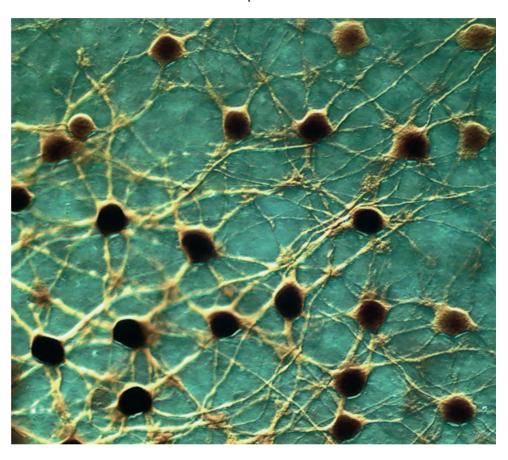


PATCHMASTER

Multi-channel data acquisition software



If you believe your job is to make new discoveries....

discover the software PATCHMASTER:

- operates on Windows and Macintosh platforms
- full software control of HEKA patch clamp amplifiers
- calibrates and tests the patch clamp amplifiers of the EPC 9 and EPC 10 series
- featuring new concepts of adaptive feedback control and global variables
- suitable for research and industry

....featuring....

- digital oscilloscope
- versatile waveform generator
- multi-channel stimulation and data acquisition
- control of external devices
- · powerful online analysis
- experiment automation and standardization
- macro programming
- · data tree editor
- full data integrity due to parameter storage

...including...

- software Lock In amplifier for capacitance measurements in whole-cell and on-cell mode
- Photometry extension to control light sources and analysis of fluorescence measurements
- Batch Control for platform independent control of PATCHMASTER from other software packages



Multi-channel data acquisition software

PATCHMASTER goes beyond the limits of conventional patch clamp software.

PatchMaster sets new standards in electrophysiological software

PATCHMASTER offers features meeting the highest standards of modern electrophysiology. This new software, running on PCbased and Macintosh computers, harbors all the comfortable functions of PULSE, but offers a variety of novel procedures aimed to make electrophysiological research even more versatile and efficient. With PATCHMASTER, experimental design, performance, and analysis become much more flexible, giving rise to a high degree of automation and providing access to experimental protocols that were thus far unattainable with commercial software

Major innovations of the PATCHMASTER program

More channels

PATCHMASTER provides up to 8(16) A/D input channels and 4(8) D/A stimulation channels that can be sampled at high speed. In addition, the number of channels is no longer limited to the number of available AD/DA channels.

Virtual traces

Acquired data traces can be processed online by mathematical functions to compile additional derived data traces.

Extended trigger possibilities

Up to 16 trigger outputs can be defined like a stimulation channel.

Individual timing for different channels

Channels can have completely independent timing and pulse patterns. This allows easy setup of complex stimulation patterns for multiple channels.

Data compression

For all channels, individual data compression factors can be specified to reduce the amount of stored data.

Global parameters for sequence editing

For definition of a pulse generator sequence, global parameters can be used, e.g. to define the duration of a segment or the amplitude of a stimulation. If these parameters are used in multiple segments or sequences, all locations can be edited by changing a single global parameter.

Multiple Telegraphing Amplifiers

More than one telegraphing amplifier can be supported by using multiple lookup tables.

Extended Lock-In

Simultaneous capacitance measurements on multiple headstages of a patch clamp amplifier (EPC 10 Double, Triple, and Quadro) and On-Cell capacitance measurements at high sine wave frequencies are now possible.

Extended Online Analysis

An arbitrary number of online functions and methods can be defined and saved. Even complex calculations are now possible. Online analysis methods can be directly linked to acquisition sequences.

Two Online Analysis windows

Up to 12 graphs can be displayed in two different Online Analysis windows. For example, one window can be used to display series derived analyses such as IVs, and the other for display of time lapse data.

Photometry feature

Multi-wavelength stimulation for multiple fluorescence excitation systems and analysis for multiple fluorescence signals is now supported.

Protocol Editor for automation

A complete experiment can be automated and standardized, including incorporation of feedback from online analysis or external devices.

PATCHMASTER – a program for patch clamp, 2-electrode voltage clamp experiments, and general data acquisition

Full support of HEKA's patch clamp amplifiers EPC 7, EPC 8, EPC 9, and the EPC 10. The novel program design of PATCH-MASTER is perfectly suited for the operation of multi-channel stimulation as used for patch clamp amplifiers with multiple headstages (e.g. EPC 9 Double / Triple, EPC 10 Double / Triple / Quadro).

Additionally, PATCHMASTER can be run with any other patch clamp amplifier, electrophysiological current clamp or voltage clamp devices (e.g. two-electrode voltage clamps used for research on *Xenopus* oocytes) or a standalone data acquisition interface for general stimulation/data acquisition purposes.

Display Buffer **Macros** Oscilloscope Change the display of the Add, subtract or accumulate Macro features allow the Digital oscilloscope with oscilloscope (e.g. grid, dimmed zoom, digital filters, and your measured traces. recording of routine functions overlay. The size of the overlay). and then accessing these Macros by a simple mouse click window can be customized. or key stroke. PatchMaster Replay File Edit Windows Replay Display Buffer Notebook Macros EPC10 Help Export your data (e.g. single - 0 × _ | | × sweep, online analysis results) to Label show Root to idle 10:42:58 00:09:08 Set printer, Matlab, IgorPro® files or Comment ASCII (MAC or WINDOWS). 3-平 Toff 3 2 25 of 25 (Measure) Scan IV Overl.Ser 4 - 🗸 i IV Inst.IV **Replay Window** 5 -▼ IV 10m Gating Trace 1 Acquired data and previous Dig. Filter -Sodium C 6 - ₹ IV recorded data files can be Off 00 Y-scale reviewed and edited. Data files 3.00 - | D | X | acquired with PULSE are Y-offs, 🤄 automatically converted to the 1.09nA 0.00 Start Time PATCHMASTER file format. 474.pA 0.0 % End 0.007 60.0mV 100.0 % **Online Analysis** -7.44pA Page Immediate analysis of the just 4 1.0 ¢ -3.15nA acquired or replayed data. The X: 50.0 ms Y: 2.00 nA analysis result is shown as a -6.29nA (Break) (Stop) (Next -60.0mW 0.000function of a variety of parameters, specified by the 2(Continuou) 3 Hinf 4 Sine 5 Tails 6(TestSeries 1 0 user. Wait Resume 1 Prot-1 2 Prot-2 3 4(Motebook_12-0ct-2005 _ | × Mean[A], Amp 12 [V], Min[A] Sweep #, 179.pAs -60.000m, -5.6006p, -7.4411p -5.7433p, -55.000m, -13.113p -15.531p, -50.000m, -32.285p -23.274p, -45.000m. -63.493p 89.4pAs -29.740p, -40.000m, -128.26p -58.630p, -35.000m, -238.89p -75.626p, -30.000m, -408.62p -652.28p -106.09p, -25.000m, 0.00As -126.66p -20.000m-1.0389n 13.0 25.0

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.

Protocol Editor

With the Protocol Editor, complex experimental procedures can be designed, stored, and executed.

5.0 mV/pA

Input ADC

Imon2

Onff

Onoise

C-fast

Rang

C-slow

Rs Comp

Leak Comp

Filter 1

Filter 2

2 Continuous 3 (

Sequence

20.0 μs (50.0kHz StartTime 9.00

Not Triggered

StartSeg

(double

0 mV

SET-UP ON-CELL WHOLE-CELL

LJ 0.0mV Uo 0.0mV Auto Track

0.00 pF

Off

Off

50.00 pF

R-series 20.0 MOhm (>) Auto

0.00 A

The virtual front panel of EPC 10 patch clamp amplifier

) Show All

Recording Mode On Cell

Length

5.0 ms

0.5 µs

Cap Track

Off

Delay Off

Off Auto Track

Bessel 10 kHz

Bessel 2.9 kHz

(>) Auto

Test Pulse | show both |

Control Selection

Three different window sizes can be set by a mouse click to show different sets of control buttons.

Macros

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

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.

Capacitance 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.

Monitoring

Gain and holding parameters can be set. The measured current and voltage as well as the resulting membrane resistance (if applicable) is always displayed.

Offset Compensation

A liquid junction potential can be specified. The offset potential can be cancelled out automatically. The Track function allows repetitive automatic cancellation of the offset potential.

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.

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

DELETE

EXECUTE

12.70 ms 3.700 ms

12.70 ms

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

No of Sweeps

Sweep Interva

Sample Interval

LOAD SAVE

Wait before 1. Sweep

200, ms

Flexible Pulse Generator configures stimulation and acquisition

51 LIST COPY MOVE

Total

Stored

Stimulus

Sweep Length

Channel Length

Timing Determines the number of sweeps and the sweep and sample intervals. **DA-channels** Multiple DA

channels and digital trigger lines can be addressed for output.

Segments

A pulse pattern consists of an arbitrary number of segments.

Template Preview

The stimulation template is also shown graphically.

Stimulus -> DA Points Store Leak Segments 🚺 🗘 🗆 Store 1 Segment Class Voltage [mV] hold V-mam n2 100 val -120 Filter Factor Duration [ms] p1 200 V-incr. Mode Increase V-fact./incr. [mV] t-incr. Mode t-fact./incr. [ms] Rel Y-sed Draw: Active Channel, all Sweeps | Delay: DA 0.00 s | AD 0.00 s V-membrane [mV] (display) ☐ Set Last Seg. Amplitude Leak Pulses No of Leaks Leak Delay -100 ms Leak Alternate 20.0mV 0.200 Leak Size Leak Hold [mV wait = abs, hold p10 n.nnn n nnnn 2 0000m 100 00m

Segment parameters are assignable by global variables.

pgf-Parameters

Leak

Determines various parameters of the leak pulses for p/n subtraction.

Sweep Length

The length of the stimulus and the amount of acquired data is displayed.

AD Channels

Multiple AD channels can be acquired simultaneously and processed (virtual trace). Data compression and store/nonstore flags on individual traces reduces the required data storage capacity.

Filter Factor

The input filter is set automatically with respect to the sampling rate.

Analysis

Direct link of the stimulation template to the corresponding analysis method.

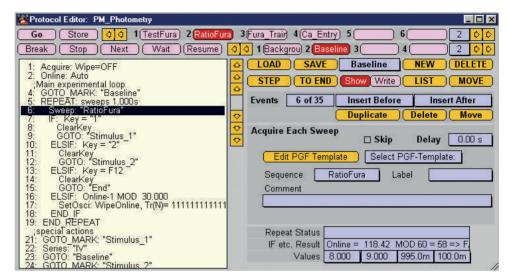
Break Condition

Break conditions can be defined for each AD channel.

Protocol Editor - Tool for automation and standardization of experiments

The protocol editor is a completely new feature of PATCH-MASTER. With this editor, complex experimental procedures can be designed, stored, and executed. This tool greatly increases the versatility of PATCHMASTER and provides means for automatic experiment performance.

The principal idea of the protocol editor is to generate a list of events or tasks, which then are executed automatically. Various functions such as REPEAT LOOPs, input queries, or conditional statements allow for the generation of complex interactive processes.

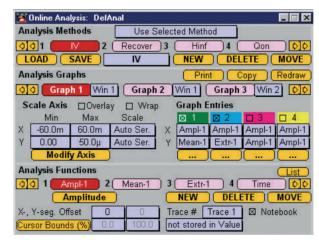


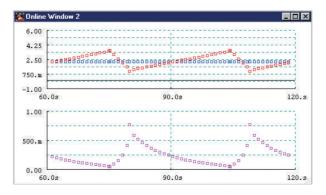
The features offered by the protocol editor will be appreciated by scientists asking for complex, precisely timed experimental protocols. In addition, the high degree of automation possibilities increases efficiency, minimizes experimental errors and is thus suited for industrial applications.

Online Analysis - Powerful on- and offline data processing

An arbitrary number of analyses can be performed on newly acquired or replayed data. Directly analyzed data or derivative data, obtained by application of mathematical functions on the analysis results, are then displayed in a versatile manner in several graphs placed in two independent windows. This allows for separation of different data types, for example, current-voltage plots are shown separately from time-based data (e.g. chart recording).

Analysis templates can be predefined and stored. Thus, several analysis procedures are available such that various incoming data types can be analyzed without extra editing just by switching between analysis procedures. A direct link between Pulse Generator sequences and analysis procedures provides definition of data acquisition and analysis prior to the experiment.





Software Lock-In amplifier for capacitance measurements

The PATCHMASTER software, combined with our EPC 9 and EPC 10 patch clamp amplifiers provide you with everything you need for time-resolved measurement of membrane capacitance (Cm).

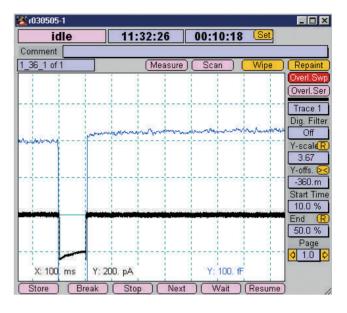
The EPC 9 or EPC 10 is the ideal patch clamp for Cm measurements since all the relevant parameters are under control of PATCHMASTER. No additional hardware such as Lock In amplifiers and filters are required.

Implemented Lock In modes

In 1982 Neher and Marty introduced the Lock In amplifier into the patch clamp field for Cm measurements using a single sine wave frequency. For determining the appropriate phase setting, they used dithering or the compensation network while changing the phase for obtaining a maximum signal for Cm. This method is appropriate under stationary conditions for measurements of changes in Cm. We refer to this method as the "piecewise-linear" method.

"piecewise-linear" method.
Since the piecewise-linear
method is prone to errors (see
Gillis in B.Sakmann & E.Neher
Eds. Single Channel Recording
2nd Edition, Plenum Press)
Lindau and Neher introduced in
1988 a method using the real and
imaginary part of the admittance
plus the DC-conductance to
determine the absolute values of
Cm, membrane conductance,
and access resistance. We refer
to this method as the "Sine+DC"
method.

The best resolution of small changes in Cm are achieved in the cell-attached patch clamp configuration. Since a different equivalent circuit applies in this recording mode, a third method, referred to as the "On-Cell" method, has been implemented.



Different modes of calibration

Digital control of the filter settings, gain and compensation networks that are featured with the EPC 9 and EPC 10 patch clamp amplifiers directly benefit the *Calculated* calibration mode. Phase shifts introduced by the measuring system can be calculated and the corrected phase of the Lock In amplifier, which is dependent upon the recording conditions, can be set automatically.

A *Measured* calibration method allows the phase and attenuation of the recording system to be determined by analysis of the admittance of a pure resistor at the amplifiers input.

In case other procedures for determination of the phase and attenuation of the measuring system are used, a *Manual* calibration mode allows the phase and attenuation of the software Lock In to be directly set by the user.

Lowest noise recordings

In the Sine+DC mode, automatic CSlow compensation cancels the bulk of the membrane capacitance and thus allows to be operated the patch clamp amplifier in the high gain (low noise) range during Lock In measurements. In the On-Cell mode signal to noise ratios can be increased by using a higher sine wave frequency (typically 20 kHz) to resolve small changes in Cm (< 100 aF) due to fusion of single vesicles.

Simultaneous measurements from multiple patch clamp amplifiers

In combination with EPC 9 or EPC 10 Multi-Patch amplifiers, simultaneous Cm measurements on multiple amplifiers are supported by our software Lock In.

Online equivalent circuit parameter calculation

Software Lock In provides online calculation of equivalent circuit parameters and offline recalculation. Customer specific calculations can be done online by using the virtual trace feature of the pulse generator in PATCHMASTER.

Key Features

Amplifier Control

- All 'clamp' amplifiers are supported. Amplifier windows for all EPC 9 and EPC 10 amplifier types and telegraphing amplifiers are available.
- Automatic test and calibration routines for all HEKA EPC 9 and EPC 10 amplifiers are provided.

Data Acquisition

- Up to 16 input channels
- Pulsed and continuous acquisition mode
- Automatic data compression (different sample rates for different channels)
- Virtual trace for mathematical online processing of acquired channels
- For each acquisition channel a break criteria can be defined.
 An Automatic Break will stop the acquisition whenever one of break criteria becomes true.
- Variable data format for storage (INTEGER, REAL, LONGREAL)

Leak Subtraction

- Leak pulses (p/n correction) supported for all output channels (important for multiheadstage clamps)
- Various leak pulse storage modes: none, average, all (important for offline leak correction)

Stimulation

- Up to 4(8) stimulation channels
- Stimulation with an arbitrary number of pulse segments
- Segment types: constant, ramp, sine, square, non-stored
- Various increment modes for segment amplitude and duration
- Segment parameters assignable by global variables (pgf-parameters)

Digital Oscilloscope

- Display of up to 20 different traces
- Individual display scaling and visual appearance for all traces
- Individual digital filter settings for all traces
- Various different labeling modes (e.g. Grids+Labels, Grids+Values, Labels only)
- Zoom
- Dimmed overlay
- Absolute and relative measurements with mouse click
- Read individual data points with scan function

Online Analysis

- Arbitrary number of user defined online methods
- More than 40 predefined analysis functions
- Mathematical standard operations allow setup of user defined analysis functions
- Trace operation functions
- Two online windows with up to 12 graphs for graphical representation of analysis results.
- Link of analysis methods to different acquisition sequences

Trace Buffer

- Four independent trace buffers are available for basic arithmetic operations on the level of a trace (add, subtract, accumulate, deaccumulate)
- Buffer traces can be shown as a reference trace in the oscilloscope window.

I/O Control

- Direct access to set digital outputs, analog outputs, input parameters and to send serial commands.
- Monitoring of digital inputs, analog inputs and input parameter values.

Standardization and Automation

Macros: A series of user defined actions can be recorded and stored as a macro.

Protocol Editor: The Protocol Editor allows standardization and automation of complete experimental procedures. Within the procedure, the system can get feedback from external inputs, amplifier controls, online analysis results or user inputs and experimental parameters can be adjusted. A protocol can be started/called from another protocol.

Batch Control: The complete PATCHMASTER acquisition system can be controlled from another application. The user can write their own application with a custom tailored user interface but still benefit from the advanced features of the HEKA system.

Data Integrity

- Acquired data are organized and stored in a data tree.
 Multiple data packages are usually stored in one data file and organized in levels of Groups/Experiments/Series/ Sweeps and Traces to allow easy review, selection and analysis of all data in that data file.
- Due to the complete software control, a complete set of parameters describing the state of the amplifier and other recording conditions is stored with the data. This allows detailed reconstruction of the experiment for exact analysis at later times.

Data Export

- Export and printout of raw data traces or online analysis results
- Supported formats: ASCII, IGOR PRO, MathLab and WMF
- High level layout features of IGOR PRO supported.

Other Modules

Software LockIn:

PATCHMASTER features a software Lock-In amplifier for time resolved measurements of membrane capacitance.

- Sine+DC, Piecewise-linear and ON-CELL capacitance measurements modes
- Calculated calibration mode allows automatic correction for phase lag introduced by the EPC 9 or EPC 10 patch clamp system
- Measured and Manual calibration modes for highest accuracy in challenging recording modes
- Lock In measurements on multiple amplifiers are supported (EPC 9 and EPC 10 multi patch amplifiers required)

Photometry Extension:

- Support of fluorescence excitation light sources TILL Polychrome, PTI DeltaRAM, SUTTER DG4/DG5, Lambda-10
- Multi-excitation and multiemission protocols
- Simultaneous high-speed fluorescence and patch clamp measurements

Compatibility

- Runs on Windows 98/NT 4.0/2000/XP (requires a parallel printer port on Windows computers) and on Mac OS 9 and Mac OS X (requires a USB port on Macintosh computers).
- PATCHMASTER can read data recorded with PULSE or generated with PULSETOOLS or PULSESIM.

Hardware Requirements

- For data acquisition: EPC 9 or EPC 10 type patch clamp amplifier or ITC-16, ITC-18 or LIH 1600 data acquisition interface in combination with any other clamp amplifier.
- Computer: see data sheet "Recommended Configurations".



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