — The open or closed state level number.

Row 3: Amplitude_Before — Amplitude of the level preceding the Event, i.e., the level before the transition point.

Row 4: Amplitude_After — Amplitude of the Event, i.e., the level after the transition point.

Row 5: Duration — Duration of the Event.

The last column of data contains a zero duration

The column after the last column of data contains the total duration for all transitions, i.e., the total analyzed time.

Row 6: Amplitude — Amplitude of the Event.

Row 7: Amplitude_Valid — Include / Exclude the Event in processing

0	=	Invalid	Not a transition Event
1	=	Valid	A transition Event.

Note: The very first transition in a signal is defined to be 'Invalid' and cannot be used in analysis.

The very last transition in a signal is defined as 'Valid', and its timing can be used to end a previous transition; however, its "following" duration is set to '0'.

 Copy to Layout — Copy the 'Table of Transitions' into a new Layout window, or append to an existing Layout page.

Copy to Clipboard	Copy the 'Table of Transitions' to the system clipboard.
Clear	Clear all transitions from the table, and reset to the first found transition.
Save Copy to Disk	Save the 'Table of Transitions' to an Igor wave (*.ibw) file.
Load Table from Disk	Load the 'Table of Transitions' from an Igor wave (*.ibw) file.

Table of Results

Copy to Layout	Copy the 'Table of Results' into a new Layout window, or append to an existing Layout page.
Copy to Clipboard	Copy the 'Table of Results' to the system clipboard.
Clear	Clear all entries from the table, and reset to the first found transition.

Table Column Labels

Gaussian Fit

Header Row	Entry
Source	Sweep_#
Analysis	Current Amplitude Histogram Fit, or
	Amplitude Distribution Fit
Label1	Amplitude
Value1	#
Label2	p(closed)
Value2	#
Label3	p(open)
Value3	#
Label4	Gauss_y0_1
Value4	#
Label5	Gauss_A_1
Value5	#
Label6	Gauss_x0_1
Value6	#
Label7	Gauss_width_1

Value7	#
Label8	Success_1
Value8	#
Label9	Gauss_y0_2
Value9	#
Label10	Gauss_A_2
Value10	#
Label11	Gauss_x0_2
Value11	#
Label12	Gauss_width_2
Value12	#
Label13	Success_2
Value13	#
Label14	Gauss_y0_3
Value14	#
Label15	Gauss_A_3
Value15	#
Label16	Gauss_x0_3
Value16	#
Label17	Gauss_width_3
Value17	#
Label18	Success_3
Value18	#

Linear Exponential Fit

Header Row	Entry
Source	Sweep_#
Analysis	Duration Distribution Fit
Label1	Exp_y0
Value1	#
Label2	Exp_A
Value2	#
Label3	Exp_Tau
Value3	#
Label4	Exp_0
Value4	#

Label5	Success
Value5	#

Logarithmic Exponential Fit

Header Row	Entry
Source	Sweep_#
Analysis	Duration Distribution Fit
Source	Sweep_#
Analysis	Duration Distribution Fit
Label1	LogNormal_k0
Value1	#
Label2	LogNormal_k1
Value2	#
Label3	LogNormal_k2
Value3	#
Label4	LogNormal_k3
Value4	#
Label5	Success
Value5	#

4.2.20 Synaptic Event Analysis

Post-synaptic potentials and currents from excitatory and inhibitory events (EPSPs, EPSCs, IPSPs, IPSCs) are analyzed with this application module. Access via the Scope analysis window 'Measurements' button, or the Data Navigator (signal) 'Available actions' menu.

Spontaneous miniature events (mEPSPs, etc.), which generate small and often overlapping events, are detected with an innovative deconvolution algorithm. This technique finds Events with high temporal fidelity, while also improving the signal-to-noise ratio (SNR).

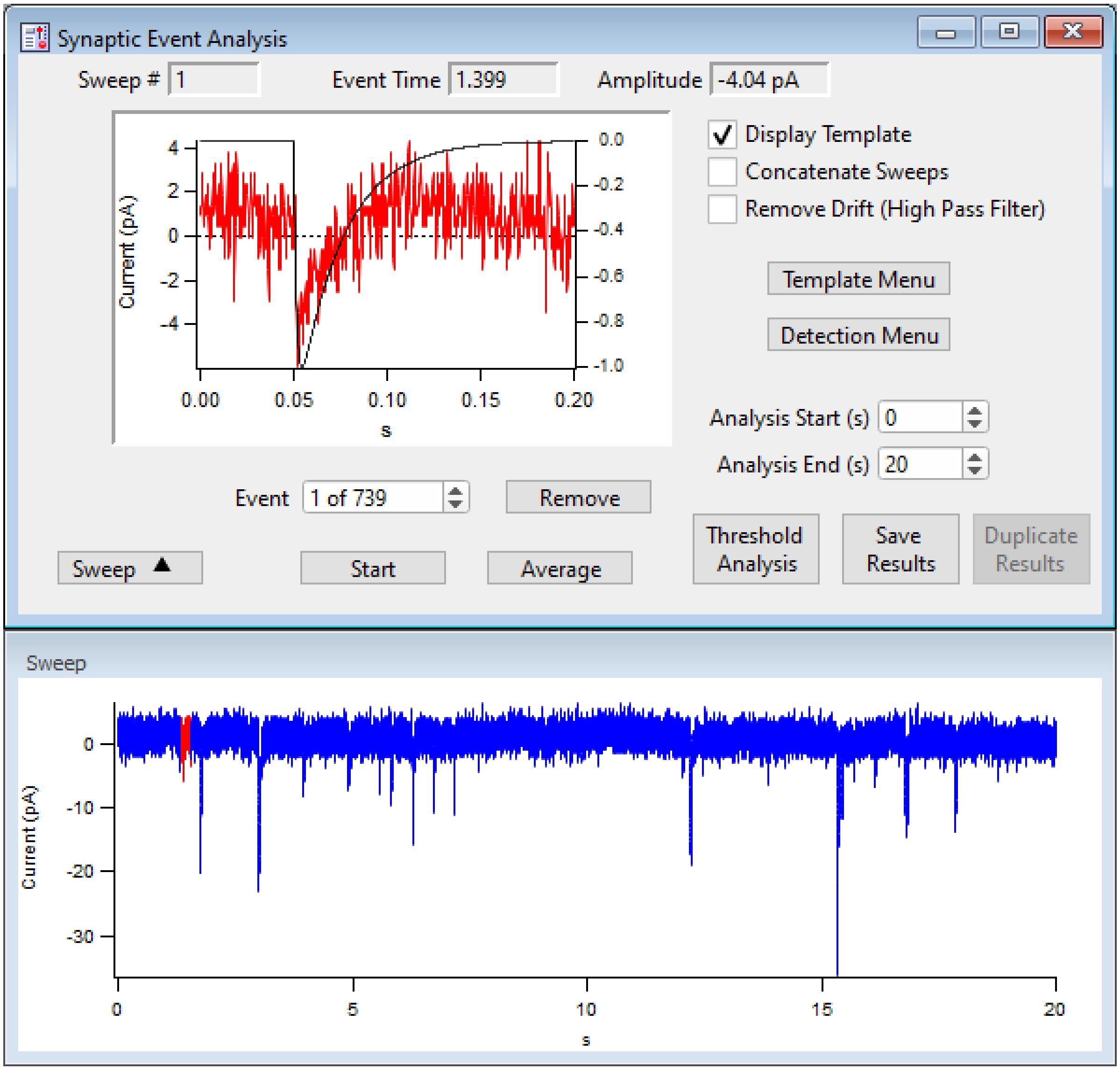


Figure 4-88. Synaptic Event Analysis

Sweep # — The sweep number of the displayed data.

Select arbitrary sweeps for processing by “marking” them in the Data Navigator tree (or during Scope acquisition/analysis.)

Event Time — The time (s)of the event’s threshold crossing relative to the start of the sweep.

Amplitude — The amplitude averaged around the peak by ±1 ms.

[Event pane] — A graph of the selected Event overlaid by the template, with the X-axis zero point reset to the template starting point.

To measure X-Y data points or set a fitting range, select ‘Toggle Cursor Info’ from the right-click menu. (See ‘Right-Click Menus’ for Scope windows.)

Display Template	Display the ideal event's template on top of the selected event in the graph - its Y-axis displays on the right edge of the graph.
Tip:	To match the template to the data, hover the mouse cursor over the right Y-axis, and use the mouse wheel to rescale the template.
Concatenate Sweeps	Combine all sweeps into a single pseudo-sweep before processing. This can sometimes improve performance of the detection algorithm signal-to-noise ratio with shorter sweeps.

Remove Drift (High Pass Filter)

A 1 Hz high-pass filter is applied to the signal to remove baseline drift.

Template Menu — Open the Template sub-panel to configure a template.

Create a template of a typical event as a double-exponential curve. The data will be deconvolved to this template for further analysis.

Event Polarity	[1, -1]	1 = positive -1 = negative
Rise Time (μs)	[10 – 5,000]	Time constant (τ) for the rising phase of the template event.
Decay Time (us)	[100 – 100,000]	Time constant (τ) for the falling phase of the template event.
Create Template	Click this button to create the template.	

Detection Menu — Open the Levels sub-panel to configure detection levels.

Threshold (xSD) — A detection threshold representing the "Event Strength". A lower ("weaker") number finds more events, while a higher ("stronger") number finds less events. Adjust this threshold based on empirical testing of your data.

[0.1 – 10]	Lower # =	more events	(false-positives)
	Higher # =	less events	(false negatives)

Note: The default threshold is set to 4 times the standard deviation of a Gaussian fit to an all-points histogram of the (Fourier) deconvolved data signal.

Ampl Threshold — [5e-13 – 1e-6]

Set an amplitude threshold for the minimum size of events.

Decay tau (us) — [<, >] [500 – 1,000,000]

Set the decay tau as "less than" or "greater than" the tau value.

Analysis Start (s)	Sweep time to start looking for an event threshold.
Analysis End (s)	Sweep time to stop looking for an event threshold.
Event	The current event number vs. total number of events.
Remove	Delete the current event from the analysis.
Sweep	Show/Hide the sweep preview panel.
[Sweep pane]	The graph of a sweep (blue) containing a selected Event (red). To measure X-Y data points or set a fitting range, select 'Toggle Cursor Info' from the right-click menu. (See 'Right-Click Menus' for Scope windows.)
Start	Find and analyze synaptic events.
Average	The averaged event displays in the graph.
Threshold Analysis	A scatter plot of the Event Strength vs. Current is displayed in a sub-panel. To measure X-Y data points or set a fitting range, select 'Toggle Cursor Info' from the right-click menu. (See 'Right-Click Menus' for Scope windows.)
Save Results	Reports are generated in a both a Layout page and a table.

Synaptic Event Analysis Results

The results are copied into their own Layout window, accessible via Windows / Layout Macros.

Series Name:	The path from the Igor internal root directory is displayed (see Data Browser.)
Total time analyzed = (s)	
	Includes the Start/End times for all sweeps
Number of events detected =	
	Total number of Events found.
Event Frequency =	(Hz)
Average Event Amplitude = (pA)	
	±1 ms peak average
Standard Deviation of Event Amplitude = (pA)	
Graphs:	Cumulative probability vs. Amplitude
	Amplitude (Average) vs. Time

Frequency vs. Sweep Number

Amplitude vs. Sweep Number

Results Table	A table of columns is created.
[blank]	Row number with one row per Event.
Sweep Number	The sweep number the Event is in.
Event Time (s)	'Time to event' from the start of the sweep.
Event Strength (xSD)	A measure of how well the signal matches the template. (Lower is weaker, higher is stronger.)
Event Amplitude (A)	The Event peak amplitude ± 1 ms average
Event Integral (A*s)	
10-90% Rise Time (s)	
Event Decay Tau (s)	
Absolute Event Time (s)	A continuous time scale from the start of acquisition, i.e., from the clicking of the 'Start' button, prior to the initial Sweep/Series external trigger.
Event Inter Event Interval (s)	

(See the SutterPatch Algorithms Appendix for a reference to the Event detection algorithm.)

4.3 General

SutterPatch general operations.

Note: Hidden unminimized windows can be brought into view with the menu command Windows / Control / Retrieve All Windows.

4.3.1 Command Window

This window is an Igor Pro code interpreter, providing programmatic interaction with SutterPatch. You can manually execute Igor Pro and user-defined assignments, functions and operations in this window.

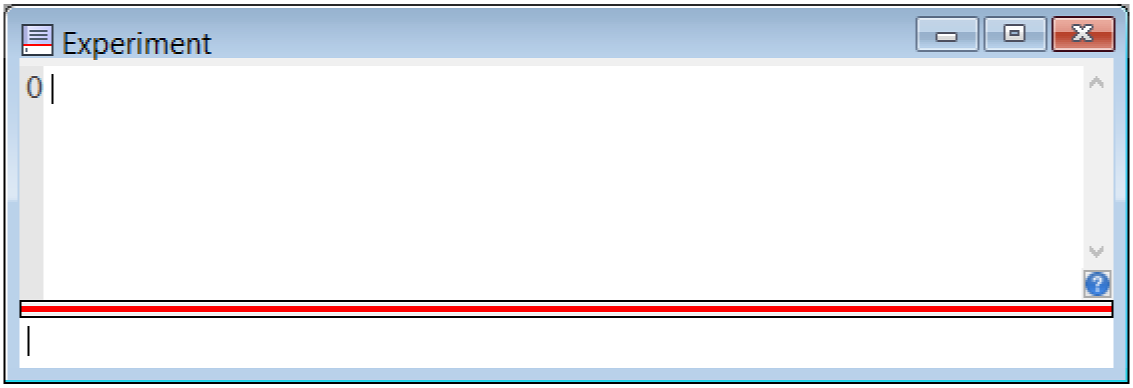


Figure 4-89. Command Window

The Command window is labeled with the currently loaded Experiment filename.

A history of commands and responses displays in the upper portion of the window. Some warning messages also display here.

The lower section is a command buffer with a "command line", where commands to be executed are entered. Commands can be entered into the command buffer in multiple ways:

- Manually type (or copy and paste) a line of text into the "command line" in the lower section of the window.
- Highlight lines in the history section and press the Enter key to copy them into the command buffer in the lower section of the window. To select the entire history, use 'CTRL-A'.
- Use the Paradigm Editor Execute step buttons 'Copy to Command Line' or 'Expand to Command Line' (for vars) to transfer the step command to the command line.

Commands in the command buffer are processed when the 'Enter' key is pressed.

A maximum of 400 characters can be entered into the command buffer, however they can be spread across multiple commands on multiple lines.

Note: Igor syntax usually requires that open/close parentheses "( )" be appended to the end of a command. However, exceptions include the "beep" and "print" commands, for which no parentheses are used.

The Command window has a resizing line between the upper history section and the lower command section – the mouse cursor will change to a double-headed arrow.

For more information, see Section II-2 of the Igor Pro manual.

4.3.2 Dashboard Panel

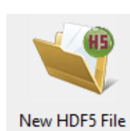
The Dashboard panel provides a convenient gateway to key areas of the SutterPatch program.

Figure 4-90. Dashboard

 Preferences

Icon Size	Large Icon
	Small Icon
Icon Orientation	Vertical
	Horizontal

Acquire Data	Live recordings and acquisition configuration.
Analyze Data	Review and analyze data in the Data Navigator.
View Last Data	Open the Experiment’s last recorded data Series.
Set Metadata	Configure metadata settings and values.
New Experiment	Start a new Experiment, and/or switch the amplifier model or emulation mode.
New HDF5 File	Start a new HDF5 file for the Experiment.



[Replaces the ‘New Experiment’ button when enabled in SutterPatch / Set Preferences / Files and Naming.]

Clicking the Acquire Data icon opens an adjoining secondary pane:

Figure 4-91. Dashboard – Acquire Data

Control Panel	Hardware control via the Amplifier Control Panel.
Membrane Test	Monitor seal formation and cell health.
Free Run	Run an oscilloscope-style signal monitor.
Routines	Configure Routine acquisition settings.
Paradigms	Control the execution of commands.

Note: While a Scope window is open, its icon remains depressed for Membrane Test / Free Run / Routine activation.

4.3.3 File Import/Export

Experiments & Data

There are two formats for saving an experiment. The packed format is recommended for most purposes.

- Packed experiment: (*.pxp file)

  A SutterPatch Experiment is saved by default as a "packed" (Igor Pro) experiment, which includes all data, analyses, graphs, routines, paradigms, etc., in one file.

  A Preferences option now allows data to be saved to an HDF5 file, which has the advantage of an unpacked experiment (like fast saving), without the disadvantage of much larger file sizes.

- Unpacked experiment: (*.uxp file, experiment Folder)

  A SutterPatch Experiment can also be saved as an "unpacked" (Igor Pro) experiment, which saves all waves, procedure windows, and notebooks as individual files in an experiment or "home" Folder, along with an instruction (*.uxp) file.to recreate the experiment.

  The advantage of an unpacked experiment is:

  1. Much faster processing of experiment recordings that include very large numbers of waves (thousands or more), as existing data waves are not resaved with each new recording.

The disadvantages of an unpacked experiment are:

2. Much more disk space is used, especially for experiments that have a lot of small waves.
3. The UXP format is more “fragile”, as you need to keep the experiment file and its corresponding folder together when you copy or move the experiment.

Saved experiments can be re-opened into the current experiment via the Data Navigator ‘Import’ button.

Note: If a SutterPatch Experiment file is opened into Igor Pro without SutterPatch running, its graphs and layouts can be displayed with the menu items Windows / Graphs, or Windows / Layouts, or Windows / Layout Macros.

Axon Files

Data can be selected and exported to the Axon Instruments pCLAMP file formats via the Data Navigator ‘Available Actions’ button or a right-click menu. Set to the ABF or ATF file formats in SutterPatch / Set Preferences / Data Export.

HDF5 Files

HDF5 is a modern efficient file format for saving and managing high volumes of data. Enable (*.h5) files for Experiments in SutterPatch / Set Preferences / Files and Naming.

HEKA Files

HEKA Elektronik PatchMaster Pulse Generator Files (*.pgf) can be opened in the Routine Editor ‘Pools and Files’ section and their Sequences merged with the current routine pool.

Igor Files

Data can be selected and exported to the Igor Pro file format (*.ibw) via the Data Navigator ‘Available Actions’ menu. Select the Igor Binary format in SutterPatch / Set Preferences / Data Export.

Igor binary waves (*.ibw) can be loaded into the current Experiment via Data / Load Waves / Load Igor Binary. Find the files in Data / Data Browser.

Template Files

Templates can be imported or exported via the Template Editor as Igor Binary Wave files (*.ibw).

Templates can also be used to export portions of data from a sweep.

Graphs and Layouts

Import graph files into or export from the current experiment via the Analysis Editor / Files options as Igor Binary Wave (*.ibw) files.

Note: Graph data for each axis can also be saved as Igor Pro 6 one-dimensional wave files, however files using this older format cannot be re-imported back into SutterPatch.

Graphs can be exported to a Layout window via:

Analysis Editor:	[To Layout] button
Scope window:	"Export a layout" button 
Paradigms:	Export step

Graphs exported to the Layout window use the SutterPatch / Set Preferences / Graphs and Layouts settings.

Individual graphs can be saved as Graph Macros - recall them via the Windows / Graph Macros menu.

Layout windows can be saved as Layout Macros - recall them via the Windows / Layout Macros menu.

4.3.4 Log Window

The Log window displays time-stamped commands, responses, administrative information and error messages that provide a history of the steps having a possible influence on the execution of the experiment and its data. The Log window can also serve as a user laboratory notebook for free-form entries.

At start up, the Log window displays the SutterPatch Version and Build numbers.

The following unnamed columns are used by the Log window:

Date & Time	Day name, month name, day date, year date, time: hours:minutes:seconds, AM/PM	
Event Type	Log entries are assigned an Event Type:	
	Data Acquisition	Acquisition operations, Membrane Test measurements.
	Metadata	Tags.
	Paradigm	Paradigm operations.
	Startup	SutterPatch version information.
	Unknown	Other operations.
Event Description	A text description of the log entry.	

Each row with a Data Acquisition, Metadata or Paradigm entry is appended with the name of the appropriate Routine or Paradigm; if there is no value to list, just the name of the Routine or Paradigm is displayed.

4.3.5 Menus

The SutterPatch main menu item contains all of the SutterPatch-specific menu items. The rest of the main menu items provide the standard Igor Pro functionality. For documentation of the non-SutterPatch features, refer to the Igor Pro online help or manual.

Window/Dialog Controls

Keyboard “Return” key	=	‘OK / Yes’ buttons
Keyboard ESC key	=	‘Cancel’ button

File

New Experiment	Unload the current Experiment and start a new Experiment. It is recommended that you create one Experiment per cell, to keep file sizes manageable.

Note: Whenever a new Experiment is started, you will be asked to re-save the old Experiment, even if there were no changes to it; this is required by an internal Igor cleanup process.

Open Experiment

Open a previously saved SutterPatch Experiment (*.pxp, *.uxp) file. If a SutterPatch experiment is opened into an Igor-only session, SutterPatch is automatically loaded.

If the SutterPatch preference for HDF5 files is enabled, a SutterPatch Question will ask how to load the matching HDF5 file:

- Load and store changes in analysis files

  Open the HDF5 file in read-write mode, i.e. the original metadata and experiment structure, analysis results, images, etc. are overwritten when closing the present experiment. However, raw data are NEVER modified.

- Load in read-only mode, i.e. don't store any change back to the file

  Open the HDF5 file in read-only mode. Nothing is stored, and the data file remains unaffected. Anything done in this session is lost when closing the Experiment.

- Cancel loading HDF5 file

  Do not open the HDF5 data file. Nothing is stored, and anything done in this session is lost when closing the Experiment.

  A normal Igor session is launched, that includes a main menu SutterPatch menu command 'Reactivate SutterPatch' to re-open the Experiment, followed by blank submenus.

Save Experiment

If the current experiment is already named, it is immediately saved. Otherwise, a 'Save experiment as' file dialog is displayed. If Preferences are enabled for automatic file naming, a default Experiment name is provided.

Stores the Experiment data and temporary (input / output) waves, but does no other cleanup; HDF5 files are not updated to disk.

Save Experiment As	If Preferences are disabled for automatic file naming, the last used Experiment file name or the default name is displayed for renaming and saving. This then stores the Experiment data and temporary (input / output) waves, but does no other cleanup; HDF5 files are not updated to disk. If Preferences are enabled for automatic file naming, an incrementing Experiment name is displayed for renaming and saving. However, nothing else in the Experiment gets saved.
Recent Experiments	A list of recently used Experiments.
Exit	An Experiment file 'Save' dialog is displayed before closing the program. If an Experiment is not saved, global variables and window sizes / positions are lost.
New HDF5 File	[Only displays when the HDF5 file in SutterPatch / Set Preferences / Files and Naming is enabled.] Stores all existing data into the present HDF5 file, cleans-up the Experiment, and creates a new HDF5 file, so that acquisition can continue as if you had started a "New Experiment", but without restarting SutterPatch.
Update HDF5 File	[Only displays when HDF5 file saving in SutterPatch / Set Preferences / Files and Naming is active.] Update the HDF5 data file without starting a new Experiment.
Compact HDF5 File	[Only displays when HDF5 file saving in SutterPatch / Set Preferences / Files and Naming is active, and data has been discarded.] Removes discarded data from an the HDF5 data file without resaving the entire Experiment.

Data

Load Waves / Packages / Install HDF5 Package

Load Waves / New HDF5 Browser

Data Browser	Access all SutterPatch objects contained in the experiment.

Analysis

The Analysis menu provides a wide assortment of mathematical transforms.

Curve Fitting	Create custom fitting equations.
Quick Fit	A variety of Igor fitting equations.

Windows

The Windows menu provides access to graphs, tables and layouts.

Control / Retrieve All Windows	Hidden unminimized windows can be brought into view with the menu command
Command Window	Process SutterPatch and Igor commands.

Layout

The Layout menu only displays when a Layout is the active window. Use it to modify the Layout window display and objects.

SutterPatch

Dashboard		Display icons for core program functions.
Acquisition Control		Open a control panel with Start/Stop and other interactive acquisition controls for Routines and Paradigms.
Scope Window		Bring an open Scope window to the front.

Hardware Control		
	Amplifier Control Panel	Open the hardware control panel.
	Reset Control Panel	Return the Amplifier Control Panel to its default settings.
	Lock-In Adjustments	Manually tune the "lock-in amplifier" system.
	Reset USB	Re-initialize USB communication with the computer. If in Demo mode, you need to start a 'New Experiment' to access 'Reset USB'.

Membrane Test		Open and run the Scope window to monitor seal formation and cell health.
Free Run (Scope)		Open and run the Scope window in oscilloscope style.
Reset Acquisition		Stop the Paradigm and/or data acquisition and clear corrupted acquisition settings.

Paradigm Editor		Open the dialog to load, edit and run Paradigms.

Routine Editor	Open the dialog to load and edit Routines.
Template Editor	Open the dialog to manage templates.
Equation Editor	Open the dialog to load and edit Equations.
Solution Editor	Open the dialog to control solutions.

Camera Module	Open the window to capture images.
Data Navigator	Open the window to organize and display the experiment Paradigm, Routine and acquisition data in a tree structure.
Analysis Editor	Open the dialog to manage analysis graphs.
Layout Page	Show Layout
	Delete Layout

Set Metadata	Open the dialog to configure user-specified experimental information.
Set Preferences	Open the dialog to modify the default program settings.
Log Window	Open the window to display a history of program actions.

Shortcuts	

Shortcut Editor	Open the Shortcut Editor dialog to manage keyboard shortcuts.

Action 1 [VHold+10mV:Right]	
Action 2 [VHold-10mV:Left]	
Action 3 [VHold+1mV:Right,shift]	
Action 4 [VHold-1mV:Left,shift]	
Action 5 [View last]	F2
Action 6 [Stop Acquisition]	F3
Action 7 [Start Routine]	F4
Action 8 [Stop Routine]	F5
Action 9 [Pause Paradigm]	F6
Action 10 [Resume Paradigm]	F7

Available Analysis Modules

- Action Potential Analysis
- Synaptic Event Analysis

Single Channel Analysis

Help

Igor Help Browser	Igor and SutterPatch Help Topics.
About SutterPatch	SutterPatch version and contact information.

Scope Right-click Menus

Different areas of the Scope windows support additional functionality through "right-click" menus in Windows, or "Command-click" menus in macOS.

Scope X-Axis (right-click the X-axis)

- Autoscale All Axes
- Full Scale Axis
- Axis Properties…

Scope Y-Axis (right-click the Y-axis)

- Autoscale All Axes
- Continuous Autoscale Axis
- Autoscale Axis
- Full Scale Y Axis
- Axis Properties…
- Hide Signal ‘<signal name>’
- Show Signal ‘<signal name>’ Only
- Stack All Signals

Scope (acquisition) main window To display a limited data modification menu, right-click the blank area in a signal.

Note: If you click too close to the data, the full data modification menu displays instead. If you are having this issue, click near a horizontal or vertical edge of the signal pane.

- Autoscale All Axes
- Add Annotation
- Export Graphics Copy the selected signal to a Graph window

- Toggle Cursor Info
  Manually measure X-Y data values or set a fitting range.
- Colors
- Hide Signal ‘<signal name>’
- Show Signal ‘<signal name>’ Only
- Stack All Signals

Scope (analysis) main window

To display a limited data modification menu, right-click in the blank area in a signal pane.

Note: If you click too close to the data, the full data modification menu displays instead. If you are having this issue, click near a horizontal or vertical edge of the signal pane.

- Autoscale All Axes
- Add Annotation
- Export Graphics
  Copy the selected signal to a Graph window
- Toggle Cursor Info
  Manually measure X-Y data values or set a fitting range.
- Colors
- Hide Signal ‘<signal name>’
- Show Signal ‘<signal name>’ Only
- Stack All Signals
- 3D View
- Zero Baselines
  The average of the first four data points is subtracted from the sweeps display. Does not affect data values in measurements
- Show All Sweeps
- Select Sweeps

Signal data

To display the full data modification menu, right-click on or near the data.

- Browse <signal name>
- Edit <signal name>
- Remove Sweep_#
- Hide Sweep_#
- Duplicate Sweep_#
- Replace Sweep_#
- Copy

- Modify Sweep_#
- Customize at Point
- Mode
- Line Style
- Line Size
- Markers
- Marker Size
- Color
- Bring to Front
- Send to Back
- Forward
- Backward
- Move to Opposite Axis
- Quick Fit
- Export Graphics
- Toggle Cursor Info — Manually measure X-Y data values or set a fitting range.
- Hide Signal ‘<signal name>’
- Show Signal ‘<signal name>’ Only
- Stack All Signals

Scope marquee window (click-and-drag in a signal)

- Expand
- Horiz Expand
- Vert Expand
- Shrink
- Horiz Shrink
- Vert Shrink

- Extract Template

4.3.6 Sample Files

Sample settings files (subject to change) are included in the ... / Documents / SutterPatch / Parameters folder:

<u>Dynamic Clamp Pool Sample Files</u>

SP_DynamicClampPool.spd	Dynamic Clamp models for dPatch systems.

<u>Equation Pool Sample Files</u>

SP_EquationPool.txt	Equations for all Sutter amplifier systems.
1. X3pi	3*pi
2. ElapsedTime	ParadigmTime - time
3. Temperature	aux[1]*1.23 – 273.15

<u>Paradigm Pool Sample Files</u>

LockIn / LockIn_dPatch.spp	Paradigms for dPatch lock-in tuning.
LockIn / LockIn_IPA.spp	Paradigms for D/IPA lock-in tuning.

1. LockIn_Adjust_500Hz
2. LockIn _ Adjust_1kHz
3. LockIn _ Adjust_2kHz
4. LockIn _DoAdjust

SP_ParadigmPool_dPatch.spp	Paradigms for dPatch systems.
SP_ParadigmPool_IPA.spp	Paradigms for D/IPA systems.
1. Amplifier_Setup	Set initial amplifier settings.
2. Start_one_Series	Start acquisition of one routine.
3. Set_amplifier_and_start_IV	Set amplifier to a known state, then start a routine.
4. Start_two_Series	Start acquisition of two subsequent routines.
5. Start_ForEachSweep	Start acquisition of a routine, individually triggering each sweep.
6. Interactive_acquisition_1	Run an interactive acquisition stopping at a given analysis condition.

7.	Interactive_acquisition_2	Run an interactive acquisition loop that selects between 2 routines, and manually stops via a Checkbox.
8.	Toggle_Persistence	Use a Checkbox to toggle Scope trace persistence while acquiring a routine.
9.	Switch_Headstages	Switch between multiple headstages.
10.	Tuning_with_Keys	Use the keyboard to increment or decrement a Routine's stimulus output by 10 mV.
11.	Tuning_with_Input	Use the paradigm "Input" control to increment or decrement a Routine's stimulus output.
12.	CellHealth_From_CC	Monitor the cell's resistance and capacitance in current clamp mode.

Routine Pool Sample Files

LockIn / LockIn_DIPA.spr — Routines for Double IPA lock-in tuning.

1. phase_delay
2. LockIn_500Hz
3. LockIn_1kHz
4. LockIn_2kHz

LockIn / LockIn_dPatch.spr — Routines for dPatch lock-in tuning.

LockIn / LockIn_IPA.spr — Routines for IPA lock-in tuning.

1. phase_delay
2. LockIn_500Hz
3. LockIn_1kHz
4. LockIn_2kHz

SP_RoutinePool.spr — Routines for IPA sytems.

1.	Amplitude Equations	Equation for a variety of stimulus waveforms.
2.	A_T_InactRec_P4	Inactivation with leak subtraction.
3.	Bowtie_Test	Multi-channel input with incrementing ramp waveforms.
4.	ContinuousNoOut	Acquisition without any output waveform.
5.	IV	I-V for voltage-clamp mode.

6. IV_CC	I-V for current-clamp mode.
7. IV_Continuous	I-V with continuous acquisition.
8. IV_P4	I-V with four leak-subtraction pulses.
9. IV_tuning	I-V for sample “tuning” paradigms.
10. Multi_Test	Multi-channel input with an incrementing square-step waveform.
11. Onset_SlowActivation	Onset Slow activation.
12. Recovery_Inactivation	Recovery from inactivation.
13. Recovery_SlowInact	Recovery from slow inactivation.
14. SS_Inactivation	Steady-state inactivation.
15. SS_SlowInactivation	Steady-state slow activation.
16. Synaptic_Stim	Synaptic stimulation.
17. Synaptic_Stim30	Synaptic stimulation for 30 s.
18. Synaptic_StimPlusDig	Synaptic stimulation with digital output.
19. Template_PlusVirtual	Template wave and recording virtual signals.
20. Template_SpontAct	Template wave from a recorded signal.
21. Template_Test	Template wave for waveform output.
22. Test_Pulse	Test pulse.

SP_RoutinePool_Dendrite.spr	Routines for Dendrite systems.

SP_RoutinePool_DIPA.spr	Routines for Double IPA systems.
1. Amplitude Equations	Equation for a variety of stimulus waveforms.
2. A_T_InactRec_P4	Inactivation with leak subtraction.
3. Bowtie_Test	Multi-channel input with incrementing ramp waveforms.
4. ContinuousNoOut	Acquisition without any output waveform.
5. IV	I-V for voltage-clamp mode.
6. IV_CC	I-V for current-clamp mode.
7. IV_Continuous	I-V with continuous acquisition.
8. IV_P4	I-V with four leak-subtraction pulses.
9. IV_tuning	I-V for sample “tuning” paradigms.
10. IV_VC_CC	IV for voltage- and current-clamp modes.

11. Multi_Test	Multi-channel input with an incrementing square-step waveform.
12. Onset_SlowActivation	Onset Slow activation.
13. Recovery_Inactivation	Recovery from inactivation.
14. Recovery_SlowInact	Recovery from slow inactivation.
15. SS_Inactivation	Steady-state inactivation.
16. SS_SlowInactivation	Steady-state slow activation.
17. Synaptic_Stim	Synaptic stimulation.
18. Synaptic_Stim30	Synaptic stimulation for 30 s.
19. Synaptic_StimPlusDig	Synaptic stimulation with digital output.
20. Template_PlusVirtual	Template wave and recording virtual signals.
21. Template_SpontAct	Template wave from a recorded signal.
22. Template_Test	Template wave for waveform output.
23. Test_Pulse	Test pulse.

SP_RoutinePool_dPatch.spr	Routines for two-headstage dPatch systems
SP_RoutinePool_dPatch_1HS.spr	Routines for one-headstage dPatch systems.

<u>ShortcutPool Sample Files</u>

SP_ShortcutPool.spo	Shortcuts for all Sutter amplifier systems.

1. VHold + 10mV	Increase the Control Panel V-holding level by 10 mV.
2. VHold – 10mV	Decrease the Control Panel V-holding level by 10 mV.
3. VHold + 1mV	Increase the Control Panel V-holding level by 1 mV.
4. VHold – 1mV	Decrease the Control Panel V-holding level by 1 mV.
5. View last	Open the last acquired Series into an analysis Scope window.
6. Stop Acquisition	Stop the acquisition of a Series.
7. Start Acquisition	Start the acquisition of a Series.
8. Stop Paradigm	Stop the execution of a Paradigm.
9. Pause Paradigm	Pause the execution of a Paradigm.
10. Resume Paradigm	Resume execution of a Paradigm.

Solution Pool Sample Files

SP_SolutionPool.spo	Solutions for all Sutter amplifier systems.
1. undefined	All solutions are blank.

Template Pool Sample Files

SP_TemplatePool.spt	Templates for all Sutter amplifier systems.

1. RoutinePreview
2. Single_actionPotential
3. HodgkinHuxley
4. Noise

Experiment Sample Files

Sample data (subject to change) are included in the ... / Documents / SutterPatch / Example folder:

ActionPotentials.pxp	Action potential data.
LargeAPs.pxp	Large action potential data.
MiniExample.pxp	Spontaneous miniature synaptic potential data.

4.3.7 Set Preferences

Preferences settings customize the default settings for several areas of the SutterPatch program. To access, go to the SutterPatch / Set Preferences menu.

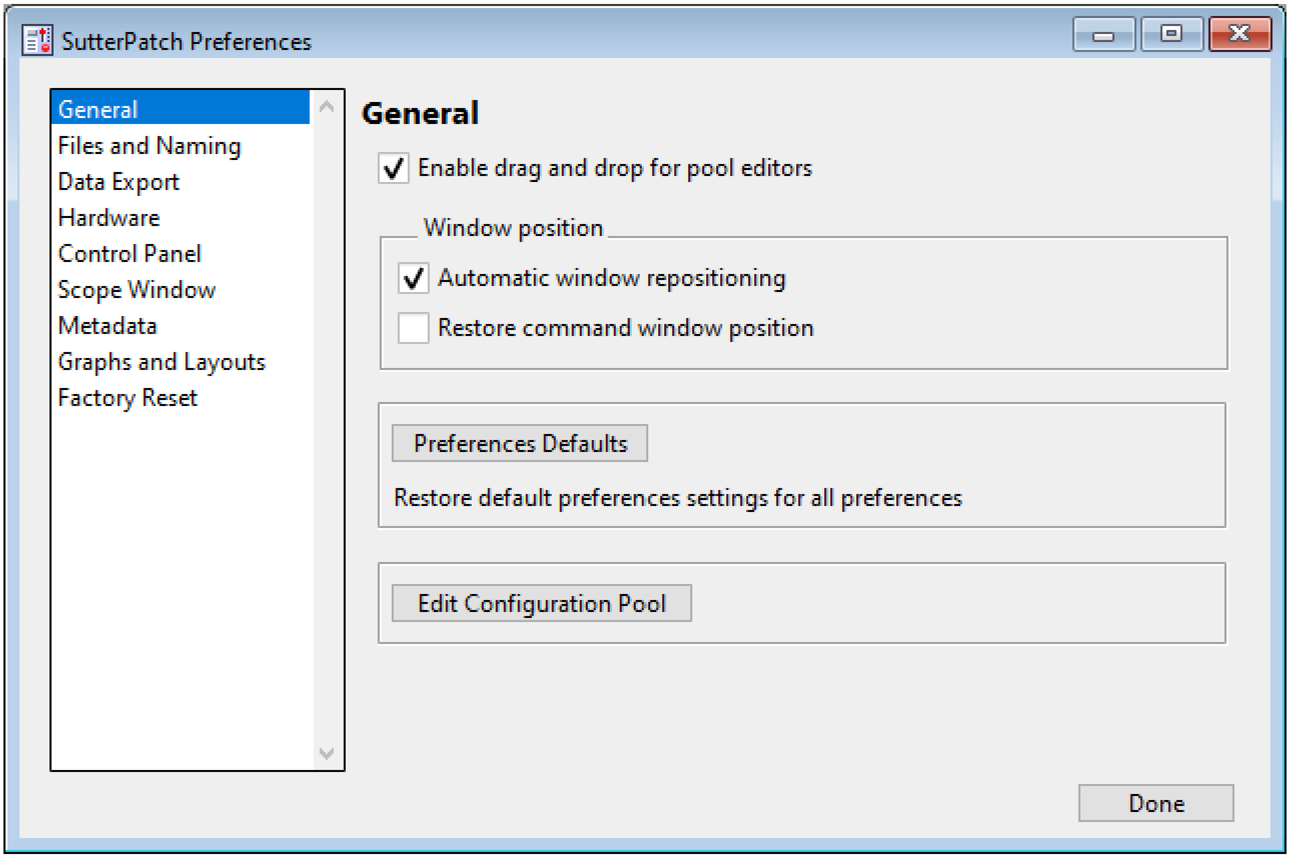


Figure 4-92. Preferences Settings

i. <u>General</u>

Enable drag and drop for pool editors

The Routine Editor loads a "pool" of Routines from a Routine Pool file. These Routines can be re-arranged in the Routine Pool list by clicking and dragging with the mouse.

The Paradigm Editor operates in the same manner, and also displays a list of Paradigm Steps, which can also be re-arranged by dragging-and-dropping.

Window Position

Automatic window repositioning

When SutterPatch windows or dialogs are opened or moved, when the action is done, they are automatically repositioned to be fully visible. If a “child” window is opened, the parent window is moved to the left until the child sub-window is fully visible or the parent window reaches the left edge of the main window/screen.

Restore command window position

Enable so the command window position is remembered for the next Experiment that opens. Otherwise, a new Experiment always returns the Command window to its default size and location.

Dual-monitor option [macOS only]

One screen Prevents a window spanning across monitors.

If a “parent” window is moved to another monitor, it fully displays in the new monitor, while any child sub-window remains behind fully displayed in the original monitor.

Note: You can also bring all unminimized windows into view with the Windows > Control > Retrieve All Windows menu command.

Preferences Defaults Restore default Preferences settings for all Preferences.

Edit Configuration Pool Edit and save user Preferences configurations.

Delete Configuration Remove the named Configuration from the list. The last remaining name is not deletable.

Add New Configuration Create a new Configuration name for the current Preferences settings.

Configurations are selected for loading at the start of a new experiment.

Description Enter the name for the Configuration (enclosed in double quotes).

Update Active Configuration (#)

The “active” Configuration is updated with the current Preferences settings.

This dialog opens with the active Configuration name listed. The number (#) indicates its position in the Configuration list.

Note: SutterPatch Configuration files use the *.spc file extension. However, this is reported by the OS as file type “PKCS #7 Certificates”.

ii. Files and Naming

Data file path: Browse to select a folder.

The data folder should reside on a local disk drive. We do not recommend using a mapped network drive, as speed/throughput bottlenecks can occur, including delays when saving experiments.

Enable automatic naming for experiment

Experiment file name example: (Maximum 34 characters)

[ ]

Note: At least one of the following file name components must be enabled.

Text: [ ]
Legal characters are A-Z, a-z, 0-9 and "_".

Include user text in the file name. At least one character is required.

Date: YYMMDD Include the date in the file name.

Time: hhmmss Include the time in the file name.

Save to separate HDF5 file Store the Experiment in HDF5 format, a modern efficient file format for managing and saving high volumes of data

By default, the Experiment file is stored in a "packed" (*.pxp) experiment, where all experimental information is conveniently stored in one file.

However, for very long experiments, this can result in delays when saving new data, as the entire experiment is re-saved with each additional recording.

Enable the HDF5 file option to avoid such delays. This setting become active after starting a New Experiment or restarting SutterPatch.

Then, whenever a new HDF5 file is created, SutterPatch stores all existing data into the active HDF5 file, cleans up the Experiment, and creates a new HDF5 file so that HDF5 acquisition can continue as if you had started a "New Experiment", but without starting a new SutterPatch session.

The raw signal data are stored to the HDF5 disk file during acquisition after each sweep, instead of storing all data at the end of an Experiment, which can be a time-consuming experience.

Other waves from the SutterPatch Data folder (including metadata, Experiment structure, analysis results, Routines, Log, images) are stored to the HDF5 file at the end of an Experiment; items outside of the SutterPatch Data folder (such as graphs and layouts) are stored to the ".pxp" Experiment file.

Note: It is strongly advised to enable the "automatic naming" option above, so that the '*.h5' file and its paired '*.pxp' Experiment file are kept "in sync".

Keep only one Sweep in Memory

[Only displays when HDF5 is enabled.]

For the leanest operation, only hold the wave of one sweep in memory, so

memory buffers do not need to be re-allocated for the Experiment.

The downside is that multi-sweep data cannot be processed online, such as subtracting the data of a reference sweep from other sweeps.

Update HDF5 file after each routine

[Only displays when HDF5 is enabled.]

The raw signal data are written during acquisition, while the other SutterPatch Data folder information (metadata, Experiment structure, analysis results, Routines, Log, images) are written to the HDF5 file at the end of the Routine.

<u>Save Options</u> [Only displays when HDF5 is disabled.]

- Save to temp file after each routine

The raw data are saved into a temporary file after each recording. This can help to speed up file-saving time for large Experiments composed of several smaller recordings.

The temporary file starting size is based on the starting size of the Experiment. The temporary data are re-saved to the main Experiment when the Experiment is closed and/or saved.

- Save entire experiment after each routine

  This default option re-saves the entire Experiment after each recording (all data and Experiment information). This is the safest method of operation for data integrity, but can produce significant post-recording file-saving delays in larger Experiments.

- Don't save to temp file after each routine

  Data and information are held in memory until the Experiment is explicitly saved; there are no file-saving delays after a recording is stopped. This provides the fastest method of operation when making multiple recordings, but is also the least secure, as data loss can occur if the computer encounters problems.

iii. Data Export

- Igor Binary — Save the signal formatted as an "Igor Binary Wave" file (*.ibw).
- ABF Format — Save each signal of the routine formatted as an "Axon Binary File" v.1.8 (*.abf).
  - Export all selected sweeps to one file per signal
    - Ignore unselected sweeps
    - Replace unselected sweeps with NaN
  - Create individual files for each sweep

  Note: If the new filename is the same as an existing filename in the target folder:

  - Choose a different folder, or
  - Delete the older file via the OS file browser.

- ATF Format — Save the signal formatted as an "Axon Text File" (*.atf).

iv. Hardware

- Prompt for hardware on startup (if no Sutter hardware is found).

  When a new Experiment is started, if Sutter patch-clamp hardware is not connected to the computer and turned on, you are prompted to retry the USB connection or select an emulation mode.

- If no Sutter hardware is found, emulate:
    - IPA — Integrated Patch Amplifier system.
    - Double IPA — Dual-headstage IPA system.
    - dPatch — Ultra-fast Digital Patch-clamp system.
    - Dendrite — Low-noise Data Acquisition system

  When a new Experiment is started, if Sutter patch-clamp hardware is not connected to the computer and turned on, automatically start up in the selected hardware emulation mode.

Stability Control

In CC mode reduce electrode compensation by [0.0 – 3.0] pF.

During whole-cell patching, if the Electrode Compensation control is set too high, oscillations can occur, and the patch-clamp seal can become unstable and be lost. As the Voltage Clamp mode typically operates with higher electrode compensation values than the Current Clamp mode, this preference promotes “safe” switching between the Voltage Clamp and Current Clamp modes.

If you are routinely losing cells when switching into Current Clamp mode, increase this setting from the default ‘0.5’ to ‘1’ or ‘2’.

Note: The electrode compensation reduction is done in the background, and does not affect the Control Panel current-clamp settings.

v. Control Panel

Customize the active headstage tab's color in the Control Panel.

Headstage background color

Headstage 1	Color palette displays.
Headstage2	Color palette displays.

vi. Scope

Scope setting changes are only applied when a Scope window is opened.

General

Time axis unit:

- Auto-set Sweep duration < 120 s, use "s"
  Sweep duration >= 120 s, use "min"
  Sweep duration >= 7200 s, use "h"
- SI unit Always use standard SI base units, such as "s" for time.

Acquisition

Y axis initial settings This setting is applied when a new Scope window is created. If autoscaling is selected, it is also applied to the Scope Y-axis and Y-axes autoscale buttons.

- Continuous autoscale
  The Y-axes limits are rescaled with each sweep so that all data are visible.
- Continuous autoscale from zero
  The Y-axes limits are rescaled with each sweep so that all data are visible. If the signal does not cross the zero line, the Y-axes ranges are extended to the zero amplitude,
- Full scale The full-scale range is used.
- Use last y-scale The last setting of the Scope window Y-axis scaling is used.

Maximal sweeps displayed in persistence display

[2 – 100]

All

Improve data readability and system performance by restricting the number of sweeps displayed, which reduces the display processing load.

Allow Display Compression Display compression is applied to the data in all "live" Scope windows (Acquisition / Free Run / Membrane Test). This reduces the display processing load, and can improve system performance when resources are low.

When there are four times as many data

points as the Scope width in pixels, the number of data points plotted are reduced, as the minima and maxima from two groups of up to 50 samples are displayed per screen pixel.

Reanalysis

Y axis initial settings — This setting is applied when a new analysis Scope window is created. If autoscaling is selected, it is also applied to the Scope Y-axis and Y-axes autoscale buttons.

- Autoscale — The Y-axis limits are rescaled with each sweep so that all data are visible.
- Autoscale from zero — The Y-axis limits are rescaled with each sweep so that all data are visible. If the signal does not cross the zero line, the Y-axis range is extended to the zero amplitude, Full scale The full-scale range is used.
- Use last y-scale — The last setting of the Scope window Y-axis scaling is used.

Maximal sweeps displayed in persistence display

[2 – 100] (30 = default value)

The last 'N' sweeps are displayed

[All] All sweeps are displayed.

Improve data readability by restricting the number of sweeps displayed. This can also improve system performance by reducing the display processing load.

Allow Display Compression — Display compression is applied to the data in the Scope analysis window. This reduces the display processing load, and can improve system performance when resources are low.

When there are four times as many data points as the Scope width in pixels, the number of data points plotted are reduced, as the minima and maxima from two groups of up to 50 samples are displayed per screen pixel.

Note: Display compression is not applied to Single Channel Analysis.

Show event tags — Display tag lines in the Scope reanalysis window; also display tag text boxes in the Continuous and Concatenated display modes.

Tag Position

- Frozen
- Movable

Tag types to show

- User
- Input-triggered
- System

Tag text box

- Time
- Tag sample
- Description

Tag appearance

- Color by type
- Transparent

Appearance — Use a color palette to choose colors.

Active signal panel color — [light gray]

Inactive signal panel color — [dark gray]

Active sweep color — [blue]

Inactive sweep color — [red]

Drop-down color palette — Click on a color square to set it as the active color.

Other — Use a color dialog with more options.

Opaque — < unused >

[Preview Pane]	The selected signal and sweep colors are displayed in a preview pane.

Tip: For dark-room experiments, the window background color can be adjusted by the operating system:

- Windows: In the Control Panel / Appearance / Personalization window, scroll down and select the High Contrast Black theme, or use the Windows Magnifier tool with option 'Turn on color inversion' enabled.
- macOS: Press 'Control-Option-Command-8' to set the System Preferences / Accessibility / Display / Invert Display colors option, or open its menu with 'Command-Option-5'.

vii. Metadata

Metadata detail level: Select levels for metadata setup categorization complexity.

Select which metadata groups and parameters are visible for configuration in Set Metadata and in Data Navigator / Build Hierarchy.

1	Basic	Show only the most essential parameters.
2	Extended	Show additional detail.
3	Full	Expose all available metadata parameters.

Note: This setting does not affect the Metadata Review dialog - all defined metadata are displayed.

This setting does not affect data acquisition metadata prompts – all configured prompts are always executed.

viii. Graphs and Layout

These settings apply to Layout windows and stand-alone graph windows (not graph files).

Copy "To Clipboard" graph format

Several popular file formats are supported:

- PNG Portable Network Graphics
- PDF Portable Document Format

- TIFF Tagged Image File Format
- JPEG Joint Photographic Experts Group

Note: When pasting, not all formats may be supported by other programs.

Layouts

Changes are applied when a new Layout window is created.

Default graphs per new layout page

- 1
- 2
- 3
- 2 x 2 (Column x Row)
- 2 x 3 (Column x Row)
- 2 x 4 (Column x Row)

Graphs

left (typically the Y-axis)

Tick location:

- Outside
- Crossing
- Inside
- None

Labels:

- On
- Axis only
- Off

Grid:

- Off
- On
- Major only

bottom (typically the X-axis)

Tick location:

- Outside
- Crossing
- Inside
- None

Labels:

- On
- Axis only
- Off

Grid:

- Off
- On
- Major only

ix. Factory Reset

[Factory Reset] Click this button to reset SutterPatch to it default settings. SutterPatch will need to be exited and restarted to complete the factory reset.

Warning! When you do a factory reset, you will lose ALL your configuration data (including Metadata, Scope, Preferences, etc.)

4.3.8 Shortcut Editor

Keyboard control of SutterPatch is available by configuring keyboard shortcuts.

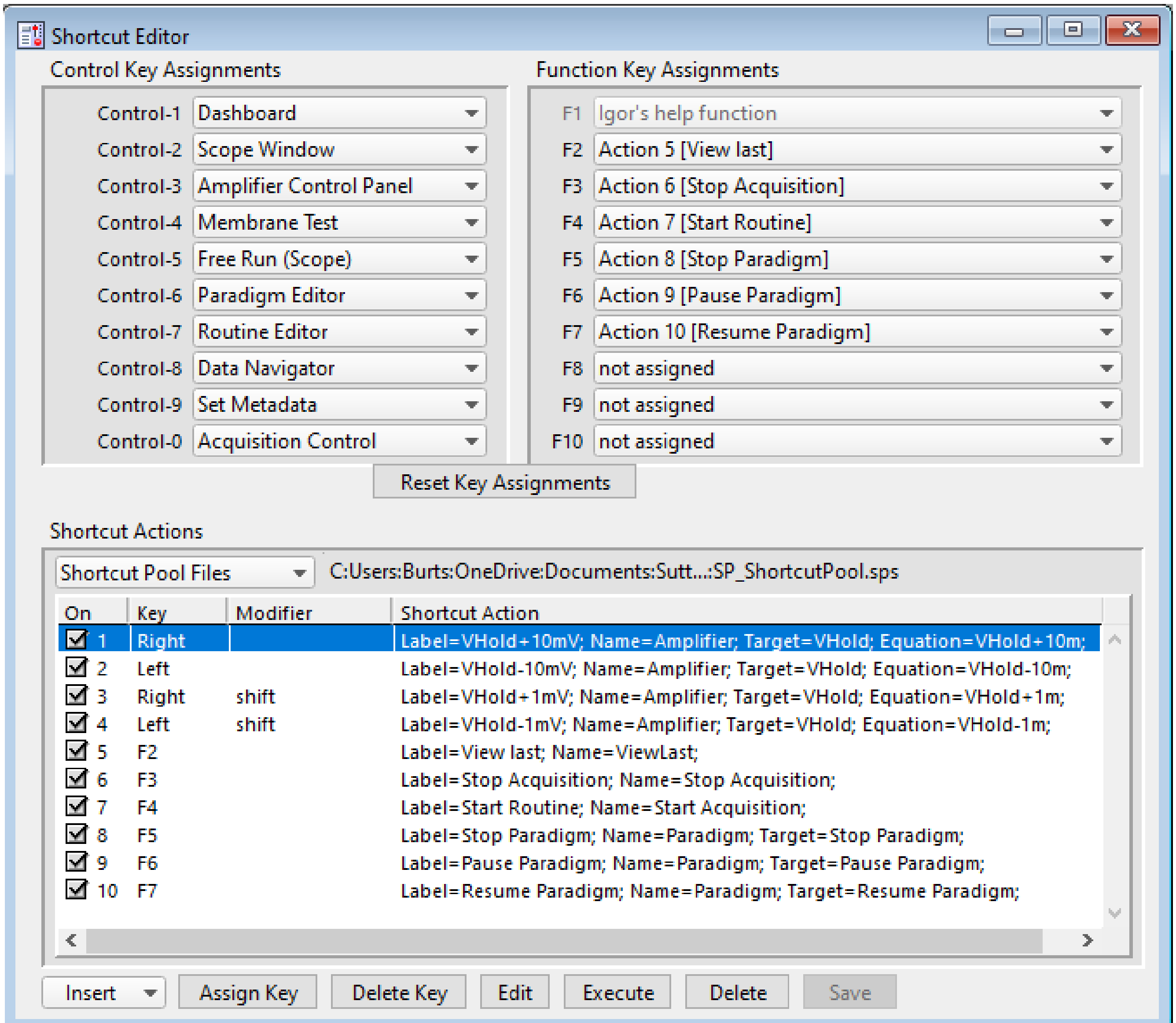


Figure 4-93. Shortcuts Editor

All assigned Control (Command), Function and Shortcut Action key combinations are automatically added to the main menu SutterPatch / Shortcuts submenu. All Control and Function Key Assignments are automatically saved when the program is closed.

The ShortCuts main menu list is shared by the Control and Function keys, so each menu item can only be .assigned to a single key., Prior duplicate entries are replaced by “not assigned”.

Control Key Assignments / Command Key Assignments

A Control key assignment can be easily made by clicking on its drop-down list and selecting a new item from the list of SutterPatch menu items.

Use Control keys by holding down:

- Windows: Control key: Ctrl
- macOS: Command key: ⌘

and clicking its assigned number.

Control keys and their default SutterPatch menu assignments:

Control-1	Dashboard
Control-2	Scope Window (Scope window must be open.)
Control-3	Amplifier Control Panel
Control-4	Membrane Test
Control-5	Free Run (Scope)
Control-6	Paradigm Editor
Control-7	Routine Editor
Control-8	Data Navigator
Control-9	Set Metadata
Control-0	Acquisition Control

Other SutterPatch menu items available for assignment:

Analysis Editor

Camera Module

Equation Editor

Lock-In Adjustments

Log Window

Reset Acquisition

Set Preferences

Shortcut Editor

Solution Editor

Template Editor

Function Key Assignments

Computer keyboards usually include a set of Function keys [0 – 9] for special functionality. Configure a 'Function Key' assignment by clicking on its drop-down list and selecting a new menu item or Action item.

Function keys and Control keys share the same menu items list, and each menu item can only be only be assigned to a single key. So, after any new assignment, any duplicate key is changed to "not assigned".

Function keys can also be assigned from Shortcut Actions – see below.

Note: On some keyboards, you also need to press the 'Fn' key to use Function keys.

Also, macOS reserves nearly all Function keys for itself. In order to use Function keys for a macOS application, you must first check a checkbox in the macOS Keyboard control panel. Even then, macOS will intercept some Function keys.

Function Key F1 is not available for assignment, as it is reserved for Igor's help function.

Default Functions

F2	Action 5	[View last]
F3	Action 6	[Stop Acquisition]
F4	Action 7	[Start Routine]
F5	Action 8	[Stop Routine]
F6	Action 9	[Pause Paradigm]
F7	Action 10	[Resume Paradigm]
F10	Action 11	[Cursor Info]

Reset Key Assignments

Reset all Control to their default settings.
Function key "menu" items are reset to 'not assigned', while 'Action' items are unaffected.

Shortcut Actions

Up to 10 additional custom keyboard Actions can be created, managed and stored in a file.

Shortcut Pool Files

New Shortcut Pool	Create a default Shortcut Pool.
Load Shortcut Pool	Load the Shortcuts of a previously saved Shortcut Pool file into the Shortcut Pool.

Revert to Last Saved	Undo any unsaved changes to the Shortcut Pool.
Save Shortcut Pool	Save the Shortcut Pool using its existing file name and path.
Save Shortcut Pool As…	Save the Shortcut Pool to a new file, and switch to the new file. The default file name is the same as the original file name.
Save Shortcut Pool Copy…	Save the Shortcut Pool to a new file, but do not switch to the new file. The default file name has 'Copy of" prepended to it.
Merge Shortcut Pools	Append the Shortcut Actions from a previously saved Shortcut Pool file into the loaded Shortcut Actions table.

[Shortcut Pool file path]

<u>Shortcut Actions Table</u>

Columns

On	Enable/disable the Shortcut Action.
Key	The assigned keyboard key.
Modifier	The keyboard "modifier key" used in a key combination - all keys are simultaneously pressed.

.

<u>Windows</u>

• Ctrl	Only for use with keys '0 – 9'.
• Alt	Keys '0 – 2' reserved by Igor for File / Recent Experiments.
• Shift	Shift key.
• Fn	Function key.
• Caps Lock	Ignored.

<u>macOS</u>

• Command	Only for use with keys '0 – 9'.
• Option	Option key.
• Shift	Shift key.
• ctrl	Keys '0 – 2' reserved by Igor for File / Recent Experiments.

- Caps Lock   Ignored.

Shortcut Action	An Action's instructions and settings.

Click a field in the pool to highlight an Action and make it the active entry. Click-and-drag a field to reposition an Action in the pool.

Insert — Adds an Action to the 'Shortcut Action' list and opens its Shortcut Actions Editor for setup.

These Actions operate similarly to Paradigm steps, with an additional Label field to name the Action in the Shortcuts menu.

Amplifier	Control an IPA amplifier's settings.
Analysis	Append, average, display and save analyses.
Camera	Take a photo or run live video.
Execute	Run an Igor or SutterPatch command.
Export	Send graphs to the Layout window.
Front Window	Set the specified window as the front window.
Hide Window	Hide the specified window.
Paradigm	Load & Run, Stop, Pause or Resume a Paradigm.
Reset Timer	Reset the Paradigm Editor Timer to zero.
Routine	Record data from a Routine.
Scope Operation	Control the display of the Scope window signals and sweeps.
Set Axis	Modify the axis scaling of a signal.
Set Checkbox	Set local and global checkboxes for conditional processing in Paradigm 'If' steps.
Set Mark	The selected sweep in the Scope (analysis) window is "marked" so that it is highlighted for group averaging or export in the Data Navigator.
Set Variable	'Label' entry only displays in the Shortcut Action column.
Set Tag	Write a comment tag to the Paradigm metadata.
Start Acquisition	In an open Scope (acquisition) window.
Stop Acquisition	In an open Scope (acquisition) window.

View Last	Display the last recording in a Scope (analysis) window.
Write Log	Write a note to the Log window.

Assign Key — This button opens the Shortcut Key Input dialog (or double-click in a “Key” or “Modifier” field) to input the desired keyboard combination for a letter, number, or symbol.

Note: Available keyboard letters, numbers, and symbols can vary from computer to computer, depending on the computer OS and Igor Pro’s key usage. (Reserved keys typically open another window type, or are non-responsive.)

Tips: If the CAPS LOCK button is on when assigning a key, the key is case insensitive.

Keyboards often have a Function (FN) button to allow special access to the Function keys.

Although the F1 function key is reserved in Igor Pro, it can be assigned if used with a modifier key.

Delete Key	Remove the Key entry for the selected Action.
Edit	Open the Shortcut Editor dialog (or double-click in a “Shortcut Action” field) to change the Action’s parameters.
Execute	Run the selected Action.
Delete	Remove the selected Action from the ‘Shortcut Action’ list.
Save	Saves any changes to the current Shortcut Pool file.

Predefined Actions

Action 1	[Hold+10mV|10pA:Right]	
Action 2	[Hold-10mV|10pA:Left]	
Action 3	[Hold+1mV}1pA:Right,shift]	
Action 4	[Hold-1mV|1pA:Left,shift]	
Action 5	[View last]	F2
Action 6	[Stop Acquisition]	F3
Action 7	[Start Routine]	F4
Action 8	[Stop Routine]	F5
Action 9	[Pause Paradigm]	F6
Action 10	[Resume Paradigm]	F7
Action 11	[Cursor Info]	F10

4.3.9 Startup

The SutterPatch application startup sequence:

1. Click on the Igor Pro 8 icon to launch SutterPatch.
2. The application opens an Igor 'Command' window along with a "splash" screen displaying file opening information; both windows are then closed.
3. Next, a 'Welcome to SutterPatch' screen displays a few launch options:
   - Igor Only Run Igor Pro (without launching SutterPatch).
   - Open Launch SutterPatch from a saved Experiment.
   - Start Launch SutterPatch for a new Experiment.
4. Click 'Start', an Igor "Command" window opens, and the Welcome screen displays a progress bar while compiling the SutterPatch code; the Welcome screen is then closed.
5. A 'Save experiment as" dialog displays with standard file saving controls.
6. Next, if no Sutter amplifier is detected, the 'No USB Connection' pane allows you to try re-establishing the USB connections, or to select a hardware-emulation demonstration mode (IPA, Double IPA, dPatch, Dendrite).

   In demonstration mode, or "demo" mode, the Amplifier Control Panel and acquisition Scope window are labeled with "Demo", the input and output channels use simulated data, and most SutterPatch functions are available.
7. The SutterPatch files are initialized, the Dashboard panel displays, and additional SutterPatch windows display if they were open in the prior experiment.

5. PROGRAMMING

5.1 Data Format

SutterPatch data are written in a 64-bit double-precision binary floating-point format. This supports a decimal precision of 17 significant digits.

The data are stored within an Igor Experiment (*.pxp) file.

For large data sets, an optional HDF5 file format will be available for streaming data acquisition without resaving the experiment at the end of a recording.

5.2 Data Structure

SutterPatch recorded data are stored as multidimensional data waves, and are listed per signal in the Data Browser. Select a data wave in the Data Browser and right-click to 'Edit' the Signal data in a spreadsheet-style table. The two-dimensional data wave is displayed with one row per sample point and one column per trace, with the number of data table columns increasing with the number of sweeps.

Warning! The raw data can be directly edited in the Data Browser – this is not recommended, as it permanently alters the data.

Note: While SutterPatch does not read the older Igor one-dimensional wave data-format, graph data for each axis can be separately exported to it. See the Analysis Editor / Files menu.

5.3 Data Paths

The Data Browser path references an internal Igor "root" folder, and not the computer's file system. The Data Browser right-click 'Copy Full Path' command copies a Signal's data wave path to the system clipboard.

For advanced users, the object's path name can be used in user functions and executable commands. However, when referencing an active Scope window, the path name to the data wave can be substituted by "t[#]", where '#' refers to the signal position number in the Scope window.

5.4 User Functions

SutterPatch functionality can be extended through the use of user-defined Functions.

To create a user Function:

1. Open the menu for Windows / Procedure Windows / Procedure Window.
2. Enter your user code into the Procedure window, following its '#pragma' and '#include' lines.

Example:

```
#pragma TextEncoding = "Windows-1252"
#pragma rtGlobals=3    // Use modern global access method…
#include "SP_Globals", optional
```

Function SayHello()

DoAlert 0, "Hello World!"

End

Note: The Function name must include trailing open/close parentheses "()"

3. Click on the Compile button at the bottom of the window.
4. Enter the Function name (including parentheses) into the Command window and press 'Enter', or use it in a Paradigm 'Execute' step.

For more information on creating your own functions, see the Igor Help topics on Programming / User-Defined Functions, and Procedure Windows.

Warning! User-defined functions only exist during the Experiment. They are not stored when the Experiment is closed. If you plan to re-use them in other Experiments, save them to a separate file, such as with a word processor.

Also, while user-defined functions are stored internally by Igor, there is no visible list, so you will need to maintain such a list manually.

6. TROUBLESHOOTING

6.1 Technical Support

Technical support is provided to customers at no charge.

Support hours:	8:00 AM - 5:00 PM PST (Pacific Standard Time).
Telephone:	(+1) 415.883.0128
Fax:	(+1) 415.883.0572
E-mail:	info@sutter.com
Address:	Sutter Instrument Company One Digital Drive Novato, CA 94949

When contacting us for technical support, please provide your SutterPatch version and "build" numbers to help us troubleshoot your situation. These numbers are found in the Start splash screen during program loading, or in the Log Window Startup events.

For issues regarding Igor Pro features (all non-SutterPatch menu items), please contact Wavemetrics, Inc. for technical support.

6.2 Manual

The IPA manual is installed as a PDF file along with the SutterPatch software. The latest version of the manual can be downloaded from our web site:
https:www.sutter.com/AMPLIFIERS/SutterPatch.html.

You can navigate through the PDF document using Table of Contents links, accessed via the Bookmarks tab on the left side of the PDF screen.

6.3 Online Help

Online help is available via the main Help menu, under 'Igor Help Browser' or 'Help Topics'. The SutterPatch Help Topics / Help File names start with "SP_". The online Help includes the same information as found in the PDF manual.

Most items in SutterPatch also include a short description as a tool tip. Hover the mouse over an item to see the tool tip.

6.3.1 Error Messages and Notifications

Some SutterPatch error messages or notifications will flash to get your attention, and automatically close after several seconds, and then write to the "History" window. To review such messages, see the Command window (Windows / Command Window).

6.4 Startup Issues

6.4.1 Installation Fails

Problem: The SutterPatch installation on Windows fails due to language pack incompatibilities.

Solution: Support for foreign language packs has been added. If language versions still cause problems, please contact Sutter Technical Support.

6.4.4 Startup Compiler Errors

Problem: The SutterPatch loading on Windows fails due to compiler errors.

Solution: Instead of using the SutterPatch updater, run the full SutterPatch installer.

6.4.4 Application Not Loading

Problem: The SutterPatch application does not load – the startup sequence only loads Igor Pro.

Solution: If available, execute the Igor Pro menu command 'Macros / Autocompile'.

6.4.5 Startup EEPROM Errors

Problem: Starting up SutterPatch and simultaneously powering on the amplifier generates an EEPROM error. Attached hardware such as headstages might use incorrect settings.

Solution: Close SutterPatch, power cycle the amplifier, and relaunch SutterPatch.

6.4.6 Startup Odd Errors

Problem: When starting up or running SutterPatch, odd program errors display.

Solution: If after a SutterPatch update, closing and re-opening SutterPatch sometimes clears up these errors. If after an OS update, roll back the OS software update.

6.4.6 USB Communication Fails

Problem: When starting up SutterPatch in Windows, there is no USB communication with the computer.

Solution: Power off the amplifier, then reseat the USB cable on both ends, and then power on the amplifier.

Or, the Windows "power plan" might have disabled the USB ports.

1) Go to the Windows Start screen, and enter "edit power plan" in the Windows Search box.

2) Click on "Change advanced power settings".

3) Scroll down to "USB settings" and click on its [+] box.

4) Click on the "USB selective suspend setting" [+] box.

5) Change the "On battery" and "Plugged in" settings from 'enabled' to 'disabled', and click "OK".

Or, a Windows 10 update can sometimes disable the computer's USB ports. Either browse the web to find a Device Manager driver solution to the problem, or restore the OS to an earlier version of Windows 10, until a Windows 10 update fix is released.

6.4.3 Sample Parameter Files Not Installed

Problem: The SutterPatch installer fails to install the sample parameter files, as access is blocked to the Program Files or Users\..\Documents folders.

Solution: Disable any virus scanners or firewalls. If that does not help, then manually copy the sample parameter files into the Users\..\Documents\SutterPatch \Parameters folder.

6.5 Acquisition Issues

6.5.1 Acquisition In Progress

Problem: Unable to start an acquisition because the system thinks it is already running.

Solution: Use the menu command SutterPatch / Reset Acquisition to clear the acquisition status.

6.5.2 Acquisition Windows Lock Up

Problem: The Scope window, Routine Editor or Paradigm Editor lock up during acquisition.

Solution: Use the menu command SutterPatch / Reset Acquisition to halt acquisition.

A combination of SutterPatch-related and computer-related issues can contribute to your system's performance. For suggestions to improve it, see the Troubleshooting item Sluggish Acquisition below.

6.5.3 Acquisition Terminates

Problem: During acquisition, the recording terminates unexpectedly.

Solution: Close the Analysis / Data Browser window, if it is open.

This window can consume a large amount of system resources, which can interfere with data acquisition.

6.5.4 Signal Flat

Problem: A Scope input signal is completely flat during acquisition, i.e., zero amplitude.

Solution: The corresponding headstage might not be attached to its port. The headstage HDMI connectors do not lock on - make sure they do not disconnect from their port.

Power off the dPatch system and reconnect the headstage.

6.5.5 Signal Saturated

Problem: A Scope input signal is completely saturated during acquisition.

Solution: The corresponding headstage is not attached to its port. Power off the IPA system and reconnect the headstage cable.

If the headstage is attached, the Gain setting might be set too high. Reduce the output gain on the Amplifier Control Panel.

Note: The headstage HDMI connectors do not lock on - make sure they do not disconnect from their port.

6.5.6 Noise I

Problem: The noise levels of the instrument suddenly and erratically increase.

Solution: If the headstages are touched, the noise level will greatly increase. Make sure you are grounded or working in a Faraday cage.

6.5.7 Noise II

Problem: The (5 kHz) noise levels of the instrument suddenly and permanently increase from ~0.8 pA to 2 pA.

Solution: If early model headstages are "hot swapped", i.e. plugged in or unplugged while the instrument power is on, their headstage operational amplifiers might be damaged. Contact Sutter Technical Support for service.

Best practices is to turn off the instrument power before (un)plugging its headstages.

6.5.8 Paradigm Sound Reduced

Problem: The paradigm 'Sound' step volume is attenuated at lower frequencies.

Solution: Upgrade the computer speaker, such as with add-on speakers.

6.5.9 Offset Zero Delay

Problem: The Offset button in the IPA Control Panel has a short delay before it responds.

Solution: This can occur after running the Membrane Test due to internal processing.

6.5.10 Post-Acquisition Delay

Problem: Every time acquisition completes, there is a delay with the program operations, as the entire *.pxp Experiment file is resaved when a recording stops.

Solution: Create new Experiments more often, so that file sizes are smaller and more manageable.

Or, change the file saving settings in Set Preferences / Files and Naming.

6.5.11 Sluggish Acquisition

Problem: Data acquisition is sluggish.

Solution: The computer's available resources need to be increased to handle the system load.

A combination of SutterPatch-related and computer-related issues can contribute to your system's performance. Here are some suggestions to improve it:

Close:	Data Browser window – in Igor Pro 8, it consumes a lot of CPU time.
Disable:	Computer screen saver, Power Save or Sleep modes.
Disable:	Scope window persistence display.
Disable:	Virtual input channels.
Reduce:	Routine / Acquisition & Routine Parameters / Output sampling rate.
Close:	Background software.
Remove:	Software for certain license protection USB keys (dongles).
Optimize:	Hard disk (defragment).
Upgrade:	Computer graphics card.
Increase:	Computer RAM, cache size or CPU speed.

6.5.11 Routine Loading Delays

Problem: Selecting a Routine in the Routine Editor temporarily hangs the SutterPatch program.

Solution: Hide the Routine Editor Preview pane.

If there is a very large number of data or sweeps to display, the Preview pane can take a long time to redraw, and the program becomes temporarily unresponsive.

6.5.12 USB Errors

Problem: A USB communications error occurs.

Solution: In the amplifier Control Panel, click the Reset USB button. If the button does not turn from red to green, then try to isolate the problem.

a) Unplug and re-plug both ends of the USB cable from the amplifier to the computer.

b) Try another USB cable.

c) Try another USB port

d) Remove any USB hubs.

6.5.13 System Freezes

Problem: The system hangs up after changing the filter setting or VC/CC mode selection.

Solution: Reset the USB port via the amplifier Control Panel USB Reset button or the SutterPatch / Hardware Control menu.

6.5.14 IPA Amplifier Control Panel Issues

Problem: The IPA/DIPA Amplifier Control Panel is having odd problems.

Solution: Reset all hardware controls by mouse-clicking almost anywhere in the Control Panel or via the menu SutterPatch / Hardware Control./ Reset Controls.

:

- Reset Amplifier Controls — Reset all hardware settings of all attached Sutter IPA/DIPA amplifiers and their headstages to factory defaults.

6.6 Analysis Issues

6.6.1 Analysis Not Deleted

Problem: An analysis cannot be deleted in the Analysis Editor.

Solution: The analysis is still in use, i.e. displayed in another window, such as a graph window - close the window to allow the analysis to be deleted.

6.6.2 Signal Axes Overlay

Problem: The X-axes and units are overlaid in the Scope window.

Solution: There is not enough room for the X-axes and units due to the number of signals displayed. Switch to a tiled signal layout, or reduce the number of visible signals by right-clicking a signal and selecting 'Hide Signal'.

6.6.3 Graphs & Layouts Not Visible

Problem: Cannot see SutterPatch Experiment graphs or layouts on non-SutterPatch computers.

Solution: Use the Igor menu command Windows / Graph, or Windows / Layout or Layout Macros, to see the object. Right-click it to modify with Igor options.

6.7 General Issues

6.7.1 Buttons Unresponsive

Problem: When using a slower computer in emulation (demo) acquisition mode, acquisition-related actions might be difficult, such as clicking the Stop button.

Solution: You may need to click the button more than once or hold it down longer than usual.

A combination of SutterPatch-related and computer-related issues can contribute to your system's performance. For suggestions to improve it, see the Troubleshooting item Sluggish Acquisition above.

6.7.2 Slow Display of Sweeps

Problem: When displaying a large number of sweeps, the display slows down.

Solution: Disable Persistence display in the Scope window.

6.7.3 Window Maximizing

Problem: Maximizing a window only maximizes the title bar.

Solution: Certain fixed-size windows and panels will not maximize (Action Potential Analysis, Amplifier Control Panel, Dashboard, Synaptic Event Analysis, Log, Paradigm Editor, Set Metadata, Set Preferences.) This is a reported Igor issue.

Also, if the active window is maximized, creating a new window might automatically "maximize" the new window.

6.7.8 Slow Window Opening/Closing

Problem: Window opening and closing is slow on the macOS.

Solution: Close the SutterPatch Data Browser. This function consumes a lot of system resources.

6.7.5 Command Window Frozen

Problem: The Command Window is blank and/or unresponsive.

Solution: Use Ctrl-J, or click on the Amplifier Control Panel, and the Command window is redrawn as an active window. This is a reported Igor issue.

6.7.6 File Operations Crash

Problem: In Windows 10, file opening or saving crashes SutterPatch.

Solution: Remove the Dell Backup and Recovery utility v1.8, or upgrade it to a newer version.

6.7.9 Wrong Preference Settings

Problem: Program preferences are non-standard or corrupted.

Solution: Reset the SutterPatch preferences to their defaults via the SutterPatch / Set Preferences / General / Preferences Defaults button.

6.7.10 Font Size Too Large

Problem: The font size is too large when using the display scaling in 'Scale and Layout'.

Solution: This can occur on high-resolution monitors running on older versions of Windows 10. Upgrade to the current version of Windows 10.

6.7.11 Weird Behavior

Problem: There is weird or buggy behavior with the SutterPatch program.

Solution: Reset the SutterPatch program settings to their factory defaults via the SutterPatch / Set Preferences menu command, by performing a Factory Reset.

6.7.12 Igor Pro Features

Problem: There are a large number of standard features in Igor Pro that can be used in conjunction with the SutterPatch application.

Solution: Refer to the Igor Help browser, or to Wavemetrics, Inc., regarding issues with Igor Pro features.

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APPENDIX C: ACCESSORIES

• Ground Point GP-17	Grounding point hardware
• Patch Panel	Rack-mountable analog and digital I/O BNC panel
• Quartz Pipette Holder	Quartz pipette holder
• Rack Pack	Rack mounting hardware

APPENDIX D: FUSE REPLACEMENT

In the event that the instrument fails to power up when it is switched on, the power-line fuses should be checked to determine whether they have blown. Two fuses are located in the fuse holder in the power cord module on the rear of the amplifier.

To replace a fuse:

1. Unplug the power cord from the power entry module, revealing the fuse holder below.
2. Remove the fuse holder.
3. If a fuse is blown, it is recommended to replace both fuses.
4. Insert appropriately-rated replacement fuses (see below).
5. Replace the fuse holder in the power entry module and reconnect the power cord.

Mains Power Source	Fuses (Type: Time Delay/Time Lag, 5mm x 20mm, glass tube)	
	Fuse Rating	Manufacturer Examples
100 – 240 VAC	T2.0A, 250V	Bussmann: GMC-2-R, S506-2A, Littelfuse: 239.002.P

Table D-1. IPA Fuses

APPENDIX E. TECHNICAL SPECIFICATIONS

General Specifications

IPA & Double IPA Amplifiers	
Dimensions (in.) (includes handles & connectors)	IPA: 18.8 (W) x 11.8 (D) x 1.8 (H) DIPA: 18.8 (W) x 11.8 (D) x 3.5 (H)
Dimensions (cm) (includes handles & connectors)	IPA: 48.25 (W) x 30.0 (D) x 4.5 (H) DIPA: 48.25 (W) x 30.0 (D) x 9.0 (H)
Weight (lb) (with headstages)	IPA: 9.05 DIPA: 8.1
Weight (kg) (with headstages)	IPA: 4.1 DIPA: 3.7
Case	IPA: steel DIPA: aluminum
Communications	USB 2.0 (High Speed)
BNC Channels	2 auxiliary analog outputs (impedance: 16 Ω) 4 auxiliary analog inputs (impedance: 1 MΩ) 1 digital input trigger 1 digital output trigger 8 auxiliary digital outputs
Rack use	IPA: 19” rack-mount (1U) DIPA: 19” rack-mount (2U)
Benchtop use	Rubber feet
Signal Ground	4 mm Banana socket
Earth Ground	4 mm Banana socket
Safety	CE marking (Conformité Européenne)
Auxiliary I/O	DB-15 female connector

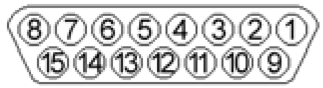
Port		
Auxiliary I/O Pinout	Pin	Definition
	1	Digital Output 1
	2	Digital Output 2
	3	Digital Output 3
	4	Digital Output 4
	5	Digital Output 5
	6	Digital Output 6
	7	Digital Output 7
	8	Digital Output 8
	9	Auxiliary Analog Input 1
	10	Auxiliary Analog Input 2
	11	Auxiliary Analog Input 3
	12	Auxiliary Analog Input 4
	13	Auxiliary Analog Output 1
	14	Auxiliary Analog Output 2
	15	Signal Ground
Configurations	Voltage-clamp Current-clamp	
Whole Cell Capacitance Compensation	0 – 100 pF	
Current-Clamp Rise Time (with 20 kHz low-pass filter & 100 MΩ load)	17.5 µs	
Output Gain	0 – 25x	

Table E-1. IPA & DIPA Amplifier Specifications

IPA Headstage - Physical	
Construction	Anodized aluminum case
Dimensions (in.)	4.000 (L) x 1.375 (W) x 0.825 (H)
Dimensions (cm)	10.160 (L) x 3.493 (W) x 2.096 (H)
Cable Length (feet)	6
Cable Length (m)	1.83

Weight (lb) w/o cable w/cable	0.21 0.294
Weight (kg) w/o cable w/cable	0.095 0.133
Ground Socket (mm)	1
Feedback Resistor	500 MΩ

Table E-2. IPA Headstage - Physical

IPA Headstage - Noise (measured with 8-pole Bessel filter)	
Bandwidth	**Open-Circuit Noise (RMS)**
0.1 – 1 kHz	< 0.25 pA
0.1 – 5 kHz	< 0.75 pA
0.1 – 10 kHz	< 1.40 pA

Table E-3. IPA Headstage Noise

IPA Data Acquisition	
Analog I/O Channel Type	Full Differential
Analog I/O Channel Amplitude (voltage)	±10 V
Analog I/O Channel Amplitude (current)	±20 nA
Analog I/O Channel Resolution	16-bit
Headstage Input Sample Rates	0.1 - 50 kHz
Headstage Input Filter Bandwidth	0.5 - 20 kHz
Headstage Output Sample Rates	0.1 -10 kHz
Auxiliary In Sample Rates	0.1 - 200 kHz

Auxiliary Out Sample Rates	0.1 - 100 kHz
Digital In States	0 - 0.8 V = Low 2.0 – 5.5 V = High
Digital Out States	0 – 0.4 V = Low 2.4 – 3.3 V = High
Digital In Trigger Width	Edge triggered (ns)
Digital Out Trigger Width	100 µs
Digital In Sample Rates	0.1 – 50 kHz
Digital Out Sample Rates	0.1 – 10 kHz
Digital Out Current (max)	20 mA

Table E-4. IPA Data Acquisition

IPA Electrical	
Power consumption	18 Watts maximum
Mains fuse	250V 1A Slow Blow (5 mm x 20 mm) T2.0
Cables	Shielded grounded power line cord
Line Voltage	100 VAC – 240 VAC

Table E-5. IPA Electrical

IPA System Components

Carefully remove all components from the shipping container. The following should be included:

• (1) IPA Amplifier	IPA/E-1	
• (1) IPA Headstage	IPA-HS	
• (1) Polycarbonate Pipette Holder	EH-P170	(standard)
or (1) Quartz Pipette Holder	EH-Q170	(optional)
• (1) Model Cell	MCELL	
• (1) Auxiliary I/O Adapter Cable		
• (1) Rack-Mount Kit	RACK-PK	
• (1) Power Cord		

- (1) USB 2.0 Cable
- (1) Quick Start Guide (with Igor Pro 8 Serial #)
- (1) USB Flash Drive (with SutterPatch and Igor Pro 8 software)

Double IPA System Components

Carefully remove all components from the shipping container. The following should be included:

• (1) Double IPA Amplifier	IPA/E-2	
• (2) IPA Headstages	IPA-HS	
• (1) Auxiliary I/O Adapter Cable		
• (2) Polycarbonate Pipette Holders	EH-P170	(standard)
or Quartz Pipette Holder(s)	EH-Q170	(optional)
• (1) Model Cell	MCELL	
• (1) Power Cord		
• (1) USB 2 Cable		
• (1) Rack Mount Kit	RACK-PK	
• (1) Quick Start Guide	(with Igor Pro 8 Serial #)	
• (1) USB Flash Drive	(with SutterPatch and Igor Pro 8 software)	

Pipette Holder Parts

- End Cap
- Silicone Gaskets (O-rings, 6 ea.)

Gasket ID	Color
1.1 mm	Clear
1.2 mm	Green
1.5 mm	Orange-Red
1.75 mm	Blue

- Silver Wire
- Body/Barrel (standard: polycarbonate; optional: quartz)
- Wire Seal (tubing)
- Gold Pin
- Pin Cap
- Lockdown Ring

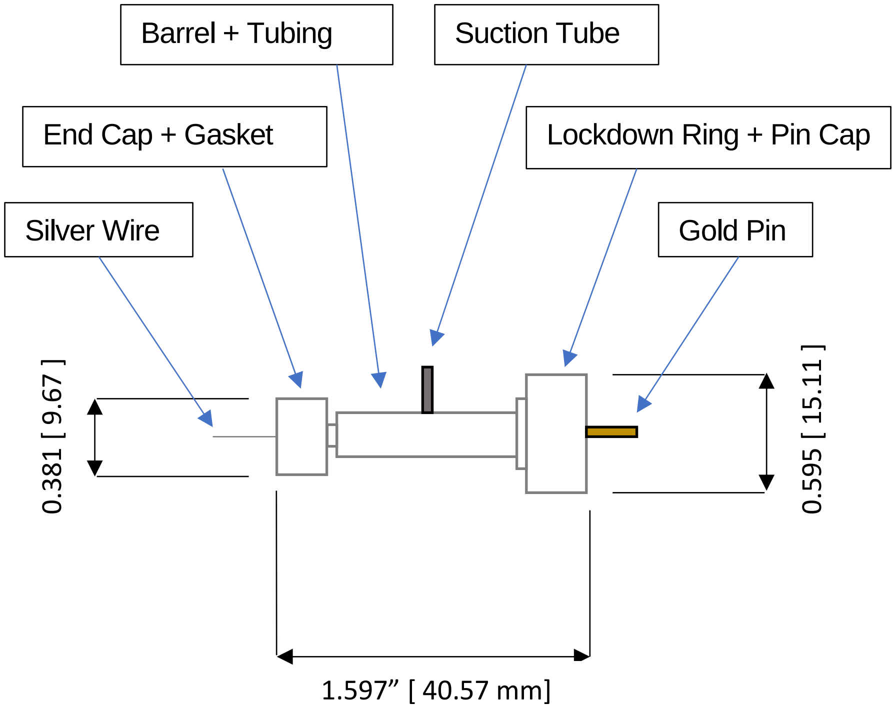


Figure E-0-1. Electrode Holder

Figure dimensions are in "inches [mm]".

Model Cell Parts

- Model Cell
- Connector pins with crimp
- Ground wire

IPA Accessories

Patch Panel IPA-PCH

This optional rack-mountable panel provides BNC ports for easy access to digital and auxiliary I/O connections.

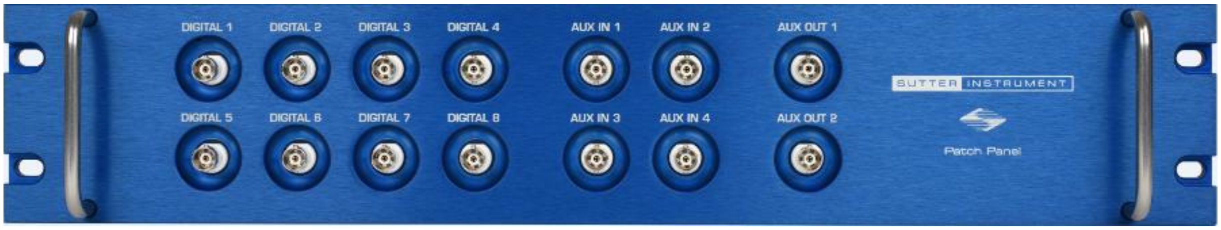


Figure 0-2. Patch Panel

IPA Patch Panel	
Dimensions (in)	18.8 x 2 x 3.5
Dimensions (cm)	48 x 5 x 9
Weight (lbs)	3.5
Weight (kg)	1.6
Digital Output BNCs	8
Auxiliary Analog Input BNCs	4
Auxiliary Analog Output BNCs	2

Table E-6. IPA Patch Panel

Ground Point GP-17

For system grounding, this optional machined brass tower provides reliable low-resistance connections for electrophysiology setups. The base plate mounts directly to air table tops (imperial and metric) with the included ¼-20 and M6 screws. The plated connectors accept up to 9 banana plugs and 8 bare wires (up to 10 gauge). A "star" ground configuration is used to avoid ground loops.

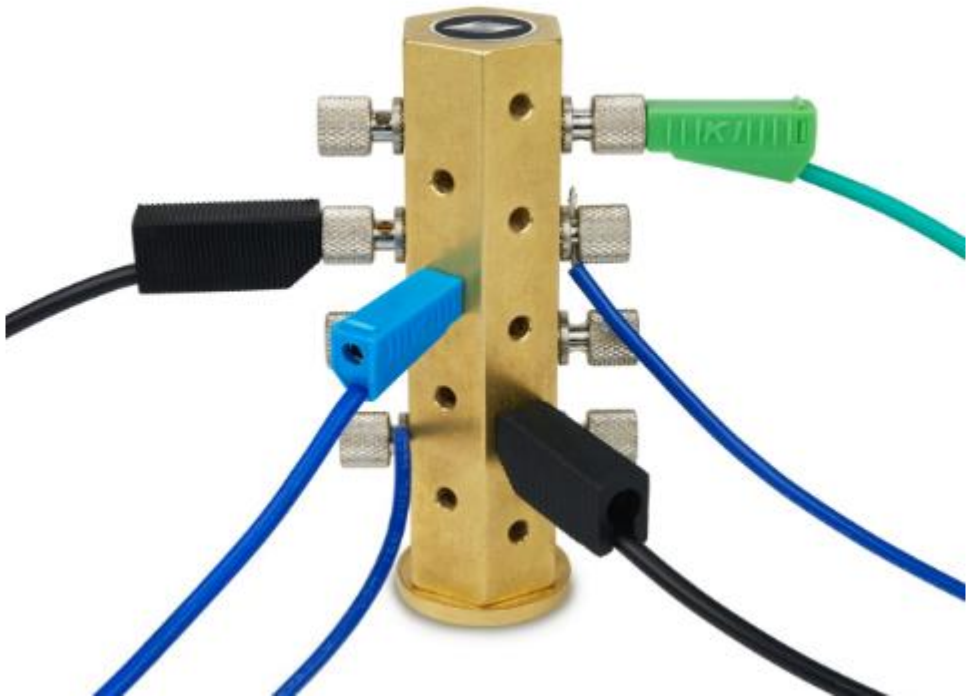


Figure 0-3. Ground Point

The Ground Point 10-item kit includes cables with banana plugs and alligator clips. For very complex rigs, two sets of cables might be needed.

APPENDIX F. SUTTERPATCH ALGORITHMS

Action Potential Threshold Algorithm

[for Action Potential Analysis]

Math used in the Results pane 'Threshold potential' computation:

The Event starts when the signal slope is > 1 mV/100 µs (10 V/s), or when 25% of the maximum slope is reached, whichever is smaller.

The exact 'Threshold potential' timepoint is based upon differentials using a central differences algorithm.

LockIn Computation

[for Routine Editor Virtual Input Channels]

Math used in the LockIn computation:

Factor = (2.0 / SinePointsPerCycle) / sine_amplitude^2

A = Factor * ∑(current * stim_real)
∑ over one SinePointsPerCycle

B = Factor * ∑(current * stim_imag)
∑ over one SinePointsPerCycle

DC = 1/SinePointsPerCycle * ∑(current)
∑ over one SinePointsPerCycle

	VC-mode	CC-mode
Phase =	atan(B/A)	atan(B/A)
RealY =	A	A / (A^2 + B^2)
ImagY =	B	B / (A^2 + B^2)

Omega = (2 * pi) / SineCycleDuration

Gt = Idc / (Vdc – Et)

$$Gs = (A^2 + B^2 - A * Gt) / (A - Gt)$$

$$Gm = Gt * Gs / (Gs - Gt)$$

$$Cm = (A^2 + B^2 - A * Gt)^2 / ((A - Gt)^2 + B^2) / (Omega * B)$$

Single Channel Fitting

[for Single Channel Analysis]

Math used in single channel fitting:

<u>Gaussian Fit</u>

$$y = y0 + A * \exp(-((x – x0) / width)^2)$$

y0 = offset

A = height of curve's peak

x0 = position of center of peak

width = $\sqrt{2} * \sigma$

σ = standard deviation of the peak

<u>Linear Exponential Fit</u>

$$y = y0 + A * \exp(-(x – x0) / tau$$

<u>Logarithmic Exponential Fit</u>

$$y = k0 + k1 * \exp(-(\ln (x / k2) / k3)^2)$$

Standard Error of the Mean (SEM) Algorithm

[for Analysis Editor Error Bars]

Math used in the 'Standard Error of the Mean' computation:

$$SEM = \sqrt{} (SumSq – Mean^2 * N) / (N-1)$$

SumSq = sum of all squared samples

Mean = sum of all samples / N

Note: The SEM algorithm is similar to the Standard Deviation "√ (variance)", but using 'Mean' vs. 'sum of all samples'.

Synaptic Event Detection Reference

[for Synaptic Event Analysis]

Deconvolution paper:

Pernía-Andrade AJ, Goswami SP, Stickler Y, Fröbe U, Schlögl A, Jonas P. A Deconvolution-Based Method with High Sensitivity and Temporal Resolution for Detection of Spontaneous Synaptic Currents In Vitro and In Vivo. Biophys J. 2012 Oct;103(7):1429–39.