doric

Fiberless & Wireless Optogenetically Synchronized Electrophysiology System

User Manual

Version 2.1.0

Contents

System Overview

1.1 Fiberless Wireless Optoelectrophysiology System

The Fiberless Wireless Optoelectrophysiology System (FiWi Oephys) combines optogenetics and electrophysiological recordings. The system delivers an optical signal to the region of interest and detects electrical signals and fields that arise from neural activity.
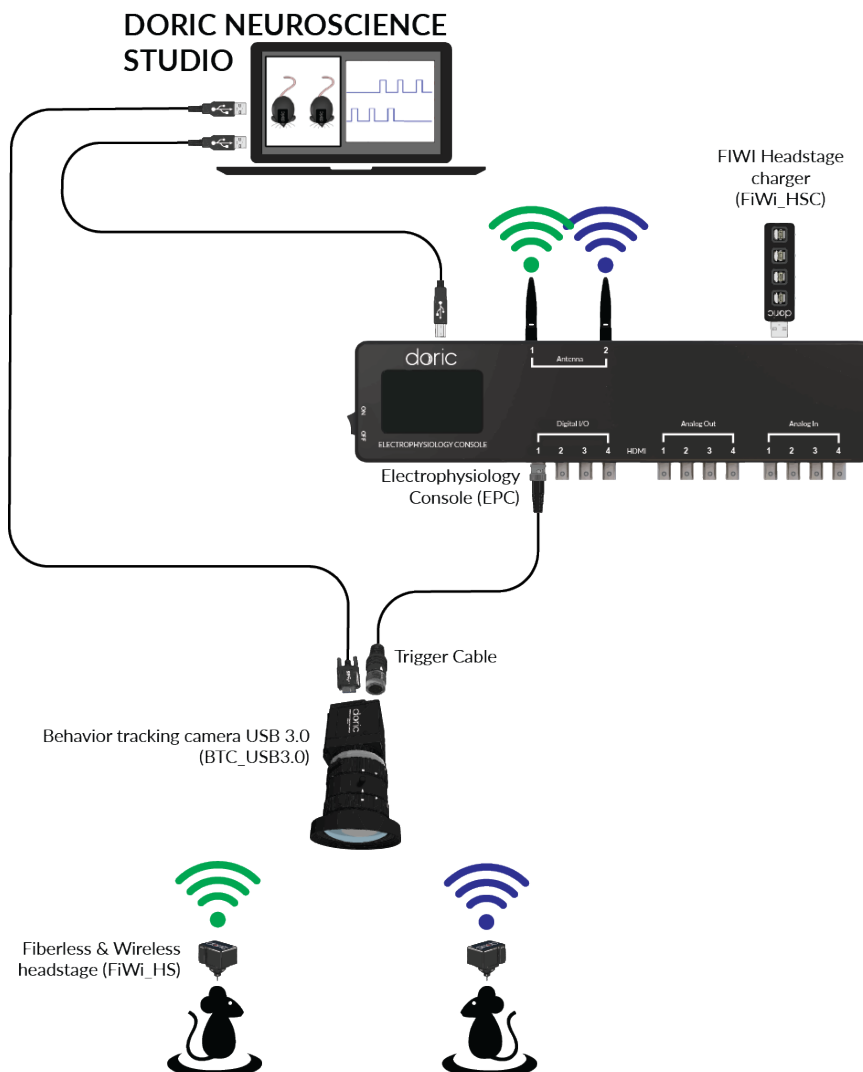

Figure 1.1: *Optogenetically Synchronized Electrophysiology System Layout*

The system uses a *Fiberless & Wireless Headstage* (Fig. 1.2) to transmit wireless data between the *Fi-Wi Opto-electric Cannula* and the *Electrophysiology Console*. This allows small animal subjects high mobility while performing optogenetic and electrophysiological recording experiments. The system can easily be integrated alongside other *Doric Lenses* products, including our *Behavior Tracking Camera*.

A standard FiWi Oephys system (Fig. 1.1) contains the following elements:

- The *Fiberless & Wireless Headstage* x2.

- The *Fi-Wi Opto-electric cannula* x3.

- The *Electrophysiology Console* x1.

- The *Dummy Headstage* x1.

- The *USB Charger* x1.

- The *Test Cannula* x1.

- The *Implantation Holder* x1.

- The *Behavior tracking camera* x1 (optional).

- The *Doric Neuroscience Studio*.
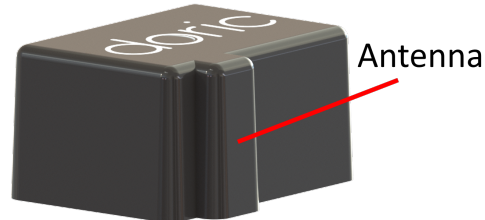
1.1.1   Fiberless & Wireless Headstage

Figure 1.2: *Fiberless & Wireless Headstage*

The *Fiberless & Wireless Headstage* (Fig. 1.2) is an electronic device that creates the link between the opto-electric cannula and the electrophysiology console. Its primary use is to record, amplify and digitize the electrophysiological signal. This is done using the **Intan RHD 2132** chip. Its secondary use is as an LED driver. It sends pulse sequences to the stimulation LED contained in the cannula. The headstage also contains a battery that powers the LED as well as all wireless transmissions. The headstage is secured to the cannula using a snap-on connector.

- The **Antenna** (Fig. 1.2) is a small ridge on the side of the headstage used to transmit wireless electrical signal.

- The **Headstage connector**, found on the underside of the headstage, links it to the cannula transmitting data and power.
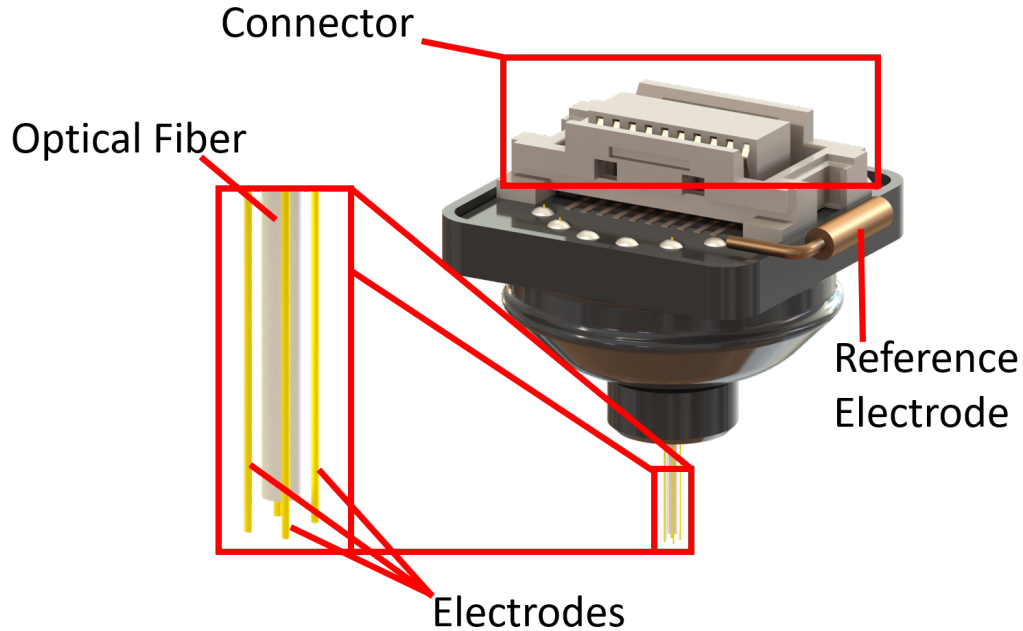
1.1.2 Fi-Wi Opto-electric Cannula

Figure 1.3: *Fi-Wi Opto-electric Cannula*

The *Fi-Wi Opto-electric cannula* (Fig. 1.3) contains an LED connected to a single optical fiber, allowing light to be sent into the region of interest. The cannula also contains the electrodes, positioned around the optical fiber, up to a maximum of 4. Electrode-only and optical-only cannulas are available on request.

- The **Cannula connector** links the cannula and the headstage, transmitting data and power.

- The **Recording Electrodes** receive electrical signal from the brain region in which their tips are located.

- The cannula also includes a single pin connector (0.3mm) for a **Reference electrode** that can be intra- or extra-cranial.

- The **Optical Fiber** guides light from an LED integrated into the cannula.

Much care should be taken when unpackaging the **Fi-Wi Opto-electric Cannula**. The **Electrodes** and the **Optical Fiber** are very fragile. While removing the protective cap, ensure the movement used is parallel to the axis of the **Optical Fiber**. Removing the cap with a gentle unscrewing motion can help keep the cap in the right orientation.
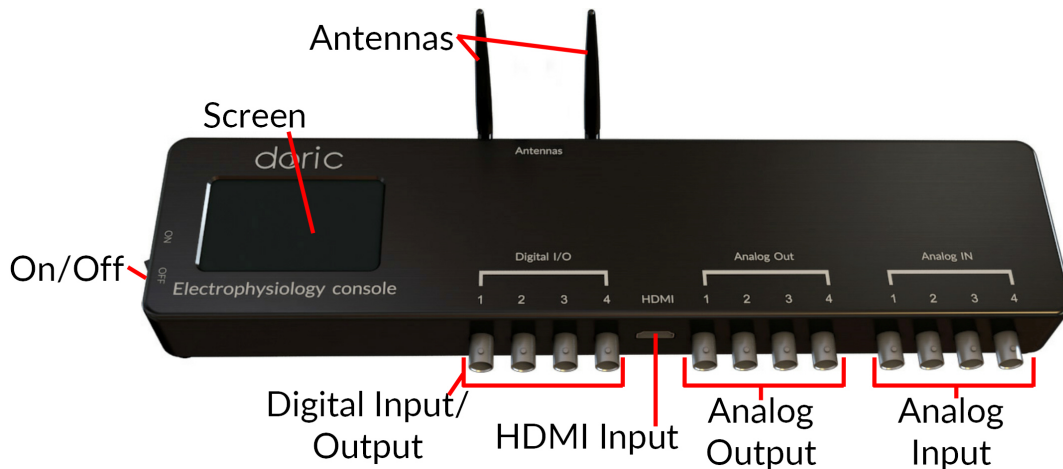
1.1.3 Electrophysiology Console

Figure 1.4: *Electrophysiology Console, Front*

The *Electrophysiology Console* (Fig. 1.4) allows control of the different elements, as well as reception and transmission of information from the control computer.

- The **ON/OFF** (Fig. 1.4) switch activates the console.

- The **LCD Screen** (Fig. 1.4) shows the status of the system's channels.

- The **DIGITAL I/O** (Fig. 1.4) ports control, send and receive 0-5 V TTL signals.

- The **HDMI** (Fig. 1.4) port acquires digital signals and digital communications via a custom pinout HDMI connector.

- The **Analog-Out** (Fig. 1.4) ports send a variable ± 4.75 V signal.

- The **Analog-In** (Fig. 1.4) ports acquire analog signals coming from various inputs.

- The two **Antennas** (Fig. 1.4) connect to the **SMA Connectors** (Fig. 1.5) and allow wireless connection to the headstages.

- The **12 VDC power input** (Fig. 1.5) connects to the 12 VDC power supply.

- The **USB Computer Connection** (Fig. 1.5) port allows connection to a computer using a USB-A/USB-B cable.

- The **USB Charging Port** (Fig. 1.5) is used by the *Fi-Wi Headstage Charger* to recharge the headstage battery. It is best to connect the charger to the console using the *USB-A Male/Female cable*.

- The **Service** (Fig. 1.5) port is a USB-B input through which the firmware of the channels can be updated (for the motherboard firmware update, it's necessary to use the USB Computer connection port).
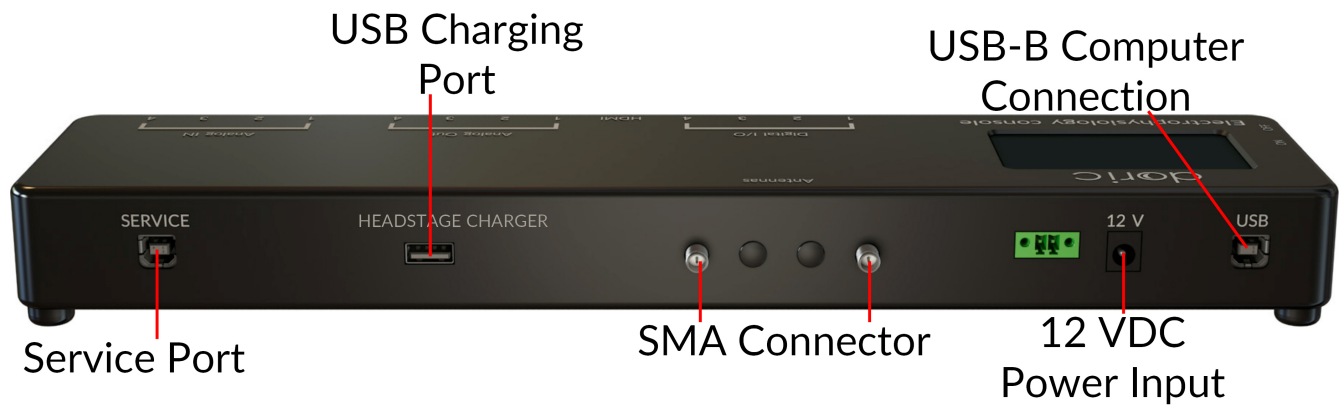
USB Charging
Port

USB-B Computer
Connection

SERVICE   HEADSTAGE CHARGER   12 V   USB

Service Port   SMA Connector   12 VDC
                                                               Power Input

Figure 1.5: *Electrophysiology Console, Rear*

1.1.4   FiWi Headstage Charger

Charging Headstage

Docking
Port

USB
Connector
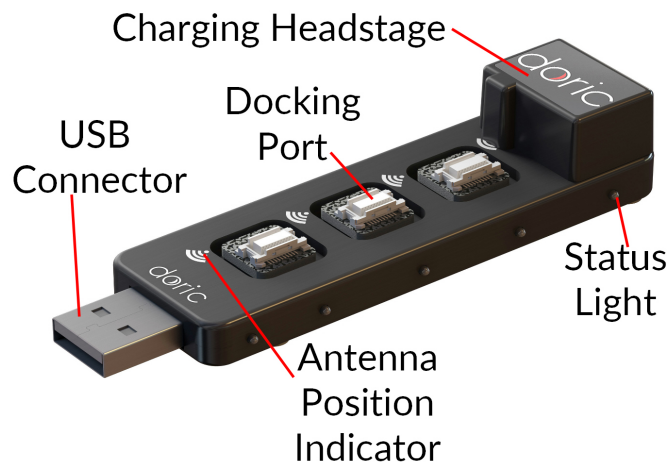
Status
Light

Antenna
Position
Indicator

Figure 1.6: *USB Charger*

The *FiWi Headstage Charger* (Fig. 1.6) is used to recharge the headstage batteries and shut down the wireless con-
nection. During the pairing process, all unused headstages must be docked on the charger, otherwise they will be
detected in-place of the target headstage. The charger uses the same snap-in connectors as the cannula.

- The **USB Connector** connects the charger to the console or directly to a computer.

- The **Docking Ports** are used to recharge docked headstages. When a headstage is connected to a **Docking Port**,
  its wireless connection is shut down.

- The **Status Lights**, on the side of the charger, displays the connection stage of the headstages. The one closest
  to the connector is blue when the charger is ready for usage. The others are yellow when a headstage is charging
  on a corresponding docking port. Once a headstage battery is fully charged, the light goes from yellow to green.

- The **Antenna Position Indicator** shows the proper placement of the antenna when docking a headstage to
  recharge the battery.

1.1.5   Test cannula

The *Test Cannula* is a simplified version of the *Fi-Wi cannula* that is used to test headstage performance. It can be used
to test illumination sequences and ensure proper wireless connectivity, as the headstage alone provides no external
indicators of activity. It otherwise has all the same functions as the *Fi-Wi cannula*, with the exception that it is not
designed to be implanted.

1.1.6 Dummy Headstage

The *Dummy Headstage* is a simplified version of the *Fi-Wi Headstage* that is used to habituate an animal subject to the weight of the headstage. It contains no electronics or other valuable components, and will not function as a *Fi-Wi Headstage*.
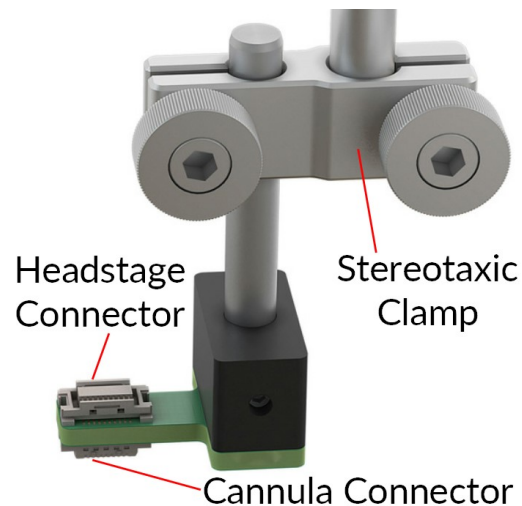
1.1.7 Implantation Holder

Figure 1.7: *Fi-Wi Cannula Implantation Holder*

The *Implantation Holder* is used to hold the *Fi-Wi Cannula* during surgery. The holder is designed to integrate with a stereotaxic apparatus. The holder is comprised of the following elements:

- The **Stereotaxic Clamp** is used to attach the holder to a rod in a stereotaxic apparatus.

- The **Headstage Connector** is used to hold a *Fi-Wi Headstage*. A headstage in this position will be able to send/receive signals from a cannula connected to the **Cannula Connector**.

- The **Cannula Connector** is used to hold a *Fi-Wi Cannula*. A cannula in this position will be able to send/receive signals from a headstage connected to the **Headstage Connector**.

1.1.8 Behavior Tracking Camera

Figure 1.8: *Behavior Tracking Camera*

The **Behavior Tracking Camera** (Fig. 1.8) is an optional addition that allows the observation of the subject in real-time. See the product page for more information.

Operations Guide

2.1 System Setup

The following steps assume the *Fi-Wi Opto-electric Cannula* is already implanted in the region-of-interest of the animal's brain (Fig. 2.1). The process of implantation is not described in this document.
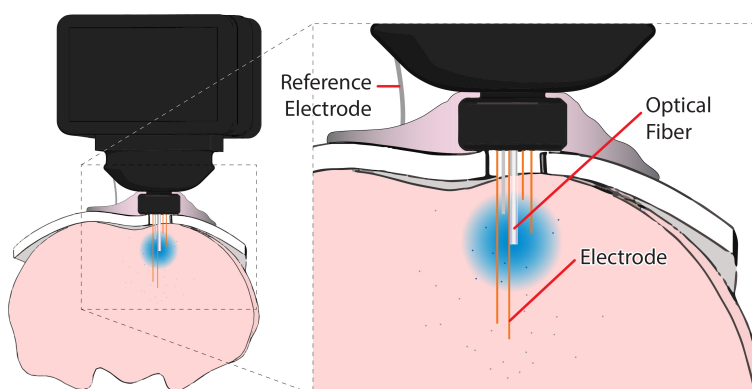

Figure 2.1: *Implanted Fi-Wi Opto-electric Cannula*
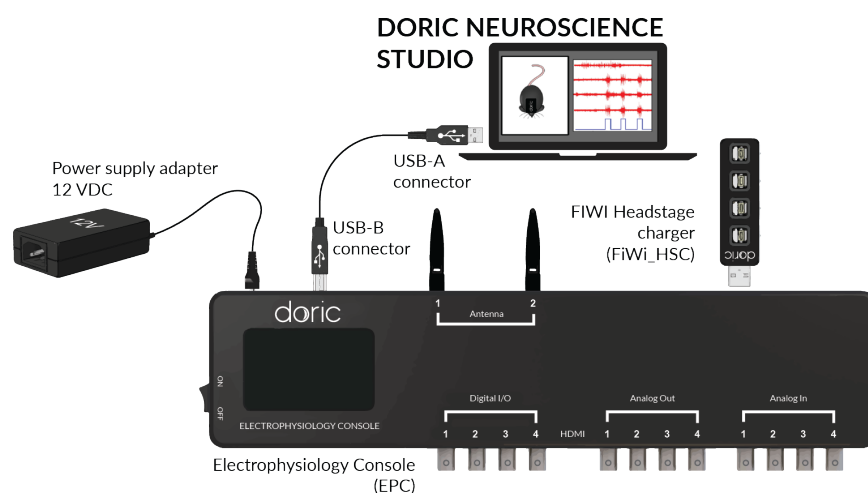
2.1.1 Electrophysiology Console Setup

Figure 2.2: *Electrophysiology Console Setup*

1. Screw each antenna into the **SMA Connector**. Fold the antenna upwards for optimal reception.

2. Connect the console to the **12 VDC power supply** and turn **ON** the console.

3. Connect the *Fi-Wi Headstage Charger* to the **USB Docking Port**. Install the headstage(s) onto the charging port.

4. Connect the console to the computer using the **USB-A/USB-B Cable**.

2.1.2 Fiberless & Wireless Headstage Setup

The *Fiberless and Wireless Headstage* is the bridge between the *Fi-Wi Opto-electric Cannula* and the *Electrophysiology Console*. Some preparation is required for optimal use.
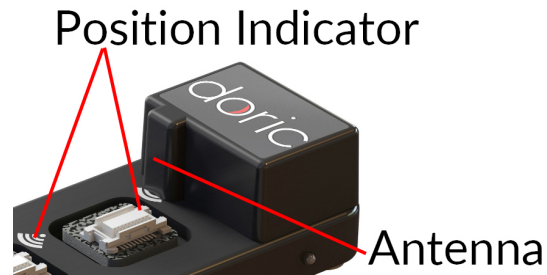

Figure 2.3: *Headstage charger installation*

1. The *Fiberless and Wireless Headstage* must first be charged. Place a headstage onto the *USB Charger* so the antenna is on the position indicator.

   - The console must be **ON** for the headstage to recharge, which requires > 60 min.
   - The headstage cannot connect wirelessly while being charged.
   - While recharging, the **Status Lights** of each **docking port** are yellow. These turn green once the headstage is sufficiently charged for use, which typically takes approximately an hour.

2. Remove a headstage from the *USB Charger*.

3. Click the **Pair** button in the **Wireless Settings**. Once properly paired, the section will display the **Headstage ID**, the **Battery Level** and the **Signal Strength**.
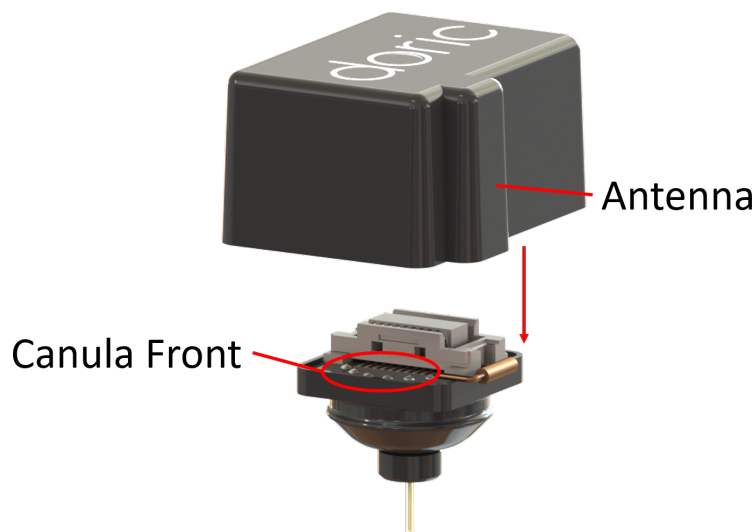

Figure 2.4: *Headstage placement*

4. The headstage is connected to the cannula using a snap-on connector. For the two elements to connect properly, the headstage antenna ridge must be oriented towards the front of the cannula, as shown in Figure 2.4.

5. Connect the headstage to the **Test Cannula** and place it inside the experimental space. Test your **Signal Strength** and the LED to ensure proper function of the headstage. This is used to validate transmission quality between the headstage and the console during an experiment.

6. The headstage can then be installed onto the *Fi-Wi Opto-electric Cannula* for experimental use. Ensure the protective cap is removed from the cannula using a motion that is parallel to the **Optical Fiber**, as described in section 1.1.2.

7. After use, place the **Fiberless and Wireless Headstage** back onto the **USB Charger** to reset the wireless connection.

8. To ensure optimal transmission:

    - The experimental subjects should be on the same plane as the console antenna.
    - Avoid placing metallic objects between the console and the **Headstage**. For proper transmission, the vertical faces of the animal cage must be non-metallic.
    - If the signal is not properly received, change the wireless **Channel**. Note that nearby Wi-Fi signals can interfere with transmission.
    - Ensure the antenna is fully screwed into the **SMA Connector** (Fig. 1.5).
    - Before starting an experiment, place the *Fi-Wi Headstage* in the experimental area to verify transmission quality.

9. It is possible to connect a second **Fiberless and Wireless Headstage** to the console at the same time. When using a second headstage, ensure that each headstage is using another **Channel**.

Doric Neuroscience Studio

The Doric Neuroscience Studio's Fiberless and Wireless *Electrophysiology Console* module takes care of pairing the headstage to the Console, collecting electrophysiological data and synchronizes of electrophysiology with optogenetics, behaviour and other measurements. Through its intuitive user interface, scientists can configure channels with great flexibility, fitting a wide range of experimental setups.
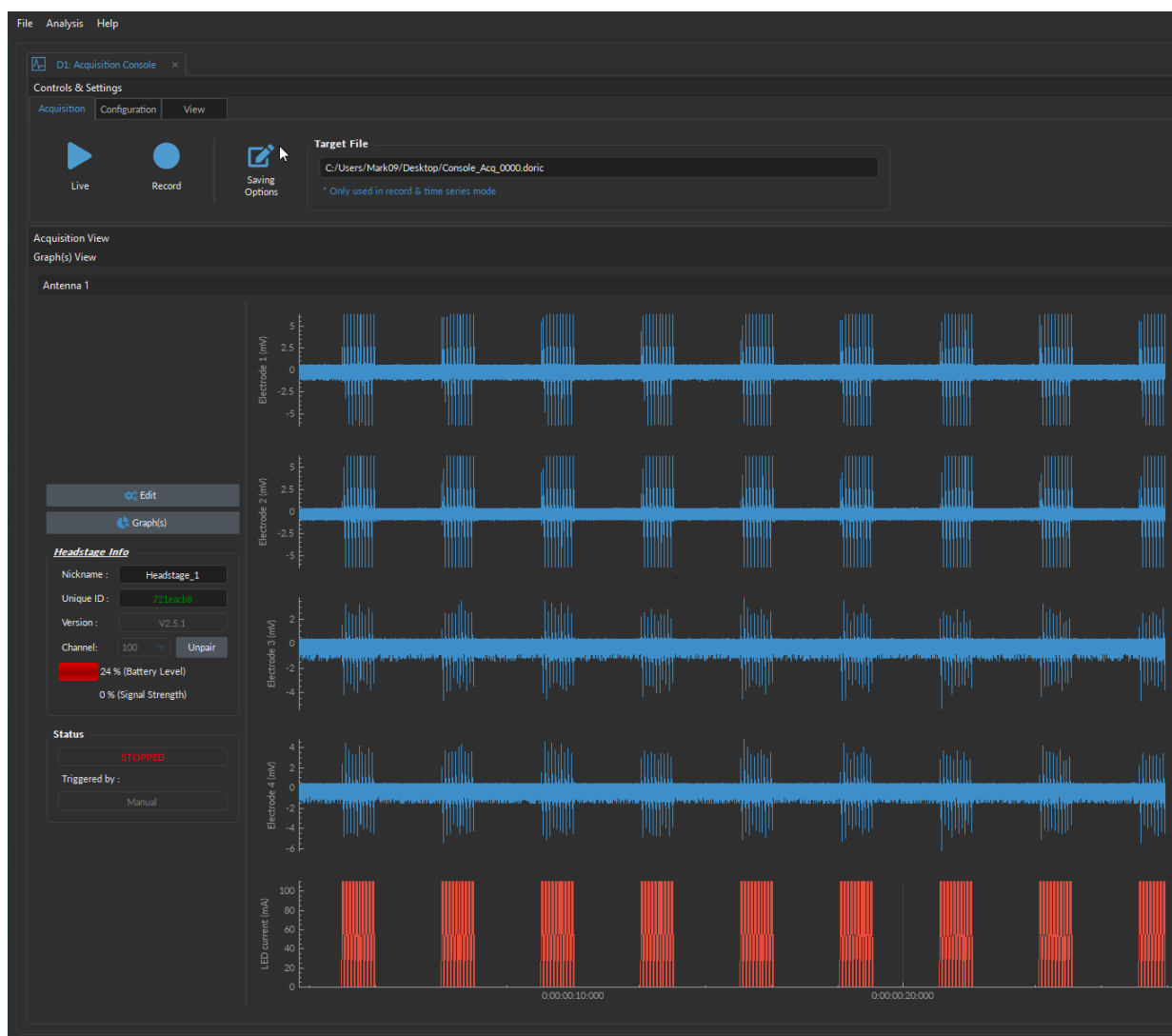

Figure 3.1: *Electrophysiology console user interface*

3.1 Connecting devices to DNS

Once *Doric Neuroscience Studio* is opened, the *Device Selection* window should automatically pop up, if the device is turned ON and properly connected to the computer with USB port (as in Fig. 3.2).

To add a device to the studio, **double click** on the device of choice in the *Available device(s)* sections (bottom half of window). If the device in question does not show up, double-check that it is indeed turned ON and the two ends of the USB cord are properly connected within the USB port. Then click *Refresh*. When properly connected to the system, the device will appear in the *Connected/Opened device(s)* section of the Window (see the green checkmark in Fig. 3.2).
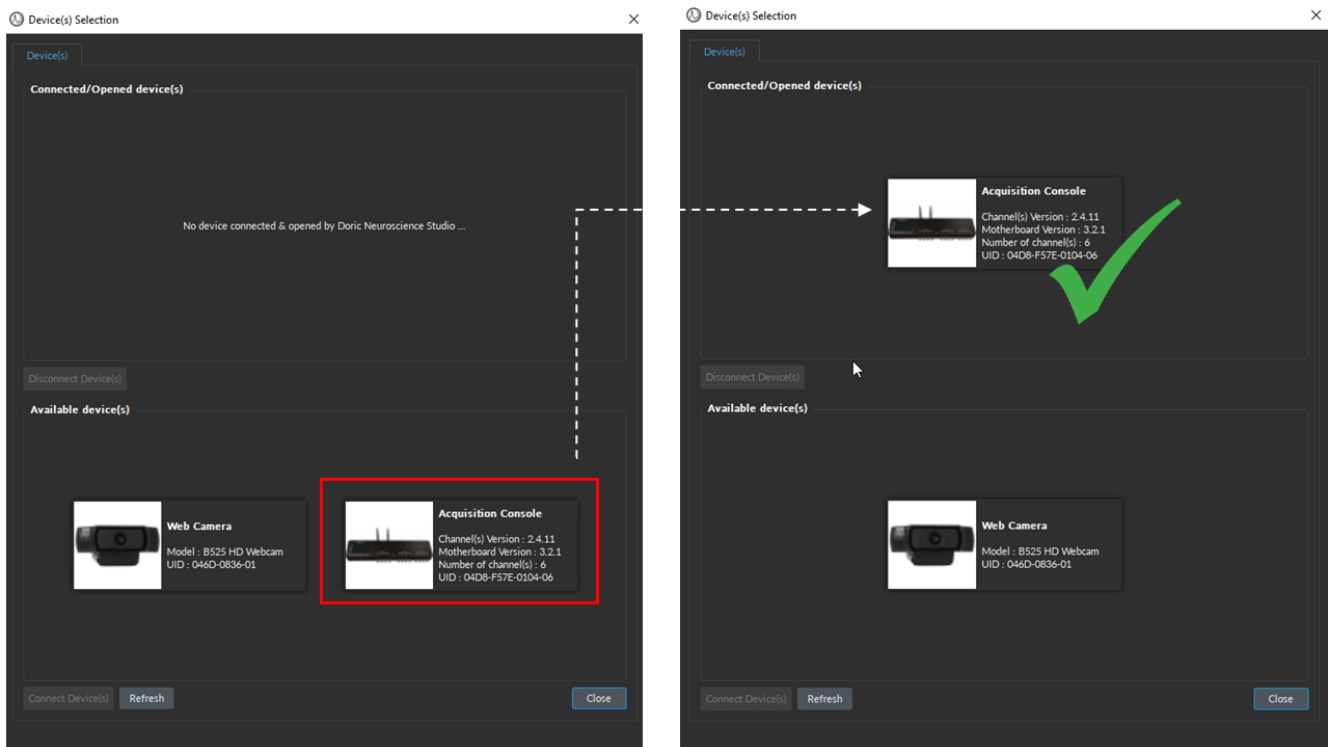

Figure 3.2: *Double click on the device of choice to connect it to DNS*

NOTE: If you have switched to DNS v6, older devices will require a firmware update to be recognized by the new version of the software. This update can be easily done using *Doric Maintenance Tools (DMT)* application and must be done one by one for each device. Further instructions can be found **HERE**.

Manually opening the *Device(s) Selection* window:
To manually open the *Device(s) Selection* window, select the *File*, then *Device Selection* (as per Fig. 3.3) or use the hot key: *Ctrl+N*.
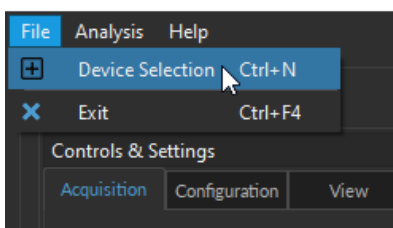

Figure 3.3: *Open Device Selection Window*

3.2 Overview

The **Acquisition Console** Module for the FiWi System of *Doric Neuroscience Studio* software is split into three sections: **(1) Control and settings tabs** (Section 3.3) are used to manage different elements of the software (Acquisition, Configuration, and View); **(2) Headstage Info** (Section 3.5) is used to pair the Headstage to the console; **(3) Acquisition view** (Section 3.5) displays the input and output traces for visualization.
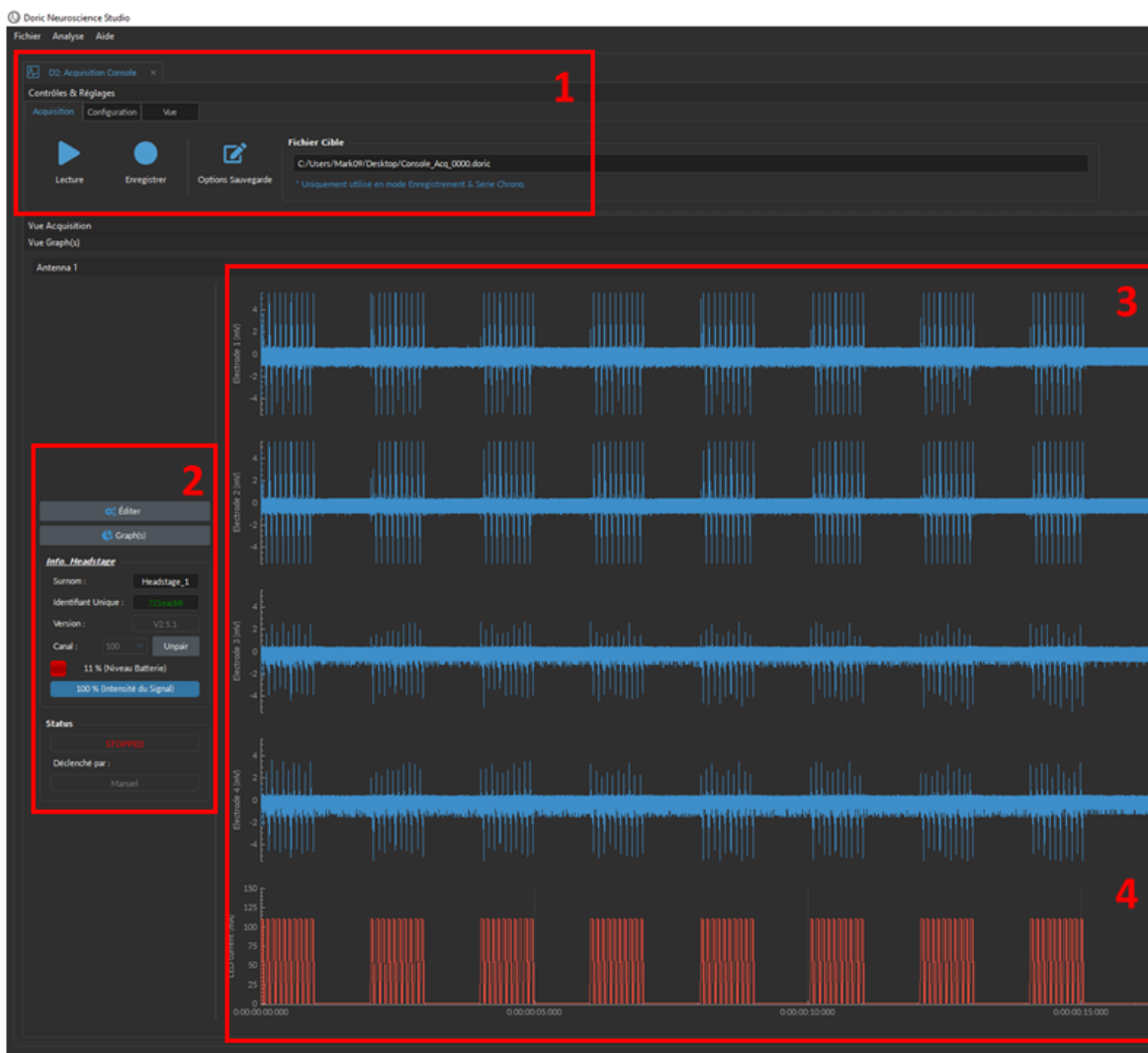

Figure 3.4: *DNS user interface*

3.3 Control and Settings tabs

The three **Control and settings tabs** are used to manage the different parts of the software. There are three tabs, **Acquisition** (Section 3.3.1), **Configuration** (Section 3.3.2), and **View** (Section 3.3.3).

3.3.1 Acquisition Tab

The **Acquisition** tab is used to start a live/recording session and set the saving parameters. The **Live** and **Record** buttons will not function if channels have yet to be set-up. See section 3.4.1 to configure channels for recording.
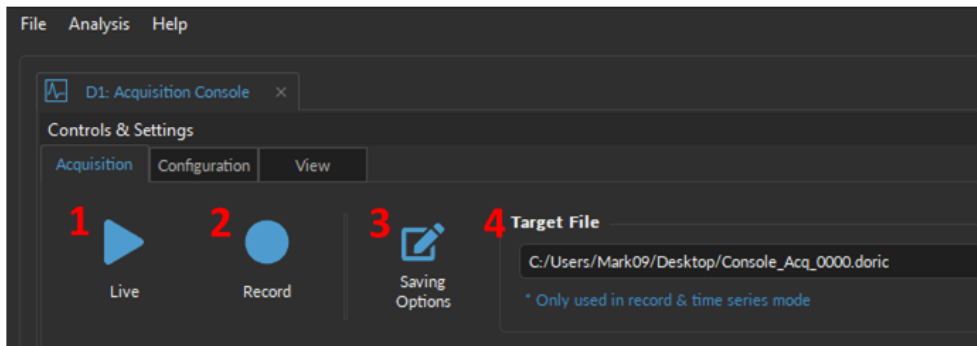

Figure 3.5: *Acquisition Tab*

1. The **Live** button (Fig. 3.5, 1) activates all prepared channels. This mode does not save data, keeping only the most recent 700 000 data points in memory. This mode is not recommended for long or critical measurement sequences. **Live** mode is useful to quickly test the recording software and to ensure that the parameters were properly set.

2. The **Record** button (Fig. 3.5, 2) activates all prepared channels while periodically saving recorded data to the disk. This mode is recommended for long measurement sequences.

3. The **Saving Options** (Fig. 3.5, 3) button opens the **Saving Parameters** window (Fig. 3.6). See section 3.3.1.1 for greater details.

4. The **Target File** (Fig. 3.5, 4) displays the path and file name where the data will be stored once the **Record** button is selected. Select the **Saving Options** button to change the path and file name.
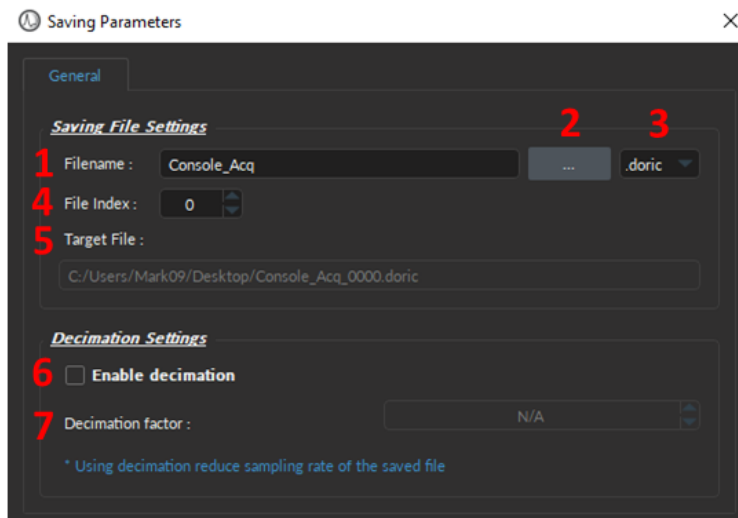

Figure 3.6: *Saving Menu Window*

3.3.1.1 Saving Parameters

The **Saving Parameter** window is used to define how and where the file is saved. This window is opened by selecting the **Saving Options** button in the Acquisition Tab (Fig. 3.5, 3).

1. The **Filename** text-box lets users specify the name of the data file that will be saved (Fig. 3.6, 1).

2. The **[...]** button opens a File Explorer window where users can select the folder where the data will be saved (Fig. 3.6, 2).

3. The **File format** (Fig. 3.6, 3) is **.doric**, an HDF5-based format that supports metadata (signal, video, images, tables, parameters, etc.). Version 6 of *Doric Neuroscience Studio* is no longer compatible with other file formats (.csv, .excel, or .tiff). We provide Matlab, Python, and Octave codes to read **.doric** files HERE. While not recommended, it is possible to export a *.doric* file into .csv format through the **Doric File Reader** module.

4. The **File Index** (Fig. 3.6, 4) box is used to define the current indexation number used for multiple files saved during the same measurement session. The suffix is incremented automatically when recording multiple files.

5. The **Target File** (Fig. 3.6, 5) displays the absolute path and filename where the data will be saved.

6. The **Enable decimation** checkbox (Fig. 3.6, 6) provides a way to reduce the file sizes. This method conserves one point over a number of data points equal to the **Decimation Factor**.

7. The **Decimation factor** text-box (Fig. 3.6, 7) is used to define the number of points saved.[1]

3.3.2 Configuration Tab

The **Configuration** tab is used to set the channels and the global settings (such as sampling rate and Master trigger options), as well as save and load the preset channel configurations.
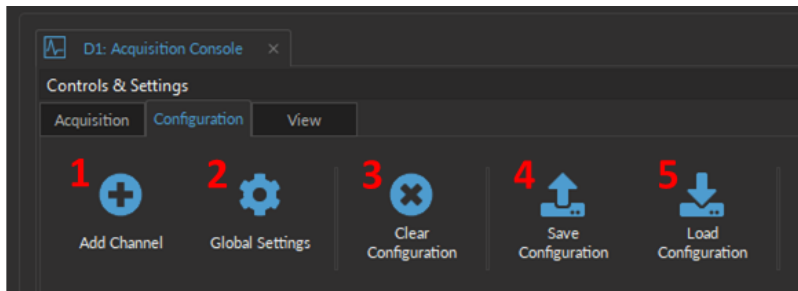

Figure 3.7: *Configuration Tab*

1. The **Add Channel** button (Fig. 3.7, 1) opens the **Channels configuration** window. How to *add* and *configure* a channel is detailed in Section 3.4. Table 3.1 describes different types of channels available, their use cases and their individual sections.

2. The **Global Settings** (Fig. 3.7, 2) opens the **Global Options** window in Fig. 3.8, where user can set the acquisition sampling rate and specify the master trigger options. See Sections 3.3.2.1 for more details.

3. The **Clear configuration** button (Fig. 3.7, 3) resets the acquisition view and all other parameters set. Any configurations not saved will be lost.

4. The **Save configuration** button (Fig. 3.7, 4) allows a console configuration to be saved in the **.doric** format. This file preserves the current channel configuration/parameters, the Acquisition View window organization, and any custom trace colors and names.

5. The **Load configuration** button (Fig. 3.7, 5) imports a pre-configured **.doric** file into the module.

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[1]For a data set of 10 points, saved with a **Decimation Factor** of 2, the first point will be saved, the third ... This produces a file of 5 points of data.

3.3.2.1 Global Settings

Through the **Global Settings**, user can set the acquisition **Sampling Rate** and specify the **Master Trigger Options** that will start recordings.
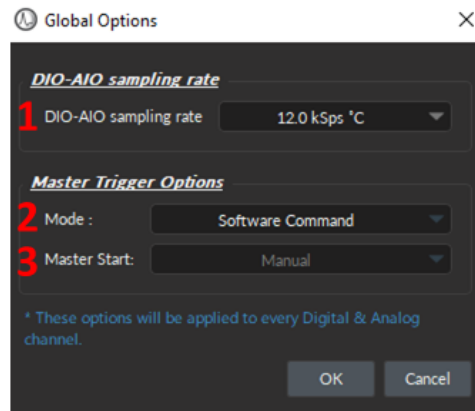

Figure 3.8: *Global Options Window*

1. The **DIO-AIO sampling rate** (Fig. 3.8-1) is 12 kSps°C by default. This value was selected because it is the highest value that still produced reliable data given the hardware limitations of the devices. See section 3.3.1.1 to enable the *Decimation* and effectively reduce the saving sampling rate and restrict the data file size.

2. The **Mode** (Fig. 3.8, 2) of the **Master Trigger Options** sets the origin (internal, external or time-series) of the trigger that will start the recording session and synchronize all the external and internal devices. Four options are available for different use cases:

   - *Software Command* - The recording will start when the **Record** button is selected in the **Acquisition Tab** (Fig. 3.5, 2). The **Master Start** is, by definition, always **Manual**.

   - *Triggered* - The recording session starts when a trigger signal is received (from the **Master Start**, either manual or from an external digital source), and continues even if the trigger signal stops. Thus, the **Triggered** mode only controls the START of the recording session (and NOT the endpoint).

   - *Gated* - The recording session starts when a high TTL signal (>4 V) is detected (from the **Master Start**, either manual or from an external digital source), and will stop when a low TTL signal (<0.4 V) is detected. Thus, the **Gated** mode controls both the START and the END signals of the recording session.

   - *Timeseries* - This mode allows users to record pre-defined series over longer periods of time (that can span several days) (Fig 3.9a). This mode works similarly to the *Sotware Command* mode, however, when the **Record** button is selected, the **Time Series Window** (Fig 3.9b) pops up. See section 3.3.2.2 for more details.

3. The **Master Start** (Fig. 3.8, 3) defines the source that will automatically start the recording. This source can either be:

   - *Manuel* - the user ultimately starts the recording session by clicking **Record** within *Doric Neuroscience Studio*;

   - *Digital I/O Channel (1-4)* - The specified channel will automatically begin the recording session when it receives a digital trigger pulse from an external device. ***However, this mode still requires that the **Record** button is selected BEFORE the TTL trigger signal is received.***

3.3.2.2 Time Series

The **Time Series** Window (Fig 3.9b) can be opened by clicking on the **Record** button (Fig. 3.5, 2) when the **Master Trigger** is in **Time Series** mode in the **Global Settings** window (Fig. 3.8, 2). Every **Time series** sequence is automatically saved to the *.doric* file defined in **Saving Options** (Section 3.3.1.1).
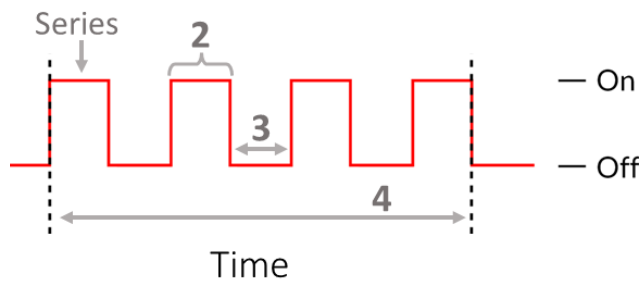
(a) *Time Series Acquisition Timing Diagram*
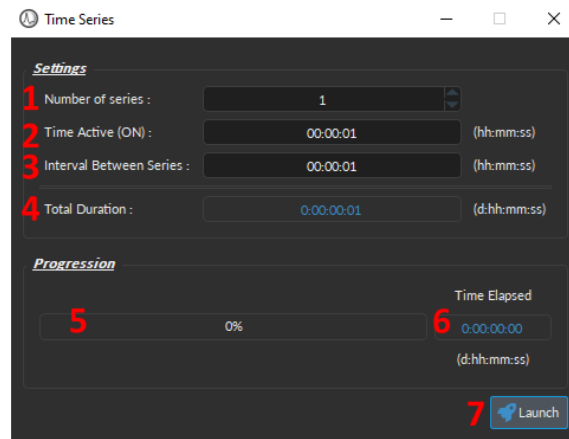
(b) *Time Series Window*

Figure 3.9: *Time Series Mode can be set through Global Settings*

The **Time Series** window (Fig. 3.9b) sets the following parameters:

1. The **Number of series** (Fig. 3.9b, 1) defines the amount of times the series is repeated.

2. The **Time Active (ON)** (Fig. 3.9b, 2) defines the duration of the series.

3. The **Interval Between Series** (Fig. 3.9b-3) defines the amount of time between each series, if the **Number of series** is greater than 1.

4. The **Total Duration** (Fig. 3.9b, 4) displays the total amount of time that the timeseries recording will take place.

5. The **Progression bar** (Fig. 3.9b, 5) indicates the progression of the timeseries (in %).

6. The **Time Elapsed** (Fig. 3.9b-6) counter indicates the amount of time that has already passed in d:hh:mm:ss.

7. The **Launch** (Fig. 3.9b, 7) button start the series. While the series is active, it is impossible to add channels or change the configuration, though **View** settings can be modified.

3.3.3 View Tab

The **View Tab** (Fig. 3.10) is used to modify the presentation of graphs in the **Acquisition view**.
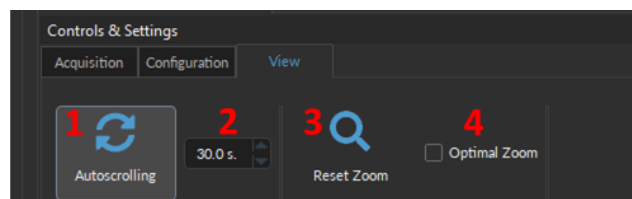

Figure 3.10: *View Tab*

The **View** parameters are as follows:

1. The **Autoscrolling** button (Fig. 3.10, 1), when selected, automatically set the graphs to scroll as new data appears.

2. the **Zoom range** (Fig. 3.10, 2) sets the graph zoom to the value of choice, specified in the text-box.

3. The **Reset Zoom** button (Fig. 3.10, 3) readjusts the graph zoom to the value specified in the zoom range text-box.

4. The **Optimal Zoom** check-box (Fig. 3.10, 4) automatically adjusts the graph range based on the values of the data collected. Smaller values will lead to greater zoom, and vice versa.

3.4   Channel Configurations

Before pairing the Headstage of the *FiWi electrophysiology system*, users first need to set up the required channels and specify the data acquisition parameters required for the experiment.

3.4.1   Add Channel:

To create a new channel, regardless of the input and/or output type, select the **Add Channel** button, which can be found under the **Configuration** tab (Fig. 3.11). This will open the **Channel(s) Configuration** window (Fig. 3.13).
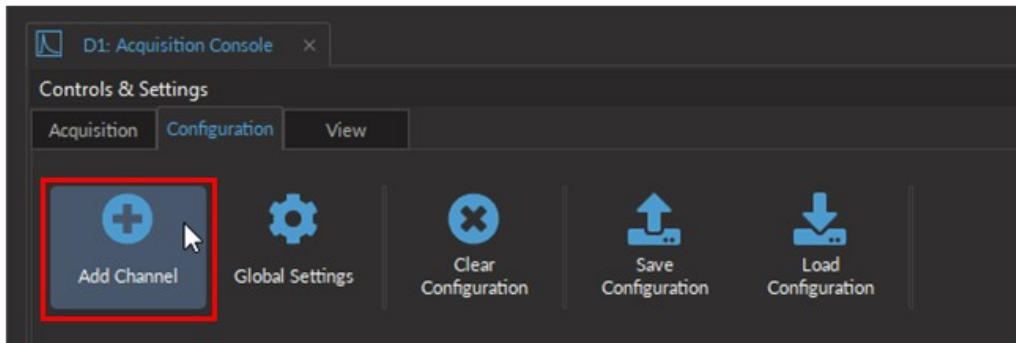

Figure 3.11: *Add Channel button opens the Channel Configuration window*

To generate a new **Channel** using the **Channel(s) configuration** window (Fig. 3.12):

1. Select one of the available **Channel Type** icons from the left most column of the **Channel(s) Configuration** window (Fig. 3.12). Table 3.1 describes the use case of each type.

2. Clicking on the icon will display the **Channel Type**-specific options on the right side of the window. Each **Channel Type** has a number of parameters which can be configured to fit the needs of the experiment(s). Details of the parameters and their options will be covered in the following sections. See Table 3.1 for hyperlinks to the relevant sections.

3. Select the **Add** button (Fig. 3.12) to generate the defined channel or to update an already configured channel, but does not automatically close the *Channel Configuration* window. This allows the user to conveniently set up all required channels one after the other.

4. Select the **Close** button to shut the window once all channels are configured.

Figure 3.12: *Channel(s) configuration window, Digital I/O input*

3.4.2 Channels Types

Different types of input and output can be configured for the particular of the experiment by creating a new Channel in the Configuration tab or editing an existing one (Fig 3.11). Table 3.1 details the types of inputs and output the console and the software can handle and gives a quick access to their sections.

Table 3.1: *Types of channels and their use cases*

Icon	Channel Type	Use Case	Section
	Antenna	Pair and record electrophysiological signal from the FiWi Headstage	3.4.3
	Digital I/O	For input and output of TTL signals	3.4.4
	Analog Output	For the output of analog signals, such as sine, stair or customized	3.4.5
	Analog Input	To collect the fluorescent signal (such as GCamp, RCamp, Isosbestic or FRET)	3.4.6
	Camera(s)	To collect video for behaviour experiments	3.4.7
	Keypress Event(s)	To manually flag events time-locked to the current recording using customized keys	3.4.8

3.4.3   Antenna Channel

The **Antenna** Channel is required to pair the wireless Headstage to the software, to record the four electrophysiology inputs, in addition to output the digital triggers that control the LED current.

The *Channel(s) Configuration* window for the Antenna Channel is divide into four sections (Fig. 3.13): (1) the **Channel Options** (Section 3.4.3.1), (2) the **Sequence Options** & (3) **Preview** (both treated in Section 3.4.3.3), and (4) the **LED Trigger Options** (Section 3.4.3.2).
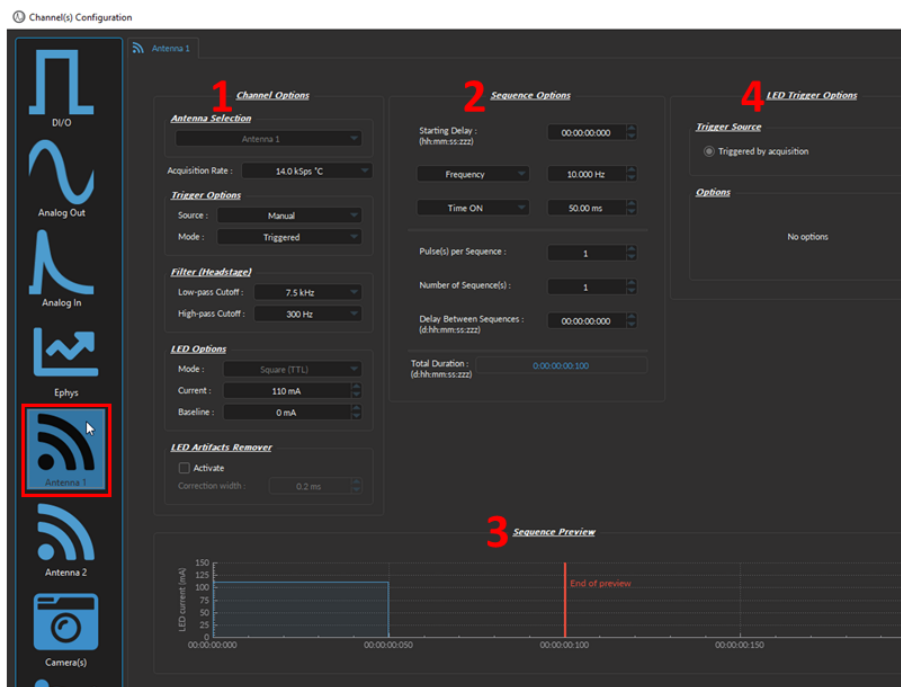

Figure 3.13: *Electrophysiology channel configuration window*

3.4.3.1   Channel Options

The **Channel Options** (Fig. 3.14) of the Antenna *Channel Configuration* window sets the trigger and recording parameters, as follows:

1. The **Antenna selection** (Fig. 3.14, 1) box shows the currently selected antenna.

    - The **Acquisition Rate** is the sampling rate of the electrophysiological data and can be a value between 0.1kSps°C and 14kSps°C. The default value is set to 14kSps°C.

2. The **Trigger options** (Fig. 3.14, 2) box allows the selection of trigger parameters for data acquisition.

    - The **Trigger source** can either be **Manual**, or can come from the **Digital I/O** channels.
    - The **Trigger mode** can be one of two types. In **Triggered** mode, the measurement sequence starts when a trigger signal is received, and continues even if the trigger signal stops. In **Gated** mode, the measurement sequence starts when a high TTL signal (>4 V) is detected, and will stop when a low TTL signal (<0.4 V) is detected.
        - **NOTE:** If the source is **Manual**, then only the **Trigger** mode is available. Whereas, if the source is one of the four **Digital I/O** channels, the mode can either be **Triggered** or **Gated**.

3. The **Filter (Headstage)** (Fig. 3.14, 3) filters define the high and low-pass frequency cutoff for electrical signal received by the headstage.

    - The **Low-pass Cutoff** (Fig. 3.14, 3) is used to attenuate the noise and smooth the waveform of the spikes. The cut-off value can be set between (1-500Hz), depending on the need of the experiment.

- The **High-pass Cutoff** (Fig. 3.14, 3) is used filter the multi-unit activity that occurs from the average / synchronized spiking of a small population of neurons in the vicinity of the electrodes. It can also removed DC noise. The cut-off value can be set between 0.1-500Hz.

4. The **LED options** (Fig. 3.14, 4) are all parameters used to control the light source of the cannula connected to the *Fi-Wi headstage*, such as:

- The **Mode** (Fig. 3.14, 4) allows the selection of the pulse sequence mode.
- The **Maximum current** (Fig. 3.14, 4) defines the current sent to the cannula LED. For proper function of the cannula, the current should always be greater than 10 mA.
- The **Baseline** (Fig. 3.14, 4) leaves a small offset to the current sent to the LED. It is reccomended to use a small offset, as a complete shut-down of the LED will induce a spike in the electrical acquisition signal.
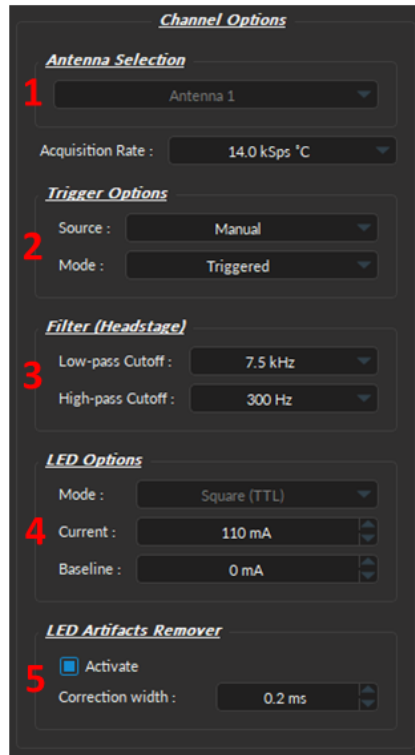

Figure 3.14: *Antenna Channel Options*

5. The **LED Artifacts Remover** reduces the number of photoelectric artifacts that occurs when the electrodes pick up the electrical trigger signals of the LED. These photoelectric artifacts can often look like spikes, but their timing are locked to the initiation (Fig. 3.15a) and termination (Fig. 3.15b) of the LED.

To remove the artifacts, an algorithm detects the first spike occurring after the initiation or termination of the LED (Fig. 3.15). All the value between the time of the first spike until the specified *Correction width* are set to 0V.

To turn on the **LED Artifact Remover**:

- Check the **Active** box (Fig. 3.14, 5);
- Specify the size of the **Correction width** (Fig. 3.14). The minimum value is **0.1 ms**. And while the maximum value can reach 1 second, it is important to note that the larger the **Correction widths**, the more likely true spikes are removed from the signal. Thus, the value must be selected carefully, taking into account this trade-off.

WARNING: Using a large *Correction width* can result in real **signal deletion**.
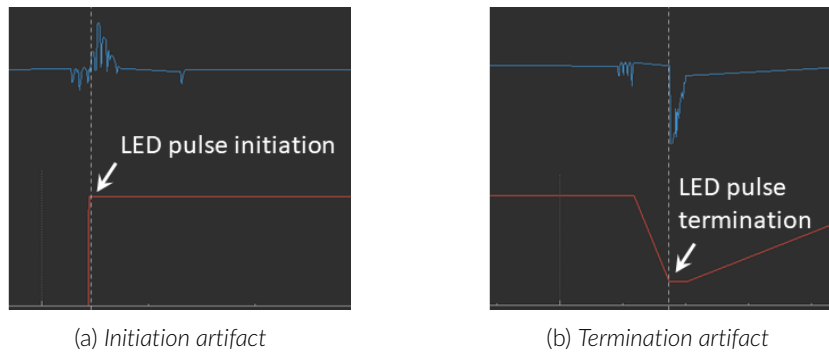**SELECT *Correction width* CAREFULLY!**

(a) *Initiation artifact* (b) *Termination artifact*

Figure 3.15: *Electrical artifacts caused by LED pulses*

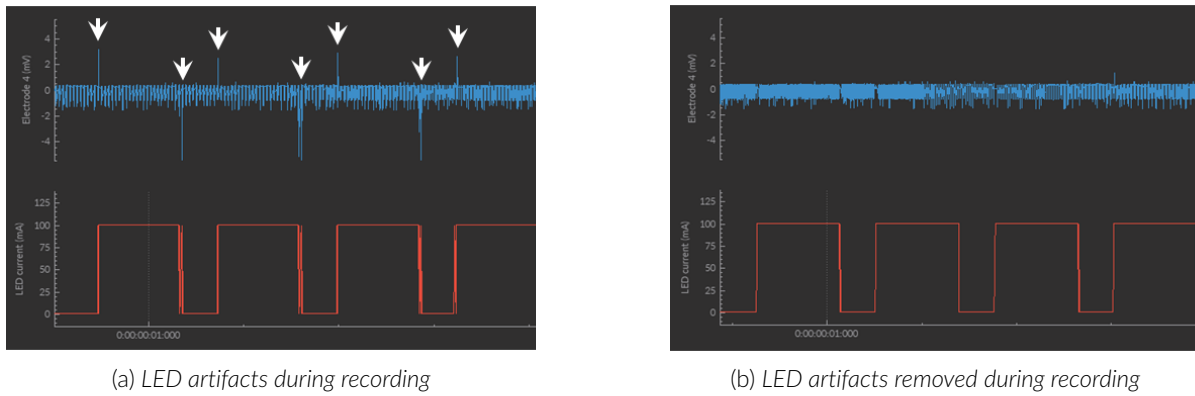(a) *LED artifacts during recording* (b) *LED artifacts removed during recording*

Figure 3.16: *LED Artifact Remover Option Example*

3.4.3.2 LED Trigger Options

The **LED Trigger Options** (Fig. 3.17), allow users to select what device triggers the LED source. By default the **Trigger Source** is the DNS acquisition system.
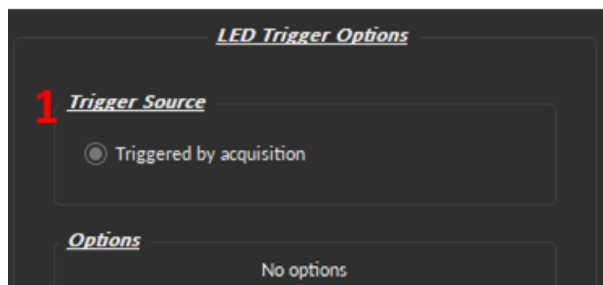

Figure 3.17: *Antenna LED Trigger Options*

3.4.3.3 Sequence Options & Preview

The **Sequence options** section (Fig. 3.18a) contains the LED pulse sequence parameters, while the **Sequence Preview** section (Fig. 3.18b) displays the corresponding shape and timing of the sequence.

1. The **Starting Delay** (Fig. 3.18) sets the delay (in hh:mm:ss:zzz format) before the first pulse.

2. The **Frequency/Period** (Fig. 3.18a) sets the frequency (in Hz) or period (in ms) for the pulse within the sequence. For example, a signal at 10 Hz (frequency) will output one pulse every 100 ms (period), whereas a pulse sequence at 0.5 Hz (frequency) will output one pulse every 2000 ms (period).

3. The **Time ON/Duty Cycle** (Fig. 3.18) sets the time (in ms) or the duty cycle (in %) for each pulse. The **Time ON** must be lower than (1/frequency)+0.005 ms, while the **Duty cycle** must be below 100 %. These squares will appear red should an impossible **Frequency**/**Time ON** be selected.

4. The **Pulses per sequence** (Fig. 3.18a) sets the number of pulses per sequence. If it is set to 0, the pulse will be repeated indefinitely.

5. The **Number of sequences** (Fig. 3.18a) sets the number of times that the sequence will be repeated. If it is set to 0, the sequence will be repeated indefinitely.

6. The **Delay between sequences** (Fig. 3.18) sets the delay (in hh:mm:ss:zzz format) between each sequence if the **Number of Sequences** is greater than 1.

7. The **Total Duration** (Fig. 3.18) displays the total time of the experiment. The different values can be *Inf* for infinite, a valid time value or *Err* if the **Time ON** value is greater than 1/frequency.
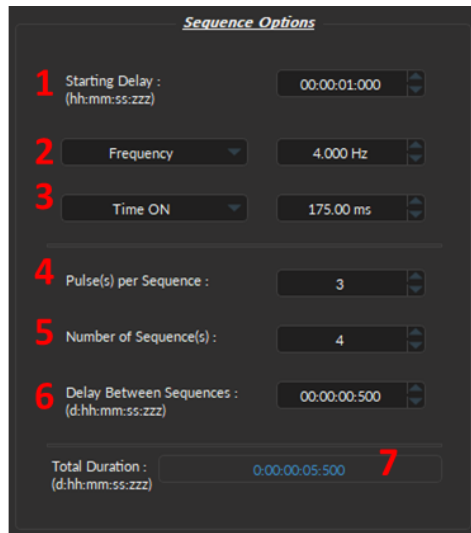
(a) *Sequence Options*
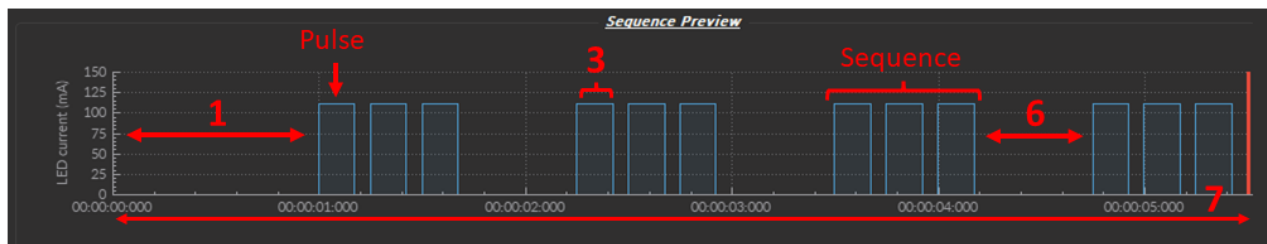
(b) *Sequence Preview*

Figure 3.18: *Channel(s) configuration window, Antenna - Sequence Options and Preview*

3.4.4   Digital I/O Channels

Each **Digital I/O** channel can be configured as an output or an input to create TTL (On/Off) pulse sequences. **Digital Outputs** can provide triggers to external devices (such as light sources) required for the experiment, while remaining synchronized with to recording system. In addition, **Digital Inputs** can record a copy of the trigger of an external driven device used during the experiment (such as the timing of a displayed stimuli or a measured behavior).

The *Channel(s) Configuration* window for the **Digital I/O** Channel is divided into three sections (Fig. 3.13): (1) the **Channel Options** (Section 3.4.4.1), (2) the **Sequence Options** & (3) **Preview** (both treated in Section 3.4.4.2).
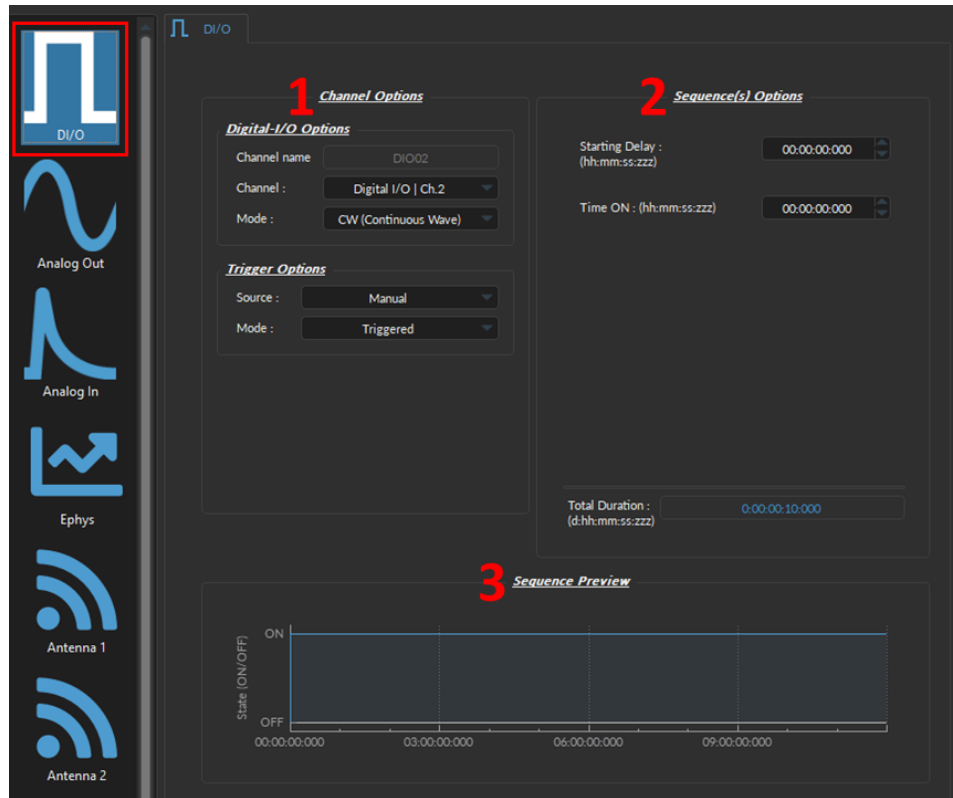

Figure 3.19: *Channel(s) configuration window, Digital I/O - CW mode*

3.4.4.1   Channel Options

The **Channel Options** defines the channel, source and mode of the digital signal, through **Digital I/O Options** and **Trigger Options**.
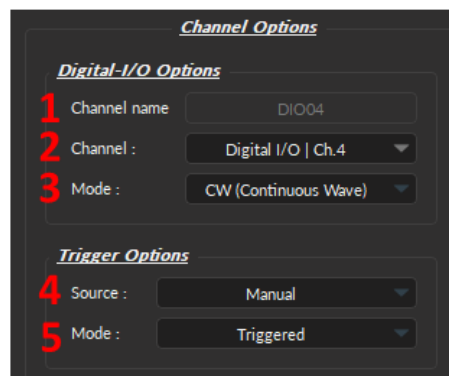

Figure 3.20: *Channel(s) configuration window, Digital I/O Channel Options*

Digital I/O Options:

1. The **Channel name** (Fig 3.20, 1) allows user to specify a label for each channel.

2. The **Channel** (Fig 3.20, 2) identifies the channels available to create a Digital I/O. The channel can be changed by selecting a new one from the drop-down list. Each numbered channel on the physical console corresponds to the same number of the digital channel within the software.

3. The **Mode** (Fig 3.20, 3) identifies the type of signal sent (for output channels) or the way the signal is measured (for input channels). Three modes are available:

   - The **Continuous wave (CW)** Mode (Fig. 3.21a);
   - The **Square (TTL)** Mode (Fig. 3.21b);
   - The **Input** mode receives a signal that are either 0 (**Off**) or 1 (**On**). The channel can then be used as a trigger source for all the other channels of the console (See Section 3.3.2.1). No **Sequence Options** or **Sequence Previews** are available for this mode.
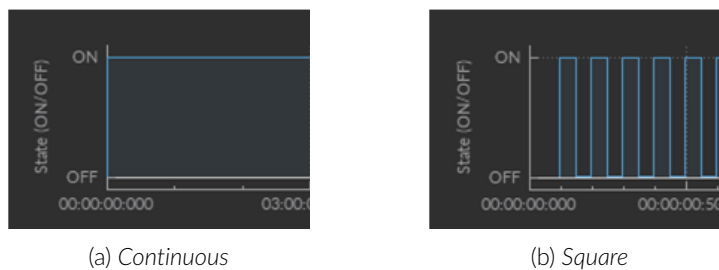

(a) *Continuous*   (b) *Square*

Figure 3.21: *Channel Options - Output Modes*

Trigger Options:

1. The **Source** trigger option (Fig 3.20, 4) allows the choice of a **Manual Trigger** (activated by a user) or an **Input** trigger, coming from a **Digital I/O** channel set in input mode.

2. The **Mode** (Fig 3.20, 5) defines how the trigger activates a sequence. This includes input sequences, which can be triggered/gated by an outside source.

   - In **Triggered** mode (Fig. 3.22a), the sequence is started manually or by a trigger source from another digital input channel. Once the trigger source is received, the sequence will continue until the end or until **Stop** is pressed.
   - In **Gated** mode (Fig. 3.22b), the sequence will start once the voltage reach a high TTL signal (4 V or more) on the input modulation BNC. When the TTL signal reaches a low TTL signal (0.4 V or less), the sequence stops and waits for another high TTL signal to continue. This mode can cut pulses, once the high signal returns. ***ONLY AVAILABLE FOR SQUARE CHANNEL MODE***
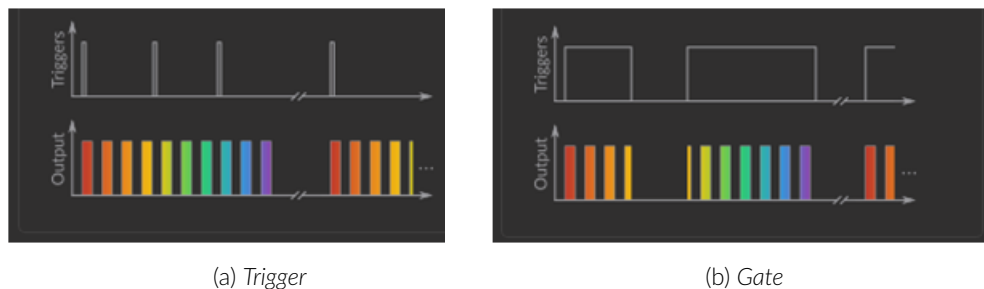

(a) *Trigger*   (b) *Gate*

Figure 3.22: *Trigger Options Modes*
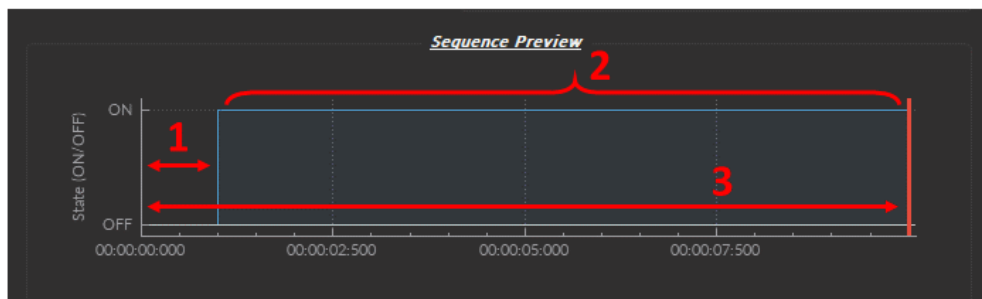
3.4.4.2   Sequence Options & Preview

The **Sequence options** section (Fig. 3.23a) contains the TTL pulse sequence parameters, while the **Sequence Preview** section (Fig. 3.23b) displays the corresponding shape and timing of the sequence. Should a parameter chosen be impossible to apply to a sequence (for example, a **Time ON** greater than 1/**Frequency**), the color of the option boxes will turn **RED**.

The parameters contained in the **Sequence Options** depend on the **Channel Mode** (selected in **Channel Options**, Fig. 3.20), as following:

- The **CW (Continuous Wave)** channel mode (Fig. 3.19) allows the creation of a continuous TTL pulse sequence. The following elements appear in the **Sequence Options** box.

  1. The **Starting Delay** (Fig 3.23, 1) defines the time between the activation of the pulse sequence and the beginning of the signal.

  2. The **Time ON** (Fig 3.23, 2) defines the length of time the continuous signal is active. Should the time chosen be 0, the signal will continue until the pulse sequence is stopped manually.

  3. The **Total Duration** (Fig 3.23, 3) shows the total expected duration of the pulse sequence. Should the duration be infinite, the box will display ∞. If there is an error in parameter selection, this box will display **N/A**.
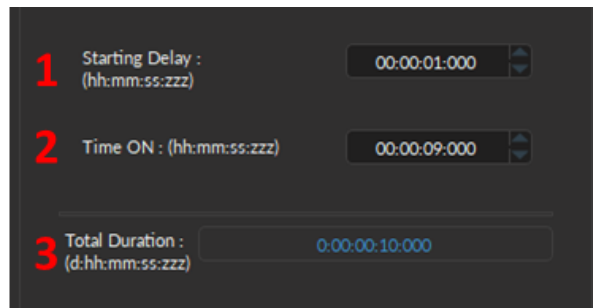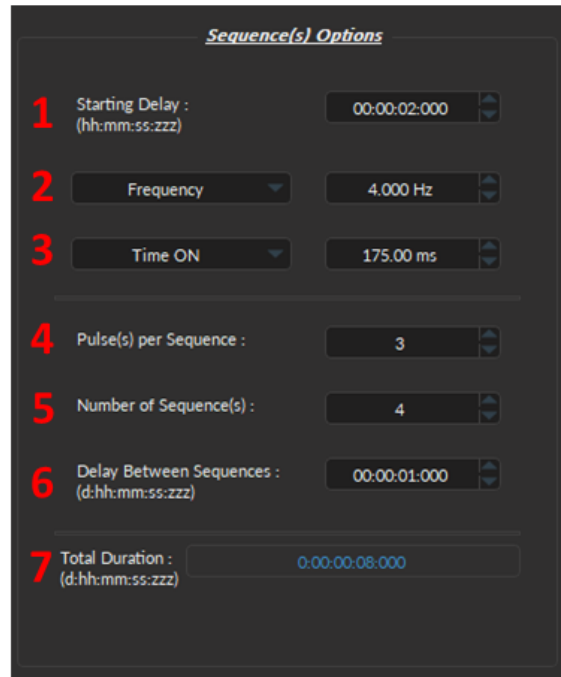

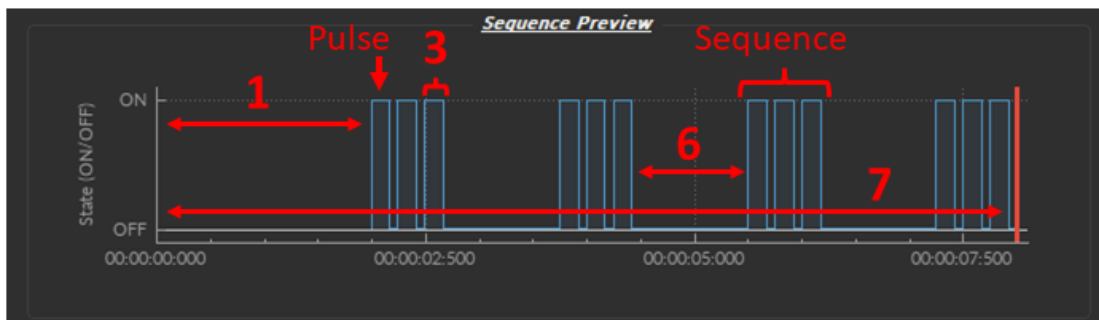(a) *Sequence Options*

(b) *Sequence Preview*

Figure 3.23: *Channel(s) configuration window, Digital I/O - CW Mode*

- The **Square** channel mode (Fig. 3.20) allows the creation of a square TTL pulse sequence. This includes all sequence options as the **CW** mode (Fig. 3.23, 1-3), with the following additions:

  1. The **Starting Delay** (Fig 3.23, 1) defines the time between the activation of the pulse sequence and the beginning of the signal.

  2. The **Frequency** (Fig. 3.24a, 2) sets the frequency (in Hz), which is the number of pulses per second. The frequency can also be changed to the **Period** (Fig. 3.24a, 2). For example, a signal at 10 Hz (frequency) will output one pulse every 100 ms (period), whereas a signal at 0.5 Hz (frequency) will output one pulse every 2 seconds (period).

3. The **Time ON** (Fig. 3.24, 3) defines the length of a single pulse. This time can also be converted to a **Duty Cycle**, which indicates the % of the period the pulse duration corresponds to.

4. The **Pulse(s) per sequence** (Fig. 3.24, 4) sets the number of pulses within a single sequence. If it is set to 0, the number of pulses will be infinite.

5. The **Number of sequence(s)** (Fig. 3.24, 5) sets the number of times that the sequence will be repeated.

6. The **Delay between sequences** (Fig. 3.24, 6) sets the amount of time separating any two sequence (excluding the **Starting Delay**).

7. The **Total Duration** (Fig 3.23, 3) shows the total expected duration of the pulse sequence. Should the duration be infinite, the box will display ∞. If there is an error in parameter selection, this box will display **N/A**.

(a) *Sequence Options*

(b) *Sequence Preview*

Figure 3.24: *Channel(s) configuration window, Digital I/O - Square Mode*
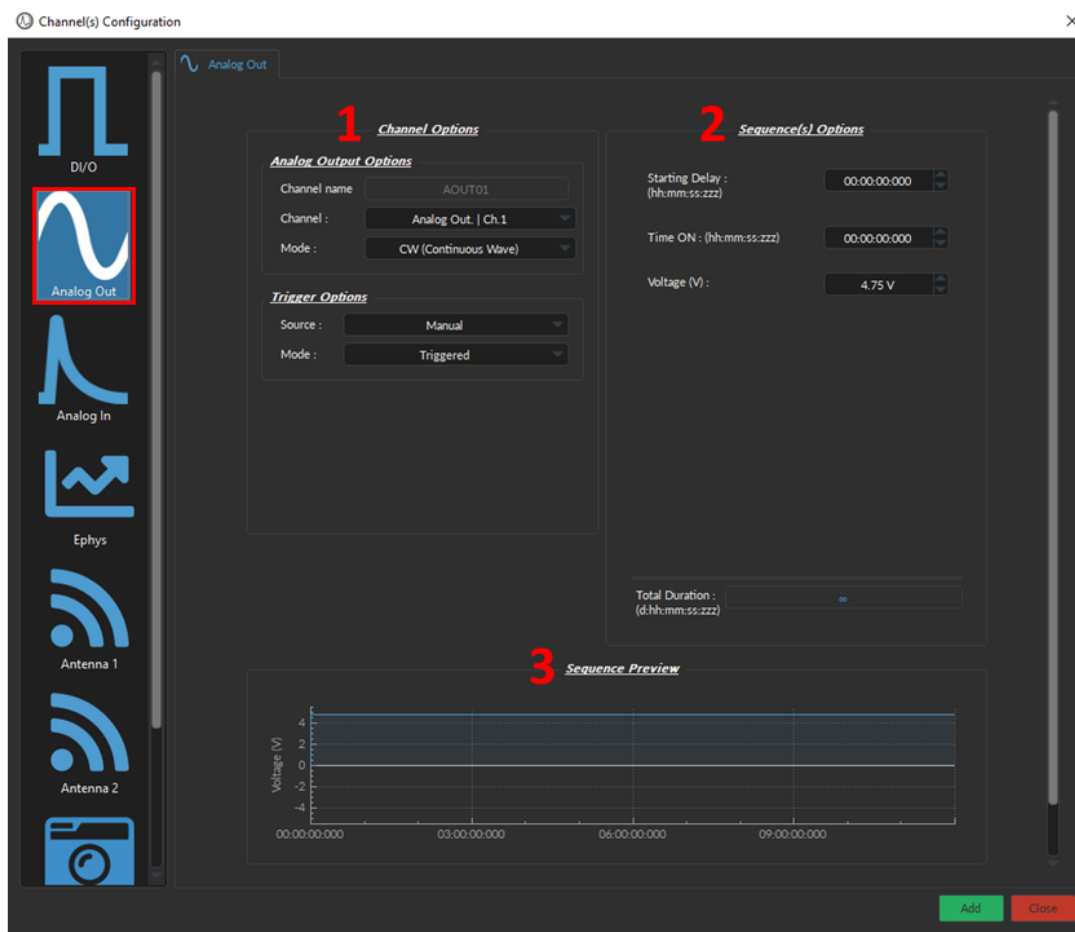
3.4.5 Analog Output

Figure 3.25: *Channel(s) configuration window, Analog Output CW*

The **Analog Output** channel type creates analog pulse sequences. Each numbered channel corresponds to the same analog channel number on the console. Pulse sequences have different parameters depending on the channel **Mode**, which can be **Continuous**, **Square**, **Sine**, **Stair** and **Custom** (Fig. 3.26).
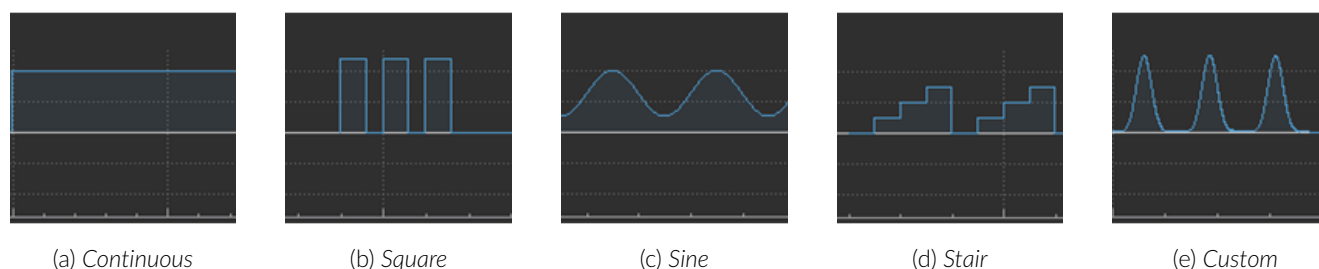

(a) *Continuous* (b) *Square* (c) *Sine* (d) *Stair* (e) *Custom*
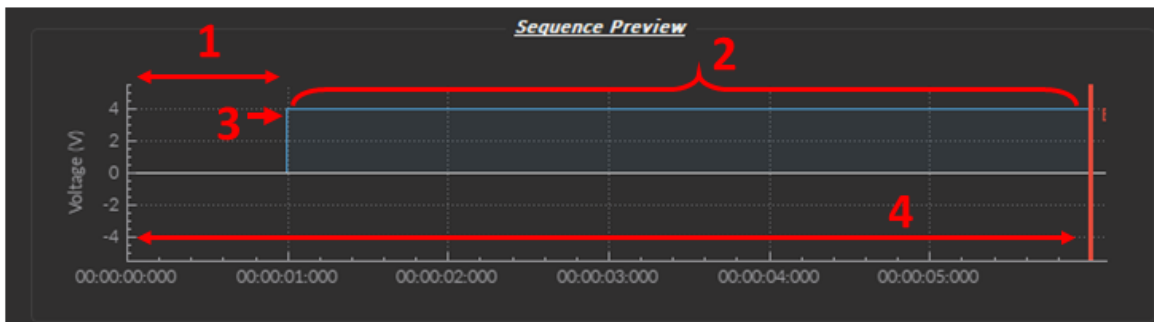
Figure 3.26: *Analog Output Modes*

3.4.5.1 Continuous Wave (CW) Mode

The **CW (Continuous wave)** channel mode (Fig. 3.27) allows the creation of a continuous analog signal. The following elements appear in the **Sequence Options** box (Fig. 3.27a).

1. The **Starting Delay** (Fig. 3.27) defines the time between the activation of the pulse sequence and the beginning of the signal.

2. The **Time ON** (Fig. 3.27) defines the length of time the continuous signal is active. Should the time chosen be 0, the signal will continue until the pulse sequence is stopped manually.

3. The **Voltage** (Fig. 3.27) defines the voltage of the continuous signal, in volts. The signal cannot go beyond ±4.75 V.

4. The **Total Duration** (Fig. 3.27) shows the total expected duration of the pulse sequence. Should the duration be infinite, the box will display ∞. If there is an error in parameter selection, this box will display **N/A**.
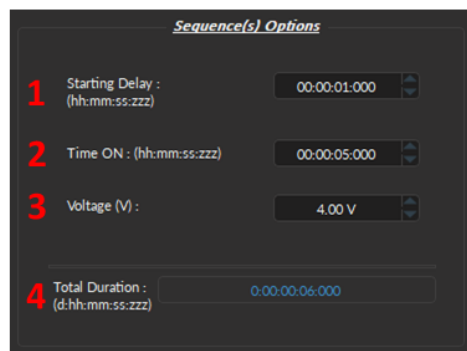
(a) *Sequence Options*

(b) *Sequence Preview*

Figure 3.27: *Channel(s) configuration window, Analog Output CW*

3.4.5.2 Square Mode

The **Square** channel mode (Fig. 3.28) creates a sequence of pulses with the minimum of the pulses at **V-** and the maximum of each pulse at **V+**.

1. The **Starting Delay** (Fig. 3.28, 1) defines the time between the activation of the pulse sequence and the beginning of the signal.

2. The **Frequency** (Fig. 3.28, 2) sets the frequency (in Hz), which is the number of pulses per second. The frequency can also be changed to the **Period**. For example, a signal at 10 Hz (frequency) will output one pulse every 100 ms (period), whereas a signal at 0.5 Hz (frequency) will output one pulse every 2 seconds (period).

3. The **Time ON** (Fig. 3.28, 3) defines the length of a single pulse. This time can also be converted to a **Duty Cycle**, which indicates the % of the period the pulse duration corresponds to.

4. The **Maximum Voltage (V+)** (Fig. 3.28, 4) defines the maximum voltage of each pulse, in volts. The signal cannot go beyond +4.75 V.

5. The **Minimum Voltage (V-)** (Fig. 3.28, 5) defines the minimum voltage of each pulse, in volts. The signal cannot go below -4.75 V.

6. The **Pulse(s) per sequence** (Fig. 3.28, 6) set the number of pulses per sequence. If it is set to 0, the number of pulses will be infinite.

7. The **Number of sequence(s)** (Fig. 3.28, 7) sets the number of times that the sequence will be repeated.

8. The **Delay between sequences** (Fig. 3.28, 8) sets the delay between each sequence.

9. The **Total Duration** (Fig. 3.28, 9) shows the total expected duration of the pulse sequence. Should the duration be infinite, the box will display ∞. If there is an error in parameter selection, this box will display **N/A**.

(a) *Sequence Options*
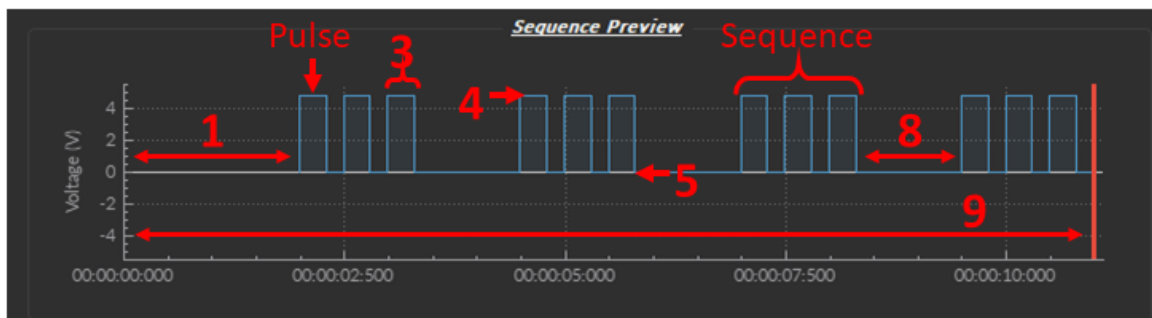
(b) *Sequence Preview*

Figure 3.28: *Channel(s) configuration window, Analog Output Square*

3.4.5.3 Sine Mode

The **Sine** mode (Fig. 3.29) creates a sinusoidal pulse sequence with peaks at **V+** and **V-**.

> **Note:** The **Starting Delay** is not available for this mode (Fig. 3.29a).

1. The **Frequency** (Fig. 3.29, 2) sets the frequency (in Hz), which is the number of pulses per second. The frequency can also be changed to the **Period**. For example, a signal at 10 Hz (frequency) will output one pulse every 100 ms (period), whereas a signal at 0.5 Hz (frequency) will output one pulse every 2 seconds (period).

2. The **Phase** option (Fig. 3.29, 2) replaced **Time ON** (Fig. 3.28, 3). This allows the choice of the sine wave phase, in degrees.

3. The **Maximum Voltage (V+)** (Fig. 3.29, 4) defines the maximum voltage of each pulse, in volts. The signal cannot go beyond +4.75 V.

4. The **Minimum Voltage (V-)** (Fig. 3.29, 5) defines the minimum voltage of each pulse, in volts. The signal cannot go below -4.75 V.

5. The **Period per Sequence** (Fig. 3.29, 5) is similar to the **Pulse per Sequence** parameter in Square mode (Section 3.4.5.2, Square), but where the period (a single sine wave from peak to peak, Fig. 3.29b, 1) replaces the pulse.

6. The **Number of sequence(s)** (Fig. 3.29, 7) sets the number of times that the sequence will be repeated.

7. The **Delay between sequences** (Fig. 3.29, 8) sets the delay between each sequence.

8. The **Total Duration** (Fig. 3.29, 9) shows the total expected duration of the pulse sequence. Should the duration be infinite, the box will display ∞. If there is an error in parameter selection, this box will display **N/A**.

(a) *Sequence Options*
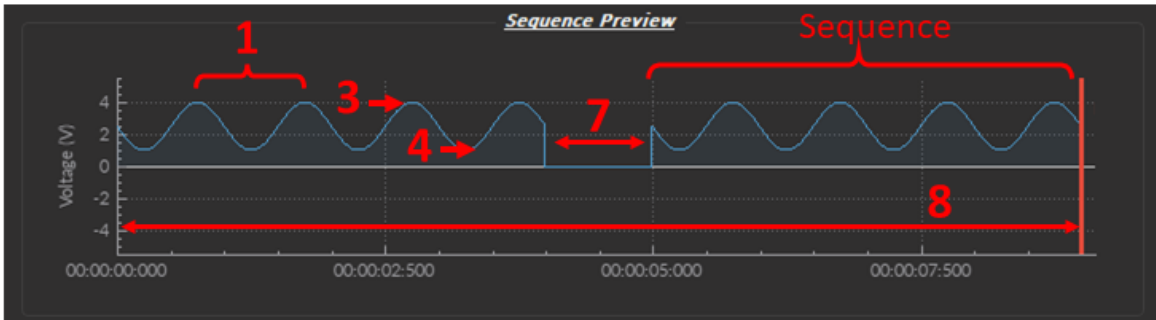
(b) *Sequence Preview*

Figure 3.29: *Channel(s) configuration window, Analog Output Sine*
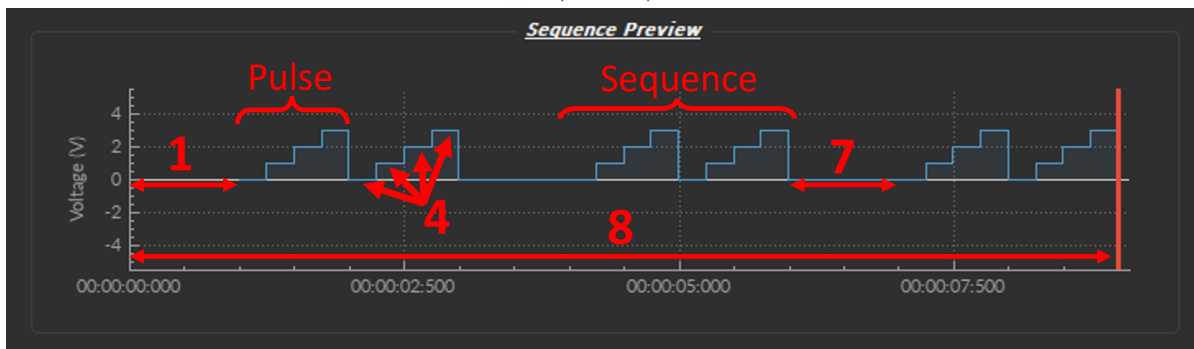
3.4.5.4 Stairs Mode

The **Stairs** mode (Fig. 3.30) creates a stepwise pulse sequence with peaks at several different Voltage levels **V+**.

1. The **Starting Delay** (Fig. 3.30, 1) defines the time between the activation of the pulse sequence and the beginning of the signal.

2. The **Frequency** (Fig. 3.30b, 2) option replaces the **Time ON** . This parameter applies to a whole pulse, which include all the voltage steps (up to a max of four).

3. The **Number of Steps** sets the amount of voltage levels of a single pulse, up to a maximum of four (Fig. 3.30b, 3). Increasing the number of steps automatically adds an additional parameter to specify the voltage of the added step below.

4. The **Step Voltage** sets the value of stair level X between **-4.75V** and **4.75V** (Fig. 3.30, 4).

5. The **Pulse(s) per sequence** (Fig. 3.30, 5) set the number of pulses per sequence. If it is set to 0, the number of pulses will be infinite.

6. The **Number of sequence(s)** (Fig. 3.30, 6) sets the number of times that the sequence will be repeated.

7. The **Delay between sequences** (Fig. 3.30, 7) sets the delay between each sequence.

8. The **Total Duration** (Fig. 3.30, 8) shows the total expected duration of the pulse sequence. Should the duration be infinite, the box will display ∞. If there is an error in parameter selection, this box will display **N/A**.

(a) *Sequence Options*
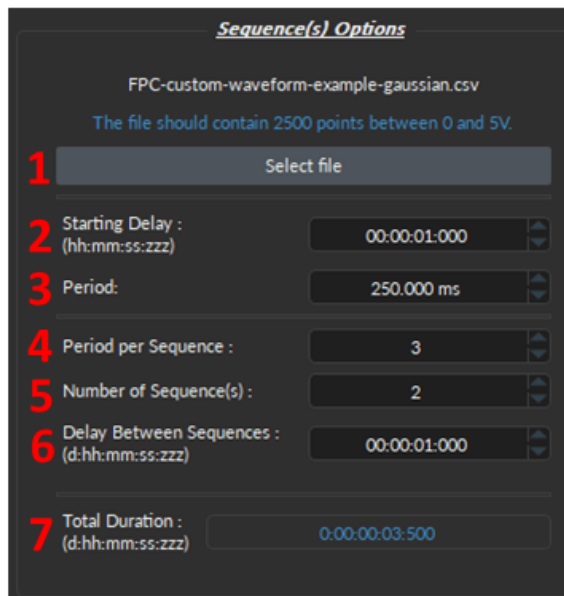

(b) *Sequence Preview*

Figure 3.30: *Channel(s) configuration window, Analog Output Stairs*

3.4.5.5 Custom Mode

The **Custom** mode (Fig. 3.31) provides users with the ability to design a pulse with non-standard shape to fit experimental needs.

1. The **Select File** button (Fig. 3.31a, 1) is used to input a custom .csv file containing the data for the pulse sequence. This must be a .csv format and requires 2500 values in column vector format (i.e. with *line break* between values), as in Fig. 3.31b. The values can be any value between **-4.75V** and **+4.75V**.

2. The **Starting Delay** (Fig. 3.31, 2) defines the time between the activation of the pulse sequence and the beginning of the signal.

3. The **Period** option (Fig. 3.31, 3) replaces the **Time ON** option (Fig. 3.28, 3). This option will stretch or shrink the 2500 value sequence to fit the specified amount of time.

4. The **Period per Sequence** (Fig. 3.31, 4) is similar to the **Pulse per Sequence** found in **Square** modes (Fig. 3.4.5.2, 6), where the pulse is replaced by the period sequence (Fig. 3.31c, Sequence).

5. The **Number of sequence(s)** (Fig. 3.31a, 5) sets the number of times that the sequence will be repeated.

6. The **Delay between sequences** (Fig. 3.31, 6) sets the delay between each sequence.

7. The **Total Duration** (Fig. 3.31, 7) shows the total expected duration of the pulse sequence. Should the duration be infinite, the box will display ∞. If there is an error in parameter selection, this box will display **N/A**.
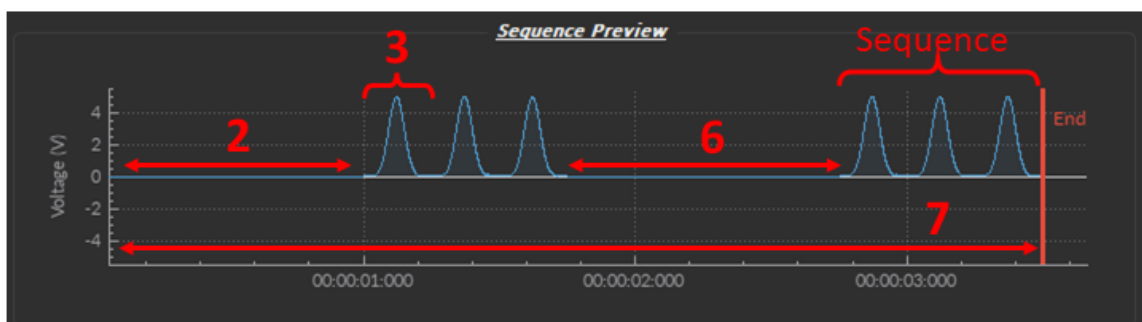
(a) *Sequence Options*

(b) *Example .csv file*

(c) *Sequence Preview*

Figure 3.31: *Channel(s) configuration window, Analog Output Custom*

3.4.6   Analog Input

The **Analog Input** channel type acquires signal from the **Analog Input** BNC connector ports. Each numbered channel corresponds to the same analog channel number on the console.

The *Channel(s) Configuration* window for the **Analog Input** is divided into two sections (Fig. 3.32): (1) the **Channel Options** (Section 3.4.6.1) and (2) the **Mode-specific Options** (*Linear*, Section 3.4.6.2; *Interleaved*, Section 3.4.6.3; *Lock-In*, Section 3.4.6.4).
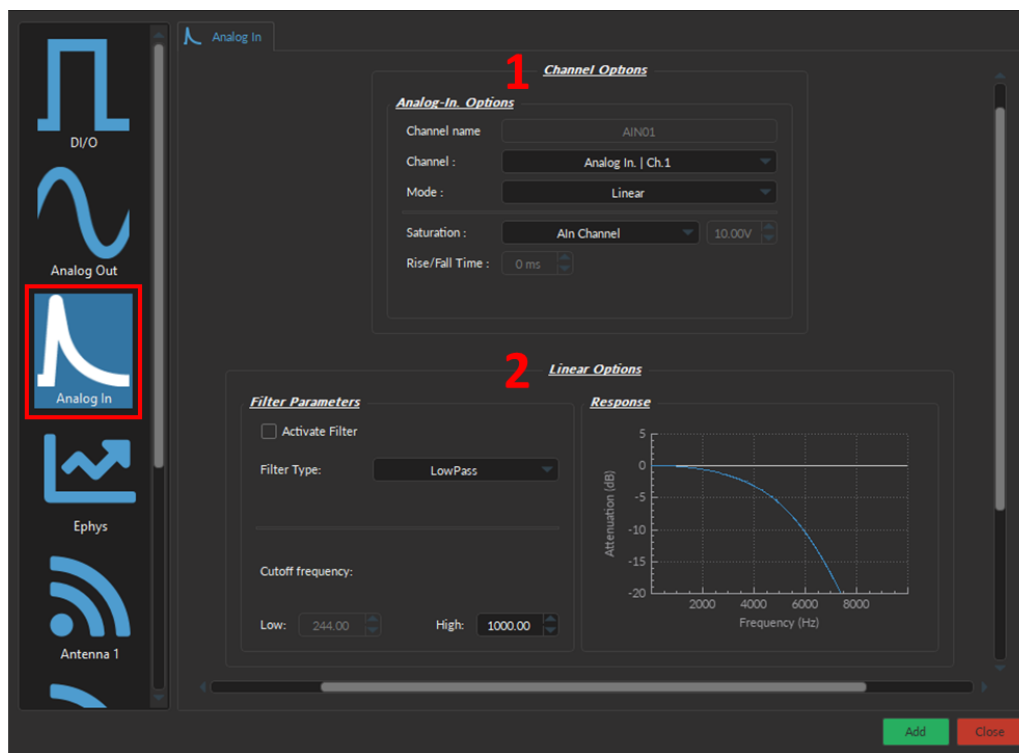

Figure 3.32: *Channel(s) configuration window, Analog Input*

3.4.6.1   Channel Options

The **Channel Options** (Fig. 3.33) defines the channel, source and mode of the digital signal, as following:
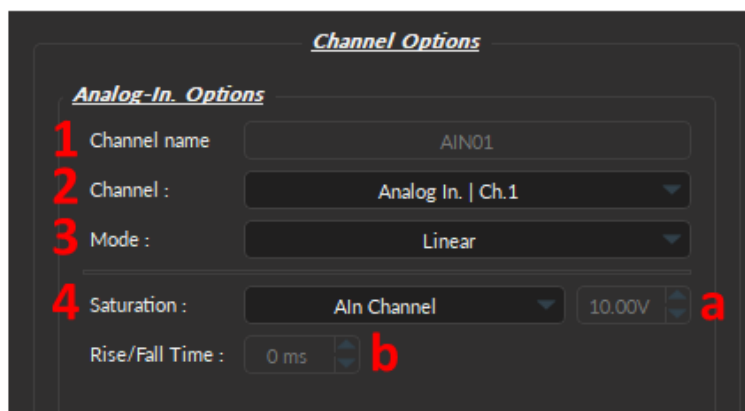

Figure 3.33: *Channel(s) configuration window, Analog Input - Channel Options*

1. The **Channel name** (Fig 3.33, 1) allows user to specify a label for each channel.

2. The **Channel** (Fig 3.33, 2) identifies which of the channels available for each channel type is currently being modified. The channel can be changed by selecting a new one from the drop-down list. Each numbered channel on the physical console corresponds to the same number of the digital channel within the software.

3. Three **Mode** are available to record the input signal, each of which have their own defined parameters in the Option box below **Channel Options** (Fig. 3.32, 3):

   - *Linear* - Section 3.4.6.2;
   - *Interleaved* - Section 3.4.6.3;
   - *Lock-In* - Section 3.4.6.4.

4. The **Saturation** (Fig. 3.33, 4) automatically sets the following parameters and depends on the detector acquiring the data (Detectors: *Doric detector*, *Newport Detector*, *Hamamatsu C10709*, and *AIn Channel*):

   a) The **Maximum Voltage** (Fig. 3.33, 4a)
   b) The **Rise/Fall Time** (Fig. 3.33, 4b)

   To manually set either parameter, select **Custom** in the drop-down menu.

Each **Analog-In Mode** has a specific set of parameters. The function of each **Mode** is described here.

3.4.6.2   Linear

The **Linear** channel mode (Fig. 3.34) allows the direct measurement of signal received by a channel. The linear mode-specific parameters are as follows:
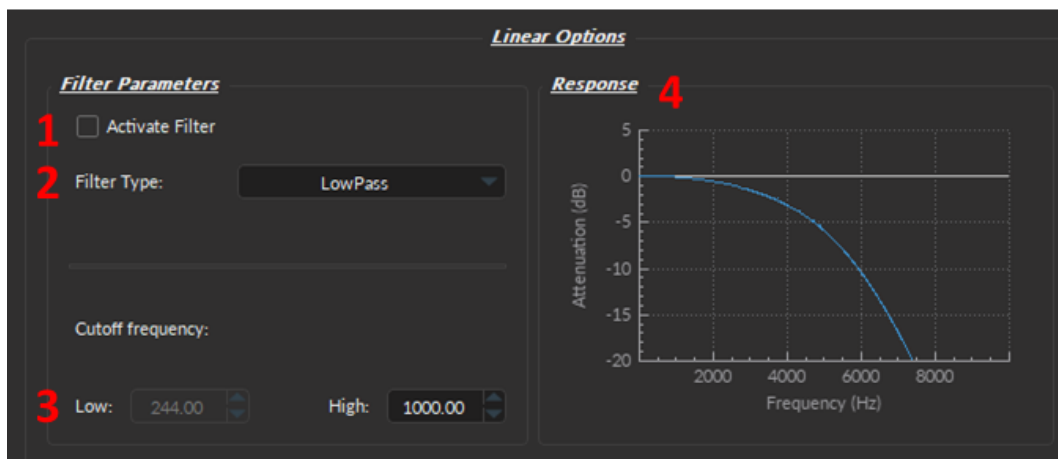

Figure 3.34: *Channel(s) configuration window, Analog Input Linear*

1. When the **Activate Filter** checkbox is selected, the defined filter is applied on all input data and displayed on a new trace. The filtered data is for display only, and will not be saved.

2. The **Filter Type** drop-down list allows the choice of a filter type from **High-Pass**, **Low-Pass**, **Band-Pass** and **Band-Stop**.

3. The **Cutoff Frequency** boxes are used to define the low/high cutoff values for the filter, depending on the type used. The cutoff frequency must be less than half of the sampling rate. Note: the true cut-off value is, by definition, always 3 dB below (Low Cutoff) or above (High Cut-off) the specified value.

4. The **Response** box displays Frequency (Hz) vs Attenuation (dB) trace of the filter according to both the filter type and the cut-off values.

3.4.6.3 Interleaved

The **Interleaved** channel mode allows two channels to send an alternating pulsed signal of opposite phase for two separate light sources. Each source can excite a different fluorophore, which allows the detection of two separate fluorescence signals coming from the same sample using a single channel (Fig. 3.35).
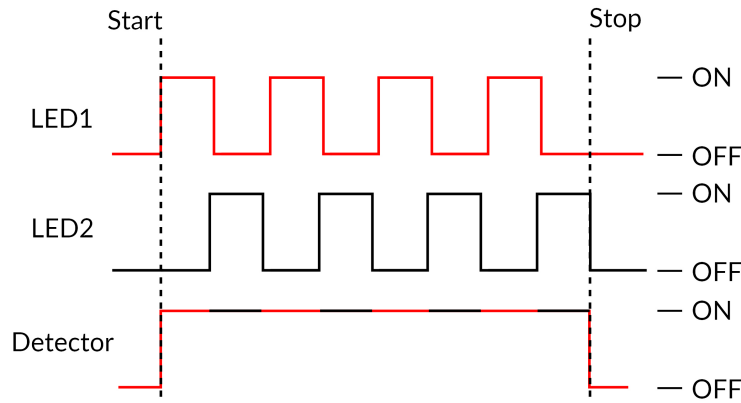

Figure 3.35: *Interleaved Acquisition Timing Diagram*

The interleave preset is using 50% duty cycle for each LED, without delay between them (Fig. 3.35). Thus, depending on the Rise/Fall time of the detector in use (Fig. 3.33, 4b, Detector Rise/Fall Time), there will be more or less crosstalk between the interleaved channels (Fig. 3.36).

> **WARNING:**
> **Crosstalk** occurs between **two interleaved** Digital I/O channels. If possible, use **Lock-In mode** instead, or **switch to a detector** will smaller Rise/Fall Time.

Specifically, when one of the digital channels is ON, it will pick up when the other is turn ON or OFF (Fig. 3.36). Figure 3.36 shows how the Digital Output channel of LED 1 has a small increase in voltage when the LED 2 is turned ON. And, conversely, there is a small dip in voltage in the LED 2 channel when LED 1 is turned OFF (Fig. 3.36).
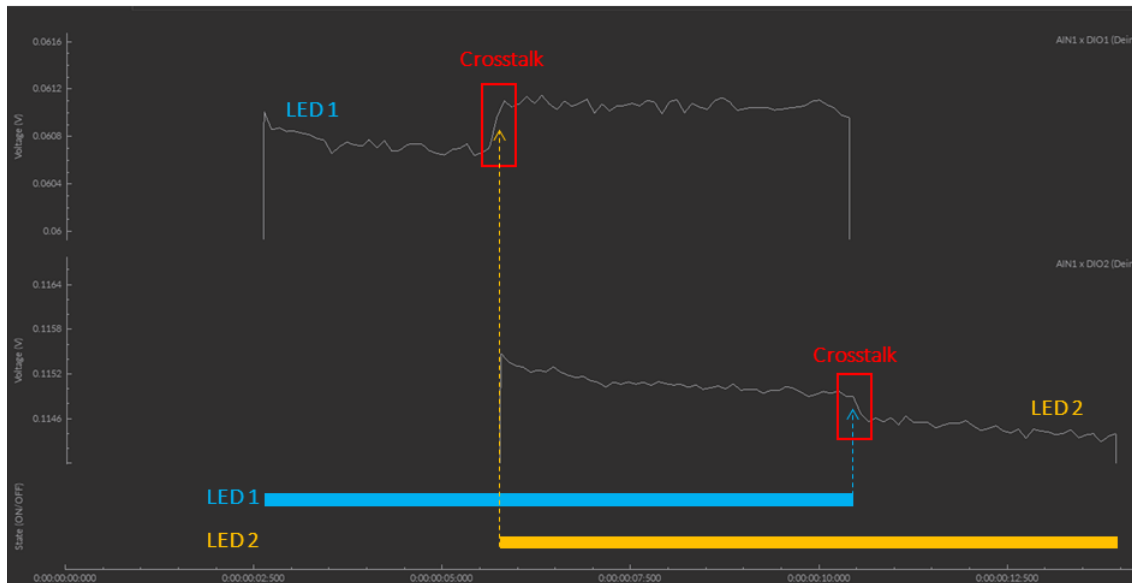

Figure 3.36: *Interleaved Cross-talk*

Strategies to Mitigate Crosstalk:

1. If the sampling rate of the triggered device(s) is high enough (>120Hz), use the **Lock-In mode** (Section 3.4.6.4) instead of the **Interleaved mode**;

2. Switching to a detector with a smaller Rise/Fall Time will reduce the crosstalk. For instance, the *Doric* and *Newport Detectors* have a Rise/Fall Time of 15 ms, while Hamamatsu C10709 has one of 1 ms.

Regardless of the Detector in use, **care should be taken not to misinterpret crosstalk as real signal during data analysis.**
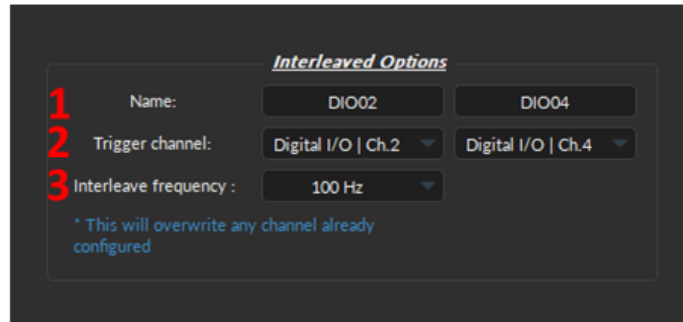

Figure 3.37: *Channel(s) configuration window, Analog Input Interleaved*

To use the **interleaved mode**, specify the parameters in the **Interleaved Options** section of the *Channel Configuration* window (Fig. 3.37):

1. The **Name** (Fig. 3.37, 1) lets users customize the label of the channel to increase clarity of the acquisition system.

2. The **Trigger channel** (Fig. 3.37, 2) drop-down list allows the choice of interleaved outputs (can be either digital or analog). However, once the first channel is selected, the user will only be allowed to select the same type of output (analog or digital) for the second channel.

3. The **Interleave frequency** (Fig. 3.37, 3) drop-down list allows the choice of a pre-configured frequency (either 10, 20, 50 or 100Hz) for the interleaved channels. The two selected trigger channels will be configured to function at the chosen frequency.

> ⚠️ **WARNING:**
> Specifying the interleave frequency will **overwrite** any channel already configured. ⚠️

3.4.6.4   Lock-In

The **Lock-In** mode can be used to detect fluorescence signals embedded in strong noise (e.g. Isosbestic and a fluorophore) or to separate multiple signals (e.g. Green and red fluorophores) during fiber photometry.

Each *LED light source* emits a sinusoidal illumination at a given frequency (Fig. 3.38a & 3.38b). The detector collects the fluorescent data at a frequency corresponding to the summation of the LED frequencies (Fig. 3.38c).
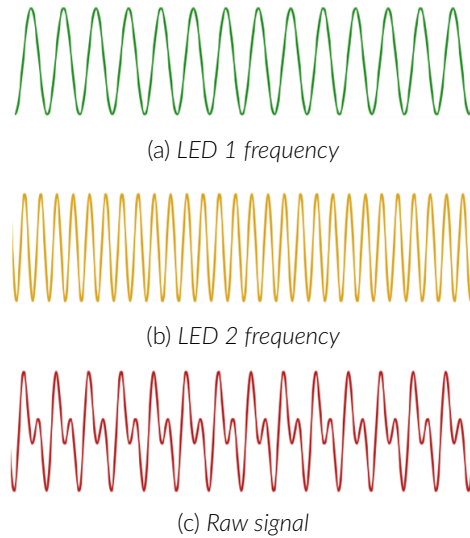

(a) *LED 1 frequency*

(b) *LED 2 frequency*

(c) *Raw signal*

Figure 3.38: *Lock-In Acquisition Timing Diagram*

The amplitude changes of the raw signal are due to the collected fluorescence and are dependent of the frequency (Fig. 3.39a). By targeting the known LED frequencies in the raw signal using filters, it is possible to demodulate the fluorescence based on the emission wavelength (Fig. 3.39). The result is separated from the ambient noise that occurred at different frequencies (Fig. 3.39b). The same principle can be applied to demodulate two fluorescent signals.
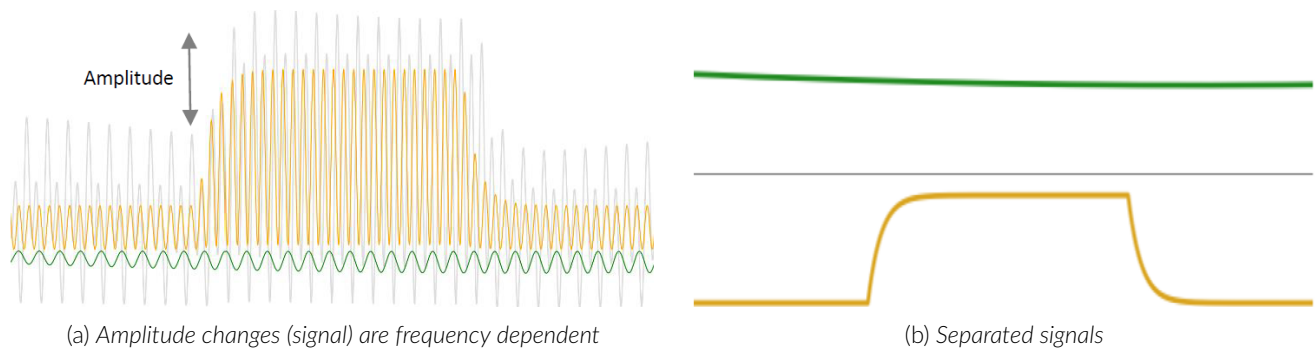

(a) *Amplitude changes (signal) are frequency dependent*

(b) *Separated signals*

Figure 3.39: *Demodulation separates noise from signal or two signals from each other*

WARNING:

To properly set-up the Lock-In mode, users must have a complete understanding of the wiring of inputs and outputs of they photometry system.

The **Lock-In Mode** parameters are as follows (Fig. 3.40):

1. The **Enable** (Fig. 3.40, 1) row lets users select which output channel to include in the Lock-In settings by clicking the respective check boxes. Each column corresponds to an Analog Out channel of the console (in order, such

that left most column = AOUT1). Users should enabled the output(s) channels that will be driving the input specified in Fig. 3.33, 2.

2. **Trace Name** (Fig. 3.40, 2) is the identity of the Input and Output Channel(s) enabled for this Lock-In configuration.

- The **AIN #** corresponds the console Analog In port number that receives the raw (non-demodulated) signal from the detector. To change the **AIN #**, select a different **Channel** number from the **Analog In Options** box (Fig. 3.33).

- The **AOUT #** number corresponds the the Analog Out port on the console that sends eletrical information (including the reference frequency) to the *LED Driver*. While you cannot change the **AOUT #** since it is native to each column of the **Carrier Frequency Options** (Fig. 3.40), changing which port is enabled using the checkbox (Fig. 3.40) or physically moving the cable to a different port on the console allows user to specify the connection of the output.

3. **Reference Frequency** (Fig. 3.40, 4) is the oscillating trigger signal that drives the LED (or device(s) of choice). We recommend using the default values since they are optimised for fiber photometry. But, if modified, frequencies will be re-adjusted in steps of 5.96 Hz. In addition, the reference frequency should not be a multiple a of known noise frequency (e.g. 50 and 60 Hz), or a multiple of another reference frequency.

4. **LED maximum current** (Fig. 3.40, 5) is the largest current that the LED can handle. This value should be set either in low power mode (recommend) or based on the intrinsic maximum current of the LED in use (500 mA or 1000 mA, depending of the type of LED).

- **Low Power Mode (200 mA)** - allows reduced power for the same voltage. This allows low-power signals to be more stable in time. The **maximal current** is reduced to one tenth of light source normal maximal current. For example, a driver with a normal maximum current of 2000 mA for a 5 V signal (400 mA/V) will have a maximum current of 200 mA for a 5 V signal (40 mA/V).
*Recommended for Fiber Photometry using *Doric FMC or RFMC* systems*

- **500 mA** - the **LED maximum current** for the following LEDs: 365 nm, 385 nm & 405 nm.

- **1000 mA** - the **LED maximum current** for most *Doric* LEDs, except the three mentioned above.

- **Custom** - this setting allows users to manually adjust the Vmax and Vmin of the LED, regardless of LED maximum current. Care should be taken to remain below the maximum voltage, or the excitation signal will be cropped at the true maximum value (see Fig. 3.41).
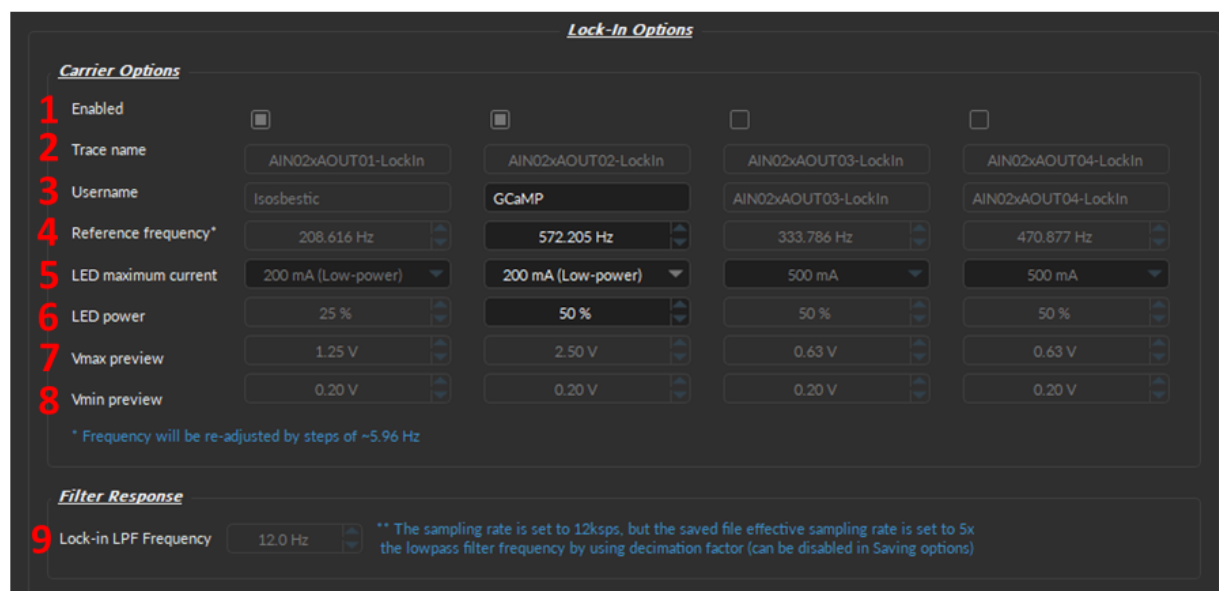

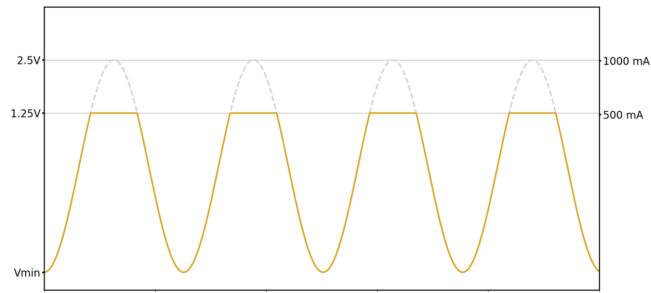Figure 3.40: *Channel(s) configuration window, Analog Input Lock-In*

Figure 3.41: *Cropped LED excitation signal*

5. **LED power** (Fig. 3.40, 6) is the percentage of maximum current (converted to voltage) that will be used as **Vmax** and in **External Mode** during the recording, since the LED driver outputs a current proportional to the voltage with a conversion factor of 400 mA/V in standard operation mode, and 40 mA/V in low-power mode.

   - **Note:** The **LED current** should always be set to its maximum on the *LED driver* (and in **External Mode**), while increasing or decreasing **Vmax** should always be done by changing the *FP console* **LED power**.

6. **Vmax Preview** - automatically displays the maximum voltage based on the **LED maximum current** and the **LED power** selected above (Fig. 3.40, 7). **Vmax** can be changed if the **Custom** LED maxiumum current mode is selected. The **Vmax** should never be below 0.3 V, nor above 4.7 V.

   - **Note:** If you are using GCaMP and its isosbestic, we recommend that the isosbestic demodulated trace be about half the power of the GCaMP demodulated trace to reduce the risk of photobleaching (as in Fig. 3.40, 6).

7. **Vmin Preview** (Fig. 3.40, 8) - the default value is set to 0.2 V, but can be changed if the **Custom** LED maxiumum current mode is selected. The **Vmin** should never be below 0.1 V.

8. **Lock-in LPF Frequency** (Fig. 3.40, 9) define the **Cutoff Frequency** of the low-pass filter that extracts the signal and is set to 12 ksps by default. This value was selected because in photometry experiments, the greatest source of noise to the filter is around the carrier frequency above 200 Hz. Thus, with the current filtering algorithm, a cutoff frequency of 12 Hz (corresponding to a decimation factor of 200x) gives the best filtering results.

   - **Note 1:** The saved file **effective sampling rate** is set to 5x the lowpass filter frequency, using a decimation factor (which can be disabled in Saving Options, see Fig. 3.42).

   - **Note 2:** The **Cutoff Frequency** (the frequency at which a -3 dB attenuation will occur) should be chosen as a value close to that of the phenomena observed. A lower cutoff frequency may not result in smaller noise figures.
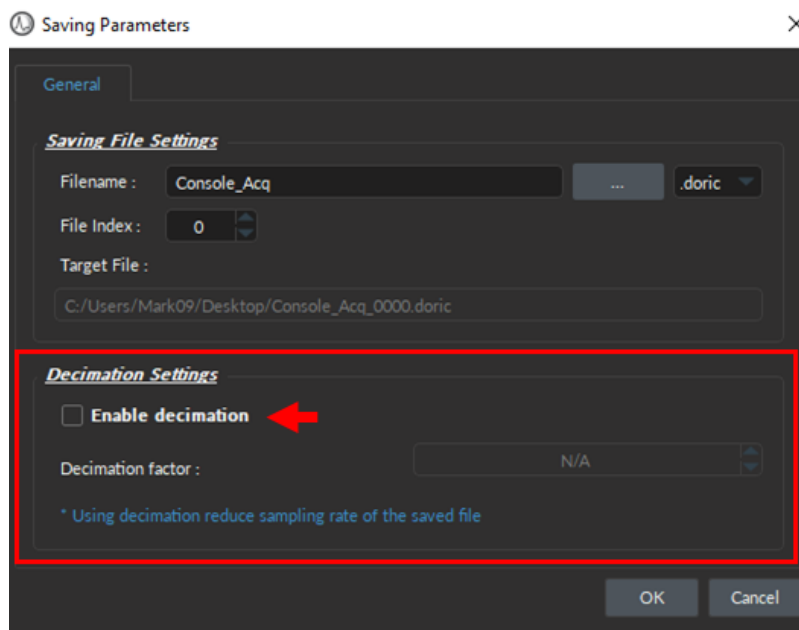
Figure 3.42: *Enable/Disable the decimation factor that reduces Sampling Rate of the saved files*

3.4.7 Camera Channel

It is natural to pair Doric neural recordings with behaviors. Many behaviors, especially freely moving behaviors, require camera inputs for its measurement.
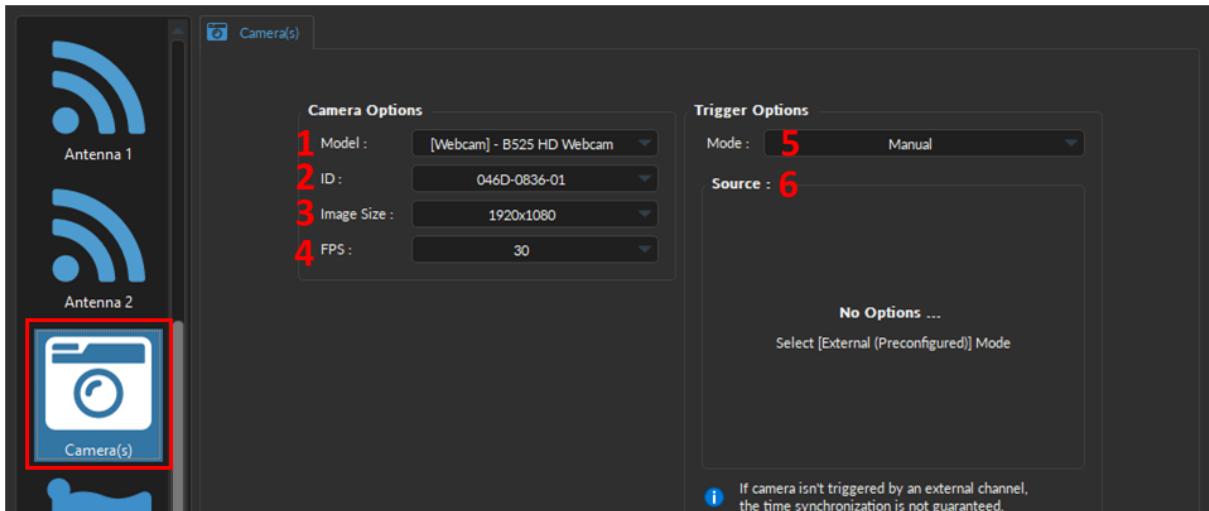

Figure 3.43: *Channel(s) configuration window, Camera*

> **WARNING:**
> A camera cannot be used for <u>BOTH</u> **Acquisition Console** and **Camera** modules.
> When creating a Camera Channel, if *"No available camera detected..."*, <u>disconnect</u>
> the camera in the **Device Selection** window to close the extra module.

Camera Options:

1. The **Model** (Fig. 3.43, 1) allows you to select the camera of choice based on the type of camera.

2. **ID** drop-down list (Fig. 3.43, 2) is used to select a camera based on its unique ID. The ID is particularly useful if multiple cameras of the same model are required for the experiment.

3. The **Image Size** (Fig. 3.43, 3) is used to set the resolution of the image. The large the number of pixels used for width x height, the better the resolution. Currently, image size can ranges between 160x120 to 1920x1080 pixels.

4. The **FPS** (Fig. 3.43, 4) is used to specify the frame rate of the camera (i.e. the number of images displayed per second). FPS can be any value between from 5-30 for web cameras and up to 60 FPS for the *Doric Behavior Camera*.

Trigger Options:

5. The **Mode** (Fig. 3.43, 5) sets the type of trigger that will control the camera. Depending on the type of camera, at most three modes are available:

> **WARNING:**
> If the camera isn't triggered by an external channel,
> the **time synchronization is NOT guaranteed**.

- **Manual** - Selecting the *Live* or *Record* buttons located in the Acquisition Tab will the trigger the start of the camera recording. ***The time difference between the actual start time and when the first frame is received depends on the camera itself.*** Around a 1 second delay is pretty common for web cameras.

The time delay (in ms) between the photometry and video data is recorded in the *DifferenceMasterStart-ToFirstImage* attribute, located in *.doric* file under the **Web Camera ID** folder (Fig. 3.44). This attribute can be used to retroactively align the video and fiber photometry data during analysis.
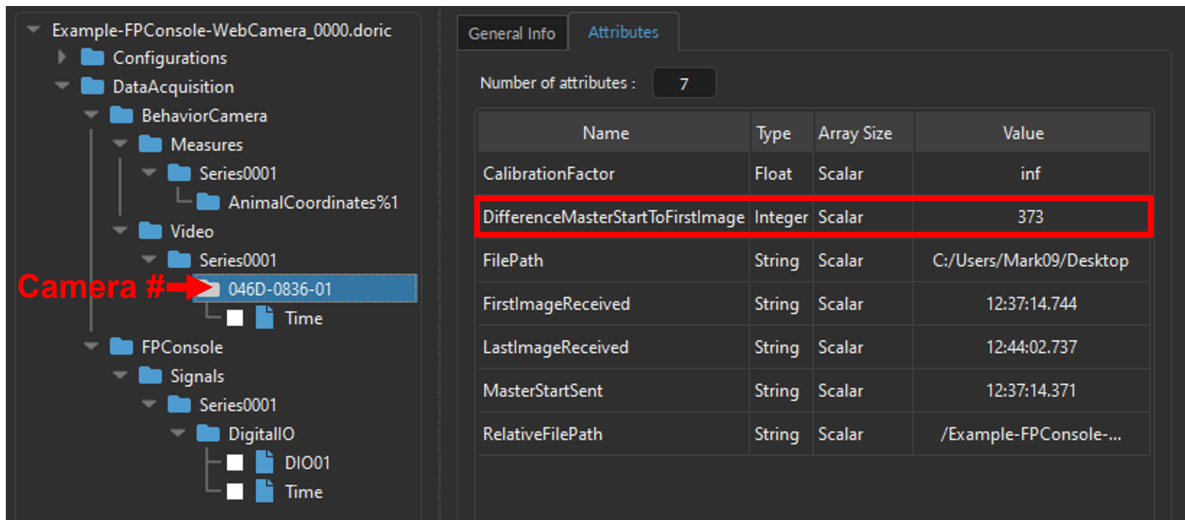

Figure 3.44: *Doric File Viewer, Web Camera Attributes - Video Alignment Variable*

- **External** - Will drive the camera using external TTL signal through the trigger cable (Frequency: 30 Hz (or camera FPS); Time ON: 5 ms). This signal can come from any external device connected to the opposite end of the trigger cable. If using *Doric Neuroscience Studio* to synchronize the recording, use *External (Preconfigured)* mode below instead. ***ONLY offered for the *Doric Behavior Camera*.***

- **External (Preconfigured)** - This is the recommended mode to synchronize the camera with the rest of the Acquisition system. This mode automatically creates an additional Digital I/O channel configured to drive the camera at the proper frequency and Time ON. ***ONLY offered for the *Doric Behavior Camera*.***

6. The **Source** (Fig. 3.43, 6 & Fig. 3.45) is only used for the **External (Preconfigured)** mode, and displays the **Digital I/O** channel with the preconfigured parameters that will be created at the same time as the **Camera Channel** (Fig. 3.45). For detailed description of each Digital I/O parameter see the corresponding section in the Fiber Photometry System Manuel. Briefly, key parameters include:

   a) The **Channel** (Fig. 3.45, a) corresponds to the physical Digital I/O channel number on the Console that is connected to the trigger cable of the *Doric Behavior Camera*.

   b) The **Mode** (Fig. 3.45, b) is by default set to the *Square (TTL)* which provides the external trigger signal to the camera. This parameter cannot be changed.

   c) The **Frequency** (Fig. 3.45, c) corresponds to the **FPS** set in the **Camera Options**. Changing the **FPS** will automatically change the **Frequency** in the **Sequence(s) Options**.

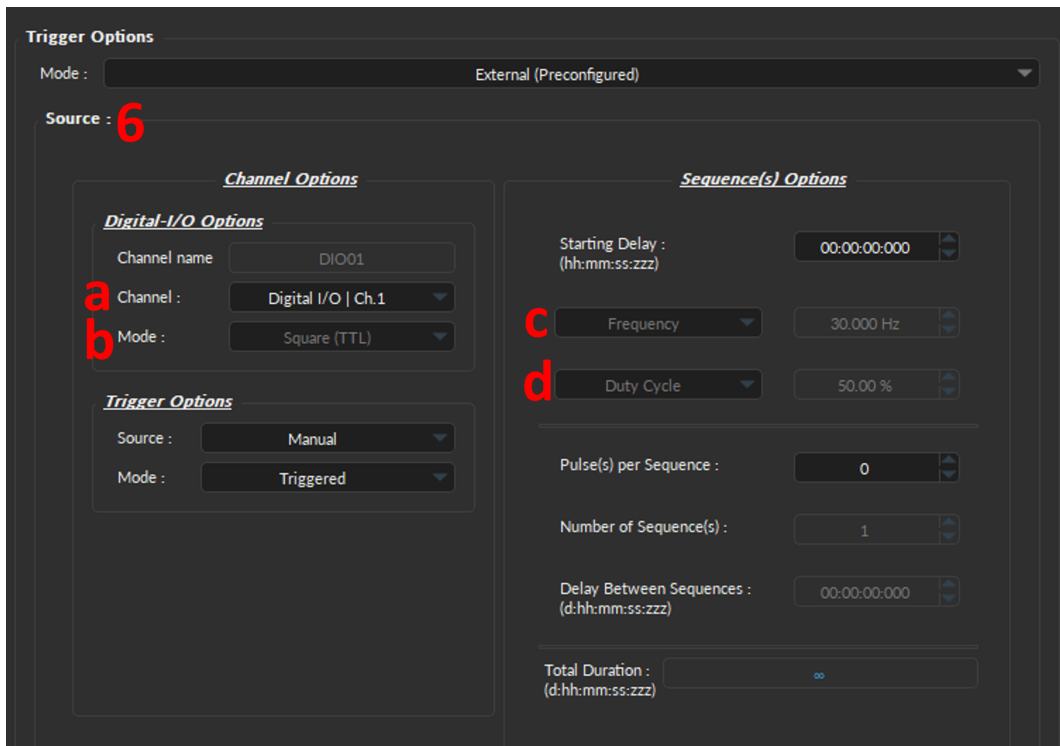   d) The **Duty Cycle** (Fig. 3.45, d) is by default 50%. The frame will be taken at the start of each square pulse.

Figure 3.45: *Channel(s) configuration window, Camera - External (Preconfigured)*

3.4.8 KeyPress Event(s)

Keypress Event(s) are ideal when manually labelling or annotating events during experiments. Specifically, selecting any keyboard key during a recording will save the output synchronized to other measurements. Keypress events can be used to:

- Flag disruptions during the experiment, such as lights on, door opened, construction noise, etc.

- Record experimentally relevant events/stimuli, such as airpuff, licks or any other behavior.

WARNING:
Keyboard event(s) timing are **accurate within 1 second** due to variations in Windows priority management and buffering of the signals.

3.4.8.1 Adding/Removing KeyPress Event(s)

To add a new **Keypress Event**, select the + sign at the botton of the window (Fig. 3.46, left). To remove a KeyPress, use - button (Fig. 3.46, right).

- **NOTE:** Selecting the + button (without clicking the *Add* button or the *Close* of the *Channel Configuration* window) will **automatically** add the Keypress Event channel at the **bottom** of the Acquisition View window, below any pre-existing channels (Fig.3.46).
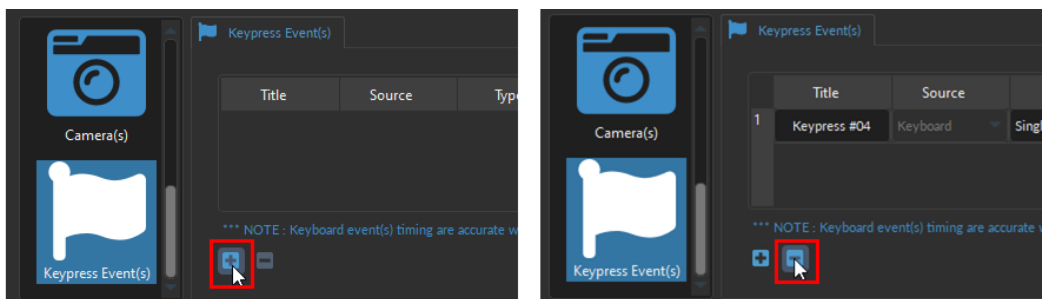

Figure 3.46: *Adding and Removing Keypress Events*

To edit a pre-existing **Keypress Event** Channel, select the left button (Fig. 3.47) in the **Acquisition View**.
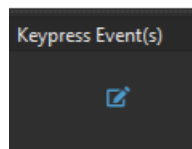

Figure 3.47: *Edit Keypress Event(s) Channel*

The following are the configurable parameters of a **Keypress Event,** per Fig. 3.49:

1. The **Title** allows you to give a name for the Keypress event.

2. The **Source** is by default *Keyboard*.

3. Three **Types** of Keypress Event(s) can be specified with the drop-down list:

   - **Single** - Records single event at the touch of a key (Fig. 3.48a).
   - **Toggled** - Records the start and end of an event using the same key. First press denotes the start of the event while a second press denotes the end of it (Fig. 3.48b).
   - **Timed** - Records an event for a predetermined duration of time (Fig. 3.48c). Every keypress is a new event, with the start of the event occuring when the key was depressed.
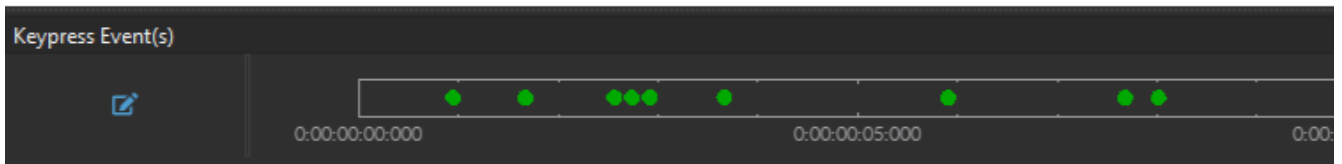
(a) *Single*
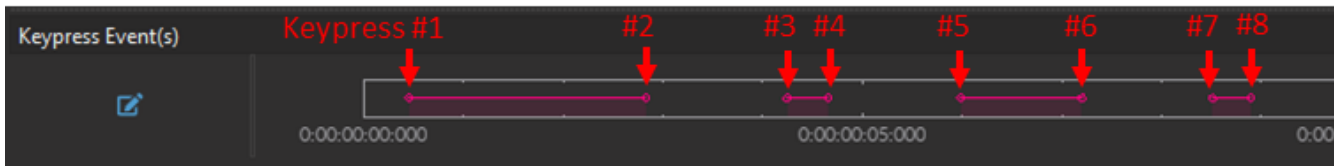
(b) *Toggled*
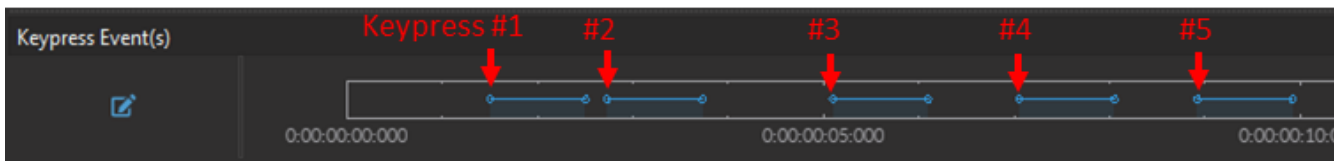
(c) *Timed*

Figure 3.48: *Three types of Keypress Event(s)*

4. The **Duration** is only used for the **Timed** Keypress type to specify the predetermined amount of time a Keypress Event will span. The duration is set in hh:mm:ss:zzz.

5. Select the **Color** field to open the **Select Color** window. Basic colors are provided, in addition to custom colors can be created and stored.

6. The **Shortcut Key(s)** can be any keyboard key, including space bar, enter, backspace, any letters, number and special characters (*, !, ? etc.). To specify the key, click inside the *Shortcut Key(s)* cell, then press the keyboard key of choice. If a key was properly set, it will appear in the *Shortcut Key(s)* cell (as in Fig. 3.49, column 6).

7. The **Information** column provides space to make notes or write a short description of the Keypress Event.
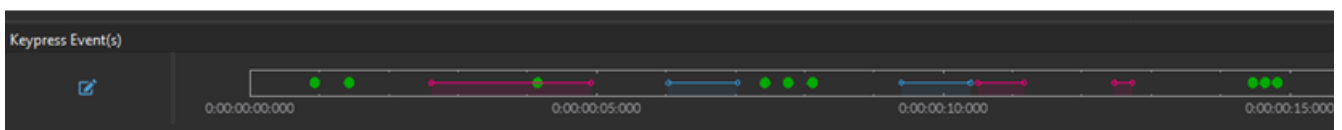

Figure 3.49: *Channel(s) configuration window, KeyPress Event(s)*

3.5 Acquisition View

The **Acquisition View** displays all the information concerning active channels: **Control box** (Fig.3.50a) and the **Graphs** (Fig.3.50b).

If neither **Control Box** nor **Graphs** are displayed in the **Acquisition View**, this means channels have yet to be configured. User can either use the **Load Configuration** button (see Section 3.3.2) to load a *.doric* file with previously saved channel parameters, or user can manually add channels using the **Add Channel** button (see Section 3.3.2).

(a) *Channels*   (b) *Common parameters*

Figure 3.50: *Acquisition View*

3.5.1 Channel Control Box

Each channel **Control box** shows the following basic elements (Fig.3.50b), with additional elements available for specific channel types:

1. The **Channel name** (Fig. 3.50b, 1) is located on the upper left of the **Control box**, identifying the type of channel and it's number, corresponding to that on the console. This name can be modified in the **Graph options** window (Fig. 3.51).

2. The **Edit** button (Fig. 3.50b, 2) opens the **Channel Configuration** window, were channel parameters can be modified (See section 3.4.1).

3. The **Graph(s)** (Fig. 3.50b, 3) button opens the **Graph Options** window (Fig. 3.51) corresponding to the channel whose graph will be modified. This window allows users to configure visualization and naming parameters of each channel graph (Fig. 3.51). If a channel has multiple traces, parameters to configure each trace individually will appear automatically on different rows (Fig. 3.51). **Graph(s) Options** parameters (Fig. 3.51) are as follows:
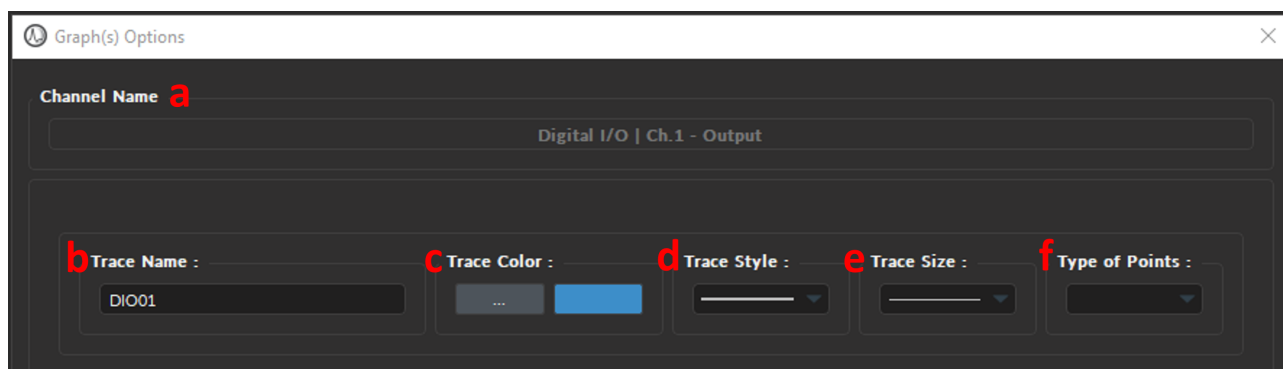
Figure 3.51: *Graph(s) Options Window*

a) The **Channel Name** (Fig. 3.51, 1) is the default name assigned by the software, which includes the type of channel (Digital / Analog In or Out) and the location of said channel on the console (BNC connector 1-4).

b) The **Trace Name** text-box (Fig. 3.51, 2) allows users to specify a name for the trace, instead of the default name generated by the software.

c) The **Trace Color** button (**...**) (Fig. 3.51, 3) opens the **Color Select** window (Fig. 3.52), which allows the selection of a trace color from a wide palette. The **Pick screen color** in this window allows the selection of any color displayed on the computer screen.

d) The **Trace style** drop-down list (Fig. 3.51, 4) allows the selection of the type of trace, from full to dashed lines. If the style chosen is empty, the trace will not be displayed.

e) The **Trace size** drop-down list (Fig. 3.51, 5) allows the selection of the trace size. Using a bigger **Trace size** than the default may result in slower display and performance degradation.

f) The **Type of points** drop-down list (Fig. 3.51, 6) allows the selection of what type of point used to indicate data points on the trace. Using different point types than the default (none) may result in slower display and performance degradation.
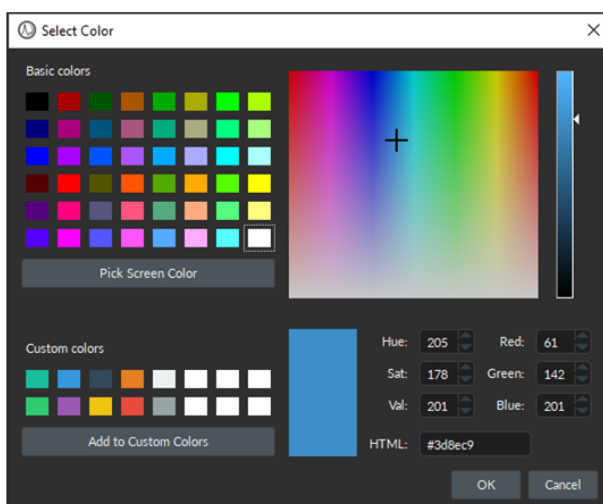

Figure 3.52: *Select Color Window*

4. The **Status** bar (Fig. 3.50b, 4) displays acquisition status. **STOPPED** is displayed when the acquisition is inactive, and **STARTED** when acquisition is active.

5. The **Triggered by:** (Fig. 3.50b, 5) text-box displays the source of the trigger for that channel, which can either be Manuel (i.e. selecting the **Record**/**Live** button) or a specific channel that provides external trigger signal.

The **Headstage Info** box (Fig. 3.53) displays the *EP Console*-specific elements which are important to pair, control and monitor the wireless headstage. The following elements are included
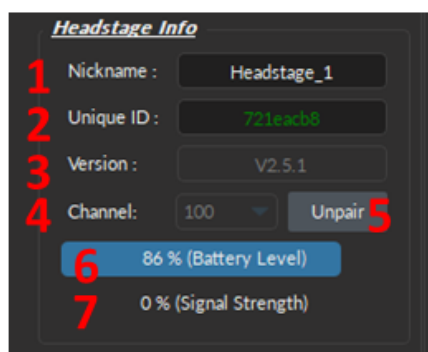

Figure 3.53: *Acquisition View, Controls*

1. The **Nickname** text-box (Fig. 3.53, 1) allows the user to change the name used for the connected headstage.

2. The **Unique ID** box (Fig. 3.53, 2) displays the unique ID sequence associated with the headstage currently in use.

3. The **Version** box (Fig. 3.53, 3) displays the headstage firmware version.

4. The **Channel** drop-down list (Fig. 3.53, 4) shows the channel currently in use by the headstage. When using two headstages, the channel must be different for each headstage.

5. The **Pair** button (Fig. 3.53, 5) is used to pair an active headstage to a console. When a headstage is paired, it becomes the **Unpair** button, which unpairs the active headstage associated with the given antenna.

6. The **Battery Level** bar (Fig. 3.53, 6) displays the headstage battery level, in %, at all times.

7. The **Signal strength** bar (Fig. 3.53, 7) displays the percentage of data packets lost during the WiFi transmission. If the signal strength is acceptable (100-76%) the bar appears blue. If the signal strength is low (75-50%), it will appear yellow. If the signal strength is critically low (<50%), it will appear red. Note that the signal will weaken if the animal/headstage is too far from the antenna or if there are obstructions between them.

Note: To obtain a value different from 0% for the signal quality, it is necessary to launch **Read** or **Record**.
To pair the headstage (Fig. 3.54), (1) click **Pair** and (2) unique ID in green will appear if correctly paired.
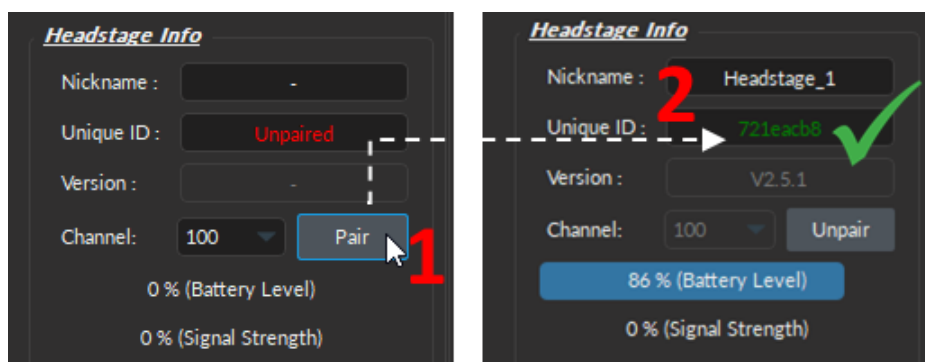

Figure 3.54: *Pairing Headstage*

3.5.2 Channel Graph Visualisation

Besides editing the trace of the channel **Graph**, which can be done through the **Edit** button of the **Control box** (section 3.5.1), other features of **Graph** view can be directly manipulated by selecting elements of the **Graph** itself. This section includes changing axis properties, manual zoom, and determining instantaneous values.
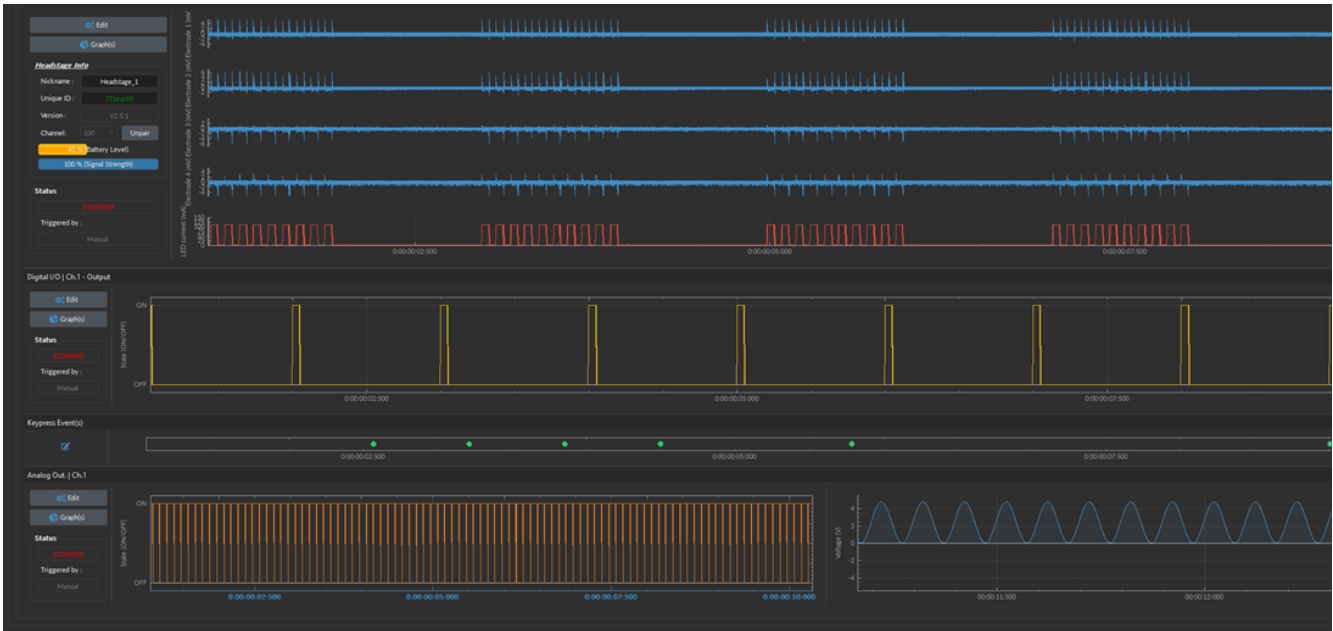

Figure 3.55: *Acquisition View - Graph*

- *Axis Options* - Each **Graph** (Fig. 3.51) has both a **Voltage** or **State** as the vertical axis and **Time** as the horizontal axis. Double-clicking either axis will open an **Axis Options** window where the axis limits can be set, similar to the **Zooming Range** in the **View Tab**. Any changes done on a horizontal axis will change the axis limits for every channel.
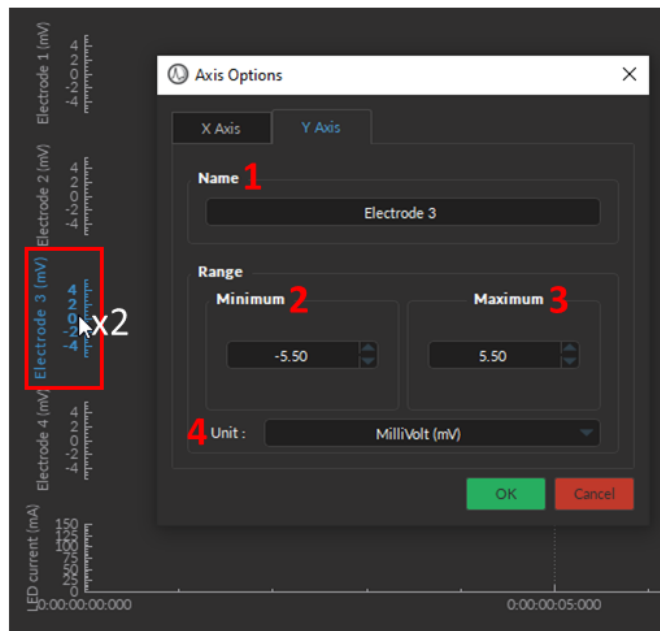

Figure 3.56: *Double click on any axis to open its Axis Options window*

- By clicking and **dragging the graph sideways or upwards**, one can scroll through nearby values on either axis, keeping the zoom range constant. Any changes done on a horizontal axis will change the axis limits for every channel.

- Using the **Mouse Scroll Wheel**, one can change the zoom range of the graph. Any changes done on a horizontal axis will change the axis limits for every channel.

- The **Instant values** box can be activated by double-clicking the **Input graph** box and selecting **Show instant values** (Fig. 3.57). This box shows the current value detected by the console for each trace on the selected channel. This box cannot be activated on **Preview graphs**. To remove instantaneous value, double click on the dot.
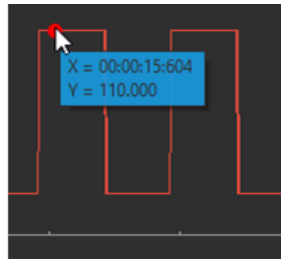

Figure 3.57: *Acquisition View - Instant values*

- The **Channel tabs** appear in certain input modes (such as **Interleaved** and **Lock-in**) where the input automatically sets the output values on separate channels. It is possible to create a **Channel tab** by undocking one channel and moving it above another until it turns blue, then releasing it.

- Analog output channels display an **Active state** graph (Fig. 3.58, left panel). This graph displays whether the channel is outputting a signal (On, V≠0) or not (Off, V=0).

- Output channels display a **Preview** graph (Fig.3.58, right panel), showing a preview of the pulse sequence.
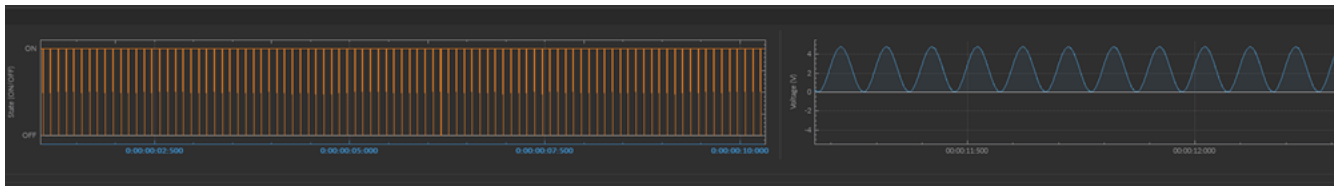

Figure 3.58: *Acquisition View - Output graph*

Analysis

Doric Neuroscience Studio includes basic data processing modules for electrophysiology. These features include the **visualizing** previous recordings, **filtering** data, **artifact removal**, and basic **spike detection** for multi-unit analysis. Since several excellent open source spike sorting algorithms (which divide spikes into single units) are available online, DNS version 6 no longer includes this feature.

The **Signal Analyzer** module can be accessed through the **Analyse** Tab at the top right of the main window (Fig. 4.1).
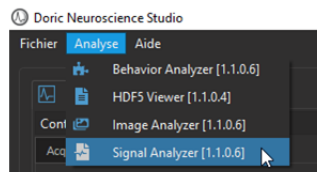

Figure 4.1: *Open the Signal Analyzer module*

The **Signal Analyzer module** (Fig 4.2) is separated into two main sections.

1. The **Control section** (Fig. 4.2, 1) contains all controls, separated into the **File**, **View**, **Spikes** and **Processing** tabs.

2. The **Graph section** (Fig. 4.2, 2) contains all currently displayed graphs as well as timestamped notes.
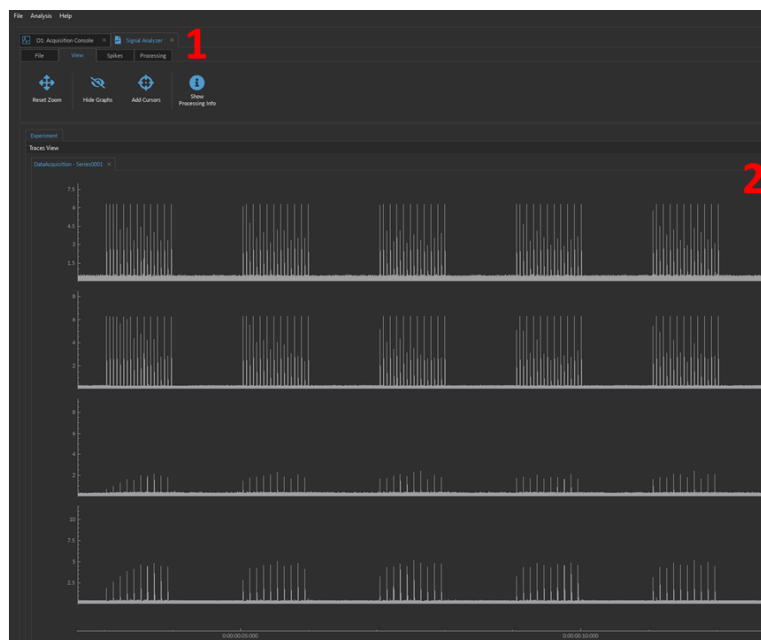

Figure 4.2: *Electrophysiology Analysis Module*

4.1   File Tab

The **File** tab (Fig. 4.3) is primarily used to load and save the data. The following details the specific features of each button, as per Fig. 4.3:
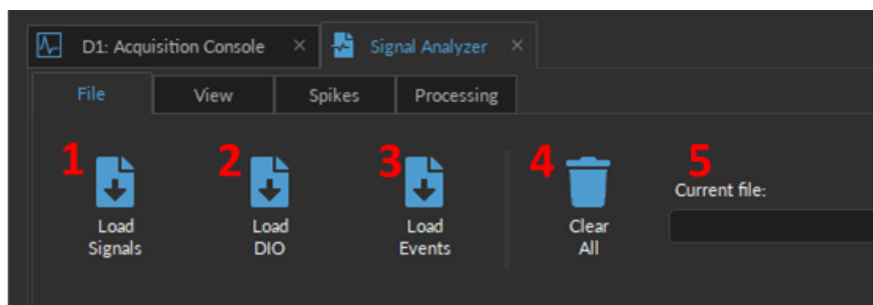

Figure 4.3: *File Tab*

1. The **Load Signals** button (Fig. 4.3, 1) opens a File Selection Window where users can select the *.doric* file of a previous recording and import it into the module. The file must contain both time and signal data. Once the file is selected, a second window will pop up (Fig. 4.4), allowing user to specify which channels to include for data processing. Only the selected channels will be displayed in the graph box. Multiple channels can be selected at once.

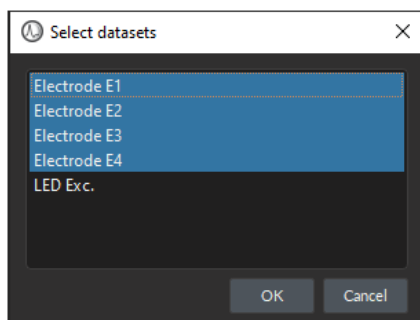   **Note:** Additional *.doric* files can be loaded into the module for comparison between recordings.

Figure 4.4: *Load selected channels*

2. The **Load DIO** button (Fig. 4.3, 2) allows users to import Digital Input/Output signals from a .doric file into *Trace View*.

3. The **Load Events** (Fig. 4.3, 3) allows users to display **Keypress Events** from the a .doric file.

4. The **Clear all** button (Fig. 4.3, 4) deletes all data currently loaded in the module. This data cannot be recovered, so ensure the data is properly saved before clearing it.

5. The **Current File** box (Fig. 4.3, 5) displays the name and path of the most recent file loaded into the analysis module. If no file has been imported, the box will be blank.

   Notes:

   - The **Signal Analyzer** module can display data from multiple files, simultaneously. If time values are missing, they will be left blank in within the **Graph** window.

   - To merge several data files together, (such to use the *Doric's* **DANSE** data analysis software, for example) use the **Doric File Editor** module.

4.2   View Tab

The **View** tab (Fig. 4.5) is used to adjust the view in the **Graphs box**. The following details the specific features of each button, as per Fig. 4.5:
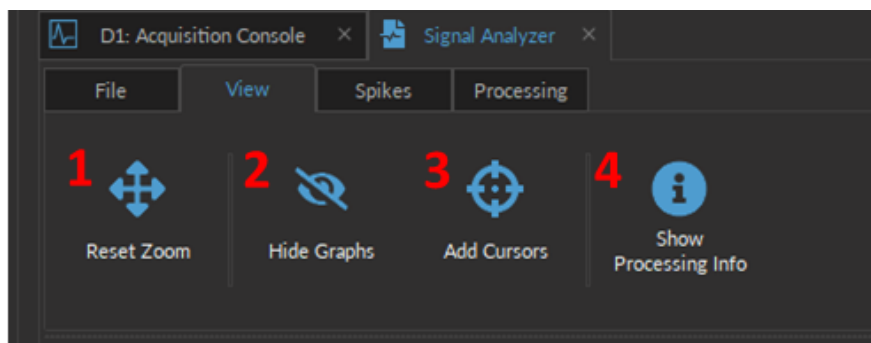

Figure 4.5: *View Tab*

1. The **Reset zoom** button (Fig. 4.5, 1) resets the axis so that the entire recording is visible.

2. The **Hide graph** button (Fig. 4.5, 2) opens the **Show/hide graphs** window (Fig. 4.6). Any checked data sets will be displayed in the **Graph box**.
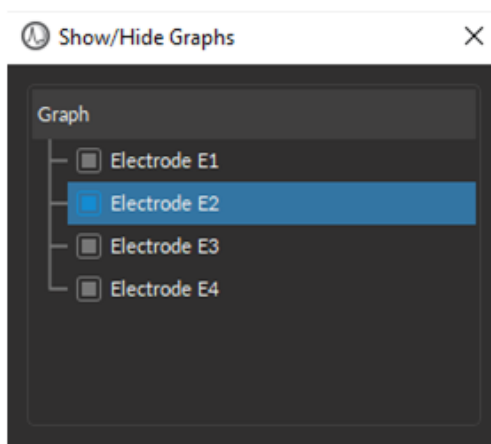

Figure 4.6: *Show/hide Graphs Window*

3. The **Add cursors** button (Fig. 4.5, 3), when selected, allows users to add 1-2 markers on the graph and displays the coordinates of the chosen point (Fig. 4.8). A left click will activate a blue cursor, while a right-click will activate an orange cursor. If both cursors are used, the **Time Difference** between the two cursors will be displayed at the top of the graph (Fig. 4.8, red box). To remove the cursors, click the **Remove cursors** button (Fig. 4.7) (previously the **Add cursors** button).
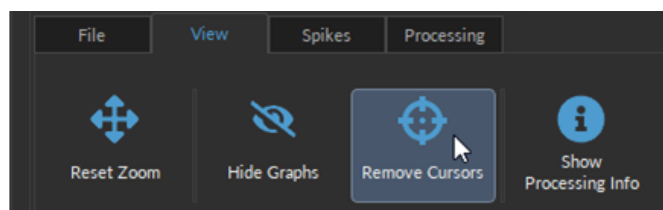
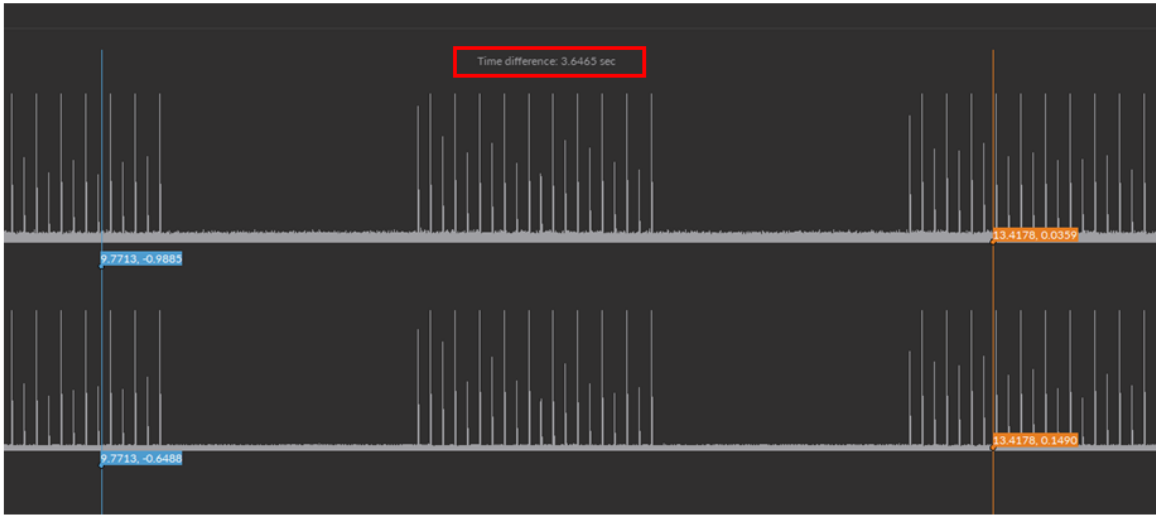

Figure 4.7: *Remove Cursors Button*

Figure 4.8: *Add Cursors*

4. The **Show Processing Info** button (Fig. 4.5, 4) opens a box on the right side of **Traces View** (Fig. 4.9). This box records all the data manipulation and parameters that a user has performed on the raw data. This includes the processing algorithms used (Operation Name), device that collected the data (Source), and parameter specified for the each algorithm.
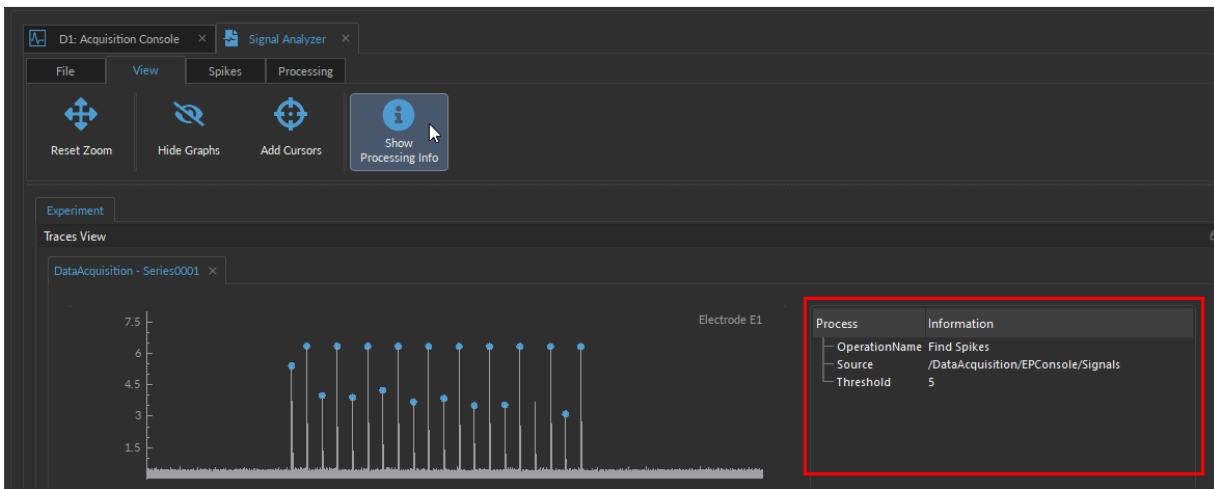

Figure 4.9: *Processing Info Box*

4.3 Spikes Tab

The **Spikes** tab (Fig. 4.10) is used to save, load and edit data files containing process data within the module.
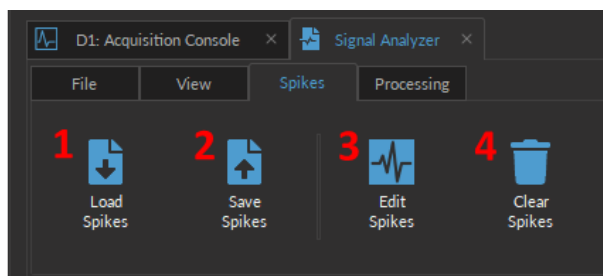

Figure 4.10: *Spikes Tab*

1. The **Load Spikes** button (Fig. 4.10, 1) will open a file selector window were user can pick which _Spikes#.doric data set to import into the module.

2. The **Save Spikes** button (Fig. 4.10, 2) will automatically output a new file containing the processed data in the same folder as the raw data. This new file will share the name of the raw data file, plus a _Spikes between the old name and .doric. If a spike file of that name already exists, an additional Spikes file is generated as _Spikes#.doric, instead of overriding the data.

3. The **Edit Spikes** button (Fig. 4.10, 3) allows users to update the spikes information in the current _Spikes.doric file without generating a new Spikes file. Instead, the new and old processed data are combined into a folder of the same name.

4. The **Clear Spikes** button (Fig. 4.10, 4) will erase the Spike file currently loaded in the module.

4.4 Processing Tab

The **Processing** tab (Fig. 4.11) contains all the operations that can be run over the raw data. Since this module can be used both to process electrophysiology and fiber photometry data, below selectively covers the operations that are applicable to electrophysiology data:
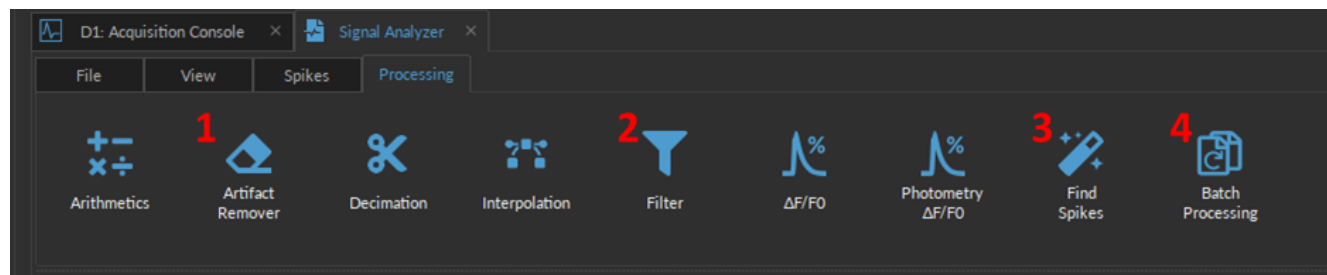

Figure 4.11: *Processing Tab*

1. The **Artifact Remover** button (Fig. 4.11-1) opens the **Artifact Remover** window (Fig. 4.12). This window enables users to delete any remaining LED artifacts from individual channels by finding the first "spike" (artifact) that occurs around LED onset or offset and forcing all the data points associated with the artifact to zero. To use this operation, the LED Exc. trace must be loaded into the module (using the **Load signals** button from the File Tab, see Fig. 4.3-1 and Fig. 4.4). For more information on this algorithm and the principles behind the artifacts, see section 3.4.3.1, LED Artifacts Remover).

   a) Using the electrode selector box (Fig. 4.12, a), select one or more **Electrode** channel(s) on which the artifact remover algorithm will be applied.

b) Select the *LED Exc.* signal for the **LED trace** (Fig. 4.12, b). This should be a Digital Output signal and NOT electrophysiological data. This trace tells the algorithm when the LED was on and off so that onset and offset artifacts can be appropriately detected.

c) Specify the **Artifact Width** (Fig. 4.12, c), which can be any value between 0.1 ms and 1 sec. However, since spikes of a neurons are on the order of 1-2 ms, we do not recommend values larger than 2 ms, as it will likely remove real spikes.
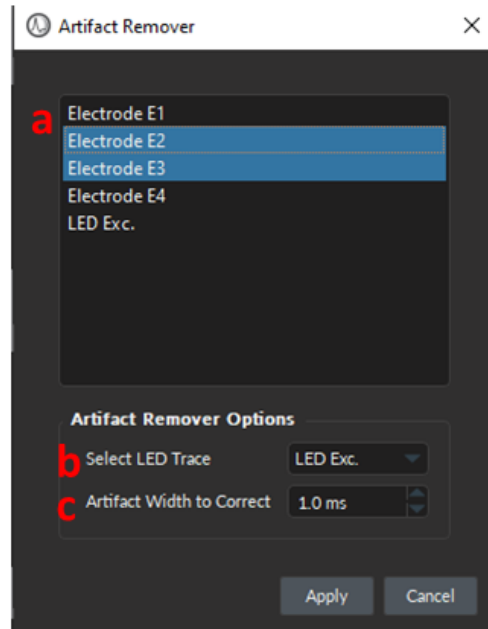

Figure 4.12: *Artifact Remover Window*

2. The **Filter** button (Fig. 4.11, 2) is used to filter out specific frequencies from the data. This can be used to either smooth the data or remove noise. Selecting the button opens the **Filter window** (Fig. 4.13) where users can choose a filter parameters. By default the Butterworth Filter (order 10) is used to process the data. The following parameters can be specified:
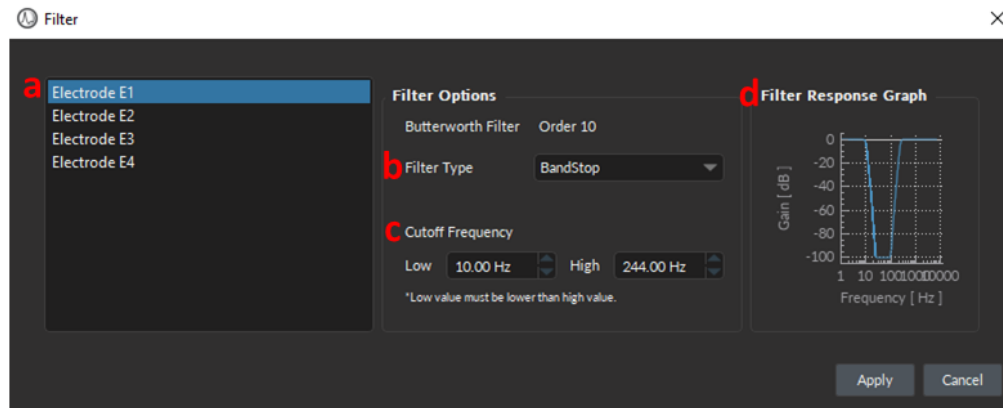

Figure 4.13: *Filter Window*

a) Using the electrode selector box, select one or more **Electrode** channel(s) on which the filter will be applied.

b) The **Filter type** defines whether the filter is low-pass, high-pass or bandwidth.

c) The **Cutoff frequency** defines which frequencies are filtered. Which values are accessible depends on the **Filter type**. Either/or both **Low** and **High** cutoff values can be defined. The low pass value must always be smaller than the high pass value.

d) The **Filter Response Graph** displays a visualization of the filter in use, with the specified low and/or high pass filter cutoffs.

3. The **Find spikes** button (Fig. 4.11, 3) identifies peaks in the data over one or multiple electrode/channels (Fig. 4.14). This operation can be used for both electrophysiology and fiber photometry to identify peaks in the data above a threshold value. Peaks will be identified as spikes if they cross a certain **Threshold** value. The **Threshold** value must be specified by the user at the bottom of the **Find Spikes** window (Fig. 4.14). This value represents the number of standard deviations over the mean baseline activity. Once detected, the spikes are displayed as blue dots over the traces in **Trace View** (Fig. 4.15).

- **NOTE:** For electrophysiological recordings, this algorithm does NOT sort spikes into single units. Thus, all detected spikes correspond to multi-unit activity (or to artifacts if they were not removed).
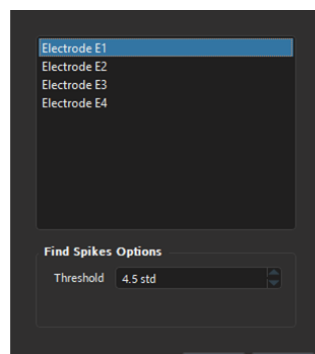

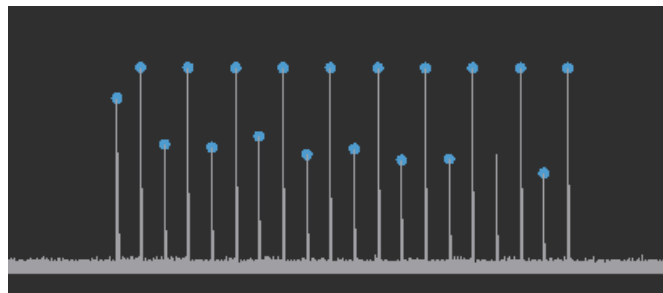Figure 4.14: *Find Spikes Window*

Figure 4.15: *Spikes are identified on the signal trace with blue dot*

4. The **Batch Processing** button (Fig. 4.11, 4) allows data from multiple recordings to be automatically processed with the same operations in a specified sequence and without user inputs.

a) The **Select Folder** button (Fig. 4.17, a) will open a file explorer window where users can specify the folder that contains the data for batch processing.
- *Batch Processing will not run* if other files (such as FILENAME_Spikes.doric files are included in the folder). Make sure only raw data file are contained in the selected folder.

b) The **Select Datasets** button (Fig. 4.17, b) is not required for FiWi data since each file should contain the same four electrode channels and LED excitation signals. If the automatic selection occurred, a path will be displayed as per Fig. 4.16. However, if other inputs/outputs are included during the recordings, make sure each file has identical channels, in order for the batch processing to work.
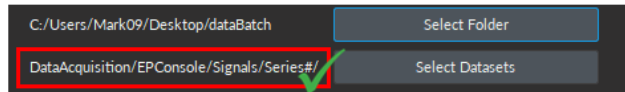
Figure 4.16: *Automatic FiWi Dataset selection*

c) If the **Save Intermediate Files** options (Fig. 4.17, c) is selected (**Yes**) batch processing will generate a file after every operation, and for each recording (Fig. 4.18). Select **No** if intermediate files are not required.

d) The **Available Operations** box (Fig. 4.17, d) contains all the possible operations that run in batches over the data. These are the same operations that can be manually selected over a single recording using the **Processing** Tab (Fig. 4.11).

e) The **Workflow** box (Fig. 4.17, e) displays the operations that will be run over each recording during batch processing, following the order of the operations. To add an operation to the **workflow**, click on the operation of choice in the **Available Operations** box. Note that the order that operations are added to the **Workflow** is the order they will be run during batch processing. To remove an operation from the **Workflow** click on that operation and it will return to the **Available Operations** box.

f) The **Options** section (Fig. 4.17, f) displays a box to specify the parameters for each operation. Details concerning parameters of each operation can be found in the non-batch processing section of the operation in question (Section 4.4: 1-3).

- **NOTE:** Some operations use data-set names. Make sure that the names are the same across all the files.
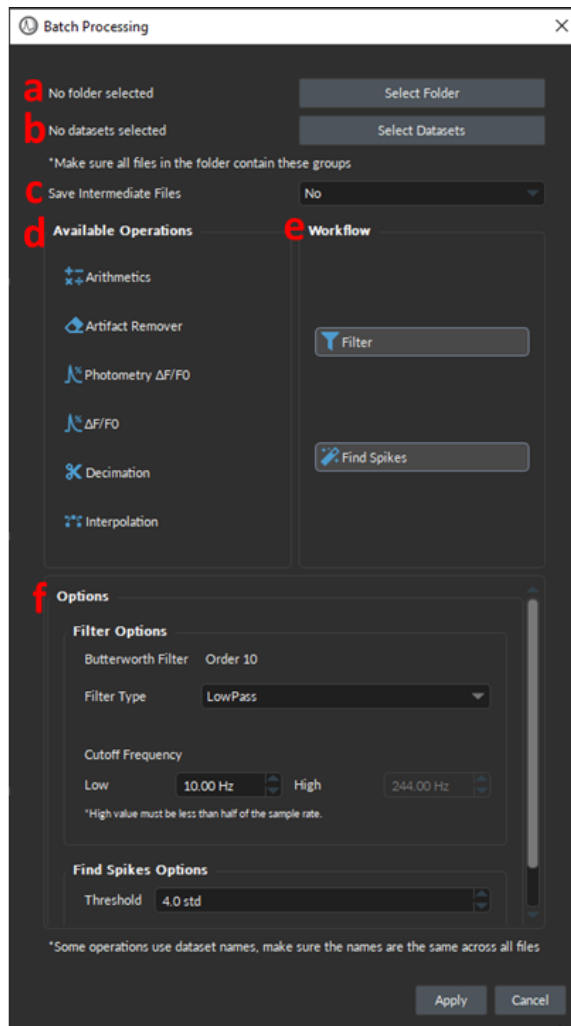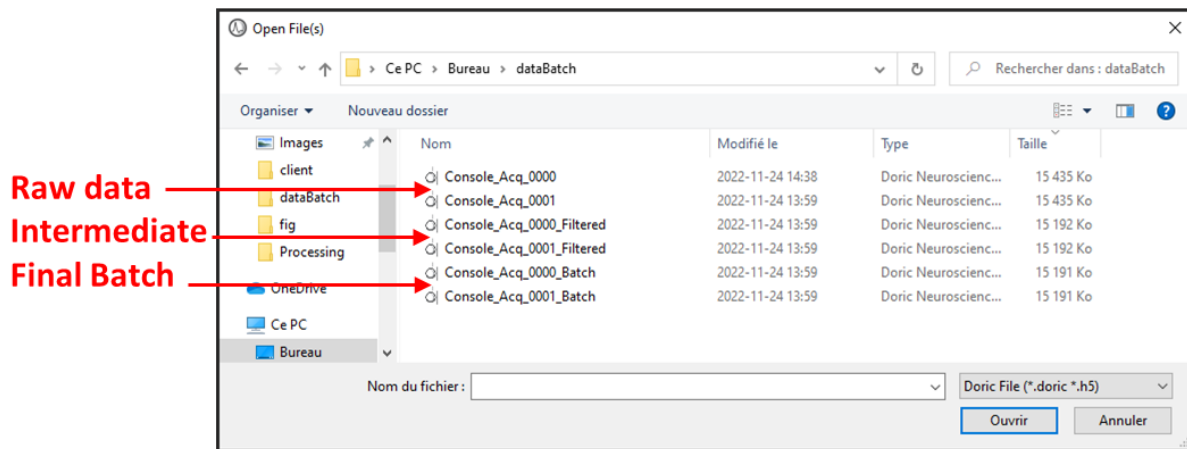
Figure 4.17: *Batch Processing Window*

Figure 4.18: *Batch Processing Output*

Specifications

Table 5.1: *Fi-Wi Opto-electric Cannula General Specifications*

SPECIFICATIONS	VALUE	NOTES
Peak optical output	8 mW	with blue (465 nm wavelength) LED
Input current	Min: 0, Max: 150 mA	
Dimensions	7.5 x 9.7 x 9.7 mm^3	Without fiber and electrodes
Mass	0.4 g	
Electrodes material	Tungsten	
Electrodes diameter	25 or 50 μm	
Optical fiber NA	0.66	
Optical fiber core diameter	200 or 400 μm	

Table 5.2: *Fiberless & Wireless Headstage General Specifications*

SPECIFICATIONS	VALUE	NOTES
Dimensions	9.6 x 14.9 x 18.6 mm^3	
Mass	2.8 g	Including 40 mAh battery
Transmission Frequency	2.400 to 2.525 GHz	
RF Data Speed	2 Mbps	
Electrophysiology Sampling Rate	14 kHz	On each channel
Operation distance	0-3 m	For optimal performance
High-pass frequency filter loss	-30 dB/decade	
Low-pass frequency filter loss	-40 dB/decade	
Battery charge duration	90 min	60 mA current at 10% duty cycle, 4 channels recording
Battery recharge time	> 60 min	

Table 5.3: *Electrophysiology Console General Specifications*

SPECIFICATIONS	VALUE	NOTES
Power supply	110-240 VAC ; 50-60 Hz	Power supply adapter included
DC Power supply	12 VDC	
Dimensions	360 x 140 x 37 mm³	Including connectors
TTL input voltage	0 to +5 V	Min Hi Level: 4 V; Max Low Level: 0.55 V
TTL output voltage	0 to +5 V	
TTL input impedance	3 kΩ	
TTL output impedance	30 Ω	
TTL time propagation delay	4.6 ns	
TTL time input resolution	82 μs	
TTL time output resolution	10 μs	
Analog output voltage	± 4.75 V	
Analog output sample frequency	25 MS/s	
Analog output max. frequency	50 kHz	
Analog output impedance	6 Ω	
Analog time propagation delay	20 ns	
Analog Input voltage	±10 V	
Analog input sample frequency	12 kS/s	
Channel-to-channel isolation	110 dB	
Analog input impedance	100 to 124 kΩ	

Table 5.4: *Electrophysiology Console Analog input noise and resolution specifications*

Data rate	Real Rate	Time resolution	- 3dB Frequency	RMS noise	Effective resolution[1]
0.3 kSps*C	372 Hz	2686 μs	200 Hz	9.6 μV	20.0 bits (9.5 μV/bit)
1.0 kSps*C	1 001 Hz	999 μs	520 Hz	15.5 μV	19.3 bits (15.5 μV/bit)
2.5 kSps*C	2 534 Hz	395 μs	1 300 Hz	26.1 μV	18.5 bits (27.0 μV/bit)
6.0 kSps*C	6 041 Hz	166 μs	3 100 Hz	46.0 μV	17.7 bits (47.0 μV/bit)
12.0 kSps*C	12 166 Hz	82 μs	6 300 Hz	120 μV	16.3 bits (123.9 μV/bit)

Table 5.5: *Console Recommended Environmental Specifications*

DESCRIPTION	OPERATION	STORAGE
Use	Indoor	Indoor
Temperature	0-40 ° C	0-40 ° C
Humidity	40-60% RH, non condensing	40-60% RH, non condensing

[1]Effective resolution for linear mode at ± 10 V input range. Input resolution is 16 bits (0.152 mV). Lower data rates provides higher effective resolution. The lock-in mode allows higher effective resolution. In this case, effective resolution is higher than RMS noise for every listed frequency.

Support

6.1   Maintenance

The product does not require any maintenance. Do not open the enclosure. Contact Doric Lenses for return instructions if the unit does not work properly and needs to be repaired.

6.2   Warranty

This product is under warranty for a period of 12 months. Contact Doric Lenses for return instructions. This warranty will not be applicable if the unit is damaged or needs to be repaired as a result of improper use or operation outside the conditions stated in this manual. For more information, see our Website.

6.3   Contact us

For any questions or comments, do not hesitate to contact us by:

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