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ePatch

Setup and functional checkout

Revision History

Date	Version	Description
02/02/2024	1.0	First version of the document

ePatch technical specifications

Voltage Clamp
- Open input (RMS) noise: 300 fA rms @ 625 Hz – 1.5 pA rms @ 10 kHz – 12 pA rms @ 100 kHz
- Current ranges: ±200 pA (Gain 2.25 GΩ), ±2 nA (Gain 225 MΩ), ±20 nA (Gain 22.5 MΩ), ±200 nA (Gain 2.25 MΩ)
- Max applicable voltage: ±500 mV
- C fast compensation range: 0-11 pF
- C slow compensation range: C in 0-250 pF – Tau in 0-2500 µs
- R series compensation range: 0-25 MΩ

Current Clamp
- Short circuit input (RMS) noise: 11 µV rms @ 625 Hz – 22 µV rms @ 10 kHz – 350 µV rms @ 100 kHz
- Parametric current protocols for pulse generator in the range of ±2.5 nA (res. 0.2 pA) and ±100 nA (res. 10 pA)
- Voltage range: ±700 mV
- Pipette neutralization range: 0-31 pF
- Bridge Balance compensation range: 0-40 MΩ
- True-zero current mode

Other technical specifications
- Available bandwidth between 625 Hz to 100 kHz
- Max sampling rate: 200 ksps
- Data output format .abf
- 0-5V programmable digital output
- Size & Weight: 42 x 18 x 78 mm, 200 g

ePatch package content

Before starting the setup, check the following components have been included in the package:
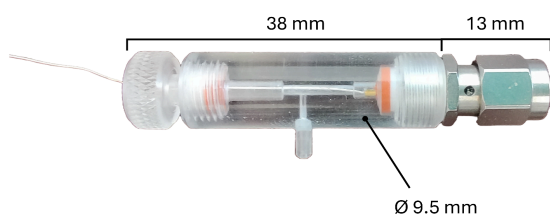
- ePatch amplifier

- USB cable

- Ground (GND) cable to connect to Faraday cage

- Reference electrode wire (1mm gold pin)

- SMA Pipette holder

38 mm   13 mm

Ø 9.5 mm

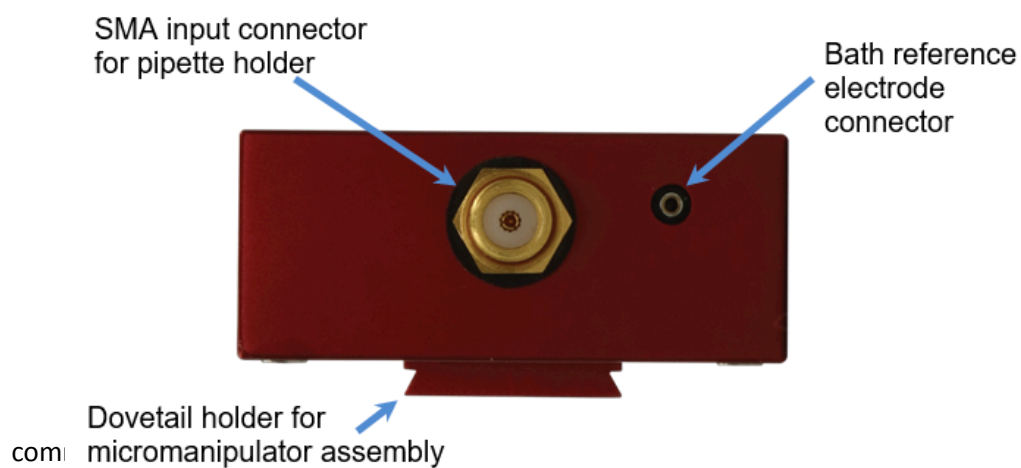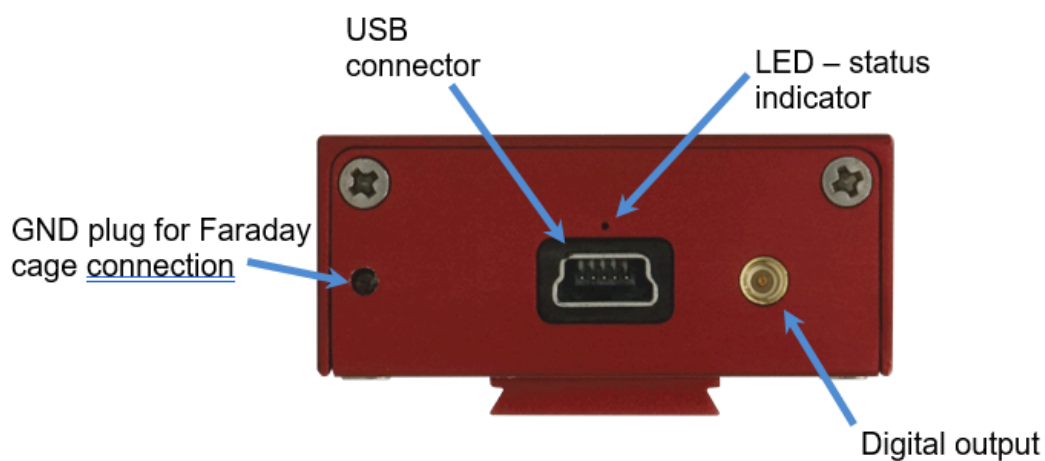- Digital-out port to BNC cable

- Model cell

- Cable to connect the model cell to the reference connector

ePatch ports description

USB connector

LED – status indicator

GND plug for Faraday cage connection

Digital output

SMA input connector for pipette holder

Bath reference electrode connector

Dovetail holder for micromanipulator assembly

Software installation instructions

Download and install the latest version of EZ Patch software from https://elements-ic.com/downloads/. Connect ePatch to the computer using the USB cable and wait for the driver installation. If successful, the small LED light above the USB port should be green.

Functional checkout

Besides all the units are thoroughly tested at the factory just before shipping, you should run a functional checkout before installing the amplifier within the patch clamp rig. Should you encounter any problem with the functional checkout or if the noise levels are beyond the declared range value (see first page), please write an email to support@elements-ic.com.

Noise test:

Test the open circuit noise by placing ePatch on a clean bench away from other equipment and take a large piece of aluminum foil. The only connections required for this test are: i) the USB cable from the amplifier to the computer: ii) the ground cable from the ground port to the aluminum foil. Wait to connect the USB to the computer and wrap a large piece of aluminum foil loosely and completely around the device. You may leave some room at the front side for the model cell connection in the next step. Clamp the alligator clip to the aluminum foil just before wrapping completely. Open EZpatch software and press connect. From the protocols list found at the right of the screen, select the protocol "holding voltage" and press the green display button above. The noise test widget should pop up after some seconds. From the "controls" widget located at the left of the screen, make sure the 200pA range is selected. Read the rms noise displayed in the noise report widget at 1.25 kHz, 10 kHz and 100 kHz and make sure they are equal or smaller than the values reported in the chart below (select the desired sampling rate from the dedicated option located within the "controls" widget).

The noise in current clamp mode is usually measured in short circuit, as reported in the feature section of the ePatch webpage. Since short circuiting the amplifier out of the production laboratory could seriously damage the device, we discourage performing the same test. Performing the safe noise test in current clamp mode can be done with the model cell, set in bath mode (see next paragraph to learn how to use the model cell). After installing the model cell, select the current clamp modality (the switch to the current clamp mode in the software interface is located at the top of the "controls" widget) and run the "holding current" protocol. Read the rms noise displayed in the noise report widget and compare them with those reported in the table below.

| Sampling rate (bandwidth: SR/2)	Open circuit rms noise in Vclamp	Noise in Cclamp (5 MΩ input)
1.25 kHz	300 fA	15 µV
20 kHz	1.5 pA	37 µV
200 kHz	12 pA	352 µV

Compensation operation check:

The model cell contains resistors and capacitors designed to emulate three patch clamp experimental conditions: the pipette immersed in the bath, the cell attached, and the whole cell configuration. Connect the model cell to the SMA input connector of ePatch (see image below). Use the supplied pin reference cable to connect the model cell to the reference port of the amplifier.
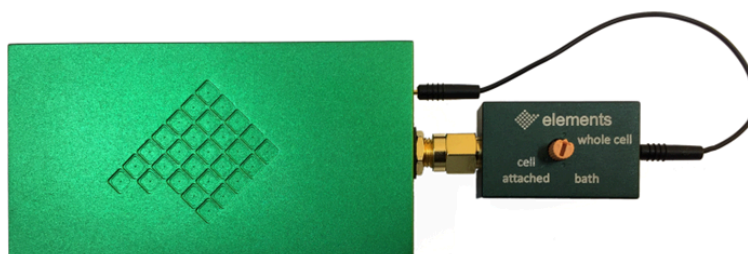

Rotate the selector of the model cell until the small arrow points to the "bath" configuration label.

In "Bath" mode, the model cell electrically mimics the experimental condition in which the pipette is immersed in the bath without touching the cell. In this configuration, offset potentials can be corrected, and the resistance of the pipette can be measured. The electrical circuit of the bath configuration of the model cell is shown in the following figure.
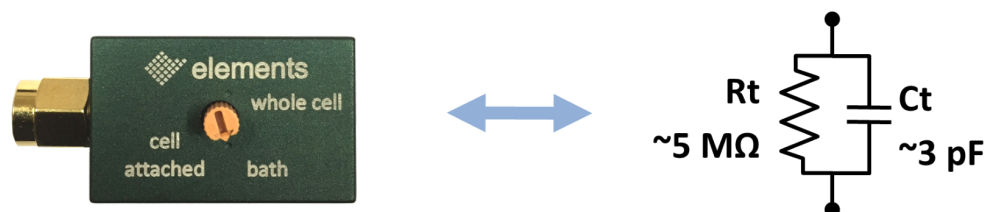
Click the **offset compensation** button in the compensations widget to eliminate any voltage offset between the bath electrode and the patch electrode. After making a brief series of steps, the software automatically finds the value that is needed for the compensation. Check that the current level is close to the zero value.
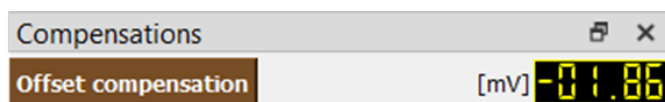

Afterwards, double click on the "seal-test" protocol found within the protocols list to open the protocols parameters. In the "Vamp" box set 20 mV and click "OK". Run the protocol by clicking the green display button (if needed, use the magnification glasses next to the oscilloscope window to adjust the scale of the current and voltage signals).

The **membrane test widget** appears below the protocols list to show the resistance and capacitance values calculated from the current response (the values might differ from the theoretical ones due to the 15% tolerance of the resistors and capacitors of the model cell circuitry).
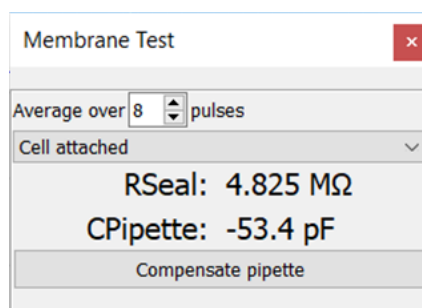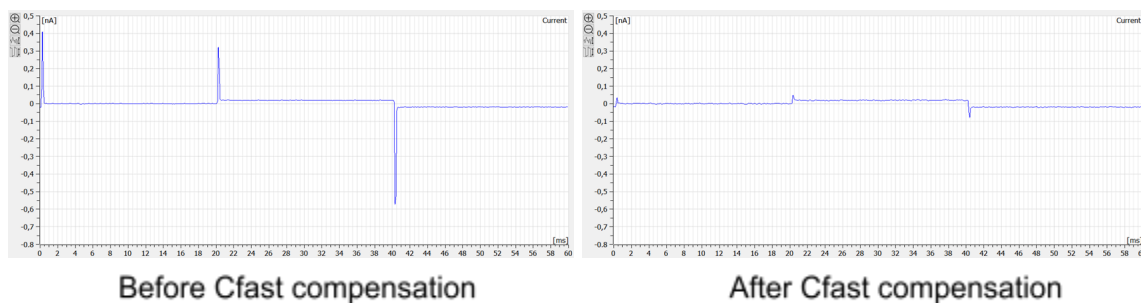

Rotate the selector of the model cell to the cell attached mode. The electrical circuit of the bath configuration is shown in the following figure.
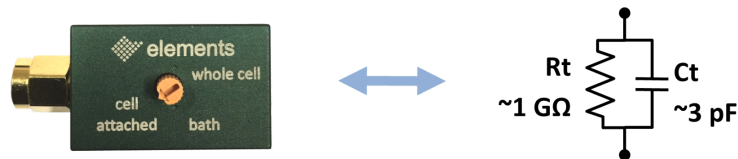
Two **capacitance transients** should arise in synchrony with the rising and falling edges of the voltage stimulus. The membrane test widget should display the calculated R and C of the circuit, namely 1GOhm and 3pF (the values might differ from the theoretical ones due to the 15% tolerance of the resistors and capacitors of the model cell circuitry).

In the "compensation" widget, click on the "pipette" button to activate the **Cfast compensation** and rotate the digital knob to adjust the value and minimize the transients. The knob can be rotated by clicking on the knob and using the keyboard arrows or alternatively using the mouse wheel.


Before Cfast compensation

After Cfast compensation

Rotate the selector of the model cell to whole cell mode. The electrical circuit of the bath configuration is shown in the following figure.
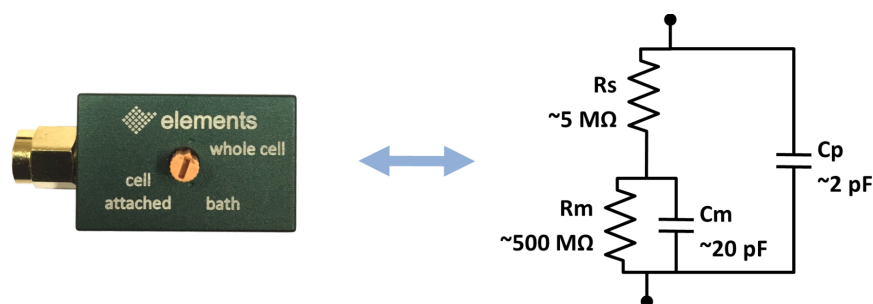

Use the drop-down menu of the "membrane test widget" to display the analysis of the membrane transient (whole-cell option). The real time analysis of the current response defines the values of the membrane capacitance (CMembrane), the membrane resistance (RMembrane) and the series resistance (RAccess).

In the "compensation" widget, click on the "membrane" button to activate the **Cslow compensation** and rotate the membrane capacitance and access resistance knobs to minimize the transients.

Before C slow compensation · After Cslow compensation
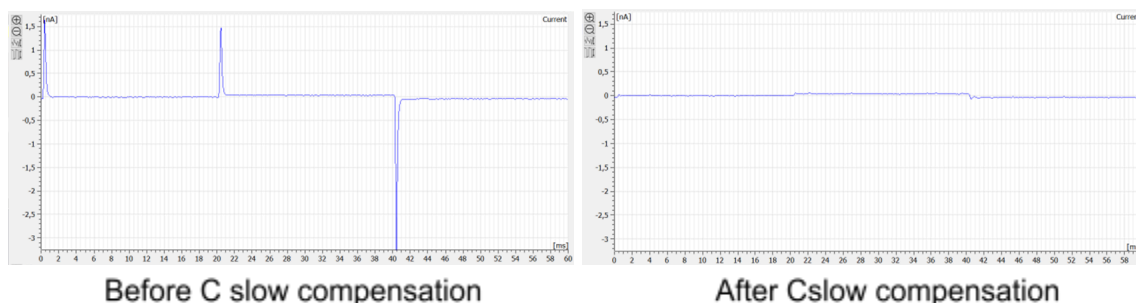
Setup in the patch clamp rig
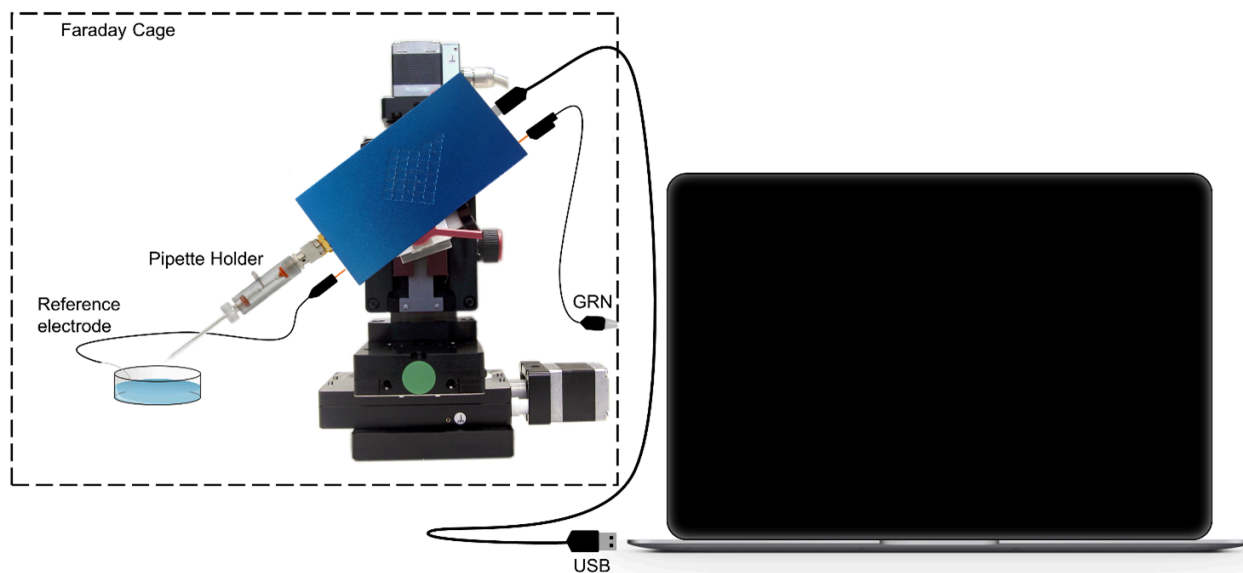
Mount the ePatch on the micromanipulator using either the dovetail or the rod bar mounting system.

Check that the silver wires of the pipette holder and the reference electrode are chlorinated. It is recommended to perform a chlorination every 5 hours of continuous experiments. The simplest method to chlorinate the silver wires is to immerse them into pure bleach until they become light gray coloured (typically 15 to 30 minutes are enough). The commercial bleach purchased in any supermarket is fine for this purpose.

Important notes:

- Reference electrode: make sure not to chlorinate the portion of the wire that fits into the ref electrode wire plug by keeping it outside the bleach. At the end of the treatment, rinse the wire with distilled water and dry.
- Pipette holder: disassemble the pipette holder as shown in the figure below, being careful not to lose the seals. Immerse only the silver wire leaving unchlorinated the section close to the soldering point. At the end of the treatment, rinse the wire with distilled water, dry and re-assemble the pipette holder (do not wash the SMA connector).
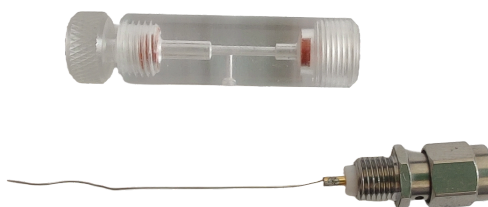
Screw the provided pipette holder to the SMA input connector of ePatch. Do not use hex wrenches to tighten the I-holder to the SMA connector. Hand-tightening is more than enough to ensure proper installation and prevents damages to the SMA connector. You might use a third part pipette holder, provided that is equipped with a standard SMA connector. We are not responsible for any damage caused by using holders other than the one provided.

Insert a Ø 0.5 mm Ag/AgCl wire into the socket of the "reference electrode wire". For a more stable connection, you may solder the silver wire to the socket. Insert the 1mm pin into the bath reference electrode port located on the front side of the amplifier (see fig at page 1).

Connect the GND cable to the port on the back of the amplifier and attach the alligator clip to the Faraday cage. You may test different points of the Faraday cage to find the place that gives the best grounding (i.e. the lowest noise).

Lastly, connect the USB to and open the software to start the recording session.

Warnings:

- Do not short-circuit the pipette electrode or the reference electrode neither with the Faraday cage nor with the GND! The ePatch reference electrode voltage is different to the GND voltage. Short circuits or improper usage/handling could damage the amplifier. While handling the amplifier (e.g. changing the glass electrode for the next patch) we suggest wearing an anti static wristband to prevent that possible ESD to damage the amplifier ASIC.

- Do not connect anything to the SMA input that is not an SMA connector. Inserting a tip (like the 1mm pin of the reference wire) causes an inner break of the SMA input.