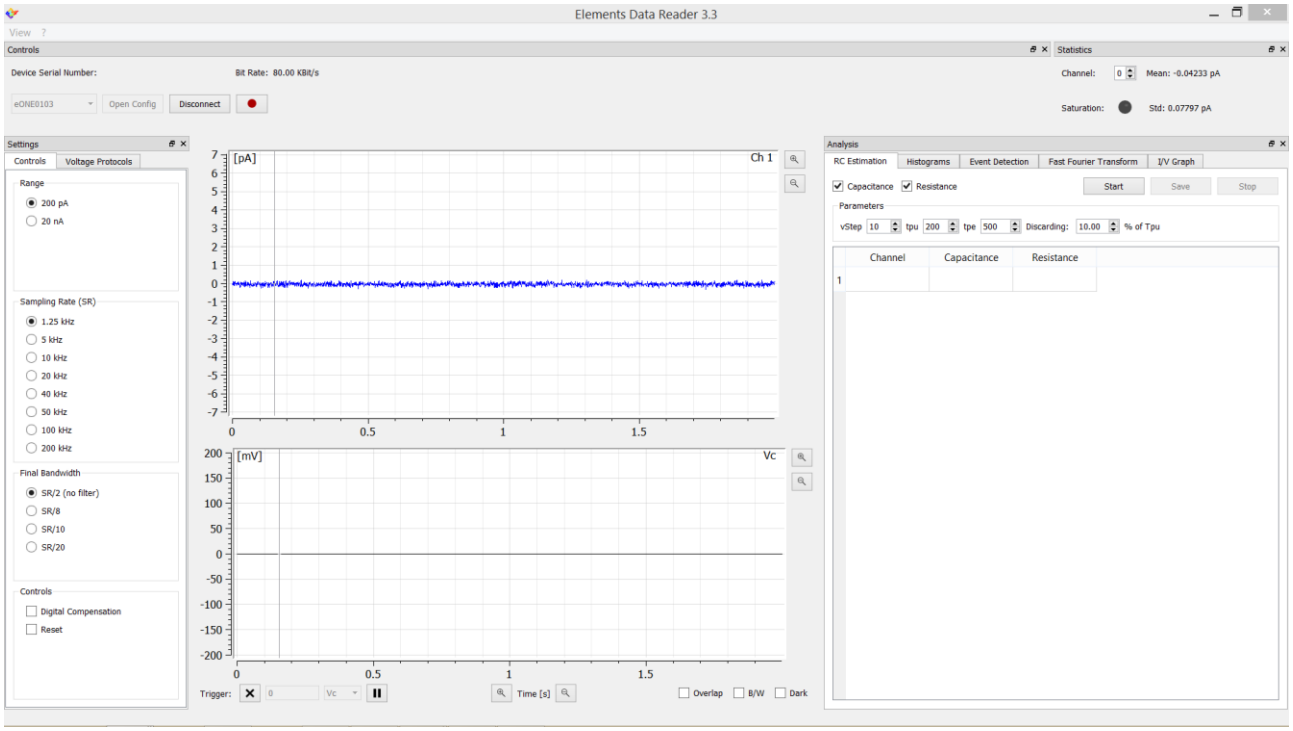
EDR 3

Elements Data Reader 3.X.X

USER'S GUIDE



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elements

enabling technologies for Life Science

Periodically check the following link for the most updated manual version: http://elements-ic.com/downloads/

Revision History

Version Preliminary	Initial Manual Created	30th October 2015
Version 1.0	Manual Released	20th June 2016
Version 1.1	Edited page headings and summary	7th October 2016

The EDR 3 software has been developed and optimized to control all the Elements devices. It was designed in a modular fashion with some features enabled or disabled depending on the particular Elements device connected. In particular devices can be divided in two families:

- **Single channel devices, such as eONE eONE-VP and eONE-HS;**
- **Multichannel devices, such as eFOUR and e16, respectively embedded in the Nanion's Orbit Mini and Orbit 16 devices.**

Summary

1.1 Software Operating Procedure

- When started, the Elements Data Reader (EDR) software appears as in the following picture:

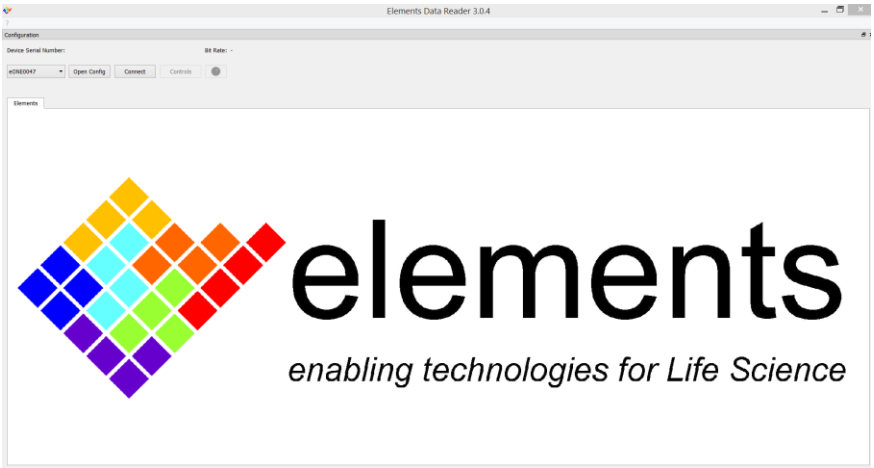


Figure 1
Elements Data Reader Main Menu Window

Note:
Multiple instances of the software can run on the same PC enabling to control several Elements devices connected to the same PC. If more than one Elements device is connected, you must first choose the correct device Serial Number to be controlled.

- In the top-left corner there is a drop-down list of the serial numbers of all the recognized Elements devices connected to the PC.
- The first operation is to select the proper Elements device.
- To start acquiring data, click on the "**Connect**" button.

Note:
*The configuration file (which contains all the necessary setups to communicate with the Elements hardware) is automatically detected from the EDR 3 installation folder (usually C:\Program Files (x86)\EDR 3\configs) or can be loaded by clicking on the **"Open Config"** button.*

1.2 Graphical User Interface

After connecting the device and loading the config file, the GUI appears as follows:

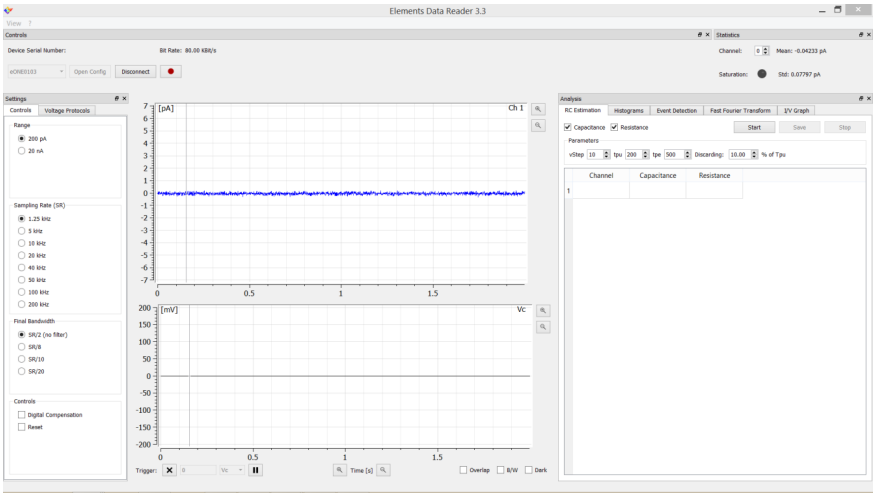


Figure 2
Elements Data Reader Complete GUI

Note:
If a menu has been closed by mistake, right click on one of the menu bar to visualize it again.

The default GUI is organized in multiple menus all embedded in a single window; however single menus can be removed from the main window and split in different windows to be placed where preferred in the screen.

To split the selected menu click on the window restore icon of the menu or double click on the menu bar.
Right click of the mouse on a menu bar opens a menu which enable the selection of the various menus to be visualized.

In particular four menus are present in the GUI:

- Controls Menu;
- Statistics Menu;
- Settings Menu;
- Analysis

1.2.1 Controls Menu

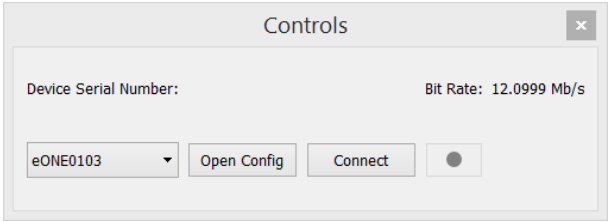


Figure 3
Configuration Menu

The Controls menu has the following buttons:

- **Open Config**– Enables the user to select the proper configuration file for the selected device, as already described in the note of previous section.
- **Connect** – Starts the communication with the eONE hardware.
- ● **Recording** – Saves data in the format and path selectable in the EDR Save Preferences menu that is opened when the button is clicked.

The window contains also an indication of the estimated input bitrate.

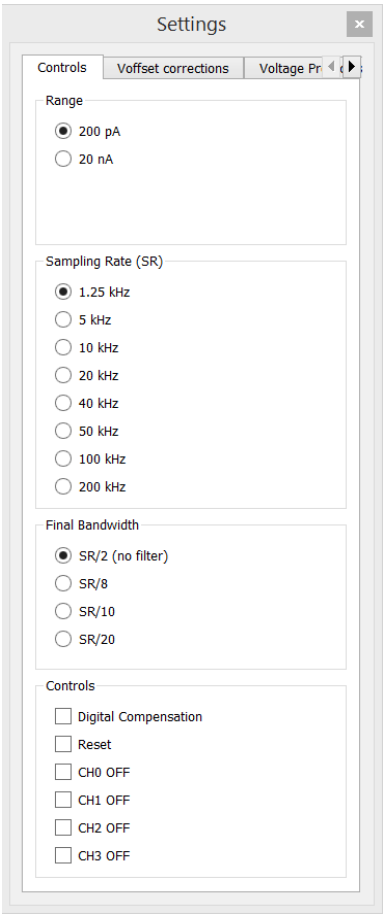
1.2.2 Statistics Menu

Figure 4
Statistics Menu

The Statistics menu indicates some estimated statistic values on the selected signal channel (in case of multichannel device), calculated in real-time during the current acquisitions.
These estimated statistic data are the **Mean** and **Standard Deviation** (**Std)** values. The input acquired data are split in temporal windows whose length is dependent on the input data bitrate and memorized in a buffer. The mean and std values are calculated on these buffers. When the user changes the bandwidth, it can be necessary to wait some seconds to have valid estimated statistic data.
The menu has also a **Saturation** indicator that switches on when the input current exceeded the maximum or minimum current value for the selected Range.

1.2.3 Settings Menu

Figure 5
Settings Menu



The Settings menu contains controls in a modular fashion with some features enabled or disabled depending on the particular configuration file and Elements device connected. Below is described the Orbit mini menu which includes all the possible features.

1.2.3.1 Controls tab

The Controls tab contains the following controls:

- Range
- Sampling Rate (SR)
- Final Bandwidth
- Digital Compensation
- Reset

The **Range** buttons enable the users to choose between two ranges:

- from -200 pA to +200 pA
  or
- from -20 nA to + 20 nA

The **Sampling Rate (SR)** buttons enable the users to choose between the different sampling rate values up to 200 kHz (for the HS version), respectively:

- 1.25 kHz
- 5 kHz
- 10 kHz
- 20kHz
- 40kHz
- 50kHz
- 100kHz
- 200kHz

Note:
Users can achieve a large variety of effective signal bandwidth using different combinations of the SR and Final Bandwidth controls. For example, selecting a sampling rate of 20 kHz and a Final Bandwidth of SR/20 (or a 10 kHz and a Final Bandwidth of SR/10), a 1 kHz bandwidth can be obtained.

The **Final Bandwidth** buttons enable the users to select the final signal bandwidth between four different digital filters settings:

- Sampling Rate/2, the Nyquist frequency
- Sampling Rate/8
- Sampling Rate/10
- Sampling Rate/20

The **Digital compensation** checkbox enables or disables the offset compensation function (see the "Digital Offset Compensation" section for details).
The **Reset** checkbox keep the device in reset mode. This function can be useful to reset the digital offset compensation value to the initial value.
The **CHx OFF** checkboxes switch off the selected channel, the channel Vc is set to 0 mV and the channel input is disabled form any kind of voltage stimulus.

1.2.3.2 Voffset corrections tab (for multichannel devices only)

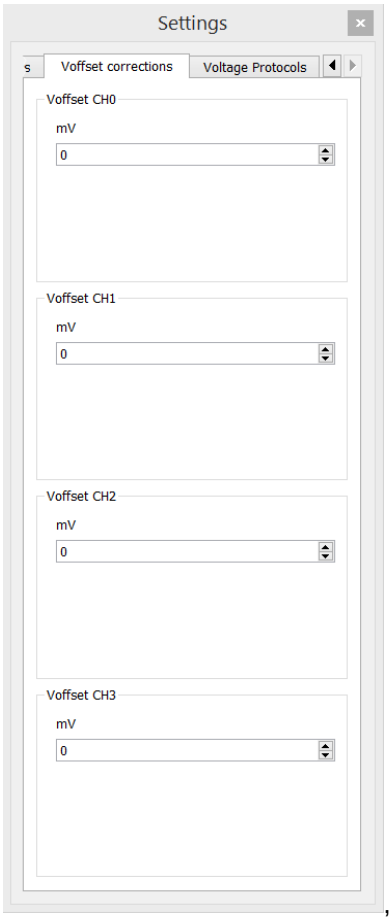


Figure 6
Voffset corrections tab

The Voffset corrections tab contains the controls to individually and manually adjust the voltage offsets of the different input channels to reduce as much as possible the small offset differences between the channels.
This manual offset correction can be done in the range from -10 mV to 10 mV for each channel and the selected value is added to the value applied by the automatic digital offset compensation function described in section 1.3.

1.2.3.3 Voltage Protocols tab

The Voltage Protocols tab (only enabled for eONE-VP, eONE-HS and multichannel devices) contains a complete set of seven standard voltage stimulus protocols. For each automatic voltage protocol the different parameters represented in figure can be set.

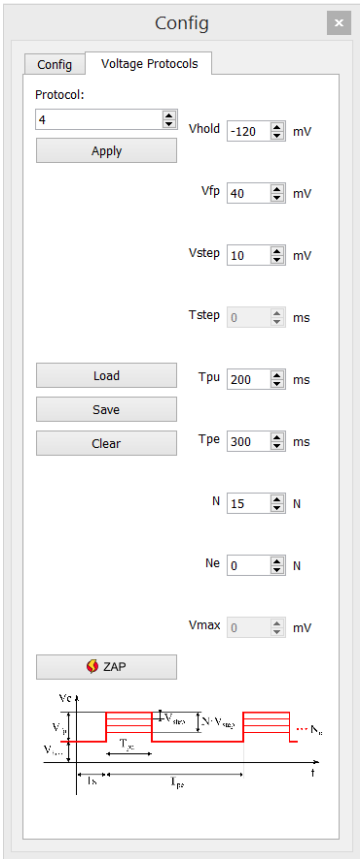


Figure 7
Voltage Protocols tab

The tab also contains the following buttons:

- The **Apply** button: starts the application of the selected stimulus protocol using the pre-set parameters.
- The **Load** button: enables to load the protocols (and the parameters related to that) saved in the selectable .trial files
- The **Save** button: enables to save all the parameters related to standardly used protocols in a proprietary .trial file.
- The **Clear** button: cancels all the parameters and set them to the default values.
- The **ZAP** button: applies a ZAP pulse of 1,65 V for 100 ms simultaneously to all the channels (only enabled in multichannel devices).

Here are listed all the setting parameters; some of them can be enabled or disabled depending on the particular protocol. The described parameters have similar meaning in all the protocols:

- **Vhold** is the constant holding potential applied at the Elements device input pin in the rangefrom -384 mV to +384 mV with 1mV steps;
- **Vfp** is the first pulse amplitude. It can be set in the range from -315 mV to +315 mV with 5mV steps;

Note:
A popup window with the enlarged figure of protocol and parameters opens by going with the mouse on protocol representation.

Note:
To quickly apply your custom and standardly used protocol save it as .trial file, load it and click the "Apply" button. See also section 1.6 for the keyboard shortcuts.

Note:
In order to be sure of the correctness of the applied holding potential, a digital offset compensation procedure must be followed. The digital compensation function is described in section 1.3.

Note:
The software also performs a rules check for the correctness of the set parameters. A warning message compares, if set parameters are invalid.

- **Vstep** is the voltage step between two consecutive pulses. It can be positive or negative to obtain increasing or decreasing consecutive pulses;
- **Tstep** is the time step in terms of variation of between two consecutive pulses.
- **Tpu** is the single pulse duration. It can be set in the range from 1 ms to 9000 ms;
- **Tpe** is the time period between two consecutive pulses. It can be set in the range 10 ms to 90000 ms;
- **N** is the total number of pulses; each one differs from the previous one by Vstep mV;
- **Ne** is the total number of identical sweeps. It can be from 1 to 127 and by setting Ne to 0 the endless repetition of the selected protocol can be obtained.
- **Vmax** is the maximum voltage variation from the Vhold potential. It can be set in the range from -315 mV to +315 mV with 5mV steps.

Below the different voltage protocols and their parameters are briefly described:

- **Constant voltage** protocol:

The constant voltage is considered as a protocol with only the **Vhold** parameter.

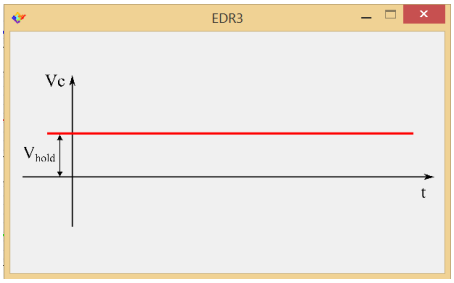
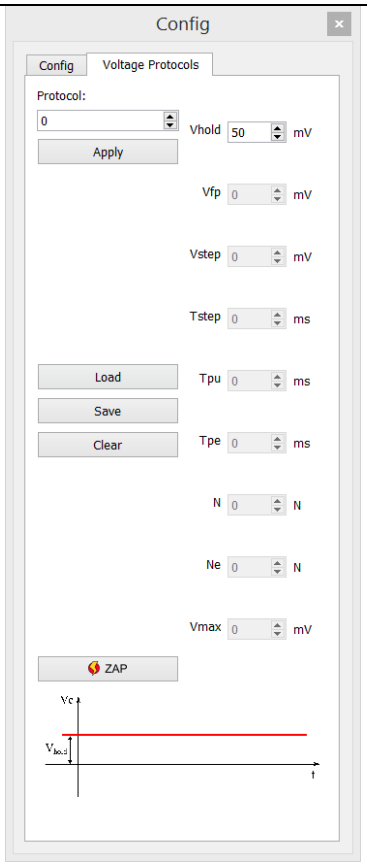


Figure 8
Voltage Protocols
Constant Voltage protocol



- **Triangular wave** protocol:

It can be used to control the bilayer formation and it is used for the Capacitance estimation feature (see details section1.2.4).

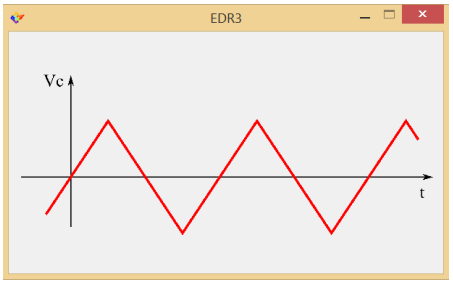
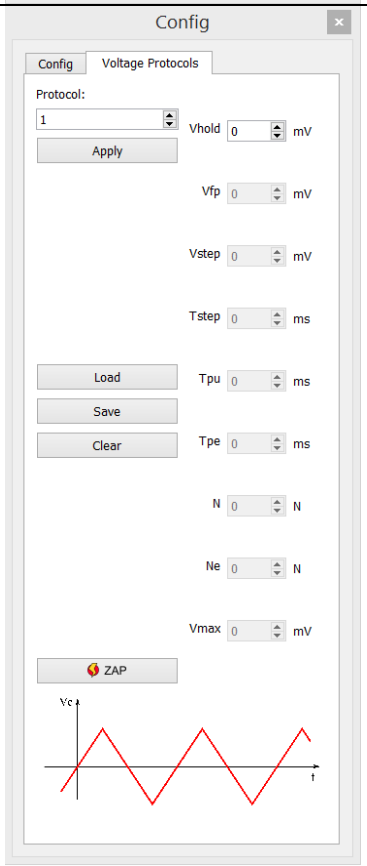


Figure 9
Voltage Protocols
Rectangular Voltage Pulses protocol



- **Seal test** protocol:

It is commonly used for setting up the sealing of the pipette to the cell in patch clamp studies. Usually the **Ne** parameter is set to 0 to repeatedly apply the stimulus to the cell.

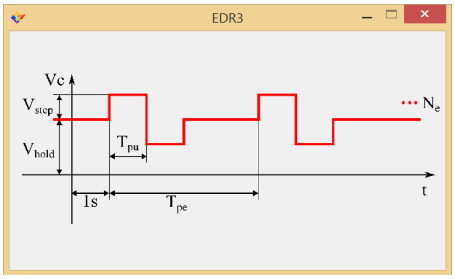
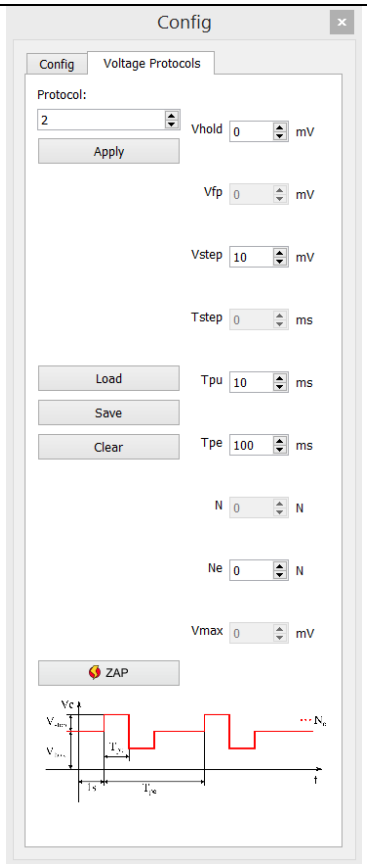


Figure 10
Voltage Protocols
Seal Test protocol



Note:
Users can activate the trigger (as explained in section 1.5) in order to trigger signal using the input current or voltage pulses.

- **Conductance test** protocol:

It can be used to semi-automatically perform a channel's conductance characterization.

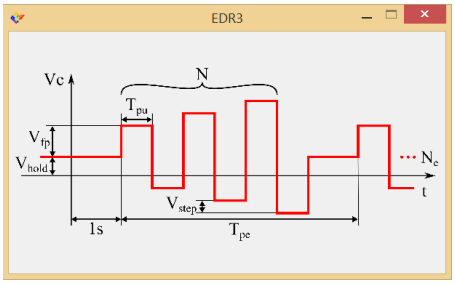
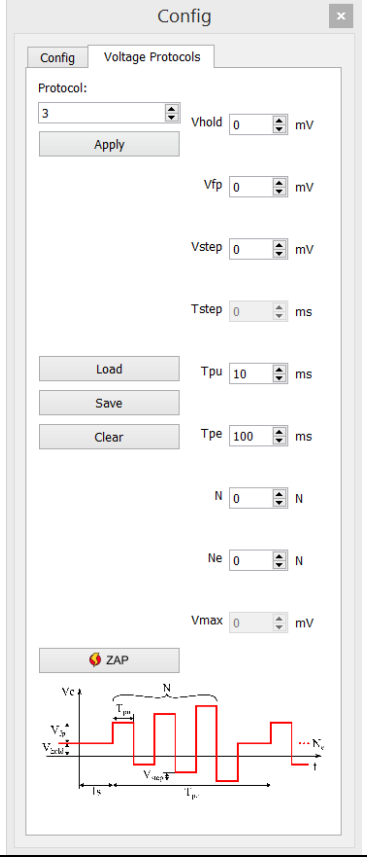


Figure 11
Voltage Protocols
Conductance test protocol



- **Rectangular voltage pulses** protocol:

It is the most common protocol to perform an analysis of channel's voltage sensitivity.

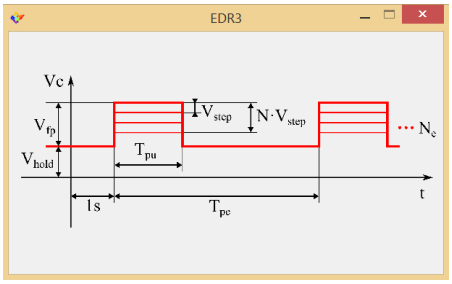
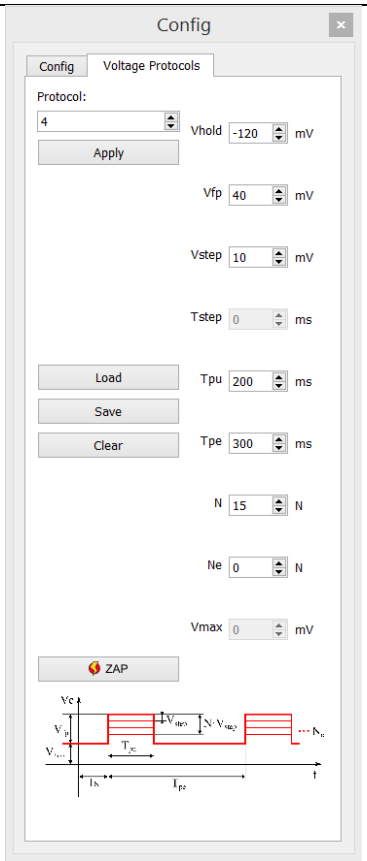


Figure 12
Voltage Protocols
Rectangular voltage pulses protocol



- **Recovery from inactivation** protocol:

It consist of a series of identical rectangular voltage pulses **Tpu**ms long with an amplitude of **Vfp** mV. The time interval between two consecutive pulses (interpulse interval) changes of the value of the **Tstep** parameter.It can be positive or negative to obtain increasing or decreasing interpulse intervals.

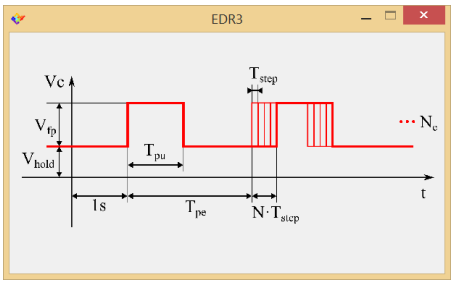
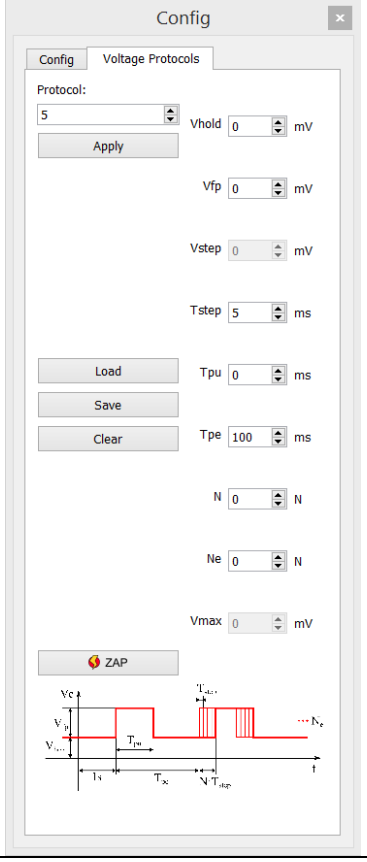


Figure 13
Voltage Protocols
Recovery from inactivation protocol



- **Variable duration pulses** protocol:

It consist of a series of rectangular voltage pulses (**Vfp** mV of fixed amplitude) of a duration that changes of the value of the **Tstep** parameter.

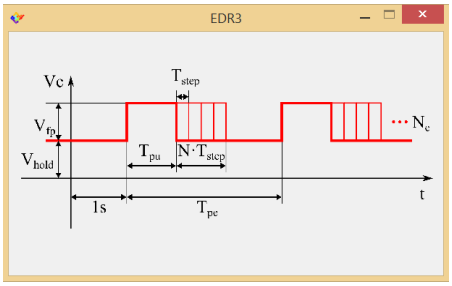
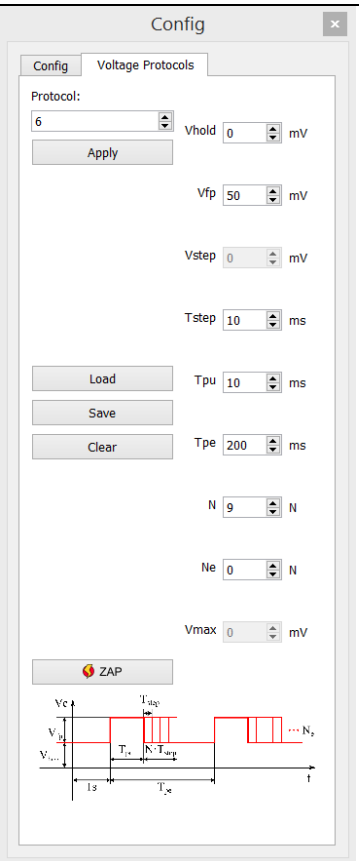


Figure 14
Voltage Protocols
Variable duration pulses protocol



- **Voltage ramp** protocol:

It is the most common protocol to generate the I-V curve for the current-voltage characterization.
The setting parameters are similar to the previously described rectangular voltage pulses protocol, with the difference that, instead of the number of pulses, users can select the maximum voltage variation from the Vhold potential, **Vmax**.

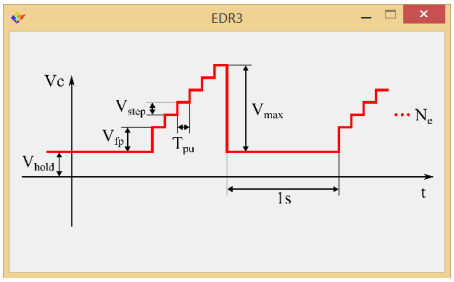
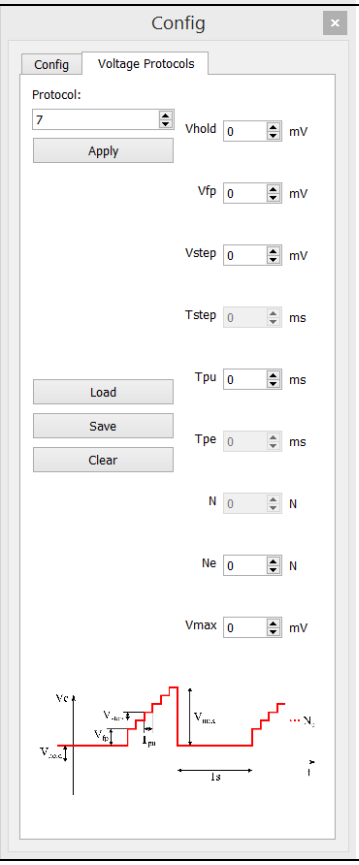


Figure 15
Voltage Protocols
Voltage ramp protocol



Note:
*To obtain the most achievable linear ramp, users have to set **Vstep** and **Tpu** parameters to 1 mV.*

1.2.4 Analysis Menu

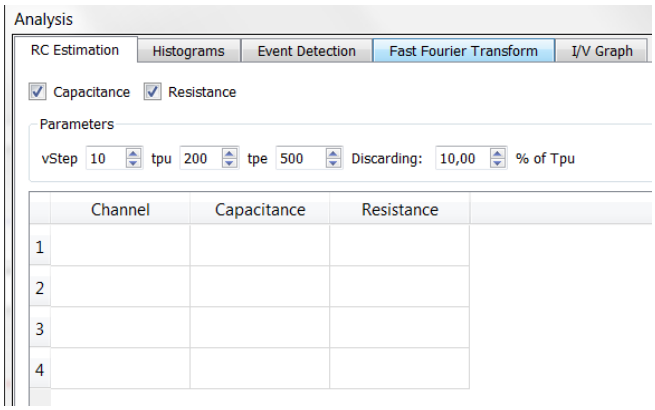


Figure 16
Analysis Menu – Base Version

The analysis menu contains the real-time statistical analysis. The EDR 3 base version allows only the Resistance & Capacitance Estimation.
Some other real-time statistics are under licence and are described in the 1.4 "Real- time statistical Analysis" section.
To obtain capacitance and resistance estimations click on the "Start" button. The EDR software automatically and alternatively calculates the equivalent input capacitance and resistance by applying the Triangular Wave (Protocol 1) and Seal test pulse (Protocol 2). Values of estimated R and/or C are continuously visualized for every channel.
The "Start and record" button starts the estimation and, at the same time, starts saving the raw data from all the input channels in the format and path selectable in the EDR Save Preferences menu that is opened when the button is clicked. It is the same as clicking on the ● **Recording** button.
To have a more accurate estimation of the Resistance value it is possible to select a percentage of Tpu parameter (see the voltage protocol tab description)of the current signal to be discarded after the voltage protocol application, in order to neglect the capacitive effects.

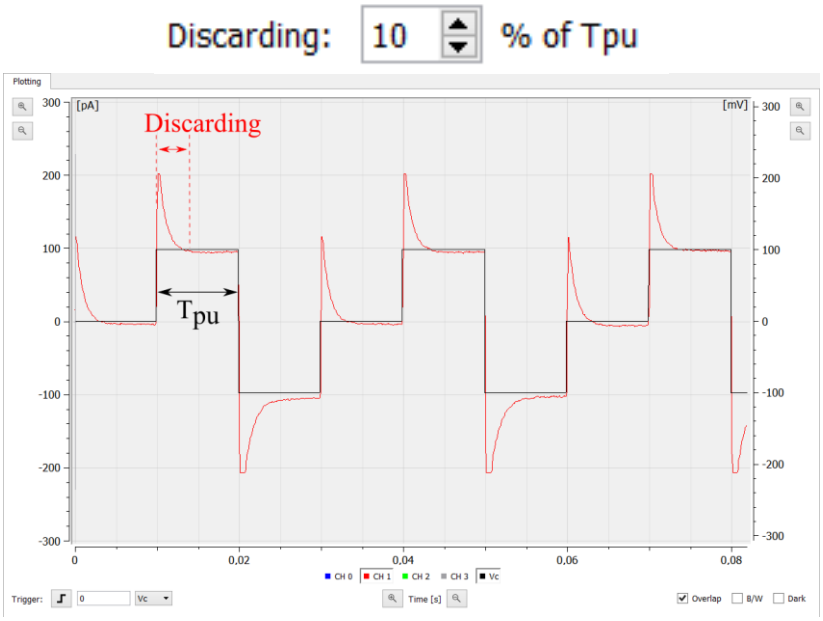


Figure 17
Discarding option for better R estimation

Note:
For capacitance values higher than about 17pF select the 20nA range in order to avoid amplifier saturation.

1.3 Digital Offset Compensation

To compensate the voltage offset due to internal opamp offsets or unwanted electrode potentials, Elements devices have an internal calibration procedure that can be activated with the **Digital Compensation** checkbox.

The compensation circuit, when activated, sets the Vc to 0 V and adds to it a variable voltage in the range from -128 mV to +128 mV until the current is 0 A (1 mV step resolution).

Note:
To successfully compensate the offset, a conductive path between the two electrodes must be present.

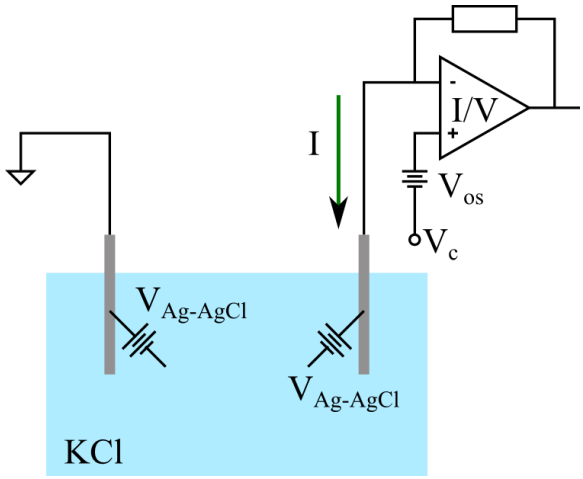


Figure 18
Digital Offset Compensation

In order to successfully compensate the offset, a conductive path must be present on the input.

Do NOT compensate the voltage offset with open input.

The compensation need only few seconds. To return to normal operation, UNCHECK the **Digital Compensation** checkbox.

The compensation value remains stored internally and added to the Vc.

1.4 Real-time statistical Analysis

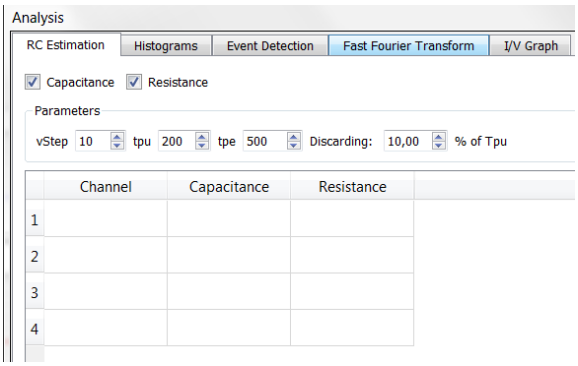


Figure 19
Complete Statistic Analysis Menu

The real-time statistical analysis module enables the users to perform five common statistical analysis, usually done by scientists during the post processing analysis of data.
Each enabled real time statistical analysis has a dedicated tab in the Analysis menu:

- Equivalent input Resistance & Capacitance Estimation
- Histograms
- Event Detection
- Fast Fourier Transform
- I/V Graph

The Input Resistance & Capacitance Estimation has already been described in section 1.2.4; below are described the other statistical analysis enabled under licence.

- The **Histograms tab:**

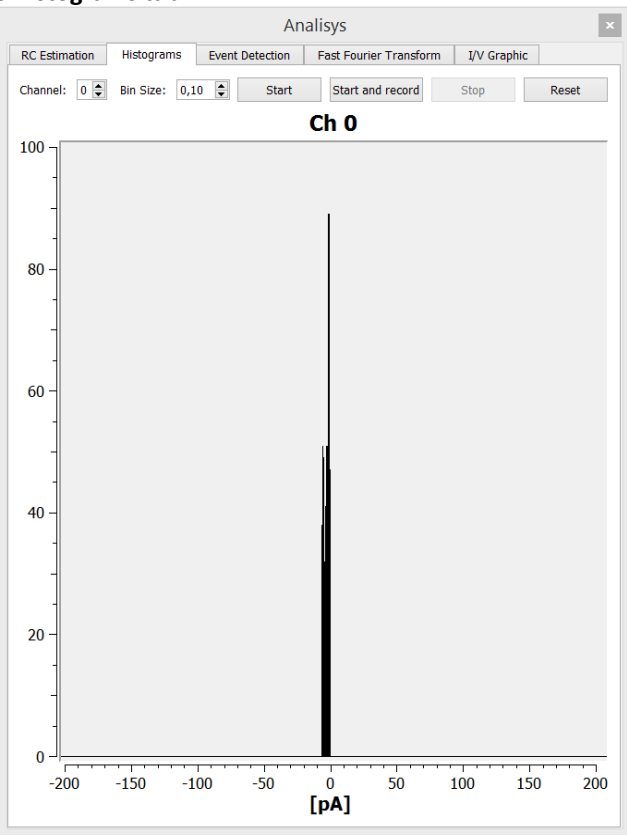


Figure 20
Histograms tab

The first step is to select the Bin size, which is the size (in pA) of the interval by which the entire range is divided and that defines the resolution of the histogram.
The "Start"(or "Start and record")button starts the construction of the histogram using the selected "Bin size" (pA) for all the channels.
Channel value can be modified in real time to visualize the selected input channel (for multichannel devices)
Using the Reset button the count is reset to zero and graph is rebuilt starting from that instant.

- The **Event Detection tab:**

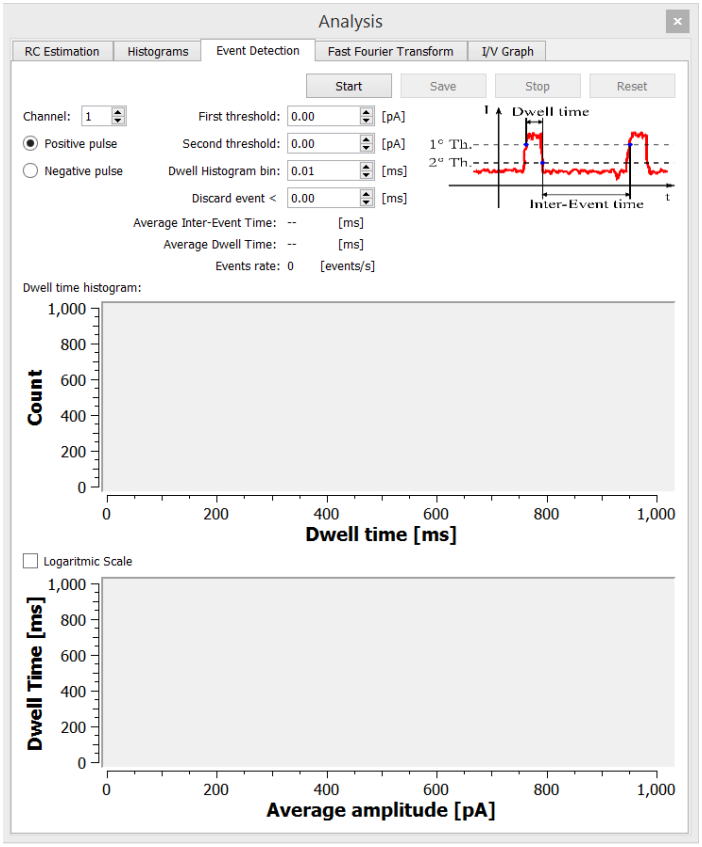


Figure 21
Event Detection tab

This analysis allows to know in real time the dwell and inter-event time related to events of interest by setting two thresholds.
The first step is to select between positive or negative pulse events and setting the two thresholds; then clicking on "Start" (or "Start and record")button, the software starts to detect events crossing the first threshold and lasting up to the second threshold, related to the selectable input channel (for multichannel devices).
Below are two figures that explain the Dwell and Inter-Event time values meanings in case of positive or negative pulse:

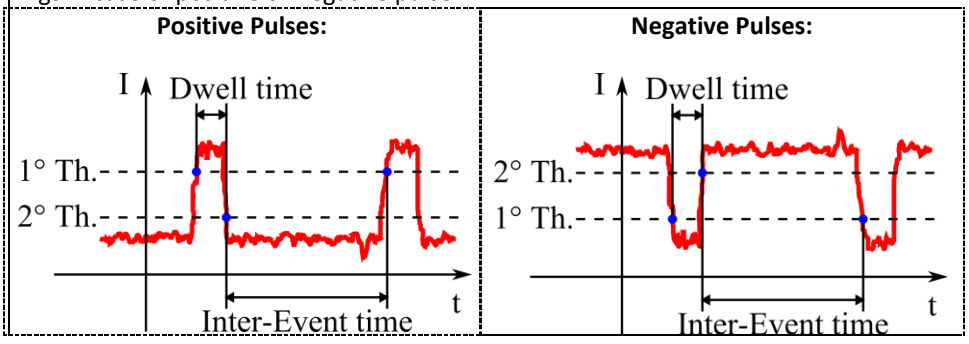


Figure 22
Inter-Event and Dwell time meanings for Positive and Negative pulses

- The **Fast Fourier Transform (FFT) tab:**

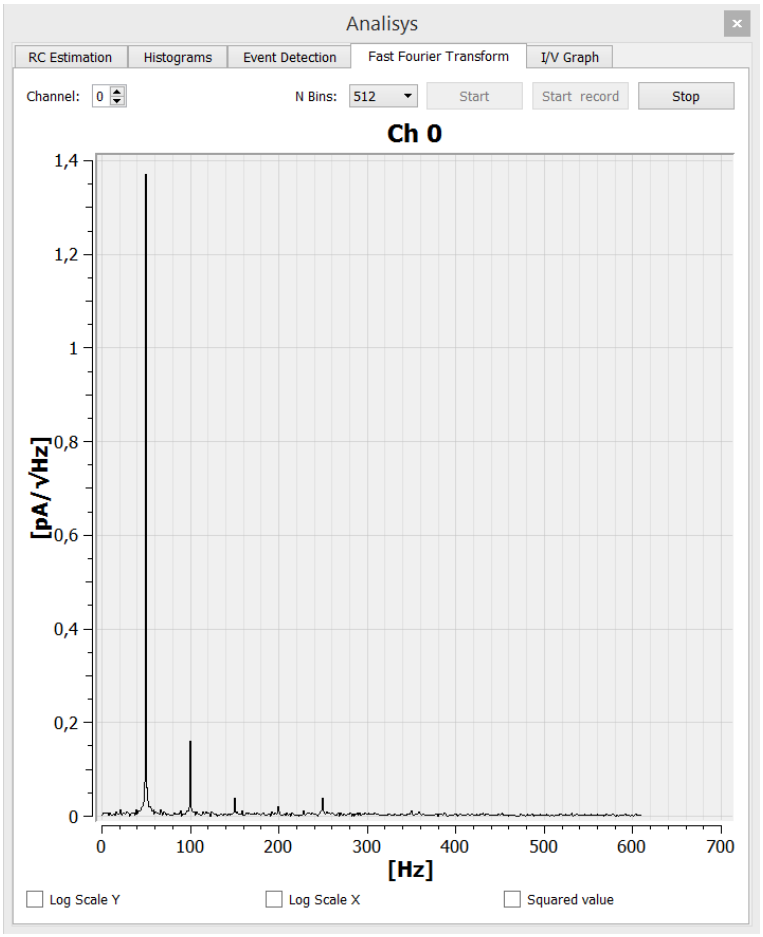


Figure 23
Fast Fourier Transform tab

This feature enables users to perform the Fast Fourier Transform of the selectable current signal channel (for multichannel devices) in real time and allows to detect events in the frequency domain.
The FFT size is defined by the N Bins parameter which is the number of Bins used for dividing the window into equal strips. This parameter defines the frequency resolution of the window.
The FFT is calculated after clicking on the "Start" (or "Start and record") button and data can be plotted in linear or logarithmic scale by checking the "Log Scale X" or "Log Scale Y" checkboxes.
Power Spectral Density instead of Amplitude spectral density can also be obtained by checking the "Squared value" checkbox.

- The **I/V Graph tab:**

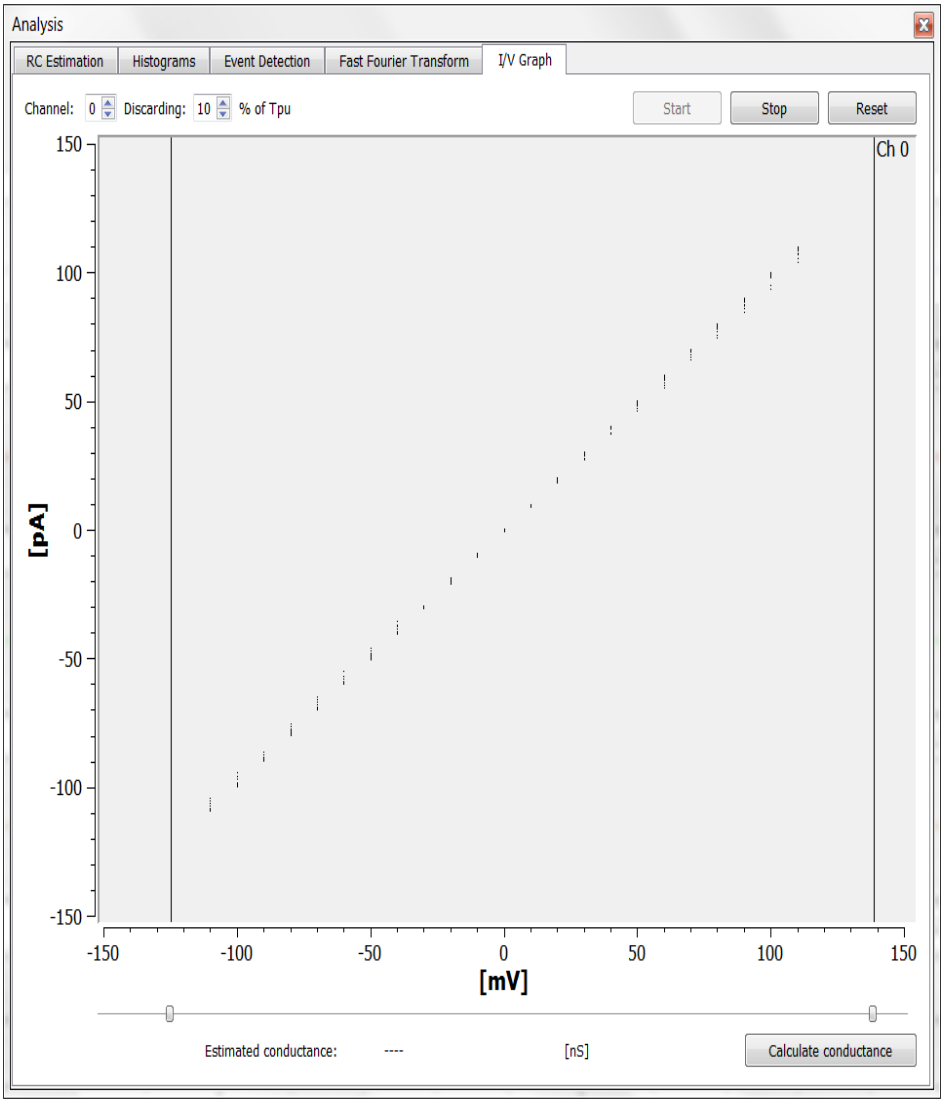


Figure 24
I/V Graph tab

This analysis is useful to visualize in real time the I/V curve of the device under test (DUT) or ion channel under study. For instance and to have a quick information of the rectification behaviour or the linearity, or not, of the response.

The first step is to select the **Conductance test** protocol (Protocol 3) or the **Voltage Ramp** protocol (Protocol 7), set the protocol parameters (see Voltage Protocols tab description for details) and apply the stimulus.

Then select the input channel to be analysed and the percentage of Tpu parameter (see the Voltage Protocol tab description) of the current signal to be discarded after the voltage protocol application, in order to neglect the capacitive effects. Clicking on the "Start" (or "Start and record") button, the I/V Graph starts to be constructed and plotted.

1.5 Trace Visualization

The central window of the Graphical User Interface displays the acquired input current channel (or channels one below the others, for multichannel devices) and the control voltage in a oscilloscope like approach.

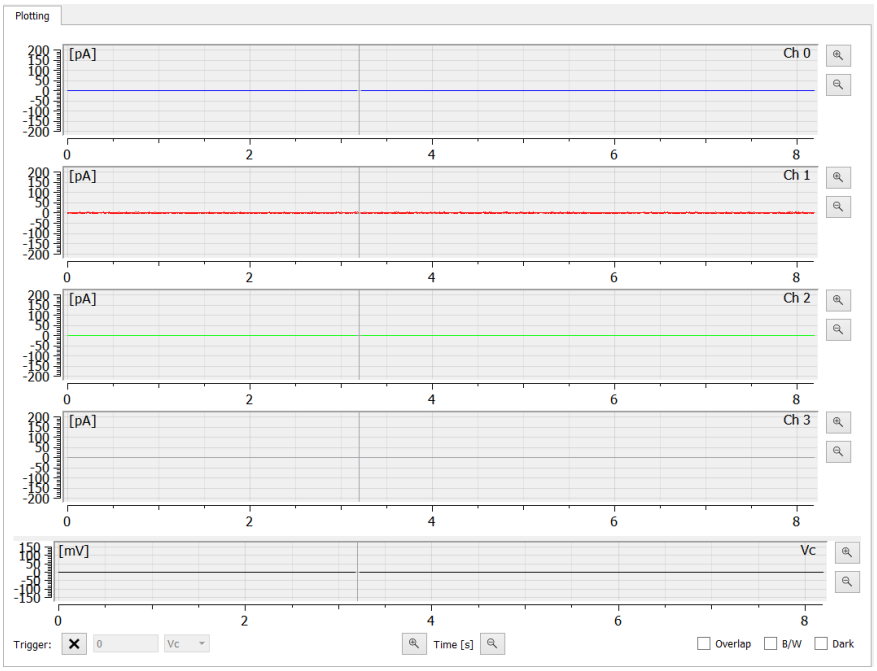
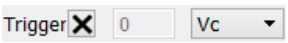
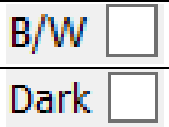


Figure 25
Data plotting window

In the data plotting window there are the buttons necessary to control the visualization.
In particular these are the button functions:

	Increases/decreases vertical zoom	*NB: click and drag in the window to zoom only the interested part of the display*
	Increases/decreaseshorizontal zoom	
	Activates the trigger function using the selected value as threshold	*NB: it is possible to select the positive or negative edge of the Vc or of the input current signals*
	Displays all the data signals (input currents and Vc) overlapped in the main window	
	Sets data traces to black and white	
	Changes from white to black the windows background	

Added to these controls, the vertical offset visualization (shift) of each individual channel can be changed selecting a data channel by clicking on it and by scrolling up or down the mouse wheel

1.6 Keyboard shortcuts

The EDR software has keyboard shortcuts to easily control the amplifier while experiments are running. Here is the list of the shortcuts that can also be found in the "?" menu:

"+"/"-"	Increase/Decrease current scale
Ctrl +"+"/"-"	Increase/Decrease time scale
Ctrl + Page Up/Down	Increase/Decrease current offset
Shift + "+"/"-"	Increase/Decrease voltage scale
Shift + Page Up/Down	Increase/Decrease voltage offset
Ctrl + R	Reset scales
Ctrl + Arrow Up/Down	Increase/Decrease Vhold by 10 mV
Ctrl + Arrow Left/Right	Increase/Decrease input current range
Ctrl + 0	Start voltage protocol 0
…	...
Ctrl + 9	Start voltage protocol 9
Ctrl + G	Start/Stop recording
Alt + Z	Activate ZAP function
Alt + S	Save voltage protocol
Alt + L	Load voltage protocol

1.7 Save & Read Data

Note:
Be careful to have enough disk space to save data and avoid to save data directly into network drive or Cloud storage service. Consider that, at the maximum sampling rate of 200KHz, to save data using.abf format requires about 50MB for every minute of recording; using .DAT format about 100MB/min and using .CSV format about 150MB/min.

The EDR software can save data in three file formats:

- Proprietary .DAT files - It's a binary format with data represented in single precision values.
- Comma separated values .csv files - It's a simple text format with data represented as text. For instance .csv files can be imported by Origin® software from OriginLab®.
- Axon™ binary format v2 .abf files – It's the Axon™ file format used by commercial pClamp® software from Molecular Devices®; data are saved in "gap-free" acquisition mode.

To save data, click on the "● **Recording**" button. Data will be saved with the filename and path selected in the "EDR Save Preferences" menu that is opened when the button is clicked.
Default path is " C:\Users\Username\EDR". Default filename is "Data_X", where X is an incremental number, automatically assigned in order to have subsequent filenames.
When saving begins, a timer in the GUI starts and indicates the total saving time.

Note:
Acquisition Range, Sampling Rate and Bandwidth cannot be changed while saving data.

To stop saving, click on the "■ **Stop recoding**" button.

The EDR software creates a new folder in the location chosen by the user with the same name of the saving files. In this new folder the EDR software creates some files:

- An header .txt file containing some eONE setup information (Range, Bandwidth, etc.).
- The saved data files. To limit the maximum file dimensions, the EDR software splits the files every X minutes of acquisition selectable in the "EDR Save Preferences" menu. Default value is 5 minutes of acquisition.

The proprietary .dat files can be handily read by EDA® (Elements Data Analysis, coming soon) or by Matlab® with the function **elementsRead.m** file. To get the function please contact Elements sending an e-mail at support@elements-ic.com

Note:
Be careful to disable sleep or hibernation state in the power plan settings of your computer when performing long-time experiments.

To import data into **Origin®**:

- Save data in **.csv** format selecting it in the "File→Preferences" menu of EDR software;
- Open Origin® and click on "Import→ Comma delimited (CSV)" and then set "Comma" as delimiter in the "Import Option" menu;
- The first imported column is the current signal (in pA or nA) while the second column is the control voltage (in mV)

To read data using **Clampfit®** software:

- Save data in **.abf** format selecting it in the "File→Preferences" menu of EDR software;
- Open Clampfit® and click on "Open Data" (or Ctrl+O shortcut) and select the saved Axon Binary File to read the two acquired data channels: current signal and control voltage;

Technical support & repairs

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