# PATCHMASTER Basics Training Webinar

If you have any Patch-Clamp related questions or comments, please feel free to send an email to the HEKA Support Team at:

support@heka.com



Frank P. Elsen, Ph.D. Electrophysiology Application Scientist Martin Oberhofer, Ph.D. Senior Support Specialist





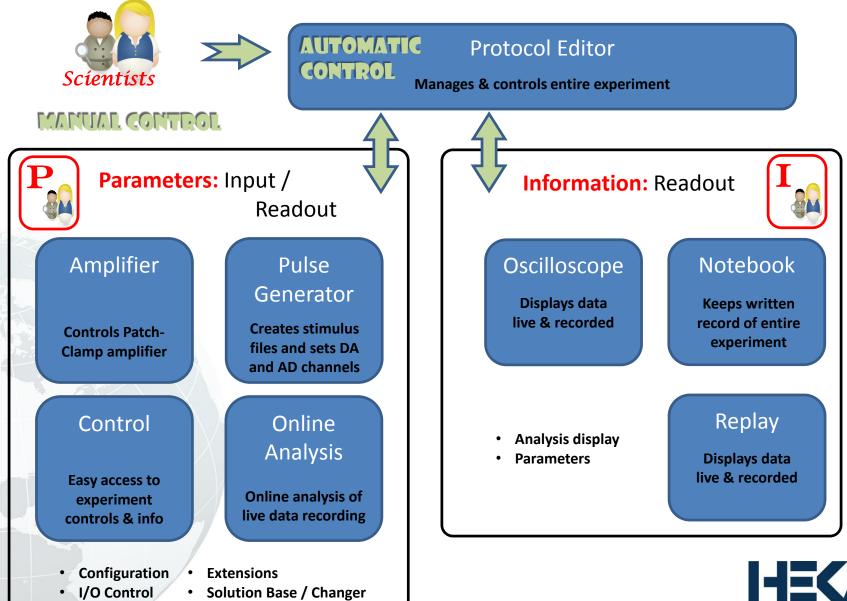
## Overview

- > PATCHMASTER Simple Handling & Easy Work Flow
- Installation Procedure (PATCHMASTER & EPC10\_USB)
- PATCHMASTER Windows (Purpose & Function)
  - Graphical User Interface (GUI)
  - Amplifier
  - Oscilloscope
  - Replay
  - o Pulse Generator
  - Analysis (online & offline)
  - **Protocol Editor** ("Assistant")
  - Control

> PATCHMASTER – Record, Display, Save and Analyze Data



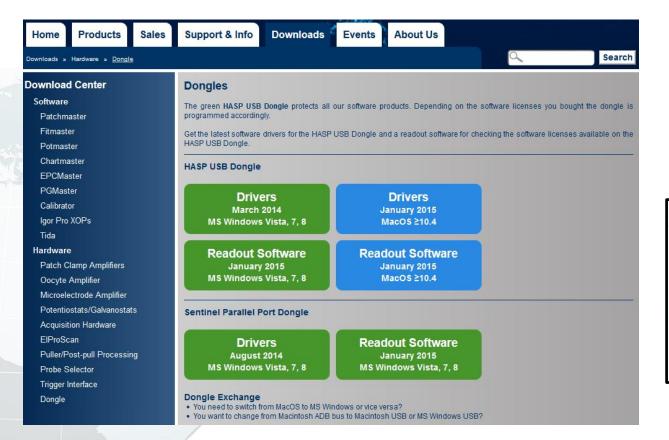
## **PATCHMASTER – Simple Handling & Easy Work Flow**



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## **Installation Procedure**

- □ Hardware Installation: Plug-in EPC10\_USB to direct USB port (PC or Mac)
- Download (www.heka.com) & install driver for protection key (dongle)
- Download & install PATCHMASTER
  - Run "Installer.exe" as administrator
  - 1<sup>st</sup> time run "PATCHMASTER" as administrator, afterwards not required



(Example: PC)





Illuminated LED indicates "fully functional"



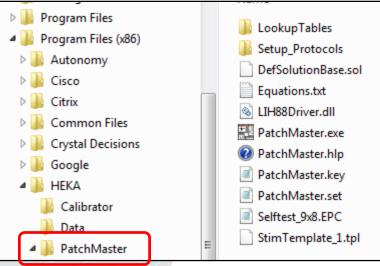


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#### C:\\Program Files (x86)\HEKA\PatchMaster



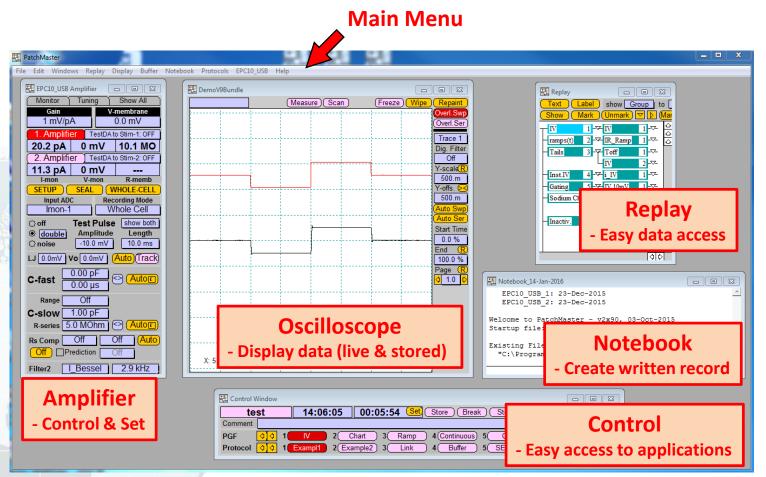
Important Files:

- PATCHMASTER.exe
- $\circ$  PATCHMASTER.set
- Default Files
  - Online Analysis (onl)
  - Pulse Generator (pgf)
  - Protocol Editor (pro)
- $\circ$  Other Files



## **PATCHMASTER – Graphical User Interface**

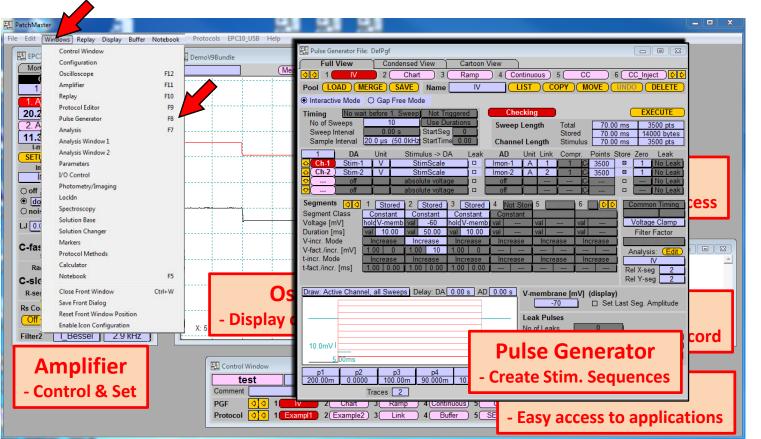
- Different windows for different functions
- □ Fully customizable and modifications are automatically saved in the PATCHMASTER.set file (Save & Exit)





## **PATCHMASTER – Graphical User Interface**

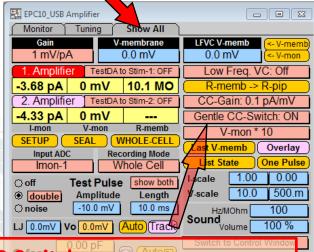
- Different windows for different functions
- □ Fully customizable and modifications are automatically saved in the PATCHMASTER.set file (Save & Exit)



### "Windows" menu

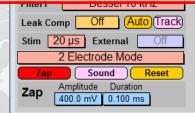


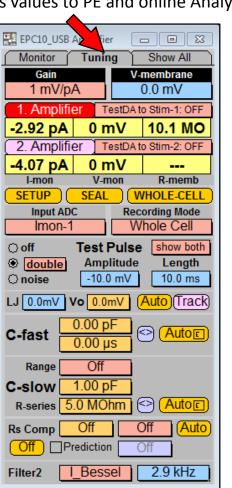
- Full Control: EPC9, EPC10 and EPC800 amplifiers (iTEV90) (limited control (through telegraphing outputs): EPC7, EPC8, AxoPatch, Warner PC-505, etc.)
   Controls: Gain, V- and I-membrane, mode, test pulse, compensation, filter, etc.
- Can be fully controlled by Protocol Editor (PE) & reports values to PE and online Analysis

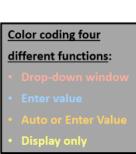


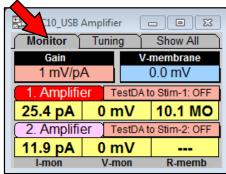
## Gentle CC-Switch

- Enables or disables "Gentle Switch" function
- Keeps Vm unchanged when switching to CC by injecting the appropriate amount of current
- Save & Easy switch between VC and CC mode





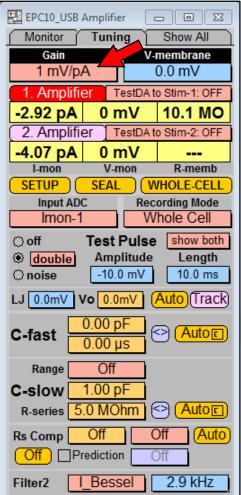








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	0.005 0.010 0.020 0.050 0.1 0.2	Feedback Resistor: I-max: C-slow range: CC mode: Rs compensation:	± 2 μΑ
✓]	0.5 1 2 5 10 20	Feedback Resistor: I-max: C-slow range: CC mode: Rs compensation:	± 20 nA
	50 100 200 500 1000 2000	Feedback Resistor: I-max: C-slow range: CC mode: Rs compensation:	50 GΩ ± 200 pA 30 – 100 pF no yes

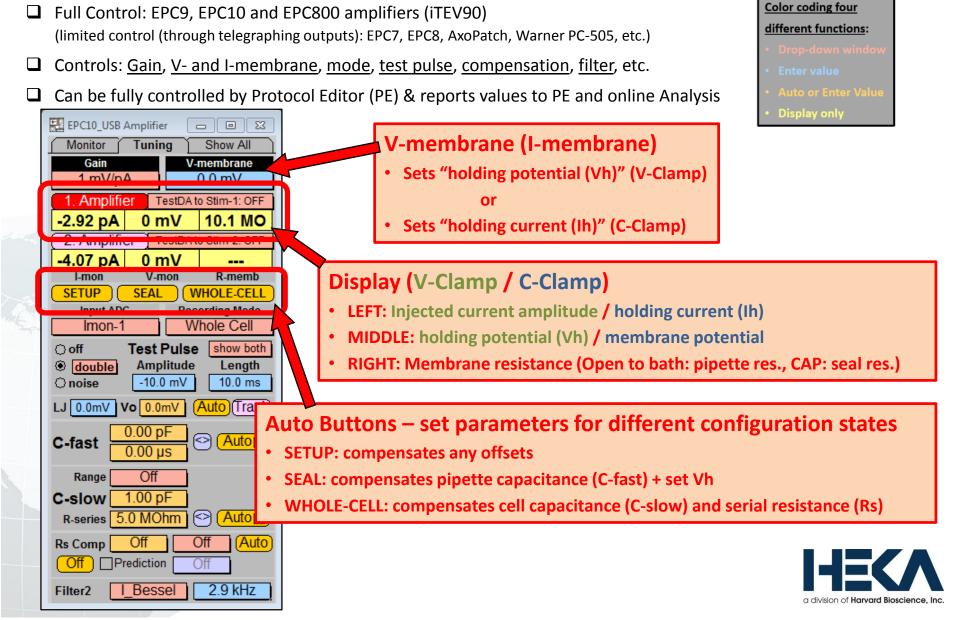


#### Color coding four

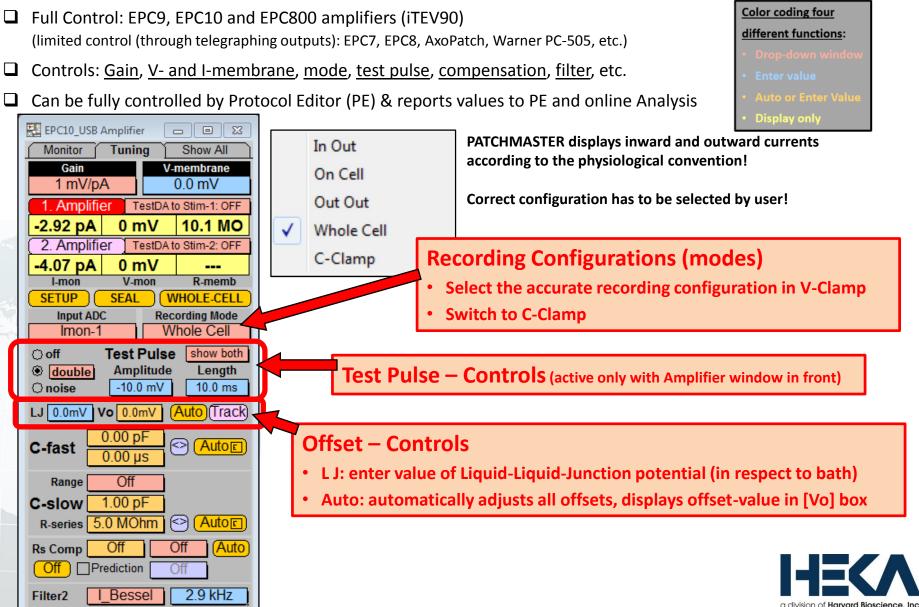
#### different functions:

- Drop-down window
- Enter value
- Auto or Enter Value
- Display only







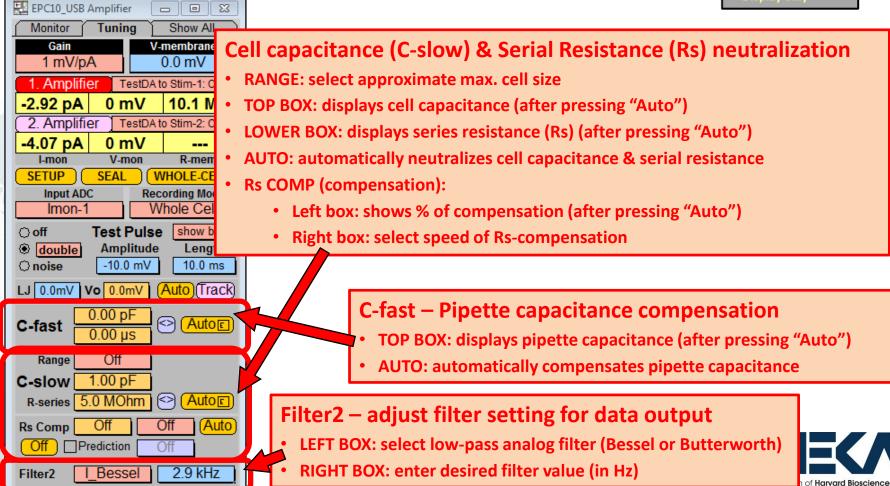


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E XX

□ Can be fully controlled by Protocol Editor (PE) & reports values to PE and online Analysis

Parameters

Marked Items

D. C. alarra D

C-slow

R-series

Rs-comp.

V-offset

CC-gain

Rs-comp. Tau

Liquid Junction

Leak Comp.

#### Main Menu – Windows - Parameters

Group Items

Series Items

Calibration Date
 Serial Number

Lockin Phase

Lockin Mode

Lockin Attenuation

Lockin Calibration

Sweep Items

08-Feb-2016

590783-T

0.000

1.000

Off

Measured

Root Items

Amplifier State	Titles	LF	Flag Group 1 Fl	ag Group 2 ) ( I
List with Traces			Copy Flags from Gr	.1) Gr.2) Gr.3)
Amplifier	EPC10 USB		Default Flags	Target to Notebo
Clamp Mode	V-Clamp		Clear all Flags (	Marked to Noteb
V-pipette	0.000 V			
🖾 Gain	1.000 mV/pA			
Ext.Stimulation	0.000		Low Freq VC	LFVC O
Filter 1	Bessel-10kHz		LFVC V-memb	0.000 V
Filter 2	2.873 kHz		CC-fast	CC-fast
Filter 2 Type	Bessel		Auto C-fast	success
Stimulus Filter	10.00 kHz		Auto C-slow	success
C-fast	5.923 pF		V-mon Scale	10x
C-fast Tau	855.9 ns		Stim Scale	10x
C-fast Range	Normal		Electrode Mode	2

22.00 pF

5.013 MOhm

79.93 %

100.0 µs

0.000 S

0.000 V

195.2 uV

0.1pA/mV

### Color coding four

Trace Items

Flag Group 3 ) (Flag Group 4 )

Target to File Marked to File

п

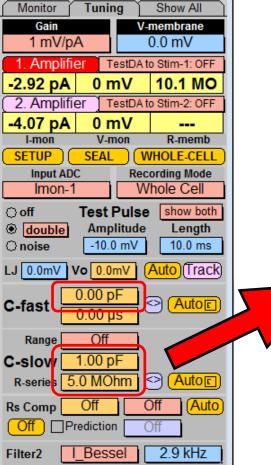
Info to Notebook Gr.1

- different functions:
- Drop-down windov
- Enter value
- Auto or Enter Value

- - X

Amplifier Items

Display only



EPC10\_USB Amplifier

- Respective parameters are updated by pressing "Auto"
- Parameter values at time of data recording are saved with data file





## **PATCHMASTER – Oscilloscope Window**



- Displays LIVE current response to:
  - Test Pulse
  - Stimulation sequences (from Pulse Generator)
- Displays data traces offline from Replay window



/indows Replay Display I	Buffer Notebook
Control Window	
Configuration	
Oscilloscope	F12
Amplifier	F11
Replay	F10
Protocol Editor	F9
Pulse Generator	F8
Analysis	F7
Analysis Window 1	
Analysis Window 2	
Parameters	
I/O Control	
Photometry/Imaging	
Lockin	
Spectroscopy	
Solution Base	
Solution Changer	
Markers	
Protocol Methods	
Calculator	
Notebook	F5
Close Front Window	Ctrl+W
Save Front Dialog	
Reset Front Window Positi	on
Enable Icon Configuration	

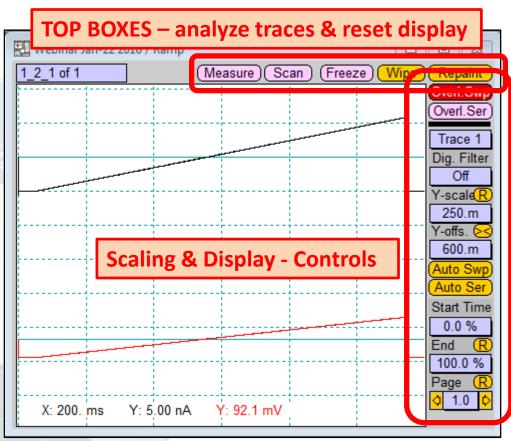


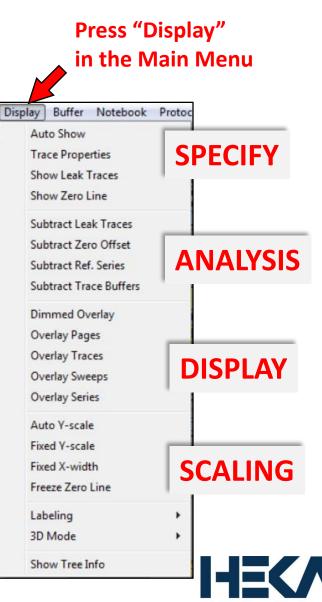
## **PATCHMASTER – Oscilloscope Window**



- Displays LIVE current response to:
  - Test Pulse
  - Stimulation sequences (from Pulse Generator)
- Displays data traces offline from Replay window

## Open data file (\*.dat) & Select a series (ramp)





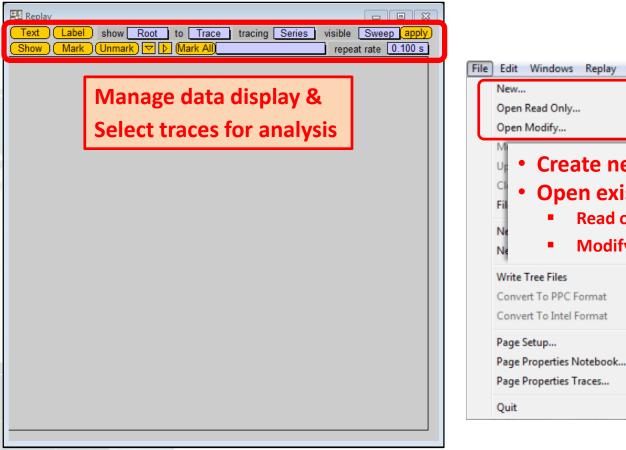
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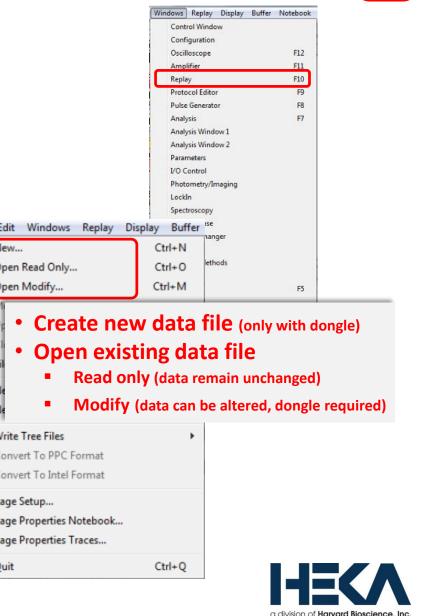
# ndow



# PATCHMASTER – Replay Window

- Data can easily be accessed from here (5-level data tree)
- □ Information (text) about experiment can be linked to data
- □ Raw data + analyzed traces are stored together

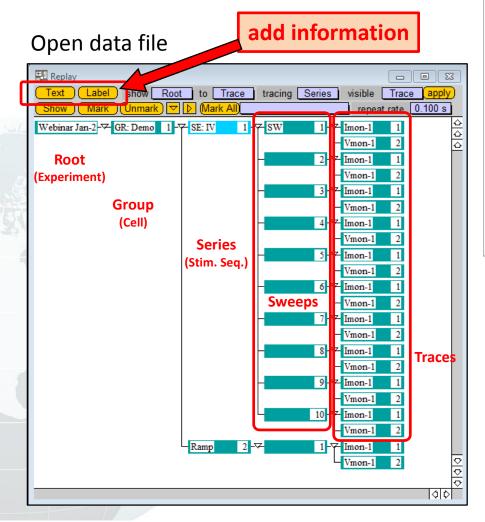




## **PATCHMASTER – Replay Window**

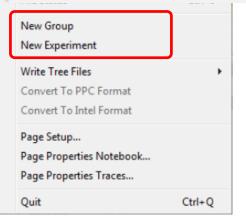


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ile	Edit	Windows	Replay	Display	Buffer
	New		Ctrl+N		
	Open Read Only			Ctrl+O	

- Insert new branch (only with dongle)
  - New Group (new cell or group of recordings)
  - New Experiment (new root or project)



#### How to use the Replay window

- One dat-file (Root) per day (recommended)
- Create new "Group" for each cell/project
- Add specific experiment related information to "Root" or "Group"



## **PATCHMASTER – Pulse Generator Window**



- □ Creating stimulus sequences (IV-curves, ramps, continuous recordings, etc.)
- □ Pool of stimulus sequences can be saved in one \*.pgf file
- Provides link to online Analysis

Pool of Stimulus Seque	ences
Full View       Condensed View       Cartoon View         Image: Condensed View       Continuous         Image: Condensed View       Continuous         Image: Condensed View       Cartoon View         Image: Condensed View       Continuous         Image: Condensed View       Continuous <td< th=""><th></th></td<>	
Timing         No wait before 1. Sweep         Not Triggered         Checking         EXECUTE           No of Sweeps         10         Use Durations         Sweep Length         Total         70.00 ms         7000 pts           Sweep Interval         0.00 s         StartSeg         0         Channel Length         Stored         70.00 ms         7000 pts           Sample Interval         10.0 µs         (100.kHz)         StartTime         0.00         AD         Usit         Link         Control         70.00 ms         7000 pts	
AD Unit Link Compr. Points Store Zero Leak C 7000   1 No Leak	
Segments       I       Stored       I       Stored       I       Stored       I       Stored       I       Common Timing         Segment Class       Constant       Constant       Constant       Constant       Constant       Common Timing         Voltage [mV]       hold/V-memb val       -60       hold/V-memb val        val        Voltage Clamp         Duration [ms]       val       10.00       val        val        Filter Factor         V-incr. Mode       Increase       Increa	
V-fact./incr. [mV]       1.00       0       1.00       0       1.00       0       1.00       0       Analysis:       Color         t-incr. Mode       Increase       Increase <th></th>	
0     □     Set Last Seg. Amplitude       Leak Pulses     0     □       10.0mV     Leak Alternate     Leak Alternate       Leak Size     0.100     (Alt.Leak Average)	
500ms         Leak Hold [mV]          wait = abs. hold           p1         p2         p3         p4         p5         p6         p7         p8         p9         p10           200.00m         0.0000         100.00m         90.000m         10.000m         45.000m         0.0000         0.0000         0.0000           Traces 2	

Windows	Replay Dis	splay Buffer	Notebook
Cont	trol Window		
Con	figuration		
Osci	lloscope		F12
Amp	lifier		F11
Repl	ay		F10
Prot	ocol Editor		F9
Puls	e Generator		F8
Anal	ysis		F7
Anal	ysis Window 1		
Anal	ysis Window 2	5	
Para	meters		
1/0 0	Control		
Phot	ometry/Imagi	ng	
Lock	In		
Spec	troscopy		
Solu	tion Base		
Solu	tion Changer		
Mark	cers		
Prot	ocol <mark>Me</mark> thods		
Calc	ulator		
Note	book		F5
Clos	e Front Windo	w	Ctrl+W
Save	Front Dialog		
Rese	t Front Windo	w Position	
Enab	le Icon Config	juration	



## **PATCHMASTER – Pulse Generator Window**

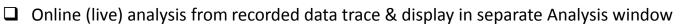


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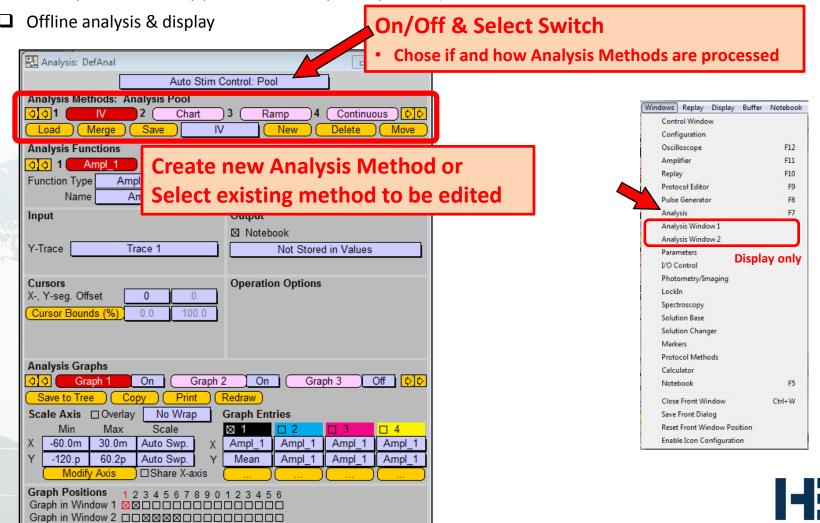
- □ Creating stimulus sequences (IV-curves, ramps, continuous recordings, etc.)
- □ Pool of stimulus sequences can be saved in one \*.pgf file
- □ Provides link to online Analysis

Pulse Generator File: DefPgf for webinar	Check "STORE" box,
Full View Condensed View Cartoon View	▲ otherwise data trace will NOT be saved !
▲ 1 V 2 Chart 3 Ramp 4 Continuous 5 CC 6 CC_Inject 👂	otherwise data trace will not be saved :
Pool LOAD (MERGE) SAVE Name IV LIST COPY MOVE UNDO DELETE	
Interactive Mode     Gap Free Mode	
Timing     No wait before 1. Sweep     Not Triggered     Checking     EXEC       No of Sweeps     10     Use Durations     Sweep Length     Total     70.00 ms     0 bytes       Sweep Interval     0.00 s     StartSeg     0     Stored     70.00 ms     00 bytes       Sample Interval     10.0 µs     (100.kHz)     StartTime     0.00     Channel Length     0 µs     0 ms     200 pts	
DA Unit Stimulus -> DA Leak AD Unit Link Compr. Points Store Zero Leak	
	Stimulation & Recording channels
Segments () () 1 Stored 2 Stored 3 Stored 4 Not S ore 5 Constant 6 C () Common Timing	
Segment Class         Constant         Constant         Constant         Constant           Voltage [mV]         holdV-memb val         -60         holdV-memb val          val          Voltage Clamp	
Duration [ms] val 10.00 val 50.00 val 10.00 val val val Filter Factor	Link to online Analysis
V-incr. Mode         Increase         Increa         Increase         Increase	Link to online Analysis
t-incr. Mode Increase Increase Increase Increase Increase Increase	Enter exact name of analysis method
t-fact./incr. [ms] 1.00 0.00 1.00 0.00 1.00 0.00 Rel X-seg 2 Rel Y-seg 2	
Draw: Active Channel, all Sweeps) Delay: DA 0.00 s AD 0.00 s	Specify relevant segments in stimulus sequence
0 Set Last Seg. Amplitude	
o of Leaks	Shape & Size
10.0mV     eak Delay     -100. µs     Leak Alternate       ak Size     0.100     (Alt Leak Average)       5.00ms     eak Hold [mV]	of Stimulation Sequence
<u>p7</u> p8 p9 p10	
200.00m 0.0000 100.00m 90.000m 10.000m 45.000m 0.0000 0.0000 0.0000 0.0000 Traces 2	

## **PATCHMASTER – Analysis Windows**

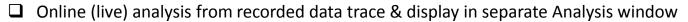


□ Vast amount (98) of analysis functions (timing, stimulation properties, measurements, LockIn, AP analysis, trace & sweep parameters, math, power specs, etc.)



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## **PATCHMASTER – Analysis Windows**



□ Vast amount (98) of analysis functions (timing, stimulation properties, measurements, LockIn, AP analysis, trace & sweep parameters, math, power specs, etc.)

□ Offline analysis & display

<u> ।</u>	lethods: Analysis I IV 2 Merge Save		ol amp4 (Contin New (Delete	uous)(¢](¢) (Move)	Define An	alysis Fun	ctions for s	elected method
	Ampl 1 2	Mean ) 3 Mea	n(start) ) 4 (Mean(	( <u>List</u> ) end))				
Function Ty Nai		Change	New Delete	Move				1
V Trace	Timing	Measurements	Lockin	Trace Param.	Math	Trace	Power Spectra	
Y-Trace	O Sweep Count	O Extremum	O Lockin CM	O Trace Count	O Equation	O Trace	O log( Frequency )	
	O Analysis Index	O Maximum	O Lockin GM	O C-slow	$\bigcirc$ Y(x): y at pos = x	O Equation	O Density	
Cursors	() Time	O Minimum	O Lockin GS	O R-series	O Constant	O Q = Integral		
X-, Y-seg. (	O Timer Time	O Extr. Amplitude	O Lockin_Phase	O Rs-value	⊖a+b	0 1 / (trace)	Histogram	
Cursor Bo	O Series Time	O Min. Amplitude	O Lockin Freq	C Leak Comp.	⊖a-b	01/(Q)	O Histogram Ampl	
	O Real Time	O Max. Amplitude		O M-conductance	⊖ a * b	O In (trace)	O Histogram Bins	
		C Time to Extremum	AP Analysis	Cell Potential	⊖ a / b	⊖ In (Q)		
	Stim. Properties	C Time to Maximum	O Baseline	O Seal Resistance	🔿 a in b	O log ( trace )		
Analysis G	Amplitude	C Time to Minimum	O AP Amplitude	O Int. Solution	O abs	O log ( Q )		
	O Duration	C Time to Threshold	O Time to AP Ampl	O Int. Sol. Value	() log	🗘 dt = Differential		
	🗘 Rel. Seg. Time	O Threshold Ampl.	O Repol Ampl	O Ext. Solution	O sqrt	O Trace x-axis (time)		
Save to T	O Abs. Seg. Time	O Thres. Crossings	O Time to Repol Ampl	O Ext. Sol. Value	O arctan	O Stimulus		
Scale Axis	O Scan Rate	O Mean	O Rise Time		🔿 1/a			
Min		O Variance	O Up Slope	Sweep Param.	O 1/log			
X -60.0m		O Integral	O Rise Time Delay	O Temperature	O 1/sqrt			
		O Anodic Q	O Decay Time	O Pip. Pressure	O 1/arctan			
Y -120.p		Cathodic Q	O Down Slope	O Digital-In				
( <u>Moo</u>		O Reversal	O Decay Time Delay	O User_1				
Graph Pos		O Slope	O Decay Tau	O User_2				
Graph in W		O Intercept						
Graph in W		🔿 Tau				Cancel	Done	
								a division of Harvard Bioscience, Inc.

## **PATCHMASTER – Analysis Windows**

- □ Online (live) analysis from recorded data trace & display in separate Analysis window
- □ Vast amount (98) of analysis functions (timing, stimulation properties, measurements, LockIn, AP analysis, trace & sweep parameters, math, power specs, etc.)
- □ Offline analysis & display

Analysis: DefAnal	
Auto Stim Control: Pool         Analysis Methods: Analysis Pool         Image: Save         Image	
Input Output	
Y-Trace Trace 1 Notebook Not Stored in Values	<ul> <li>Modify selected Analysis Function</li> <li>Available options depend on function type</li> </ul>
Cursors     Operation Options       X-, Y-seg. Offset     0       Cursor Bounds (%)     0.0	Available options depend on function type
Analysis Graphs	
Image: Constraint of the second se	Specify Display (Graph) Parameters
Scale Axis       Overlay       No Wrap       Graph Entries         Min       Max       Scale       Image: Constraint of the state of the stat	
Graph Positions         1 2 3 4 5 6 7 8 9 0 1 2 3 4 5 6         Graph in Window 1 ⊠⊠□□□□□□□□□□□□□□□□□□□□□□□□□□□□□□□□□□	

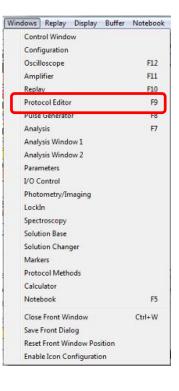


## **PATCHMASTER – Protocol Editor**



- □ Allows user to fully automate entire experiment (amplifier, acquisition and analysis)
- □ Saving settings allows user to replicate experimental conditions precisely
- **L** Exchange of protocol files between colleagues

Protect Editor Dense	tocol Files
1 SETUP 2 SEAL 3 (WHOLE-CE      Command: "E Mode 3; Whole Cell"     Command: "E CSlow 0; 0 pF"     Command: "E RSeries 0; 0 M-ohm"     Command: "E PulseMode 1, single pulse"     Command: "E PulseAmp -5.0mV"     Command: "E PulseAmp -5.0mV"     Command: "E PulseDur 10.0ms"     Command: "E Gain 9; 2.0 mV/pA"     Command: "E AutoZero"     T: Command: "E SaveRpip"	4       5       6       1       0         A       STEP       TO END       Write       LIST       MOVE         Command       Record Macros       Relative Value       non-blocking         Events       1 of 11       Insert Before       Insert After         Duplicate       Delete       Move         Macro Command       Skip       Delay       0.00 s         Command       E Reset       Enter & Modify Commands         Repeat Status       Inon-blocking         If etc. Result       If etc. Result





## **PATCHMASTER – Protocol Editor**

Protocol Sequence



- □ Allows user to fully automate entire experiment (amplifier, acquisition and analysis)
- □ Saving settings allows user to replicate experimental conditions precisely
- **L** Exchange of protocol files between colleagues

Protocol Editor: Demo 1 SETUP 2 SEAL 3 WHOLE-CI 1: Command: "E Reset" 2: Command: "E Mode 3; Whole Cell" 3: Command: "E CSlow 0; 0 pF" 4: Command: "E RSeries 0; 0 M-ohm" 5: Command: "E PulseMode 1, single pulse" 6: Command: "E PulseTrace 0; current" 7: Command: "E PulseAmp -5.0mV"	List of Events	Break Chain Protocol Clear Key Goto Goto_Mark IFThen ElselfThen Else Launch Macro Command Repeat	Data/Display Analysis Digital Filters Display Properties Export File Operation Replay Trace Buffer
	non- Repeat Status IF etc. Result	Acquire Properties Acquire Series Set Sweep Label Test Pulse Hardware Amplifier Serial Output Set DAC Set Digital Bit Set Digital Word Set Solution Changer Data/Display	Messages Annotation Beep Write Icon Value Extensions LockIn EIProScan Photometry E-Chem Methods Cyclic Voltammetry Impedance Spectroscopy Open Circuit Potential Square Wave Voltammetry

## **PATCHMASTER – Control window**



F12

F11 F10

F9

F8

F7

Windows Replay Display Buffer Notebook

Control Window

Configuration Oscilloscope

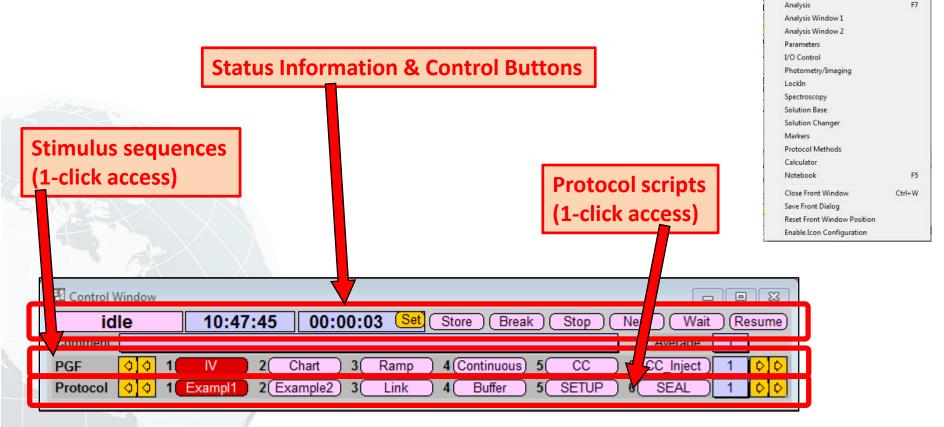
**Protocol Editor** 

Pulse Generator

Amplifier

Replay

- Provides controls to steer the experimental procedure
- Offers information on status of current experiment
- Easy access to 1-click activation of all stimulus sequences and protocol scripts



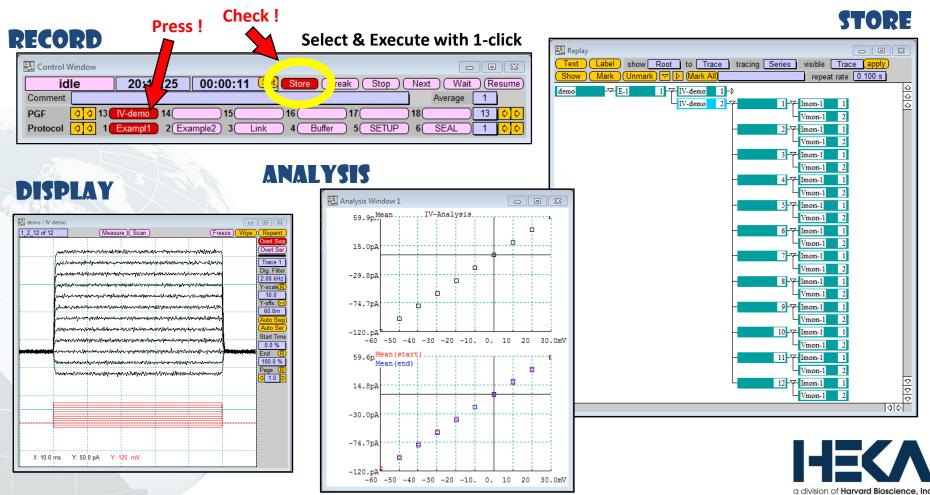
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\* For detailed information, please download "PATCHMASTER manual" from HEKA website \*

## PATCHMASTER – Record, Display, Save and Analyze Data

#### □ <u>1-click</u>:

- Records data (Control window starts stimulation sequence or protocol script)
- Displays live data traces (Oscilloscope)
- Saves (store) data (Replay window displays data tree)
- Analyzes data while doing all of the above (Pulse Generator links to online Analysis)



## **Overview of the following LIVE Demonstration**

## Maintenance

- Save changes in "PatchMaster.set" file
- Hide "PatchMaster.set" file, restart "Defaults" & Hardware selection
- Customize colors & fonts of windows

## **Establish Whole-Cell configuration (model cell)**

- Use manual control
- Use Auto buttons (modifiable with Protocol Editor)

## **Record Data & online Analysis**

- **o** Create stimulus sequence using Pulse Generator
- $\circ~$  Start experiment by using Control window
- Adjust Oscilloscope display scaling
- Enable Analysis & display in window

## **Export Data traces**

- Use copy & paste
- $\circ$  Other export options

