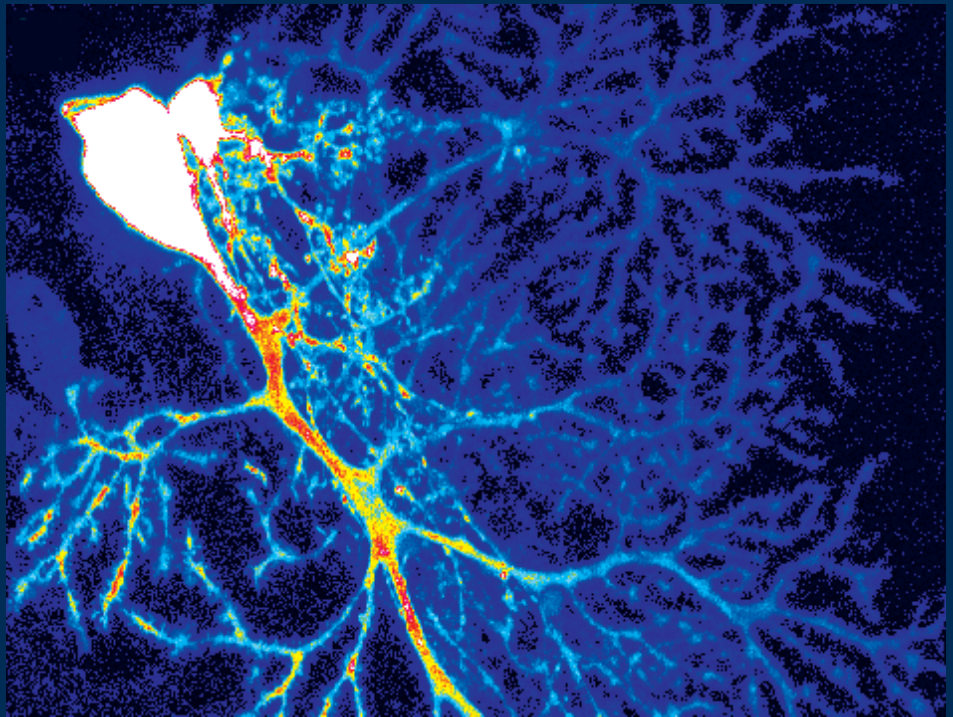




## PULSE / PULSEFIT

Software for electrophysiological research, data acquisition, review and online analysis



### Features

### PULSE

- Digital Oscilloscope
- Versatile Pulse Generator
- Parameter Monitoring
- Data Tree Editor
- Online Analysis

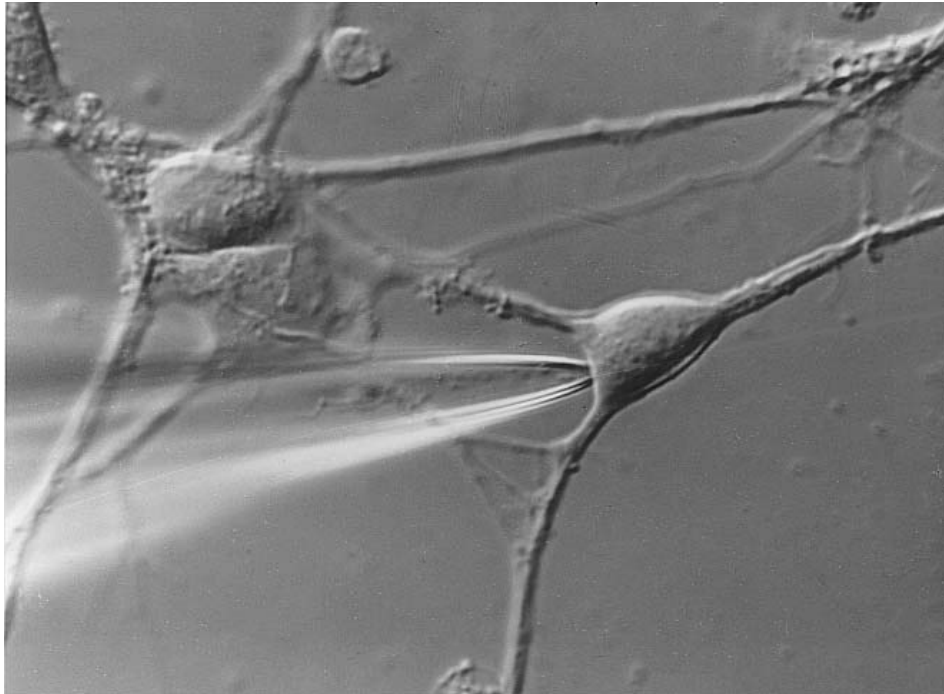
### PULSEFIT

- Data Review
- Cursor-operated measuring Features
- Fit of Data Traces
- Fit of Parameters

# HEKA

HEKA provides the finest instruments today to achieve the needed progress of tomorrow...

***PULSE is easily adaptable to any individual approach for performing electrophysiological experiments.***



**PULSE/PULSEFIT - a program which first thinks of data analysis**

The consideration of all measuring parameters has made PULSE the most powerful program for electrophysiological clamp experiments. It has been developed with the help of leading laboratories worldwide to meet the requirements of the scientific community. PULSE and PULSEFIT thus provides the optimal solution for data acquisition and evaluation. Further programs such as TAC and PULSETOOLS can immediately process the data obtained with PULSE. Export functions transfer the data to other programs such as Igor, or as ASCII (MAC and DOS). The "extensions" of PULSE (LOCK-IN, FURA, and X-CHART) provide the link from clamp to further experiments.

**PULSE/PULSEFIT - a reliable and convenient program**

PULSE and PULSEFIT constitute the perfect combination of simple operation and comprehensive functionality. For the beginner, the operation can be limited to the essential functions. Intuitive operation and uncompromising simplification of complicated adjustments quickly enable the user to perform more complex experiments and evaluations. The experienced patch-clamp investigator will be fascinated by the versatility of the program and logical structure of the functions. The outstanding user interface of the program with "virtual control keys" can be defined by the user to meet his requirements. From the very first day you can record data from your experiments with PULSE. PULSE and PULSEFIT provide the optimum in user convenience.

**PULSE/PULSEFIT - a program for your amplifier, too**

PULSE is custom-made for application with the EPC 9 and EPC 10 patch clamp amplifiers, but any other amplifier also receives a tremendous boost with PULSE, which supports all patch-clamp and voltage-clamp amplifiers. Therefore, you can operate various set-ups with different amplifiers and purposes in a laboratory with the same software. All data are available in the same documented format, and you can thus prepare filters for your own programs. Do you have questions? If so, a hotline is at your disposal!

PULSE provides sophisticated tools for electrophysiological experiments

**Tree**

Export your data (e.g. single sweep, online analysis results) to printer, IGOR files (presenting and analysing program) or ASCII (MAC or DOS).

**Buffer**

Add, subtract or accumulate your measured sweeps.

**Marks**

Tag several entries in the data file to perform one action to all of them (e.g. print, average).

**Display**

Change the display of the oscilloscope (e.g. grid, dimmed overlay).

**Replay**

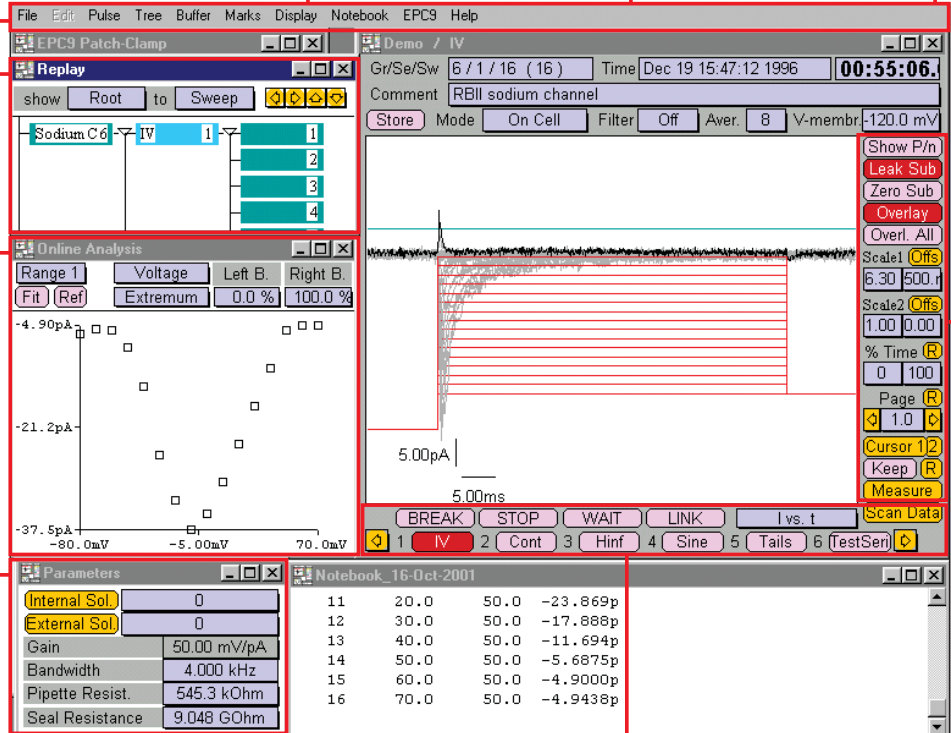
Acquired data and old data files can be reviewed and edited.

**Online Analysis**

Immediate analysis of the just acquired or replayed data. The analysis result is shown as a function of a variety of parameters, specified by the user.

**Parameters**

Parameters allow the additional data acquisition of different values as RMS noise, pipette resistance, seal resistance or user defined parameters (e.g. pH).



**Sequence Pool**

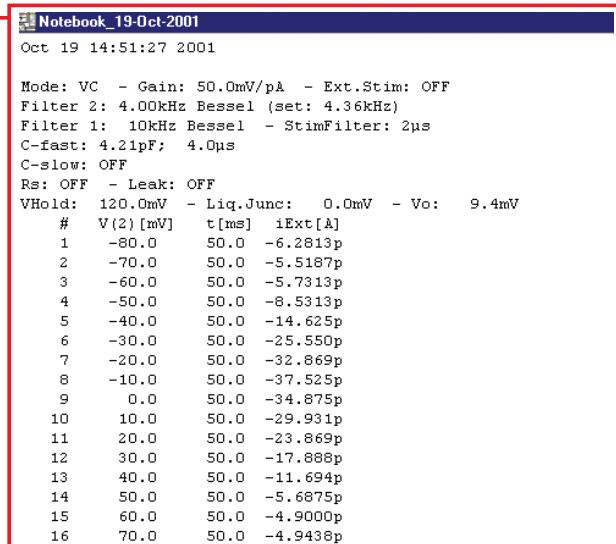
Pool of available pulse templates. Up to six template controls are displayed at a time, which can be stopped or interrupted by the tools.

**Functions**

Function features allow the displaying of measured responses like leak current...

**Notebook**

During stimulation and replay the results of the online analysis are displayed in the notebook. These data columns can be exported to disk or copied to the clipboard.



EPC 9 - the virtual front panel

The Pulse Generator window provides the layout of a stimulation pulse sequence

**Test Pulse**

There are two test pulse modes: built-in test pulses (double or single) and use of a stimulation template from the pulse generator as test pulse.

**Controls**

All values can be set automatically or manually by either the mouse or the keyboard.

**Filters**

Two built-in high quality hardware filters (Butterworth/Bessel) perform excellent signal conditioning and remove the expense of purchasing additional filter instruments.

**Stimulus**

The stimulus can be filtered by activating the stimulus switch.

**Zap Pulse**

A high voltage pulse is applied to the pipette in order to rupture the patch membrane. The parameters (duration and amplitude) can be specified.

**Sequence Pool**

It is a paging bar with a nearly unlimited number of sequences. Loads, saves, copies etc. the pool of available stimulation sequences.

**Timing**

Determines the number of the sweeps, the sweep- and the sample interval.

**Segments**

A pulse pattern consists of an arbitrary number of segments.

**AD/DA Channels**

Switch between different trigger modes and choose the right channels for acquisition.

**Sequence Cartoon**

After each editing operation the pulse pattern is displayed in the cartoon.

**Documentation**

All EPC 9 settings are stored with the raw data for easy reconstruction, analysis, and documentation of your experimental procedures.

**Automatic Compensations**

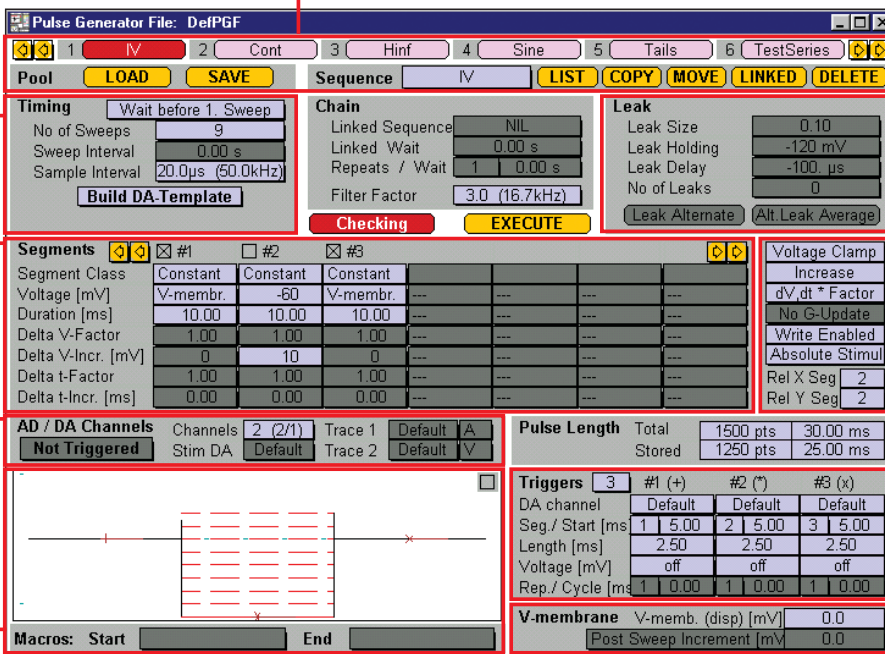
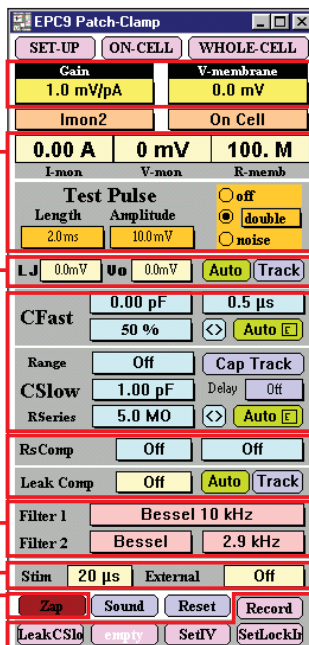
Automatic routines for leak and capacitive transient compensations, perform these tasks faster and more accurate than even the most experienced experimenter. Capacitance tracking allows continuous updating of membrane capacitance and series resistance compensation during recording sessions.

**Leak and Rs Compensation**

This controls a hardware leak compensation. The series resistance compensation corrects for membrane voltage errors under conditions of high access resistance between pipette and cell interior.

**Macros**

Macro features allow the recording of routine functions and then accessing these Macros by a simple mouse click.



**Leak**

Determines different parameters of the leak pulses.

**Increment Mode**

The variable determines the order of increment of the segment voltage/duration.

**Triggers**

There are up to 3 triggers chooseable.

**V-Membrane**

Displays the presently selected holding potential.



PULSEFIT - all common fit routines in the field of electrophysiology are available

**Sweep Fit Control**

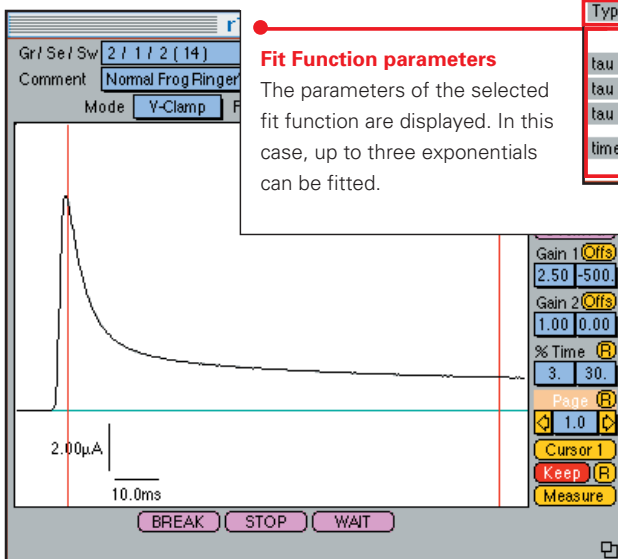
Buttons for the control of fits to raw data traces. "Auto" repeats the same kind of Sweep Fit for all Sweeps of a series automatically.

**Simplex Fit**

Control parameters for the Simplex fit algorithm are edited here. Manual fit can be performed for the determination of good starting values.

**Fit Function parameters**

The parameters of the selected fit function are displayed. In this case, up to three exponentials can be fitted.



**Fit Type**

Select a fit function (polynomial, Gaussian, function according to a Hodgkin & Huxley formalism for channel activation and in-activation) or store results of the Online Analysis or those obtained from manual mouse-operated measurements to the Analysis file.

**Sweep Fit**

The trace fit is superimposed to the raw data within a window selected by the vertical cursor lines. In this case, a double-exponential fit describes the current decay.

**Sweep Fit Results Table:**

The results determined by Sweep Fit are stored in the Analysis File and are available in Series Fit for further analysis. These parameters can be displayed as a function of other parameters or various abscissa variables like potential or duration of the relevant segment.

**Fit Types**

Fit functions supported by Series Fit are: Polynomials, Exponentials, Boltzmann functions, and current-voltage relationships. In this example recovery from inactivation is analyzed. The peak current in a test segment is plotted versus the exponentially increasing duration of an interpulse segment. The theoretical curve describes a double exponential recovery.

**Series Fit Control**

Control buttons for Series Fit. Selected data and fits can be exported in various formats. Individual data points can be skipped so they are not considered for the fit.

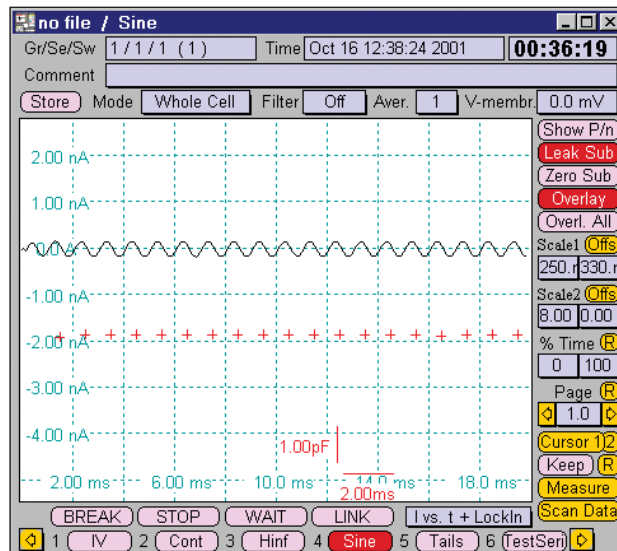
Capacitance measurements with PULSE

The PULSE software and our EPC 9 or EPC 10 patch clamp amplifier combine to provide you with everything you need if you are interested in membrane capacitance measurements. The EPC 9 or EPC 10 is the ideal patch clamp amplifier for capacitance measurements because all the relevant parameters are under its control. Digital control of filter settings, gain and compensation networks allows the admittance of the signal source at the amplifier's input to be determined based on the amplifier's own calibration. No external filters are required for standard measurements.

In 1982 Neher and Marty introduced the lock-in amplifier into the patch clamp field for measurement of membrane capacitance using a single sine-wave frequency. For determining the appropriate phase setting, they used a simple and easy trick - dithering of the compensation

network while changing the phase for obtaining a maximum signal. This method is appropriate under stationary conditions.

Commonly used extensions, however, particularly the automatic 'phase tracking', is prone to errors (see Gillis in B. Sakmann & E. Neher Eds. Single Channel Recording 2nd Edition, Plenum). For this reason several novel schemes, including 2 frequency methods, have been suggested in the meantime. The minimum requirement for determining the three unknowns of the equivalent circuit of the cell under study involves three experimental parameters. The 1/2 frequency method (or Lindau-Neher technique) uses real and imaginary part of a sine-wave signal plus the DC-conductance to determine membrane capacitance, membrane conductance, and access resistance. This technique was adopted long ago by leading patch clamp laboratories. It is fully



implemented in the PULSE software together with two further variants of capacitance measurement.

The PULSE software implements both the classical single frequency and the 1/2 frequency (or Lindau-Neher) technique in a convenient user-friendly way. It provides 'internal calibration' which automatically corrects for phase-shifts and frequency dependent attenuation, so that the admittance of the external circuit is obtained in calibrated SI units. On this basis the parameters of the equivalent circuit are calculated 'online' according to Lindau & Neher.

In addition, the EPC 9 or EPC 10 offers automatic capacitance compensation. When activated repetitively (capacitance tracking!) this provides a continuous readout of capacitance. The experimenter can observe the residual capacitive current for a validation of correct compensation.

Since the amplifier 'knows' the settings of the compensation network, the 1/2 frequency method can operate while the bulk of the capacitance is compensated. Consequently the amplifier can be operated at high gain without saturating. High gain implies low noise. The high resolution admittance measurement of the residual capacitance together with the compensation circuit provides an effective dynamic range which has yet to be achieved with any other instrument.

The screenshot shows the 'LockIn Configuration' dialog box. It has several sections: 'LockIn Mode' set to 'Sine + dc', 'Calibration Mode' set to 'Calculated', 'Phase Shift' set to '0.0°', 'PL-Phase' set to '0.0°' with a 'Compute' button, and 'Calib. Sequence' set to 'IV'. Below these is a large yellow button labeled 'Perform Measured Calibration'. There are two checked checkboxes: 'Write to Notebook' and 'Show Label'. Under 'Show Label', 'Fixed Scaling' is selected. 'Display Y-min' is set to '40.0 pF', 'Display Y-max' is set to '50.0 pF', and 'Averaged points' is set to '1'. 'Connect Markers' is unchecked and set to 'Plus'. At the bottom is a large yellow button labeled 'Done'.

**Technical specifications**

**PULSE**

*Data acquisition*

- 2 channels A/D and 1 channel D/A (stimulation)
- 3 channels D/A (Trigger, auxiliary stimulation channels)

*Amplifier*

✓ EPC9 Amplifier EPC8 Remote EPC8 Local EPC7 Amplifier Telegraphing Amplifier
EPC9 Demo Mode EPC8 Remote Demo EPC8 Local Demo EPC7 Demo Mode Teleg. Amplifier Demo
Other Amplifiers

All 'clamp' amplifiers are supported. Several amplifier windows for e.g. EPC 9, EPC 10 and Telegraphing Amplifiers are available. Other amplifier windows can be generated.

*Extensions to Pulse*

Activate LockIn Fura Configuration ⌘Y Turn X-Chart On Inactivate All Extensions
--

- X-CHART  
Multichannel chart recorder for up to 16 channels.
- FURA  
Performs synchronized ratiometric fluorescence- and electrophysiological measurements.
- LOCK-IN  
Software lock-in to measure membrane capacitance.

**PULSEFIT**

PULSEFIT is an analysis program for pulsed data files. Data can be analyzed on three hierarchical levels:

*Sweep Fit*

Fit of raw data traces. Within specified windows various functions can be fitted with a Simplex algorithm to the raw data traces:

*Raw Data Fits*

- Polynomials
- Exponentials
- Gaussian Fit
- Versatile functions according to Hodgkin & Huxley gating formalism

*Series Fit*

Fit of parameters as derived from the fit to raw traces (e.g. peak current) on the level of a series of traces as a function of various parameters (e.g. pulse, potential):

*Result Fits*

- Polynomials
- Exponentials
- Boltzmann Laws
- Various Current-Voltage Relationships

*Group Fit*

Fit of parameters as derived from the fit to data on the level of a series (e.g. reversal potentials) as a function of various parameters (e.g. concentration of an ingredient of the bathing saline):

*Parameter Fits*

- Polynomials
- Exponentials
- Boltzmann Laws
- Dose-Response Curves

**PULSE/PULSEFIT**

*In/Out & data management*

- Export to ASCII (MAC and DOS) format, IGOR
- Printout of raw data and Online Analysis results

*Graphical representation*

- Oscilloscope
- Zoom
- Dimmed Overlay
- Absolute and relative Cursor Measurements

*Processing*

- p/n Leak Correction
- Zero Correction
- Software Gaussian Filter
- Buffer (basic arithmetic Operation)
- Fit of Data Traces

*Miscellaneous*

- Runs on Mac OS up to version 9 and Windows 98/NT/2000/XP
- Macro Recording
- Online Analysis

**Hardware requirements**

- For Data acquisition  
EPC 10 or EPC 9 amplifier or LIH 1600, ITC-16 or ITC-18 acquisition interface
- Computer  
See data sheet "Recommended Configuration"

**Related Products**

**X-CHART**

Software implemented chart-recorder.

**FURA**

Extension for synchronized ratiometric fluorescence measurements

**PULSETOOLS**

Software for data reorganisation, editing and averaging. Amplitude histograms and noise analysis.

**PULSESIM**

Modeling and simulation of data based on kinetic schemes or discrete Markov models.

**EPC 10**

The fully computer controlled patch clamp amplifier with built-in interface board.

**LIH 1600**

16 bit multi-channel data acquisition system

**EPC 8**

The successor of the EPC 7. Manual or digital control selectable.

**EPC 7 PLUS**

The classic patch clamp amplifier for single channel and whole-cell measurements.

**PIP 5**

Temperature controlled micro pipette puller.

**Service & Support**

As the first manufacturer of patch clamp amplifiers in the world HEKA knows the needs of scientists. We provide exceptional pre and post sales customer support from our trained international sales representatives and our own technical support advisors. With thousands of high performance hardware and software products in daily use worldwide we understand all aspects of data acquisition systems not just the software.



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*A guarantee of one year applies on all instruments.*

