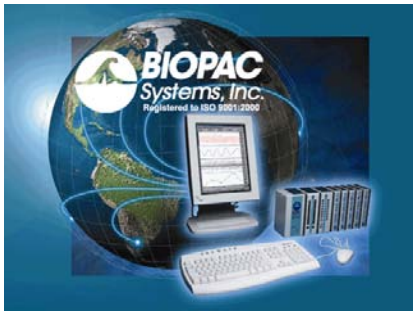
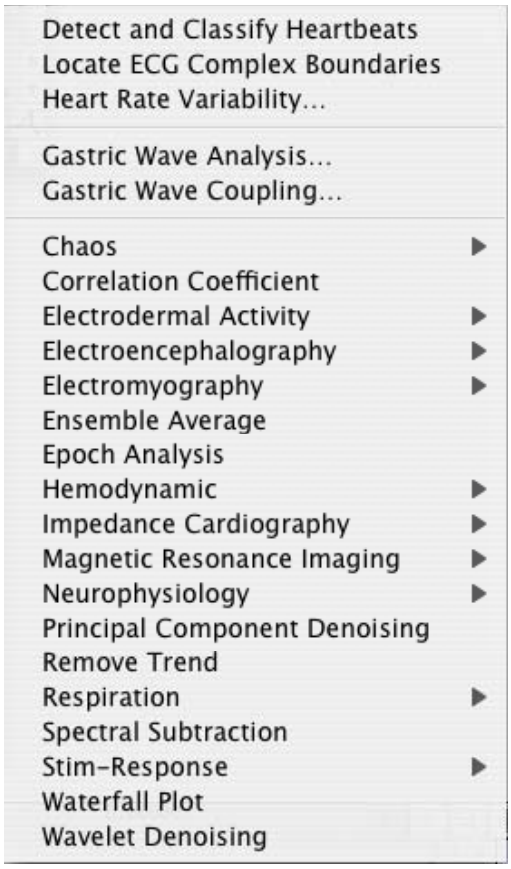


AcqKnowledge™

Analysis Scoring & Automation

For Life Science Research Applications

Data Acquisition and Analysis with BIOPAC MP Systems
Running on Windows® or Mac OS X™



The Specialized Analysis package includes comprehensive analysis tools to automate analysis to save hours (or days!) of processing time and standardize interpretation of results.

- Acq*Knowledge*™ 4 includes a courtesy copy of the Specialized Analysis Package under the Analysis menu.

Specialized Analysis provides extensive post-acquisition analysis options similar to modules from Mindware Technologies, PONEMAH Physiology Platform, EMKA Technologies, SA and other advanced analysis applications. If you need still more analysis options, save the data as MatLab, Igor Pro, PhysioNet, raw, or text format—or compress the file to reduce file size by about 60%.

Analyze data collected on MP System (MP150 or MP36R)s with Windows OS or Mac OS X.

*See these other **Analysis** menu items for more operations that derive data and measurements from the graph*:

Histogram
Autoregressive Modeling
Nonlinear Modeling
Power Spectral Density
AR Time-Freq Analysis
FFT
DWT
Principal Component Analysis
Independent Component Analysis
Find Cycle
Find Next Cycle
Find All Cycles
Find Rate

The Analysis package includes the following Analysis Packages and Classifiers:

Analysis package—bundle of transformations created to assist with analysis in a specific area of research.

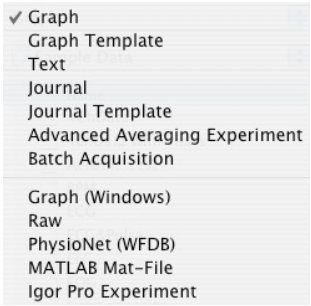
Classifier—special-purpose transformation that defines events at well-known points of interest on standard waveforms, such as the ECG wave boundary classifier and the QRS beat detector and arrhythmia detector.

- Detect and Classify Heartbeats
- Locate ECG Complex Boundaries
- Heart Rate Variability
- Gastric Wave Analysis
- Gastric Wave Coupling
- Chaos Analysis
  - Detrended Fluctuation Analysis
  - Optimal Embedding Dimension
  - Optimal Time Delay
  - Plot Attractor
- Correlation Coefficient
- Electrodermal Activity
  - Derive Phasic EDA from Tonic
  - Event-related EDA Analysis
  - Locate SCRs
  - Preferences: Output Display Format; Phasic EDA Construction Method: Smoothing Baseline Removal or High Pass Filter
- Electroencephalography
  - Compute Approximate Entropy
  - Delta Power Analysis
  - Derive Alpha-RMS
  - Derive EEG Frequency Bands
  - EEG Frequency Analysis
  - Remove EOG Artifacts
  - Preferences: Output Display Format
- Electromyography
  - Derive Average Rectified EMG
  - Derive Integrated EMG
  - Derive Root Mean Square EMG
  - EMG Frequency & Power Analysis
  - Locate Muscle Activation
  - Preferences: Output Display Format
- Ensemble Average
- Epoch Analysis
- Hemodynamic Analysis
  - Classifiers: ABP; LVP; MAP
  - Arterial Blood Pressure
  - ECG Interval Extraction
  - Left Ventricular Blood Pressure
  - Monophasic Action Potential
  - Respiratory Sinus Arrhythmia
  - Preferences: Output Display Format; LVEDP Location Method; dP/dt pk-pk %; MAP Plateau Location Method; dP/dt MAP pk-pk %
- Impedance Cardiography Analysis
  - Body Surface Area
  - ICG Analysis
  - PEP Pre-ejection Period
  - dZ/dt Derive from Raw Z
  - dZ/dt Classifier: B, C, X, Y, and O Points
  - dZ/dt Remove Motion Artifacts
  - Ideal Body Weight
  - VEPT
  - Preferences: Output Display Format; C-, B-, and X-Point Location; Stroke Volume Calculation Method; Body Measurement Units; Body Surface Area Method; Ideal Weight Estimation Method; dZ/dt Max Method
- Magnetic Resonance Imaging
  - Artifact Frequency Removal
  - Artifact Projection Removal
- Median Filter Artifact Removal
  - Signal Blanking
- Neurophysiology
  - Amplitude Histograms
  - Average Action Potentials
  - Generate Spike Trains
  - Find Overlapping Spike Episodes
  - Set Episode Width and Offset
  - Classify Spikes
  - Dwell Time Histograms
  - Locate Spike Episodes
  - Preferences: Detect Spike; Default Episode Width; Default Episode Offset; Default # of Spike Classes
- Principal Component Denoising
- Remove Trend
- Respiration
  - Compliance and Resistance
  - Penh Analysis
  - Pulmonary Airflow
- Spectral Subtraction
- Stim-Response
  - Digital Input to Stim Events
  - Stim-Response Analysis
- Waterfall Plot
- Wavelet Denoising

Acq*Knowlege* File Portability Windows←→Mac OS 10.3 or higher

Use Specialized Analysis to analyze Acq*Knowledge* data files collected on MP System (MP150 or MP36R)s running on Windows/PC or Mac OS X. Specialized Analysis allows you to open/save the following file formats:

Opening files for Specialized Analysis

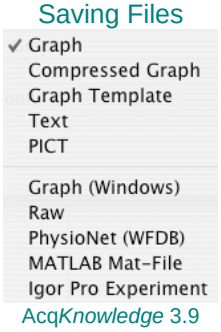


The default file formats (Graph and .ACQ) are referred to as "Acq*Knowledge*" files. The Acq*Knowledge* file format is the standard way of displaying waveforms in Acq*Knowledge*. These files are stored in a compact format that retains information about how the data was collected (i.e., for how long and at what rate) and takes relatively little time to read in (compared to text files, for instance). Acq*Knowledge* files are editable and can be modified and saved, or exported to other formats using the Save as command.

File Compatibility

- Mac Acq*Knowledge* 3.9 can open and create PC-compatible Graph (*.acq) and Graph Template (*.gtl) files. Variable sampling rate information and hardware settings are retained, and Journals can be read from and written to PC files. Files must end on a multiple of the lowest channel sampling rate to be fully PC compatible.

Saving files after Specialized Analysis



Acq*Knowledge* 3.9

The default file format for the File>Save as command is to save files as an Acq*Knowledge* file. Selecting Graph (MPWS) or .ACQ (MPWSW) from the popup menu in the Save As dialog box will save a file as an Acq*Knowledge* file, which is designed to be as compact as possible. These files can only be opened by Acq*Knowledge*, but data can be exported to other formats once it has been read in.

The Options button generates a dialog box that allows you to save only a portion of your file. When the "Selected Section only" option is enabled, only the data that has been selected with the I-beam tool will be saved. This option saves the selected area to another file and does not affect the current file that you are working in.

File Compatibility

Windows Acq*Knowledge* 3.9 and above files can be opened with MacAcq*Knowledge* 3.9 and above, but some advanced features may not transfer.

Mac Acq*Knowledge* 3.9 and above can save as "Graph (Windows)" files, but it saves in Windows Acq*Knowledge* 3.7.1 format. In this earlier format, all data is retained, but newer Acq*Knowledge* features (like dual stimulation, data views, embedded archives, etc.) are lost along with any settings specific to Mac Acq*Knowledge* (like events, adaptive scaling settings, etc.).

- Mac Acq*Knowledge* 3.9 and above can save PC-compatible Graph (*.acq) and Graph Template (*.gtl) files. Variable sampling rate information and hardware settings are retained, and Journals can be read from and written to PC files. Choose the format "Graph (Windows)" to create PC-compatible files.

The Mac version does not save PC GLP files or compressed PC files.

Files must end on a multiple of the lowest channel sampling rate to be fully PC compatible.

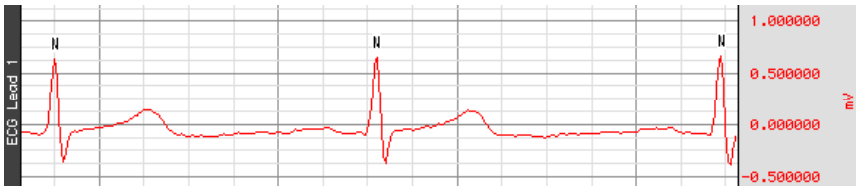
Excel Spreadsheet Export—The Specialized Analysis tools have been updated to automatically export their results to an Excel spreadsheet if desired. The spreadsheet contents mirror the tabular Journal text output. All the spreadsheets are saved as temporary files, so they need to be re-saved in order to be saved permanently.

- Also available for File > Save As, File > Save Journal Text As, and Find All Cycles journal.

Note Specialized Analysis scripts are complex and undo may not function for all steps.

Some of the specialized algorithms are very complex and processor intensive, so they may take a long (even *very* long) time to return a result.

Detect and Classify Heartbeats

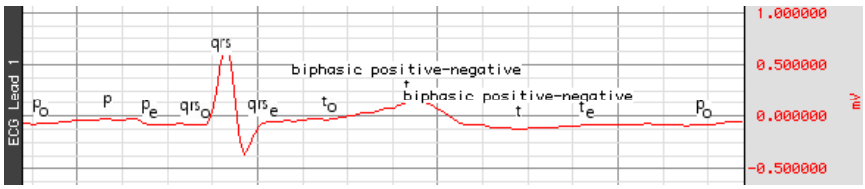


This robust QRS detector is tuned for human ECG Lead II signals. It attempts to locate QRS complexes and places an event near the center of each QRS complex to identify the type of heartbeat event:

- Normal The beat was recognizable as a valid heartbeat falling in a human heartbeat rate.
- PVC The beat was shorter than the beats around it and may be a pre-ventricular contraction. These events can be found in the "Hemodynamic > Beats" submenu of the event type listing.
- Unknown The beat wasn't recognizable as a valid heartbeat. This may occur on the first beat prior to the QRS detector locking onto the signal. It may also occur if tracking is lost due to changes in signal quality.

The Cycle/Peak detector may be used with these events to perform further cardiac analysis.

Locate ECG Complex Boundaries



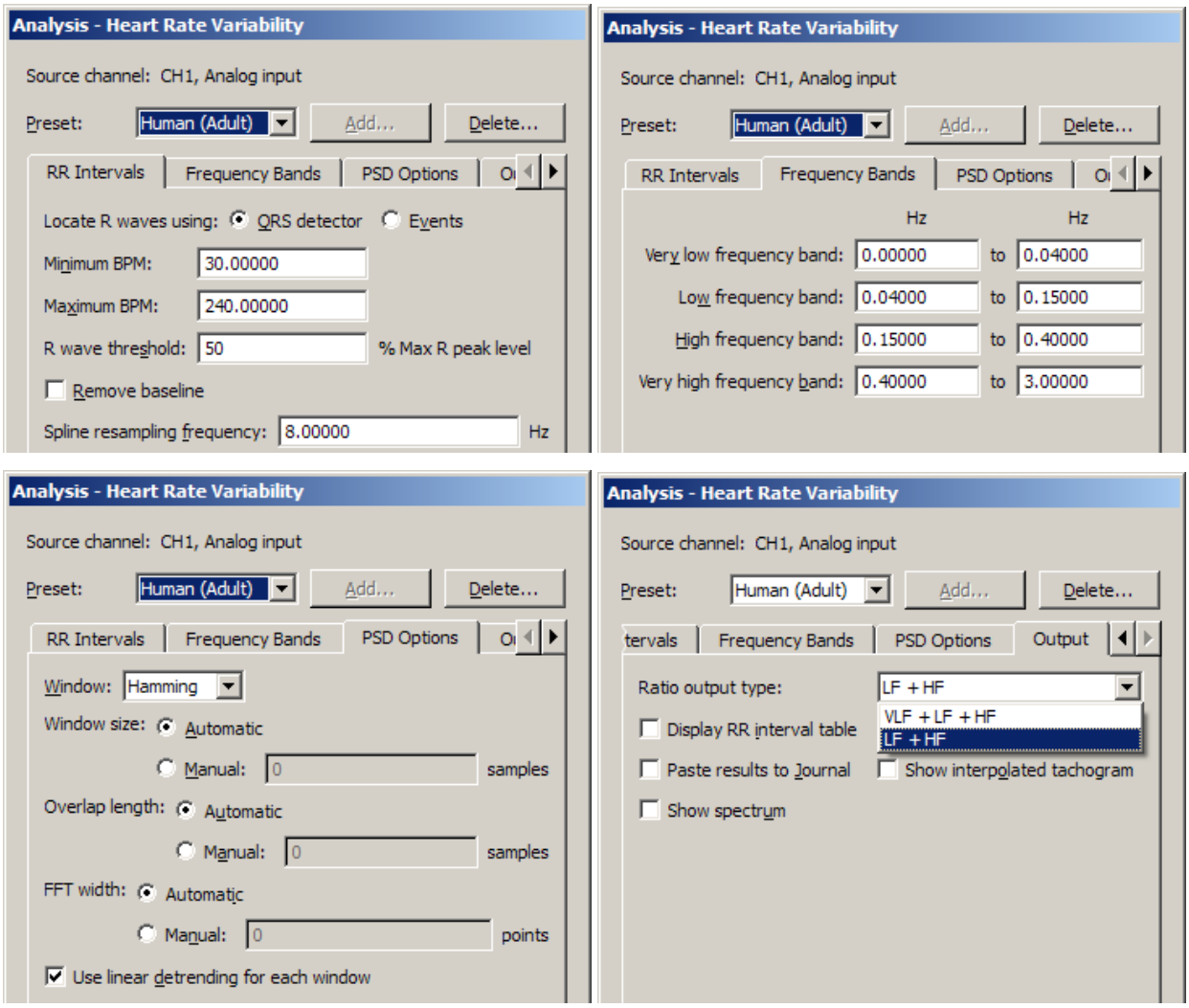
Locate ECG Complex Boundaries performs ECG waveform boundary detection for human ECG Lead II signals; ECG signals must be sampled at 5 kHz or below to be analyzed with this classifier. It will attempt to locate the boundaries of the QRS, T, and P wave and will define events for each individual complex. It will attempt to insert the following events; all of these complex boundaries can be found in the "Hemodynamic > ECG Complexes" submenu of the Event Type listing.

Wave	Type	Event Placement & Description
QRS	Onset	Before the beginning of the Q wave
	Peak	At the top of the R wave
	End	After the end of the S wave
T-wave	Onset	At the onset of T
	Peak	At the peak of the T wave *Note:* This may not be a positive peak if the T-wave is inverted. If the T-wave seems to be bi-phasic, two T-wave events will be inserted and the event description will indicate that the T-wave is bi-phasic.
	End	At the end of T
P-wave	Onset	At the onset of P
	Peak	At the top of the P wave *Note*: This may not be the absolute maximum, but rather the likely center of P.
	End	At the end of P

The Cycle/Peak detector may be used with these events to perform further cardiac analysis.

Heart Rate Variability

New parameter settings for the HRV algorithm function better on shorter ECG signals and correspond more closely with other implementations.



Heart rate variability is the examination of physiological rythms that exist in the beat-to-beat interval of a cardiac signal. Heart rate variability assists in performing frequency domain analysis of human ECG Lead II data to extract standard HRV measures. The HRV algorithm in Acq*Knowledge* 3.9 and above conforms to the frequency domain algorithm guidelines as published by the European Heart Journal. HRV processing in Acq*Knowledge* consists of three stages:

1. The RR intervals are extracted for the ECG signal.
   - A modified Pan-Tompkins QRS detector is used.
2. The RR intervals are re-sampled to a continuous sampling rate in order to extract frequency information.
   - Cubic-spline interpolation is used to generate this continuous time-domain representation of the RR intervals.
3. The frequency information is extracted from the RR intervals and analyzed to produce standard ratios. Power sums are reported in units of sec^2.
   - A Welch periodogram is used to generate the Power Spectral Density (equivalent to Transform > Power Spectral Density).

The initial implementation of the HRV algorithm was primarily for use with long duration recordings. HRV algorithm improvements allow for further customizations to the algorithm:

- Windowing type for FFTs used to construct the PSD may be changed between Hamming, Hanning, and Blackman
- Overall window length for segmenting source data for individual FFTs to include in PSD average may be modified
- Length of the individual FFTs in the average can be manually specified
- Scaling has been changed for PSDs, which are now scaled relative to the sampling frequency
- Summary of power in individual frequency bands has been changed
- Instead of a straight sum, an average power value is now reported
- Power at endpoints is halved (e.g. divided by 2)
- Sympathetic/Vagal ratios may optionally include the very low frequency band in the total power estimate
- The modifications to the HRV algorithms that affect its power spectrum estimation have also been applied to the PSD transformation.

After selecting Analysis > Heart Rate Variability, choose the appropriate tab(s) and establish settings.

Preset controls, Transform entire wave checkbox, and OK/Cancel buttons apply across all of the tabs.

Preset—The preset menu can be used to save a variety of HRV settings, including: beat detection parameters, spline resampling frequency, and frequency band ranges. Choose a preset from the popup menu to apply its settings. To construct a new preset with the currently displayed settings, choose Add New Preset. A default preset for adult human subjects is supplied.

IMPORTANT—Recording good data is essential for performing HRV analysis. The protocol for data acquisition, filtering, artifact detection and correction in Application Note 233 results in great improvements in HRV analysis.

> "Results reveal that even a single heart period artifact, occurring within a 2-min recording epoch, can lead to errors of estimate heart period variability that are considerably larger than typical effect sizes in psychophysiological studies." —Berntson & Stowell, 199

- See **Application Note 233 Heart Rate Variability—Preparing Data for Analysis Using Acq*Knowledge* (online at www.biopac.com)**

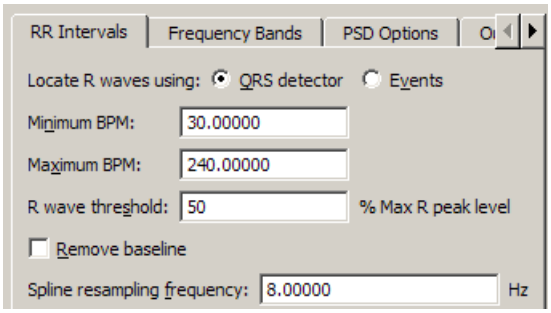
The note explains how to optimize ECG R-R interval data for Heart Rate Variability studies by using a template matching approach. It also explains how to identify erroneous R-R interval values caused by signal artifact and shows methods for correcting the errors by using the tools in the Acq*Knowledge* software. The note explains how to:

A. Record good ECG data
B. Prepare data for the tachogram
    1. Filter the ECG data
    2. Transform the data using Template Correlation function
C. Create a tachogram
D. Identify problems with the tachogram data
E. Correct tachogram data

RR intervals

Select a method to locate R waves: QRS Detector or Events.

QRS detector

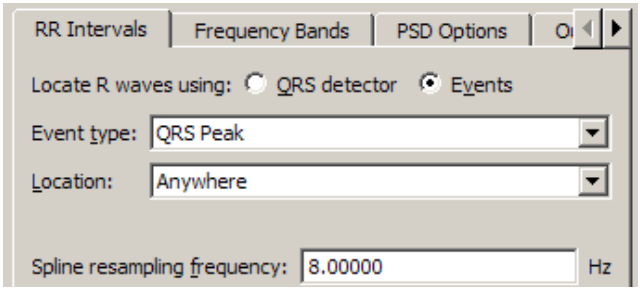


The heart rate variability implementation has a built-in QRS detector. The detector does not run on raw source data; it uses a modified Pan-Tompkins algorithm to normalize the ECG data to 1, whereby the peak amplitude of the highest R-wave represents 1. Use the tachogram output to examine the output of the QRS detector.

- *R wave threshold*—Starting with Acq*Knowledge* 4.1, the detection threshold must be specified in terms of percentage of maximum R peak level; this helps to clarify the units in which this threshold is expressed. The default threshold level of .5 should place the threshold in the middle of the R-wave, which should function on a wide range of data sets. If the R-wave amplitude varies a lot, it might be necessary to adjust the threshold level.
    - R wave threshold is expressed in normalized units, which are in the range (-1, 1): positive for positive R wave peaks. The maximum voltage in the signal maps to 1.0 and the minimum voltage in the signal maps to -1.0.

      Pan J and Tompkins WJ. A Real-Time QRS Detection Algorithm. *IEEE Transactions on Biomedical Engineering* ***32***(3):230-236, 1985.

Events



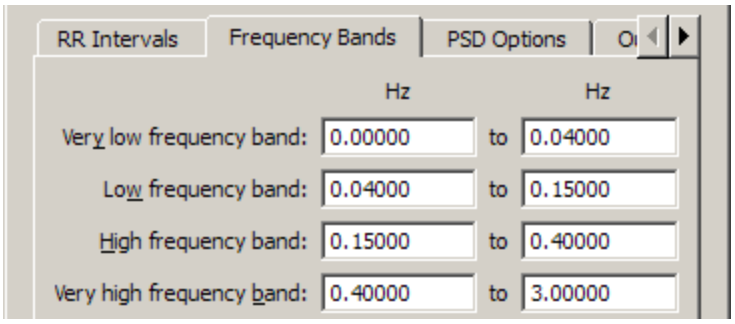
R-wave peaks will be located using events already in the graph of the channel of data to be analyzed. This assumes a single event is placed at each R-wave peak and that all of the R-peak events are of the same event type. When using events, the buit-in QRS detector is not used; the exact positioning between the events on the channel is used to extract the RR intervals.

By using events, it is possible to use other QRS detectors within Acq*Knowledge* for performing HRV analysis. It is also possible to apply spectral HRV-style analysis to data in other domains as long as intervals can be reduced to events.

Spline resampling frequency

For highest accuracy, set to no less than twice the topmost frequency of the very high frequency band.

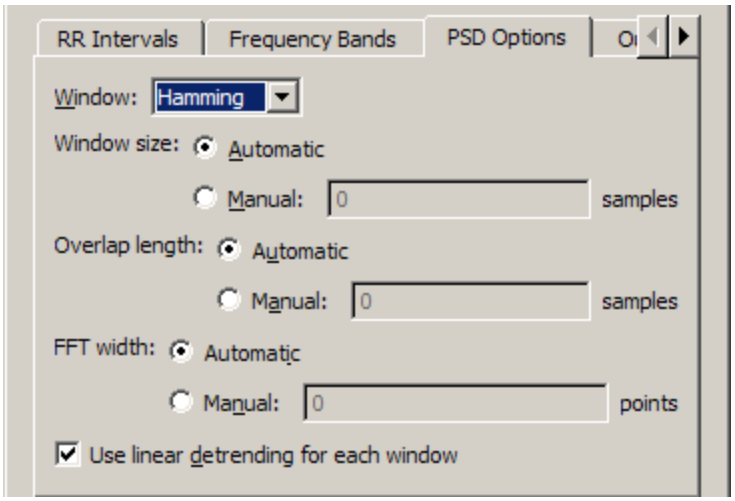
Frequency Bands



Enter the start and end of each specified frequency band to adjust the boundaries of the frequency analysis. They are preset to the frequency ranges recommended by the *European Heart Journal*. Output of derived parameters is presented in a dialog and may also be pasted as text to the Journal.

- Very high frequency band, usually used in rat studies, is disabled if the spline resampling frequency is less than the upper bound of the very high frequency range.

PSD Options



PSD Options establish parameters for the power spectral density transformation used to compute the spectrum from the interpolated tachogram; the options contained in this tab mirror the controls of the Analyis > Power Spectral Density transformation.

The use of linear detrending in each individual segment of source data prior to the windowed periodogram analysis has been allowed to be turned on and off. When off, the algorithm may be tuned to correspond to implementations that do not apply linear trending, such as MATLAB, which uses windowing only. The new PSD options have also be added to the power spectral density transformation so users can regenerate the spectrum from either the raw or interpolated tachogram output as necessary.

After the user modifies the parameters for the PSD transformation, those parameters will become the new default values each time the dialog is displayed. When the application is relaunched, the default settings will be used (user changes are not persistent).

PSD output is scaled so power values are scaled by the sampling rate. That is:

$$PSD_{new} = \frac{PSD_{old}}{f_s}$$

Window Used to change the window that is applied to each segment of the source data prior to computing the PSD to be included in the average. Includes the following options:

Hamming Hanning Blackman

Window size The specified number of samples must be a power of two. Note that the window function is applied to the entire window width of the data; using a subset of the windowed data will not include the final portion of the windowed data.

- If the FFT size is less than the window size, only a subset of the windowed sample data will be used.

Automatic If selected, the window size is selected automatically depending on the size of the source data. For a data length of *n* samples, choosing this radio button will use the window size:

$$\frac{n}{4.5}$$

Manual If selected, the window size will be input manually by the user in the associated edit field. The window size must be greater than three and must be less than the length of the data selection. Users will be warned on invalid window sizes when attempting to click OK.

Overlap length After each individual FFT, the window of source samples is shifted over by a certain amount to compute the next FFT, so there is an overlap of source samples in successive windows of source for the next FFT in the average.

Automatic If selected, the number of samples to overlap successive windows will be computed automatically. Given a window length *L* computed according to the window width choices, choosing this radio button will use an overlap number of samples:

$$\frac{L}{2}$$

Manual If selected, the number of samples to overlap successive windows of source data. Overlapping reduces windowing artifacts The overlap length must be positive and must be less than the window size. Users will be warned on invalid overlap lengths when attempting to click OK

FFT width Automatic If selected, the number of points to use for each individual FFT will be computed automatically. Given a window length *L* computing according to the window width choices, the number of points in the FFT will be set to:

$$N_{fft} = \begin{cases} 256, L < 256 \\ 2^{\left\lceil \frac{\log(L)}{\log(2)} \right\rceil}, L \geq 256 \end{cases}$$

- The number of points in the FFT is set to 256 if the window width is less than 256. Otherwise the length is set to the next power of two higher than the window width.

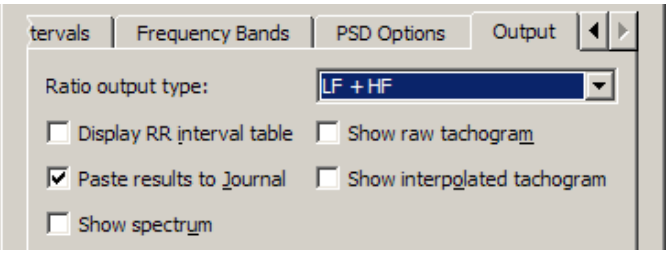
Manual — If selected, the number of points in the FFT will be specified manually in the edit box to the right of the radio. The number of points in the FFT will be required to be a positive power of two. It is recommended that the FFT length be set larger than the window size. If longer than the window size, zero point padding is used. Users will be warned on invalid FFT number of points when attempting to click OK. If the user inputs a number of points for the FFT that is shorter than the window width, a confirmation dialog will be displayed to the user warning that the windowing is shorter than the requested FFT width and asked if they want to continue.

Use linear detrending for each window — When enabled, linear regression detrending is applied for each individual segment prior to the FFT computation. When unchecked, windowing only is applied.

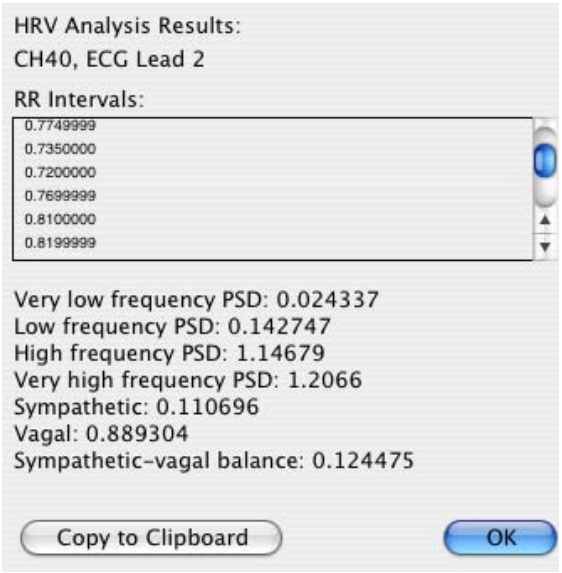
Transform entire wave — When enabled, the entire waveform is delayed. When unchecked, only the selected area is delayed.

- If there is no selection in the graph, the checkbox is enabled and dimmed.
- As the selection changes in the graph with the selection palette, the state of this checkbox is updated.

Output



Create standard result presentation graphs or assess performance of the HRV algorithm. Output options allow access to intermediate computation data for algorithm validation and/or measurements.



RR Interval table

1. If the combined output formula is selected, the analysis output will contain an additional line of text: "VLF Ratio" with the corresponding percentage.

Spectrum

Displays the power spectrum density (PSD) estimation from which the PSD summations and sympathetic/vagal ratios are computed.

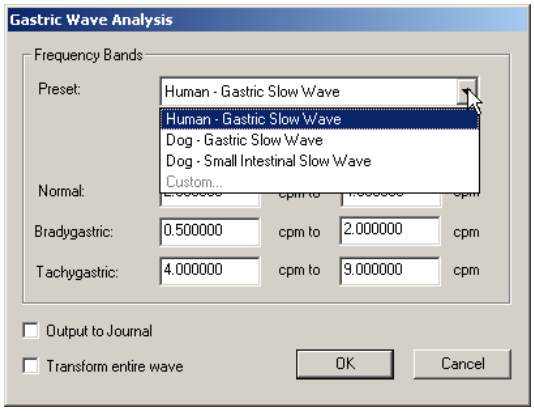
Raw tachogram

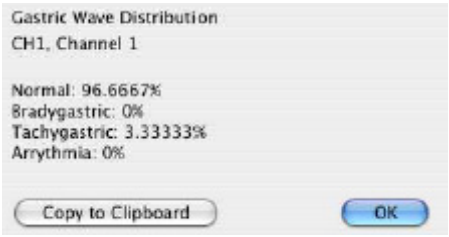
Plots the raw R-R intervals found by the QRS detector. Perform statistical HRV measures on the R-R intervals without exporting the textual R-R table to excel.

Interpolated tachogram

Plots the resampled R-R intervals after cubic spline interpolation is applied and extracts the PSD from this data.

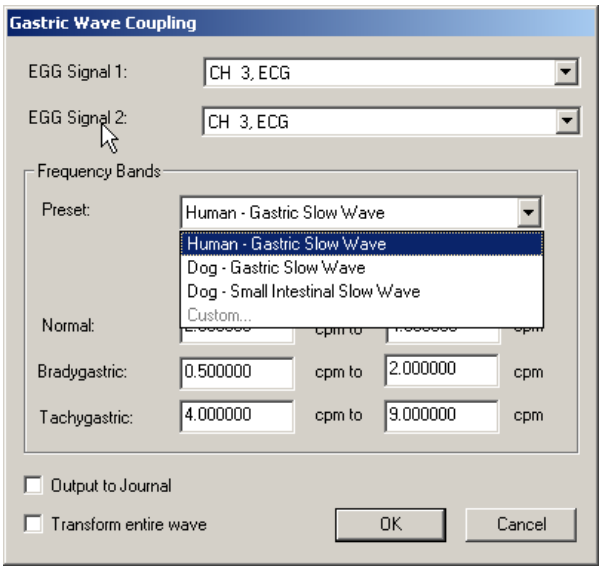
Gastric Wave Analysis

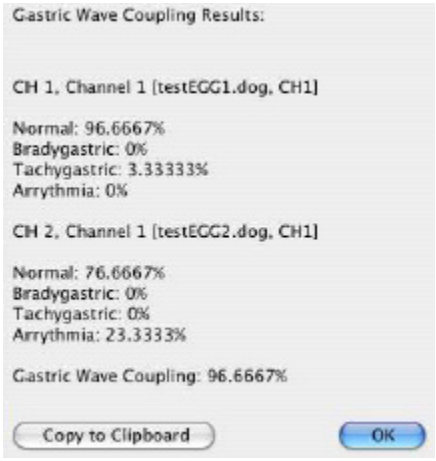




Gastric Wave Analysis uses autoregressive time-frequency analysis to determine the classifications of gastric waves present in an EGG signal. The single wave analysis determines the percentage of gastric waves that fall within the frequency bands corresponding to normal, bradygastric, and tachygastric waves. The analysis also indicates the percentage of waves that fall outside of these boundaries and are arrhythmias. The frequency bands are expressed in units of "contractions per minute" and may be adjusted by the user. Presets for commonly used subject and wave types are predefined; you may extend these presets with your own.

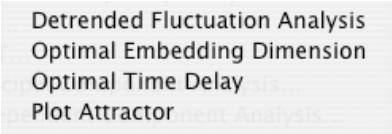
Gastric Wave Coupling





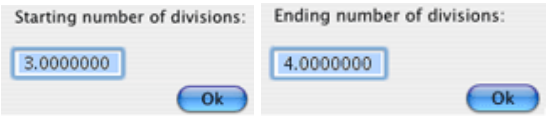
Gastric Wave Coupling takes two EGG signals and uses autoregressive techniques to classify the contractions in those signals according to user-configurable frequency bands (similar to single channel Gastric Wave Analysis). In addition to providing classification information for the two signals, Gastric Wave Coupling provides a measure of the percentage of coupling between the two signals—this measure that can be used to determine the amount of slow-wave propagation across the stomach.

Chaos Analysis



The “Chaos” analysis package assists the user in exploring the chaotic nature of data, including measurement selection and visualization of time domain attractors in the data.

Detrended Fluctuation Analysis



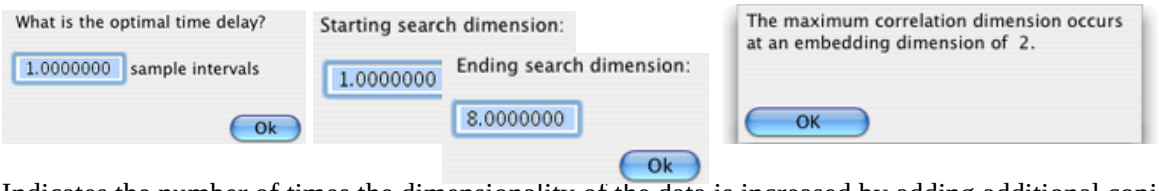
Modified root mean square analysis, useful for evaluating self-similarity in a long-term, non-stationary data series. Source data is mean-adjusted and then integrated; it is then split up into *n* segments of equal length, and in each segment, via linear regression, the best fit least squares line is computed. For a particular value of *n* and a number of samples *N*, the characteristic fluctuation of the piecewise linear fit y_n. is defined as:

$$F(n)=\sqrt{\frac{1}{N}\sum_{k=1}^{N}[y(k)-y_n(k)]^2}$$

F(n) is evaluated over a user-specified range for the number of divisions. *n* will equal the total length divided by the number of divisions. A log-log plot of the interval width *n* in samples versus the corresponding value of *F(n)* will be created. If a linear relationship appears to exist in this graph, then the source signal displays some form of self-similarity. The slope of the line in this graph is related to the scaling exponent.

➲ For more information on Detrended Fluctuation Analysis, see http://www.physionet.org/physiotools/dfa/

Optimal Embedding Dimension

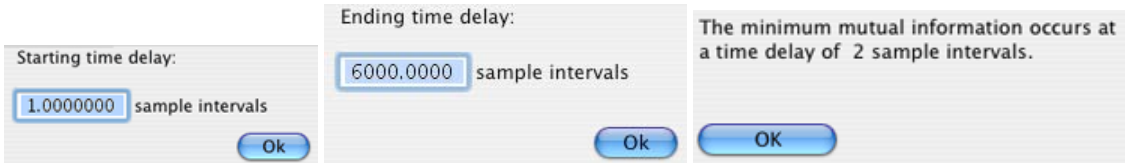


Indicates the number of times the dimensionality of the data is increased by adding additional copies of the data. Many of the fractal measurements take an embedding dimension parameter. Increasing the dimensionality of the data may improve the quality of the results. In general, embedding dimensions should always be less than 8.

After the most relevant time delay for the data has been selected, Optimal Embedding Dimension assists in choosing the embedding dimension that appears to give the most accurate results. The embedding dimension is chosen to be the earliest dimension in the search range where the fractal correlation dimension measure reaches a local maximum. This indicates the lowest dimension where the data has the potential to exhibit the most self-similarity.

- Since real data may not be fractal in nature, there may be no local maximum for the embedding dimension. In this case, it is not possible to determine the optimal dimension.

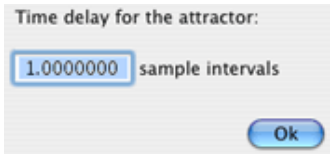
Optimal Time Delay

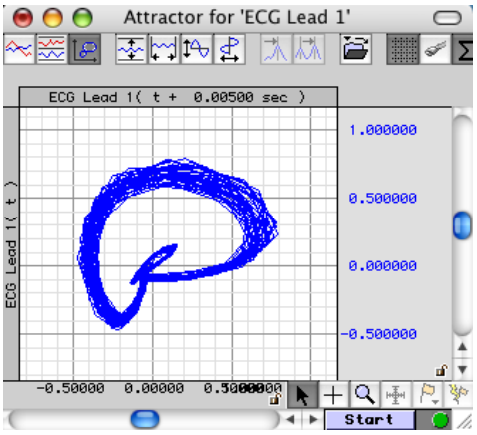


Assists in picking a time delay that is most relevant for the data. It runs through and locates the earliest time delay in the specified interval range where the mutual information measurement reaches a local minimum. Optimizing the time delay in this fashion picks the shortest delay where the signal exhibits the most independence with respect to its time-delayed version.

The fractal dimension and other chaos-related measurements operate on a single channel of data. In the process of extracting these measures, a signal is compared with a time-delayed version of itself to examine the patterns in dynamics of the data. These measures take a fixed time delay setting. The Optimal Time Delay transformation can be used to choose the best value for the parameter.

Plot Attractor

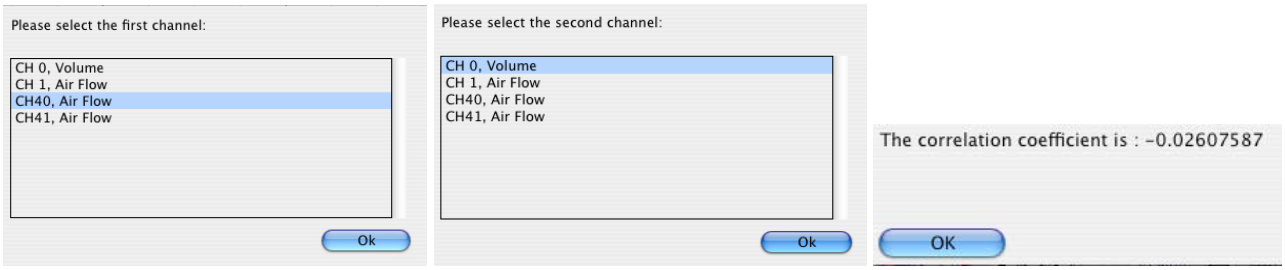




Assists in constructing X/Y plots for the attractors of time delayed data. By visually examining the shape of the attractor at a given time delay, To develop an intuitive sense for the underlying nature of the data and the dynamics of the system.

Plot Attractor functions on the active channel of the graph. It prompts the user for a time delay and then constructs a new graph window with an X/Y plot of the attractor of the original signal against the time delayed version of the signal. It does not perform any additional computation aside from assisting in the setup and configuration of the attractor plot.

Correlation Coefficient



The *correlation coefficient* is a statistical measure related to the degree of variance or covariance between two data series. Given two data series *x* and *y* of length *n*, the correlation coefficient *r* is given by the formula:

$$r = \frac{n\sum xy - \sum x \sum y}{\sqrt{\left[n\sum x^2 - \left(\sum x\right)^2\right]\left[n\sum y^2 - \left(\sum y\right)^2\right]}}$$

(see http://mathworld.wolfram.com/CorrelationCoefficient.html)

The square of the correlation coefficient can be used to determine the proportion of variance in common between the two signals. As the square gets closer to 1, the signals are a better statistical match for each other.

To derive the correlation coefficient, two channels of data are compared against each other.

- the channels must have the same length
- the channels must have the same waveform sampling rate
- all of the data of the entire graph for the two channels will be used to compute the correlation coefficient.

Electrodermal Activity

Overview

The **Electrodermal Activity** analysis routines are separated into three menu options that transform the tonic EDA signal to create a phasic waveform, locate and score skin conductance responses, or perform a detailed event-related EDA analysis by combining marker information from the Stim-Response: Digital Input to Stim Events routine (see page 67) to the event-related EDA Analysis routine. The Event-related routine will automatically derive the phasic waveform and locate SCRs.

The routines employ a scoring system that marks the waveform and the point of stimulus delivery. It's easy to manually adjust the automated scoring by relocating the event onset/peak/end before rerunning the analysis. The event-related analysis provides a variety of measures from the SCR data, including classification of specific and non-specific responses. The results are pasted into the journal file or Excel for further analysis.

Preferences must be established for each routine and can be adjusted at any time via the Preferences option (page 21).

The time to complete the analysis routine will vary based on the number of SCR reposnes and the sample rate of the data.

Definitions

The prompts and results of the Electrodermal Activity analysis package use the following terminology and units:

µmho—the unit abbreviation for micromhos, used in channel labels and analysis results; micromho is equivalent to microsiemens.

EDA (Electrodermal Activity)—the general area of skin conductance signals. Sometimes referred to by the older term "galvanic skin response."

Tonic EDA—continuous data acquired from an EDA electrode that includes all baseline offset. Sometimes referred to as "skin conductance level." Averaging the tonic EDA over a specific period of time results in the average skin conductance level over an interval. Tonic EDA is recorded using BIOPAC equipment with the high pass filtering set to off (DC mode).

Phasic EDA—a continuous signal indicative of localized changes in the tonic EDA signal. Sometimes referred to as "continuous skin conductance response." Phasic EDA can be thought of as AC coupled tonic EDA. The EDA analysis package offers multiple ways of constructing phasic EDA including smoothing and high pass filtering. The EDA analysis package performs the majority of its analysis on tonic EDA signals, so if phasic EDA is being recorded directly it is recommended that a second channel be used to record tonic EDA.

Skin Conductance Response (SCR)—an individual localized change in the tonic EDA signal. An SCR may occur in response to a stimulus or may occur spontaneously. In general, there are multiple SCRs present in a tonic EDA signal and they can be detected as deflections from the localized baseline.

Reference

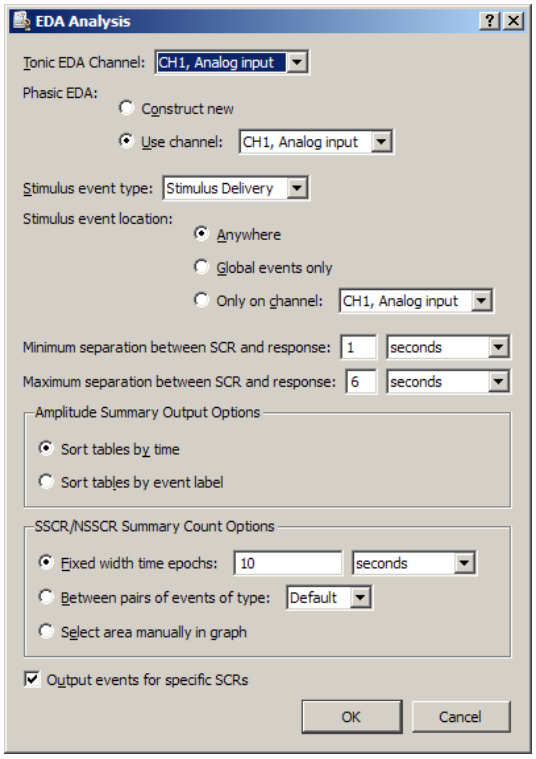
The Electrodermal Activity analysis package was developed to support the parameters established in:

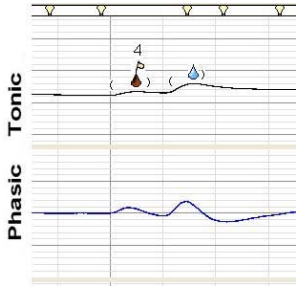
M. E. Dawson, A. M. Schell, and D. L. Filion. The electrodermal system. In J. T. Cacioppo, L. G. Tassinary, and G.B. Bernston, editors, Handbook of Psychophysiology: second edition, pages 200–223. Cambridge Press, Cambridge, 2000.

Derive Phasic EDA from Tonic

Given a tonic EDA signal, this transformation uses baseline smoothing or high pass filtering (the method currently set in Preferences) to construct a new Phasic EDA channel in the graph containing the estimate of the phasic EDA. This routine is automatically included in both the locate SCR and Event-related EDA routines.

Event-related EDA Analysis





Sample EDA Analysis Output

All SCR events are marked on the tonic waveform as follows:

(*open paren.*	The point at which the phasic signal crosses the SCR threshold level established in EDA Preferences; see page 291
blue waterdrop	blue marks the peak response of a nonspecific, event-related SCR
red waterdrop	marks a specific SCR "SRR" with a flag numbered with the stimulus event type
) *close paren.*	The point at which the phasic signal crosses the zero threshold level

The Event-related EDA Analysis transformation routine assists in the extraction of EDA measures that are linked to specific stimuli. The stimulus event marks must be included in the file BEFORE using this analysis.

This analysis routine requires four elements:

1. Tonic and Phasic waveforms.

   Tonic EDA Channel: A Tonic EDA signal must be present in the graph.

   Phasic EDA:

   Construct new: Given a tonic EDA signal, a phasic EDA will be automatically constructed using baseline smoothing or high pass filtering (the method currently set in Preferences).

   Use Channel: If the graph contains a phasic waveform, select the appropriate channel.

2. Stimulus delivery events.

   Digital markers with a common event type must be located BEFORE using this analysis.

   The Event-related EDA Analysis requires that an event be defined in the graph at the location of the delivery of each stimulus. This event may be defined using the Event Tool, hotkey insertion during acquisition, or any other method of defining events. All of the stimulus delivery locations to be extracted must have the same event type (e.g. "Flag"). To analyze multiple different event types, the transformation script must be executed multiple times.

   - If you are using E-Prime, SuperLab, or some other stimulus delivery system and have the digital events captured in the Acq*Knowledge* file, we recommend that you use the Stim-Response: Digital Input to Stim Events routine (see page 67). This routine will automatically classify and label the digital markers for use by the Event-related EDA analysis.

Stimulus event type: If using the Digital Input to Stim Events, select Stim/Response > Stimulus Delivery. Stimulus delivery events are located by event type or by specific channel of the graph.

Stimulus event location: Specify the location as anywhere, global only, or on a specified channel. See the Events section for details.

3. Skin conductance responses.

   If the tonic EDA signal does not already have SCR events defined on it, SCR events will be automatically constructed on the channel using the Locate SCRs transformation routine.

4. Specified time window between the stimulus event and the skin conductance response.

   The transformation takes a maximum allowable separation window between the stimulus event and SCR response. Each stimulus delivery event is paired with the closest SCR event. SCRs that correspond to a stimulus delivery are known as specific SCRs (abbreviated "SRR"). SCRs generally occur within a certain timeframe after stimuli. The time window allows responses too close to stimuli to be rejected and classified as non-specific.

   Minimum separation: specify in relation to the stimulus event (includes time unit options).

   Maximum separation: specify in relation to the stimulus event (includes time unit options).

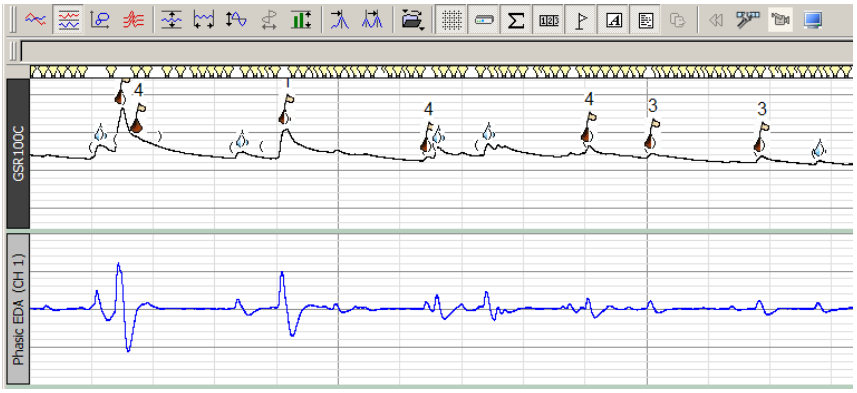
   Given a response time window [res_{min}, res_{max}], for each stimulus delivery event at a time *t*, SCR onset events that are not presently matched as SRRs will be searched for in the window [$t+ res_{min}$, $t+ res_{max}$]. The SCR onset event within this window closest in time to [$t+ res_{min}$] will be paired with the stimulus event and considered a SRR.

SRR are marked as a red waterdrop icon with a flag numbered with the corresponding stimulus event type when "Output events for specific SCRs" is enabled.

Each SRR will be matched to only one stimulus delivery event. If the closest SCR to a stimulus is farther away than this time interval, it is not assumed to be a response to the stimulus. It may be a response to a later stimulus or it may be a non-specific SCR that occurred spontaneously.

Output Events for Specific SCR

Enable this option to mark specific skin conductance events as a red waterdrop icon with a flag numbered with the corresponding stimulus event type.



Event-related EDA Analysis Output Options

Enhancements provide more options for multiple stimulus event types and unmatched events, including:

- Labels and additional measures are available in the specific stimulus and SCR analysis table
- Text and Excel tables may be optionally sorted either by time or grouped by stimulus label
- A new table has been added listing stimulus events that were not paired with an SCR
- The SRR/NS.SSR Rate analysis, which counts frequencies of SCRs in specific time periods, may now be driven by time periods defined using pairs of events or a selection in the graph
- A table has been added listing amplitude/frequency percentage statistics for all matched and unmatched stimuli events (e.g. total stimulus count, percentage of stimuli that were pared with an SCR, etc.)
- Additional optional Specific-SCR events may be defined on the tonic EDA waveform at the positions of specific SCRs with labels matching the stimulus to which they were responses. This allows for further peak-detector based runs to perform additional data reduction.

Amplitude Summary Output Options

For each specific SCR that is paired with a stimulus delivery event, the following measures are extracted in table format and can be sorted by **Time** or by **Event label**. If text output is enabled in EDA Preferences, the average value of SCL, Latency, SCR Amplitude, and SCR Rise Time will be included as the final row of the table.

Name	*Abbrev.*	*Description*	*Units*
Stimulus Delivery Time	Stim Time	The time within the recording where the stimulus delivery event was located.	seconds
Skin Conductance Level	SCL	Amplitude of the tonic EDA signal at the time when the stimulus was delivered.	µmho
Response Latency	Latency	Time separating the stimulus delivery from the onset time of the corresponding SCR. This latency will always be less than the maximum allowable latency specified as a parameter for the analysis.	seconds
SCR Amplitude	SCR Amplitude	Height of the corresponding SCR as determined by the change in the tonic EDA amplitude from the time of SCR onset to the maximum tonic EDA amplitude achieved during the SCR: $[EDA(t_{max}) - EDA(t_{onset})]$	µmho
SCR Rise Time	SCR Rise Time	Time taken for the tonic EDA to reach its maximum value within the SCR: $[t_{max} - t_{onset}]$	seconds

SSCR/NSSCR Summary Count Options

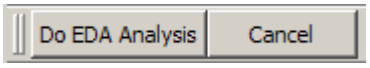
In addition to the above measures extracted for each specific SCR, the analysis performs rate extractions for specific and non-specific SCRs. By examining how the rate of SCR occurrences changes, long-term experimental trends can be investigated. This analysis is placed into a second set of waveforms (or a second table for text and Excel output).

Fixed width: fixed width window is specified as the "SCR count interval width" when performing the analysis. The entire recording is split up into fixed-width epochs of this granularity with the first epoch aligned at the start of the recording. For each fixed-width epoch, the following are extracted:

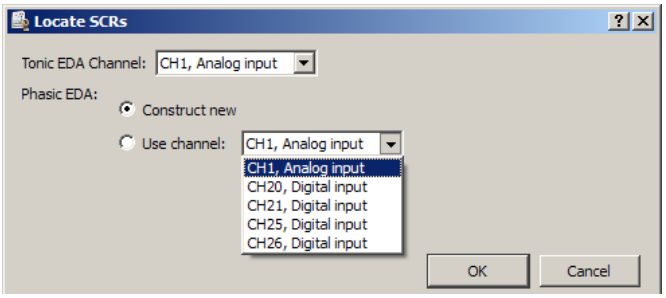
Name	*Abbrev.*	*Description*	*Units*
Epoch Start Time	Start Time	Time location in the recording of the start of the epoch being examined.	seconds
Specific SCR Rate	SRR	Frequency of the occurrences of specific SCRs within the epoch. |Specific SCRs are those SCRs that were successfully matched to a corresponding stimulus delivery event.	Hz
Non-specific SCR Rate	NS.SRR	Frequency of the occurrences of non-specific SCRs within the epoch. These are SCRs that occur spontaneously and are not paired with any known stimulus.	Hz

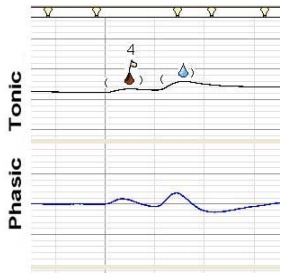
Between event pairs: Select an event type from the pull-down menu. The software will locate the event markers at the beginning and end of the region of interest and perform the analysis between the two points This option is useful if the recording is broken into defined periods—such as baseline, event, and response—using the event hotkeys..

Manually selected area: Highlight the area where NSSCR/SSCR rates should be computed and then click "Do EDA Analysis" in the graph window.



Locate SCRs





The Locate SCRs routine will identify skin conductance response and score the waveform. This analysis is useful for analyzing spontaneously occurring skin conductance responses. The routine is automatically included in the Event-related EDA routine. All SCR events are marked on the tonic waveform as follows:

(	*open paren.*	The point at which the phasic signal crosses the SCR threshold level established in EDA Preferences; see page 21
	blue *waterdrop*	The peak response point of a nonspecific, event-related SCR
)	*close paren.*	The point at which the phasic signal crosses the zero threshold level

This transformation requires a tonic EDA signal. If a phasic EDA has already been constructed for this tonic EDA, it may be used; otherwise, the transformation will create a phasic EDA automatically according to the settings in the Preferences.

Given a tonic EDA, the Locate SCRs transformation defines an event for each skin conductive response in the tonic EDA. SCR location is a two stage process. First, all potential SCR occurrences are located on the signal. Second, all potential SCR occurrences that are not large enough are rejected.

Potential SCR occurrences are detected by performing thresholding positive peak detection on the phasic EDA signal (using H and P as set via Preferences**Error! Reference source not found.**):

1. Given a detection threshold H (expressed in µmho), search for a positive threshold crossing in the phasic EDA signal. This position is recorded as the start of the potential SCR.
2. Continue examining the phasic EDA until the first negative threshold crossing of 0 µmho occurs. This position is recorded as the end of the potential SCR.
3. Return to step 1 to continue searching for more potential SCRs.

After all of the potential SCRs have been located, the set of valid SCRs is constructed as follows:

1. Determine the overall maximum amplitude of the phasic EDA signal within all potential SCRs.
2. Given a percentage P, construct a threshold level T of P percent of the overall maximum phasic EDA signal value located in step 1.
3. Examine each potential SCR. Find the maximum phasic EDA. If $m < t$, discard the potential SCR. Mark the potential SCR as a valid SCR.

If the tonic EDA channel chosen for analysis already has SCR events defined on it, the SCR events will be replaced with the newly detected SCR events. No existing SCR events will be erased without a confirmation.

Once SCR events have been defined, they can be used in conjunction with the Cycle Detector for performing further data reduction. The “event count” measurement can be used to estimate SCR frequency during individual time ranges of the experiment.

Events on Tonic EDA

After valid SCRs are located using the algorithm above, events are inserted into the graph that can allow for further data analysis around the SCR positions. Three events are defined on the tonic EDA waveform for each individual valid SCR:

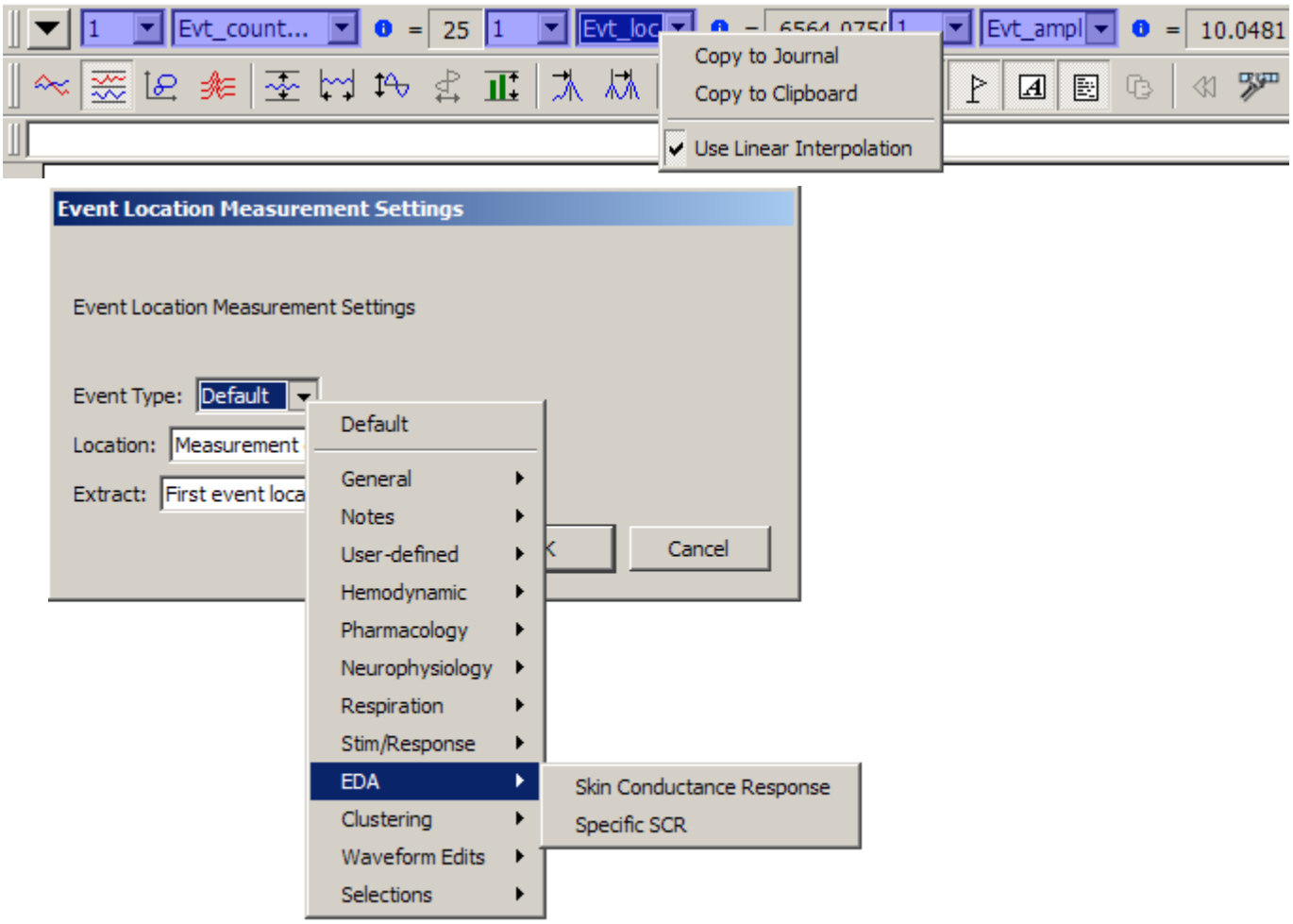
1. “General > Waveform onset” event at the SCR onset time. This is the point where the threshold H was crossed in the phasic EDA.
2. “EDA > Skin conductance response” event at the time where the tonic EDA reaches its maximum value within the SCR (max in time range).
3. “General > Waveform end” event at the ending SCR time. This is the point where the zero threshold was crossed in the phasic EDA.

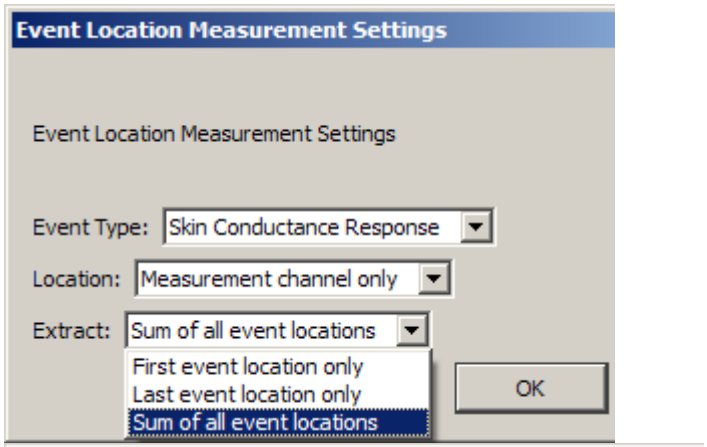
Events for SCRs will always occur as described above, in the order shown.

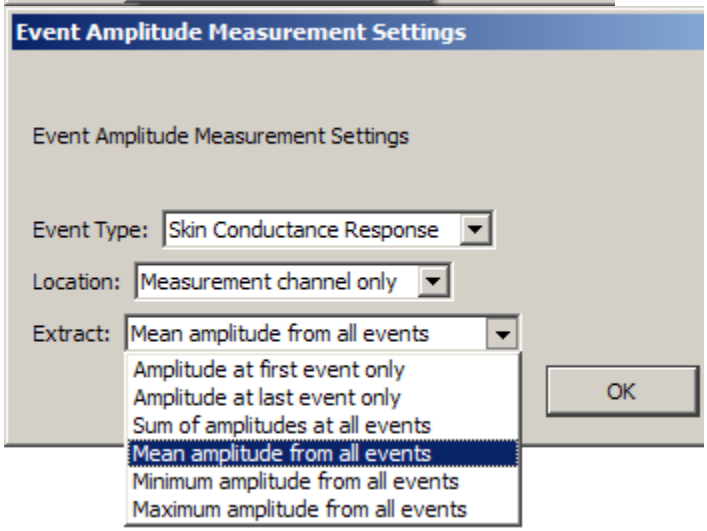
EDA Measurements

To perform Event-related EDA analysis, choose Analysis > Electrodermal Activity > Event-related EDA Analysis.

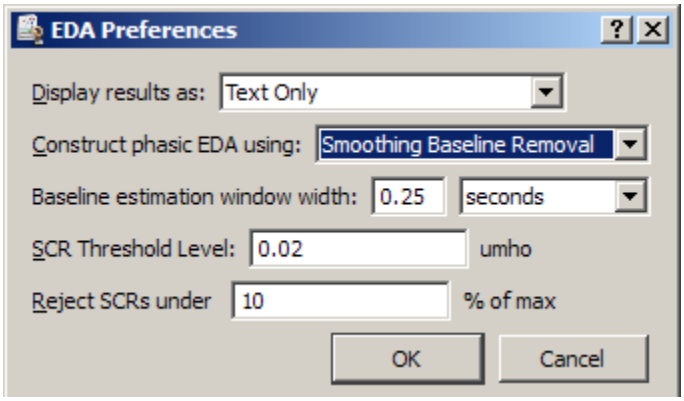
To take measurements from the skin conductance response analysis, set measurements for event count, event location and/or event frequency. Set the source channel as the Tonic EDA channel and select the location (measurement channel only, glogal events onlkty, anywhere)and measurement parameters as desired. This method is useful for spontaneously occurring skin conductance response analyses. Take measurements over a manually selected area or use Find Cycle analysis to take automatically measurements over a user-defined time interval.




Event Location Measurement Settings
Event Location Measurement Settings
Event Type: Skin Conductance Response
Location: Measurement channel only
Extract: Sum of all event locations
First event location only
Last event location only
Sum of all event locations
OK

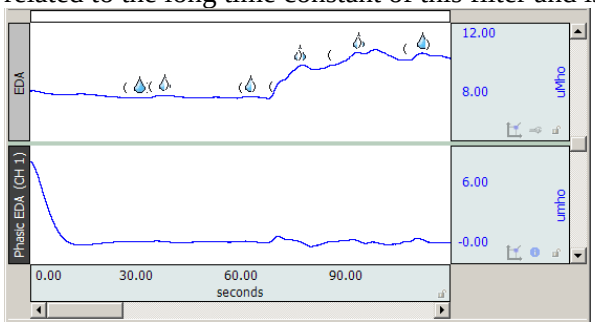

Event Amplitude Measurement Settings
Event Amplitude Measurement Settings
Event Type: Skin Conductance Response
Location: Measurement channel only
Extract: Mean amplitude from all events
Amplitude at first event only
Amplitude at last event only
Sum of amplitudes at all events
Mean amplitude from all events
Minimum amplitude from all events
Maximum amplitude from all events
OK

EDA Preferences…

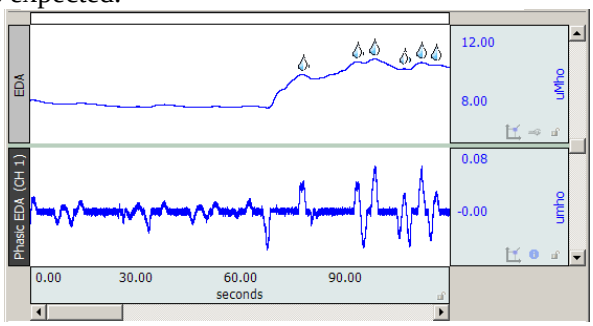


The following EDA Preferences can be configured and will be applied to all options in the analysis package:

- **Display results** as text, graph channels, or Excel
- **Construct Phasic EDA using** High pass Filtering or Smoothing Baseline Removal
  - High pass Filtering—High pass filtering constructs phasic EDA by applying a digital IIR high pass filter (f = 0.05 Hz, Q = 0.707) to the tonic EDA signal. This high pass filter essentially AC couples the tonic EDA signal similar to using the high pass hardware filter available on the GSR100C module. When using high pass filtering, the first few seconds of the phasic EDA may not be centered on zero and will appear to contain invalid data and a valid response may be excluded; this artifact is related to the long time constant of this filter and is expected.

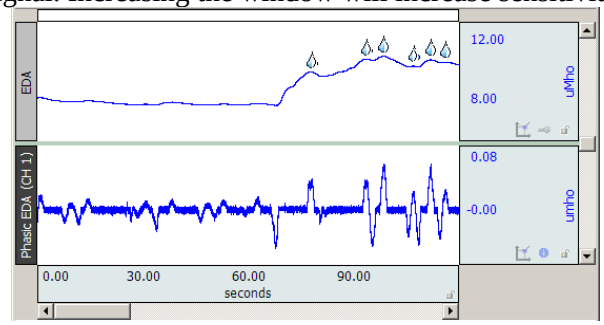


0.05 Hz High Pass Filter

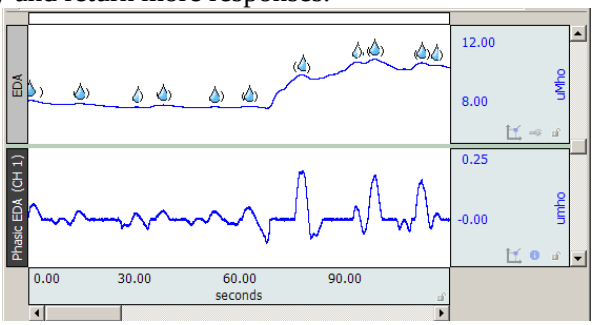


Smoothing Baseline Removal

  - Smoothing Baseline Removal—Smoothing baseline removal constructs phasic EDA by subtracting an estimate of the baseline conductance from the tonic EDA. Set the baseline estimation
- **Baseline estimation**: The estimate of the baseline is generated using median value smoothing. This is more computationally intensive than high pass filtering but does not illustrate artifact at the start of the signal. Increasing the window will increase sensitivity and return more responses.



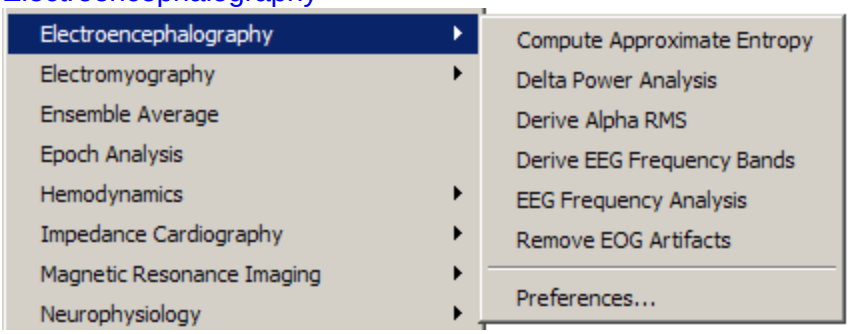
Baseline window set to 4 seconds



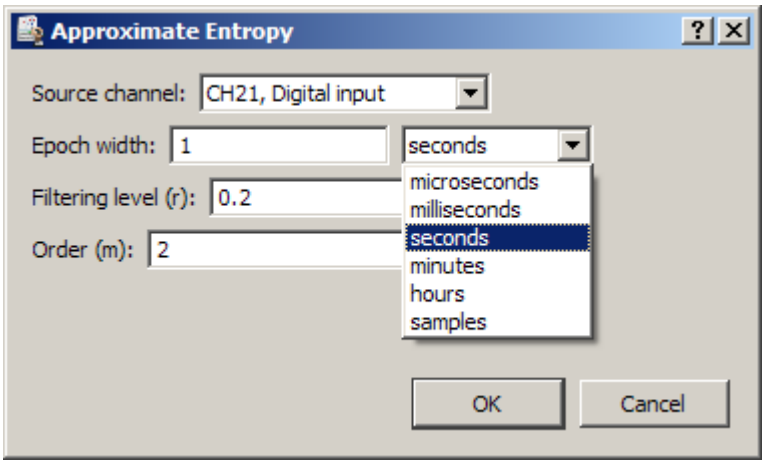
Baseline window set to 8 seconds

- **SCR detection parameters**: threshold detection level H and percentage P, see page 18.
  - The default values are H = 0.02 µmho, P = 10, where H is detection threshold and P is percentage
  - Setting H to 0 and P to 10% will approximate the SCR detection algorithm referenced in K. H. Kim, S. W. Bang and S. R. Kim, "Emotion recognition system using short-term monitoring of physiological signals," Medical & Biological Engineering & Computing, vol. 42, pp. 419-427, 2004.
  - Setting P to 0% will retain all potential SCRs (none will be rejected in the second phase).

Electroencephalography



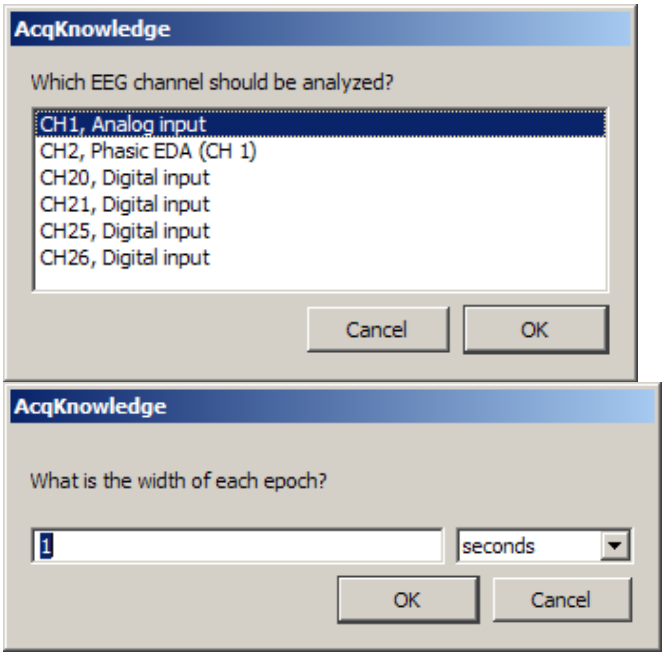
Compute Approximate Entropy



Approximate entropy is a statistical measure that attempts to quantify the predictability of a data sequence. A perfectly predictable data series (such as a pure sine wave) has approximate entropy of zero. Several studies are beginning to examine approximate entropy of EEG data and its relationship to external factors such as drugs and sleep states.

The Compute Approximate Entropy script divides an EEG signal into fixed-width epochs and computes the approximate entropy for each epoch. Derivation of the approximate entropy is a computationally intensive process and may take several minutes or hours to complete. To obtain only the sub-ranges of the EEG data, copy and paste the ranges into new graph windows to restrict the approximate entropy computations to that data range; the analysis is performed for all of the data in the graph window regardless of the selected area.

Delta Power Analysis

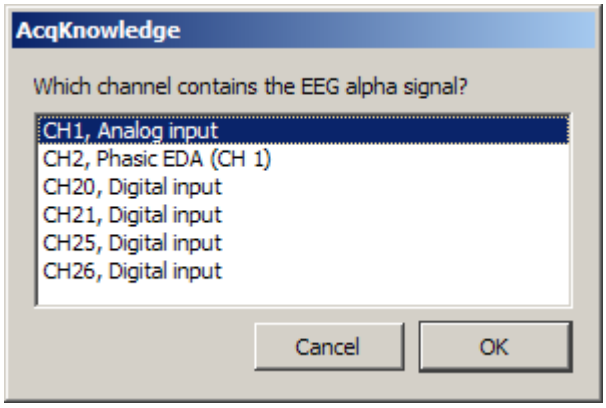


Delta power is the total power of the EEG signal that occurs within the delta frequency band as configured in the Preferences. Delta power has been examined in a number of various EEG studies as an indicator of sleep/wakefulness and other conditions. By examining changes in the delta power, it may be possible to correlate delta power with effects of external factors.

The Delta Power Analysis script divides an EEG channel into fixed-width epochs. For each epoch, the power spectral density is computed and the total power within the delta frequency band is derived from the PSD. This delta power value is then placed into the graph or into the journal as specified by the output preferences.

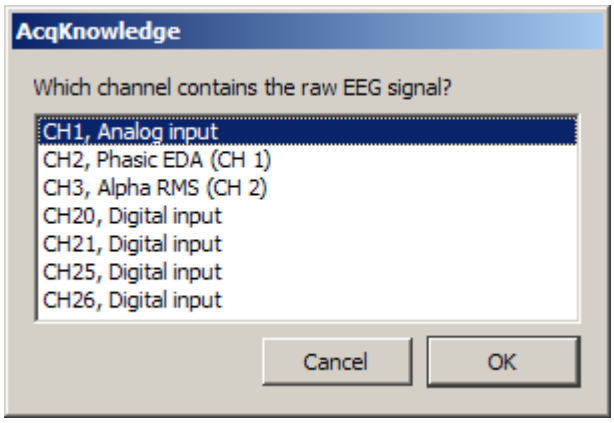
Delta power can be measured from either a filtered or unfiltered EEG channel. To compute delta power for individual frequency bands, they must be derived prior to running the Delta Power Analysis script.

Derive Alpha RMS



The Derive Alpha RMS script constructs a standard alpha RMS waveform from an alpha EEG signal. Alpha RMS is the windowed root mean square value of the signal using a window width of 0.25 seconds.

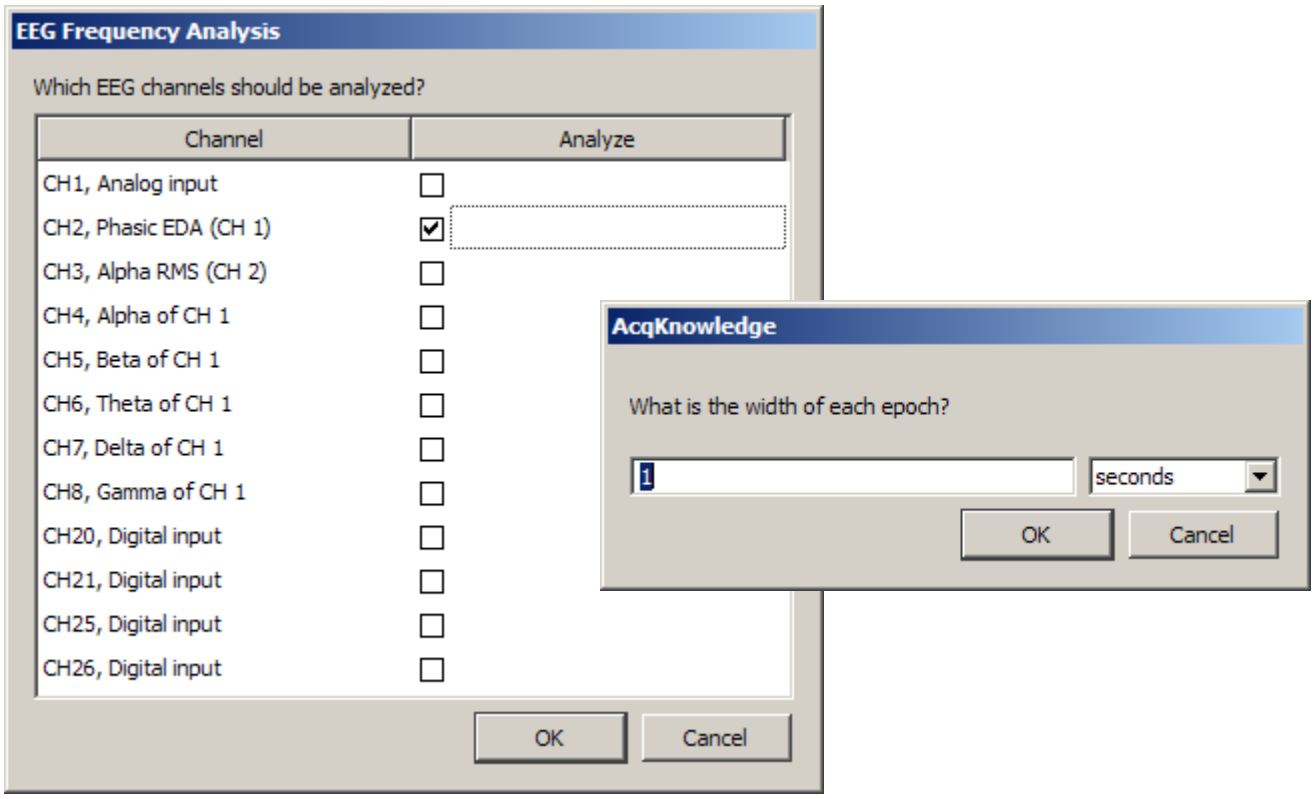
Derive EEG Frequency Bands



The Derive EEG Frequency Bands script applies filtering to an unfiltered EEG lead signal to generate the following five standard EEG bands: Alpha, Beta, Theta, Delta, and Gamma.

The frequencies used for each band are taken from the analysis package preferences. Filtering is performed using IIR lowpass+high pass combination filters.

EEG Frequency Analysis

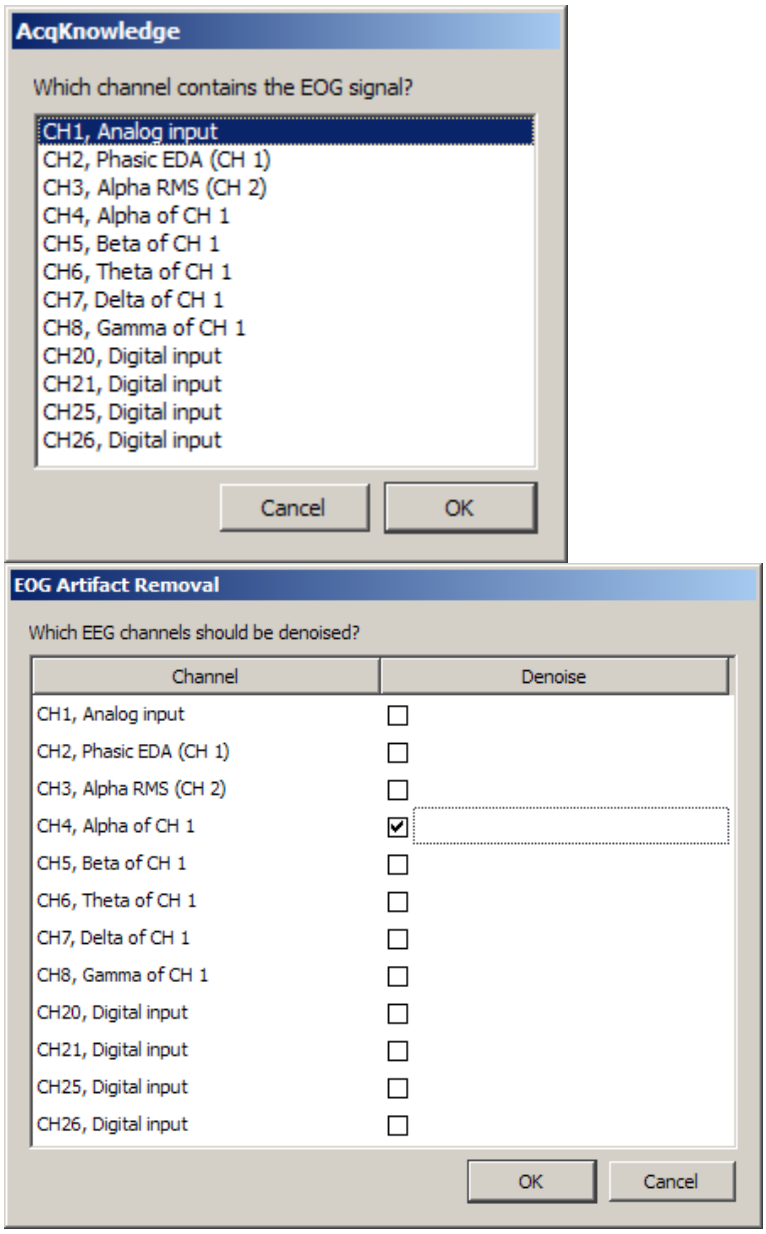


EEG may be characterized in terms of frequency and the power within specific frequency bands. The EEG Frequency Analysis script performs various feature extractions from EEG signals using FFT and other techniques to examine the power within the EEG signals. This analysis may be performed for multiple EEG leads simultaneously, allowing for either analysis of multiple leads or analysis of multiple EEG alpha, beta, theta, or delta bands from a single raw lead.

The EEG Frequency Analysis script divides the EEG signals into fixed-width time epochs. For each individual time epoch, Acq*Knowledge*'s Power Spectral Density function is used to estimate the power spectrum of that epoch using a Welch periodogram estimation method. From this PSD the following measures are extracted for each epoch:

Name	*Abbrev.*	*Description*	*Units*
Mean Power	MeanP	The average power of the power spectrum within the epoch. (Units Note: V will be replaced with the voltage units in which the EEG was recorded)	$\frac{V^2}{Hz}$
Median Frequency	MedianF	Frequency at which 50% of the total power within the epoch is reached.	Hz
Mean Frequency	MeanF	Frequency at which the average power within the epoch is reached.	Hz
Spectral Edge	Spectral Edge	Frequency below which a user-specified percentage of the total power within the epoch is reached. This percentage can be set using "Preferences" and defaults to 90%.	Hz
Peak Frequency	PeakF	Frequency at which the maximum power occurs during the epoch.	Hz

Remove EOG Artifacts



Some EEG recordings involve subjects performing various visual tasks such as reading or watching video. Under these conditions, EEG may be susceptible to interference from the much stronger EOG signal arising from eye motion, particularly if EEG is recorded from near the front of the skull. Remove EOG Artifacts helps remove EOG interference from the EEG signals, recovering the EEG data for use in further analysis.

EOG removal is performed using a blind signal separation technique known as Independent Component Analysis. ICA is used to split up statistically independent signals that have been mixed together during recording. Since EOG is independent of EEG, ICA can be used to remove it.

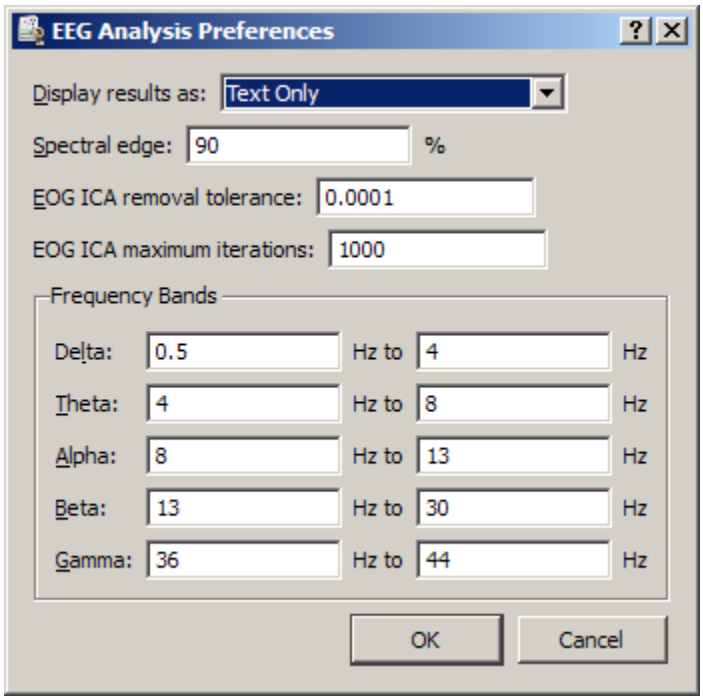
In order to use Remove EOG Artifacts, a distinct EOG signal must be acquired in addition to the EEG signals. The EOG signal is required to identify the components correlated to eye motion.

EOG artifact removal functions better when it is performed on multiple EEG leads simultaneously. Better results may be obtained by including EEG leads that do not exhibit EOG interference since the increased number of leads allows for more fine-grained signal separation. Good results can be seen with as few as two EEG leads and one EOG lead. While this technique can be performed with a single EEG lead, the results will not be as dramatic.

Note ICA is a non-deterministic technique, so it may not be possible to automatically separate the signals for every EOG/EEG data set. For ICA to be successful, it may be necessary to fine-tune the parameters

of the ICA search procedure to match the data, use a different electrode configuration, or use fewer or more leads.

Preferences…



Adjust the EOG ICA Tolerance level and the EOG ICA maximum number of iterations by accessing Transform > Specialized Analysis > Electroencephalography > Preferences. EOG ICA Tolerance is used as the termination condition of ICA signal separation. The EOG ICA maximum number of iterations is another termination condition of ICA signal separation and represents the maximum point at which the search is aborted. For more information on these settings, see the documentation for the Independent Component Analysis transformation.

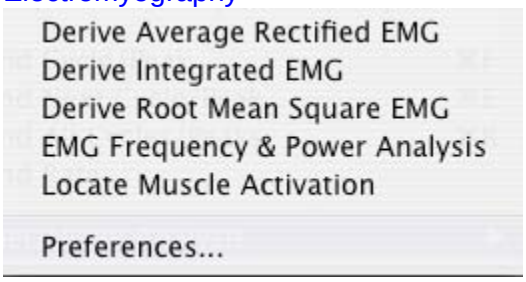
Because ICA is a statistical technique, any filtered data produced with Remove EOG Artifacts should be carefully verified against other information to ensure that the approximations produced via ICA represent information that is truly correlated to the expected ECG.

The spectral edge percentage indicates the cutoff percentage of the total power at which spectral edges will be placed. The default value is 90%.

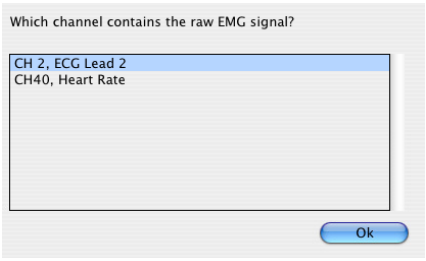
The frequency bands of alpha, beta, delta, and theta may be modified to match different analysis protocols. The default frequency ranges are:

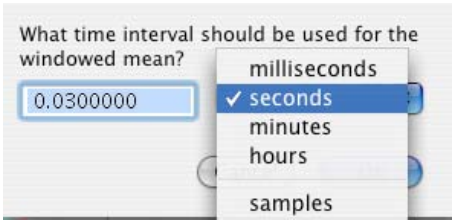
- Alpha—8 Hz-13 Hz
- Beta—13 Hz-30 Hz
- Delta—0.5 Hz-4 Hz
- Theta—4 Hz-8 Hz
- Gamma—36 Hz-44 Hz

Electromyography



Derive Average Rectified EMG



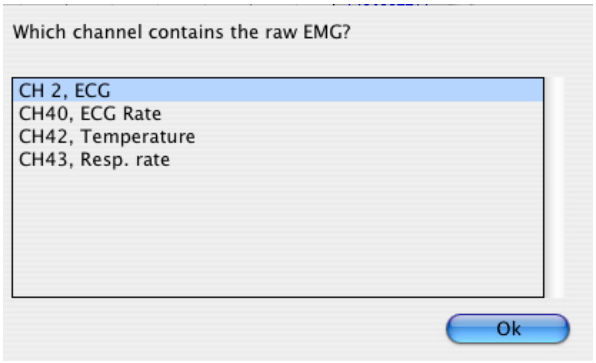


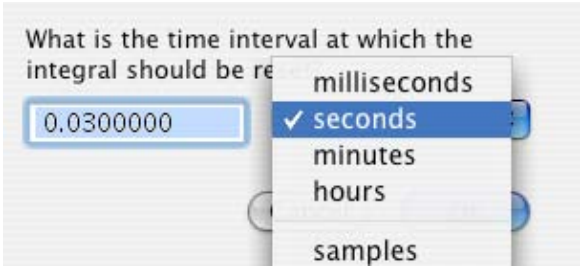
Average rectified value (ARV) is defined as a time windowed mean of the absolute value of the signal. ARV is one of the various processing methods used to construct derived signals from raw EMG data that can be useful for further analysis.

To perform ARV, a time window must be specified for the sliding mean. The default time window setting is 30 milliseconds, but this value can be adjusted depending on the desired amount of smoothing effects. It is advisable to closely examine results for time windows larger than 30 milliseconds as it is possible for delay to be introduced into the result.

The ARV is computed using the Integrate transformation with a Rectified Average over Samples configuration.

Derive Integrated EMG

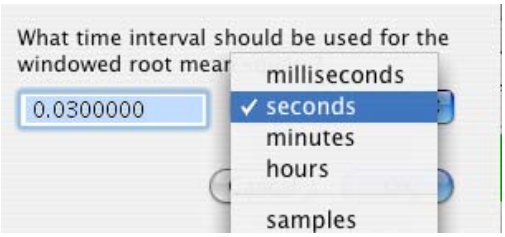




Integrated EMG (iEMG) is defined as the area under the curve of the rectified EMG signal, that is, the mathematical integral of the absolute value of the raw EMG signal. When the absolute value of the signal is taken, noise will make the mathematical integral have a constant increase. Integrated EMG splits up the signal into fixed-width timeslices and resets the integral at the start of each timeslice. To derive iEMG, the width of this timeslice must be specified. Similar to ARV, timeslices longer than 30 milliseconds may introduce delay into the result.

The integrated rectified EMG signal will appear like a “sawtooth” style wave. In addition to the true iEMG, this script will output a second waveform whose value is the maximum value of the iEMG signal in each timeslice. This Maximum iEMG is easier to interpret visually and approximates the envelope of the iEMG signal.

Derive Root Mean Square EMG



Root Mean Square EMG (RMS EMG) is defined as the time windowed RMS value of the raw EMG. RMS is one of a number of methods used to produce waveforms that are more easily analyzable than the noisy raw EMG.

To construct the windowed RMS signal, a time window must be specified for the sliding mean. The default time window setting is 30 milliseconds, but this value can be adjusted depending on the desired amount of smoothing effects in the RMS EMG. It is advisable to closely examine results for time windows larger than 30 milliseconds as it is possible for delay to be introduced into the result.

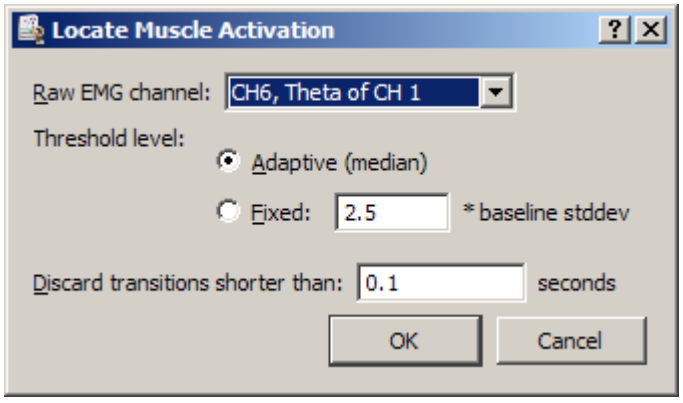
RMS EMG is computed using the Integrate transformation in a Root Mean Square Average over Samples configuration.

EMG Frequency & Power Analysis

Several frequency domain techniques may be used for data reduction of EMG signals. The EMG Frequency & Power Analysis script extracts several measures derived from the power spectrum of an EMG signal. The EMG signal is split up into a fixed number of time periods; within each window, the power spectrum is computed using the Power Spectral Density transformation. For each time period, the following measures are extracted:

Name	*Abbrev.*	*Description*	*Units*
Median Frequency	MedianF	Frequency at which 50% of the total power within the epoch is reached.	Hz
Mean Frequency	MeanF	Frequency at which the average power within the epoch is reached.	Hz
Peak Frequency	PeakF	Frequency at which the maximum power occurs during the epoch.	Hz
Mean Power	MeanP	The average power of the power spectrum within the epoch. (Units Note: V will be replaced with the voltage units in which the EMG was recorded)	$\frac{V^2}{Hz}$
Total Power	TotalP	The sum of the power at all frequencies of the power spectrum within the epoch. (Units Note: V will be replaced with the voltage units in which the EMG was recorded)	$\frac{V^2}{Hz}$

Locate Muscle Activation



When performing gait analysis, exercise physiology, or other research, identification of periods where the muscle is active can allow for correlation of external factors to muscle activity. Locate Muscle Activation attempts to identify various periods of muscle activity using statistical methods. The transformation requires a

raw, unfiltered surface EMG channel. It is important that the muscle being examined is relaxed for the first 0.25 seconds of the recording to provide an estimate of the "background noise" during areas of muscle relaxation. This quarter-second period is used to estimate baseline parameters that affect the entire process.

This transformation implements a variation of the Hodges and Bui detection algorithm as described in:

P. W. Hodges and B. H. Bui, "A comparison of computer-based methods for determination of onset of muscle contraction using electromyography," *Electroenceph. Clin. Neurophysiol.*, vol. 101, pp. 511-519, 1996.

The variation implemented is a threshold-based algorithm roughly consisting of the following steps:

1. Determine mean value μ_0 and resting standard deviation σ_0 of the first 0.25 seconds of the signal.
2. Construct a filtered ARV EMG signal, *z*.
3. Extract the variance of the signal with respect to the noise with the formula
$$g = \frac{z - \mu_0}{\sigma_0}$$
4. Using a threshold *h*, determine when the signal *g* lies below and above the threshold. Portions of time above the threshold are periods of muscle activity.
5. Discard any transitions across the threshold if they are shorter in duration than a user-specified time, *t*.

There are two methods of specifying the threshold *h*. An adaptive method examines the signal *g* and chooses the threshold to be the median of *g* over the entire waveform. Alternatively, the threshold can be specified manually. Using a manual threshold can be useful in adjusting the detection to better match specific EMG data. A suggested threshold is 2.5. By lowering the threshold, a larger quantity of data will be considered as muscle activity. By raising the threshold, a larger quantity of data will be considered to be noise.

The transition discard time *t* is specified in seconds. The default value of *t* is 0.1 seconds. If muscle activity is being inaccurately identified as inactivity for short periods within active times, try increasing the value of *t*. Do not set *t* to be greater than the smaller of either the shortest duration of a single muscle contraction or the shortest rest interval between consecutive muscle contractions.

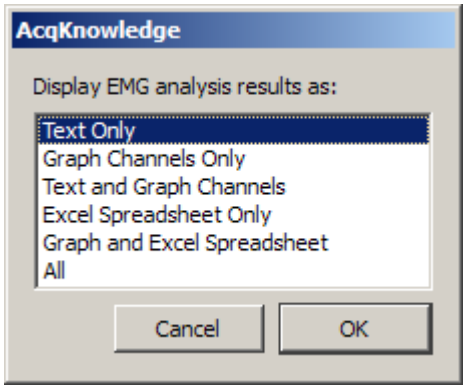
There are two outputs from the Locate Muscle Activation script.

- A new waveform, Muscle Active, will be added to the graph. The value of this wave will be zero when the muscle is at rest and one when the muscle is active. This wave can be used to quickly visually examine the record for periods of activity.
- Events are also generated on the raw EMG waveform. A Waveform Onset event is placed at each transition from inactive to active, and a corresponding Waveform End event is placed at each active-to-inactive transition. You can use these events in conjunction with the Cycle Detector to perform further data reduction based on muscle activity.

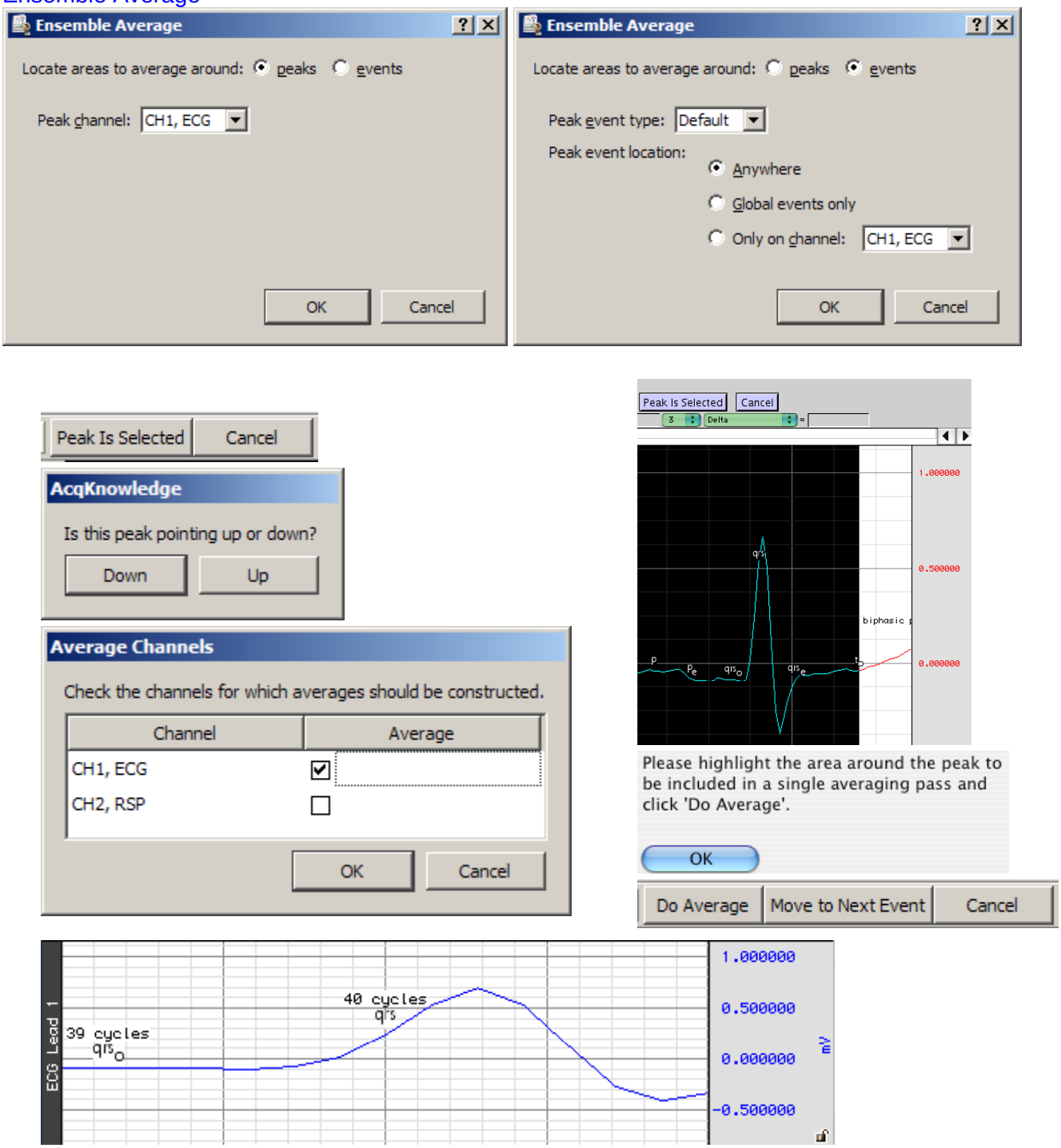
The detection of muscle activation onset and end from surface electrode EMG is an imprecise process. The output of this location should be visually examined for misidentification of activation periods that are too short, too long, overlapping, or missed.

Preferences…

The Preferences allow the type of output to be chosen for displaying results: text, graph channels, or Excel.



Ensemble Average

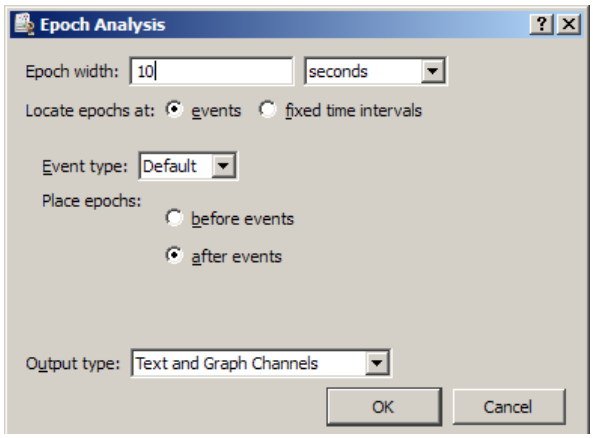


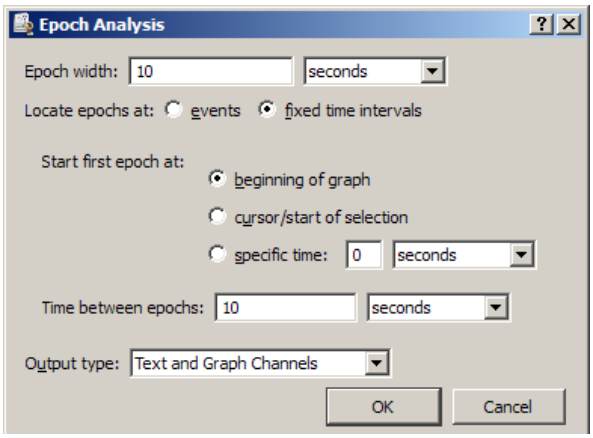
Ensemble Average assists in performing offline averaging. Offline averaging produces an average waveform from a number of cycles, also known as an *ensemble average*. Averages of multiple channels can be extracted simultaneously and be consolidated into a single graph window showing the results.

This option provides two methods for locating individual members of the ensemble.

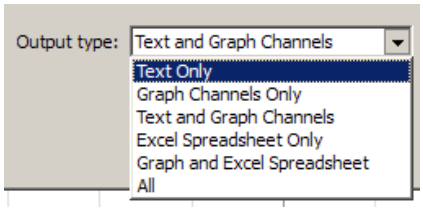
- Data-driven peak detection with positive or negative peaks in the data. This method automatically derives appropriate threshold levels from a user-selected peak and is useful for constructing averages keyed to periodic signals with strong spikes, such as ECG.
- Place members of the ensemble surrounding events in the waveform. Events must be previously defined by the user, either manually or through another automated process. This method is useful for constructing averages keyed to any types of events in a graph.

Epoch Analysis



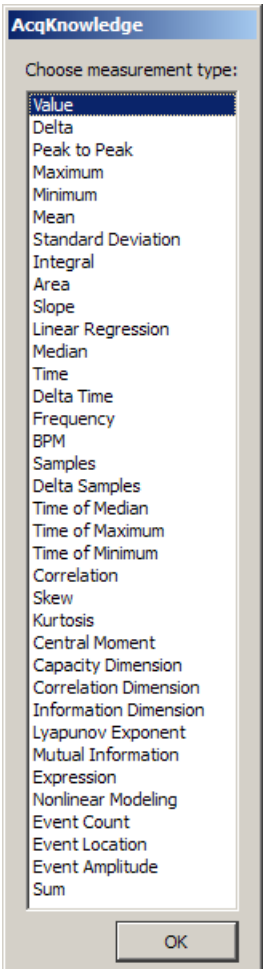


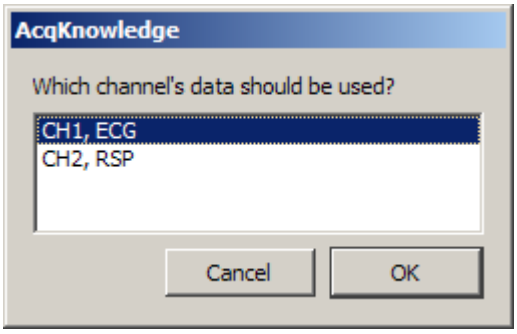
Extracts basic measures from fixed-width time segments of data. A fixed-width time segment of data is known as an *epoch*. The location of these fixed-width intervals can either be keyed off of locations of events in the graph or tied to regular time intervals (e.g. occurring at a constant frequency). All of the standard Acq*Knowledge* measurements can be extracted on an epoch-by-epoch basis with the exception of calculation measurements.

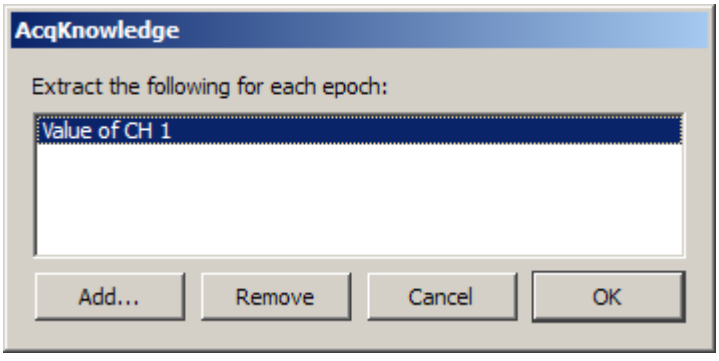


Epoch-by-epoch measurement results can be viewed either as channels of data in the graph, a textual summary, or on an Excel spreadsheet; textual summaries include a final row with an overall average of each extracted measurement.

Times output by Epoch Analysis are always expressed in seconds; all other units correspond to the current preferred measurement unit settings accessible under Display > Preferences.







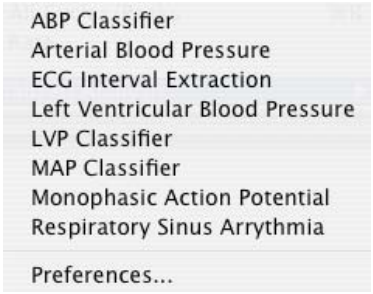
Hemodynamic Analysis

Hemodynamics is the study of blood and circulation related data. This analysis package concerns itself with interpretation of ECG, blood pressure, and monophasic action potential data; ECG signals must be sampled at 5 kHz or below to be analyzed with this package.

IMPORTANT: These routines are designed specifically for human subjects and may not function well, or at all, on animal subjects, particularly small animals.

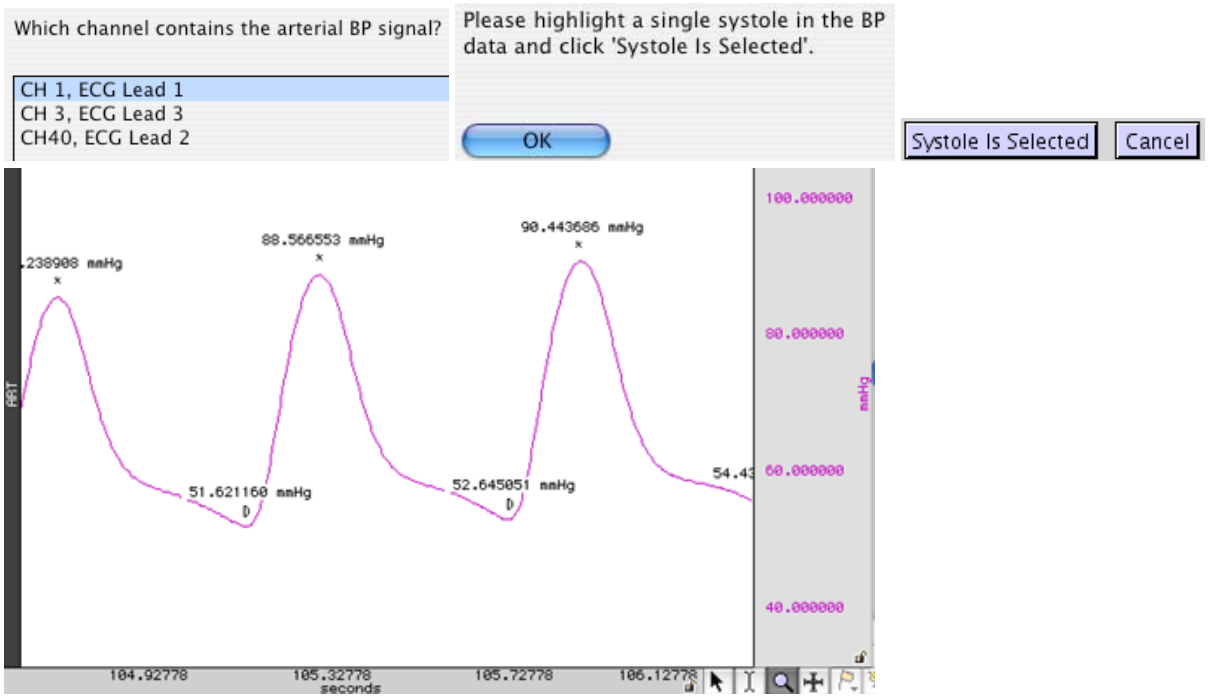
The Hemodynamic analysis package consists of:

a) ABP Classifier
b) Arterial Blood Pressure
c) ECG Interval Extraction
d) Left Ventricular Blood Pressure
e) LVP Classifier
f) MAP Classifier
g) Monophasic Action Potential
h) Respiratory Sinus Arrhythmia
i) Preferences



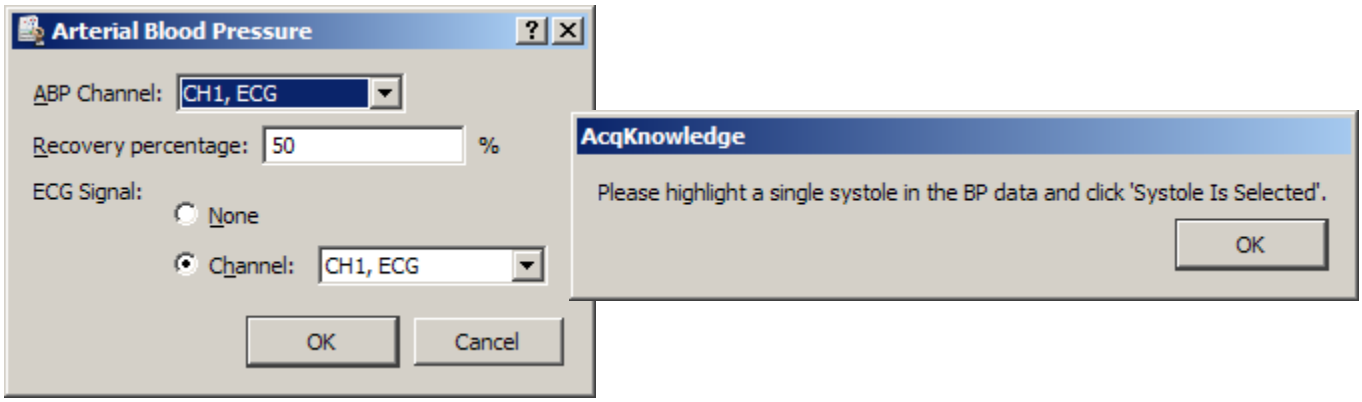
The time units reported by all of these transformations are in seconds unless otherwise noted.

ABP Classifier



Places systolic and diastolic events at appropriate locations on a continuous arterial blood pressure signal recording using either invasive means or a continuous noninvasive pressure monitoring system. The ABP classifier functions directly on the pressure data and may fail for signals that exhibit strong noise characteristics or large baseline drifts. Pre-filtering the signal may improve classification accuracy.

Arterial Blood Pressure



Extracts various cycle-by-cycle measures from arterial blood pressure (ABP) and ECG signals. It can function on an individual ABP signal or, when used in conjunction with an ECG Lead II signal, extract additional Q relative measurements.

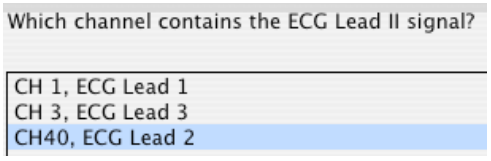
- If the ECG and ABP signals have not been classified when this analysis is performed, events for diastolic, systolic, and ECG boundaries will be inserted as necessary.
- If systolic, diastolic, and Q events are already present on the signals, however, they will be used.

On a cycle-by-cycle basis, the arterial blood pressure analysis transformation extracts the following measures:

Name	Abbrev.	Description
Diastolic	-	Minimum pressure occurring during the cycle
Ejection time	ET	Time interval between the diastolic pressure and the minimum of dP/dt
Heart rate	HR	Heart rate in BPM as extracted from the diastolic-to-diastolic time interval for a given cycle
Maximum dP/dt	dP/dt max	Maximum amount of the change in the pressure during the cycle
Mean blood pressure	MBP	Mean blood pressure: $P_{diastolic} + \frac{P_{systolic} - P_{diastolic}}{3}$
Minimum dP/dt	dP/dt min	Minimum amount of change in the pressure during the cycle
QA Interval	QA	Time interval between ECG Q wave and the diastolic pressure
Recovery interval	%REC	Time required for the pressure signal to decrease by a user specified percentage of the pulse height
Systolic	-	Maximum pressure occurring during the cycle
Time to peak pressure	TTPK	Time interval between the diastolic and the systolic pressures

When textual output is used, the average of all of these measures will be output as the last row of the table.

ECG Interval Extraction



Extracts cycle-by-cycle time and voltage measurements for various points and intervals between waveforms in the cycle on ECG Lead II signals. This interval extraction is based off of the waveform boundary locations with additional logic for defining explicit Q and S wave events. QRS peak events as output for boundary location are used as the R peak location.

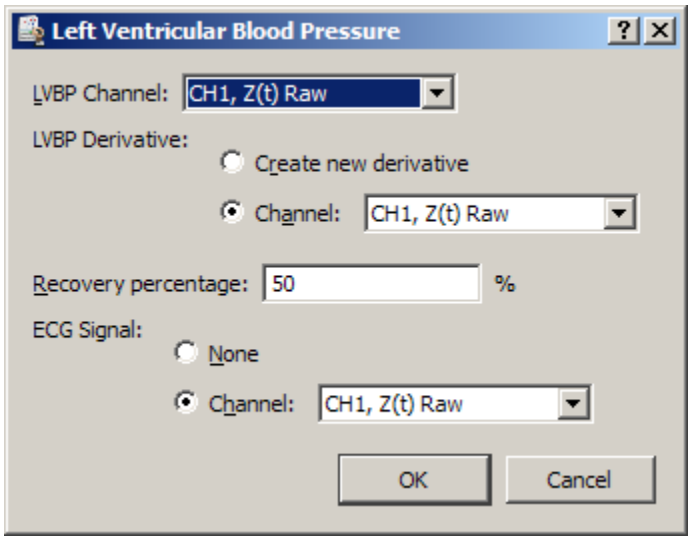
- If the ECG signal was not classified before running the interval extraction analysis, it will be classified automatically.

This analysis extracts the following cycle-by-cycle measures:

Name	*Abbrev.*	*Description*
Corrected QT interval	QTC	QT time interval divided by the square root of the RR interval
Heart rate	HR	RR time interval expressed in BPM
P height	P-H	Amplitude at the peak of the P wave in a cycle
PRQ interval	PRQ	Time between the onset of the P wave to the Q wave
QRS width	QRS	Time between onset of the QRS complex and the end of the QRS complex. Equivalent to the time between onset of Q and end of S
QT interval	QT	Time between the beginning of the Q wave and the end of the T wave
R height	R-H	Amplitude of the R wave in a cycle
RR interval	RR-I	Time between consecutive R peaks in the waveform
ST interval	ST	Time between the S wave to the end of the T wave

At the end of the text table output, the average of all of the cycles will be displayed. Additionally, both text and Excel output will indicate the number of cycles that did not have all three of the QRS, P, and T waves defined. These are cycles where the classifier missed a boundary and are listed as “Bad cycles,” which may happen due to noise or other artifacts in the signal.

Left Ventricular Blood Pressure



Extracts various cycle-by-cycle cardiac measures of left ventricular blood pressure data, optionally in conjunction with an ECG Lead II signal. Examines the LVP signal, ECG, and derivative of the LVP signal.

- If the LVP and ECG signals have not been classified before this analysis is executed, they will be classified automatically.
- Derivatives of the LVP signal can be pre-existing or can be constructed automatically.

- If an ECG signal is not included, only pressure related measures will be extracted.

The analysis outputs the following information on a cycle-by-cycle basis and the textual output cites the average of all of these cycle-by-cycle measurements:

Name	*Abbrev.*	*Description*
Contractility index	CI	maximum value of dP/dt during the cycle divided by the pressure at that time location
Developed pressure	DP	Amplitude interval between end diastolic pressure and systolic pressure
dP/dt Max	-	Maximum change in pressure over the cycle
dP/dt Min	-	Minimum change in pressure over the cycle
End diastolic pressure	LVEDP	End diastolic pressure for the cycle. This is not necessarily the minimum pressure during the entire cycle. LVEDP is located on the LVP signal using the method set in the preferences.
Minimum pressure	MIN	Absolute minimum pressure occurring during the entire cycle. This is not necessarily equivalent to the end diastolic pressure
QA Interval	QA	Time interval between the Q wave of the ECG and the end diastolic pressure
Rate	-	heart rate in BPM as extracted from the time interval between consecutive end diastolic pressure locations
Recovery time	%REC	Time it takes for dP/dt to increase from the minimum dP/dt location to a user specified percentage of that minimum value
Systolic pressure	SYS	Maximum pressure occurring during the entire cycle
Tau	-	Monoexponential time relaxation constant tau computed on a cycle by cycle basis. See “Computation of Tau” on page 35 for specifics.
Tension time index	TTI	Integral of the pressure between end diastolic and the time of minimum dP/dt

Computation of Tau

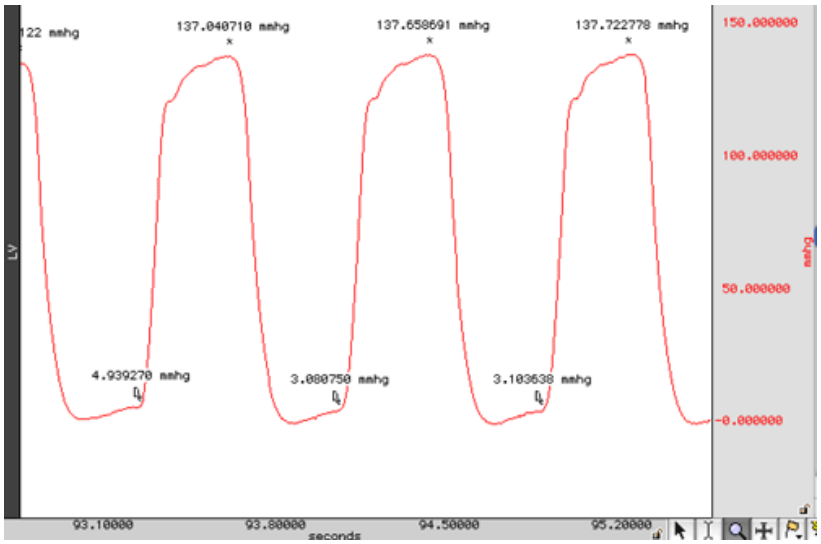
There are many different methodologies used to extract the time constant from LVP data. The time constant is extracted from a best fit parameter of a model to the trailing edge of LVP data on a cycle by cycle basis. This analysis uses a monoexponential model of zero asymptote for computing tau. The relaxation period is defined as the range of data between the time of minimum dP/dt in the cycle to the point where the LVP pressure signal drops below the previous LVEDP level. Within this range, the following model is fitted to the data using the simplex search method:

$$P_0 e^{\frac{t}{\tau}}$$

where P_0 is the value of the LVP signal at the time of dP/dt minimum and *t* is the time coordinate shifted such that *t* is 0 at the time of dP/dt minimum. The best fit value from this model is used as the value of the relaxation time constant.

LVP Classifier

Operates on left ventricular blood pressure (LVP) data to define events at the systolic pressure and the left ventricular end diastolic pressure for each cycle.

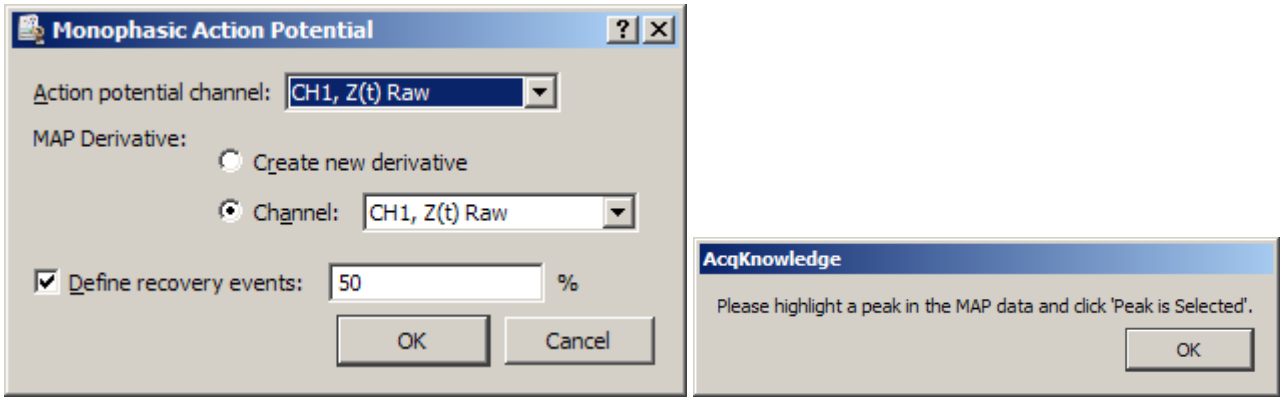


The location of these points is performed using filtered derivatives of the original LVP signal. Pre-filtering the signal (lowpass of 50 Hz or less) or smoothing the signal before running the classifier may improve accuracy.

The LVP classifier locates LVEDP (left-ventricular end diastolic pressure) by examining the derivative of the pressure signal based upon the location method specified in Preferences:

- Adaptive threshold of 0 plus a percentage of the peak to peak change in pressure. The percentage is user-specified; the default is 1%. If the LVP signals do not have “flat” valleys, this percentage may need to be increased to fine-tune positioning of LVEDP.
- First zero crossings before contraction.

Monophasic Action Potential



Performs classification of MAP data acquired from a human or animal subject and extracts various cycle by cycle intervals. Locates upstroke, maximum, 100% recovery, and user-specified recovery points on the action potential.

- Classification is performed using the action potential with its smoothed derivative; pre-filtering noise with low pass filters may improve classification.
- If upstroke, maximum, and plateau events are already defined on the MAP signal, the classifier is not invoked and only recovery events are defined.

Plateau position

To better handle animal subjects and different potential morphologies, there are two methods for locating the plateau position in monophasic action potential data; use Preferences to set the method. Each method defines recovery percentage time locations depending on the signal between its maximum and the beginning of the plateau. The plateau is located by examining the derivative of the MAP immediately following its maximum value after an upstroke.

- The first method uses an adaptive threshold of zero plus a percentage of the peak to peak change in the derivative between the maximum and the first zero crossing after the maximum. If the signal remains above the upstroke voltage in this interval, a quick algorithm is used to locate 100% recovery and user-specified percentage levels. The default percentage is 0.1%, which will place the plateau position very close to the second zero crossing. This slight window around zero helps place plateau start events better for MAP data that has plateaus that continue increasing after their starting position.
- Searches for the second zero crossing after the maximum. If the signal drops below the voltage level of the upstroke in this interval, a different (slower) algorithm is used to ensure the recovery percentage is relative to the upstroke voltage and not the minimum occurring between the maximum and plateau.

The analysis outputs the following information on a cycle-by-cycle basis and the textual output cites the average of all of the cycle-by-cycle values:

Name	*Abbrev*	*Description*
100% recovery period	100% REC	Time interval from the upstroke for the signal to recover back to the upstroke voltage level
dV/dt maximum	dV Max	Maximum change in voltage over the cycle
dV/dt minimum	dV Min	Minimum change in voltage over the cycle
End diastolic voltage	EDV	The value of the signal at the beginning of the upstroke
Max voltage	MAX	The maximum value of the signal over a single cycle
Minimum voltage	MIN	The minimum value of the signal over a single cycle. This may be less than the upstroke voltage depending on the morphology of the action potential
Plateau voltage	PLAT	The value of the signal at the start of the plateau after the completion of the upstroke
Rate	-	This is the heart rate in BPM as extracted from the time interval between consecutive upstrokes
Stroke amplitude	AMP	Voltage interval between the plateau and the upstroke voltage
User recovery period	%REC	Time interval from the upstroke for the signal to recover a specific percentage of the interval between the upstroke and the maximum voltage between the upstroke and the plateau

MAP Classifier

The classifier portion of Monophasic Action Potential only – defines upstroke, plateau, and percentage recovery events on MAP signals without performing the additional MAP data extraction.

The start of the plateau is located using either the second zero crossing of the derivative or a percentage of the cyclic peak-to-peak distance of the derivative. The plateau location method can be configured in Preferences.

Respiratory Sinus Arrhythmia

***IMPORTANT*—Respiration analysis assumes a bidirectional airflow signal that records both inhale and exhale. Unidirectional respiration signals cannot be analyzed at this time.**

Respiratory Sinus Arrhythmia is used to explore the connection between respiration and changes to heart rate. Variations in the heart rate can be directly correlated with vagal tone. The RSA index can be used to investigate changes in this connection during recording.

This RSA index is computed using the peak-valley method as outlined in:

> Grossman, P., van Beek, J., & Wientjes, C. (1990). A comparison of three quantification methods for estimation of respiratory sinus arrhythmia. *Psychophysiology*, 27, 702-714.

This method uses both a recorded ECG Lead II signal and a respiration signal. By using respiration information, this analysis method can provide breath-to-breath analysis that does not require parameter tweaking for individual subjects.

While designed for use with the RSP100C/TSD201 respiration module and transducer combination, it should be possible to use other estimates of respiration. The respiration signal is used to locate periods of inhalation and exhalation. Inhalation begins at valleys in the signal while expiration at peaks. Any respiration estimate that exhibits this morphology should be sufficient.

The RSA index outputted by this analysis is linearly scaled as per the recommendations in Grossman et. al. For comparison to other methods or studies using logarithmic scaling, Transform > Math Functions > Ln transformation can be used after analysis to convert results to logarithmic scaling.

RSA results are triggered from the respiration cycle. The RSA analysis outputs the following measures:

Cycle Index of the respiration cycle in the analysis.

Time Location of the start of the respiration cycle on the time axis.

Min Rate Minimum heart rate occurring during the inspiration window of the respiration cycle, expressed in milliseconds (IBI).

- If a respiration cycle is invalid, this measure will be set to 0.

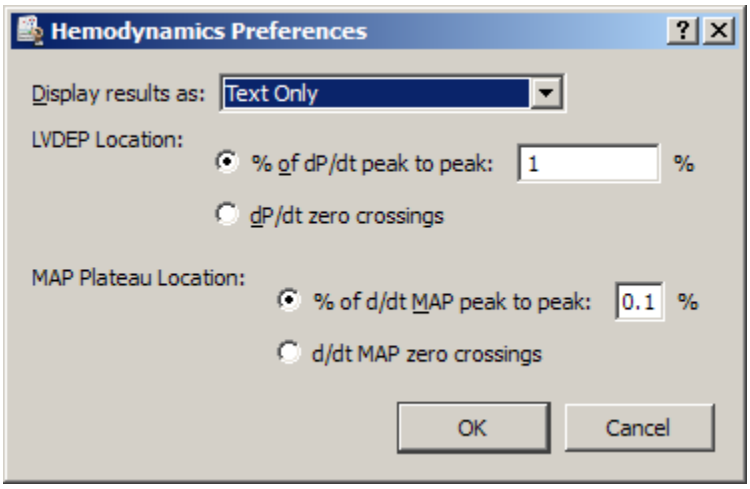
Max rate Maximum heart rate occurring during the expiration window of the respiration cycle, expressed in milliseconds (IBI).

- If a respiration cycle is invalid, this measure will be set to 0.

RSA RSA index for the respiratory cycle, expressed in milliseconds. This is the max rate minus the min rate. This is output in linear scaling. Conversion to logarithmic scaling must be performed manually, if desired.

- If a respiration cycle is invalid, this measure will be set to 0.

Preferences



Display results as

Several of these transformations produce large amounts of cycle-by-cycle derived measures. Results can be displayed as a tab delimited table in the journal, as waveforms in the graph, as an Excel spreadsheet or various combinations. Results are displayed as text-only by default.

LVEDP location method – see page 36

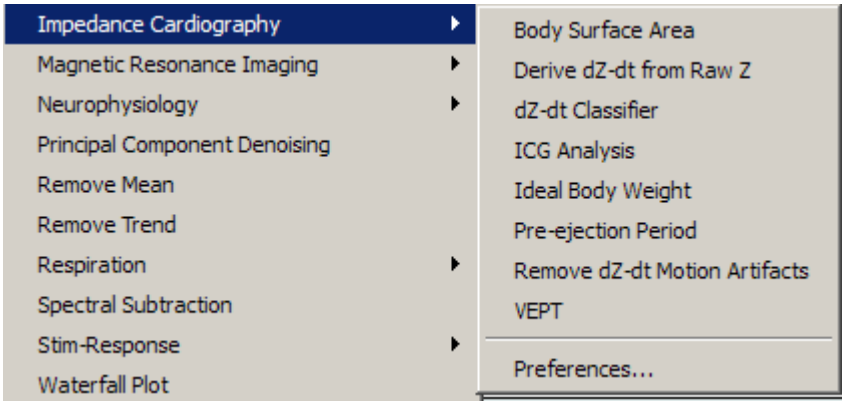
- adaptive threshold of 0 plus a % of pk-pk change in pressure
- first zero crossings before contraction on the dP/dt signal

MAP Plateau location method – see page 37

- adaptive threshold of 0 plus a % of pk-pk change in the derivative between the max and the first 0 crossing after the max
- second zero crossing after the maximum

Impedance Cardiography Analysis

The Impedance Cardiography analysis package assists in the analysis of cardiac output and other hemodynamic parameters using noninvasive bioimpedance monitoring techniques; signals must be sampled at 5 kHz or below to be analyzed with this package. It offers a variety of approaches for estimation of cardiac measures.



Body Surface Area

Determines the body surface area estimation in square meters for a subject of a given height and weight, using the formula set in Preferences. Prompts require height and weight of the subject in the preferred units. It can be used to calculate body surface area independent of any of the other analysis routines, which may be useful for validation purposes or other derived calculations.

Body Surface Area equation

Use the Preferences option to select an algorithm for estimating body surface area of a subject and deriving stroke volumes from impedance data.

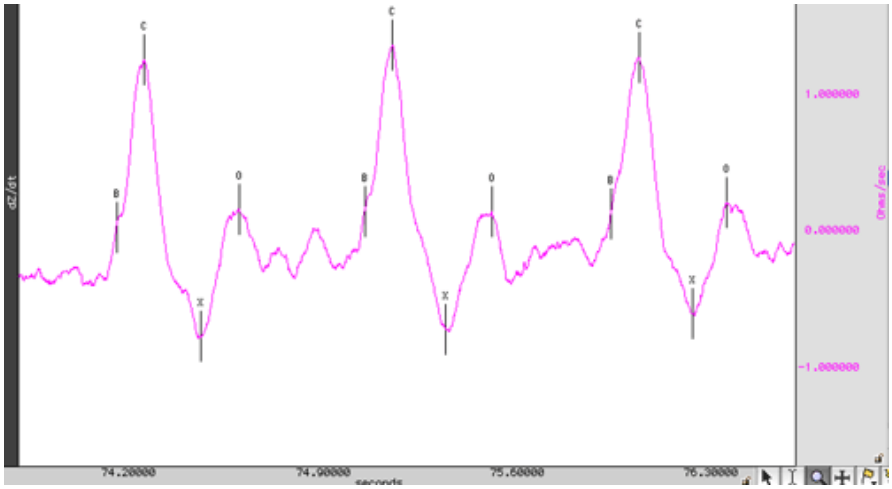
Method	*Formula*
Boyd	$BSA = 0.0003207 \times Height(cm)^{0.3} \times Weight(g)^{0.7285 - 0.0188 \log(Weight(g))}$
DuBois and DuBois	$BSA = 0.20247 \times Height(m)^{0.725} \times Weight(kg)^{0.425}$
Gehan and George	$BSA = 0.0235 \times Height(cm)^{0.42246} \times Weight(kg)^{0.51456}$
Haycock	$BSA = 0.024265 \times Height(cm)^{0.3964} \times Weight(kg)^{0.5378}$
Mosteller	$BSA = \sqrt{\frac{Height(cm) \times Weight(kg)}{3600}}$

dZ/dt Derive from Raw Z

This is a convenience utility for working with impedances recorded using the BIOPAC EBI100C amplifier or the raw impedance output of the BIOPAC NICO100C module. When computing derivatives from raw impedance signals from an EBI100C, this will apply appropriate filtering for a thoracic impedance signal and properly invert the derivative to match traditional dZ/dt presentation.

dZ/dt Classifier

Places events at common inflection points on a dZ/dt waveform to derive other measures.



The classifier will attempt to locate the following points on the ICG signal:

- B point – opening of aortic valve (set location in Preferences)
- C point – Maximum left ventricle flow (set location in Preferences)
- X point – Closing of aortic valve (set location in Preferences)
- Y point – Closing of pulmonal valve
- O point – Widest opening of mitrial valve

The algorithm for locating these points on the ICG signal examines local minima and maxima in the dZ/dt signal as well as values of its second derivative. Filtering is applied to the second derivative signal to improve accuracy.

- Pre-filtering the dZ/dt signal may improve accuracy slightly.

In a particular cardiac cycle, if there is not enough definitive change in the ICG signal to locate a particular point, the point will be omitted. This may most commonly occur with the Y point since its inflection between X and O is subtle and may be lost.

The location routine, as with impedance cardiography measurements in general, is sensitive to motion artifacts. It is intended to function on signals acquired from subjects at or near perfect rest. Swings in the dZ/dt signal may cause the classifier to fail. It is recommended that motion artifacts be removed before running the dZ/dt classifier or any other ICG analysis tools that may invoke the classifier on an ICG signal. If artifacts are present within the signal, the template matching cycle location method will exhibit better behavior than the fixed threshold method. The choice between these two methods can be made with the Preferences option of the analysis package.

B-point Location—Use Preferences to set the dZ/dt B-point location method.

There is no standard method generally accepted for programmatically locating B-points on an ICG waveform. The appropriate choice of B-point location method may depend on the data or on subjective preference. On average, all three methods will produce similar results for clean data. ICG Preferences has three options for B-point location:

- Second derivative classification – Given a C peak, it searches within a 150ms to 100ms time window before the C peak for the maximum of the second derivative of impedance ($\ddot{Z}$). The B point is placed at this maximum.
- Third derivative classification – Given a C peak, it searches for the maximum value of the third derivative of impedance ($\dddot{Z}$) within 300ms before the C peak. The B point is placed at this maximum.
- Cycle-by-cycle ‘Isoelectric’ crossings – Given a cycle defined by two C peaks, the mean of the dZ/dt signal is computed over the cycle. The B point is then placed at the closest time to the right C peak that is still underneath this baseline zero level.

C-point Location—Use Preferences to set the dZ/dt C-point location method.

In several of the ICG analysis scripts, the B, C, X, Y, and O points will need to be located on the dZ/dt waveform. The starting point of this process is locating individual cycles on the dZ/dt waveform to define the C points. Use Preferences to set the cycle location method:

- Template Matching – the user is expected to select a representative cycle of the dZ/dt waveform. The entire cycle should be selected (e.g. visually to approximate a C-C interval, a X-X interval, etc.). The entire dZ/dt signal is then correlated with that representative cycle, and individual cycles are picked out from locations of maximum correlation.
- Fixed Thresholding – the user is prompted to select one of the C peaks of the dZ/dt waveform. The voltage level of this peak is then used to compute an Ohms/sec thresholding level. Peak detection is then run on the dZ/dt waveform using that voltage level as the threshold.

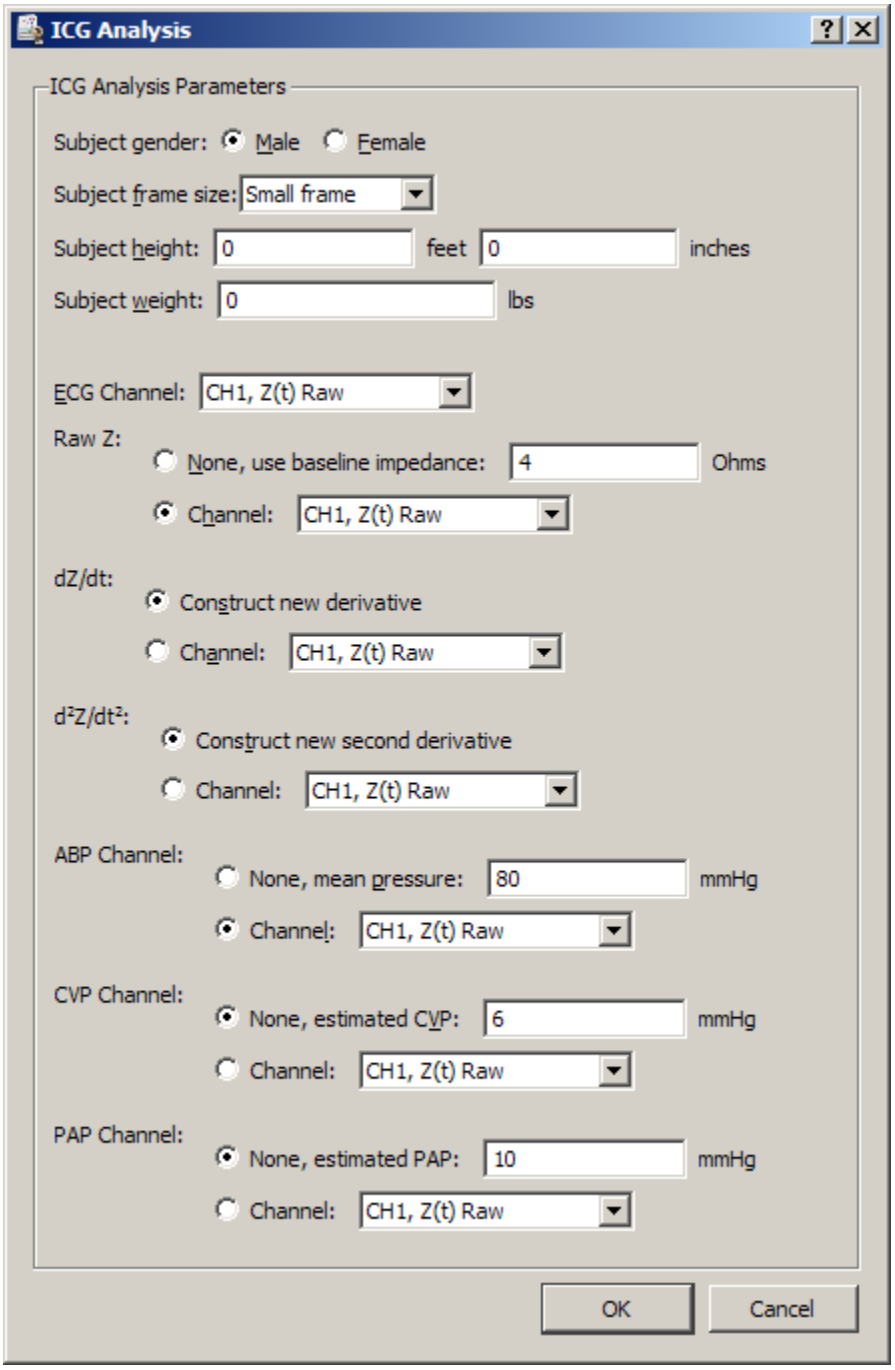
  Since ICG is subject to many artifacts such as respiration components and motion artifacts, the default method used is template matching. For extremely clean ICG signals, however, fixed thresholding can be used effectively as well and will provide a quicker analysis.
- Adaptive template matching – the user is prompted to select a representative cycle of the dZ/dt waveform. This is used as a basis for an adaptive match to locate cycles. Adaptive template matching will adapt to changes in the dZ/dt waveform as conditions change within the experiment. Two parameters may be set. The window size is the number of ICG cycles to use for estimating the next template. Smaller values will track changes more quickly; larger values will reduce interference from artifact. The correlation threshold is the value above which a match is found. It refers to the normalized cross correlation of dZ/dt with the template and should be between 0 and 1. Values closer to 1 will require precise matches and skip artifacts. Values closer to 0 will use looser match constraints and may be required if the ICG is changing rapidly.

X-point Location

There are two methods that may be used to locate the X point of the ICG waveform at the closing of the aortic valve. The choice of appropriate X point location method is dependent on the electrode configuration that is used to acquire the ICG signals. In certain electrode configurations, the dZ/dt minimum may actually occur closer to A than to X, making the first (and default) option of searching for the first turning point a more reliable solution. You may want to acquire a phonocardiogram in conjunction with ICG to help determine which method will be more accurate at locating X.

- Search for the first turning point in the dZ/dt signal that occurs after the C point location and place X at the first positive zero crossing in the second derivative of impedance (d^2Z/dt^2).This is the default X point location method.
- Locate the X point at the minimum value of dZ/dt over each cardiac cycle.

ICG Analysis



The ICG Analysis routines include 20 derived impedance and hemodynamic measures that correspond to various values that are generated by other industry-standard impedance cardiography analysis tools. Many users tend to be interested only in a subset of the various measures produced by the analysis (e.g. only heart rate and cardiac output); the extra measures can “clutter” the output and frustrate users who have to delete them manually.

The ICG Analysis output options feature adds a new step to the ICG Analysis where the user may toggle the output of individual measures on and off. This allows users to suppress generation of all output for a measure including the graph channels, column in the Excel spreadsheet, and column in the text output.

ICG Analysis performs a full impedance cardiography analysis on data, extracting intervals and derived cardiac measures. The minimal set of signals required to run this analysis is an ECG Lead II signal and either a raw impedance signal or a dZ/dt signal.

- If a raw impedance signal is present from an EBI100C or NICO100C and no derivative has been constructed, the analysis will automatically construct the appropriate derivative and perform classification.
- If both a raw impedance and a dZ/dt signal are present, the baseline impedance will be derived on a cycle-by-cycle basis to improve the accuracy of the analysis.
- If no raw impedance signal was acquired, a default fixed baseline impedance can be used.
- If a NICO100C amplifier is used, it is recommended that both the raw impedance and dZ/dt signals be acquired to improve analysis accuracy.
- To automatically apply motion filtering to the dZ/dt signal, use Preferences to enable Motion Filtering (see page 48).

In addition to the minimal set of signals, it is also possible to use arterial blood pressure, central venous pressure, and pulmonary arterial pressure signals to improve the quality of the algorithm results. If any of these signals are not present, default fixed estimated values can be substituted for the mean pressures instead of deriving pressures on a cycle-by-cycle basis.

ICG Analysis may potentially perform classification of both the dZ/dt and the ECG Lead II signals. The various notes for understanding the limitations of these classifiers apply and should be understood to properly interpret failures in the analysis.

ICG Analysis will produce the following information on a cycle-by-cycle basis:

At the end of the textual table an average of all of the cycle-by-cycle values will be appended.

Name	*Abbv.*	*Description*	*Units*	*Formula*
Acceleration index	ACI	Maximum blood acceleration	1 / sec^2	$\frac{\frac{d^2Z}{dt^2}_{max}}{TFI}$
Cardiac index	CI	Normalized cardiac output	m^2 / min	$\frac{CO}{BSA}$
Cardiac output	CO	Volume of blood pumped each minute	l / min	$SV \times HR$
Heart rate	HR	Heart rate in BPM as computed from the RR interval.	BPM	$\frac{60}{RR_i}$
Left cardiac work	LCW	Work exerted by the left ventricle each minute	kg m	$(MAP - PAP) \times CO \times 0.0144$
Left cardiac work index	LCWI	Normalized left cardiac work	kg m / m^2	$(MAP - PAP) \times CI \times 0.0144$
Left ventricular ejection time	LVET	Time interval between B and X. Time interval between aortic valve open and close.	sec	*Not applicable*
Mean blood pressure	MBP	Mean blood pressure as measured on the arterial blood pressure signal, or fixed estimate if no ABP signal is present.	mmHg	$P_{diastolic} + \frac{P_{systolic} - P_{diastolic}}{3}$
Mean central venous pressure	CVP	Mean central venous pressure over cycle, or default value if no CVP signal is present.	mmHg	*Not applicable*
Mean pulmonary arterial pressure	PAP	Mean value of the pulmonary arterial pressure of a cycle, or default value if no PAP signal is present.	mmHg	*Not applicable*

Name	*Abbv.*	*Description*	*Units*	*Formula*
Pre-ejection period	PEP	Time interval between the Q wave of the ECG and the B point of the ICG. Time interval between systole and aortic valve open.	sec	*Not applicable*
RR interval	RR-i	Time interval between R peaks in the waveform.	sec	*Not applicable*
Stroke index	SI	Normalized stroke volume	(ml / beat)/ m^2	$\frac{SV}{BSA}$
Stroke volume	SV	Volume of blood pumped by left ventricle in a single beat	ml / beat	Set equation in Preferences: Kubicek—Estimates SV from the derivative of the impedance signal and blood resistivity: $SV = \rho \times \frac{L^2}{Z_0^2} \times \frac{dZ}{dt}_{\max} \times LVET$ Note $\frac{dZ}{dt}_{\max}$ may be either the absolute maximum or the BC delta in amplitude, as set in Preferences. ▪ Sramek—Estimates SV from the derivative of the impedance signal and the estimated volume of electrically participating fluid (VEPT): $SV = \frac{VEPT}{Z_0} \times \frac{dZ}{dt}_{\max} \times LVET$ o In the ICG analysis routines, VEPT is estimated using a truncated cone model. $VEPT = \frac{(0.17H)^3}{4.25}$ ▪ Sramek-Bernstein—Estimates SV from the volume of electrically participating tissue scaled according to body habitus. The SV equation is: $SV = \frac{\delta(VEPT)}{Z_0} \times \frac{dZ}{dt}_{\max} \times LVET$ *where* $\delta(VEPT) = \frac{weight_{actual}}{weight_{ideal}} \times \frac{(0.17H)^3}{4.25}$ Ideal body weight is computed using the method set in the Preferences. To best match the original Sramek-Bernstein equation, use the Met Life Tables ideal body weight method.
Systemic vascular resistance	SVR	Afterload; arterial flow resistance	dynes sec / cm^5	$80 \times \frac{MAP - CVP}{CO}$

Name	*Abbv.*	*Description*	*Units*	*Formula*
Systemic vascular resistance index	SVRI	Normalized afterload	dynes sec m^2 / cm^5	$80 \times \frac{MAP - CVP}{CI}$
Systolic time ratio	STR	Ratio between electrical and mechanical systole	none	$\frac{PEP}{LVET}$
Thoracic fluid content	TFC	Electrical conductivity of the chest cavity	1 / Ohms	$\frac{1}{TFI}$
Thoracic fluid index	TFI	Mean value of the raw impedance over the cycle, or fixed baseline value if no raw impedance signal is present.	Ohms	*Not applicable*
Velocity index	VI	Maximum velocity of blood flow in the aorta.	1 / sec	$\frac{\frac{dZ}{dt}_{max}}{TFI}$

Note $\frac{dZ}{dt}_{max}$ may be either the absolute maximum or the BC delta in amplitude, as set in Preferences.

Ideal Body Weight

Body Weight is derived from a person's height, gender, and (for the Met Life method) frame size. It describes the ideal weight based upon various estimates. Ideal body weight is subject to much interpretation, so a number of methods are provided. Ideal Body Weight results are always expressed in kilograms.

Use Preferences to set the Ideal Body Weight computation method; the selected method is also used for ICG Analysis.

Method	*Formula*
Devine	*Men* 50 kg + 2.3 kg per inch over 5 feet *Women* 45.5 kg + 2.3 kg per inch over 5 feet
Metropolitan Life Tables	The weight is taken from the standard Metropolitan Life tables, which are based on gender, height, and frame size. The Metropolitan Life tables specify weight ranges; the ideal body weight is computed as the average of the endpoints of each weight range. Ideal weights are based on height with shoes on and are only defined for heights between *Men* 5' 2"and 6' 4" *Women* 4' 10"and 6' 0"
Miller	*Men* 56.2 kg + 1.41 kg per inch over 5 feet *Women* 53.1 kg + 1.36 kg per inch over 5 feet
Robinson	*Men* 52 kg + 1.9 kg per inch over 5 feet *Women* 49 kg + 1.7 kg per inch over 5 feet

PEP Pre-ejection Period

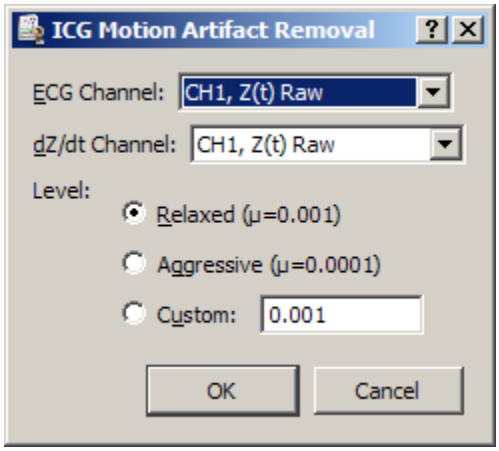
The pre-ejection period is the time interval between the electromechanical systole and the onset of ejection of blood from the left ventricle of the heart. This can be derived from standard ECG data and ICG data as the interval between the Q point on the ECG and the B point on the ICG. The Pre-ejection Period analysis tool helps extract PEP measurements from ECG Lead II and ICG data. PEP can also be computed using the full ICG Analysis tool on page 43.

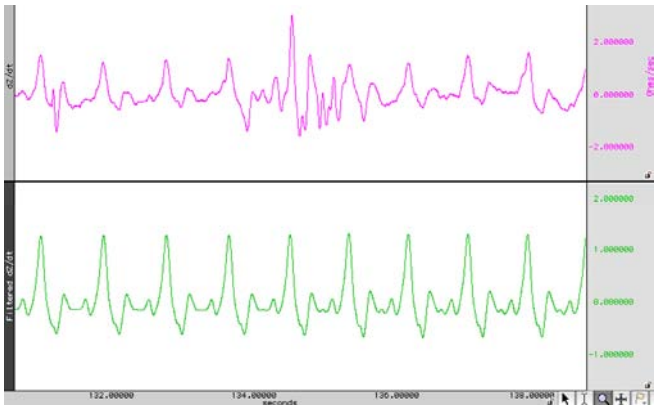
To use Pre-ejection Period analysis, both an ECG Lead II and an ICG (dZ/dt) signal must be present. If either of these signals requires classification, the analysis will run the appropriate classifier to define the relevant events on the signals. To automatically apply motion filtering to dZ/dt, use Preferences to enable Motion Filtering (see page 48).

PEP analysis will output the following information on a cycle-by-cycle basis and the final line of the textual output will be the average of all of the cycle measurements. All time unit output is in seconds unless otherwise noted.

Name	*Abbrev.*	*Description*
Heart rate	BPM	The heart rate for the cycle as indicated in BPM. Derived from the RR interval.
Pre-ejection period	PEP	Time interval between the Q wave of the ECG and the B point on the ICG for the cardiac cycle. If the PEP cannot be computed for a particular cycle, it will have the value "----" in the textual output or 0 in the graphical output.
RR interval	RR-i	Time interval between R peaks of a single cycle of cardiac data.

dZ/dt Remove Motion Artifacts





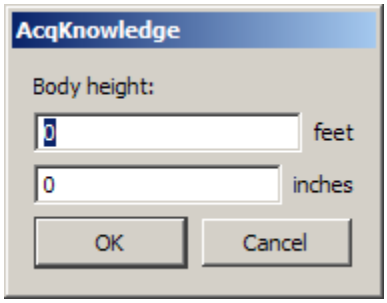
Applies SFLC motion artifact removal to a dZ/dt signal. Uses cycle information from an ECG signal to construct a sinusoidal model of the ICG signal containing only components that are correlated to the heart rate.

IMPORTANT

Motion artifact removal will affect the amplitudes of the dZ/dt signal, so results derived from a motion filtered dZ/dt signal should be additionally verified for accuracy.

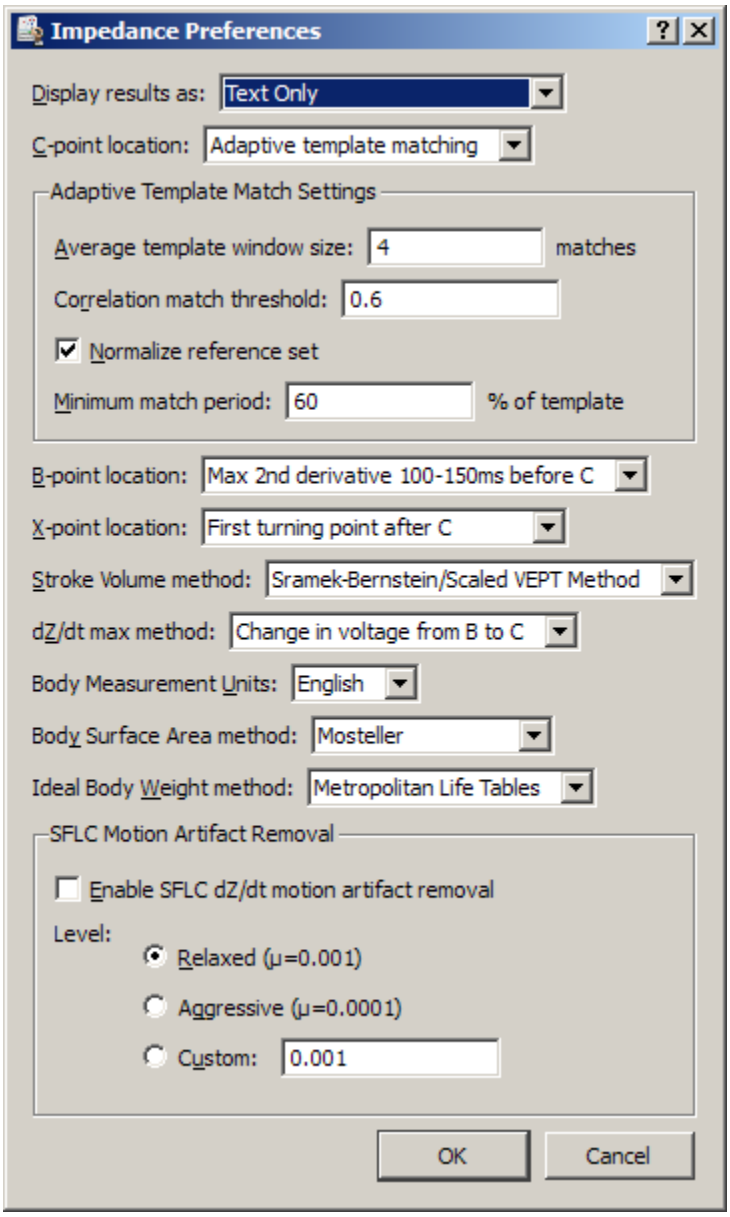
This tool performs the same type of filtering as the ICG Analysis and Pre-ejection Period tools when the Motion Filtering preference is enabled.

VEPT



Uses the truncated cone method to compute the volume of electrically participating tissue (VEPT) in cubic centimeters of a subject. You will be prompted to enter the height of the subject in the units set under Preferences. It can be used to calculate VEPT independent of any of the other analysis routines, which may be useful for validation purposes or other derived calculations.

Preferences



Display results as

- Textual tables in the journal
- Channels of data inserted into the graph.

C-point location –see page 41

B-point location – see page 41

X-point location – see page 42

Stroke volume equation – see page 45

- Kubicek, or Sramek, or Sramek-Bernstein

dZ/dt Max method – Baseline drift in ICG signals can introduce drift artifacts into stroke volume, cardiac output, and other measures that are sensitive to changes in dZ/dt max. The Preferences offer two settings. “Max dZ/dt in cardiac cycle” will extract the maximum amplitude of dZ/dt as the max value. This is the traditional way of measuring dZ/dt max. A second estimate option, “change in voltage from B to C” will take the amplitude delta between B and C as the estimate of dZ/dt max. This will produce different stroke volume results, but is useful for removing motion artifact and improving consistency.

Body Measurement Units system for inputting

- English system: body height in feet and inches, distance between measuring electrodes in inches, and body weight in pounds
- Metric system: body height in meters and centimeters, distance between measuring electrodes in centimeters, and body weight in kilograms.

Body Surface Area equation – see page 40

- Boyd; DuBois and DuBois; Gehan and George; Haycock; or Mosteller

Ideal Body Weight method– see page 46

Motion Artifact Removal

The Pre-ejection Period and ICG Analysis transformations have the ability to optionally apply motion filtering automatically to the dZ/dt signal. Motion filtering is performed using an SFLC keyed to the R waves of an ECG signal. The SFLC filtering approach is similar to performing cycle-by-cycle averaging of the dZ/dt signal. This motion filtering approach may cause errors to be introduced in derived calculations, so any results with motion filtering turned on should be validated additionally.

Filter Magnitude Level – relaxed, aggressive, and custom.

- “Relaxed” uses a SFLC step size of .001. This allows the filter to adapt moderately quickly to changes in the dZ/dt signal.
- “Aggressive” uses a SFLC setting of .0001. The filter will adapt much less quickly to changes in the ICG signal, allowing better filtering out of motion artifacts at the expense of a lessened response to changes in underlying ICG morphology.
- “Custom” allows for an arbitrary SFLC step size. The step size must be greater then zero and much less than 1 for the filter to converge.

Magnetic Resonance Imaging

Artifact Frequency Removal
Artifact Projection Removal
Median Filter Artifact Removal
Signal Blanking

Magnetic resonance imaging, or MRI, is often used to study the brain and other organs in the body. As access increases to MRI machines, researchers are beginning to combine MRI with traditional physiological signal recording. The strong magnetic fields used by MRI equipment can cause profound artifacts in physiological recordings, which can make the analysis of physiological recordings acquired in an MRI difficult. Some artifacts are external interference while other artifacts can be caused by currents being induced in electrode leads or even in the body itself.

Artifact Location and Trigger Signals

Most of the MRI analysis options require information to identify the positions of various artifacts. Event positions can be used or a "trigger signal" waveform in the graph can be used to identify periods when the MRI is active. Some MRI machines have a TTL output that is synchronized with periods where the MRI is on.

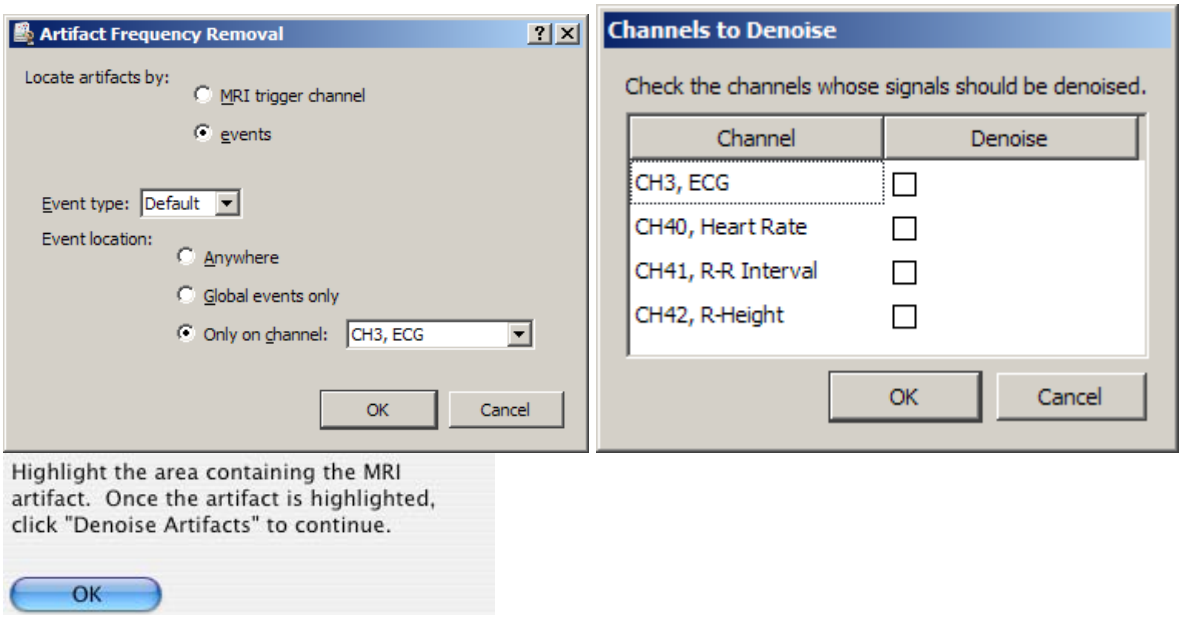
- Whenever possible, this trigger signal should be acquired with the MP unit along with the physiological data.

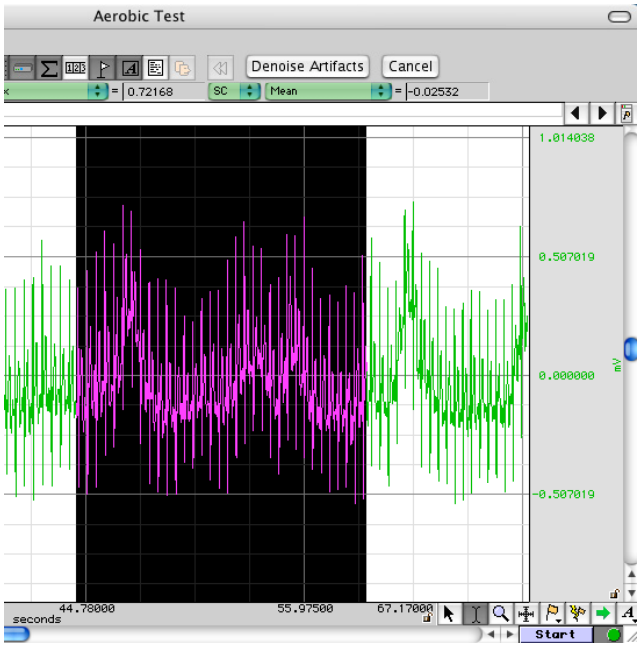
Trigger detection off of an MRI trigger signal waveform is performed using fixed level thresholding on the waveform data. The threshold level is set to be the minimum value of the entire trigger signal plus 1/10th of the peak-to-peak distance of the trigger signal. The threshold is kept data dependent to allow for artificial trigger signals to be derived from data if the MRI unit does not provide its own. The trigger signal may be acquired on either an analog or digital channel.

Event driven artifact location can be useful when trigger signals are not available from the MRI or are not recorded. A cycle detector analysis can be used to place events at the onset of each artifact, or these events may be placed manually. Event based detection is also useful for applying the procedures for artifacts that are not directly related to the MRI trigger signal, such as for removing the cardiac interference from EEG data caused by the magnetic field of the MRI machine.

Artifact Frequency Removal

MRI > Artifact Frequency Removal





Two large sources of interference in MRI recordings are the current induced by the MRI magnetic field and the RF pulses used for triggering molecule alignment. While the overlap of this interference may be difficult to separate in the time domain, the MRI interference may have a distinctive signature in the frequency domain.

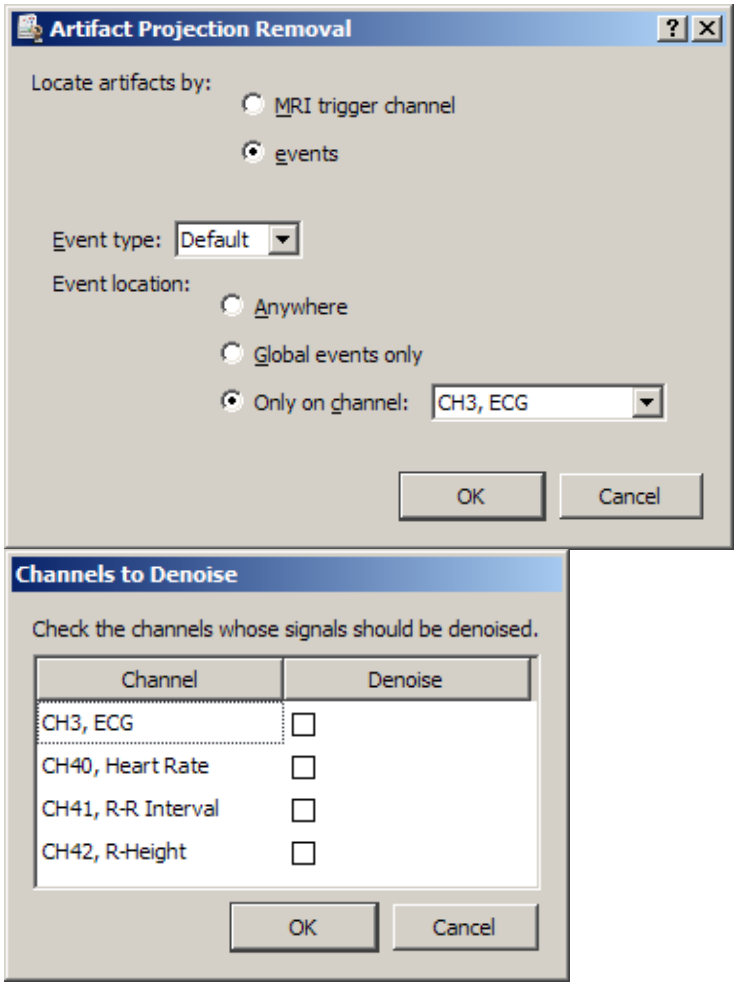
Artifact Frequency Removal is a frequency domain adaptation of the ensemble projection removal of the Artifact Projection Removal transformation. It attempts to cancel out MRI artifact by removing the frequencies most strongly associated with the MRI signal.

For each channel of data to be denoised, either the MRI trigger signal or event positions are used to locate periods of MRI activity for constructing an ensemble average. The FFT of this ensemble average is computed, and the magnitude of the average FFT is set as the reference. Cyclic mean removal is applied to each period of artifact to compensate for baseline drift or signals with expected DC offset. A second pass is then made through the data. For each individual artifact, the FFT of that artifact is computed and the projection of that FFT onto the average FFT is removed. After projection removal, negative Fourier components are discarded and a time-domain signal is reconstructed using the inverse Fourier transform. This reconstructed, filtered signal is used to replace the MRI artifact in the original data.

Application of projection removal in the frequency domain has similar limitations to applying it in the time domain, that is, it assumes that the MRI interference is stationary (which is not necessarily the case). Variations in the MRI interference may cause this method to fail.

IMPORTANT Artifact Frequency Removal requires an MRI triggering signal or artifact onset events to locate artifact positions.

Artifact Projection Removal



Artifact Projection Removal attempts to remove the noise components from the artifacts within a signal. An ensemble average is made for each period of MRI artifact in a channel. Cyclic mean removal is applied to each period of artifact to compensate for baseline drift or signals with expected DC offset. As the artifacts are averaged together, the actual interference with the physiological signal caused by the MRI should become the dominant feature if a sufficient number of artifacts are present. A second pass is made through the artifacts to remove this average MRI artifact from each individual period.

The average artifact is removed using the Remove Projection transformation. This performs a vector projection of the signal onto the averaged artifact estimation and subtracts this projection. This is an improvement over straight subtraction of the average artifact as vector projection can compensate for changes to amplitude that may occur over time.

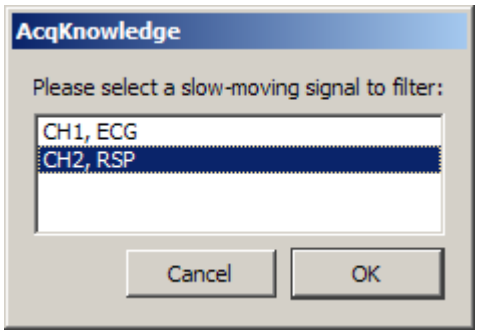
Artifact projection removal cannot compensate for MRI interference that varies in frequency due to changes in orientation of electrode leads within the MRI or other factors that may alter the MRI artifact.

Artifact projection removal is an adaptation of a denoising technique described in:

M. Samonas, M. Petrou and A. Ioannides, “Identification and Elimination of Cardiac Contribution in Single-Trial Magnetoencephalographic Signals,” *IEEE Trans. Biomed. Eng.*, vol. 44, no. 5, pp. 386-393, 1997.

IMPORTANT Artifact Projection Removal requires an MRI triggering signal or artifact onset events to locate artifact positions.

Median Filter Artifact Removal

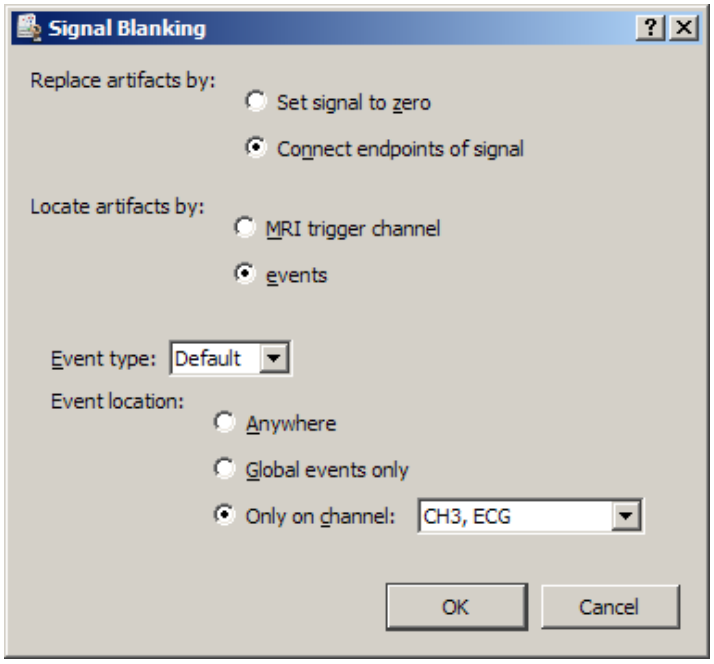


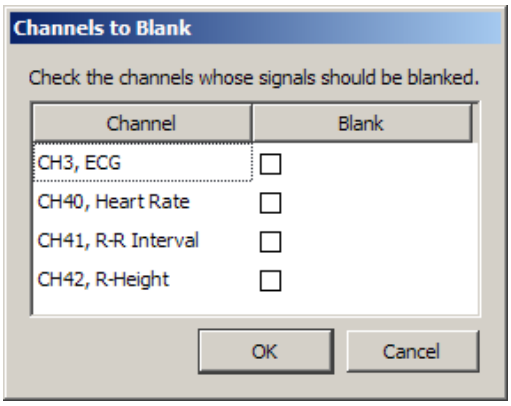
Median Filter Artifact Removal provides a basic artifact removal suitable for slow moving signals such as respiration, GSR, or temperature. It performs a windowed median transformation on the source channel with a window width of 1/10th of the acquisition sampling rate.

This median filtering approach is explained in the BIOPAC MRI application note AH223.

Median Filter Artifact Removal does not require an MRI triggering signal.

Signal Blanking





MRI artifact can grossly distort low level physiological signals, and this distortion can be several orders of magnitude larger than the signal of interest. A common practice for analyzing the physiological data is to discard the MRI artifacts and only examine the portions of the signal in between the MRI artifacts. One approach for this is outlined in BIOPAC MRI application note AH223.

Signal Blanking provides an alternate approach for discarding MRI artifacts from the signal. Using the MRI triggering signal or artifact event locations, this analysis option will locate the periods of MRI activity and "blank" the physiological signal during this period.

Two types of "blanking" can be performed:

- Set value to zero – The waveform is set to zero during each artifact.
    - For integrated measures, zeroing the signal may be preferable as it will have no effect on the running sum.
- Connect endpoints – For each artifact, a selection is made and the values within the interval are replaced with a line connecting the signal value before the MRI artifact to the signal value at the end of the interval.
    - For statistical measures or DC coupled signals, connect endpoint (linear interpolation within the interval) may be preferable to avoid causing the output to trend towards zero.

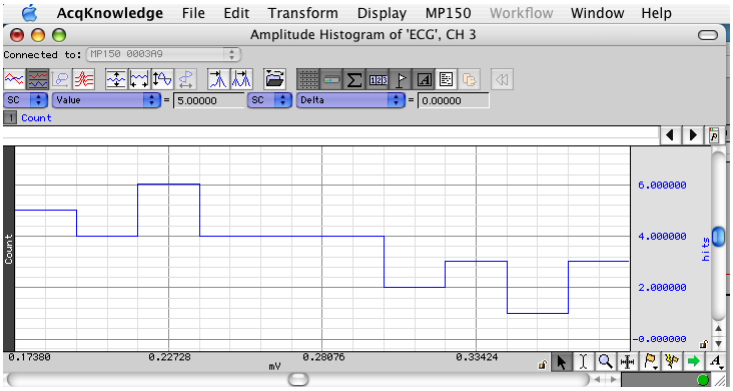
IMPORTANT Signal Blanking requires an MRI triggering signal or artifact onset events to locate artifact positions.

Neurophysiology

The Neurophysiology analysis package assists in the analysis of spikes within extracellular microelectrode recordings, such as those recorded using an MCE100C module. All of these analysis options require a continuous recorded single channel of microelectrode data.

- A *spike* is a deviation from the baseline caused by a neuron action potential. Frequently extracellular spikes will resemble exponentials. The point of maximum value of the spike will be used to locate neuron firing.
- A spike *episode* consists of a fixed time window around a spike that aims to capture the underlying neuron firing time. The episode consists both of the rise time (the time taken to reach maximum) and the relaxation period around the spike.

Amplitude Histograms

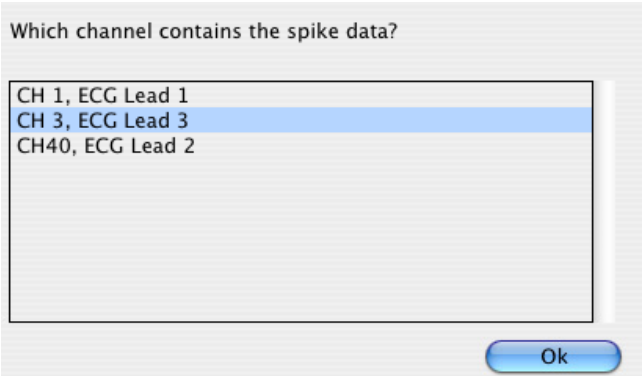


IMPORTANT To run this analysis option, the signal must first be transformed by the Locate Spike Episodes option or the Classify Spikes option.

Amplitude histograms show the population density of the maximum amplitude of neuron firing events. They may be used to interpret changes in neuron firing due to drug response or as rough indicators of the approximate number of classes of action potentials in a signal. Amplitude histograms can be generated on classified or unclassified signals.

- On classified signals, an overall amplitude histogram will be created for all of the spikes in addition to a single amplitude histogram per class (reflecting only the episodes of that class.
- On unclassified signals, a single amplitude histogram will be created from the maximum voltage within all of the spike episodes.

Average Action Potentials

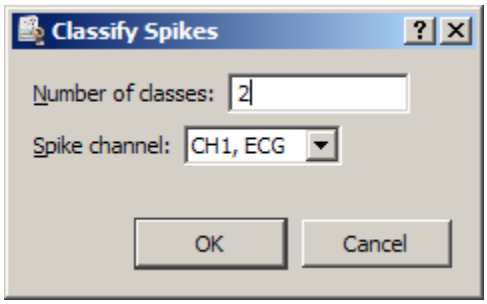


IMPORTANT To run this analysis option, the signal must first be transformed by the Locate Spike Episodes option or the Classify Spikes option.

After a classification has been completed for a spike signal, to assign spike episodes to different groups, users may wish to view the average shape of the waveforms of each class. Examining the shape of the different classes provides visual feedback as to the efficiency of the clustering, can allow for identification of certain classes as noise or artifacts, and helps to determine if the identified classes are indeed unique. Average Action Potentials can be generated on classified or unclassified signals.

- On classified signals, the resulting ensemble averages will have multiple channels.
  - The first channel will be the overall average of all of the spike episodes.
  - The remaining channels show the average of the members of each individual spike class.
- On unclassified signals, a graph will be produced with a single channel showing the average of all of the spike episodes.

Classify Spikes



IMPORTANT If cluster events from a previous spike classification are already defined on the recorded waveform, this option will erase them and replace them with the new classification of the potentials.

This analysis option will automatically classify action potentials in microelectrode data and divide them into different spike classes.

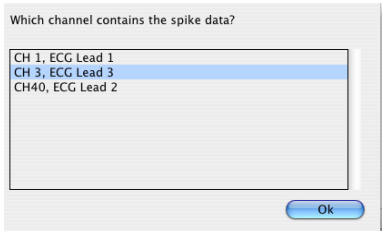
A single-feature k-means clustering classifier is used, and the entire data set is used for the clustering portion of the algorithm. The determining feature is the Sum criteria— that is, the sum of all of the data points within the waveform segment; this was one of the first features used in early action potential classifiers.

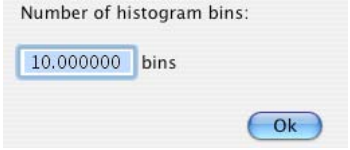
If the Locate Spike Episodes option has not been used to find spikes before this option was selected, the Locate Spike Episodes option will be automatically performed prior to the clustering.

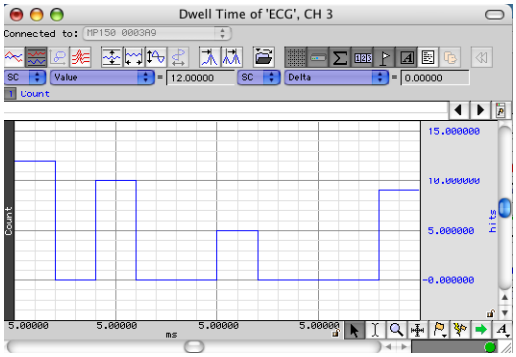
The analysis routine will ask for a number of spike classes and then use k-means clustering to group each spike episode into a class. The clustering may not produce meaningful classes, so results should be examined for accuracy.

This style of classifier is for rudimentary spike analysis. For more advanced classification techniques, use the clustering algorithm in the peak detector.

Dwell Time Histograms







IMPORTANT To run this analysis option, the signal must first be transformed by the Locate Spike Episodes option or the Classify Spikes option.

A dwell time histogram shows the population density of the duration of a neuron firing event. Dwell times can be approximated for an action potential by measuring the absolute value of the time interval between their maximum and minimum voltage levels reached during the firing of the neuron. After the minimum value in the firing recording has occurred, the neuron will be returning to its resting state, so the time difference is a good approximation for the firing duration. The dwell time histogram plots this time difference versus number of action potentials that have similar time differences. Examining varieties in dwell times can help to illustrate drug responses or to perform rudimentary classification of action potentials.

Dwell times will be defined as the time difference between the positions of the maximum signal value and minimum signal value within a spike episode. Since dwell time histograms can be used for classification purposes, they can be run on classified or unclassified microelectrode signals.

- On classified signals, an overall dwell time histogram will be constructed for all of the spikes in addition

to a single histogram per class, showing times of only the spikes in that class.

- On unclassified signals, a single dwell time histogram will be created for all of the spikes. When run on a classified signal.

Find Overlapping Spike Episodes

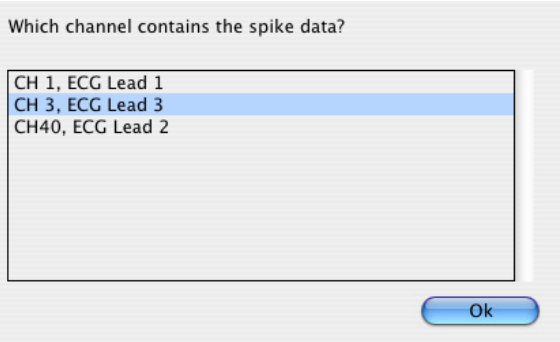
IMPORTANT To run this analysis option, the signal must first be transformed by the Locate Spike Episodes option or the Classify Spikes option.

In many extracellular recordings, it is frequent for there to be more than one neuron firing in response to the same stimulus. This can result in overlapping spike episodes when both neurons fire in close succession. Some types of analysis and spike classification are not able to produce meaningful results if too many overlapping episodes occur. “Find Overlapping Spike Episodes” can be used to locate overlapping episodes. After the spikes have been located in a signal, this option can be used to iterate only to those that are overlapping.

“Next Overlap” and “Cancel” buttons are available in the toolbar of the graph window to allow for iteration through the episodes.

Note This option is “view only.” Overlapping episodes are not affected by the analysis and will need to be manually removed manually to delete them from the file.

Generate Spike Trains



IMPORTANT To run this analysis option, the signal must first be transformed by the Locate Spike Episodes option or the Classify Spikes option.

Spike trains are good visual indicators of when action potentials are firing and are good synchronization waves for further analysis and data reduction. A spike train is a channel in a graph whose value is 0 when there is no spike and 1 when there is a spike.

Spike train generation will operate only on signals whose action potentials have already been classified.

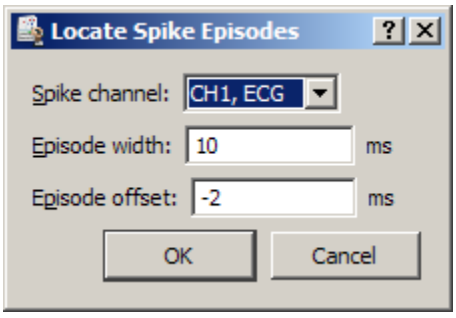
A single spike train will be generated as a channel in the graph for each class of action potential in the signal.

If text output is enabled, the spike trains will be pasted as tables in the journal with one table per spike class.

If spreadsheet output is enabled, the tables will be placed side by side so index 1 of the tables lines up for each action potential.

Locate Spike Episodes

Neurophysiology > Locate Spike Episodes



This option provides the basic spike detection for a microelectrode signal. Spike detection is performed using the following steps:

1. Obtain mean value of the entire signal.
2. Obtain standard deviation of the entire signal.

3. Detect spikes where the signal rises above a fixed threshold determined by adding a multiple of the standard deviation to the mean.
4. Position the episode around the threshold crossings according to the width and offset entered previously.

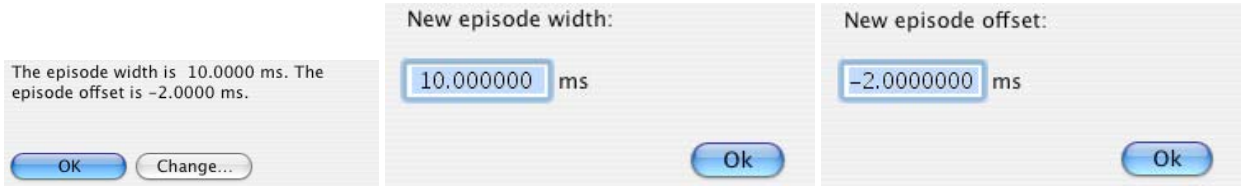
A "Spike Episode Begin" event will be placed at the start of each spike episode and will be located offset milliseconds away from the threshold crossing. A "Spike Episode End" event will be placed at the end of each episode.

If text output is enabled, a table of the start time of each episode will be placed in the graph's journal.

If spreadsheet output is enabled, a new spreadsheet will be created with the start time of each episode.

Spike episodes may also be located manually by using the Cycle Detector to define "spike episode begin" and "spike episode end" events in the graph.

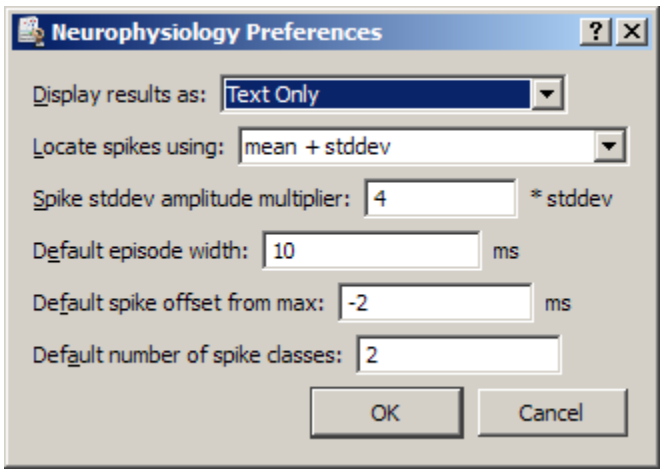
Set Episode Width and Offset



The first time spike detection is performed on a graph, the episode width and offset need to be entered. This width and offset is remembered and is used for all future spike detections in the graph performed by "Locate Spike Episodes" and other transformations. The width and offset that are entered are retained even if the file is saved and reopened.

Use this option to view or change the current width and offset.

Preferences

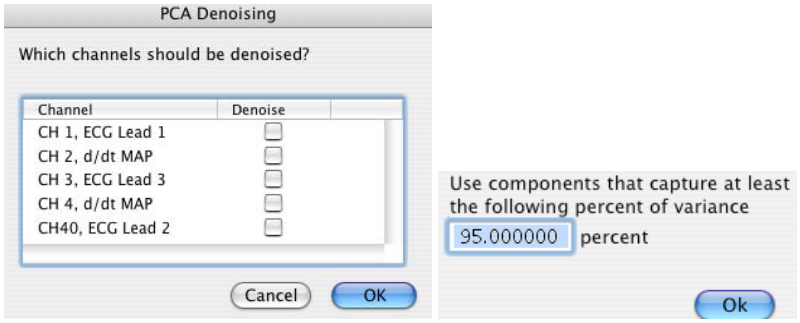


Preference	*Description*	*Default Setting*
Output type	Determines whether analysis results will be displayed as graph channels, textual tables in the journal, or Excel spreadsheets. Not all of the output types are applicable for each Neurophysiology analysis option.	Text output to journal only.

Preference	*Description*	*Default Setting*
Spike Location Method	Choose how spikes are searched for in the signal. **Mean + Stddev**—uses fixed level peak detection with a level that is computed from the mean value plus a configurable number of standard deviations of the data. **Amplitude/Half-width Discriminator**—allows for basic isolation of spike shapes that have peak voltages within a configurable range and spike half-widths within a configurable range; uses the amplitude of the spike as well as the width of the spike to determine what constitutes a valid spike event. *Half-width* For a given spike, the discriminator searches from the maximum value of the spike to both the left and right of the maximum for the sample positions where the value has dropped below 50% of the maximum. The time interval between these sample positions is defined as the estimate of the half width. The acquisition sampling rate can be increased to improve accuracy of the spike half width estimates as neuron firing events involve high frequency components. Each time the discriminator is run, the user must input the amplitude low and high values as well as the minimum and maximum spike width. The discriminator searches for spikes in a signal *x* as follows: 1. Performs regular peak detection on *x* using a fixed threshold a_{low}. This locates the local maxima occurring after each threshold crossing of the low amplitude area. This results in a sequence *p* of potential spike locations. 2. Computes the half-width time interval for each potential spike location *p*. 3. Accepts the spike for each potential spike location *p* as a valid spike *s* if, where *a* is amplitude and *t* is time window: $a_{low} \le x(p) \le a_{high} \wedge t_{low} \le t_{half}(p) \le t_{high}$ 4. For each valid spike location *s*, positions the spike episode start output at *s+offset* and the spike episode end output at *s+offset+width* . The spike discriminator generates output only for spikes that fall within the bounds of the amplitude and offset windows. If a spike candidate falls outside the windows, no output is generated.	Mean + Stddev
Spike Detection Level	Spikes are located in the signal by looking for locations where the signal deviates from its baseline by a certain number of standard deviations. This multiplier is set in this preference.	4 standard deviations
Default Episode Width	The first time that any of the spike detection is run on a graph, the time width of each fixed width episode must be specified. This preference provides the default value that is seeded in the dialog. The episode width for an individual graph does not need to match this default.	10 milliseconds

Preference	*Description*	*Default Setting*
Default Episode Offset	Each fixed width episode is located around one of the spikes in the signal. The offset allows for the episode to begin before (or after) the spike threshold crossing so the leading edge of the spike can be captured. Negative numbers indicate episodes are to start before the spike threshold crossing, positive numbers indicate episodes that start after.	-2 milliseconds
Default Number of Spike Classes	The Classify Spikes script requires the user to input the number of classes into which the spikes will be partitioned. This preference allows the default number to be modified. The number of classes that wind up being used does not need to match this default.	2

Principal Component Denoising



Removes noise from certain types of signals. For principal component denoising to be effective, more than two signals should be used as sources and all source channels must have identical waveform sampling rates. PCA denoising is most effective on signals that are known to contain a high degree of similarity, such as multiple ECG leads or multiple EEG leads. PCA denoising should not be used on signals of different types or units as all of the principal components may be needed to fully capture the differences in the signals.

➲ To determine if PCA denoising will be effective on a particular set of data or to compute a appropriate variance percentage for denoising, examine the principal components directly with “Transform > Principal Component Analysis” before selecting this option.

Given a set of signals, a principal component analysis is performed. The strengths of the components are then analyzed, and the original signals are reconstructed from a subset of the principal components that capture a certain percentage of the total variance of the signals. This essentially eliminates one or more of the higher-order principal components. For certain types of signals, these principal components are the ones that model the noise inherent in the signals.

1. Check the “Denoise” column for the channels to be denoised.
2. Enter the overall percentage of the variance.
3. After the percentage is entered, the denoising process will begin.

Remove Mean

Available in Acq*Knowledge* 4.1 and above—Remove Mean allows for mean subtraction to be performed for the selected area (or entire wave if no data is selected). It will result in the mean value being the new zero value for the waveform.

Remove Trend

Available in Acq*Knowledge* 4.1 and above—Remove Trend helps to remove baseline drift or other linear trends from data. This tool makes it easier to apply trend removal to only specific segments of a waveform. Given a selected segment of data, or an entire waveform, it computes the trend between the two endpoints (similar to the Slope measurement) and then removes this trend from the selected area such that the endpoints of the selection lie at the same voltage.

Linear Regression

Use linear regression to estimate the trend to be removed from the waveform.

Slope between endpoints

- **Left** keeps the starting point of the selection fixed at the same voltage. The software adjusts the data from left to right such that the right endpoint is aligned with the initial starting voltage.
- **Right** keeps the ending point of the selection fixed at the same voltage. The software adjusts the data from right to left such that the left endpoint is aligned with the initial ending voltage.

Respiration

***IMPORTANT*—Respiration analysis assumes a bidirectional airflow signal that records both inhale and exhale. Unidirectional respiration signals cannot be analyzed at this time.**

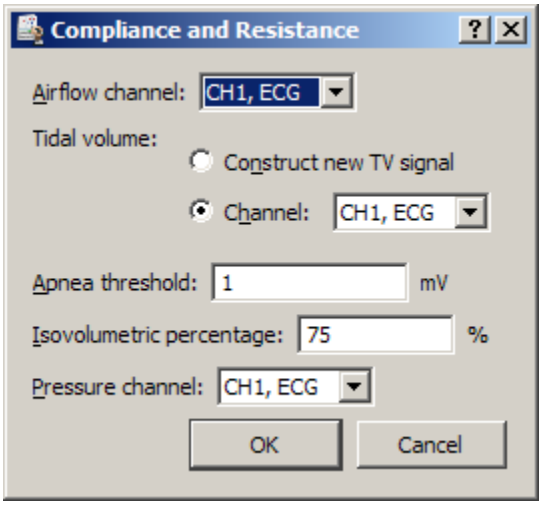
The respiration analysis package helps to analyze respiration- and airflow-related data. Other tools exist for respiration related analysis including Acq*Knowledge* transformations and the Respiratory Sinus Arrhythmia analysis in the Hemodynamics analysis package.

Compliance and Resistance

Compliance and Resistance analysis can be used to extract pulmonary resistance and pulmonary compliance in addition to basic airflow measures. This analysis requires an airflow signal and a pressure signal. The analysis will extract all of the measures of the Pulmonary Airflow analysis for the airflow signal. It also will locate apnea periods after exhalation using the same user-configurable threshold method as the Pulmonary Airflow analysis.

***IMPORTANT*— The flow signal must be recorded correctly for Compliance and Resistance analysis to work.** Compliance and Resistance analysis assumes positive flow indicates inhalation and negative flow indicates exhalation (the flow conventions of the recommended connections for a BIOPAC TSD107 pneumotach or a TSD117 airflow transducer).

- If the flow signal was recorded with exhalation positive instead of inhalation positive, multiply the flow signal by -1 to invert the signal.



Pulmonary resistance is computed using the isovolumetric method. On both sides of the tidal volume peak for a breath, the position where the volume reaches a user-specified percentage of the tidal volume is located. The pulmonary resistance is defined as the difference in pressure divided by the difference in flow at these two isovolumetric positions. Due to the discrete nature of sampled data, these points may not be exactly equal in volume. To improve the accuracy of the isovolumetric method, increase the sampling rate used to acquire data.

Dynamic pulmonary compliance is extracted on a breath-by-breath basis by dividing the tidal volume by the change in pressure between the exhale start and inhale start locations of the breath.

Individual breaths are defined as the period between consecutive Inhale Start events. Airflow units are assumed to be the standard liters/sec and pressure units mmHg. For each breath period, the analysis will define the following events:

- Inhale Start event on flow signal at start of inhale
- Exhale Start event on flow signal at start of exhale
- Apnea Start event on flow signal at beginning of apnea period (if present)
- Recovery events on volume signal at isovolumetric positions to left and right of tidal volume peak

If Inhale Start and Exhale Start events are already present on the flow signal at the start of analysis, those events will be used to define the breath periods. Apnea Start and Recovery events will always be regenerated by the analysis.

The analysis will extract the following measures from the data:

Name	*Abbrev.*	*Description*	*Units*
Cycle		Index of the breath in the data, beginning at 1.	
Time		Starting time of the inhale of the breath.	seconds
Peak Inspiratory Flow	PIF	Maximum absolute flow occurring during the inhale portion of the breath.	liters/sec
Peak Expiratory Flow	PEF	Maximum absolute flow occurring during the exhale portion of the breath.	liters/sec
Tidal volume	TV	Total volume of air inhaled during the breath.	liters
Minute volume	MV	Volume of air that would be inspired during a minute given the tidal volume and breathing rate of this breath. TV * BPM	liters/ minute

Name	*Abbrev.*	*Description*	*Units*
Breaths per minute	BPM	Breathing rate. $\frac{60}{TT}$	BPM
Inspiration time	IT	Time interval between the start of inhale and the start of exhale.	seconds
Exhalation time	ET	Time interval between the start of exhale and either: • start of apnea (if apnea present) • start of subsequent breath (if no apnea present)	seconds
Total breath time	TT	Time interval between the start of inhale and the start of inhale of the following breath. This is the sum of the inhalation time, exhalation time, and apnea time.	seconds
Apnea time	AT	Time after end of exhalation where the airflow signal remained within the apnea threshold defined at the start of the analysis.	seconds
Pulmonary resistance	RES	Change in pressure divided by change in flow at the isometric volume locations: $\frac{\Delta p}{\Delta f}$	mmHg/ (liters/ sec)
Pulmonary compliance	Cdyn	Tidal volume divided by the change in pressure between exhale and inhale locations in the breath: $\frac{TV}{\Delta p}$	liters/ mmHg

If text output is being generated, an additional row will be added containing the average values of the measures. Time and count are not output as waveforms in the graph since they can be found in the horizontal axis.

Penh Analysis

Penh Analysis script assumes standard recording methodology for a full body plethysmograph. Positive flow is treated as exhalation and negative flow is treated as inhalation.

Penh Analysis extracts measures from data recorded in a full body plethysmograph. It operates on a single channel of data recorded from an airflow transducer connected to the plethysmograph. The analysis takes a single parameter: the Rt percentage. This percentage is used to locate the plateau, or “pause,” in the airflow signal. The pause begins at the time when the Rt percentage of the exhalation volume has been reached. The Rt percentage may be adjusted by the user and is set to a default of 65%. This analysis will place Inhale Start and Exhale Start events on the airflow signal. If these events are already present when the analysis starts, the user-defined inhale and exhale events will be used. This allows for the analysis to be repeated after manual inspection and correction of inhale and exhale locations and allows for different methods to be used to define the breathing boundaries.

Penh analysis will place Recovery events on the airflow channel at the time positions where the corresponding percentage of the volume has been exhaled. The percentage used for the analysis is displayed in the label of the Recovery events.

For each exhalation period, the Penh Analysis will extract the following:

Name	*Abbrev.*	*Description*	*Units*
Cycle		Index of the exhalation cycle in the data, beginning at 1.	
Time		Starting time of the exhale for the cycle. This is the location of the Exhale Start event.	Seconds
Peak inspiratory flow	PIF	Maximum absolute airflow occurring in the inspiration cycle immediately preceding the exhalation cycle. This measure is recorded as an interval, so its value is always positive.	Airflow channel units
Peak expiratory flow	PEF	Maximum airflow during the exhalation cycle being examined.	Airflow channel units
Exhalation time	Te	Total time elapsed between the start of the exhalation cycle and the end. This is the time interval between the Exhale Start and following Inhale Start events.	Seconds
Relaxation time interval	Rt	Time required for the subject to exhale the specified percentage of the total exhaled air. This is the time interval between the Exhale Start and the subsequent Recovery event.	Seconds
Pause		Numerical factor describing the characteristics of the plateau at the end of the expiration cycle. Computed using the formula: $\frac{Te}{Rt} - 1$	
Enhanced pause	Penh	Pause scaled to be relative to the strength of the inhale and exhale. This helps take breathing variability into account. Computed using the following formula: $\frac{PEF}{PIF} * Pause$	

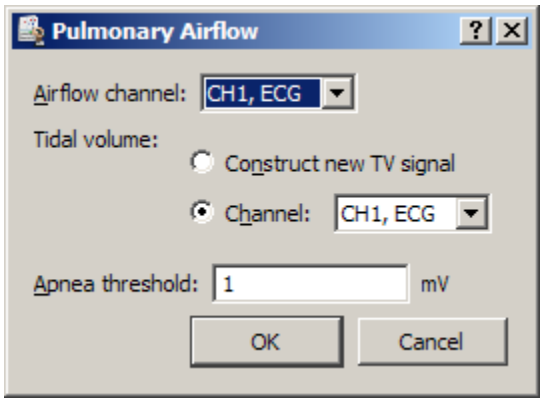
The Penh analysis excludes the following exhale cycles from the analysis:

- Exhale cycles that do not have a preceding inhale (may occur for partial cycles at the start of the data recording).
- Exhale cycles that do not have a corresponding recovery time (often occurs during apnea).

In addition, during periods of apnea, the analysis may produce invalid results, such as zero width recovery times. These results may be excluded from the analysis by either using waveform editing to remove apnea

periods, discarding all events during apnea periods and rerunning the analysis, or deleting the corresponding rows from the Excel output.

Pulmonary Airflow



The Pulmonary Airflow analysis follows the flow conventions of the recommended BIOPAC connections for a TSD107 pneumotach or a TSD117 airflow transducer. Positive flow is assumed to indicate inhalation; negative flow is assumed to indicate exhalation.

The Pulmonary Airflow analysis extracts basic parameters from a calibrated airflow signal, such as would be recorded using a pneumotach or airflow transducer. In addition to inspiration and expiration, Pulmonary Airflow also can be used to examine apnea. Apnea is defined in this analysis as pauses in breathing that occur after an exhalation.

When performing the analysis, an airflow signal f is chosen. An apnea threshold a_f is also entered.

Inhalation is defined to begin at the point where $f > a_f$. Exhalation is defined to begin at the point where $f < -a_f$. Apnea is defined to be the period between exhalation and inhalation where the flow lies within the apnea threshold: $f \in (-a_f, a_f)$. At least two consecutive samples must occur within the apnea threshold for a period of apnea to be defined. This allows for valid transitions from exhalation to inhalation to occur even if one of the samples in the transition happens to fall within the apnea threshold due to sampling.

The Pulmonary Airflow analysis will generate a tidal volume waveform if it is not present in the graph. It also will add Inspire Start and Expire Start events on the airflow signal if they are not present. New Apnea Start events will be defined each time the analysis is performed.

Individual breaths are defined as the period between consecutive Inhale Start events. Airflow units are assumed to be the standard liters/sec.

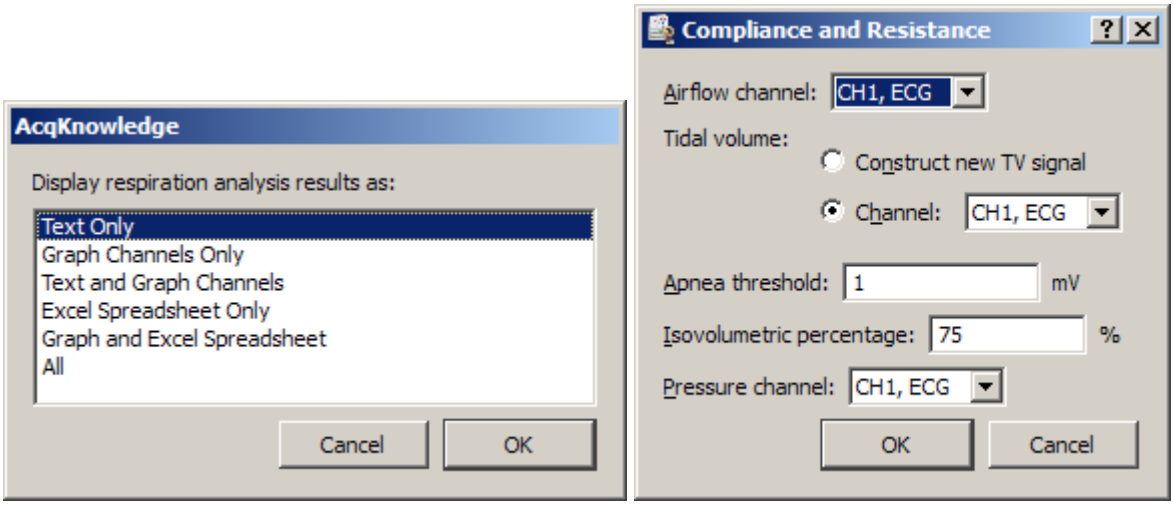
For each breath period, the analysis will extract the following:

Name	*Abbrev*	*Description*	*Units*
Cycle		Index of the breath in the data, beginning at 1.	
Time		Starting time of the inhale of the breath.	seconds
Peak Inspiratory Flow	PIF	Maximum absolute flow occurring during the inhale portion of the breath.	liters / sec
Peak Expiratory Flow	PEF	Maximum absolute flow occurring during the exhale portion of the breath.	liters / sec
Tidal volume	TV	Total volume of air inhaled during the breath.	liters
Minute volume	MV	Volume of air that would be inspired during a minute given the tidal volume and breathing rate of this breath. $TV \quad BPM$	liters / minute

Name	*Abbrev*	*Description*	*Units*
Breaths per minute	BPM	Breathing rate for the breath. $\frac{60}{TT}$	BPM
Inspiration time	IT	Time interval between the start of inhale and the start of exhale in the breath.	seconds
Exhalation time	ET	Time interval between the start of exhale and either: • start of apnea (if apnea present) • start of subsequent breath (if no apnea present)	seconds
Total breath time	TT	Time interval between the start of inhale and the start of inhale of the following breath. This is the sum of the inhalation time, exhalation time, and apnea time.	seconds
Apnea time	AT	Time after end of exhalation where the airflow signal remained within the apnea threshold defined at the start of the analysis.	seconds

If text output is being generated, an additional row will be added containing the average values of the measures. Time and count are not output as waveforms in the graph as they can be found from the horizontal axis.

Preferences



Spectral Subtraction

Spectral subtraction is a denoising technique that operates on data projected into the frequency domain. It is frequently used in speech analysis denoising applications. Spectral subtraction examines a reference noise signal and performs a Fourier transform to get the noise frequency distribution. To denoise a signal, the Fourier transform of the signal is performed. The noise estimate frequency distribution is then subtracted from the source signal. The resulting processed spectrum with the noise frequencies removed is then reconstructed into a time domain signal using the inverse Fourier transform.

Spectral subtraction performs noise removal on the entire channel in a single Fourier transformation, which allows for denoising where the noise is stationary; there is no provision for sliding window spectral subtraction at this time.

The spectral subtraction is performed using a formula with two adjustable parameters. Given a frequency spectrum F_{noise} and a mixed signal F_{mix}, the denoised frequency spectrum is computed using the following formula:

$$F_{denoise} = \left[F_{mix}{}^{\gamma} - \alpha F_{noise}{}^{\gamma} \right]^{\frac{1}{\gamma}}$$

where

Alpha is the “scaling factor” and can be used to adjust the strength of the noise estimate.
Gamma is the “power factor” and can be used to vary how the noise is removed. Gamma = 1 allows for pure subtraction, Gamma = 2 allows for Euclidean distance formulas, and so on.
Any $F_{denoise}$ that is less than zero is discarded and replaced by zero to maintain a valid set of Fourier coefficients for the reconstruction.

When spectral subtraction is being used in practice, the noise signal may not always match the length of the signals to be denoised. To define the subtraction formula, the spectrum of the noise must have the same number of points as the spectrum to be denoised. If there is a length mismatch, the noise spectrum is resampled automatically to match the length of the spectrum to be denoised. Cubic spline interpolation is used during the resampling to provide a better estimate of the overall noise spectrum.

Stim-Response

The Stim-Response analysis package can aid in analysis of stimulus-response studies. It allows for measurements to be extracted in tabular format for multiple stimulus classes. Stim-response configuration enhancements in Acq*Knowledge* 4.1 add the following functionality:

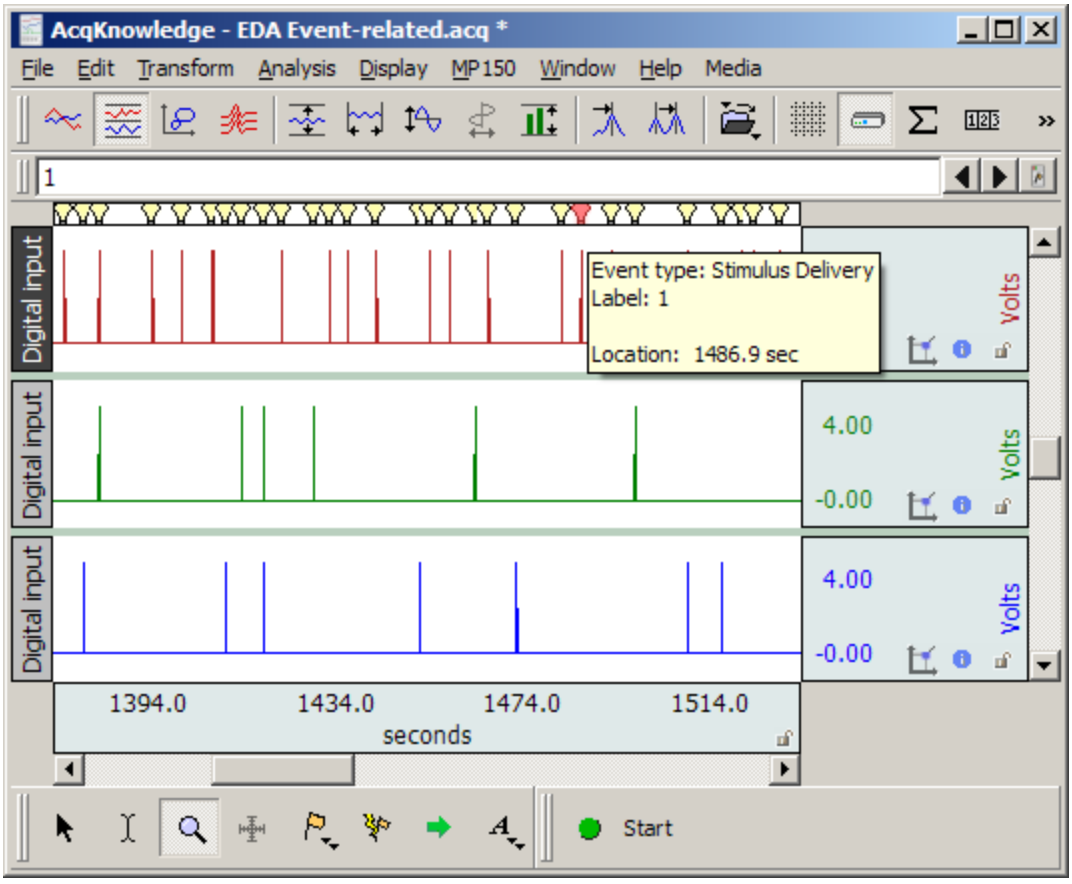
- Measurement configuration is preserved across launches of the application
- Measurement presets may be accessed directly from the specialized analysis routine
- Additional checking for invalid channels and measurement expressions that cannot be applied to the source data

The Event-related EDA Analysis routine uses the stimulus events to categorize specific and non specific responses. Responses are matched to the appropriate stimulus events using a user defined time window. See the Electrodermal Activity EDA Event-related analysis section for further information.

Note: Excel spreadsheet generation is only supported with Acq*Knowledge* 3.9.1 and higher.

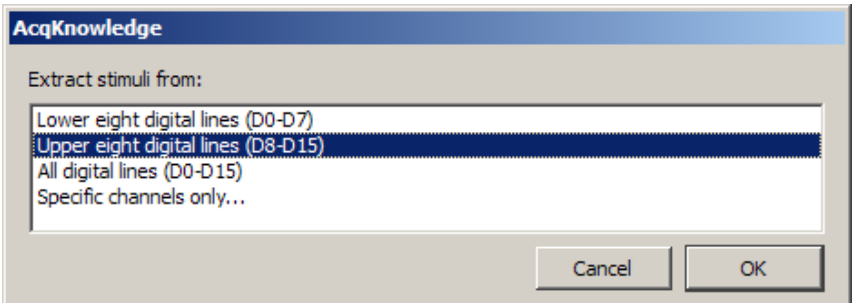
Digital Input to Stim Events

Digital Input to stim events identifies and labels stimulus events corresponding to any combination of digital inputs. A lightbulb icon is placed in the global event marker bar, the event is labeled with the stimulus event type, and the mouse-over tag includes the event time. All event information is accessible and exportable from the Event Palette.

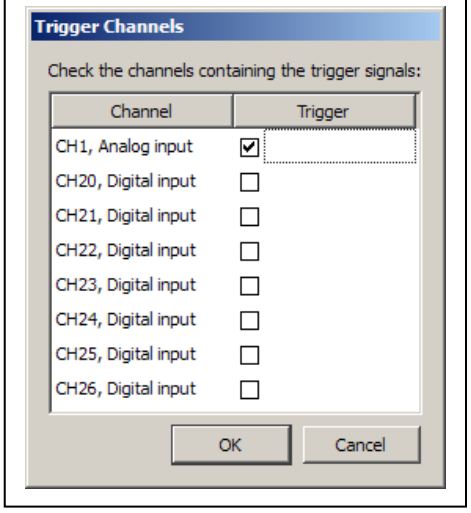


The Digital input to stim events function works with TTL trigger information coming from applications such as E-Prime®, SuperLab®, DirectRT®, MediaLab®, Inquisit®, and Presentation. It converts TTL data acquired on the digital channels of an MP device into stimulus events. The system also works with analog and calculation channel signals coming from switch transducers. Unlike TTL signals, a voltage threshold level is used to determine the transition from low to high.

This analysis option converts TTL or switch data acquired on an MP device into stimulus events. Stimulus delivery events are defined in the graph for each low to high transition of the digital data, the indications of stimulus delivery. The digital channels are interpreted as a binary number. Each stimulus event placed into the graph has the corresponding number included with its label. This allows further analysis to distinguish between different types of stimulus events by using the Cycle Detector's label matching capability.

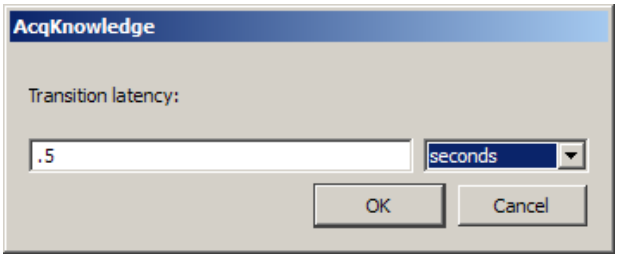


"Specific channels only" dialog

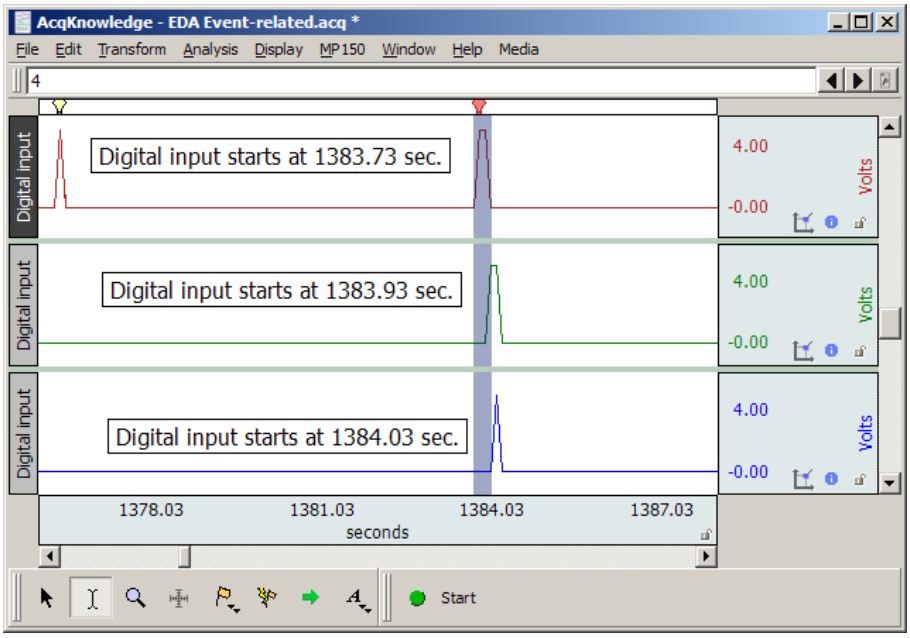


Digital line decoding can be two byte (using all 16 digital lines) or single byte (on either the low eight or high eight digital lines). Big endian bit and byte ordering are used, with digital line 0 representing the least significant bit.

When the stimulus labels are constructed, all numbers are zero-prefixed. All stimulus events will have the same number of base-10 digits with leading zeros, regardless of magnitude. This provides each stimulus event type with a unique label that can be used with the Cycle Detector (which uses substring matching).



Some systems that trigger digital lines such as parallel ports may not be able to do so instantaneously; they may require a time window before the transition from one state to another is fully complete. A "**transition latency**" time window can be given to the analysis, specified in microseconds, milliseconds, seconds, minutes, hours, or samples. If non zero, any transitions that are separated by less than this latency are treated as a single transition and only one stimulus event is inserted. The decoded value used for the transition is the maximum value observed during the transition latency window. In the following example graph, the three digital TTL inputs correspond to one event., as marked by the red icon in the global marker bar. A transition latency of .5 seconds will consider all three as part of the same event since the transitions occur within .5 seconds of each other.



Stim-Response Analysis

Stim-Response Analysis allows for extraction of measurements within fixed width intervals occurring at Stimulus Delivery events. Stimulus events must be present to perform this analysis. The Stimulus Delivery events may be defined either manually, with the Cycle Detector, or using the Digital Input to Stim Events analysis option. The information that can be extracted includes the majority of the measurements available from the graph window measurement toolbar, matching the Epoch Analysis options.

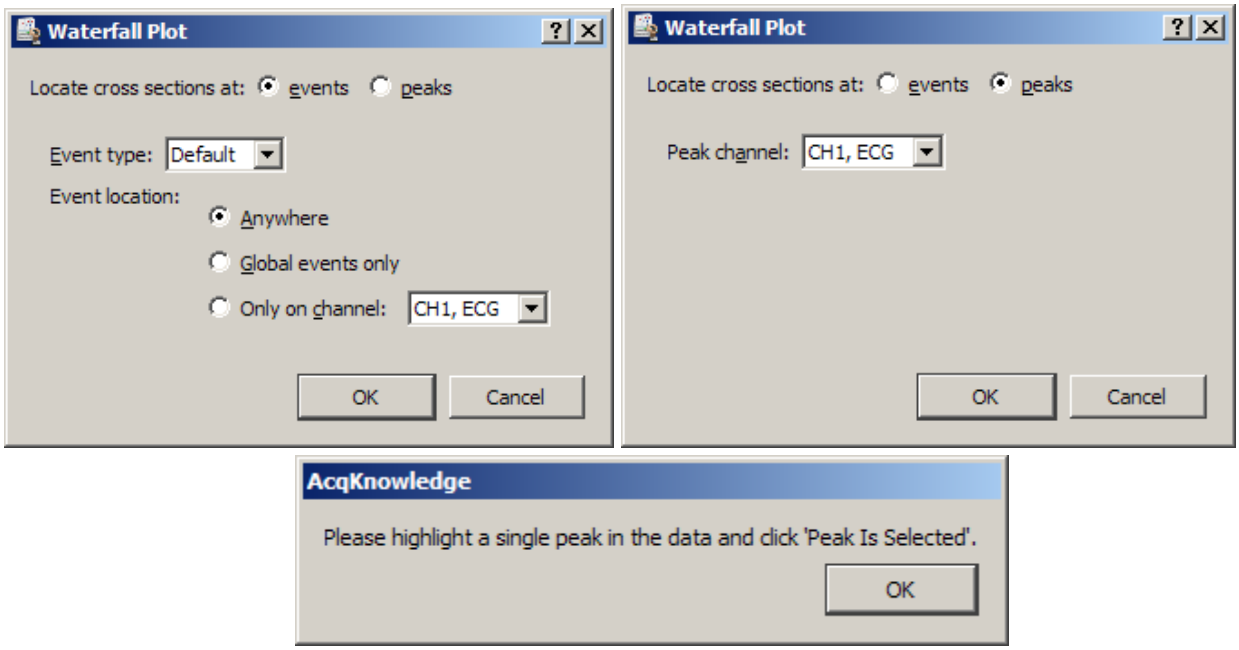
Unlike Epoch Analysis, the Stim-Response Analysis splits the analysis based upon the event labels. Stimulus Delivery events with different labels are interpreted as different stimulus types. Analysis results for each individual stimulus type are summarized in separate tables. Each independent text table has its own average of the measurements over that stimulus type.

Additional options are available for positioning the fixed width interval where measurements should be made:

- At each stimulus event – The measurement interval is aligned so the start of the measurement matches the time of the Stimulus Delivery event.
- At fixed interval offset before or after stimulus – The measurement interval begins a fixed amount of time either before (for pre-stim studies) or after the time of the Stimulus Delivery event. This allows measurements to be made at a time relative to each stimulus onset and may be useful for measurements focusing on a specific time range (e.g. P300).
- At matching response event – This option assumes that a second set of Response events have been defined for each stimulus either manually or using the Cycle Detector. Each Stimulus Delivery event is paired with the closest Response event occurring after it. The fixed width measurement interval is aligned so the start of the measurement window is the time of this matching Response event.
    - To use the “at matching response event” window positioning option, Response events must be defined in the graph. Response events are in the “Stimulus/Response” event submenu. These events are *not* defined automatically.
        - Response events can be inserted manually into the graph using the Event tool.
        - Response events can be inserted using the event output of a data-driven Cycle Detector analysis.

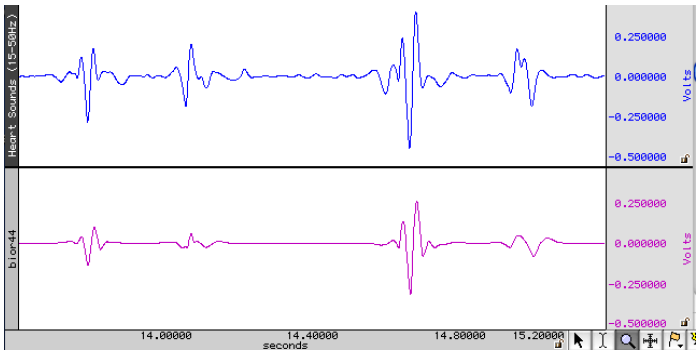
Note If EDA/SCR signals are being analyzed in response to stimulus delivery, also examine the “Event-related EDA Analysis” transformation located under Analysis > Electrodermal Activity.

Waterfall Plot



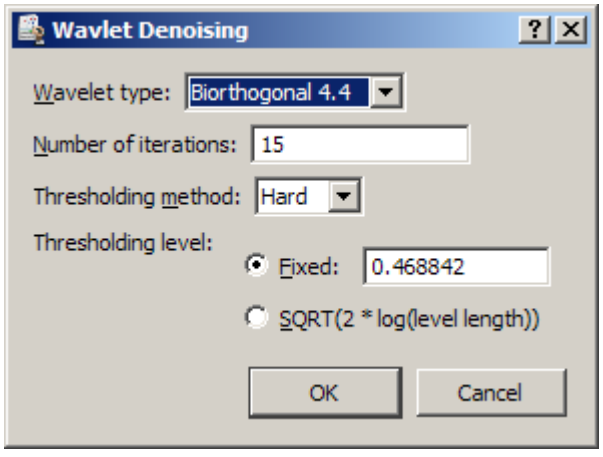
Assists in configuring the Peak Detector for 3D surface generation. These surfaces showing cycle-by-cycle data of the graph are commonly known as *waterfall plots*. Cycles are located in the graph using the same sequence of steps as the Ensemble Average transformation script. Instead of generating the averaged graph, however, a number of 3D surfaces are generated. One surface is generated for each channel that is selected by the user.

Wavelet Denoising



Sample output

Wavelet Denoising applied to heart sounds data may help clarify S_1 and S_2, as shown:



Wavelet Denoising uses the forward and reverse wavelet transformations to project source data into the wavelet domain, modify the wavelet coefficients (called “shrinking” the coefficients), and then reconstruct the data from the modified coefficients. Wavelet Denoising allows for noise to be removed from a signal while minimizing effects on portions of the signal that strongly adhere to a wavelet’s shape.

To perform wavelet denoising:

1. Choose the wavelet type to use for the denoising (Biorthogonal 4.4, Symlet 4, Coiflet 6, or Daubechies 8).

   Certain signals may work best with different wavelet types.
2. Enter the number of iterations to use in the wavelet decomposition.

   Different numbers of iterations will have different effects on the results.
3. Choose which type of thresholding should be used to shrink the wavelet coefficients.
   - Hard thresholding replaces coefficients below the threshold with zero while leaving all other coefficients unmodified.
   - Soft thresholding zeroes out coefficients below the threshold and subtracts the threshold for coefficients that are above it.

   Soft thresholding may be useful for reducing edge effects, but hard thresholding will affect amplitudes less.
4. Choose the threshold level to use for shrinkage.
   - Fixed threshold for all levels. If you choose a fixed threshold, an additional window will appear into which you can type your threshold.
   - Adaptive threshold level based on the number of coefficients in the DWT iteration (a VIS shrink procedure).

ECG Analysis Algorithm References

Acq*Knowledge* 3.9 software implements the open source ecgpuwave ECG boundary location software and the open source OSEA QRS detector and beat classification library for ECG analysis.

Automated ECG Waveform Boundary Location

➲ ecgpuwave ECG boundary location software

Acq*Knowledge* 3.9 software incorporates the ecgpuwave ECG boundary location software. Ecgpuwave is an implementation of a waveform boundary detection algorithm primarily developed by Pablo Laguna at the University of Zaragoza in Spain. This algorithm incorporates a variant of the Tompkins QRS detector, but contains additional rules that allow it to automatically extract the following characteristics of an ECG signal on a cycle by cycle basis: onset of P, P peak, end of P, onset of QRS, peak of QRS, end of QRS, onset of T, peak of T, and end of T.

The algorithm is tuned to human ECGs through comparison with manual classification. Particularly, it seems to be within the standard deviation of human examiners for the onset and end of T waves, a particularly difficult feature to extract from an ECG complex. It also has the ability to take multiple ECG leads into account to reduce errors and misclassifications and appears to function for one to twelve lead ECGs. The algorithm is well documented in a number of papers. This algorithm development was sponsored by several government agencies including CICYT in Spain and the NIH.

The ecgpuwave tool is distributed from the PhysioNet NIH servers (http://www.physionet.org). This is a tool written in Fortran that will read WFDB formatted files. It will then output a series of annotations in WFDB format indicating the locations of the various ECG complexes within each cycle. It also depends on another tool, sortann (available from PhysioNet), to perform post-processing. This software reads and writes PhysioBank formatted files. This tool is only available on OS X.
Acq*Knowledge* can automate the process of running ecgpuwave on source data and import its output back into Acq*Knowledge*. To run ecgpuwave on an ECG signal from within Acq*Knowledge*, first make the ECG channel the active channel and then choose Transform > Specialized Analysis > Locate ECG Complex Boundaries. Acq*Knowledge* will execute ecgpuwave on that signal and read in its waveform boundary location output, placing events on the channel. You will only be able to see this output if you have events shown.
Alternatively, you can save your file to PhysioBank format, run ecgpuwave manually from a Terminal, save the annotations to “atruth” and then reopen that PhysioBank file to see the ecgpuwave results; this is the same process that Acq*Knowledge* performs.

Source code for ecgpuwave detector is released under a GPL license and can be found on the Acq*Knowledge* CD.

OSEA QRS Detector

➲ OSEA QRS detector and beat classification library

Acq*Knowledge* 3.9 software incorporates the open source OSEA QRS detector and beat classification library.

The OSEA library is a set of routines provided by EP Limited (http://www.eplimited.com). This C++ based software library provides robust QRS complex detection and rudimentary beat classification. This library is well documented and tested. The QRS detector uses a standard Tompkins-based filtering and derivative detection algorithm and has been in development for about 15 years; the beat classifier has been developed for about a year or two. This algorithm development is sponsored by the NIH.

This algorithm is fairly robust against arrhythmias, baseline drifts, discontinuities, and other artifacts in the ECG signal. It achieves a 90% success rate on identifying QRS complexes on sample arrhythmia databases. The algorithm is tuned to human ECGs.

The QRS detector is optimized for 200 Hz sampled data. If the sampling rate is lower or higher, data will be internally resampled to 200 Hz before processing. The sampling rate difference may result in slightly different placement of beat events for different sampling rates.

QRS detection can be performed by selecting the desired channel of ECG data and choosing Transform > Specialized Analysis > Detect and classify beats. Acq*Knowledge* will execute the OSEA beat detector on the source data and output a sequence of events on that channel of ECG data. You will only be able to see this output if you have events shown.

Source code for the QRS detector is released under an LGPL license and can be found on the Acq*Knowledge* CD.

Open Source Licensing

The ecgpuwave and OSEA algorithms are available as open source, which means that their source code is publicly available. The source code can only be used, however, under conditions of their licenses.

- ecgpuwave is under the GPL license
- OSEA is under the LGPL license

For the full text of both licenses, visit the Free Software Foundation (http://www.fsf.org).