BOSS

Quick Start Guide

For research use only

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1.0 Table of Contents

Welcome!

Thank you for choosing Blackrock Microsystems!

Inside this manual, you will find information on our Offline Spike Sorting software (BOSS), a tool that enables the sorting and editing of previously recorded data before complete data analysis.

This manual will cover the system requirements, associated hardware, and instructions for use from basic loading of data to publishing the results to file.

For questions on this product or any other Blackrock products, contact our 24/7 support service: support@blackrockmicro.com

Installation

SYSTEM REQUIREMENTS

The specifications listed below are the minimum required by the software to run as intended.

- Microsoft Windows 7 Professional (64-bit) or higher
- A processor with 4 physical cores at 2.0 GHz
- 4 GB of RAM
- Video Card with full OpenGL compatibility

INSTALLING THE SOFTWARE

BOSS is either delivered electronically or through a USB Drive that contains the installer. In order to install the software, simply execute the installer and follow the on-screen prompts.

Software Scope

USE CASES

Blackrock's Offline Sorting Software has the following typical use cases:

- Detect action potential waveforms from recorded continuous data files.
- Sort or re-sort spikes from previously recorded neural event files or from those detected from a continuous sample into units.
- Edit the data by removing or sorting waveforms through manual or automated methods.
- Save processed data back into its original data type or export to a variety of common data formats.
- Publish summary of data by allowing the creation of a customizable PDF post-processing.

SUPPORTED DATA FORMATS

BOSS can accept a number of data formats as inputs for detecting or sorting data. These allowable inputs are shown below:

- .NSx (Blackrock Continuous Data Files)
- .NEV (Blackrock Neural Event Files)

In addition to file inputs, BOSS can also export to multiple data formats for expediting the analysis process. All export options are shown below:

- .NEV (Blackrock Neural Event Files)

AVAILABLE SORTING METHODS

After either detecting waveforms from a continuous data file or loading them from a file type that contains already extracted waveforms, BOSS offers tools to classify waveforms into clusters that correspond to different neurons.

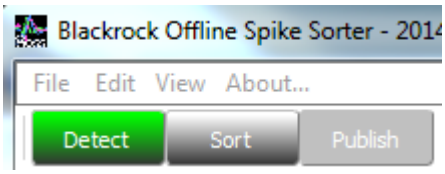
BOSS offers following methods for classification:

- T-Distribution EM
- K-Means
- Manual assignment (selection-based)
- Lines

How-To

Navigate the User Interface

The UI has been designed to guide the user through the spike-sorting process and depends on the type of file opened for analysis. The displayed widgets will depend on the step for the currently selected channel – whether it is *Preprocessing and Detection*, *Feature Extraction and Sorting* or *Publishing* - and so will the available options in the Options widget.

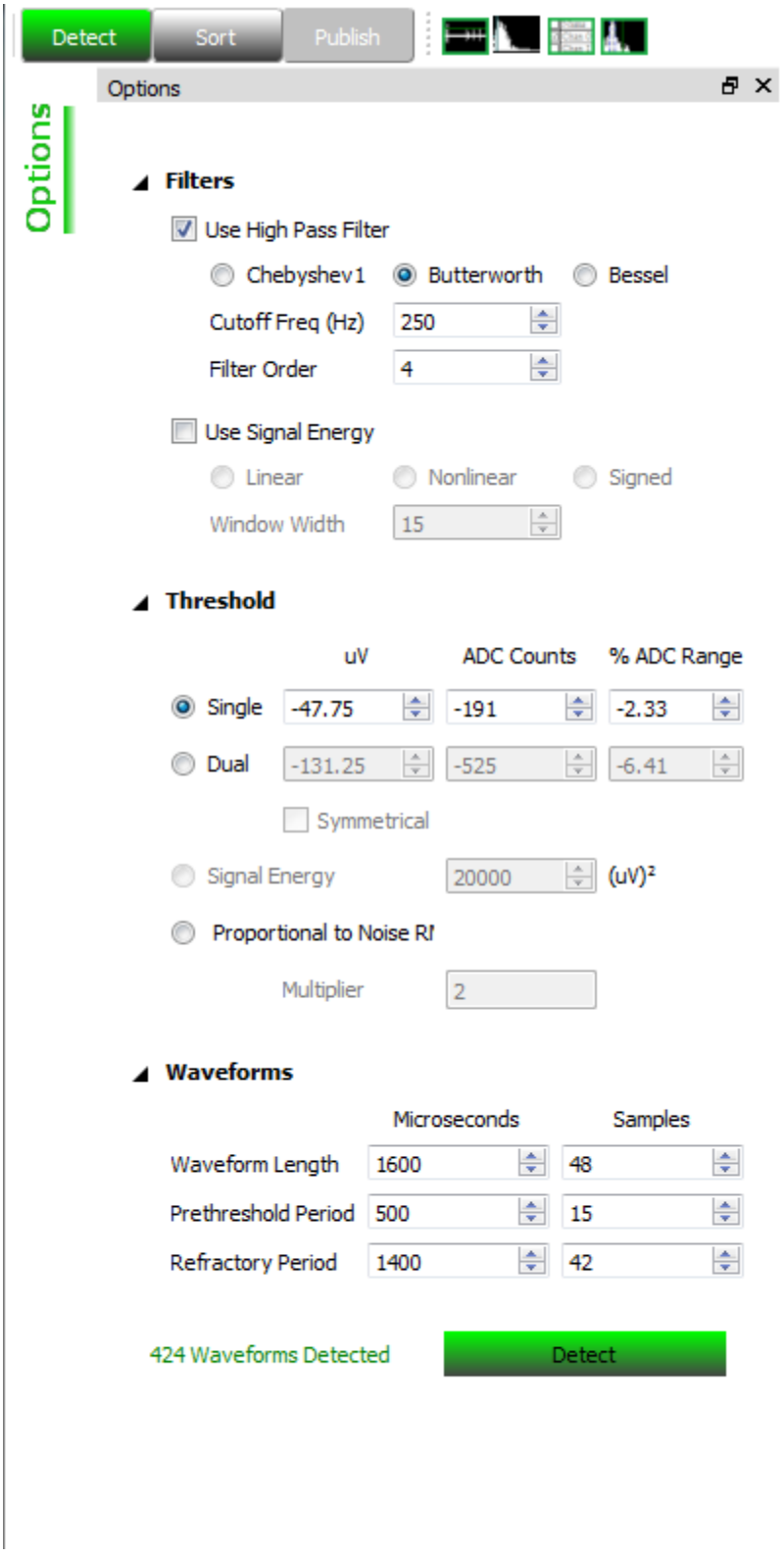


In order to advance steps, the previous steps need to be completed. For that reason if spikes have not been detected, the *Feature Extraction and Sorting* steps will be unavailable.

The UI is composed of the following widgets, which can all be undocked and arranged in any custom layout:

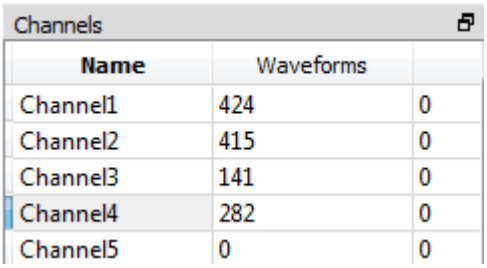
Options

The Options widget presents all available options for the current step within the sorting process. The push button on the left toolbar allows hiding and showing the widget.



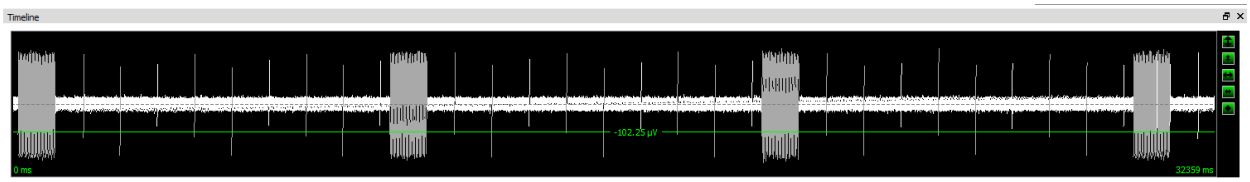
Channel Table

Use the Channel Table to peek onto channel settings and a summary of the analysis results.



Timeline

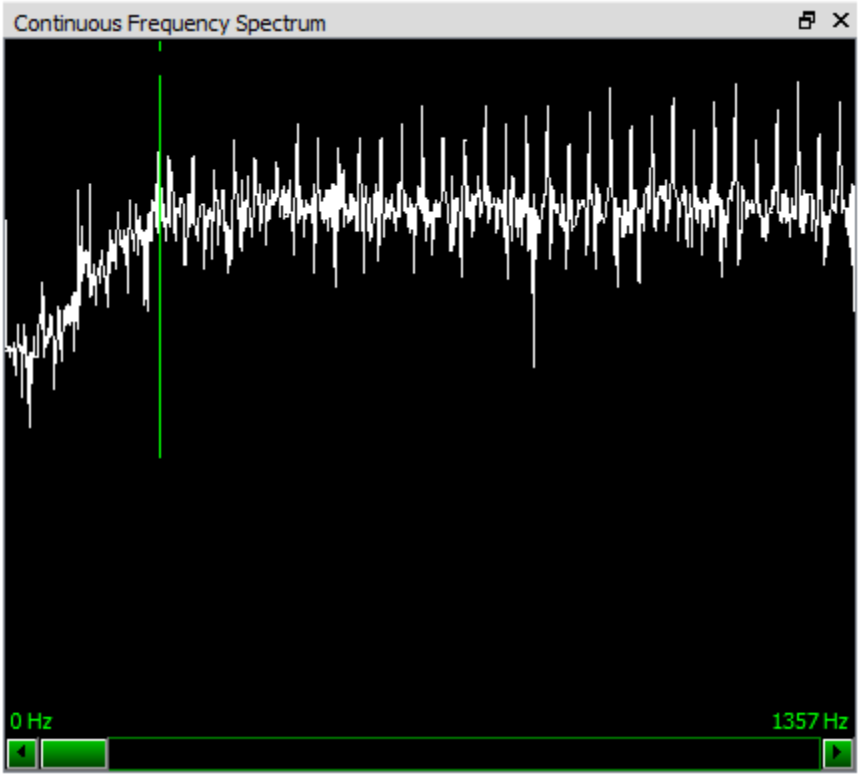
The Timeline displays a segment of the channel data. The zoom setting for the horizontal axis can be controlled using the mouse wheel; holding down the SHIFT key and using the mouse wheel will control the vertical scale. The Zoom Toolbar on the right side of the Timeline is also available for zoom control.



When opening a large file that cannot be displayed all at once, a scroll bar on top of the Timeline will appear indicating the portion of the file that is currently available in the viewport. The scroll bar on the bottom of the Timeline relates to the zoom controls.

Continuous Power Spectrum

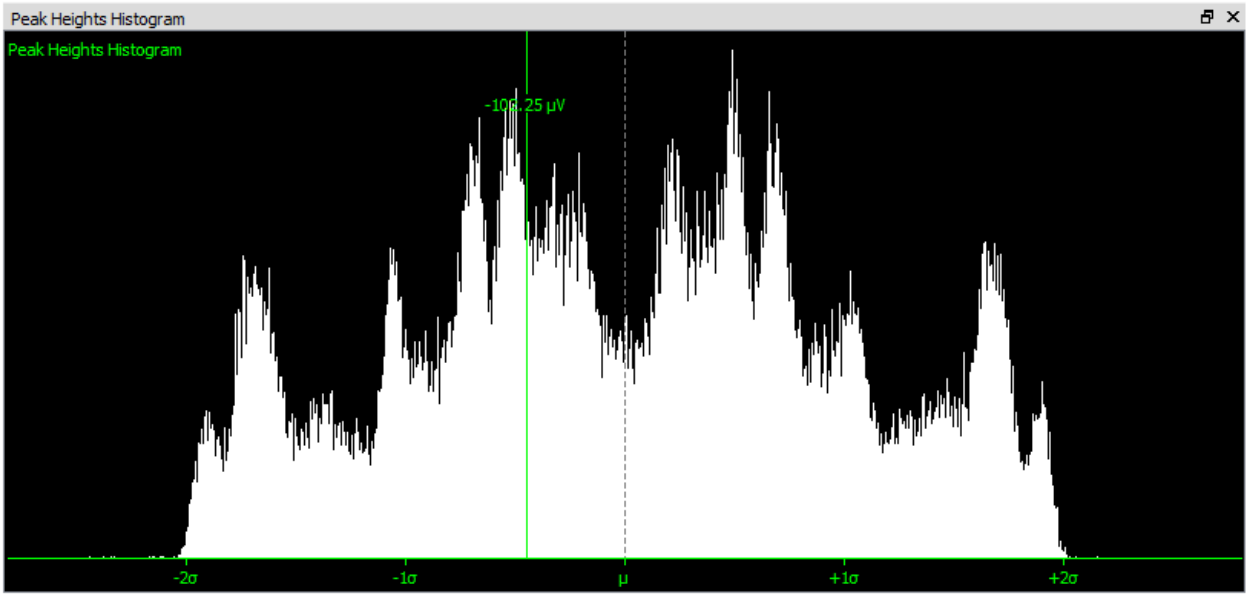
The Continuous Power Spectrum view displays the power spectrum between 0Hz and one half the sampling frequency. The vertical slider can be used to change the cut-off frequency of the HPF applied to the data.



This view is only available when a continuous data file is loaded.

Peak Heights and Energy Histograms

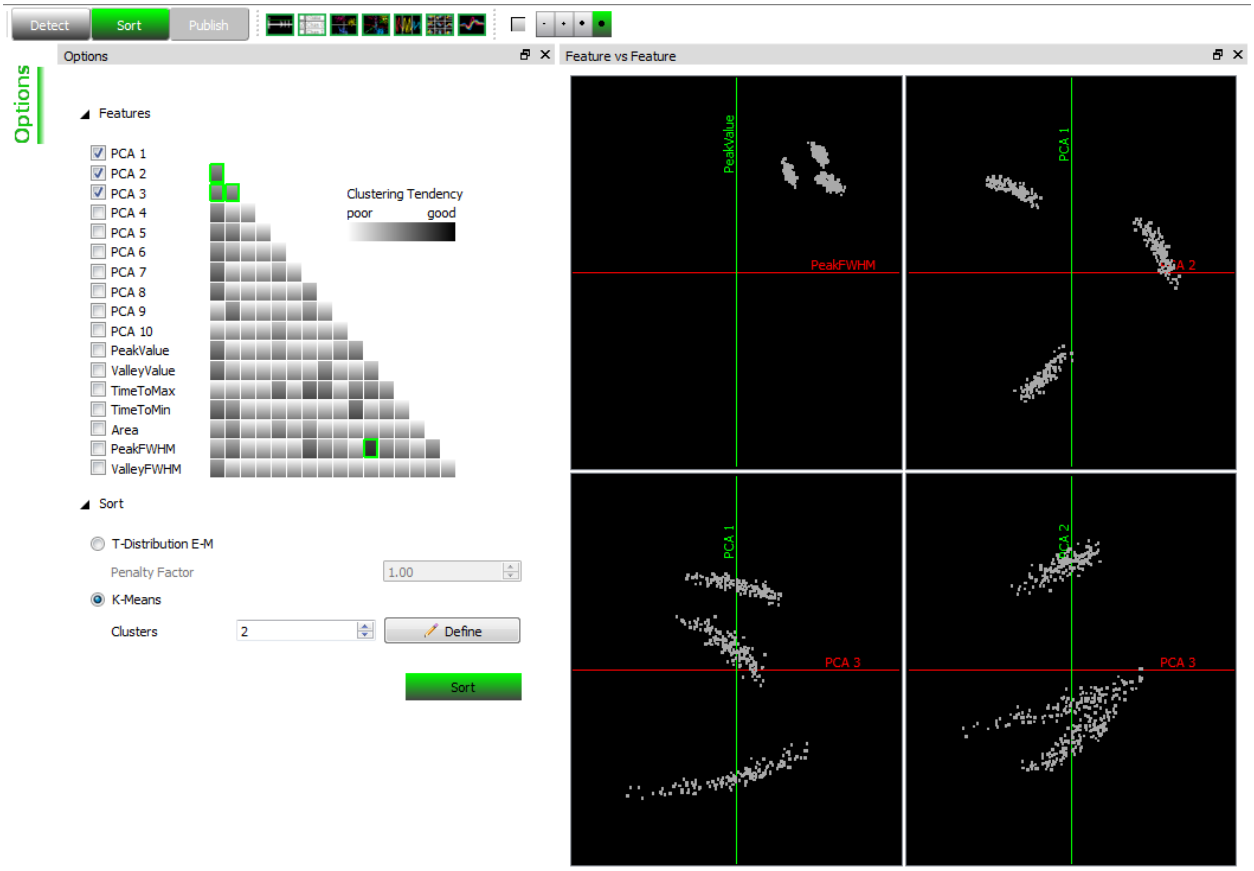
These two widgets display histograms of continuous data. The Peak Heights Histogram is displayed using a linear scale while the Energy Histogram uses a logarithmic scale.



These views are only available when a continuous data file is loaded.

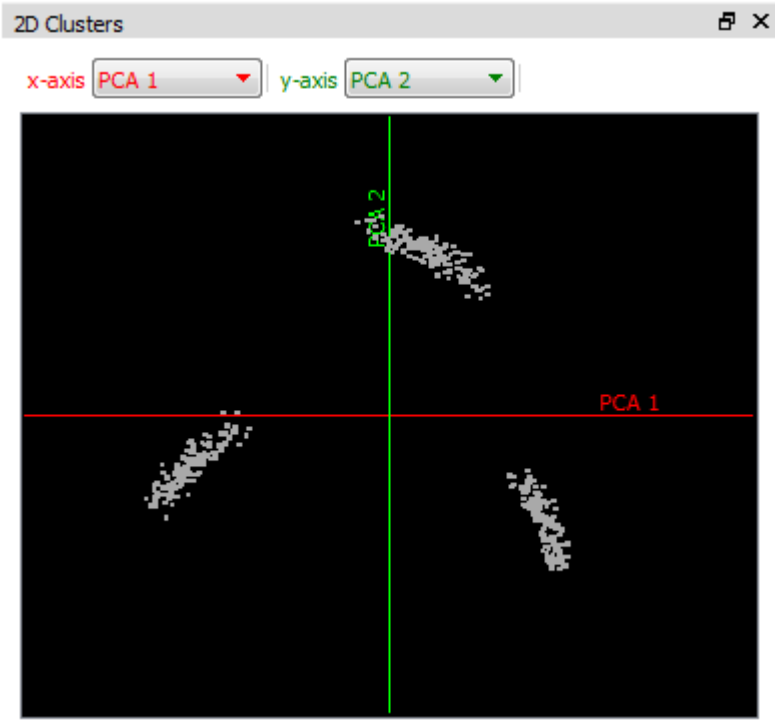
Feature vs. Feature

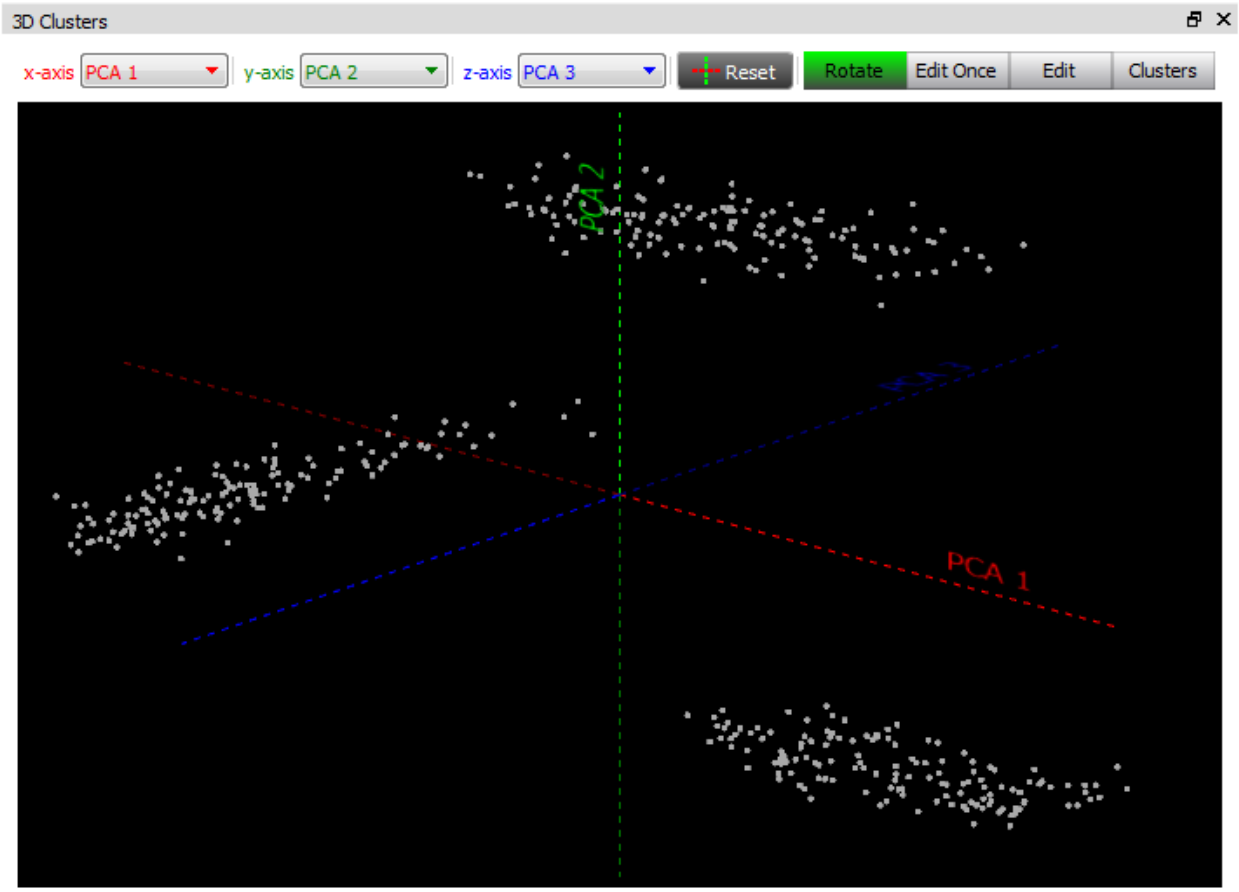
The Feature vs. Feature widget is a combination of individual Feature vs. Feature plots. The displayed plots correspond to the selected boxes in the Clustering Tendency half matrix, available in the Feature Options. You can select and unselect boxes in the matrix to display and hide those plots, respectively.



2D and 3D Features

These two widgets display the feature space defined by the combo boxes in their top toolbars.



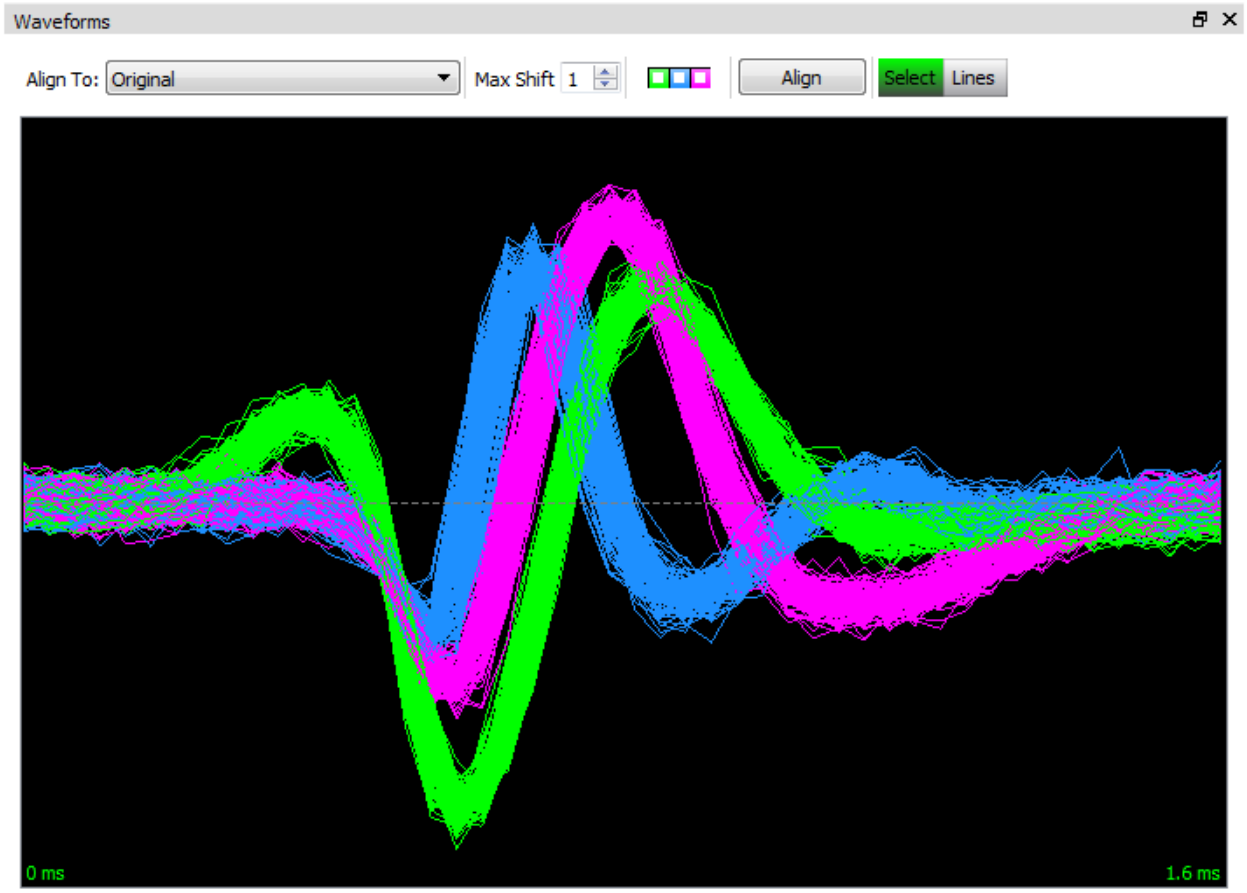


Depending on the mode of the 3D feature view (Rotate, Edit, Edit Once or Centroids), mouse movements and clicks have different effects:

- **Rotate:** Click and hold the mouse button and any movement of the mouse will cause a rotation of the display.
- **Edit:** Click the display and hold the mouse down to draw a free-hand enclosure to select waveforms. Right click to open the edit menu and chose an action to apply to the selection.
- **Edit Once:** After one editing action, the mode immediately turns back to Rotate.
- **Clusters:** Click the mouse near a cluster centroid and while pressing the CTRL key, keep defining cluster centroids in the data. These centroids can be used to initialize K-Means algorithm.

Waveforms

All waveforms corresponding to the displayed portion of the file (the data portion in the Timeline viewport) are superimposed. If units have been defined in the data, the average units are displayed with dash lines.



After detection with threshold-based methods, the waveforms are aligned to their threshold crossings. However, you can re-align the waveforms using the top toolbar within this display. The alignment controls are as follows:

- **Align To**: Select whether you want to align to the First Local Min, First Local Max, First Local Extremum, First Local Extremum After Threshold, Global Min or Global Max.
- **Max Shift**: The maximum shift for any waveform in samples. If a waveform needs to be shifted by more than this value, it will not be aligned.
- **Unit Selection Bar**: When a unit checkbox is selected (filled with color) the alignment options will apply to that unit. You can select several units for alignment and the waveforms of that unit will be aligned independently of other units. If no unit is selected, all waveforms will be aligned regardless of the unit assignments.
- **Align Button**: Click this button to apply your alignment options to the data.

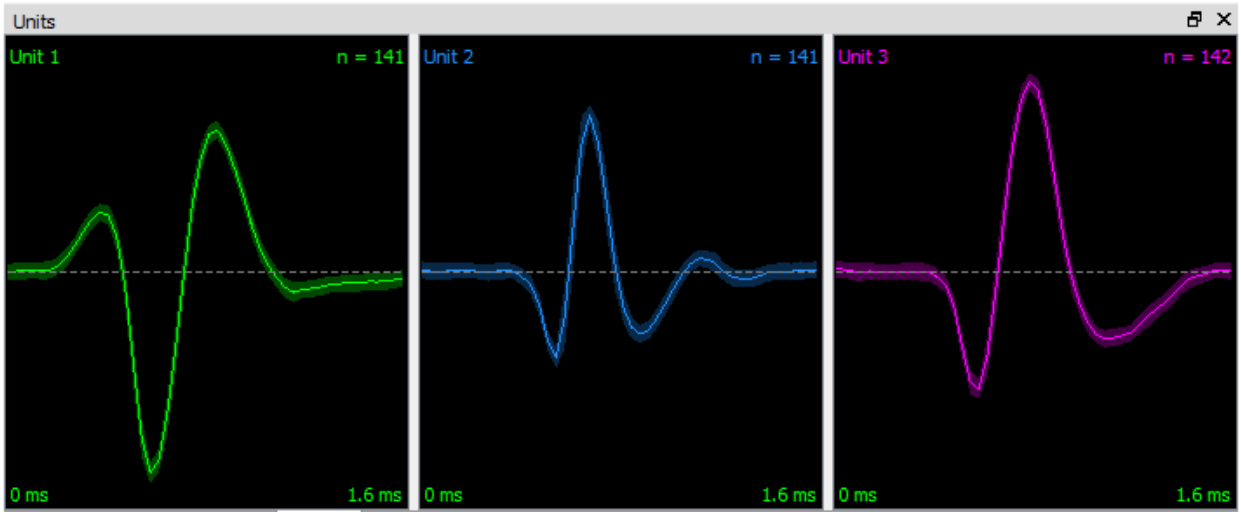
Modes of Operation for Manual Editing:

The Waveforms view has two modes of operation: *Selection* and *Lines*. When in *Selection* mode, right clicking on the view and selecting an action from the Edit Menu will only affect the selected waveforms.

In *Lines* mode, you can define several segments in order to select waveforms that go through them. When an action is selected in the Edit Menu, this action will apply to all the waveforms in the file that fall within the defined segments, not only to those visible in the view.

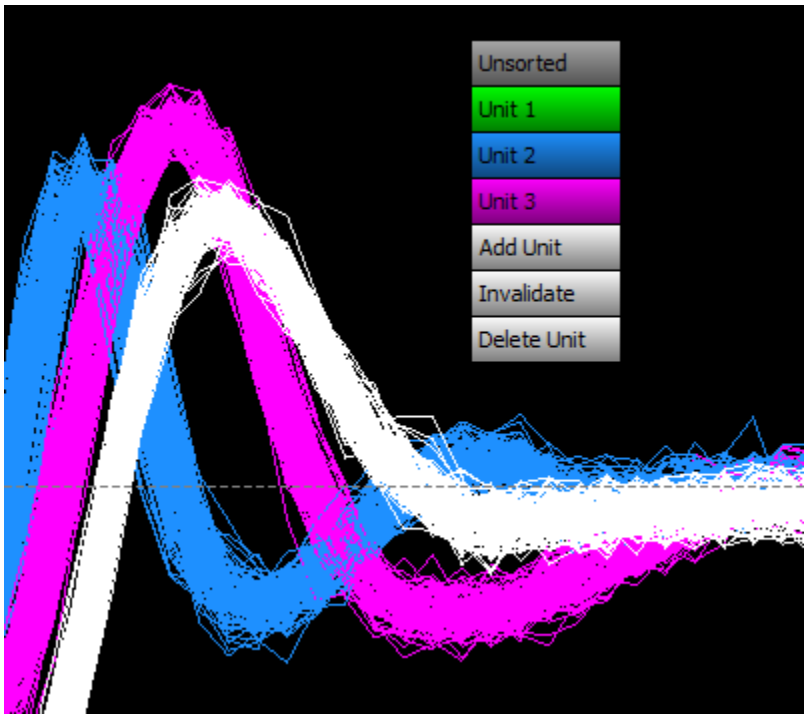
Units

The Unit view allows examining the average waveform for each unit and its standard deviation. By right-clicking on a unit, you can get a context menu with editing options for it.



Edit Menu

The Edit Menu can be accessed from any view with waveforms by right-clicking on the background. When there is an active selection, you will have the option of re-labeling the waveforms, add a new unit, invalidate the selection (mark it as noise) or delete a whole unit. When there are no waveforms selected, the only option available will be to delete a unit.

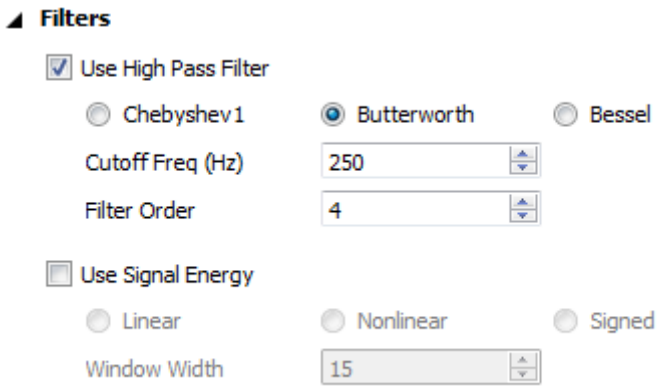


In the Unit view and the Visible Units buttons, the edit menu offers other unit-based options: invalidate, delete or combine with another unit.

DETECT SPIKES

After loading your continuous data file, select the channel that you want to analyze in the Channel Table.

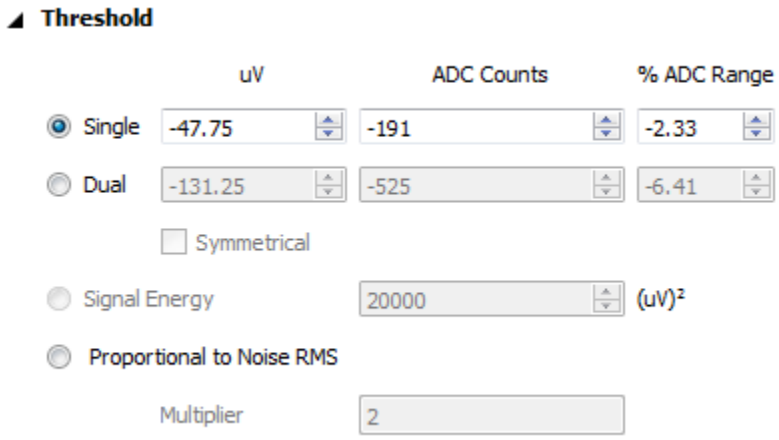
In the Filters section of the Options widget, apply the HPF of your choice by enabling the filter and selecting Chebyshev I, Butterworth or Bessel.



Select a cut-off frequency and order for the filter and the data will be updated to reflect your selection.

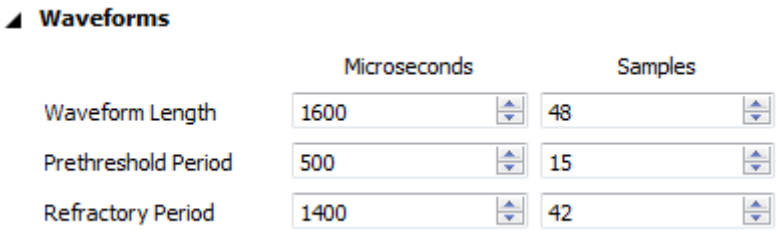
You can choose to use the energy of the signal for the detection. If that is the case, the Timeline will split to show the signal energy on top of the regular timeseries.

In the Threshold section, select the threshold that you want to use. Single or Dual thresholds are supported. You can change the threshold by typing the value on the Options widget or moving the threshold line in the Timeline or Histogram.



Also, you can let BOSS select the appropriate threshold by setting it proportionally to noise RMS.

Select the length of the snippets of data that will define a waveform in the Waveforms section and click the Detect button to detect waveforms.



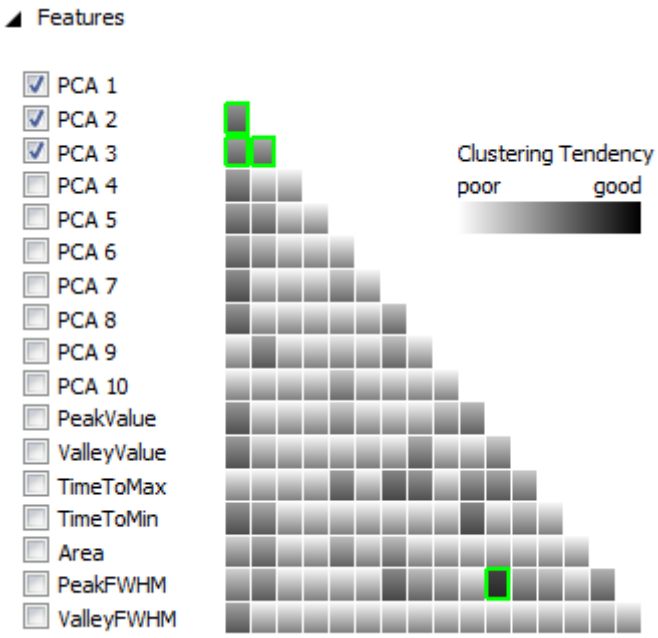
While BOSS processes the channel data, the Detect button and the channel name in the Channel Table will blink to indicate that the channel is being processed. Once finished, the blinking will stop. In the meantime, you can proceed to another channel and start processing it while the previous channel executes. BOSS will handle the processing of all channels in parallel.

ANALYZE FEATURE SPACES

Once the spikes have been detected in the continuous data or a neural event file has been loaded it is time to look at the different feature spaces and select the one that offers the best separation of clusters, eliminating all the dimensions dominated by noise. This step saves computational time as it reduces the dimensionality of the data fed to automatic clustering algorithms. Furthermore, eliminating inputs dominated by noise can certainly improve clustering outcomes. The major challenge is how to select which are the best features.

The Options widget corresponding to the Feature Extraction and Sorting steps displays a half-matrix for the different feature pairs. Each box of the matrix is colored according to its Clustering Tendency value.

This is an estimation of the pattern of the data in this feature space, representative of the quality of the separation of the clusters. You can click on these boxes to show and hide a specific feature space in the Feature vs. Feature view. Once a good subset of features have been identified for sorting, check the appropriate checkboxes to the left of the half matrix to indicate which features are going to be used for sorting.



CLUSTER SPIKES INTO UNITS

BOSS allows using manual methods as well as automatic algorithms for clustering the data. Manual methods include the definition of Lines and Manual selection of data points. Semi-automatic methods include T-Distribution EM and K-Means (with and without centroid initialization).

T-Distribution EM

This algorithm automatically determines the number of units, and identifies the neural responses from each source out of the composite signal. It implements an agglomerative mixture decompositions algorithm based on the Expectation-Maximization (EM) algorithm, where the distribution of waveforms from each unit is modeled as a multivariate t-distribution.

In order to tune this algorithm, the user can select different Penalty Factor values. The higher the Penalty Factor value, the greater the number of clusters being discovered. The default setting is 1.0.

For more details on the algorithm, please refer to:

Shoham S. et. al. Robust, automatic spike sorting using mixtures of multivariate t-distributions, Journal of Neuroscience Methods, 127(2), 111-122 2003.

K-Means

The standard K-Means algorithm is implemented in BOSS to allow for (semi-) automatic clustering of the data. In order to run it, an initial number of clusters can be defined. If no initialization of centroids is provided, the algorithm will be initialized with a random partition of the data. Providing initial centroids will ensure a faster convergence and more accurate results.

This implementation of K-Means allows empty clusters, which means that if the user requests three clusters but only two are discovered when the algorithm converges, only those two will be presented in the results. In this case, the algorithm will not try to force data points into a third cluster to meet the user request.

Lines

This algorithm uses a list of lines defined by the user in the Waveform view (i.e. time-voltage space) to classify individual waveforms into different units. A waveform will be classified into a unit when its voltage trace intersects with all the lines defined for that unit. By this criterion, if a waveform could be classified as two units, the resulting label will be the one of the first unit for which its lines intersect with it.

New lines can be defined by switching to Lines mode in the Waveform view and hand selecting the lines' ends by clicking on the view. Once defined, they can be adjusted by clicking on the ends and moving them to the desired position. To delete lines, click on a line and press Delete on the keyboard. After all lines are defined for a unit, right-click on the view and select the unit for them.

Manual Selection

BOSS allows using manual methods for cluster assignment. All views containing waveforms (Timeline, Waveforms, Feature vs. Feature, 2D Features and 3D Features) allow selecting data points or waveforms by left clicking on them. You can either click to select single waveforms or click and hold the mouse button down for free-hand drawing a line and select a subset. Also, the CTRL key can be used to add more data points to an existing selection.

Once a selection is in place, you can right-click on any view to access the Edit Menu.

SAVE RESULTS

When a file is opened in BOSS, partially processed and then closed, BOSS will automatically save the editing session so the user can later resume his analysis where it was left off. The next time the same file is opened, BOSS will offer to continue the same editing session or start a new one.

In addition, results can be saved into a .NEV file for further processing in other software packages. To do so, simply select **File Menu→Save As** and save your file.