Trodes

Getting Started

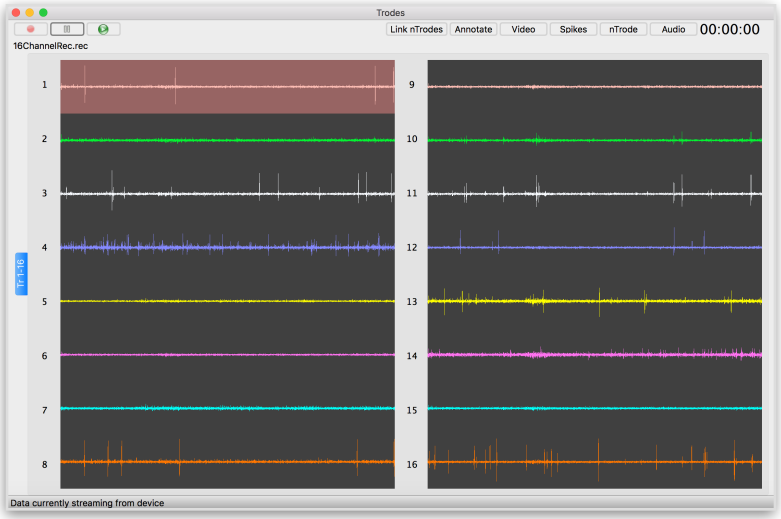
1. Installation

1) Install the FTDI D2XX driver for the USB driver.
2) Copy the downloaded Trodes directory to a desired location.
3) Double click on “Trodes” to start.

2. Controls overview

We will start the tutorial by using the playback function to give a quick overview of Trodes with real data recorded from a mouse. Download the example recording and extract the folder. The folder has two files in it: a .rec file and a .trodesConf file with the same name. In Trodes, go to **Connection->Source->File** and select ‘16ChannelRec.rec’. This file has 16 channels, where the nTrodes are single-channel bundles. Press the play button to start playback.

Now you should see data streaming across the screen, and if your computer has audio set up, you should hear audio for the currently highlighted channel. You can change the length of time displayed in the data streams by selecting a time option under **View->Streaming->Trace length**. You can toggle the coupling between the displayed streams from the spike filters under **View->Streaming->Filters**.



Along the top of the window, there are control buttons. Here are their functions:

1) **Link nTrodes** — you can either enable or disable nTrode linking with this button. When they are linked, any change in settings to one nTrode (such as reference or filter settings) are applied to all nTrodes.

2) **Annotate** — this button opens a dialog that is activated only when a recording file is open. You can type any text into this dialog, the the strings will be timestamped and saved. This allows you to take notes during the recording.

3) **Video** — this button starts the camera module

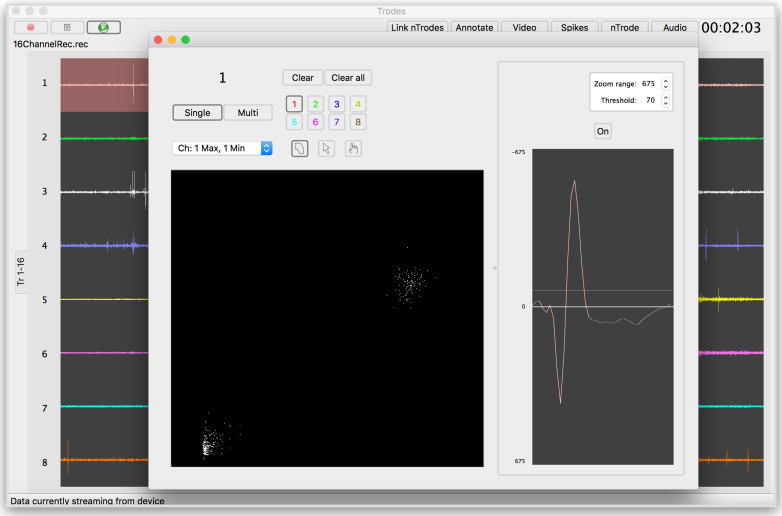
4) **nTrode** — this button opens a dialog with settings for the currently selected nTrode. To select a different nTrode, simply click on a channel in the streaming window to highlight it.

5) **Spikes** — this button open a window displaying spike trigger events for the currently selected nTrode.

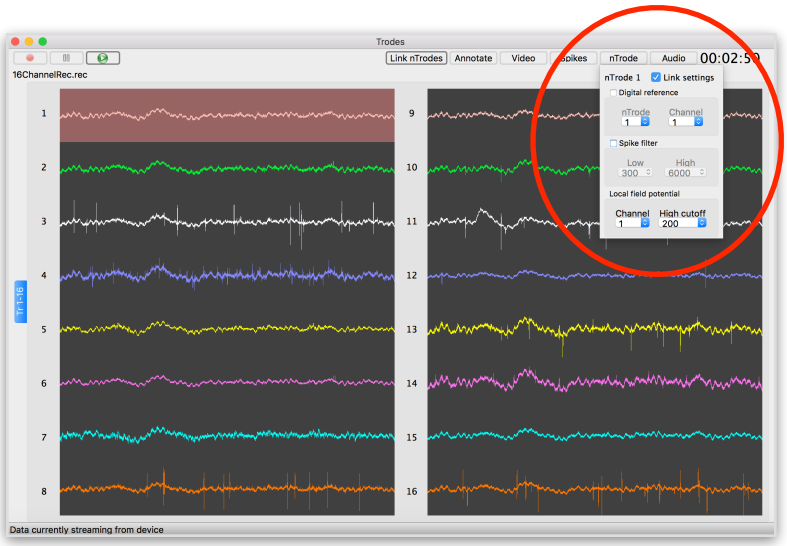
6) **Audio** — Opens a dialog to modify audio settings.

Notice that channel 1 is highlighted. This indicates that channel 1 is the currently selected channel. Audio is streamed for this channel, and the settings for the channel can be modified by pressing the ‘nTrode’ button.

With channel 1 selected, click on the ‘Spikes’ button. A window will appear showing trigger events for channel 1. You can change the voltage threshold used to define a trigger, and you can sort spikes in the scatter plot. If you click on a different channel in the main window, the open Spikes window will update to the selected channel.



Select the ‘Link nTrodes’ button to link changes across nTrodes. Then, press the ‘nTrode’ button. You’ll notice that bandpass filters are currently being applied to all channels (signified with the check mark in the spike filter section). Uncheck this, and the filters will be deactivated and you will see lower frequency signatures in the traces.



Now, turn the filters back on. You can also select a digital reference with this menu. If one or more of your channels are designated as references, you can subtract its data from any other channel.

To change the display range on any strode, simple scroll the move wheel up and down when the mouse is over the channel(s) associated with the target nTrode. You can also change the display range in the Spikes window. If you have ‘Link nTrodes’ selected, all display ranges will be modified together.

You can change the display color of any nTrode by right-clicking on it and selecting **Change nTrode color.** You can also change the background color.

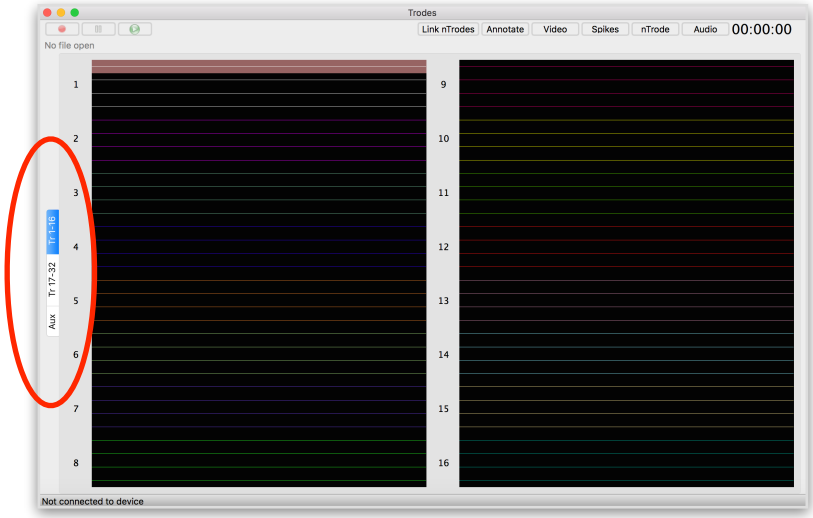
When data are processed offline, the functions use whichever settings are saved in the .trodesconf file to extract the data. So, for example, if you want to modify the filters settings, spike threshold settings, or filter settings from the ones used during the recording, you can use the file playback mode to do this. After you have modified the settings, go to **File->Workspace->Save settings as…** and make sure the name is the same as the recording file (but with the .trodesconf extension) and located in the same folder. Now, the next time you open the file for playback, the new settings will be used.

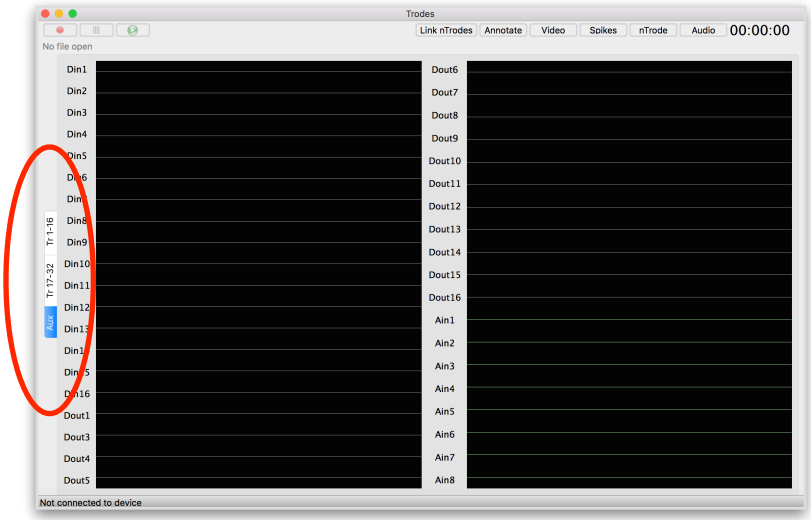
To disconnect from a source, select Connection->Disconnect. This will stop streaming when you are connected to hardware. During file playback, it stops playback and resets to file to the beginning. Now, select **Connection->Source->None** to close the playback file.

3. Acquiring data

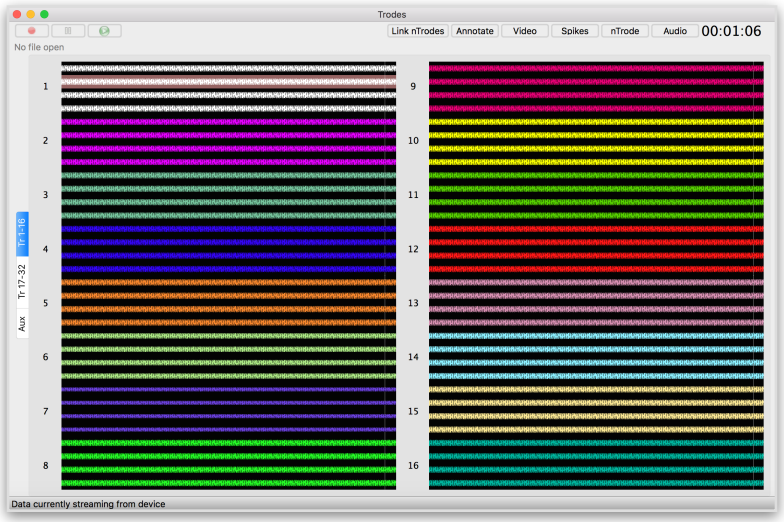
Before data can be acquired, a workspace needs to opened that tells Trodes how many channels to expect from the hardware and how to organize them (single channels, tetrodes, octrodes…). Download the workspace examples and extract the folder. For this tutorial, we will work with '128_Tetrodes_ECU.trodesconf'. This workspace sets up 128 channels for acquisition, all organized into 4-channel 'tetrodes'. It also expects data from the ECU, which streams auxiliary digital and analog data.

To open the workspace in Trodes, go to **File->Workspace->Load**, and select '128_Tetrodes_ECU.trodesconf'. You'll notice that there are multiple tabs to display all of the channels, plus an 'Aux' tab displaying the extra digital and analog lines from the ECU. Not all ECU channels are displayed— this can be changed by editing the workspace file. Also, the Spikes window now shows 4 trigger windows from every nTrode.





To connect to a data source, select a source from **Connection->Source.** In this example, we will use the Signal Generator, which produces a sine wave on all channels and does not require any hardware (to modify the generated sine wave, open up **Connection->Generator controls**). If you already have hardware available, you can select that (example: **Source->SpikeGadgets->USB**). Otherwise, select **Connection->Source->Signal Generator.** Then, to start streaming data, select **Connection->Stream from source.**



4. Recording to disk

To start recording, you must first create an output file. Select **File->Create file for**

recording to open a save dialog. You'll notice that a default name is entered into the save dialog, such as 'myAnimal03-15-2016(12_12_30).rec'. The 'myAnimal' part of the name is there because the workspace has a declared default prefix for the name. This is described in further detail in the workspace section below. The rest of the file indicates the date and time of the recording. You can use a different name, as long as you follow the following convention to make other modules and the offline file processing code behave normally:

1) Do not use the underscore character ('_').
2) Do not use spaces.

Also, each recording session should be placed in a dedicated folder. A recording session can have multiple files, in which case it is best to keep acquisition running in between file closure/creation to keep the temporal relationship of the recordings intact.

Once you have entered the desired file path and name, click 'Save'. Now, at the top of the window you will see the .rec file name, plus indicators for the current file size and amount of storage available.

Start streaming data (see above), and then select **File->Record** to start saving data. Alternatively, you can press the record button at the top of the window. You can pause recording at any time— this does not close the file but simply stops logging. When the record button is pressed, logging continues (with a gap of data in between).

You are not allowed to stop/start acquisition once logging has begun. You must close the file first. This is to ensure that no time resets occur within a single log file.

Once the file is open, you can annotate events during the recording using the 'Annotate' button. Write a line into the dialog and press 'Enter'. The line will be tagged with the current timestamp and seven to a text file. Upon the first annotation, a new file will be created in the same directory as the .rec file, but with the .trodesComments extension.

To close the file after the recording is complete, select **File->Close file**.

5. The workspace

Workspace files are written in xml format, so you can use an editor to open it and edit the contents. Here are the sections of the workspace file.

1) **GlobalConfiguration** section. This section contains general settings:

```
<GlobalConfiguration filePrefix = "myAnimal"
saveDisplayedChanOnly="1" filePath="" realtimeMode="0"/>
```

Here are the definitions for the fields:

filePrefix — text appended to the default names of created log files. Usually the animals ID

filePath — the default location where log files are stored. This should be changed to an existing directory on your machine.

saveDisplayedChanOnly — can be 1 or 0. If set to 1, then Trodes will only save the channels that are displayed. Otherwise, all hardware channels are saved regardless of whether they are displayed or not.

realtimeMode — can be 1 or 0. If set to 1, the CPU works much harder to try to retain low processing latencies for closed-loop experiments.

2) **HardwareConfiguration** section.

```
<HardwareConfiguration numChannels="128"
samplingRate="30000">
```

This section contains two fields about the data, plus child nodes describing additional hardware devices that are enabled to stream data alongside the neural data.

numChannels — a number in multiples of 32 defining how many channels are coming from the hardware.

samplingRate — the sampling frequency of the hardware. The default is 30000.

This child nodes in this section contain low-level information about what hardware is connected to the system. This information is not intended to be changed, except for one field— the "**available**" field. If set to 1, then Trodes will expect data from this device in the incoming data stream.

3) **ModuleConfiguration** section.

This section tells Trodes which modules to open along with the workspace. In this workspace, the camera and StateScript modules are defined.

4) **StreamDisplay** section.

This section controls details about the display of streaming data. It contains two fields:

```
<StreamDisplay columns="4" backgroundColor="#030303"/>
```

columns — the number of display windows to divide the channel streams into
backgroundColor — the background. This can be change within Trodes by right-clicking inside any of the stream window columns.

5) **AuxDisplayConfiguration** section.

This section defines which digital I/O and auxiliary analog channel to display. Channels are selected from the available menu defined in the HarwareConfiguration section. Each DispChannel section defined one channel:

```
<DispChannel id="Din1" maxDisp="2" color="#aaaaaa"
device="ECU"/>
```

Here are the field definitions for a DispChannel:

id — The displayed name of the channel. This should match the id defined in the HarwareConfiguration section.
maxDisp — the maximum display range of the trace.
color — the displayed trace color
device — The name of the hardware device. This should match the device name in the HarwareConfiguration section.

6) **SpikeConfiguration** section.

This section defines how channels are bundled and processed for spike detection. Each child node is a **SpikeNTrode,** which has the following fields:

```
<SpikeNTrode highFilter="6000" color="#ffffff"
lowFilter="600" LFPHighFilter="200" refChan="1" refOn="0"
id="1" LFPChan="1" moduleDataOn="1" filterOn="1"
refNTrode="1">
```

id — defines the name of the nTrode. This should be a 1-based number

refOn — 1 or 0. If 1, then the defined reference channel is subtracted from all channel in the nTrode.

refNTrode — the nTrode number of the reference

refChan — the channel in the reference nTrode to use for the reference

filterOn — 1 or 0. If 1, the spike filter is turned on.

lowFilter — this is the lower range of the bandpass spike filter (200, 300, 400 500, 600 allowed)

highFilter — this is the upper range of the bandpass spike filter (5000 or 6000 allowed)

moduleDataOn — 1 or 0. Defines if LFP data from moduleDataChan should be sent to other modules

LFPChan — Defines the LFP channel to process offline, and which channel to send to other modules for online LFP pattern detection.

LFPHighFilter — define the low-pass filter for the LFP channel (100, 200, 300, 400, and 500 allowed)

color — the display color of the channel

Each SpikeNTrode section has at least one **SpikeChannel** child node. If there are n SpikeChannel children, then all n channels are bundled together, where if any one channel goes above threshold, then a spike is triggered on all channels of the nTrode. Here is one SpikeChannel:

```
<SpikeChannel thresh="30" maxDisp="200" hwChan="63"
triggerOn="1"/>
```

Each SpikeChannel has the following fields:

thresh — the trigger threshold for this channel. All channel of an nTrode should have the same threshold.

maxDisp — the maximum display range of the channel

hwChan — the 0-based hardware channel that this channel maps to.

triggerOn — 1 or 0. Defines whether or not this channel can cause a trigger event for the nTrode. This option is available if the channel is too noisy to be a reliable trigger channel.

Here is the entire first SpikeNTrode node (with four SpikeChannel child nodes to define a tetrode):

```
<SpikeNTrode highFilter="6000" color="#ffffff"
lowFilter="600" LFPHighFilter="200" refChan="1" refOn="0"
id="1" LFPChan="1" moduleDataOn="1" filterOn="1"
refNTrode="1">

   <SpikeChannel maxDisp="200" hwChan="63" triggerOn="1"
   thresh="30"/>

   <SpikeChannel maxDisp="200" hwChan="61" triggerOn="1"
   thresh="30"/>

   <SpikeChannel maxDisp="200" hwChan="57" triggerOn="1"
   thresh="30"/>

    <SpikeChannel maxDisp="200" hwChan="59" triggerOn="1"
   thresh="30"/>

</SpikeNTrode>
```

6. Using modules

If one or more modules are defined in your workspace, they will open automatically when the workspace is created. Some modules can be launched without being configured in workspace by using the menu bar.

All modules receive receive notifications when sources are selected, acquisition is started/stopped, files are created/closed, and when logging is started/paused. Custom-written modules should be written to respond appropriately to these notifications.

1) Camera Module

The camera module allows users to collect video from web cams or GIGE cameras and timestamp the frames. It also will perform rodent position tracking in multiple modes (black on white background, single LED, and dual red-green LED's for head direction). The module can also be opened on its own for offline position tracking.

When a file is created in Trodes, the camera module will create a video file (.h264 extension) and a frame timestamp file (.videoTimeStamps extension) with the same name as the .rec file. If acquisition is paused in Trodes, it is also paused in the camera module.

Along the top of the window, there are control buttons similar to the layout in the Trodes main program. These buttons are slightly different during 'slave mode' and 'standalone mode'. During 'slave mode', the camera module acts as a slave to Trodes, and the buttons have the following functions:

Track — used to toggle position tracking (logged to disk if saving is underway)

Tools — opens the tool selector window. Available tools include a manual position input tool, an include region tool, and an exclude region tool.

Source — used to select which available camera to use. USB webcams and Allied Vision GIGE cameras are automatically detected.

Settings — opens a dialog to adjust position tracking settings

In 'Standalone mode' the program is opened directly (not through Trodes). The control buttons are different in this case:

Log — initiates and ends offline position tracking and logging to file

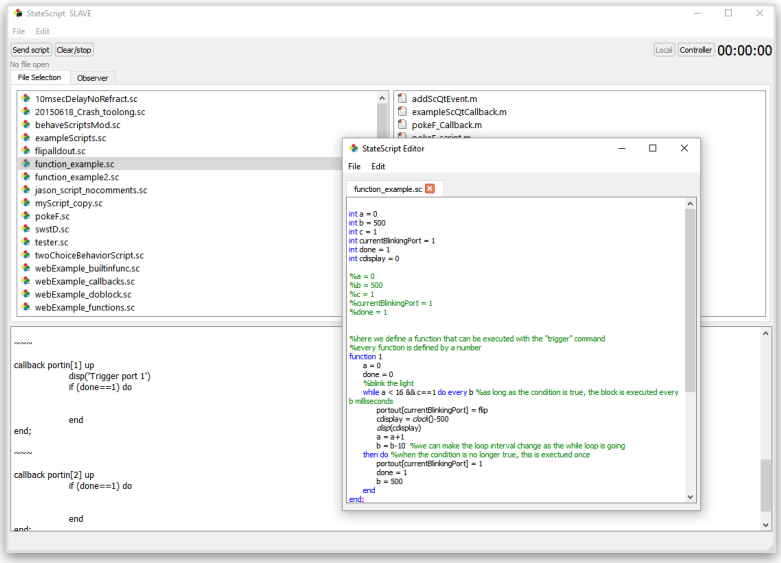
Tools — save as above

File — used to open a video file

Settings — same as above

2) StateScript Module

The StateScript Module is used to send scripts and commands to the ECU, which is running StateScript control firmware. Like the camera module, it can be opened in two modes: slave mode (through Trodes) or standalone mode (opened directly). Standalone mode is useful for controlling the experimental environment when no electrophysiology is being performed.



When the module is opened, it displays three sub panels. The top two windows are file selection windows. They should display your directories with StateScript scripts (left window) and your observer scripts (right window). To change the default folders shown, select **File->Script folders->Scripts** (for StateScripts) and **File->Script folders->Observer scripts** (for observer scripts based in matlab or python). If you double-click on a file in the left (StateScript) window, the StateScript editor will open the script for editing, as shown in the screenshot above. The bottom window is the console window, which is inactive until the module is connected to hardware running StateScript.

To connect to the hardware, press the Source button and choose your hardware from the drop down list (if using the ECU, select 'ECU') and press Connect. The window on the bottom will turn white and an vitiation sequence will appear. If connection goes smoothly, you should be able to type ';' followed by RETURN in the console window and a '~~~' will be returned indicating that your command was compiled successfully.

In slave mode, there are four control buttons at the top of the window:

Send script — when pressed, the selected StateScript is sent to hardware for compilation.

Clear/Stop — stop execution of the script and clears variables.

Local — starts the observer scripting environment (Matlab or Python). Click on the 'Observer' tab to see the console for the observer environment. To set up the observer environment select **Edit->Observer language…**

Controller — used to select and connect to hardware.

In slave mode, logging to file is controlled from Trodes, where file creation, logging start/stop, and file closing occur alongside the same actions for the recording files.

In standalone mode, the buttons at the top of the window are different:

Start — sends the selected StateScript to hardware and automatically sets up a log file for recording events.

Clear/stop — same as above

Local — same as above

Camera — open the camera module to record video of the experiment

Controller — same as above