UAMS
UNIVERSITY OF ARKANSAS FOR MEDICAL SCIENCES

Department of Neurobiology and Developmental Sciences
Center for Translational Neuroscience
College of Medicine

Neuronal Signals - NBDS 5161
Session 7: Detecting Events

Abdallah HAYAR

Lectures can be downloaded from
http://hayar.net/NBDS5161

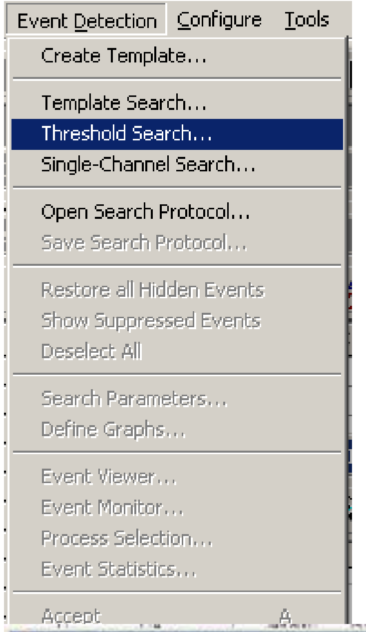
Updated Tentative Schedule for Neuronal Signals (NBDS 5161)
One Credit–Hour, Summer 2010
Location: Biomedical Research Building II, 6th floor, conference room,
Time: 9:00 -10:20 am

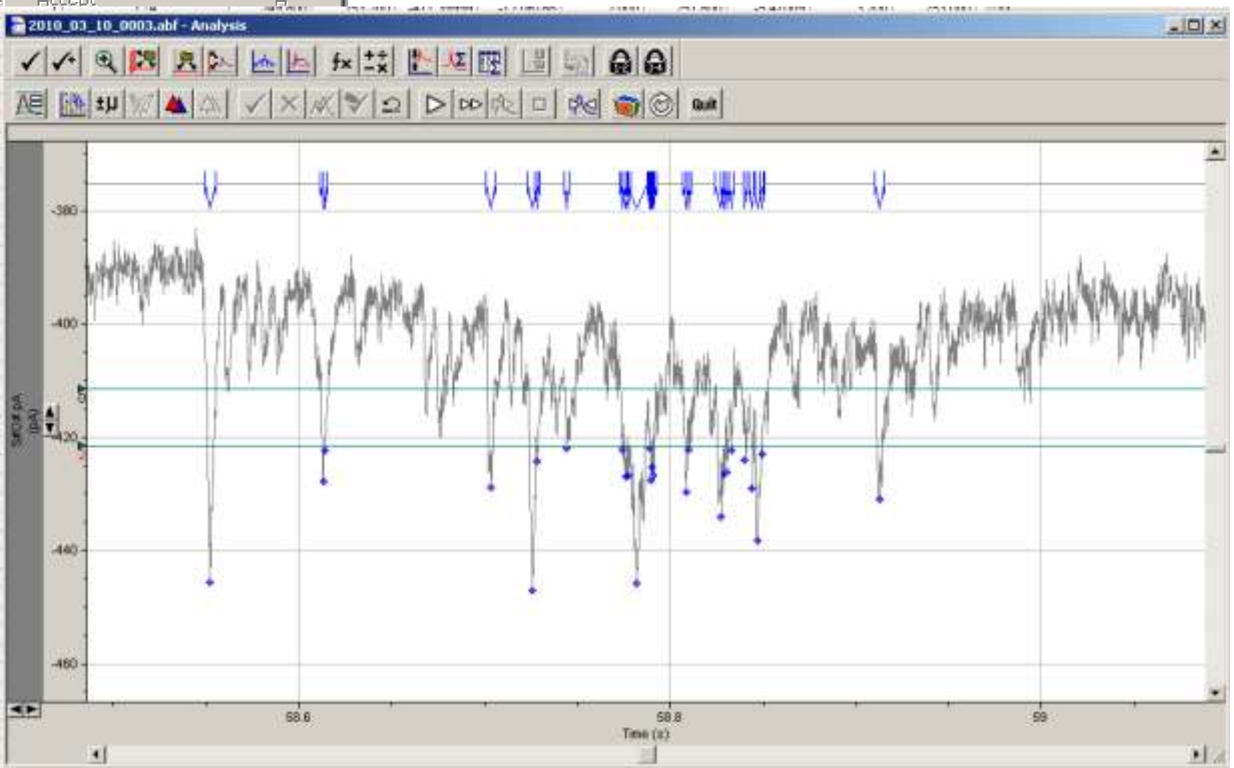
Session	Day	Date	Topic	Instructor
1	Tue	6/1	Design of an electrophysiology setup	Hayar
2	Thu	6/3	Neural population recordings	Hayar
3	Thu	6/10	Single cell recordings	Hayar
4	Fri	6/11	Analyzing synaptic activity	Hayar
5	Mon	6/14	Data acquisition and analysis	Hayar
6	Wed	6/16	Analyzing and plotting data using OriginLab	Hayar
7	Fri	6/18	Detecting electrophysiological events	Hayar
8	Mon	6/21	Writing algorithms in OriginLab®	Hayar
9	Wed	6/23	Imaging neuronal activity	Hayar
10	Fri	6/25	Laboratory demonstration of an electrophysiology and imaging experiment	Hayar
11	Fri	7/9	Article presentation I: Electrophysiology	Hayar
12	Mon	7/12	Article presentation II: Imaging	Hayar
13	Wed	7/14	Exam and students' survey about the course	Hayar

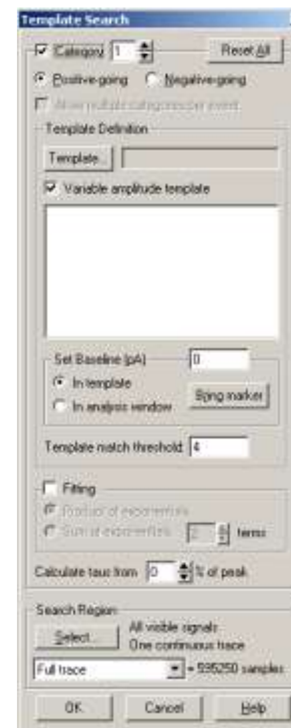
Student List

	Name	E-mail	Regular/Auditor	Department	Position
1	Simon, Christen	CSimon@uams.edu	Regular (form signed)	Neurobiology & Developmental Sciences	Graduate Neurobiology – Mentor: Dr. Garcia-Rill
2	Kezunovic, Nebojsa	NKezunovic@uams.edu	Regular (form signed)	Neurobiology & Developmental Sciences	Graduate Neurobiology – Mentor: Dr. Garcia-Rill
3	Hyde, James R	JRHyde@uams.edu	Regular (form signed)	Neurobiology & Developmental Sciences	Graduate Neurobiology – Mentor: Dr. Garcia-Rill
4	Yadlapalli, Krishnapraveen	KYadlapalli@uams.edu	Regular (form signed)	Pediatrics	Research Technologist – Mentor: Dr. Alchaer
5	Pathan, Asif	APATHAN@uams.edu	Regular (form signed)	Pharmacology & Toxicology	Graduate Pharmacology – Mentor: Dr. Rusch
6	Kharade, Sujay	SKHARADE@uams.edu	Regular (form signed)	Pharmacology & Toxicology	Graduate Pharmacology – 4th year - Mentor: Dr. Rusch
7	Howell, Matthew	MHOWELL2@uams.edu	Regular (form signed)	Pharmacology & Toxicology	Graduate Interdisciplinary Toxicology - 3rd year - Mentor: Dr. Gottschall
8	Beck, Paige B	PBBeck@uams.edu	Regular (form signed)	College of Medicine	Medical Student – 2nd Year - Mentor: Dr. Garcia-Rill
9	Atcherson, Samuel R	SRAtcherson@uams.edu	Auditor (form signed)	Audiology & Speech Pathology	Assistant Professor
10	Detweiler, Neil D	NDDETWEILER@uams.edu	Auditor (form not signed)	Pharmacology & Toxicology	Graduate Pharmacology – 1st year
11	Thakali, Keshari M	KMThakali@uams.edu	Unofficial auditor	Pharmacology & Toxicology	Postdoctoral Fellow – Mentor: Dr. Rusch
12	Boursoulian, Feras	FBoursoulian@uams.edu	Unofficial auditor	Neurobiology & Developmental Sciences	Postdoctoral Fellow – Mentor: Dr. Hayar
13	Steele, James S	JSSTEELE@uams.edu	Unofficial auditor	College of Medicine	Medical Student – 1st Year – Mentor: Dr. Hayar
14	Smith, Kristen M	KMSmith2@uams.edu	Unofficial auditor	Neurobiology & Developmental Sciences	Research Technologist – Mentor: Dr. Garcia-Rill
15	Gruenwald, Konstantin	kjoachimg@gmail.com	Unofficial auditor	Neurobiology & Developmental Sciences	High school Student – Mentor: Dr. Hayar
16	Rhee, Sung	RheeSung@uams.edu	Unofficial auditor	Pharmacology & Toxicology	Assistant Professor
17	Light, Kim E	LightKimE@uams.edu	Unofficial auditor	Pharmaceutical Sciences	Professor

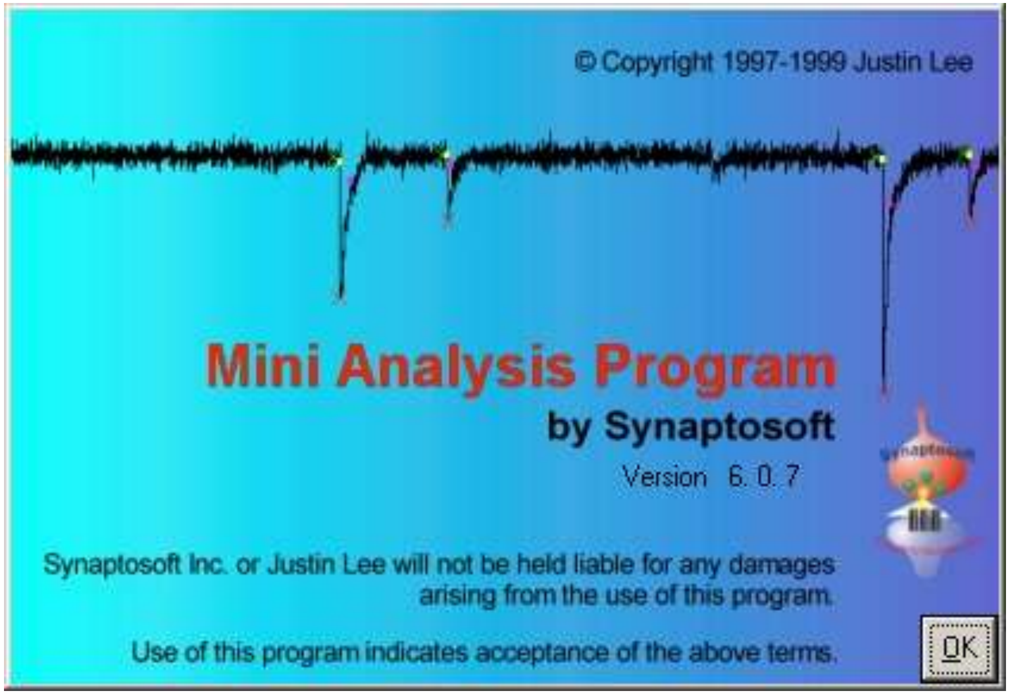
Event detection in Clampfit

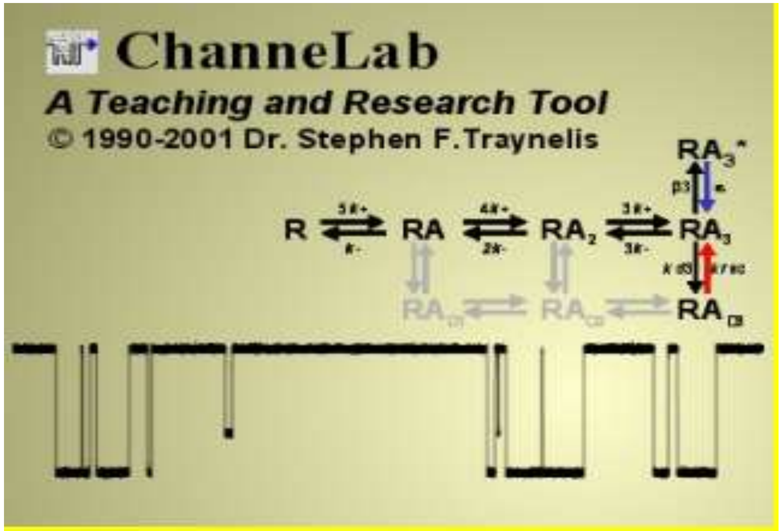






Event Detection





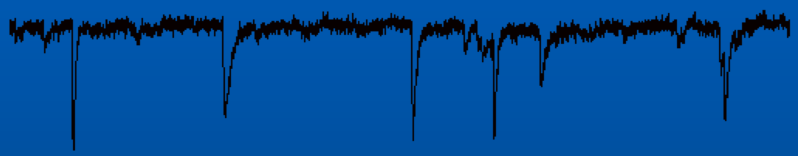
Synaptosft, Inc.
3098 Anderson Place, Decatur, GA 30033, USA
Tel: 770-939-4366 Fax: 770-939-9478
http://www.synaptosoft.com/

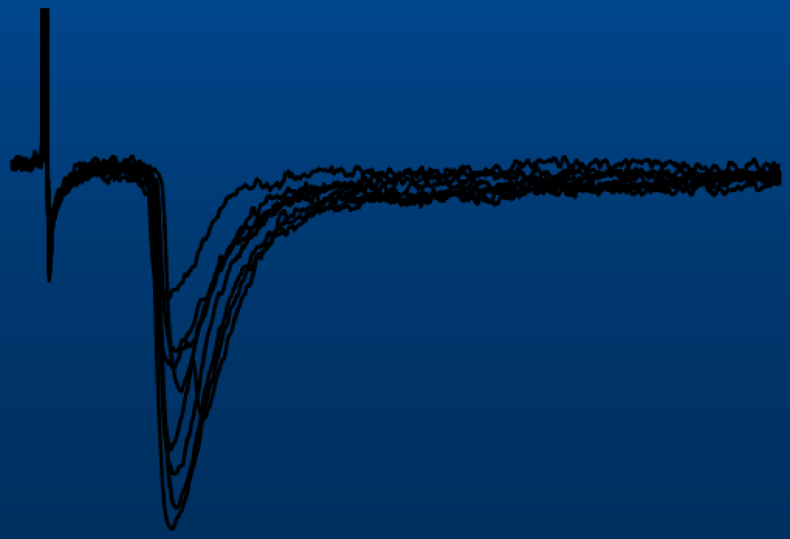
Synaptosoft Inc.

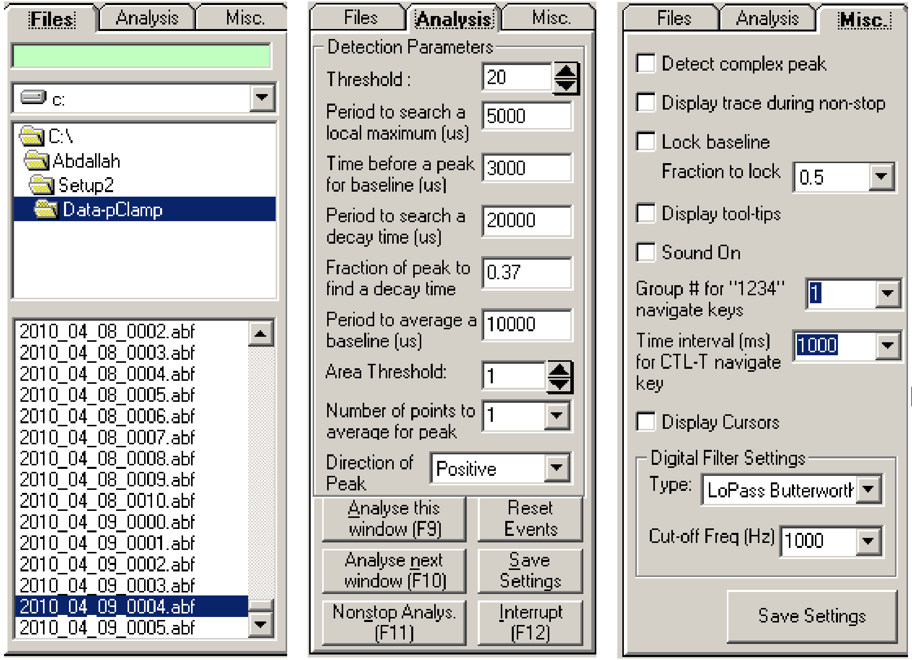
Mini Analysis Program v.6 (Full) Electrophysiological data analysis For Windows 95/98/2000/NT/ME/XP	**$250.00**
Mini Analysis Program v.6 **(Lab Site License, up to 10 copies)**	**$1000.00**

Spontaneous Activities vs. Evoked Responses

- Spontaneous activities are cell-driven activities. Usually recorded as "gap-free" or continuous records.
- Evoked responses are man-driven responses. Usually recorded as episodic records.

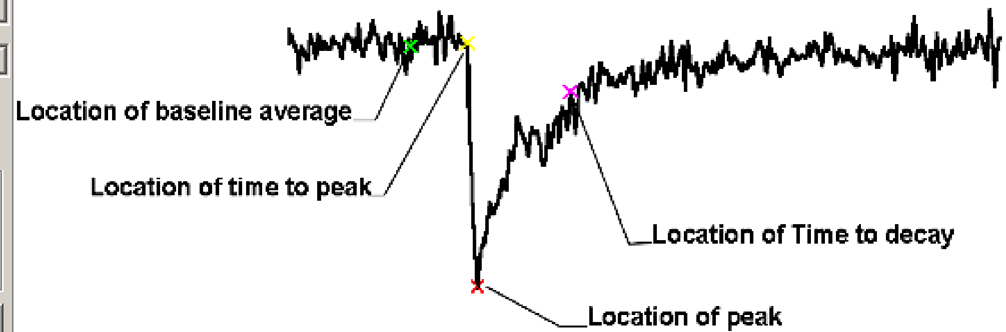






http://www.synaptosoft.com/MiniAnalysis/Tutorial

Single peak



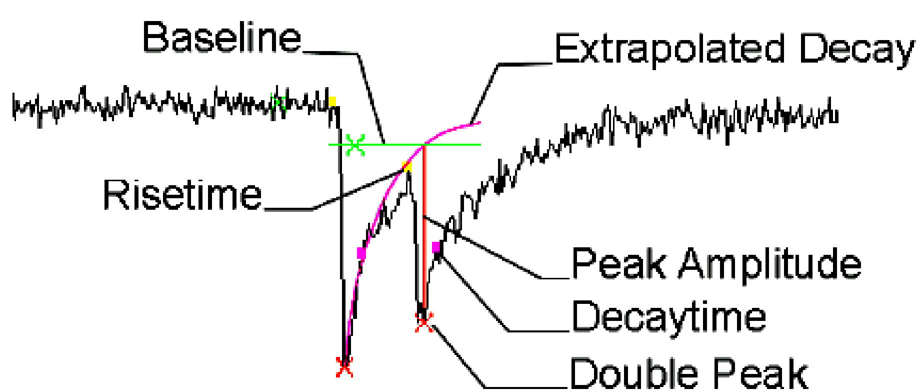
Data Conversion: by ABF Utility

Various file formats are converted
-Binary files (Pulse dat, Igor wave, Tida, Chart, etc.)
-ASCII (tab delimited text, Ca^{2+} imaging)

Unifying file format – ABF
-File header: 2048 bytes
-Data block: 16 bit integer (2 byte) array.

Convert pclamp10 to pclamp9 so that it can be read by MiniAnalysis Program: File-Save as- Type ABF 1.8 (integer)

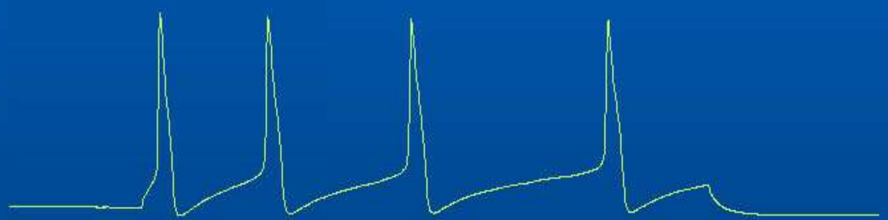
Double peak



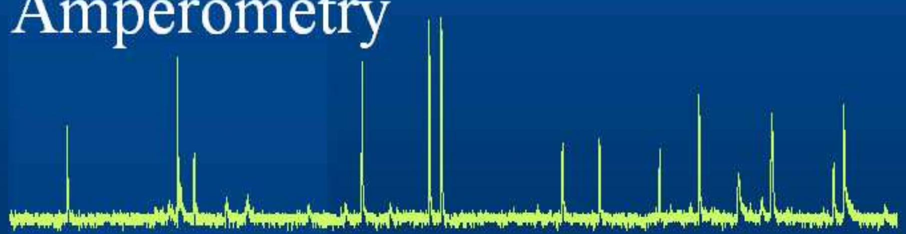
Peak Detection I : Types of Peaks

Post-synaptic currents

Action Potentials



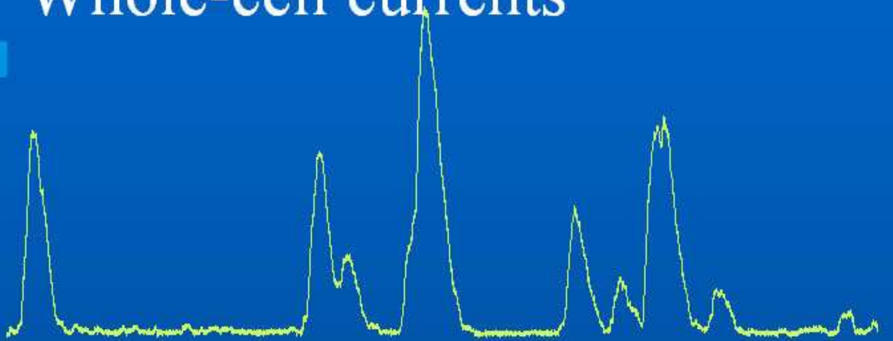
Amperometry



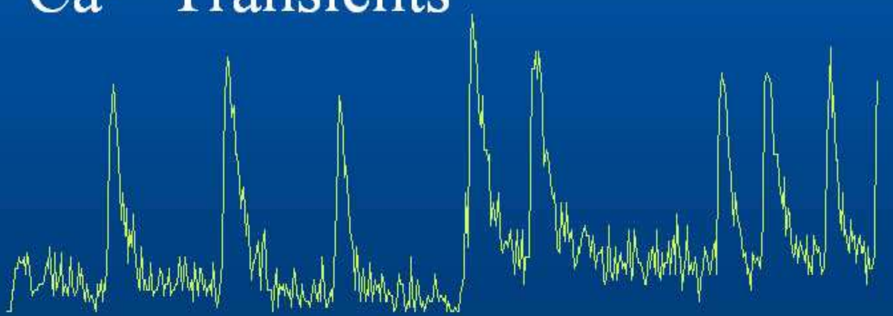
Universal detection algorithm requires...

Sensitivity and Flexibility.

Whole-cell currents



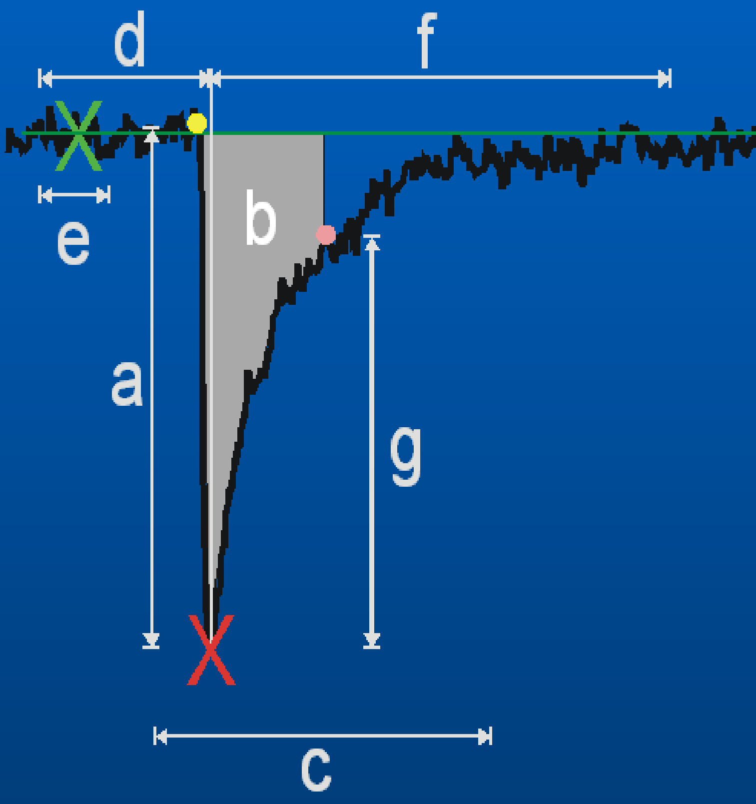
Ca^{2+} Transients



ECG

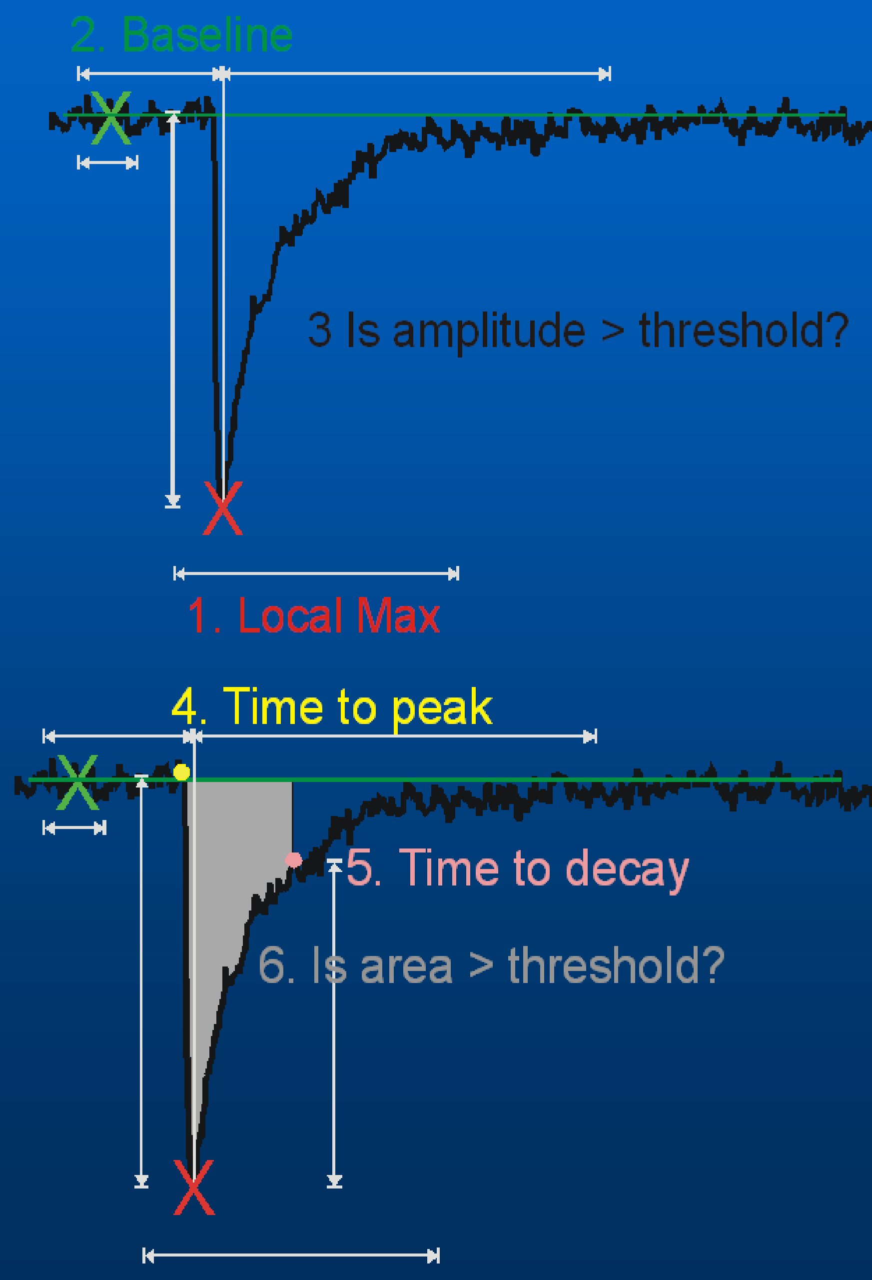
Detection Parameters

- Amplitude threshold (a)
- Area threshold (b)
- Peak direction
- Number points to average peak
- Period to search local maximum (c)
- Time before peak for baseline (d)
- Period to average baseline (e)
- Period to search decay time (f)
- Fraction to find decay (g)



Sequence of Peak Detection

1. Finds a local maximum
2. Finds a baseline
3. Amplitude is compared to the threshold
4. Time to peak is calculated
5. Time to decay is calculated
6. Area is calculated and compared to the area threshold.



Baseline fluctuations are overcome by finding the peak first and then baseline.

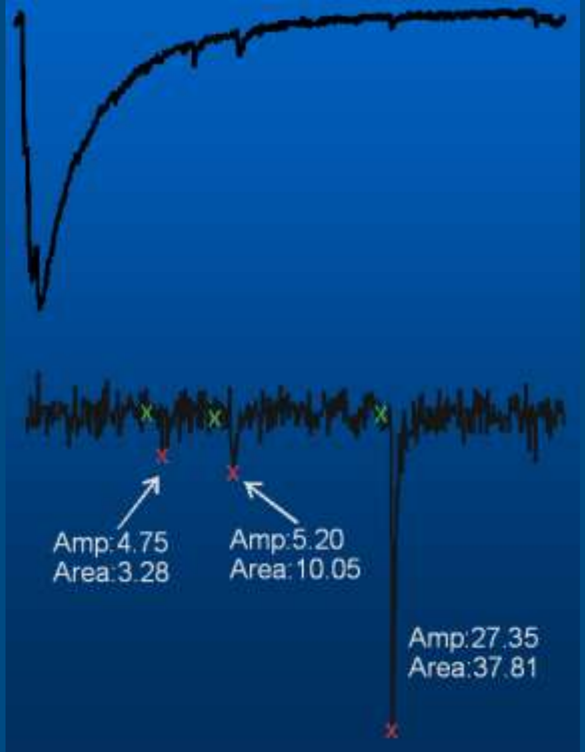
Real peaks are differentiated from noise-peaks by the area and amplitude thresholds.

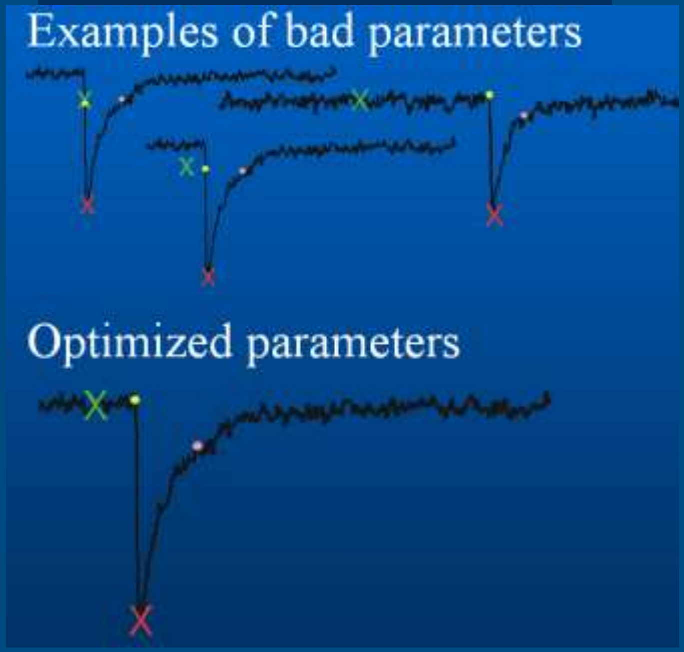
Set of time parameters allow detection of peaks, regardless of their shape.

- First use mouse- click to detect
- Examine the location of the X’s and dots. Adjust time related parameters.
- Examine the amplitude and area. Adjust thresholds.
- Non-stop analysis
- Examine detected peaks using shortcut keys

If there are multiple peaks detected for one EPSCs, Increase the period to search for a local maximum. This usually happens when you have broad EPSCs. Adjust this parameter to be little less than the duration of an event. The disadvantage is that multiple superimposed peaks will not be detected. Try activating multiple peaks analysis

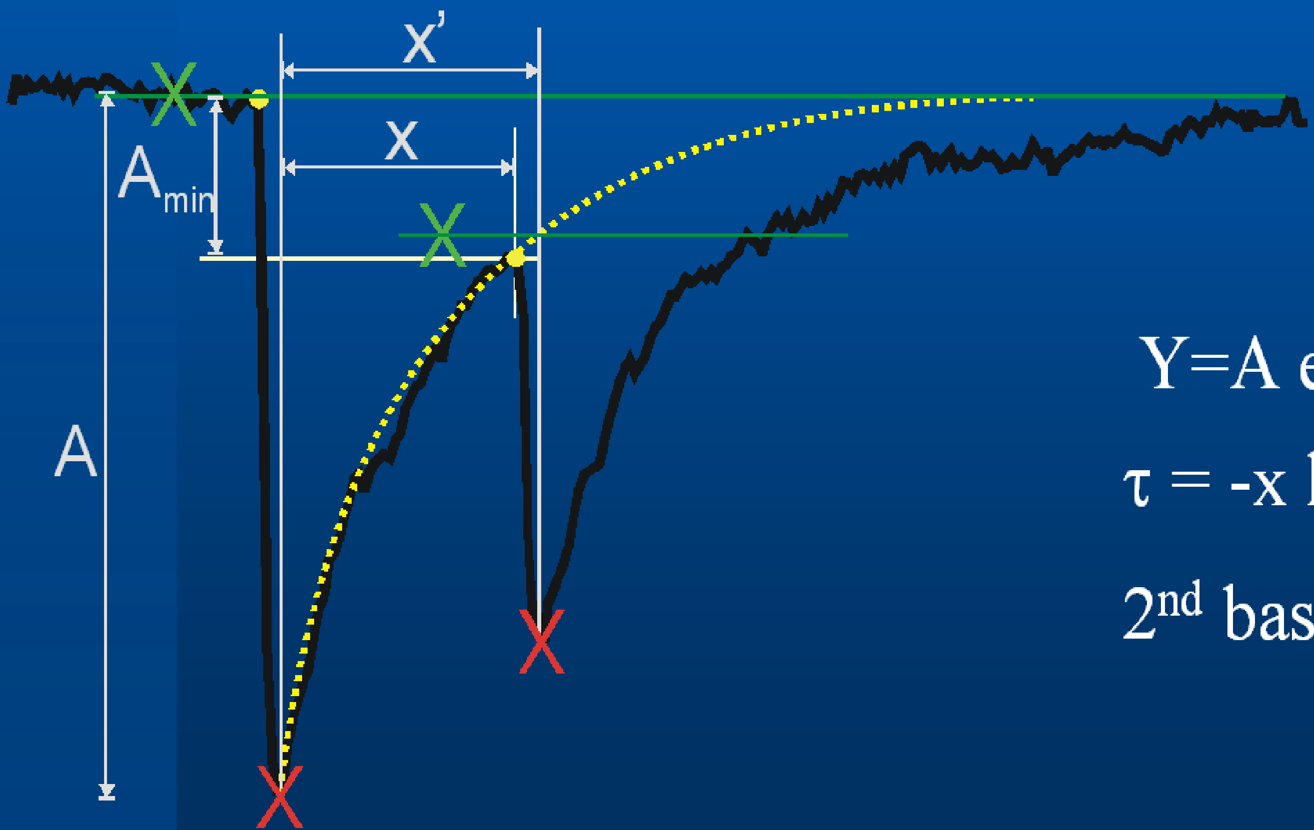
If event risetime is slow, decrease the time before a peak for a baseline and decrease the period to average for a baseline.





Peak Detection III : Multiple Peaks

- **Adjustment of baseline using single exponential decay extrapolation.**

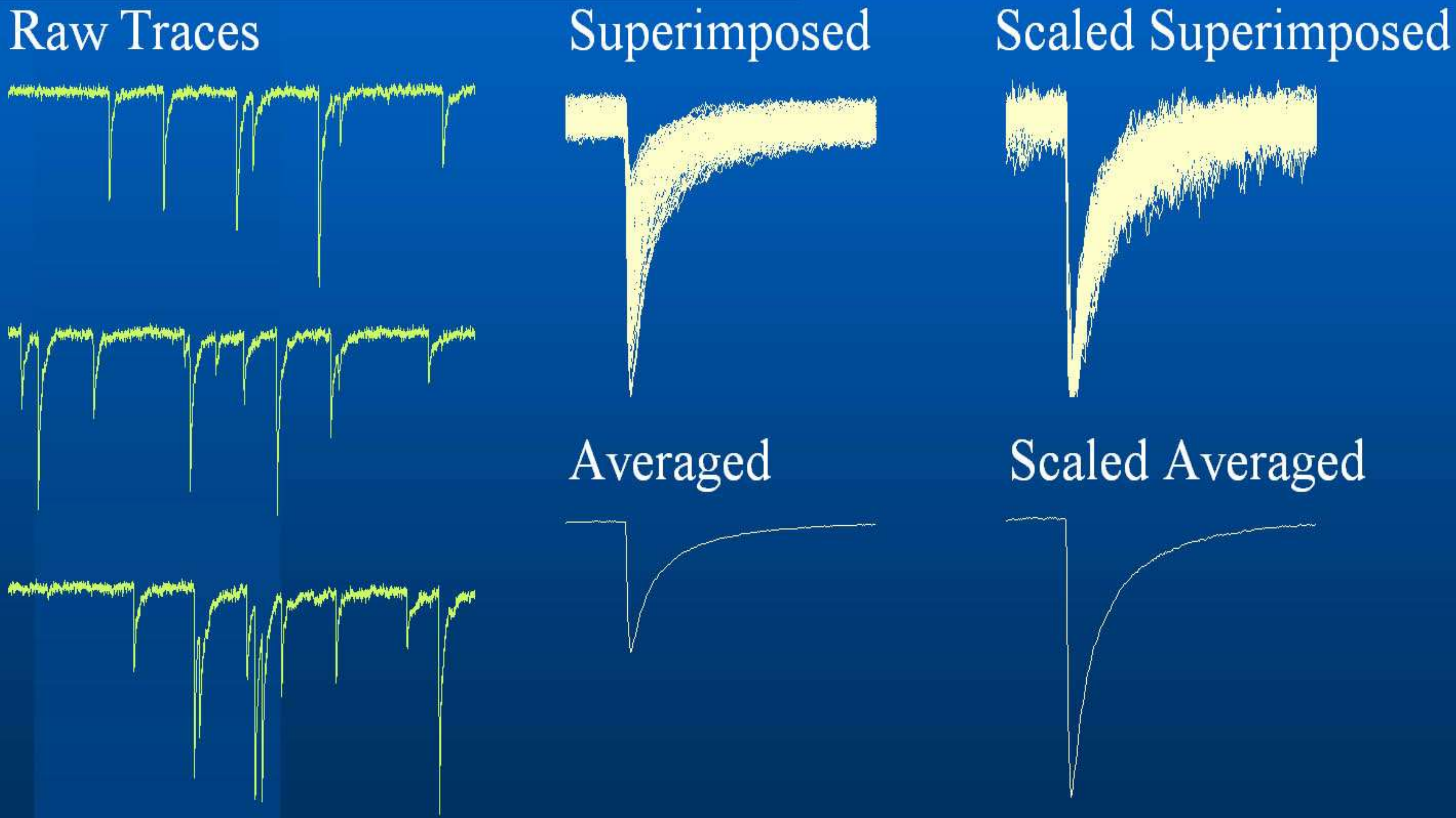


$$Y = A\, e^{(-x/\tau)}$$

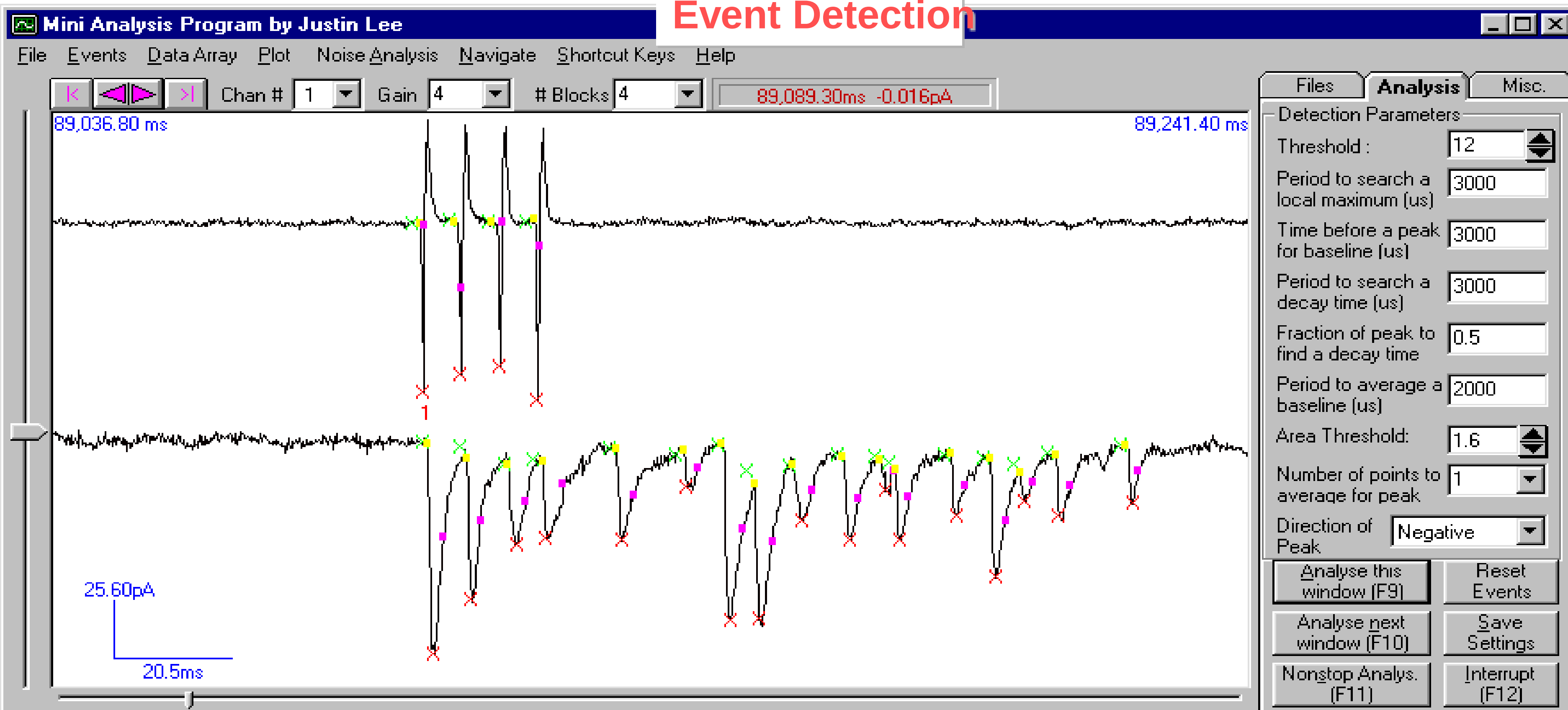
$$\tau = -x \ln(A_{min} / A)$$

$$2^{nd}\ \text{baseline} = A\, e^{(-x'/\tau)}$$

Display of Grouped Traces



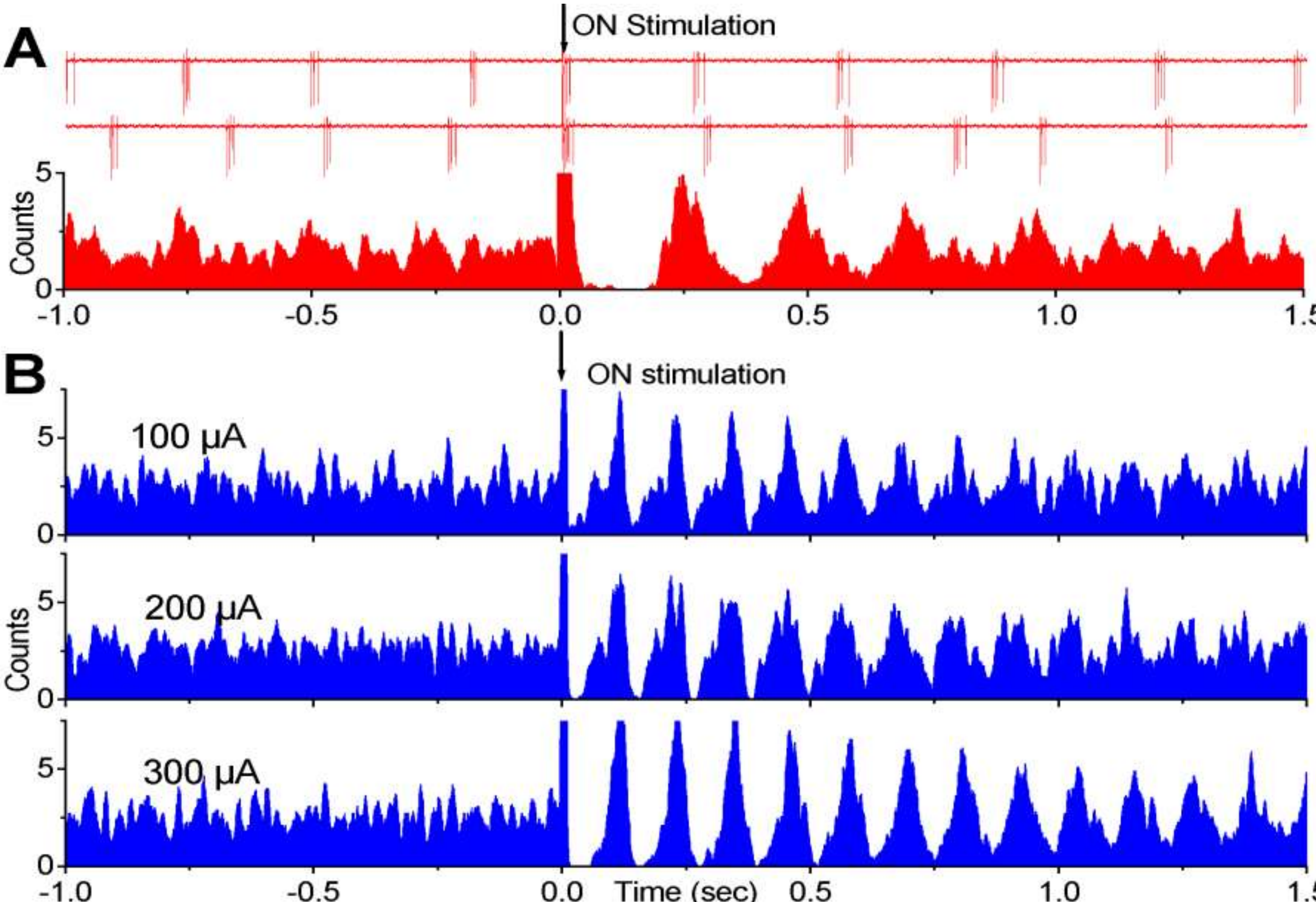
Event Detection



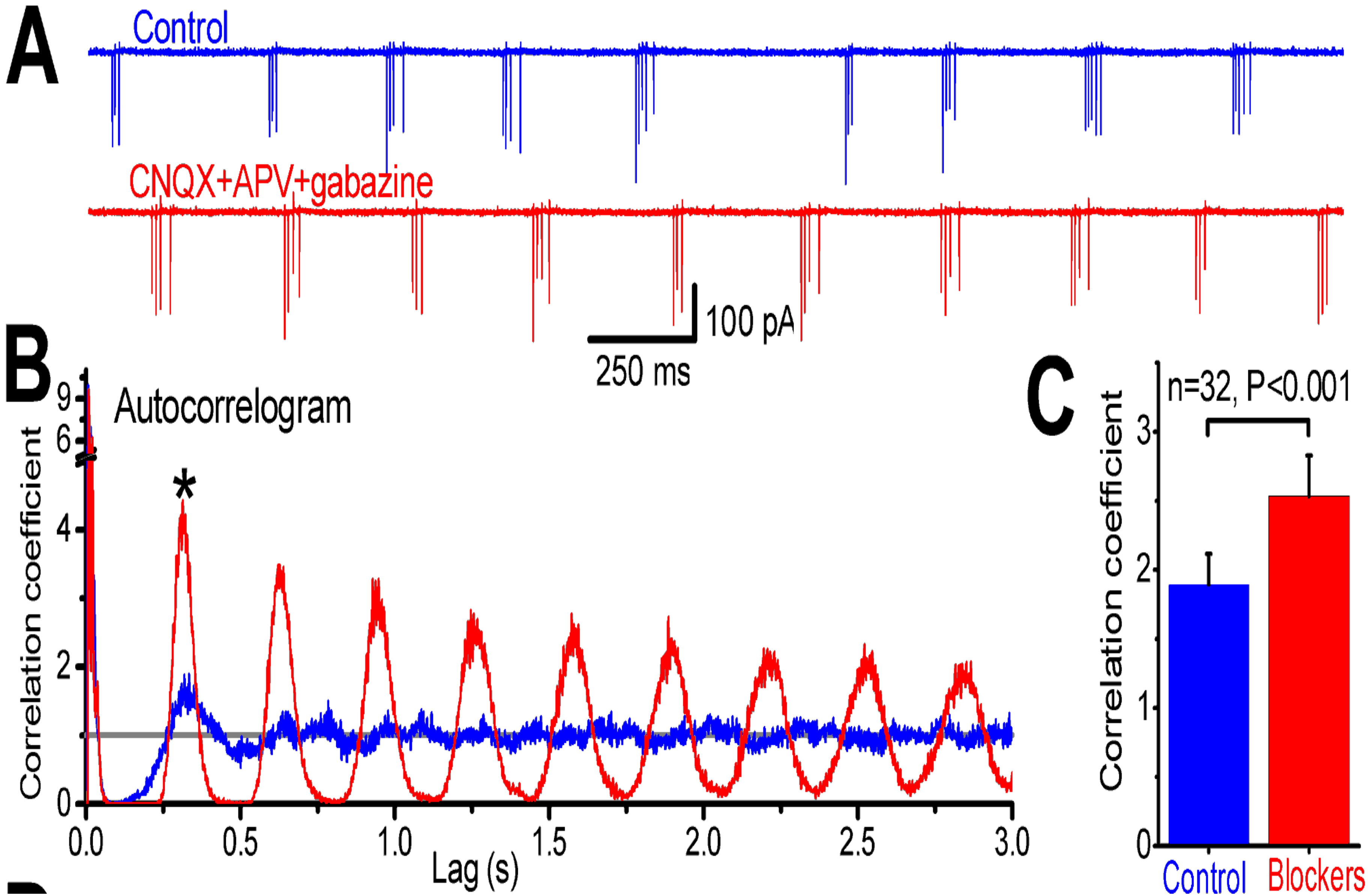
21	Time (ms)	Amplitude	Rise (ms)	Decay (ms)	Area	Baseline	Noise	Group	Channel	10-90Rise	HalfWidth	Rise50	Peak Dir	Burst#	BurstE#
1	89,100.20	100.33	0.60	0.20	33.94	-0.67	1.92	1	0	0.35	0.27	0.20	-1	0	0
2	89,106.60	91.62	1.20	0.20	32.18	1.13	1.13	0	0	0.31	0.31	0.20	-1	0	0
3	89,113.40	85.92	1.40	0.20	33.54	0.42	-0.92	0	0	0.44	0.30	0.20	-1	0	0
4	89,119.80	105.80	0.60	0.20	33.15	0.30	-0.80	0	0	0.31	0.25	0.20	-1	0	0
5	89,102.00	95.17	1.20	1.60	186.42	-2.83	-1.67	0	1	0.58	2.15	0.80	-1	0	0
6	89,108.40	68.20	0.80	1.60	119.88	-5.55	-1.67	0	1	0.61	1.95	0.60	-1	0	0
7	89,116.20	35.50	1.60	1.60	79.04	-13.50	5.75	0	1	0.91	2.62	1.20	-1	0	0
8	89,121.20	35.16	0.60	3.00	94.49	-10.84	5.75	0	1	0.42	3.31	0.40	-1	0	0
9	89,134.20	41.42	1.00	2.20	88.45	-5.83	0.83	0	1	0.57	2.67	0.60	-1	0	0
10	89,145.40	14.70	0.60	1.80	26.61	-8.55	0.80	0	1	0.44	1.96	0.40	-1	0	0
11	89,152.80	78.09	1.40	2.20	184.68	-4.66	-0.34	0	1	0.52	2.73	0.80	-1	0	0
12	89,157.80	65.94	0.80	2.20	152.80	-16.06	-0.34	0	1	0.58	2.61	0.60	-1	0	0
13	89,165.00	23.97	1.40	1.80	50.71	-14.28	2.28	0	1	1.08	2.26	0.80	-1	0	0
14	89,173.40	38.08	1.40	1.40	72.28	-8.67	-0.33	0	1	0.50	2.22	1.00	-1	0	0
15	89,179.60	15.53	1.20	1.20	26.28	-9.47	-0.33	0	1	0.92	1.41	0.40	-1	0	0
16	89,182.00	34.48	0.80	1.40	59.06	-12.27	-0.33	0	1	0.36	1.94	0.80	-1	0	0
17	89,191.60	27.13	1.00	1.60	50.97	-8.87	-1.38	0	1	0.46	2.32	0.80	-1	0	0
18	89,198.40	52.95	1.20	1.60	100.68	-10.55	0.30	0	1	0.61	2.32	0.80	-1	0	0

Peristimulus Time Histogram (PSTH)
ON stimulation causes synchronous burst oscillations in ET cells

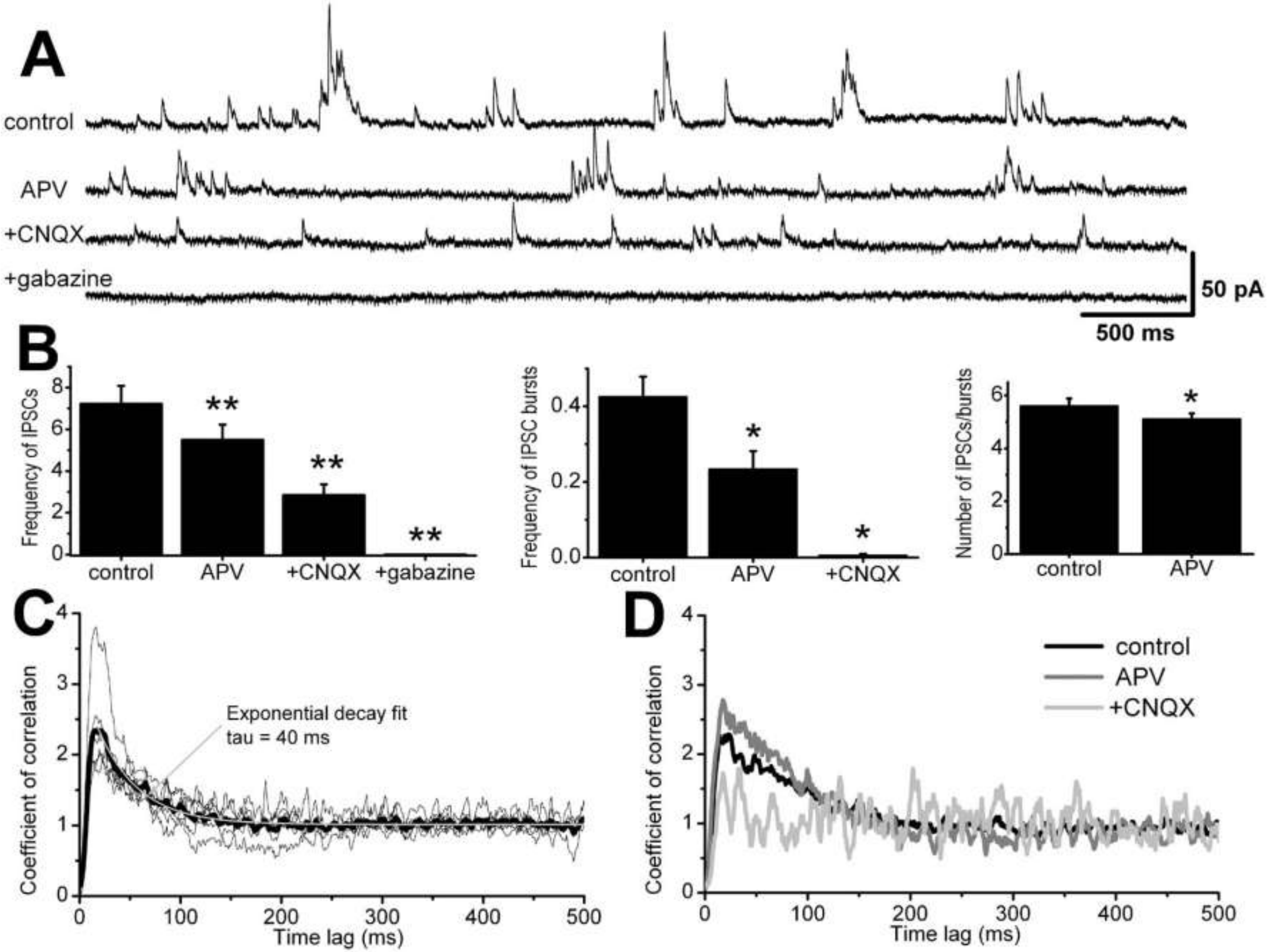
PSTH are histograms of the times at which neurons fire. They are used to visualize the rate and timing of neuronal spike discharges in relation to an external stimulus or event.



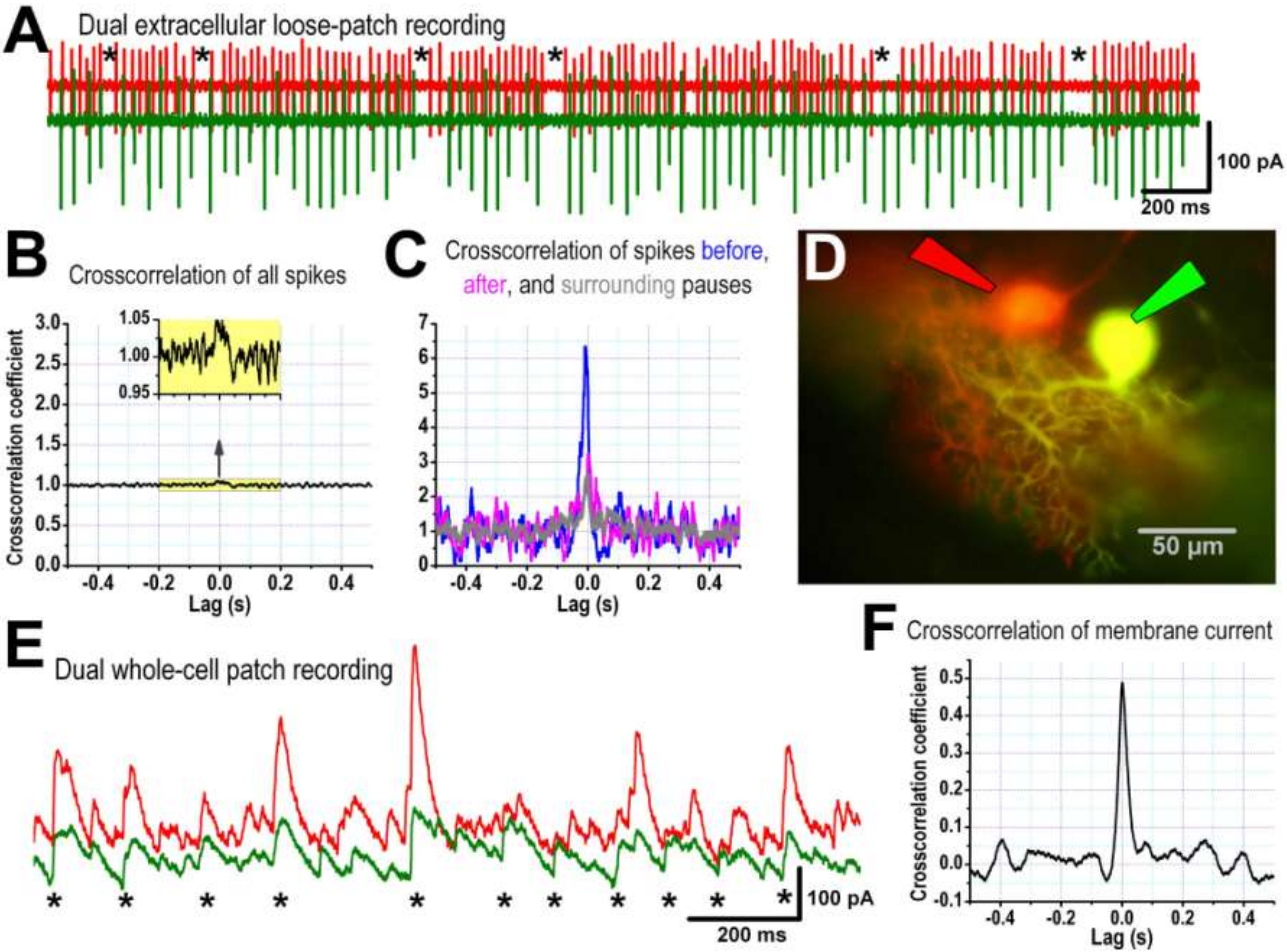
Event autocorrelograms can be used to study regularity



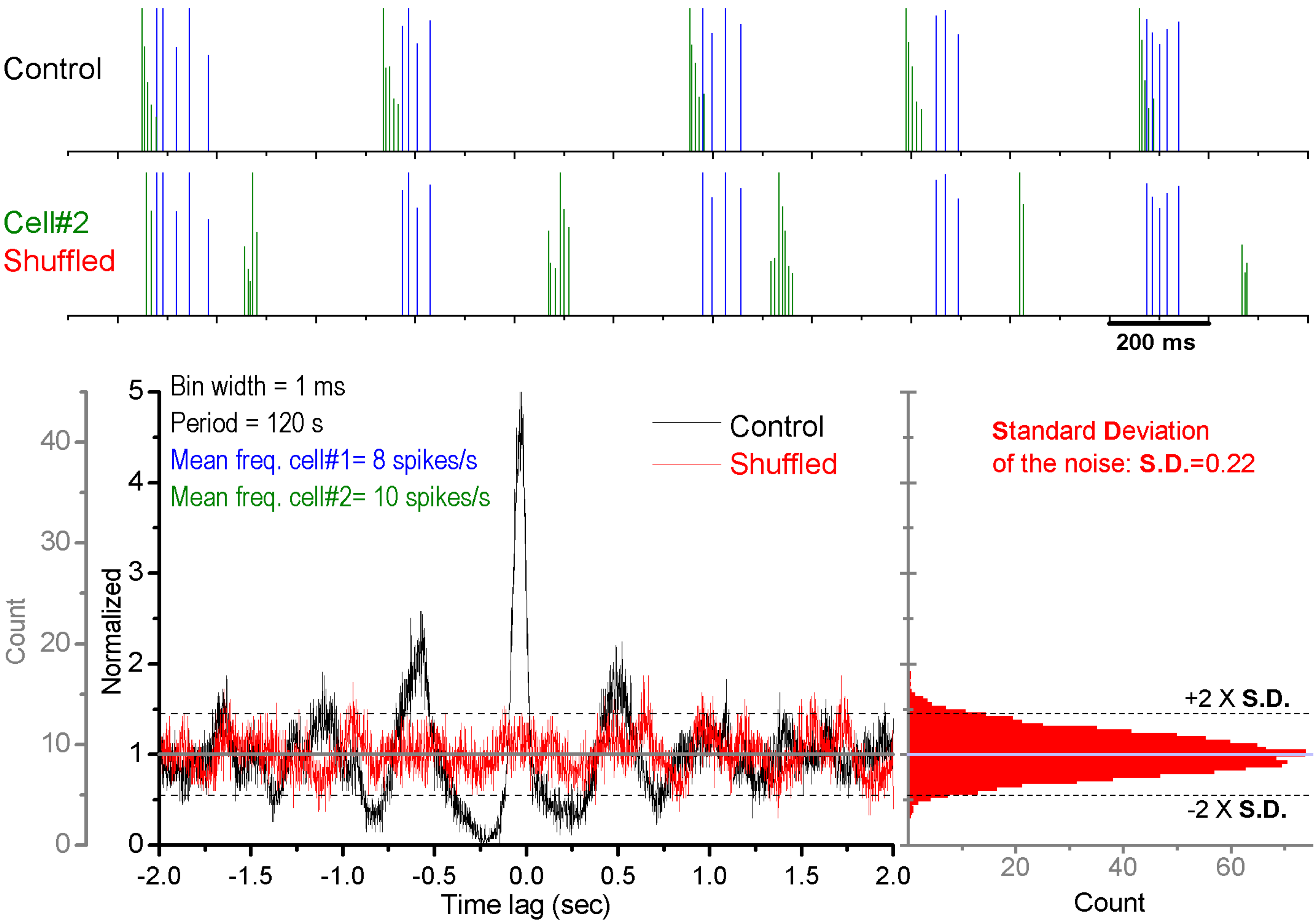
Event autocorrelograms can be used for burst analysis



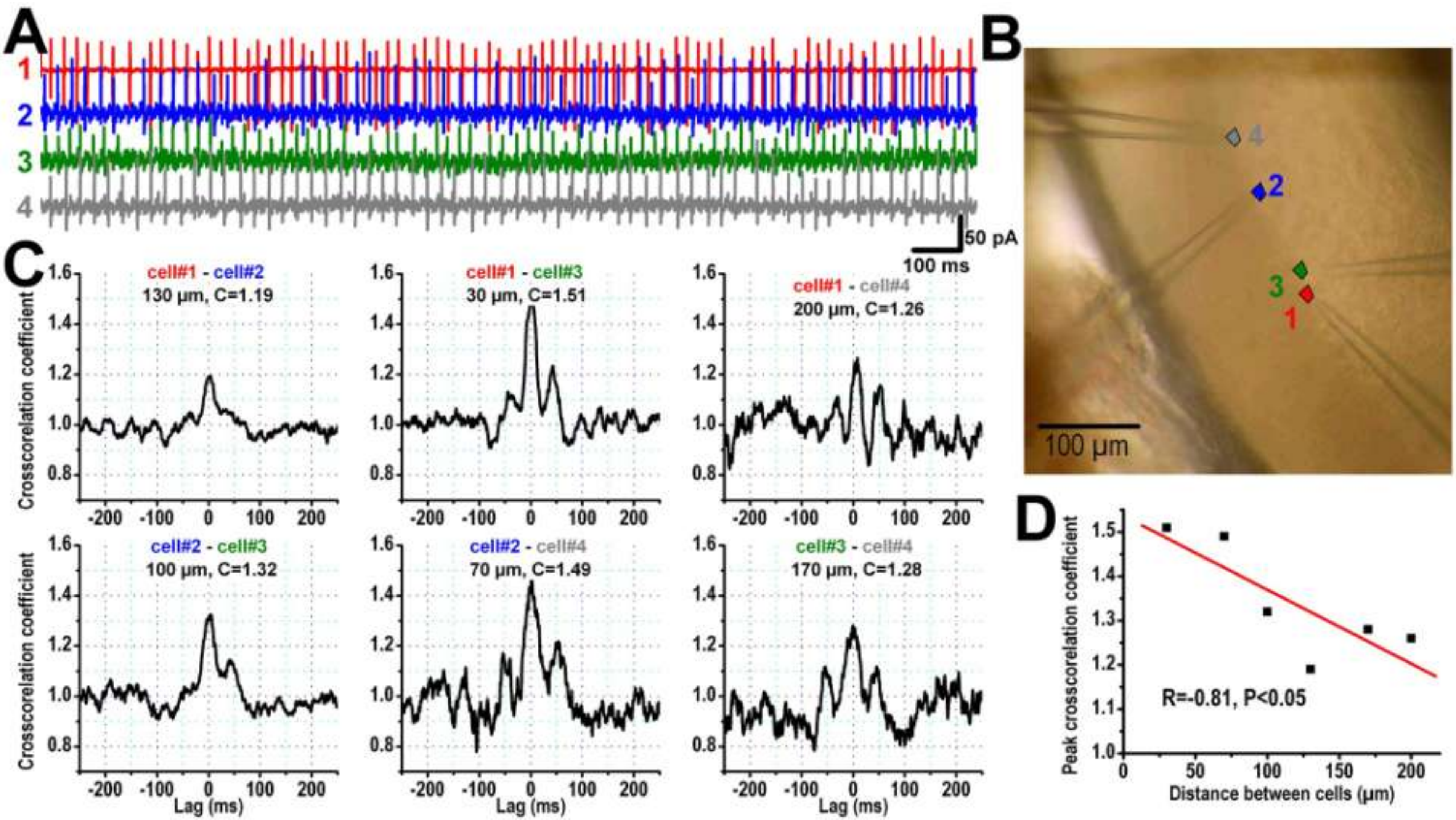
Event Crosscorrelogram vs. Wave Crosscorrelogram



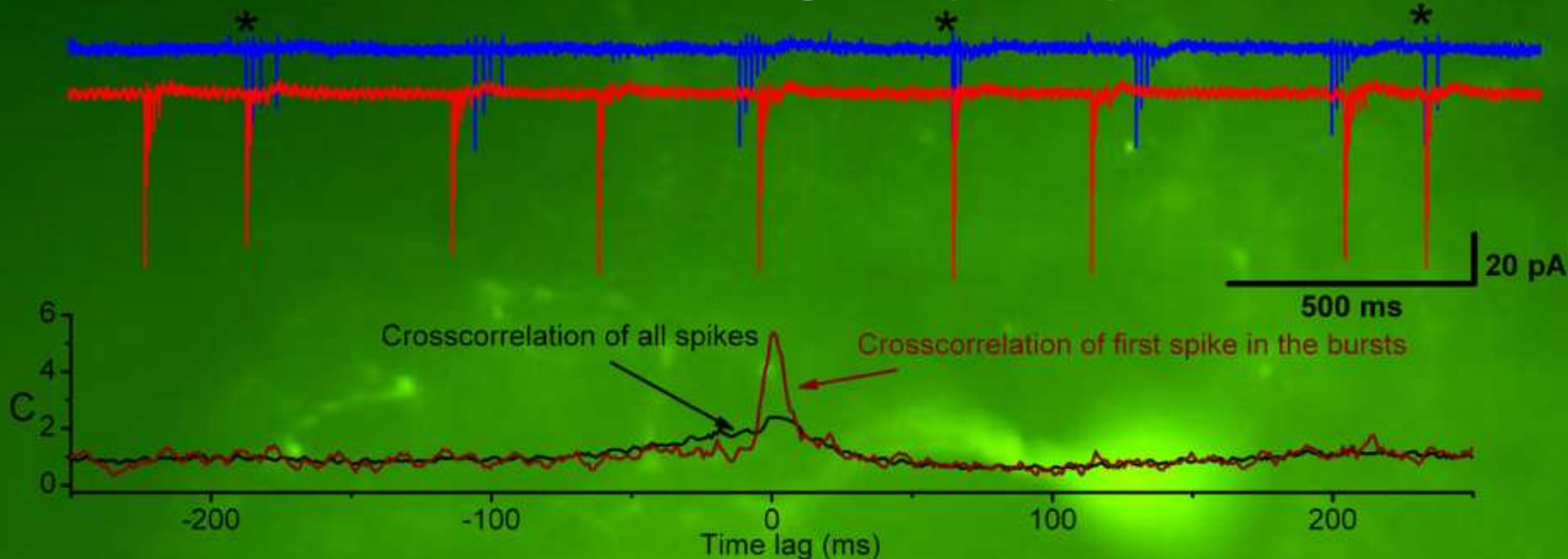
Events (spikes, EPSCs) cross-correlograms: Normalization and significance



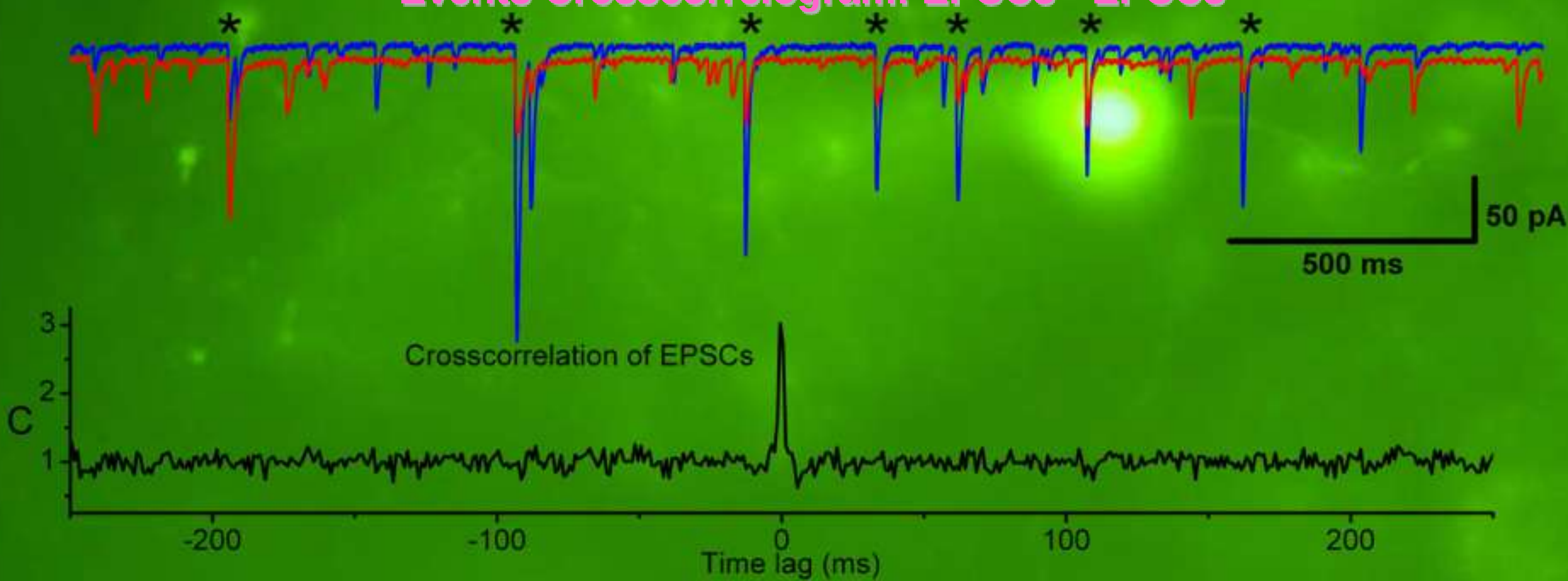
Synchrony decreases as a function of distance

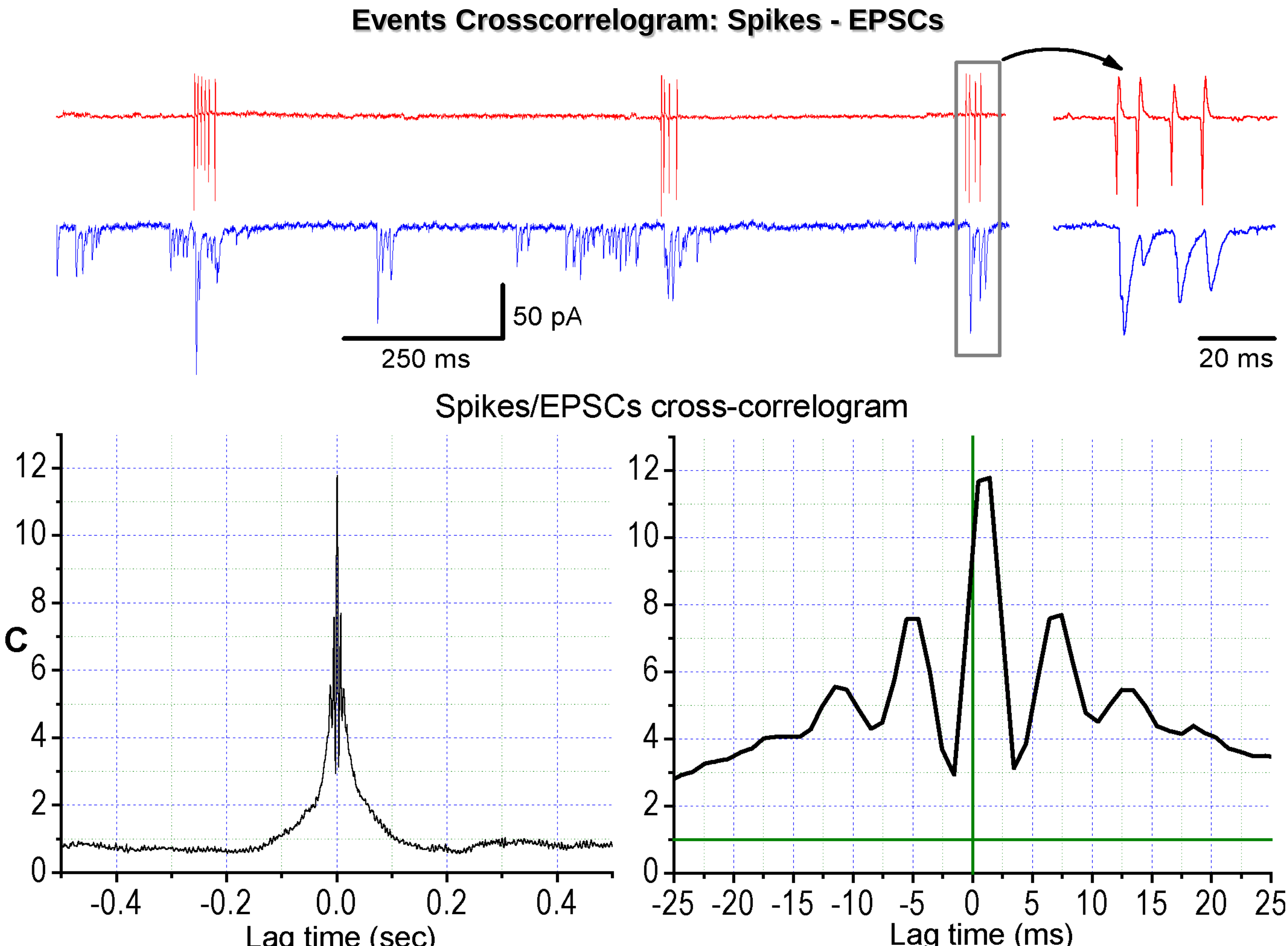


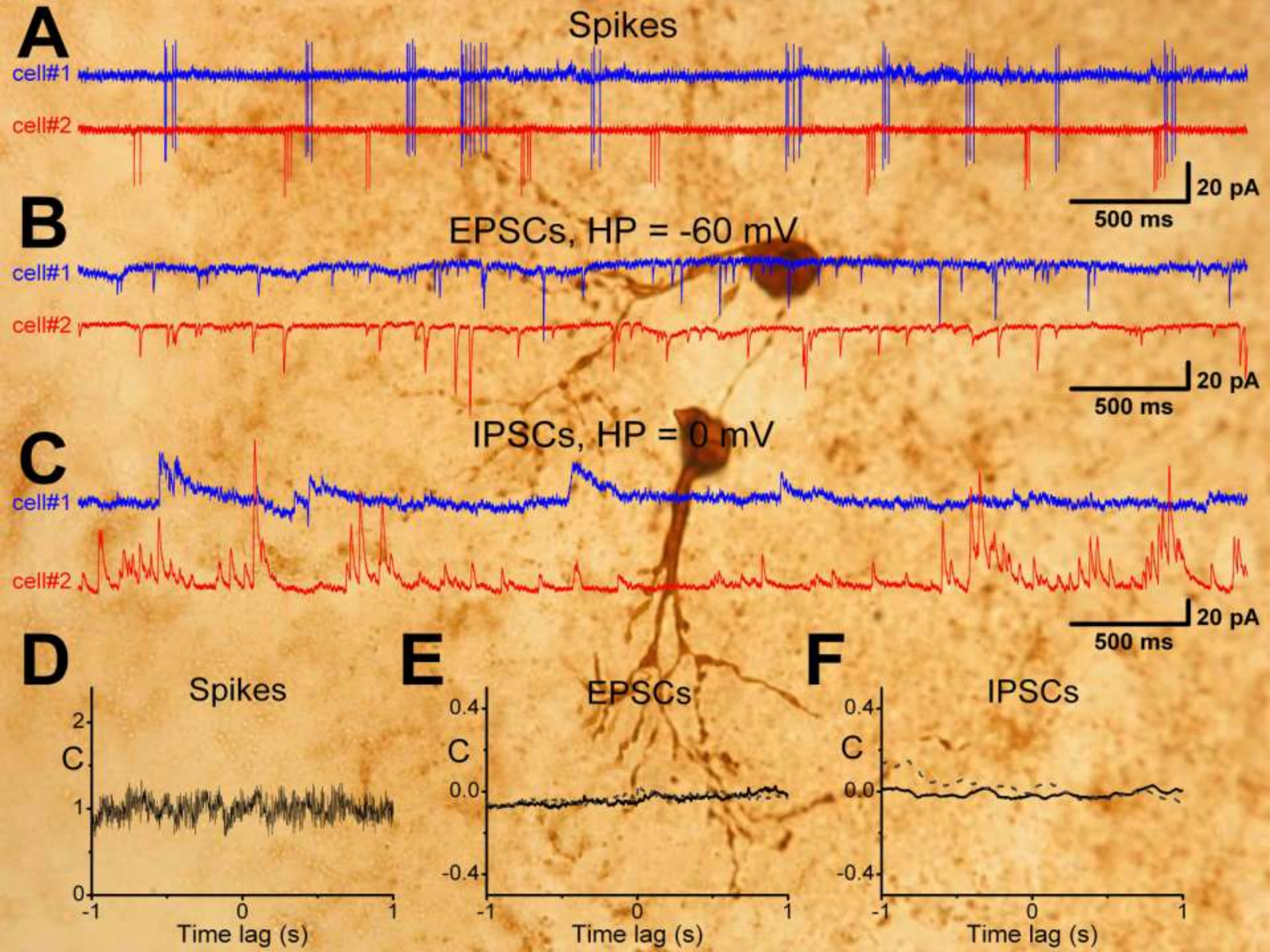
Events Crosscorrelogram: Spikes- Spikes



Events Crosscorrelogram: EPSCs - EPSCs



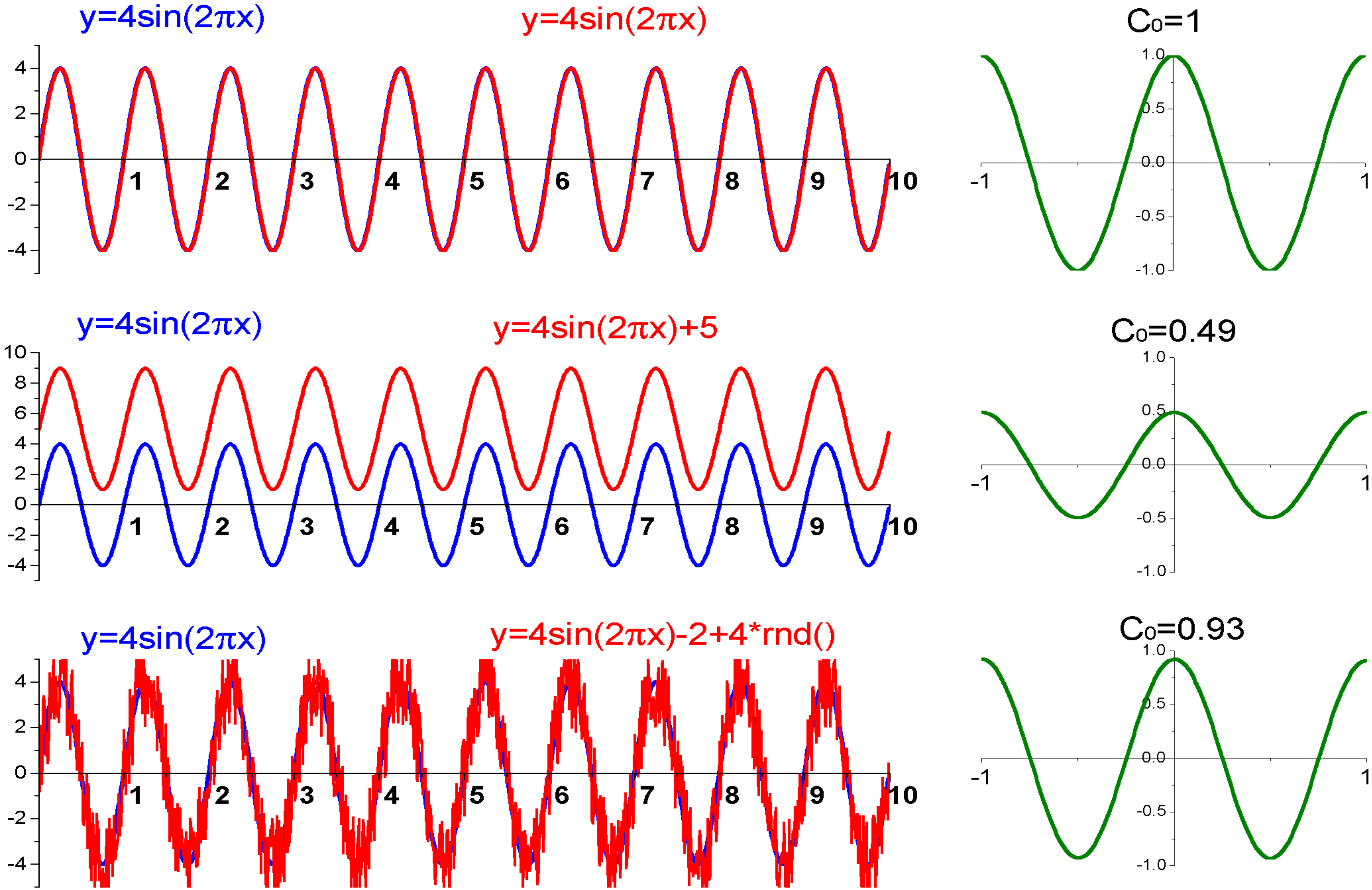

Events Crosscorrelogram: Spikes - EPSCs
50 pA
250 ms
20 ms
Spikes/EPSCs cross-correlogram
C
Lag time (sec)
Lag time (ms)


A
Spikes
cell#1
cell#2
20 pA
500 ms
B
EPSCs, HP = -60 mV
cell#1
cell#2
20 pA
500 ms
C
IPSCs, HP = 0 mV
cell#1
cell#2
20 pA
500 ms
D
Spikes
C
0
1
2
-1
0
1
Time lag (s)
E
EPSCs
C
0.4
0.0
-0.4
-1
0
1
Time lag (s)
F
IPSCs
C
0.4
0.0
-0.4
-1
0
1
Time lag (s)

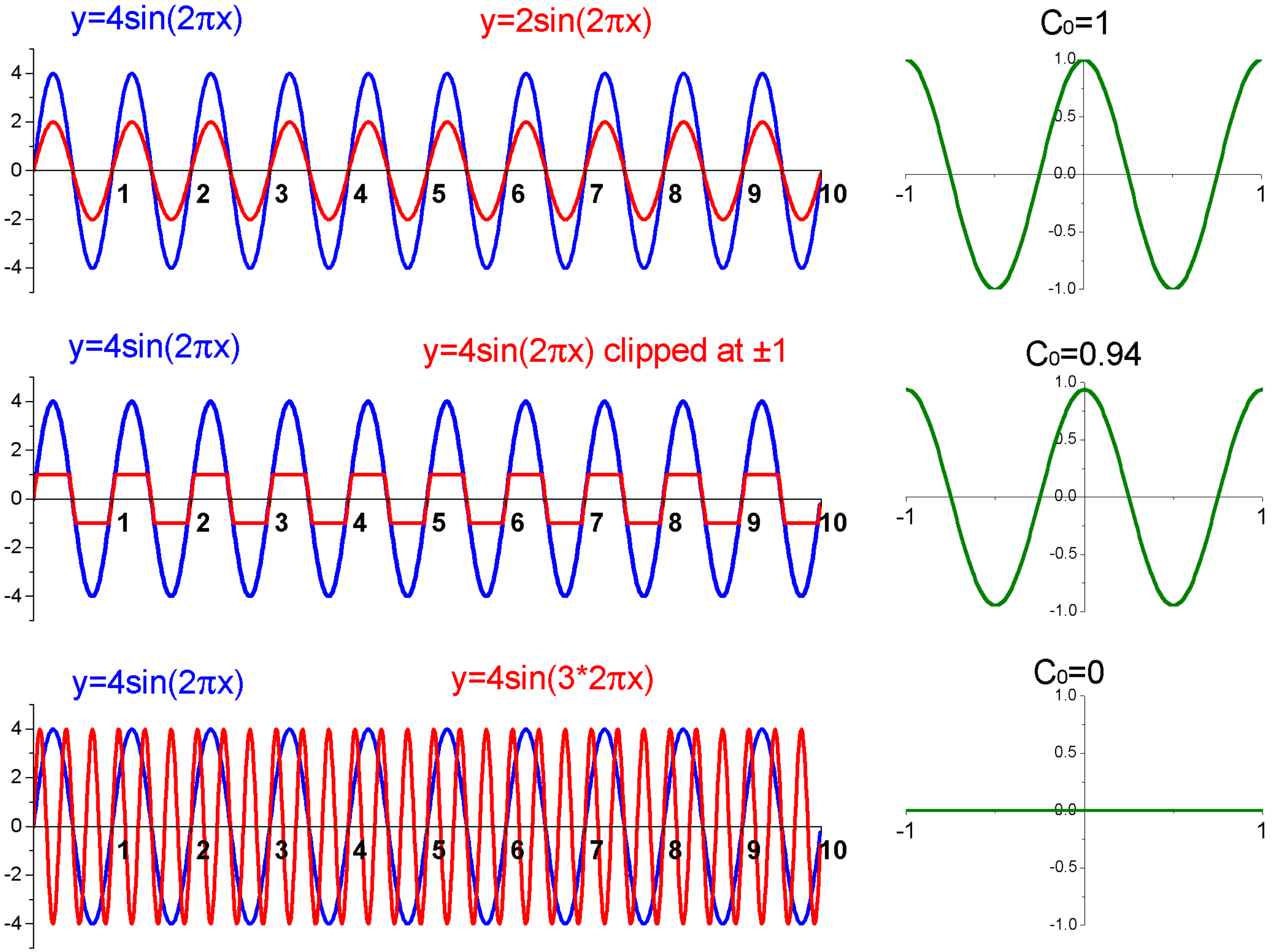
Waveform cross-correlograms: Offset and filtering

Waveform1: 1, 2, 3, 4, 5
Waveform2: 1, 2, 3, 4, 5

Crosscorrelation (Lag 0) C0= 1*1+2*2+3*3+4*4+…
Crosscorrelation (Lag 1) C1= 1*2+2*3+3*4+4*5+…



Waveform cross-correlograms: Amplitude, shape, frequency



Waveform cross-correlograms: Phase-shift

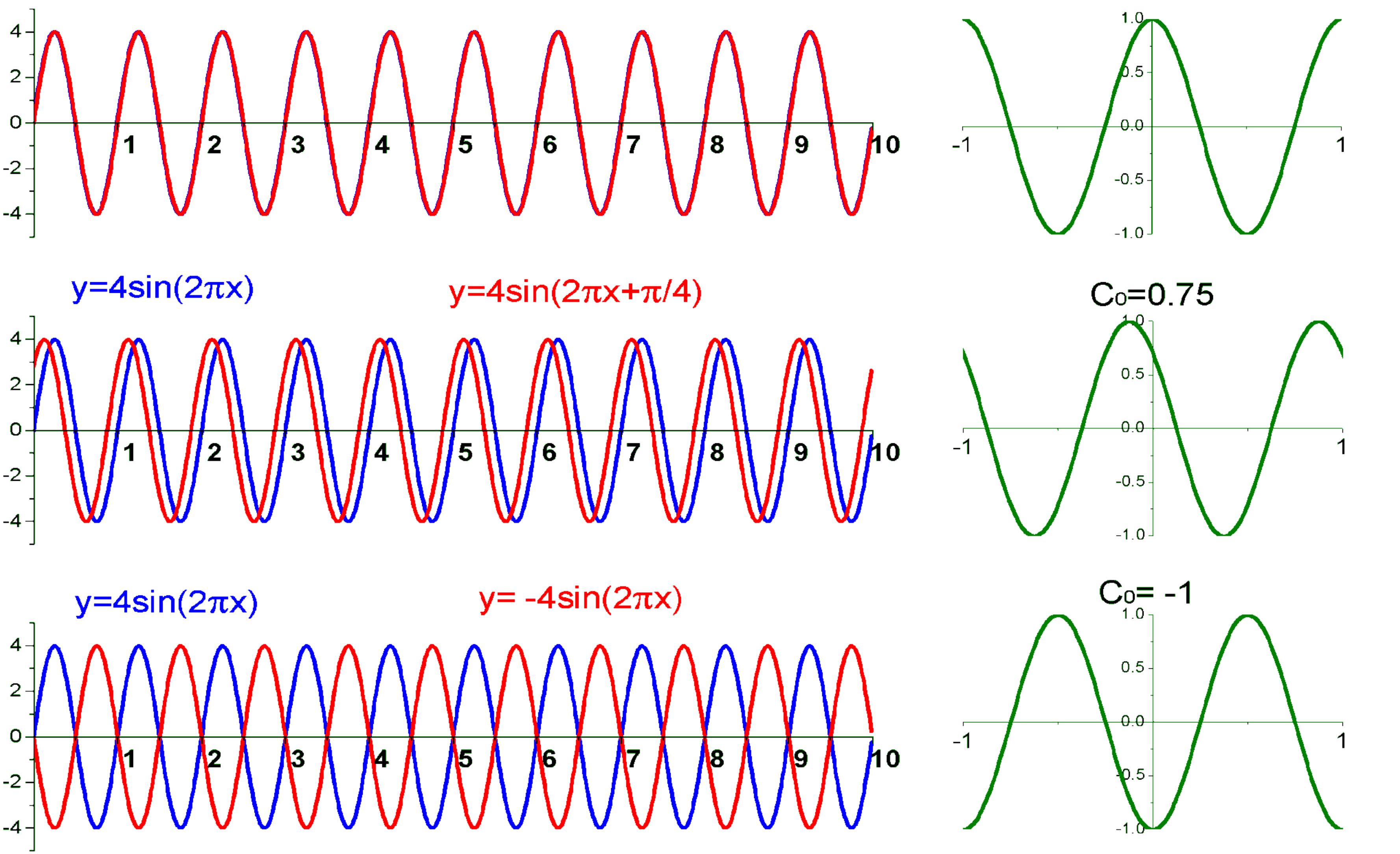
Waveform1: 1, 2, 3, 4, 5

Waveform2: 1, 2, 3, 4, 5

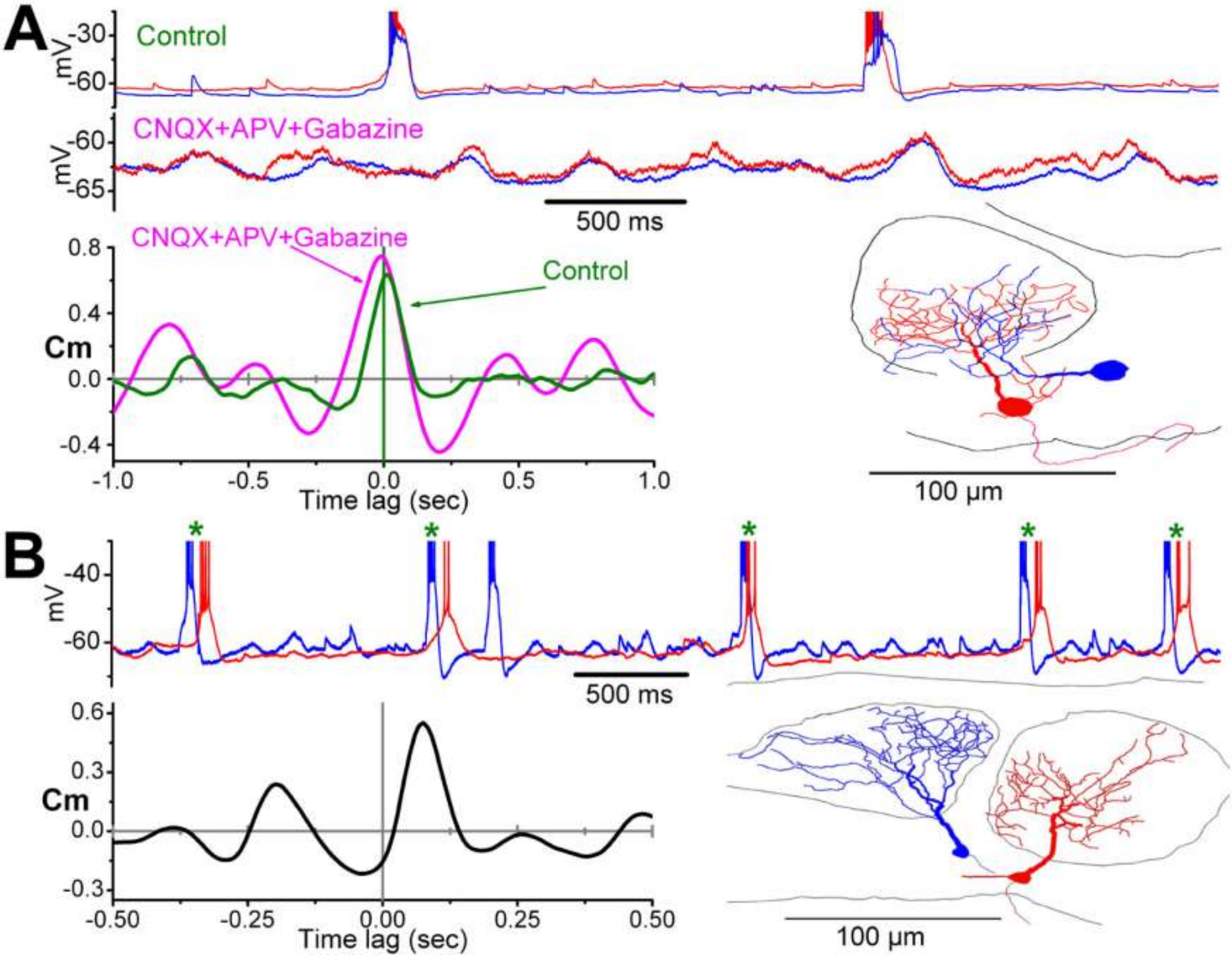
Crosscorrelation (Lag 0) C0= 1*1+2*2+3*3+4*4+…

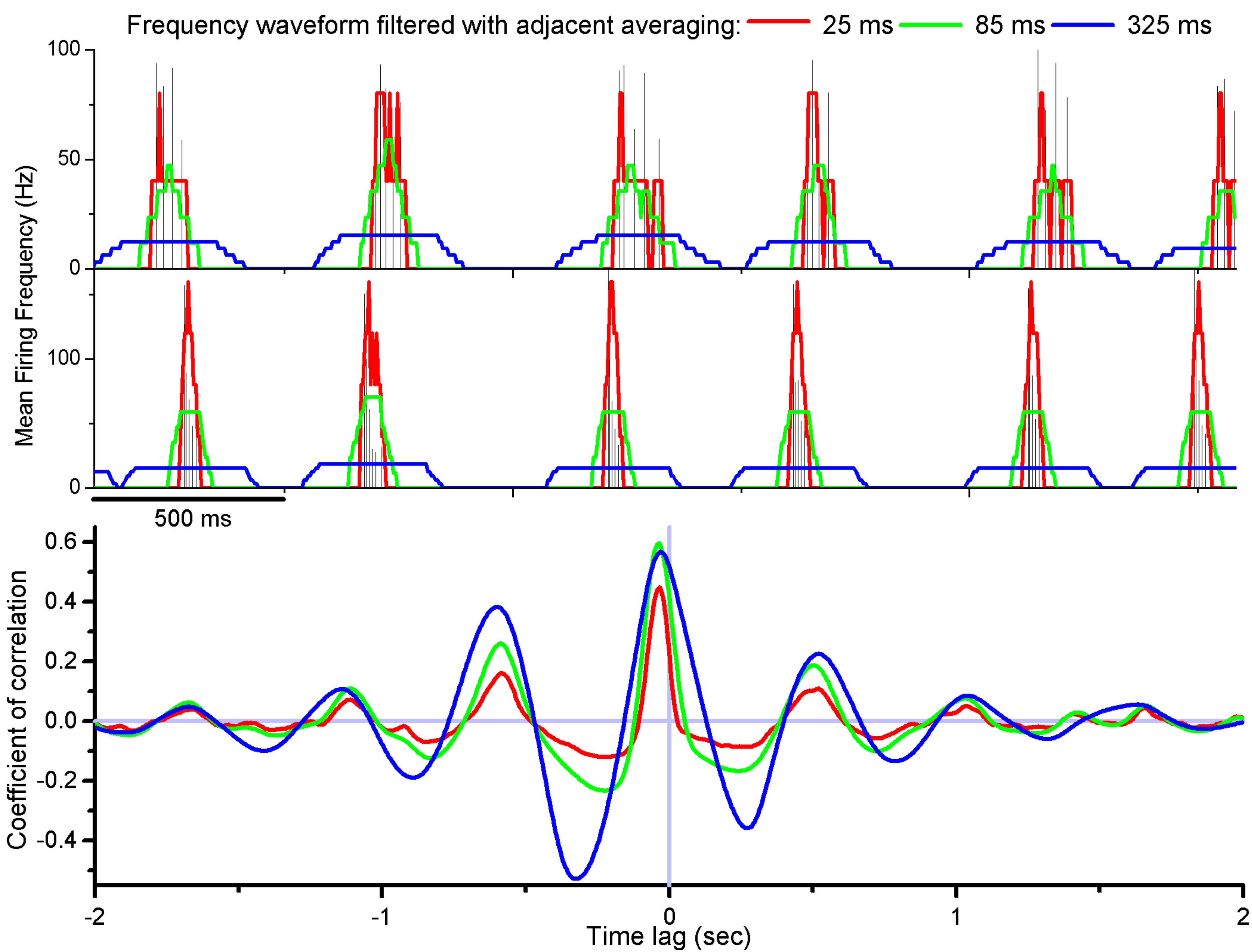
Crosscorrelation (Lag 1) C1= 1*2+2*3+3*4+4*5+…

y=4sin(2πx) y=4sin(2πx) $C_0=1$



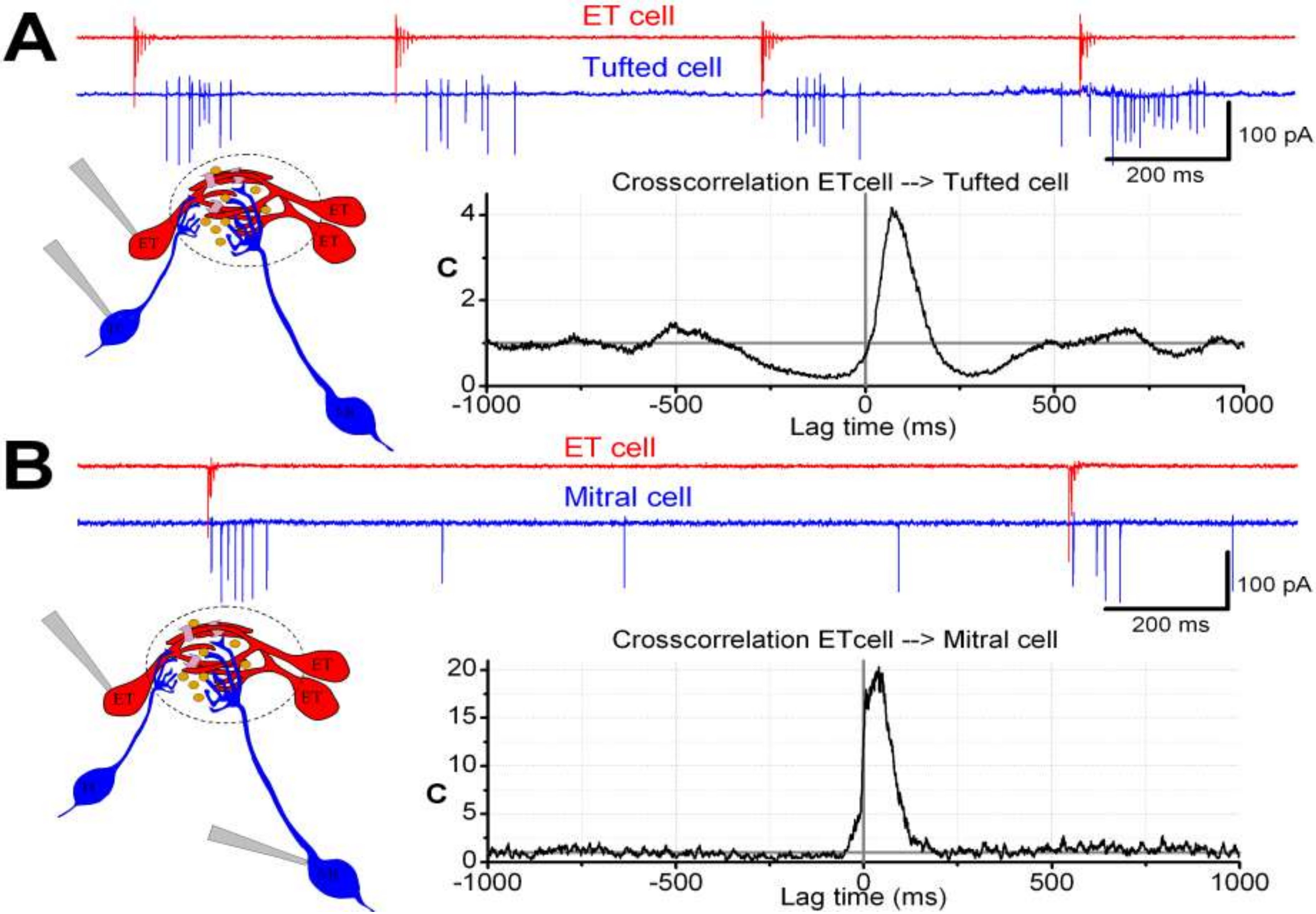
Bursting is highly synchronous among ET cells the same glomerulus



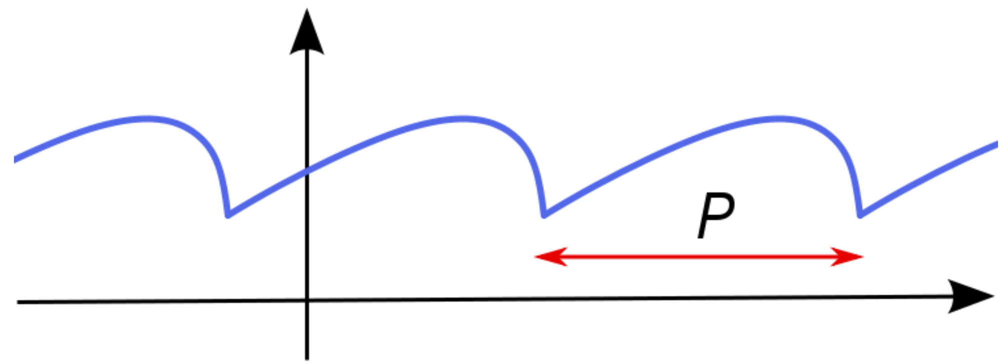


Frequency waveform filtered with adjacent averaging: 25 ms 85 ms 325 ms
100
50
0
100
0
Mean Firing Frequency (Hz)
500 ms
0.6
0.4
0.2
0.0
-0.2
-0.4
Coefficient of correlation
-2
-1
0
1
2
Time lag (sec)

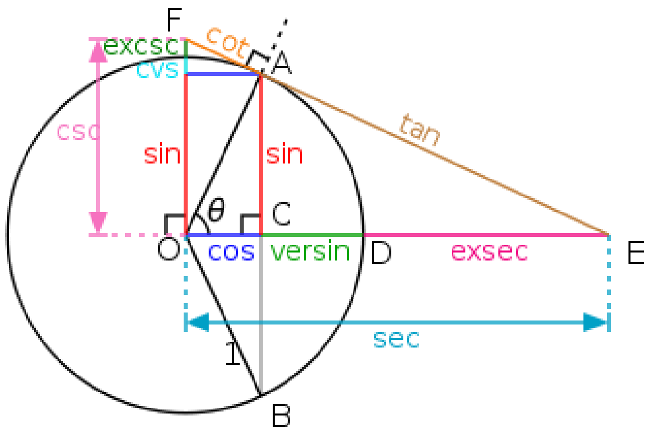
ET cell bursting coordinate tufted/mitral cell spiking

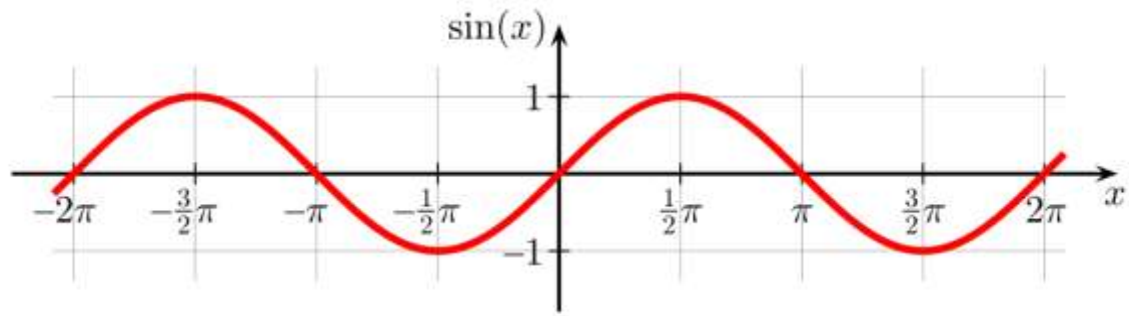


A periodic function is a function that repeats its values in regular intervals or periods.

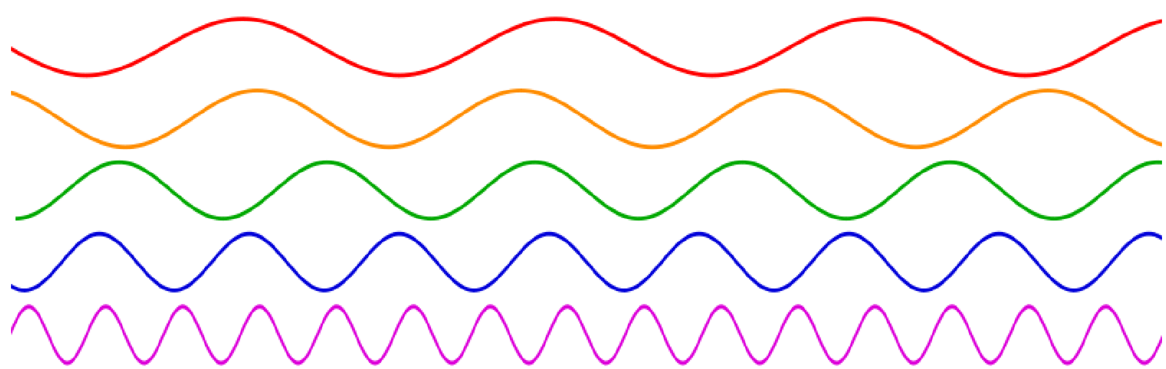


$$T = \frac{1}{f}$$





Frequency is the number of occurrences of a repeating event per unit time.



The fundamental tone, abbreviated f0 or F0, is the lowest frequency in a harmonic series.

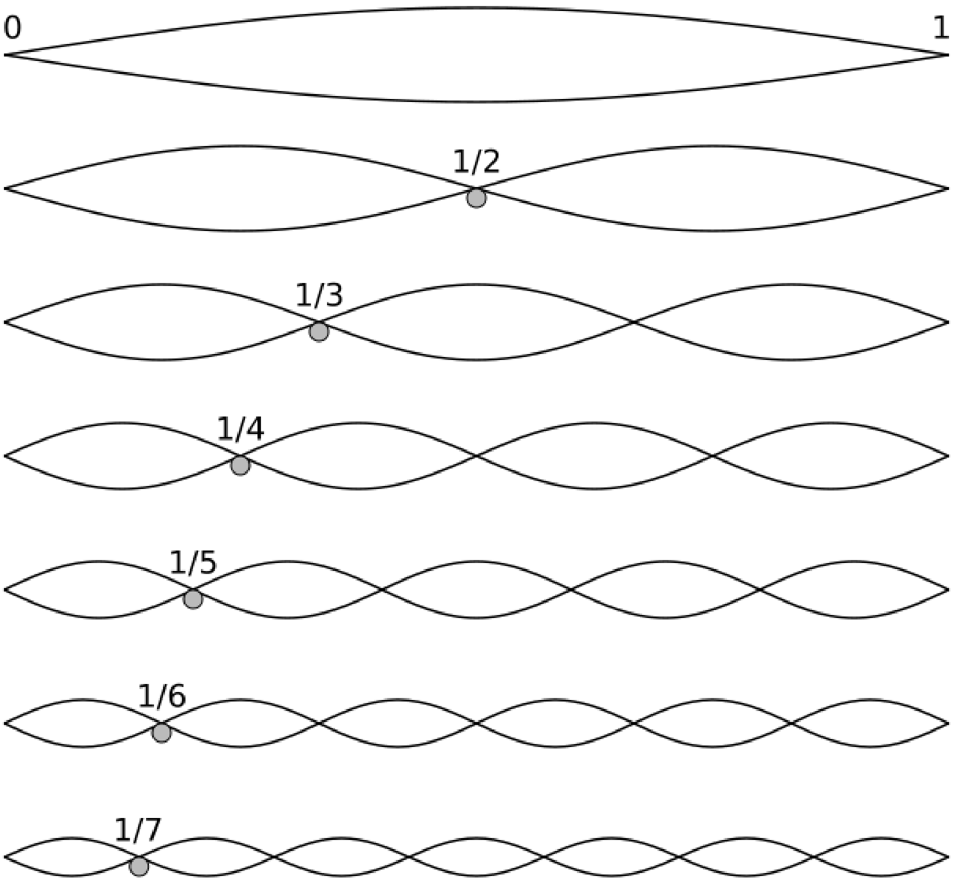
The fundamental frequency of a periodic signal is the inverse of the period length.

The period is, in turn, the smallest repeating unit of a signal. One period thus describes the periodic signal completely.

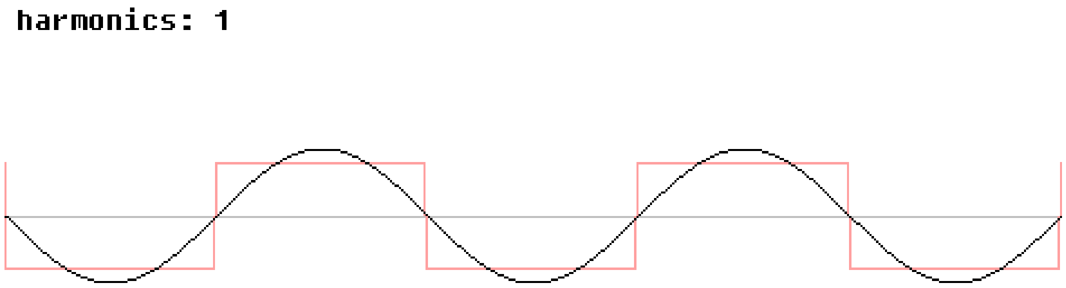
The significance of defining the period as the smallest repeating unit can be appreciated by noting that two or more concatenated periods form a repeating pattern in the signal. However, the concatenated signal unit obviously contains redundant information.

The fundamental frequency is the lowest frequency component of a signal that excites (imparts energy) to a system.

In terms of a superposition of sinusoids (for example, fourier series), the fundamental frequency is the lowest frequency sinusoidal in the sum.

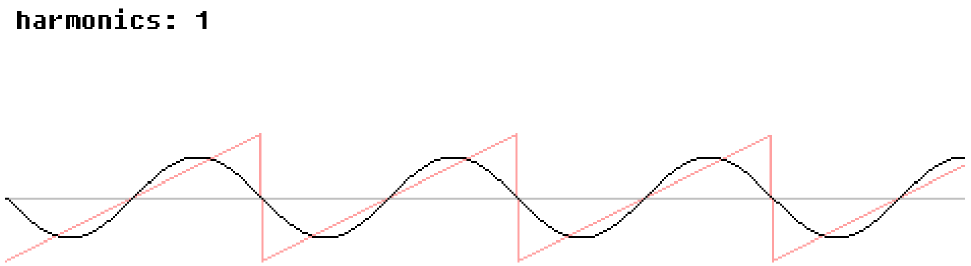


Fourier series decomposes a periodic function or periodic signal into a sum of simple oscillating functions, namely sines and cosines



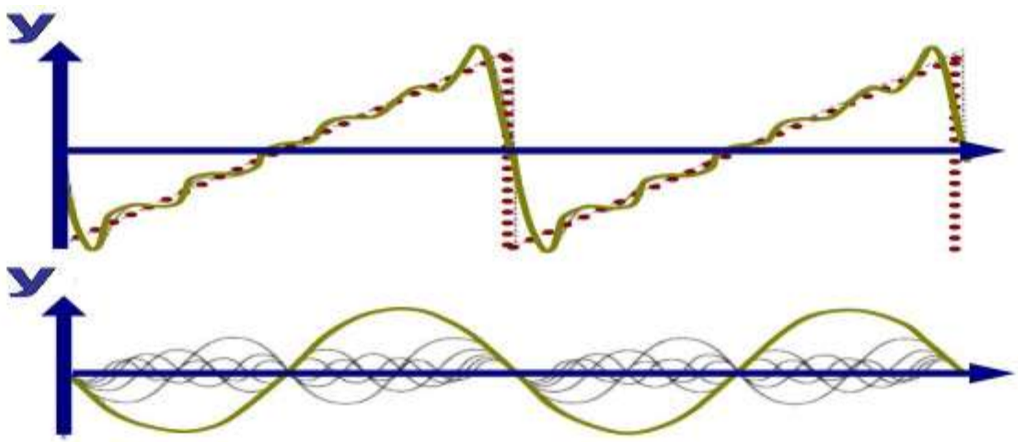
The additive synthesis of a square wave with an increasing number of harmonics

$$\sin(x) + \frac{1}{3}\sin(3x) + \frac{1}{5}\sin(5x) + \cdots$$



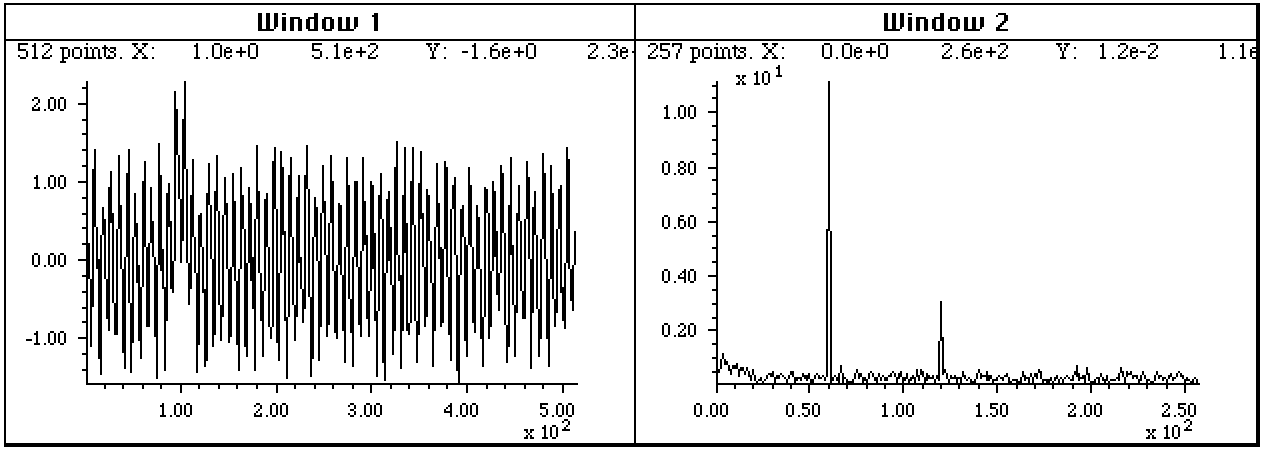
The convention is that a sawtooth wave ramps upward and then sharply drops.

Superposition of sinusoidal wave basis functions (bottom) to form a sawtooth wave (top); the basis functions have wavelengths λ/k (k = integer) shorter than the wavelength λ of the sawtooth itself (except for k = 1).



$$x_{\text{sawtooth}}(t) = -\frac{2}{\pi}\sum_{k=1}^{\infty}\frac{\sin(2\pi k f t)}{k}$$

The power spectrum or frequency spectrum is a simple way of showing the total amplitude at each of frequency; it is calculated as the square root of the sum of the squares of the coefficients of the sine and cosine components.



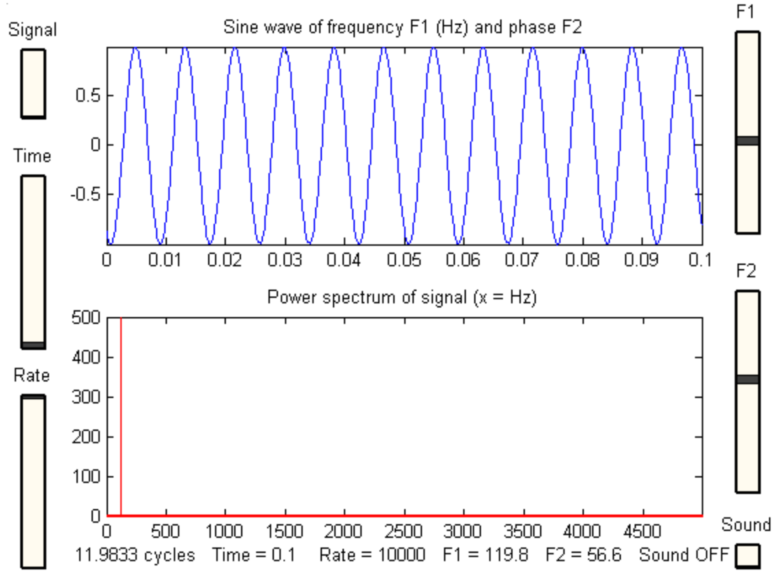
The signal on the left (x = time; y = voltage), which was expected to contain a single peak, is clearly very noisy. The power spectrum of this signal (x-axis = frequency in Hz) shows a strong component at 60 Hz, suggesting that much of the noise is caused by stray pick-up from the 60 Hz power line. The smaller peak at 120 Hz (the second harmonic of 60 Hz) probably comes from the same source.
The units of the x-axis of the power spectrum are simply the reciprocal of the units of the x-axis of the original signal (e.g. sec^{-1} = Hz for a signal whose x-axis is in sec).

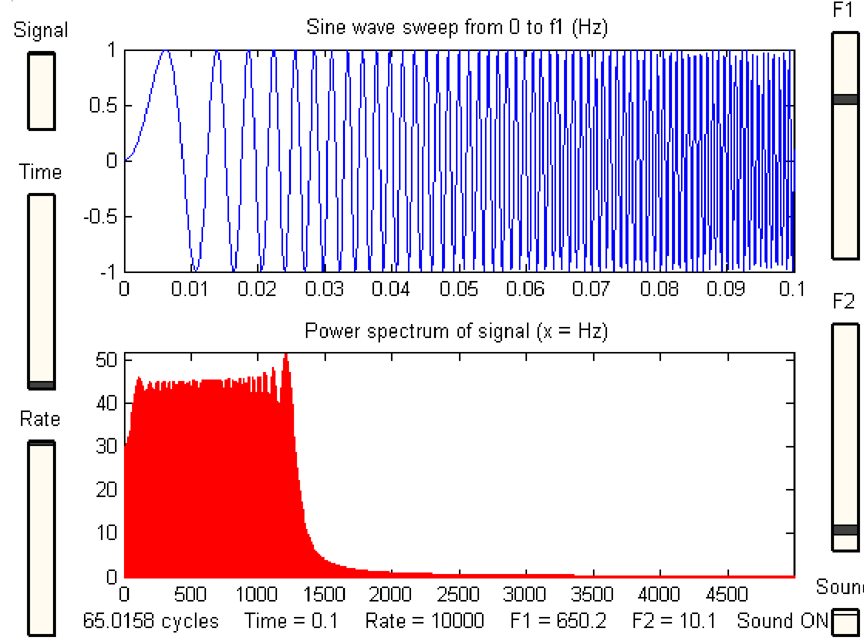
It is possible to analyze any arbitrary set of data into periodic components, whether or not the data appear periodic.

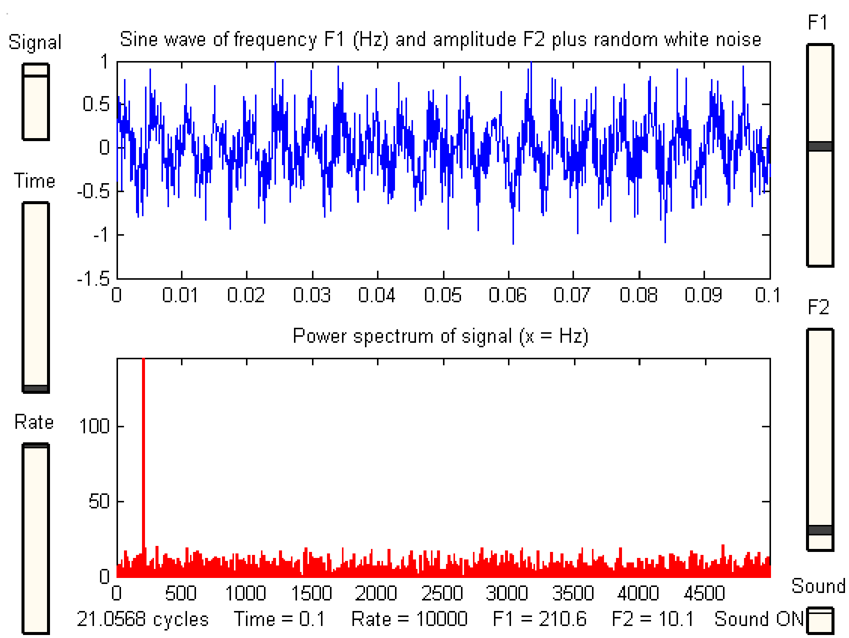
Harmonic analysis is conventionally based on the Fourier transform, which is a way of expressing a signal as a sum of sine and cosine waves.

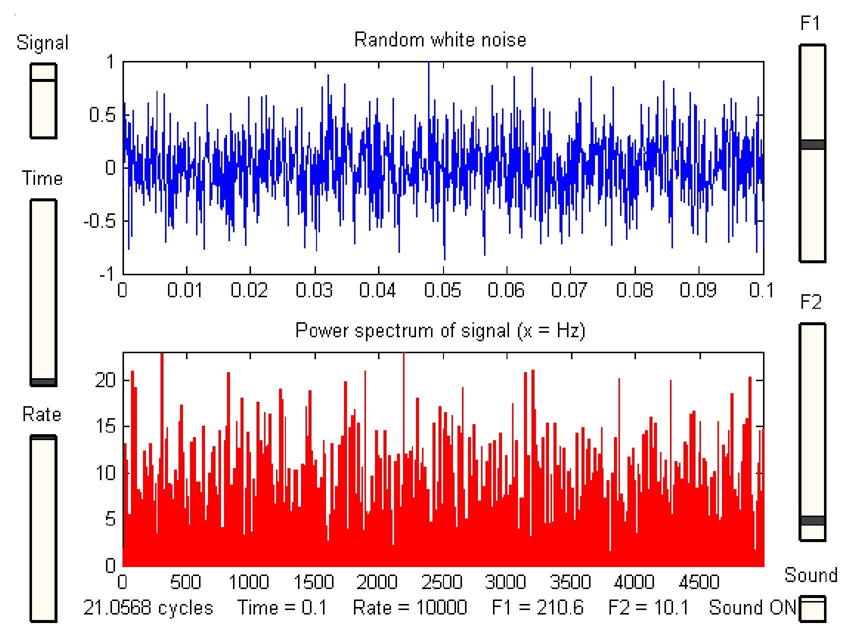
Any arbitrary discretely sampled signal can be described completely by the sum of a finite number of sine and cosine components whose frequencies are 0,1,2,3 ... n/2 times the fundamental frequency f=1/nx, where x is the interval between adjacent x-axis values and n is the total number of points. The Fourier transform is simply the coefficients of these sine and cosine components.

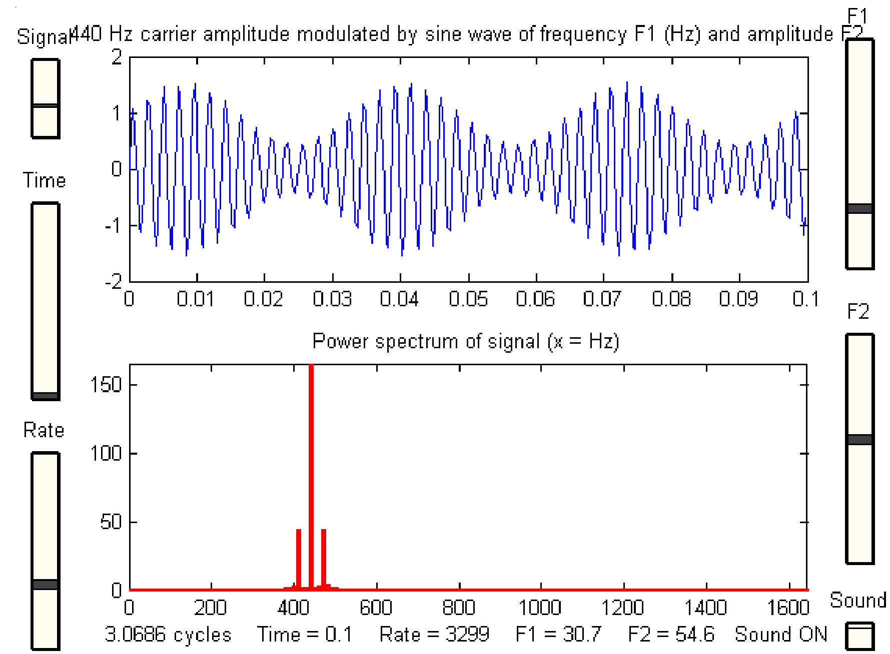
For example if you have a trace of 2 sec sampled at 1 kHz (n=2000 points- duration of each point: x=1 ms),
The fundamental frequency f=1/nx = 1/ (2000 X 0.001sec) =1/2 =0.5 Hz
The power spectrum will be from 0, 0.5,1, 1.5, 2, 2.5,……., n/2=1000 Hz

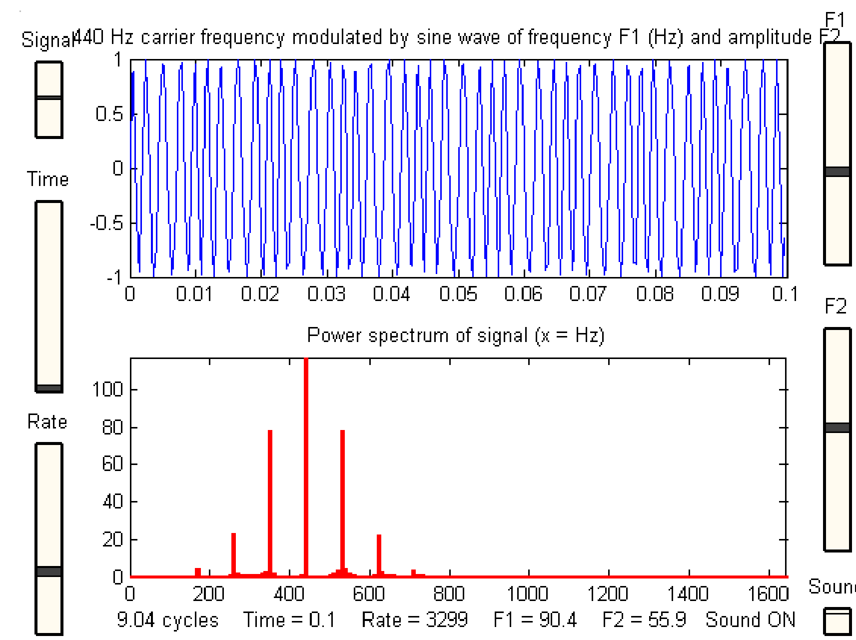

Signal
Time
Rate
Sine wave of frequency F1 (Hz) and phase F2
Power spectrum of signal (x = Hz)
F1
F2
Sound
11.9833 cycles
Time = 0.1
Rate = 10000
F1 = 119.8
F2 = 56.6
Sound OFF

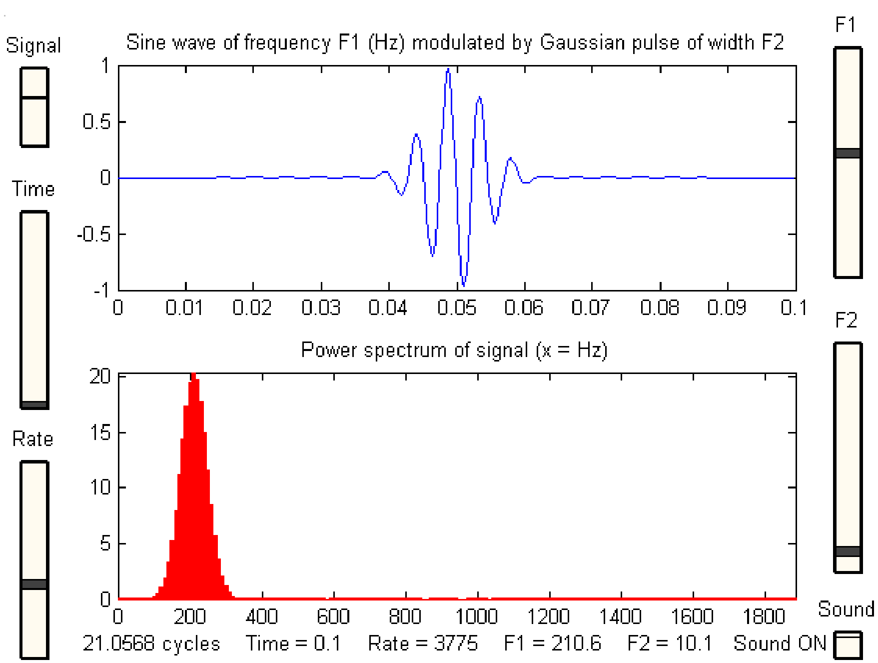

Signal
Time
Rate
Sine wave sweep from 0 to f1 (Hz)
Power spectrum of signal (x = Hz)
F1
F2
Sound
65.0158 cycles
Time = 0.1
Rate = 10000
F1 = 650.2
F2 = 10.1
Sound ON

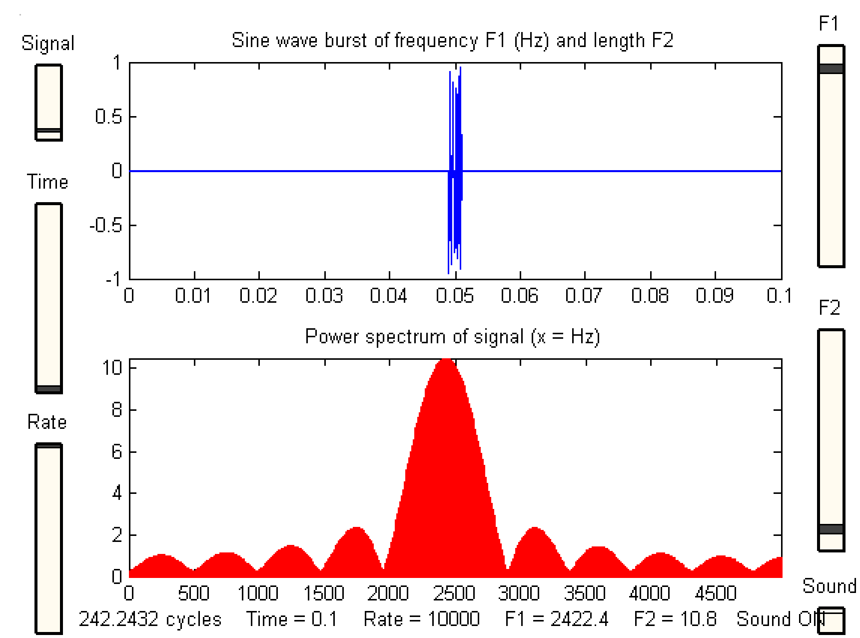

Signal
Time
Rate
Sine wave of frequency F1 (Hz) and amplitude F2 plus random white noise
Power spectrum of signal (x = Hz)
F1
F2
Sound
21.0568 cycles
Time = 0.1
Rate = 10000
F1 = 210.6
F2 = 10.1
Sound ON

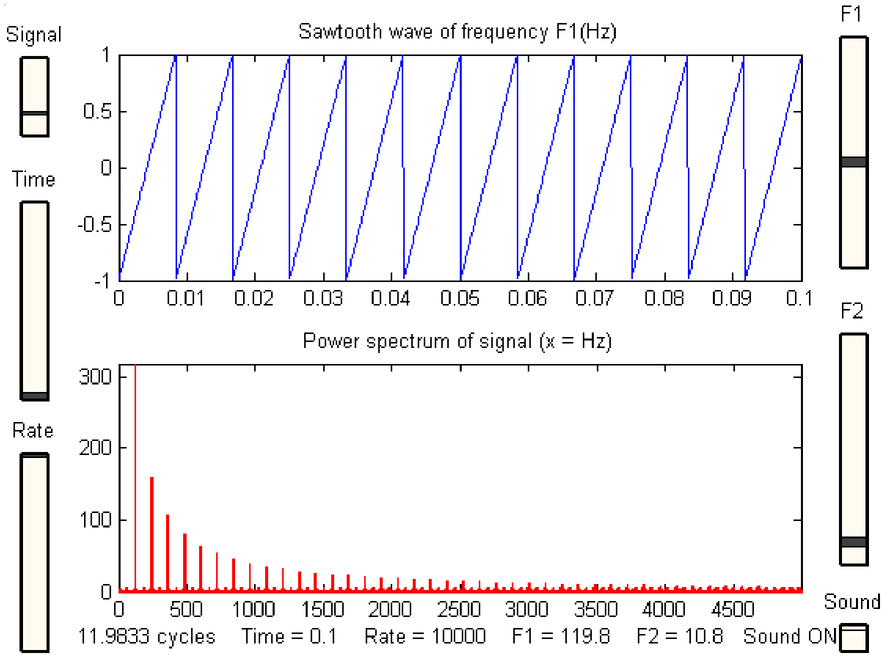

Signal
Time
Rate
Random white noise
Power spectrum of signal (x = Hz)
F1
F2
Sound
21.0568 cycles
Time = 0.1
Rate = 10000
F1 = 210.6
F2 = 10.1
Sound ON

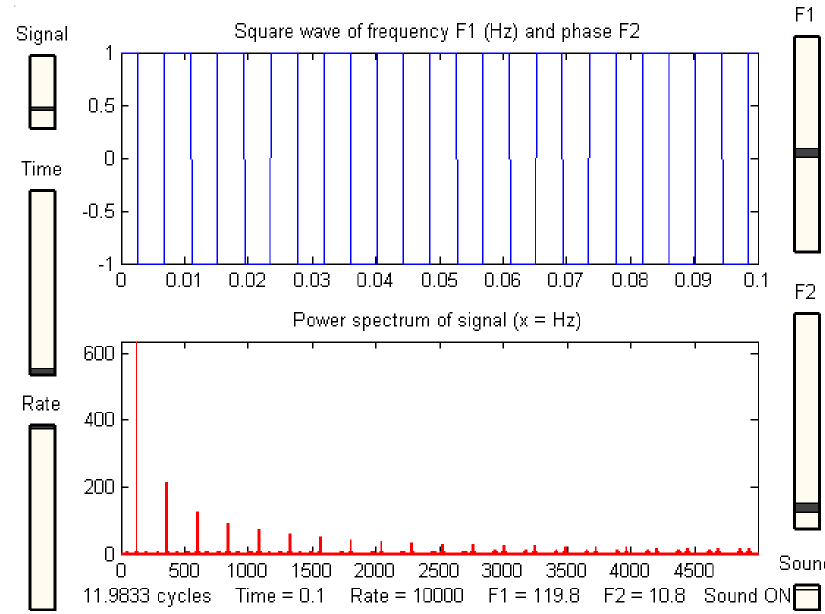

Signal
Time
Rate
440 Hz carrier amplitude modulated by sine wave of frequency F1 (Hz) and amplitude F2
Power spectrum of signal (x = Hz)
F1
F2
Sound
3.0686 cycles
Time = 0.1
Rate = 3299
F1 = 30.7
F2 = 54.6
Sound ON


Signal
Time
Rate
440 Hz carrier frequency modulated by sine wave of frequency F1 (Hz) and amplitude F2
Power spectrum of signal (x = Hz)
F1
F2
Sound
9.04 cycles
Time = 0.1
Rate = 3299
F1 = 90.4
F2 = 55.9
Sound ON

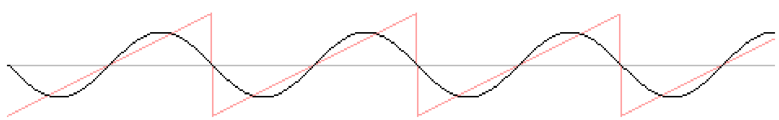

Signal
Time
Rate
Sine wave of frequency F1 (Hz) modulated by Gaussian pulse of width F2
Power spectrum of signal (x = Hz)
F1
F2
Sound
21.0568 cycles
Time = 0.1
Rate = 3775
F1 = 210.6
F2 = 10.1
Sound ON


Signal
Time
Rate
Sine wave burst of frequency F1 (Hz) and length F2
Power spectrum of signal (x = Hz)
F1
F2
Sound
242.2432 cycles
Time = 0.1
Rate = 10000
F1 = 2422.4
F2 = 10.8
Sound ON

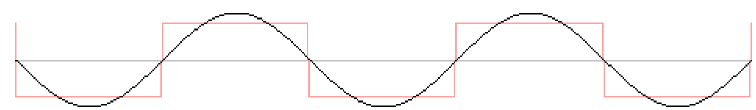


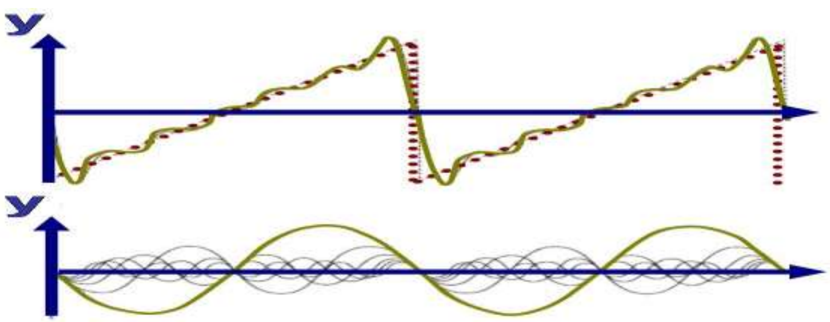


harmonics: 1



harmonics: 1

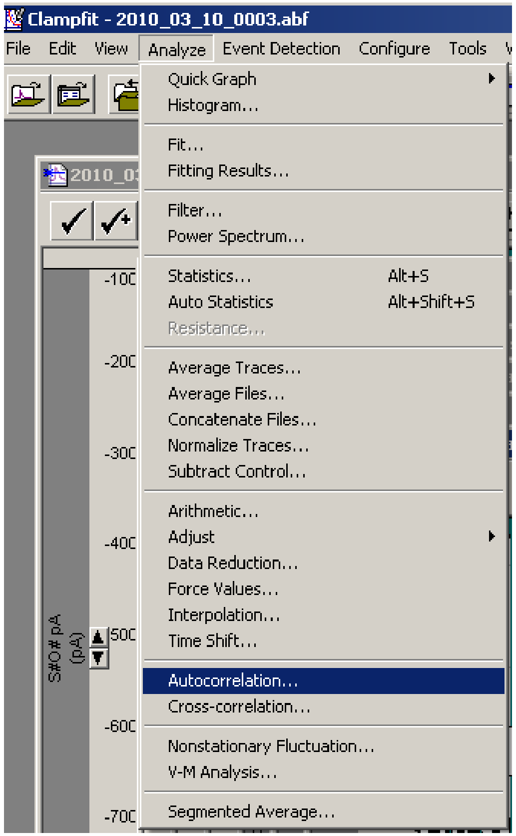


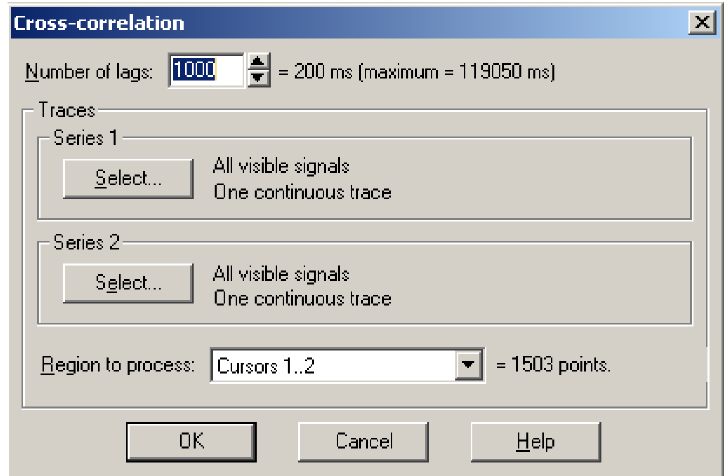


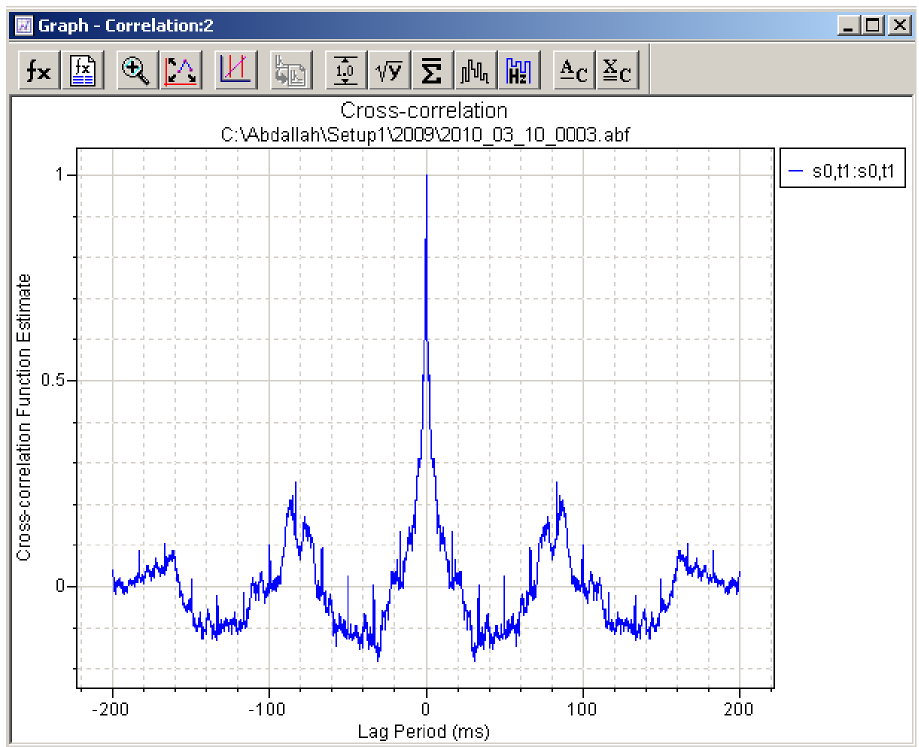
$$\sin(x) + \frac{1}{3}\sin(3x) + \frac{1}{5}\sin(5x) + \cdots$$

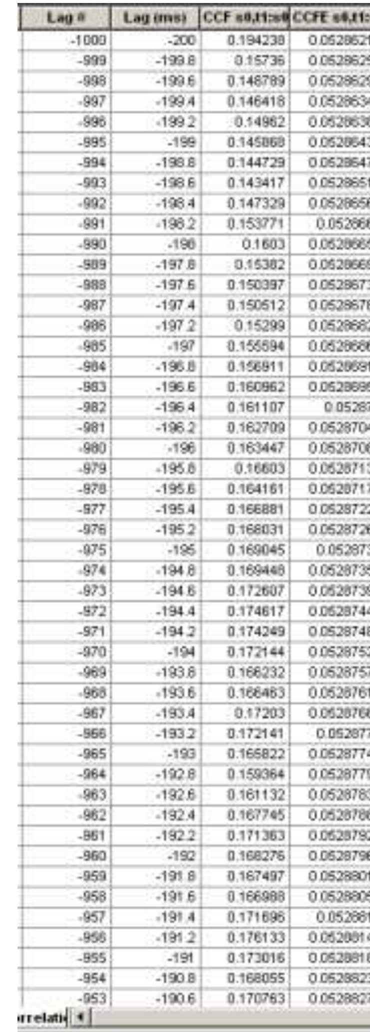
$$x_{\text{sawtooth}}(t) = -\frac{2}{\pi}\sum_{k=1}^{\infty}\frac{\sin(2\pi k f t)}{k}$$

Autocorrelation and crosscorrelation in Clampfit

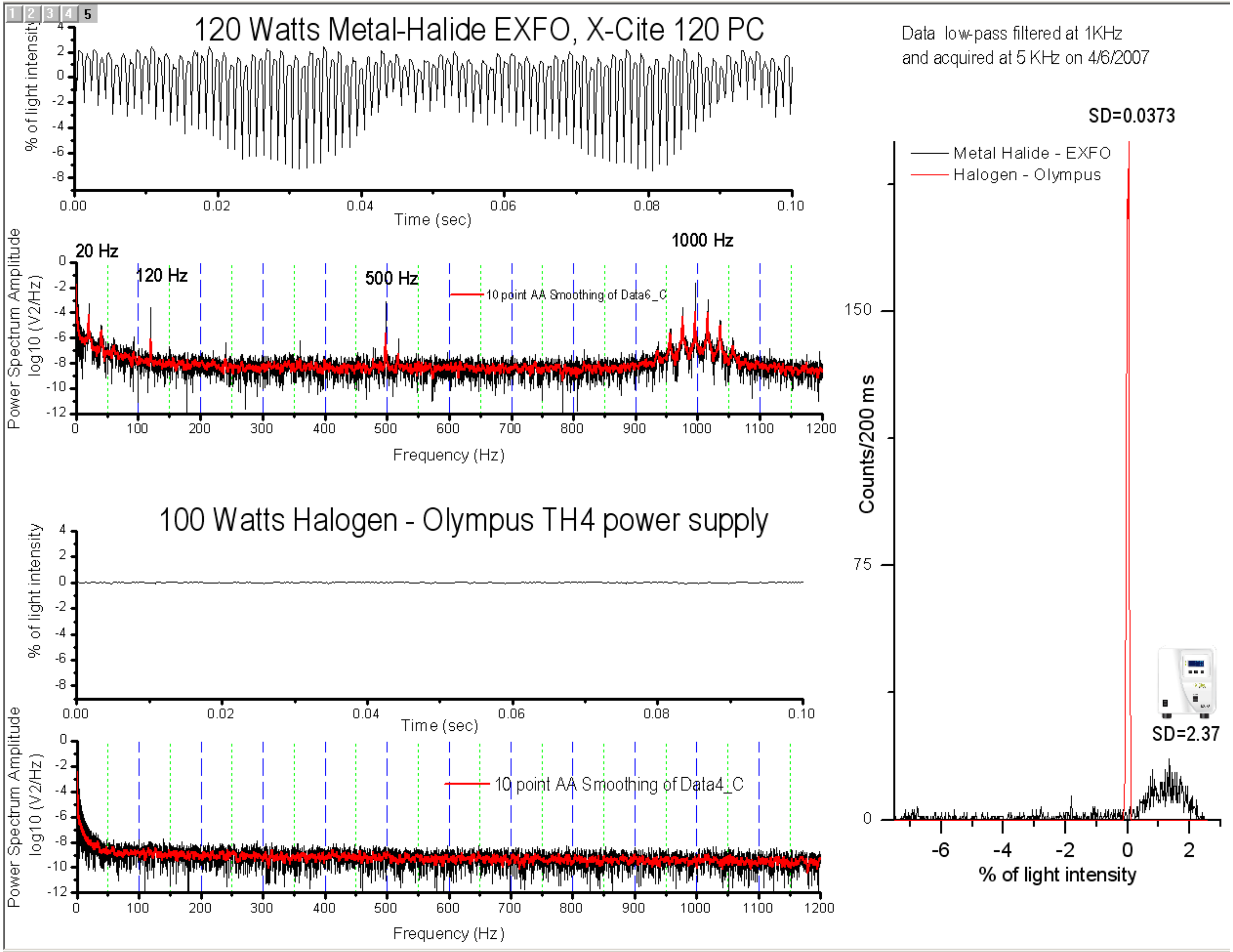








Lag #	Lag (ms)	CCF s0,t1:s0	CCFE s0,t1:s
-1000	-200	0.194238	0.0528621
-999	-199.8	0.15736	0.0528625
-998	-199.6	0.148789	0.0528629
-997	-199.4	0.146418	0.0528634
-996	-199.2	0.14962	0.0528638
-995	-199	0.145868	0.0528643
-994	-198.8	0.144729	0.0528647
-993	-198.6	0.143417	0.0528651
-992	-198.4	0.147329	0.0528656
-991	-198.2	0.153771	0.052866
-990	-198	0.1603	0.0528665
-989	-197.8	0.15382	0.0528669
-988	-197.6	0.150397	0.0528673
-987	-197.4	0.150512	0.0528678
-986	-197.2	0.15299	0.0528682
-985	-197	0.155594	0.0528686
-984	-196.8	0.156911	0.0528691
-983	-196.6	0.160962	0.0528695
-982	-196.4	0.161107	0.05287
-981	-196.2	0.162709	0.0528704
-980	-196	0.163447	0.0528708
-979	-195.8	0.16603	0.0528713
-978	-195.6	0.164161	0.0528717
-977	-195.4	0.166881	0.0528722
-976	-195.2	0.168031	0.0528726
-975	-195	0.169045	0.052873
-974	-194.8	0.169448	0.0528735
-973	-194.6	0.172607	0.0528739
-972	-194.4	0.174617	0.0528744
-971	-194.2	0.174249	0.0528748
-970	-194	0.172144	0.0528752
-969	-193.8	0.166232	0.0528757
-968	-193.6	0.166463	0.0528761
-967	-193.4	0.17203	0.0528766
-966	-193.2	0.172141	0.052877
-965	-193	0.165822	0.0528774
-964	-192.8	0.159364	0.0528779
-963	-192.6	0.161132	0.0528783
-962	-192.4	0.167745	0.0528788
-961	-192.2	0.171363	0.0528792
-960	-192	0.168276	0.0528796
-959	-191.8	0.167497	0.0528801
-958	-191.6	0.166988	0.0528805
-957	-191.4	0.171696	0.052881
-956	-191.2	0.176133	0.0528814
-955	-191	0.173016	0.0528818
-954	-190.8	0.168055	0.0528823
-953	-190.6	0.170763	0.0528827



120 Watts Metal-Halide EXFO, X-Cite 120 PC
% of light intensity
Time (sec)
Power Spectrum Amplitude log10 (V2/Hz)
20 Hz
120 Hz
500 Hz
1000 Hz
10 point AA Smoothing of Data6_C
Frequency (Hz)
100 Watts Halogen - Olympus TH4 power supply
% of light intensity
Time (sec)
Power Spectrum Amplitude log10 (V2/Hz)
10 point AA Smoothing of Data4_C
Frequency (Hz)
Data low-pass filtered at 1KHz
and acquired at 5 KHz on 4/6/2007
SD=0.0373
Metal Halide - EXFO
Halogen - Olympus
Counts/200 ms
SD=2.37
% of light intensity