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 2: Neural population recordings

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

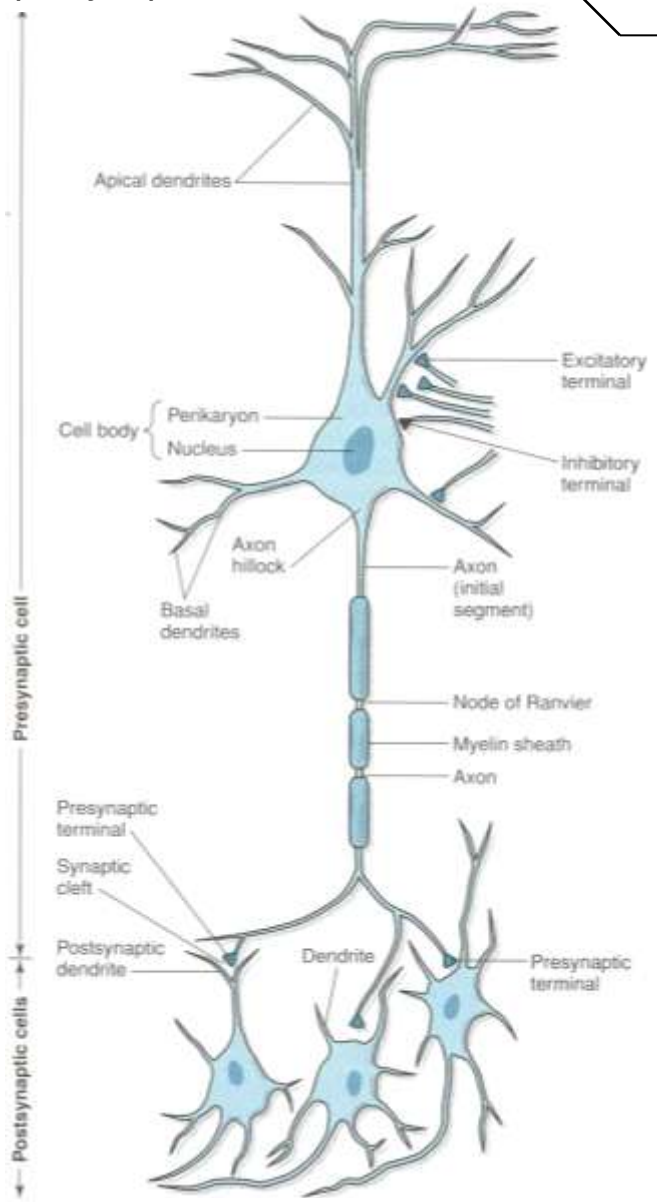
Student List

	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
	Yang, Dong	YangDong@uams.edu	Unable to attend	Pediatrics Pulmonary	Research Assistant – Accepted in Neuroscience

The synapse is a point of information processing

Greek, “connection, junction”

presynaptic neuron



postsynaptic neurons

An adult human brain contains ~ 10^{11} neurons = 100 billions,

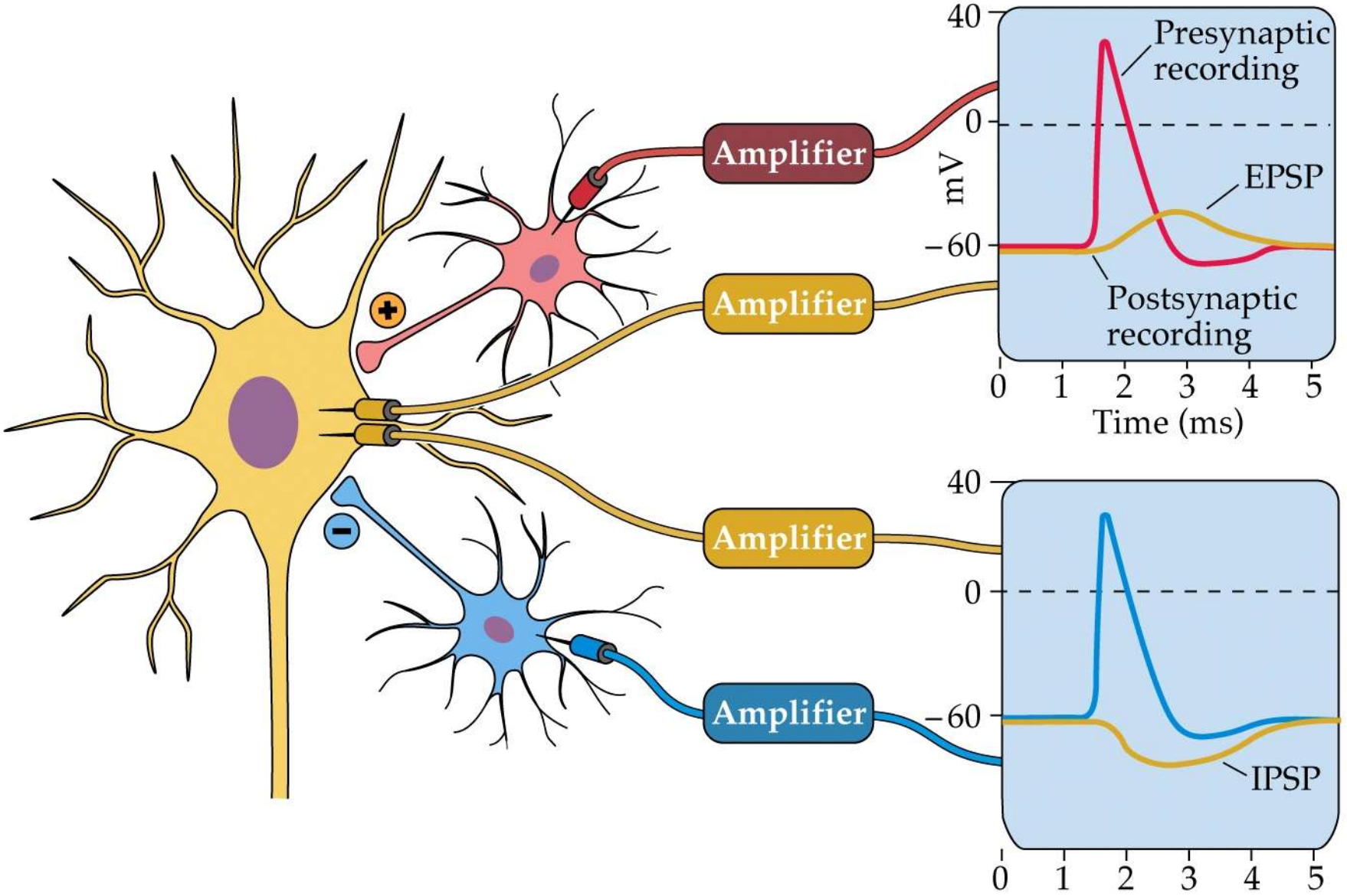
(there’s about 180 million more on the left side)

and each neuron might receive 10^3 synapses apiece, for a total of 10^{14} synapses.

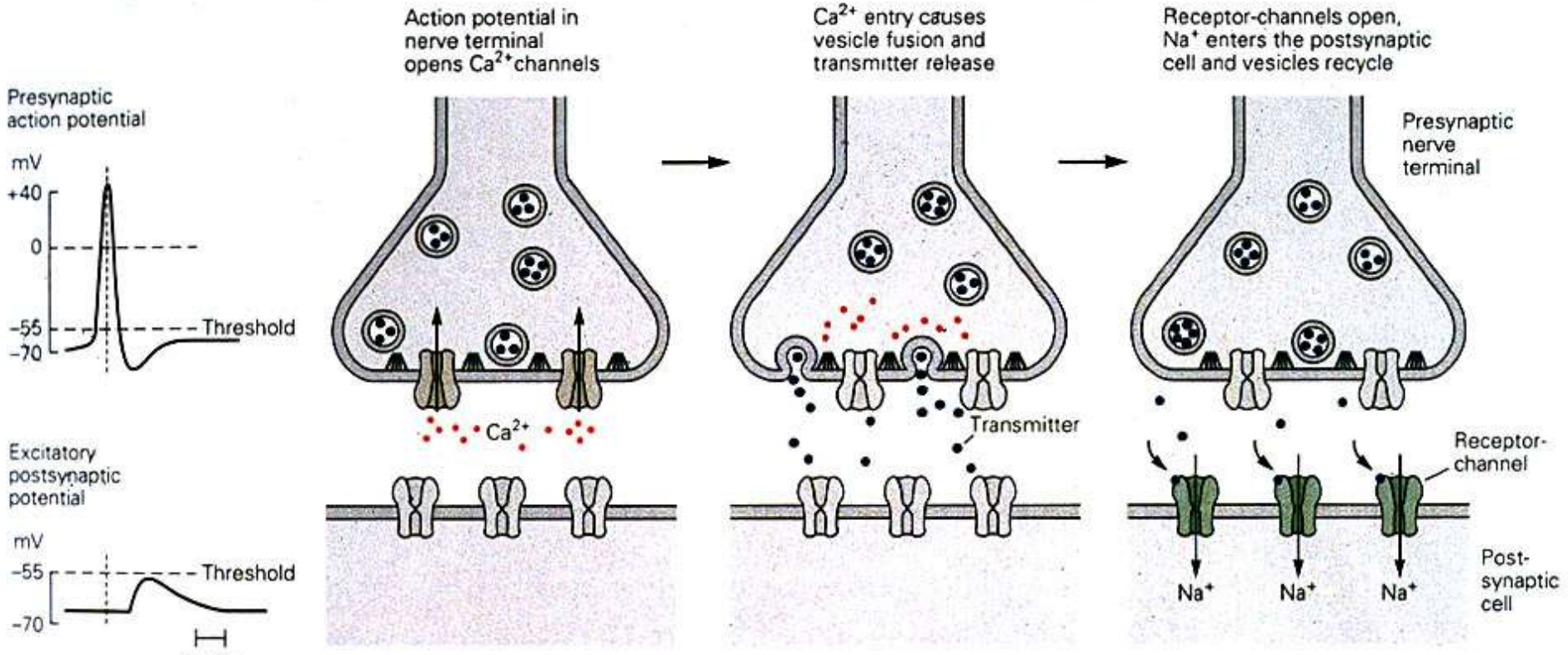
Most of these synapses form during the first 2 year of life.

Thus 10^{14}synapses/10^8 s = 10^6 synapses/s form in a fetus and infant!

Conduction velocity of action potential (2 - 400 km/hour)



Steps in Chemical Neurotransmission

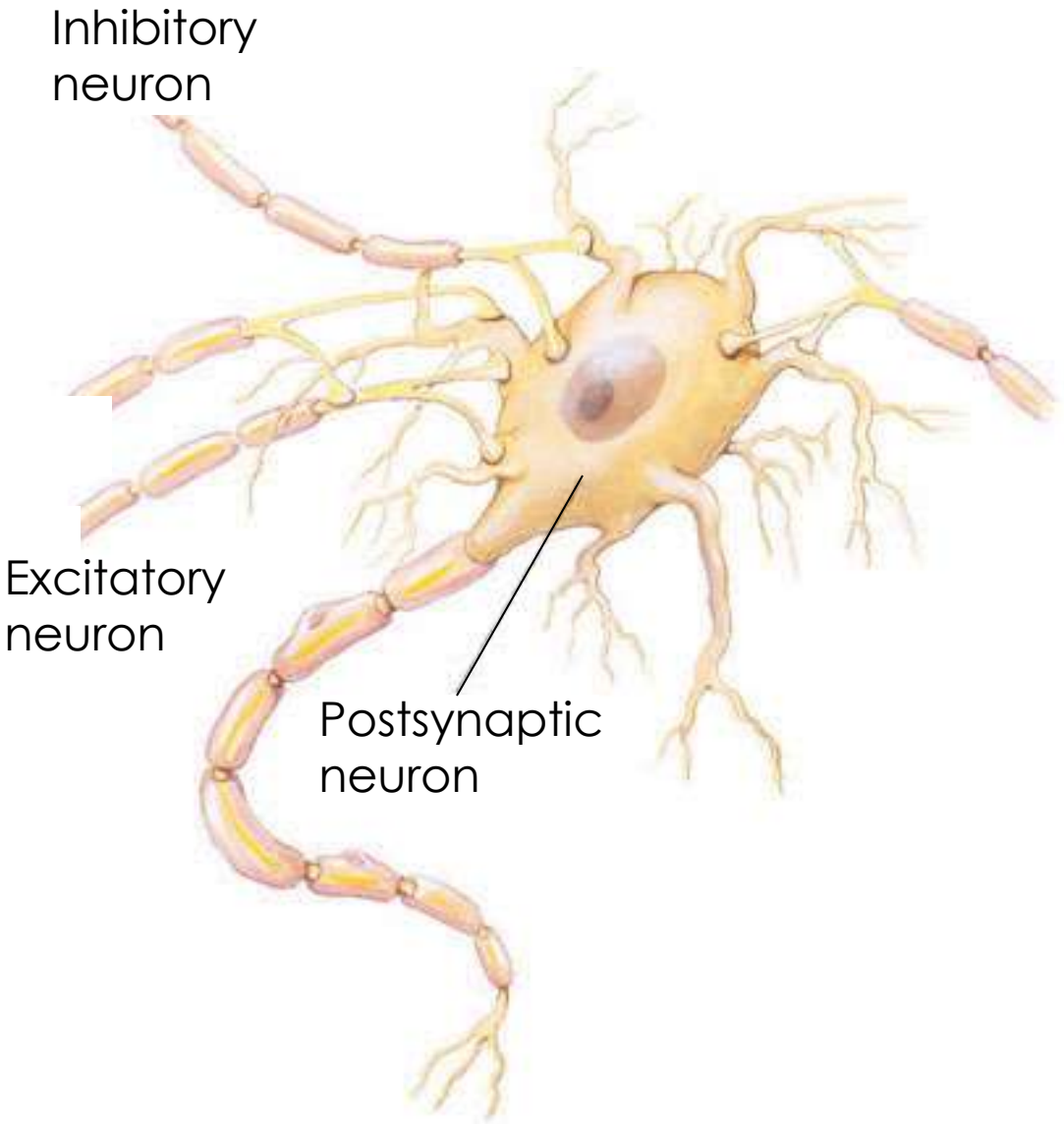


1. Action potential invades, and depolarizes, the nerve terminal.
2. Depolarization causes voltage-dependent Ca^{2+} channels to open and Ca^{2+} flows down its concentration gradient into the terminal.
3. Vesicles fuse with the presynaptic membrane and release their contents into the synaptic cleft (quantal release).
4. Neurotransmitter diffuses across the cleft and binds to its postsynaptic receptors.
5. Receptor activation leads to opening of associated ion channel and alters membrane potential of postsynaptic cell (for, e.g., ACh, GABA, glycine) or activation of certain enzyme systems (e.g., norepinephrine in the brain) that have a wide variety of effects not necessarily restricted to changes in membrane potential.
6. Transmitter dissociates from receptor.
7. Transmitter is inactivated or cleared from the synaptic cleft.

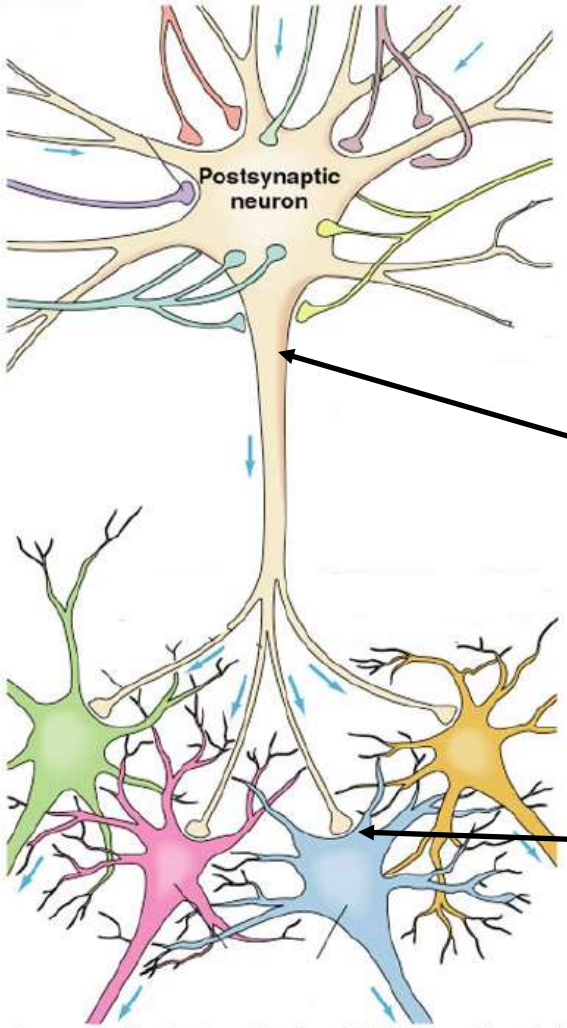
a_neuro1c synapse.mov

Integration

Many different axons synapse with the cell body of the post-synaptic neuron. The summed influence of their input determines whether or not an action potential will be sent down the axon extending below.



Convergence, Integration & Divergence



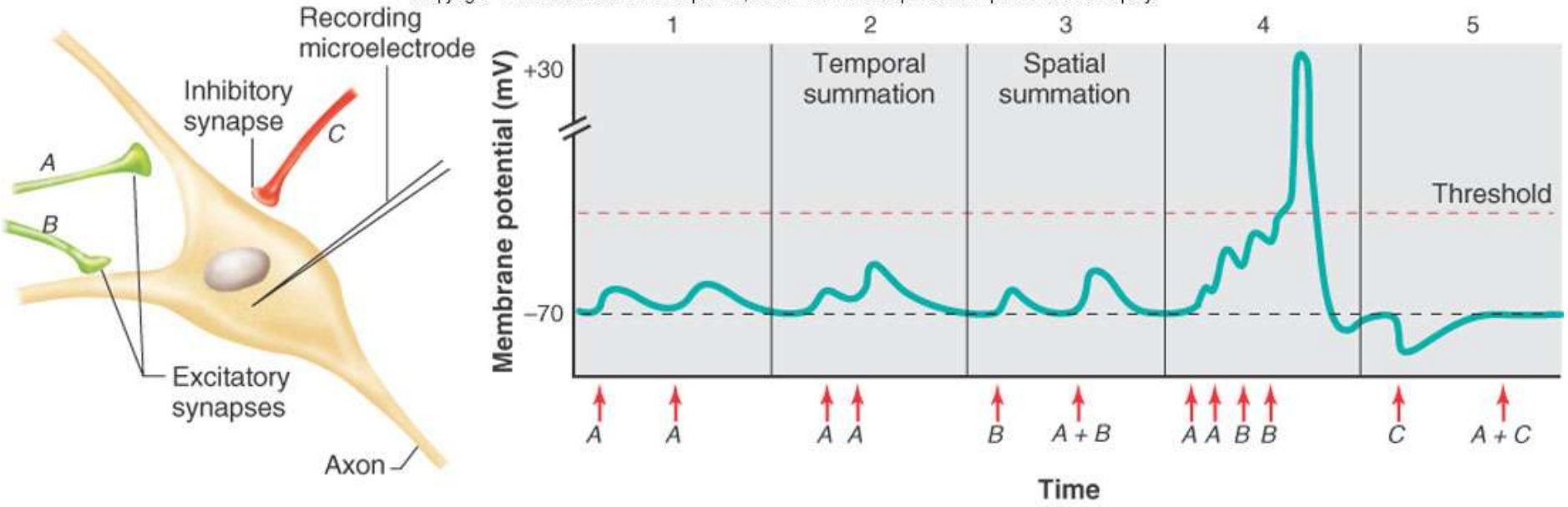
Arrows indicate direction in which information is being conveyed.

A single neuron receives thousands of inputs from other neurons = CONVERGENCE

A single neuron 'Integrates' all these inputs

A single neuron synapses with thousands of other neurons = DIVERGENCE

Spatial and Temporal Summation of PSPs

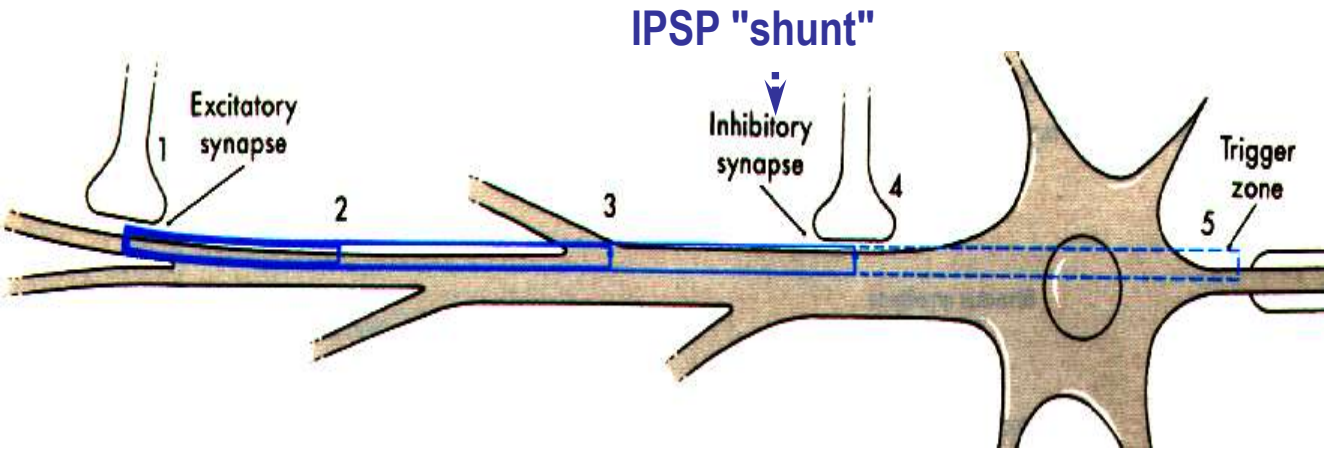


Spatial Summation Occurs when synaptic potentials arising from different synapses spread through the dendritic tree, to produce a new effect on the cell that is different than either would produce alone.

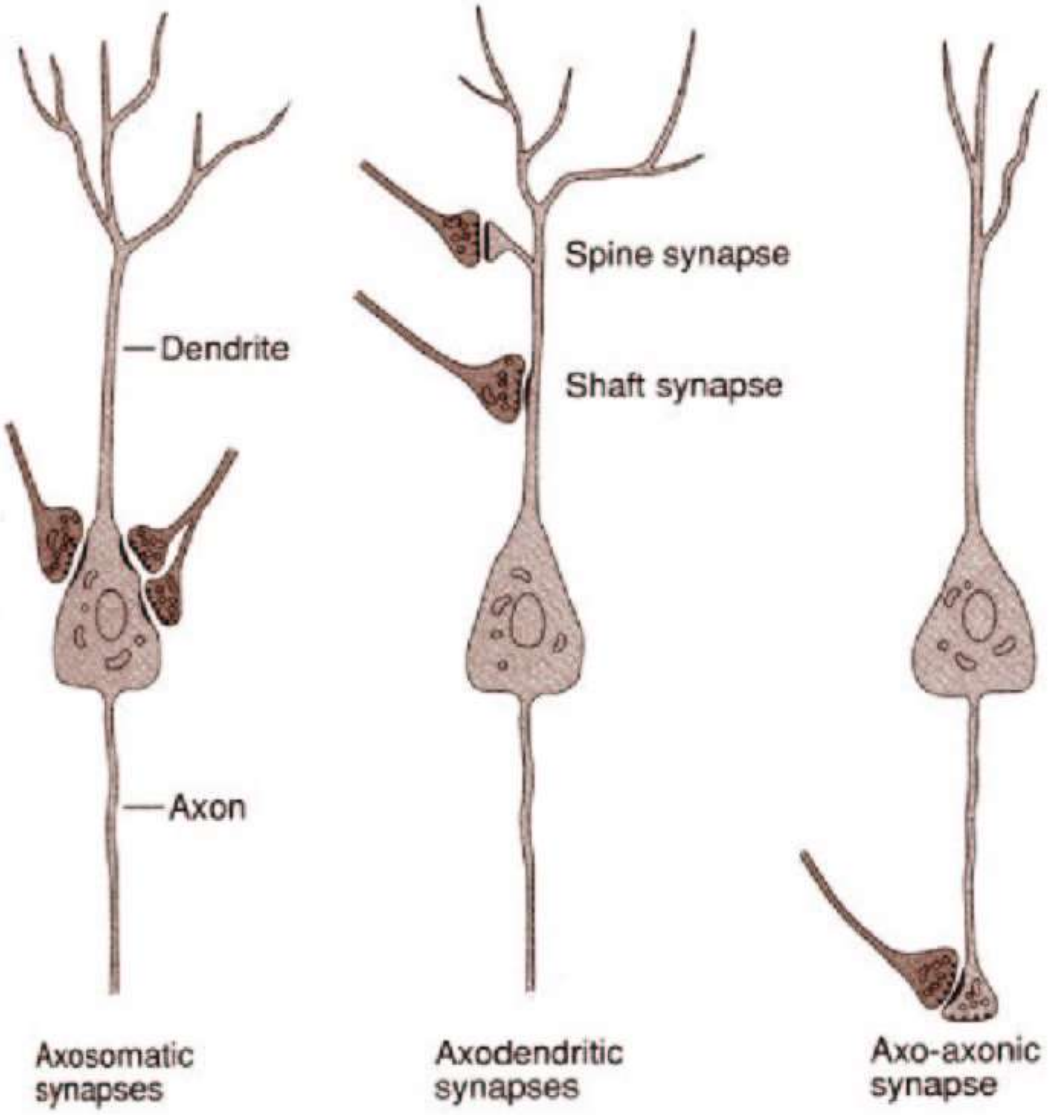
Temporal Summation Occurs when postsynaptic potentials arise at the same synapse in such rapid succession that the effects become additive.

- IPSPs closer to the trigger zone can "shunt" EPSPs

- Effectiveness therefore depends on synapse *location*

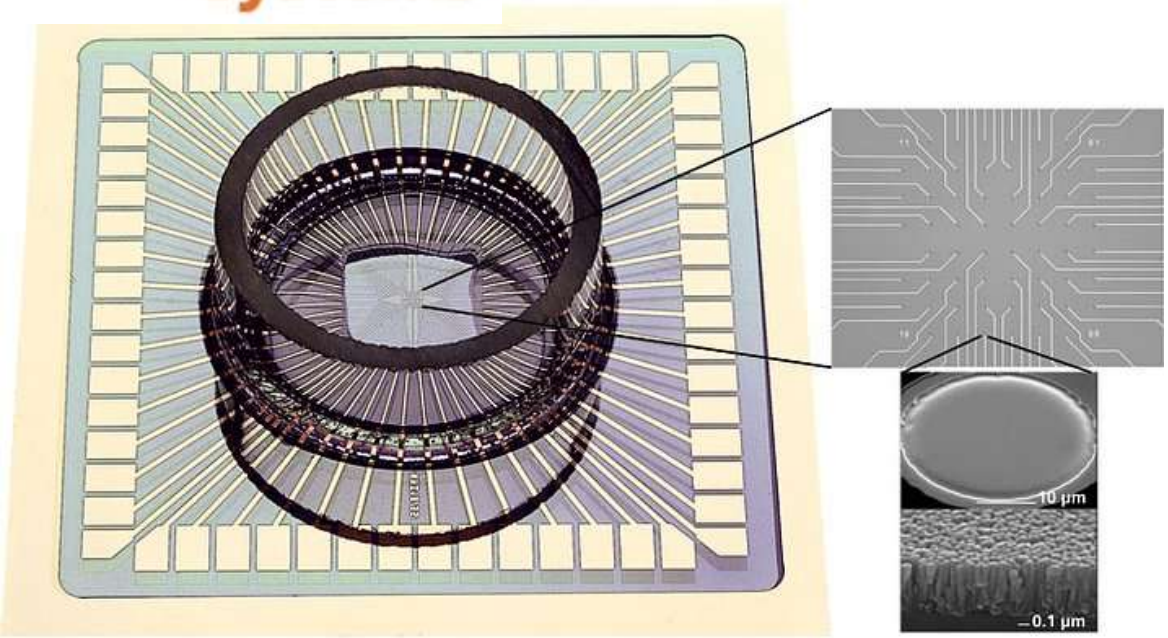


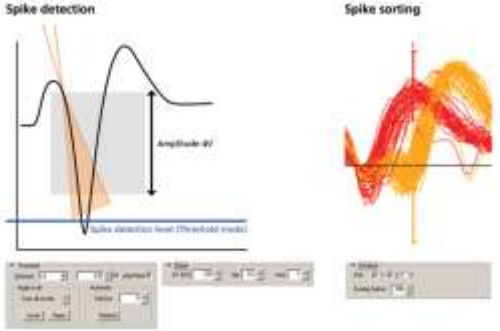
Types of Synaptic Contacts

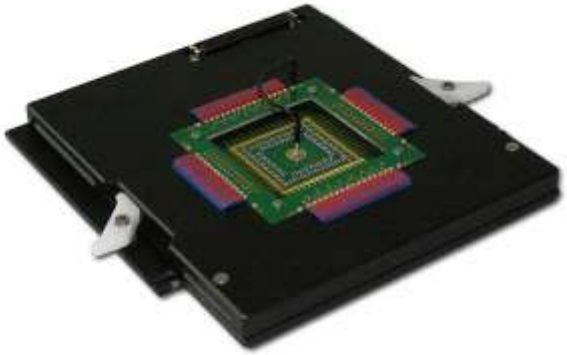


multichannel systems

In vitro Microelectrode Arrays

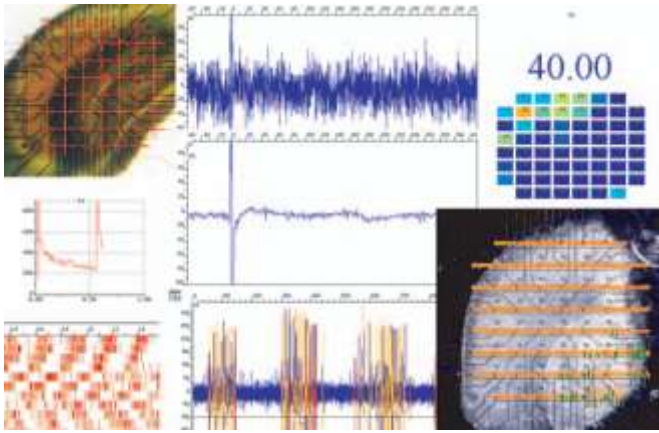






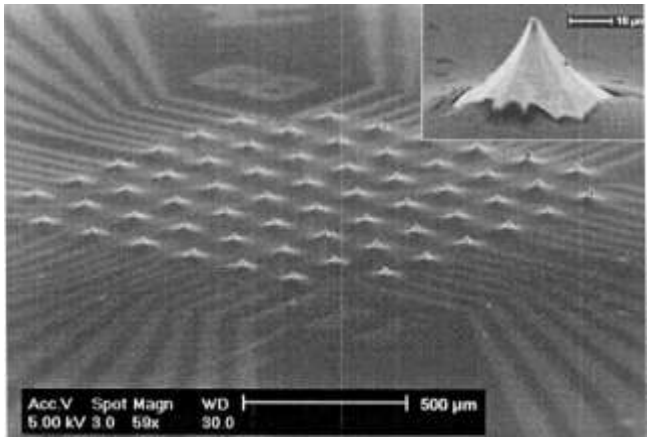
Amplifier

Data Acquisition System

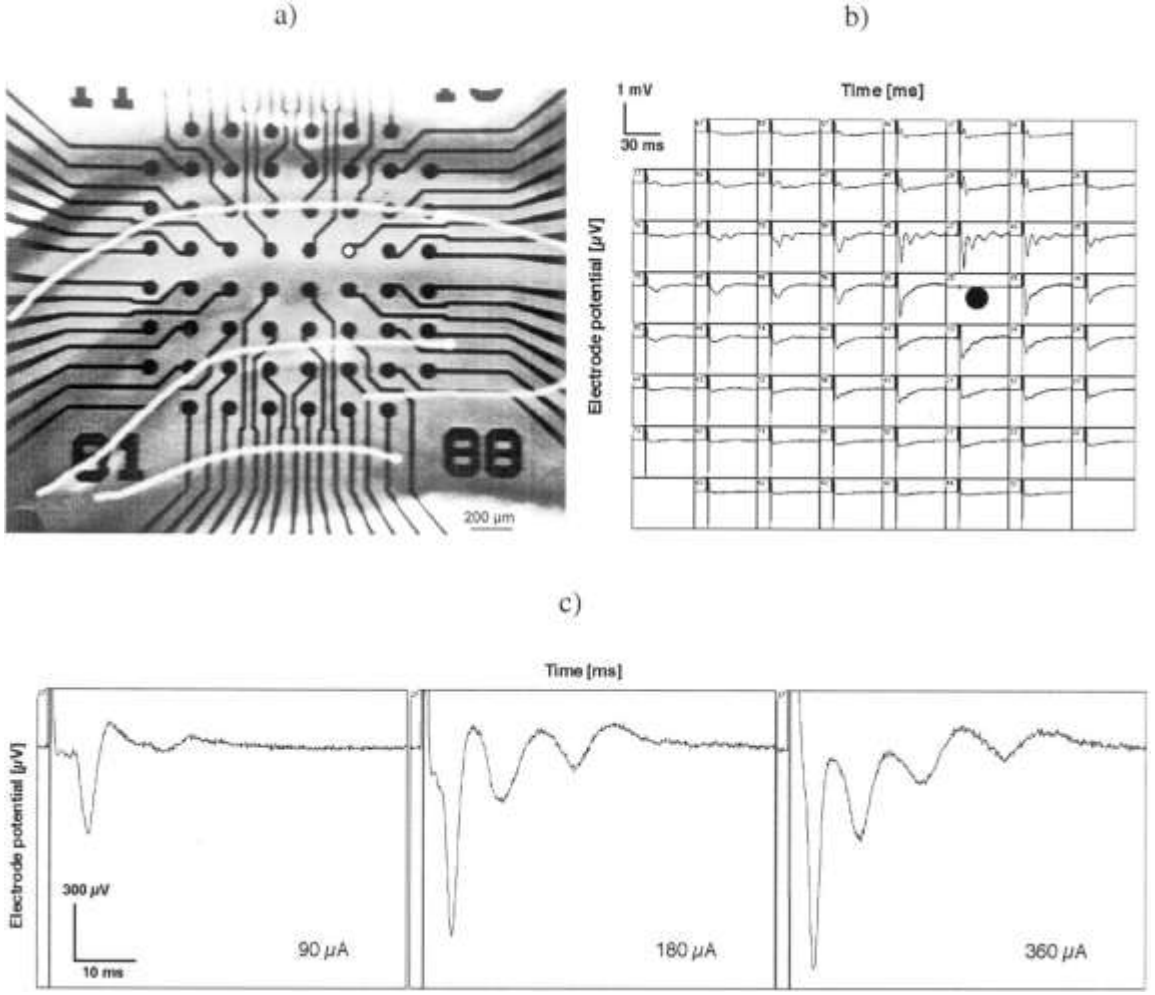


Software

A three-dimensional multi-electrode array for multi-site stimulation and recording in acute brain slices (Heuschkel et al. 2002)



SEM picture of 3D MEA recording area. It is composed of 60 tip-shaped protruding platinum electrodes. The height of the glass tips is about 60 µm, however, only 40 µm at the top of the tips form the effective recording electrodes. A thin epoxy layer insulates the electrode leads.



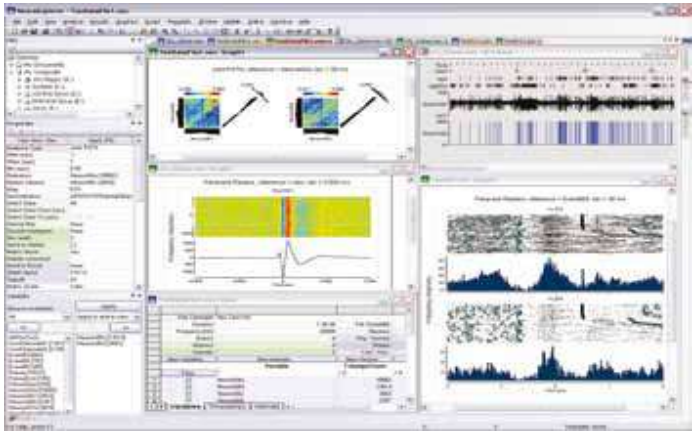
Recording of field potentials in an acute hippocampal slice. A slice was placed onto the electrodes (no nylon mesh was necessary to hold the slice in place) and current pulses were applied through one MEA electrode to locally stimulate the slice. The other electrodes were used for extracellular recording of electrical activity in the slice. (a) Picture of a hippocampal slice on a 3D MEA. The stimulation electrode is shown in white. (b) Responses evoked in the shown hippocampal slice by −/+ 360 µA, 120 µs lasting stimulation pulse on Schaffer collateral fibers in the CA1 region (black dot). In each case, the signal collected at the corresponding electrode is represented. (c) Evoked responses obtained at the electrode above the stimulation electrode (case no. 27) located in CA1 region, 200 µm away stimulation electrode, when applying −/+ current pulses having amplitudes of 90, 180 and 360 µA, respectively, and lasting 120 µs. Note that the evoked population spike was multiple.

In vivo Microelectrode Array Systems

Data Acquisition and Recording

Miniature Preamplifiers

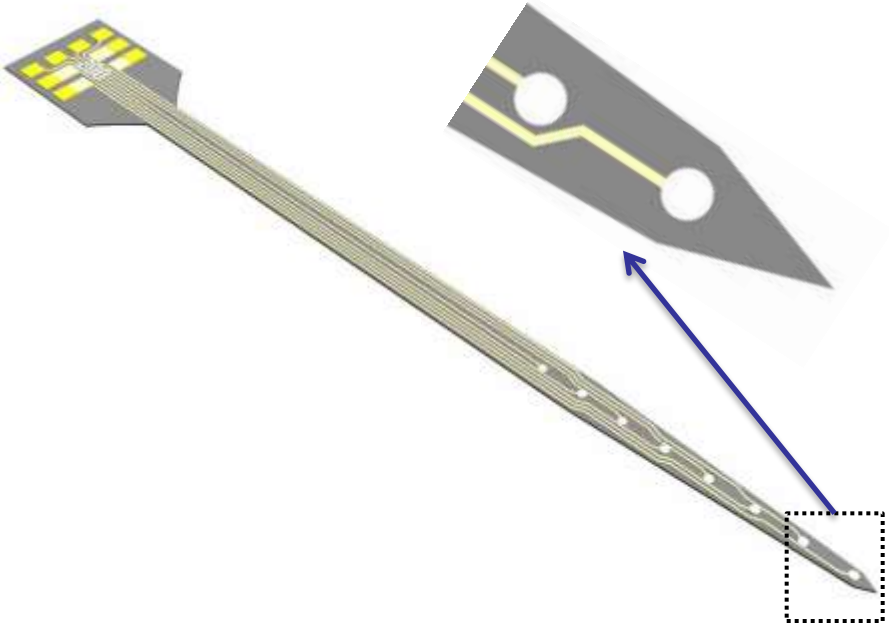
Microelectrode array

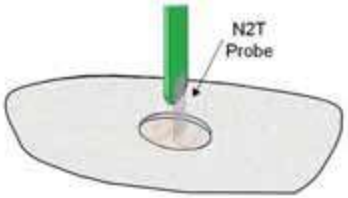


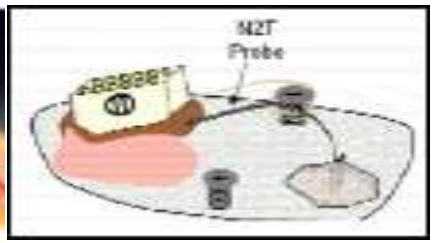
NeuroExplorer Software: This analysis software package is for statistical analysis of spike trains and other event data. It features standard histogram, raster, and correlational analyses for both point events (e.g. spikes, behavioral events) and continuous variables (e.g. field potentials), joint PSTH, burst analysis and many more.

ME-System Adapters

Silicon Microelectrode array

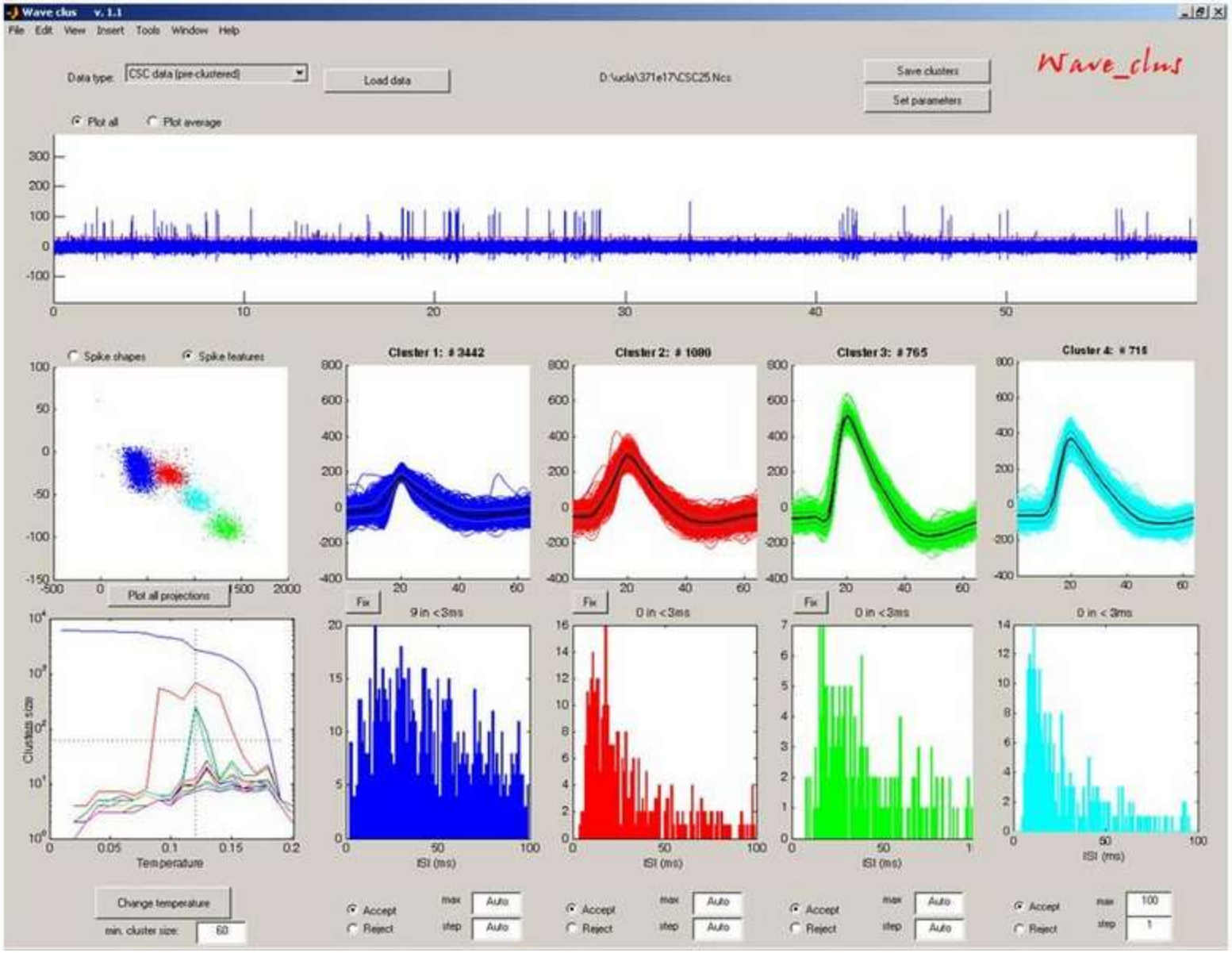




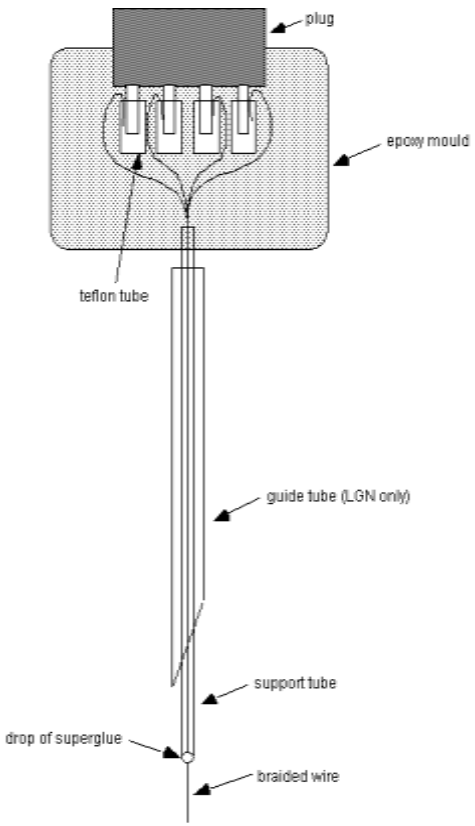


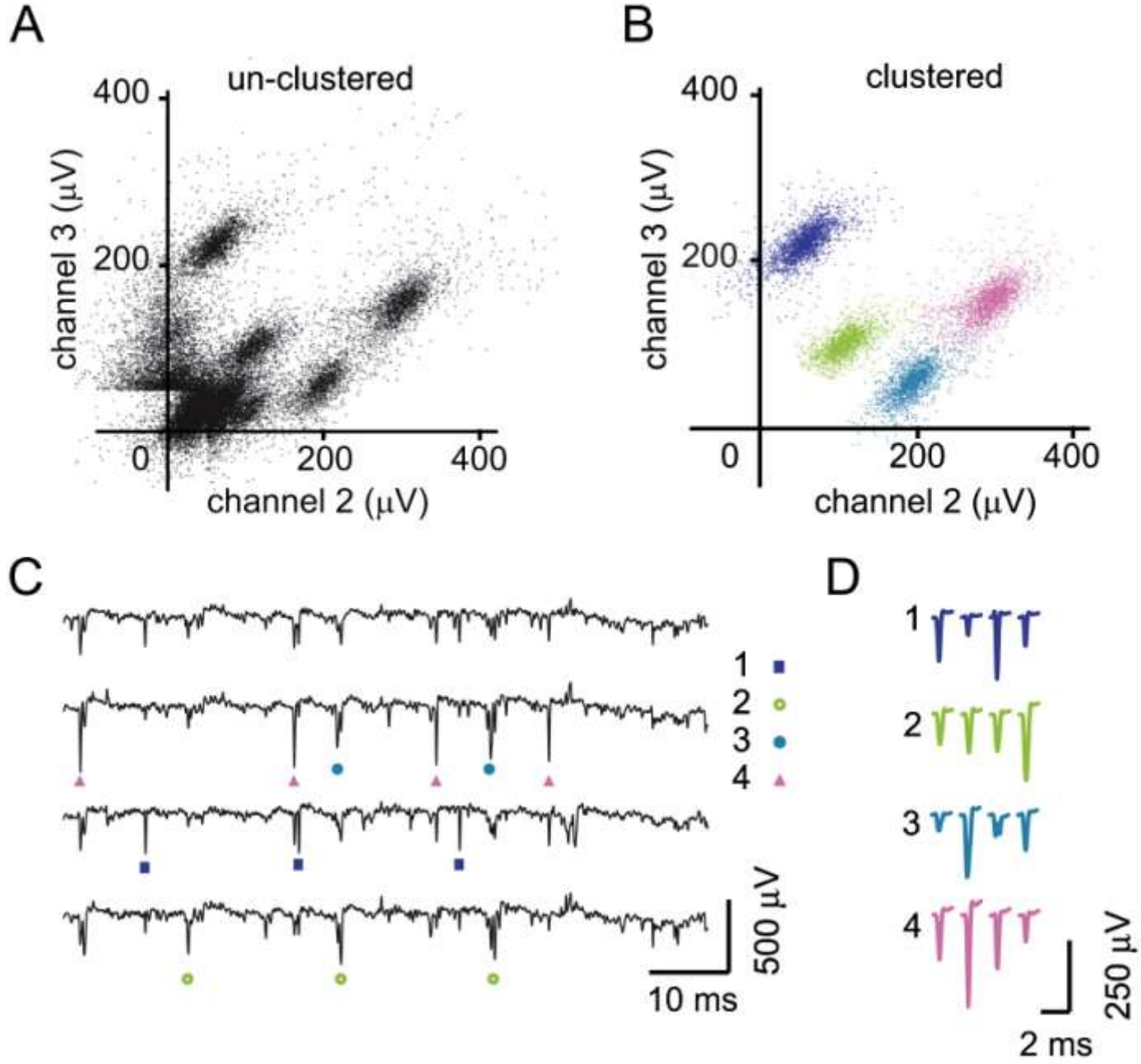
NeuroNexus Technologies
Making the connection

Spike Sorting



Simultaneous Recordings of Multiple Cerebellar Purkinje Cells with Tetrodes





de Solages et al. 2008

Spontaneous Local Field Potential (LFP)

The local field potential is believed to represent the synchronized input into the observed area, as opposed to the spike data, which represents the output from the area. In the LFP, quick fluctuations in the potential difference are filtered out, leaving only the slower fluctuations. The quick fluctuations are caused by the short inward and outward currents of the action potential. Therefore the action potential plays no part in the LFP. The LFP is thus composed of the more sustained currents in the tissue, typical of the somato-dendritic currents. The major slow current is the postsynaptic potential (PSP). It was thought until recently that EPSPs and IPSPs were the exclusive constituents of LFPs.

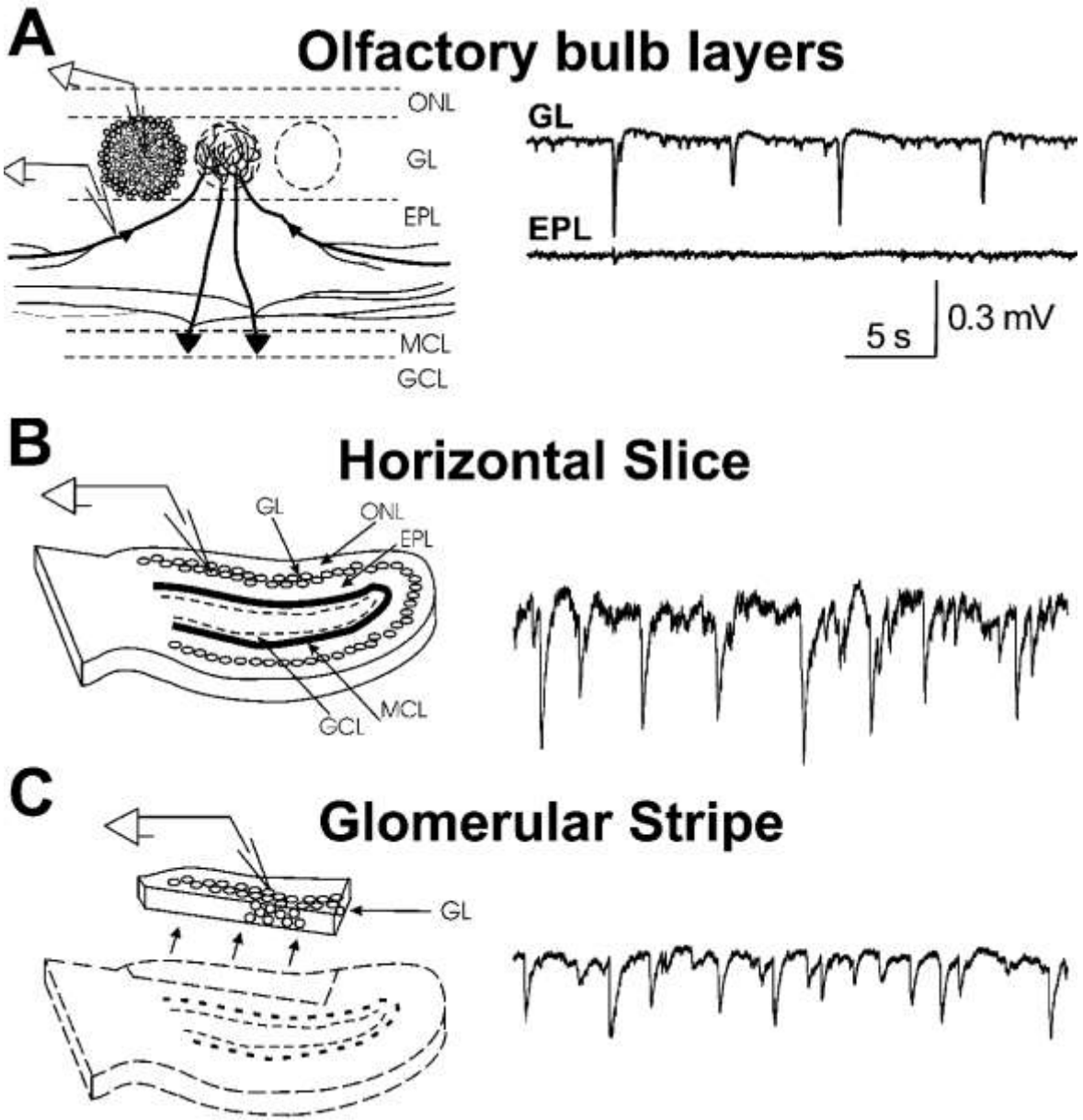
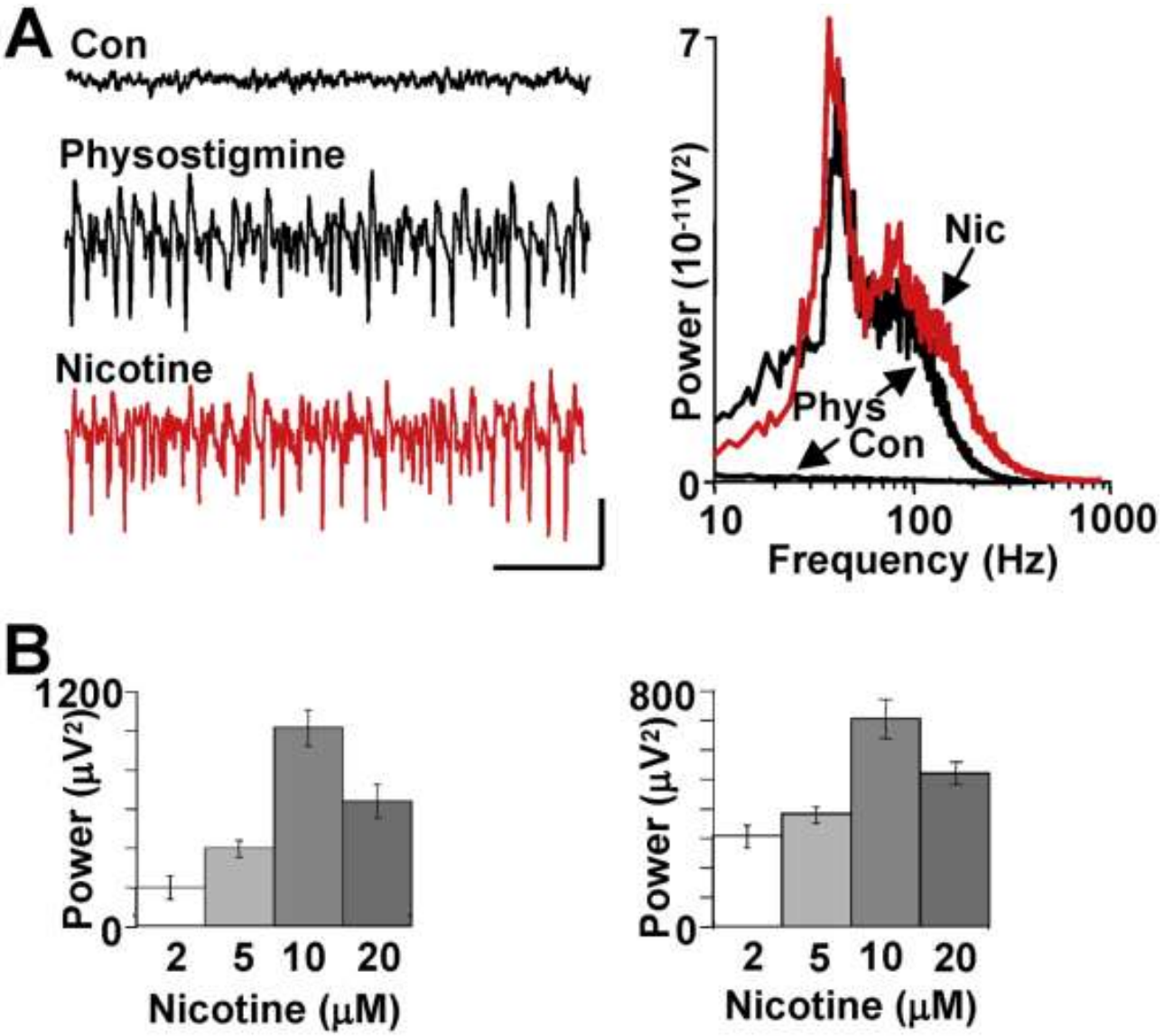


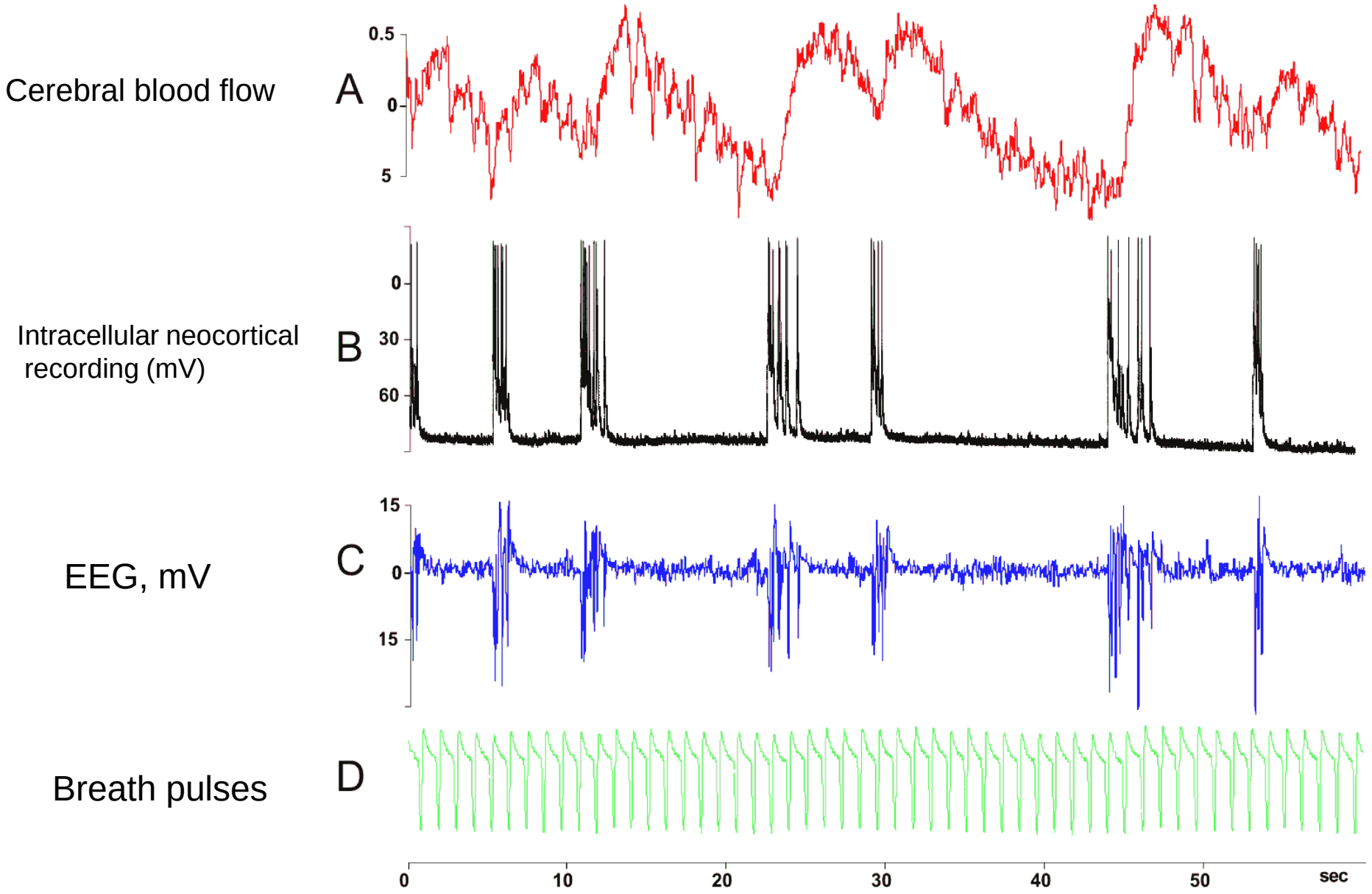
Figure 1. High-Frequency Oscillations Are Induced by Nicotinic Receptor Activation and Do Not Require Ionotropic Glutamate Receptors



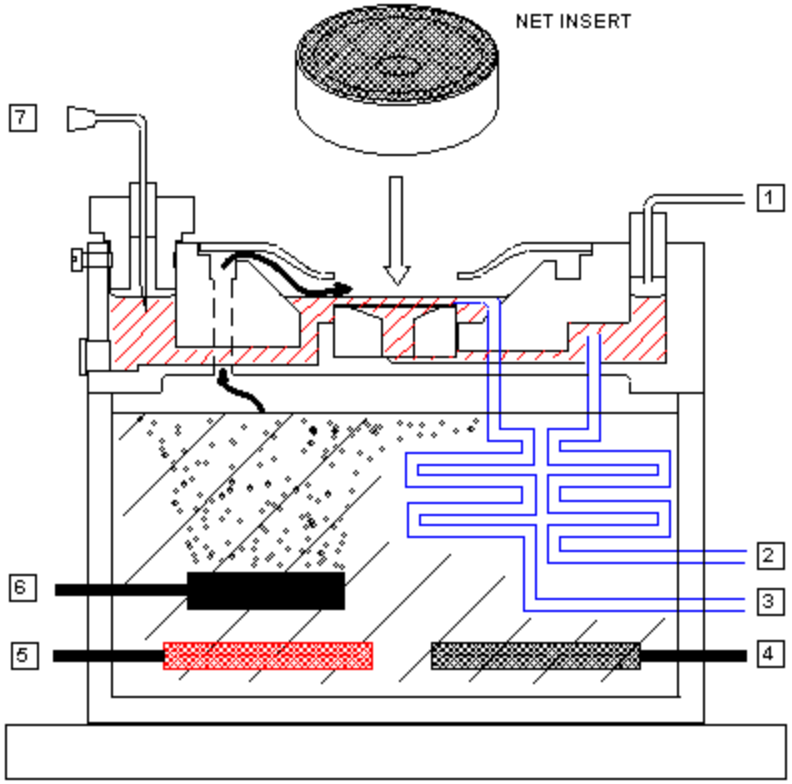
(A) Enhanced levels of endogenous acetylcholine with physostigmine (10 μM) generate concurrent gamma and VFO rhythms. Example traces show data from a field electrode in the Purkinje cell layer. The spectrum of frequencies generated by physostigmine (black line) was reproduced by nicotine (10 μM) alone (red line). Scale bars: 0.1 mV, 100 ms.

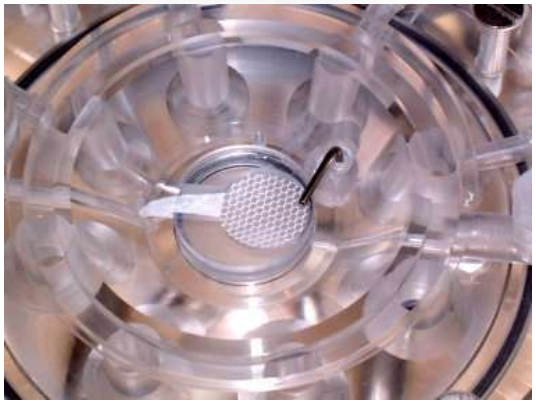
(B) Concentration response for nicotine on gamma rhythms (left) and VFO (right). Note that maximal power in either band was obtained with 10 μM nicotine.

Neuronal activity is synchronized with cerebral blood flow (CBF) under isoflurane anesthesia in a freely breathing rat.



Brain Slice Interface Chamber



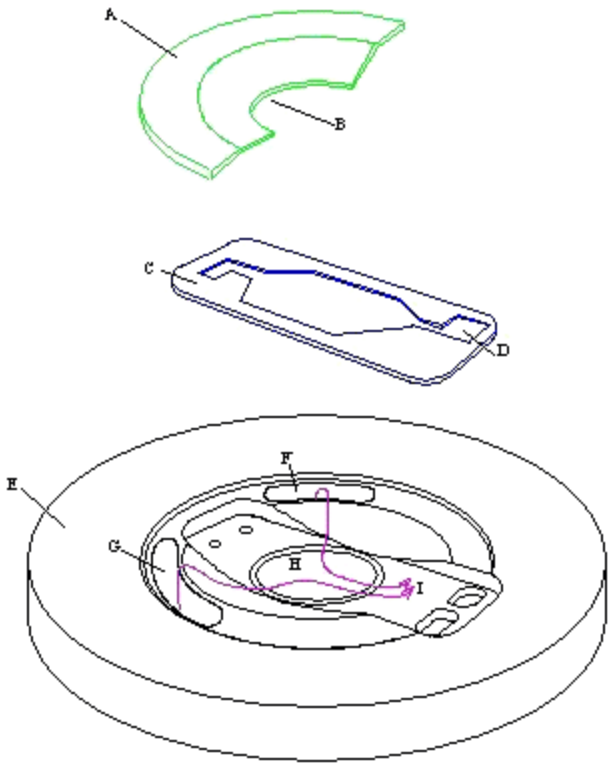


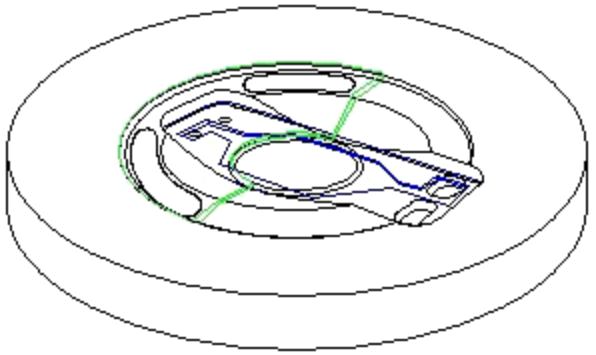
Parts Description

1. Bubble Trap
2. Heat Exchanger for Perfusion Fluid Submerged/Interface
3. Heat Exchanger for Perfusion Fluid to Sloped Insert
4. Control Temperature Sensor
5. Heating Element
6. Oxygen/Carbon Dioxide Gas Bubbler
7. Exit for Perfusion Fluid via Suction Line

AutoMate Scientific
READY FOR RESEARCH

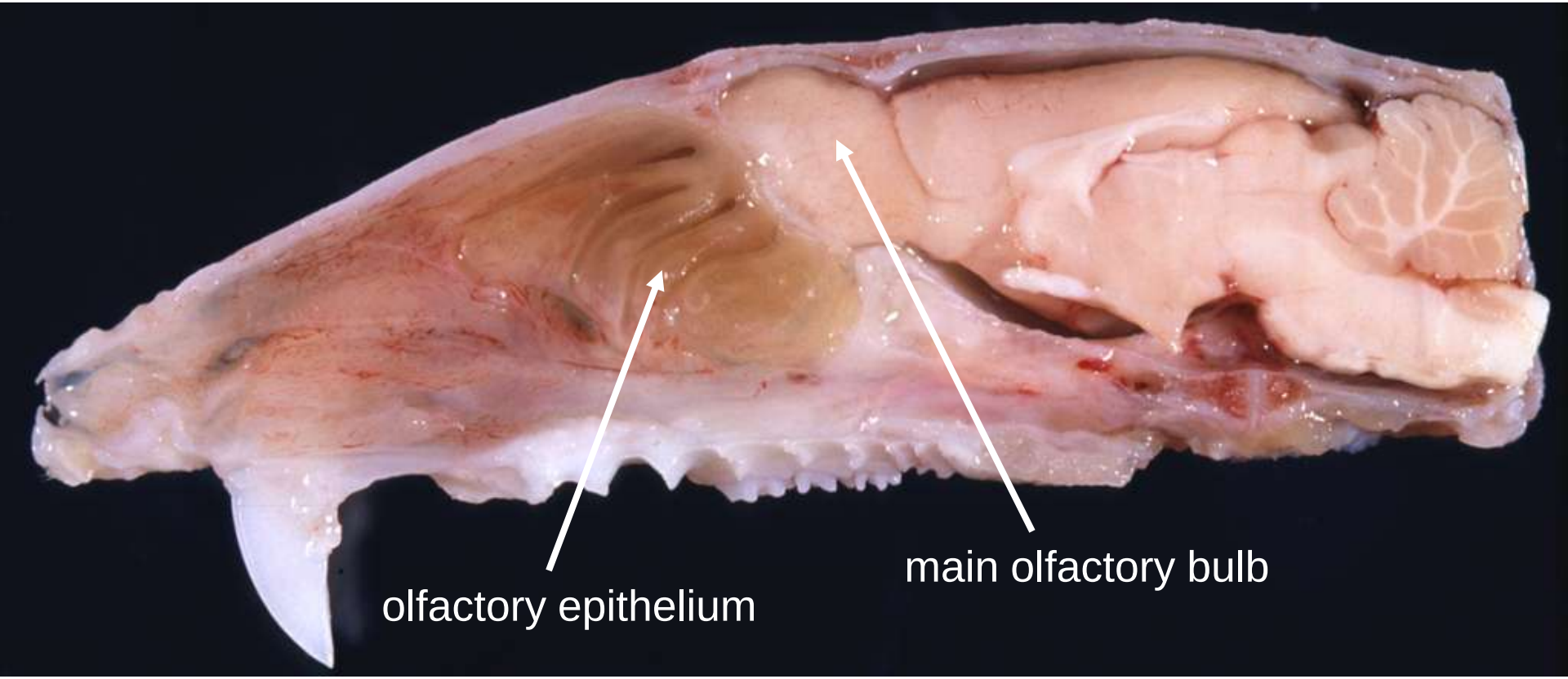
MSC3 - INTERFACE





The MS3 stage chamber allows interface methods of slice maintenance at room temperature on a compound microscope stage. Constructed from clear acrylic, it can be mounted in close proximity to the microscope condenser optics for optimal visualization. Slices rest in a flat channel with a 20mm central coverslip base. Fluid flows in a diamond-shaped channel above the slice with high oxygen tension above.

The Main Olfactory System



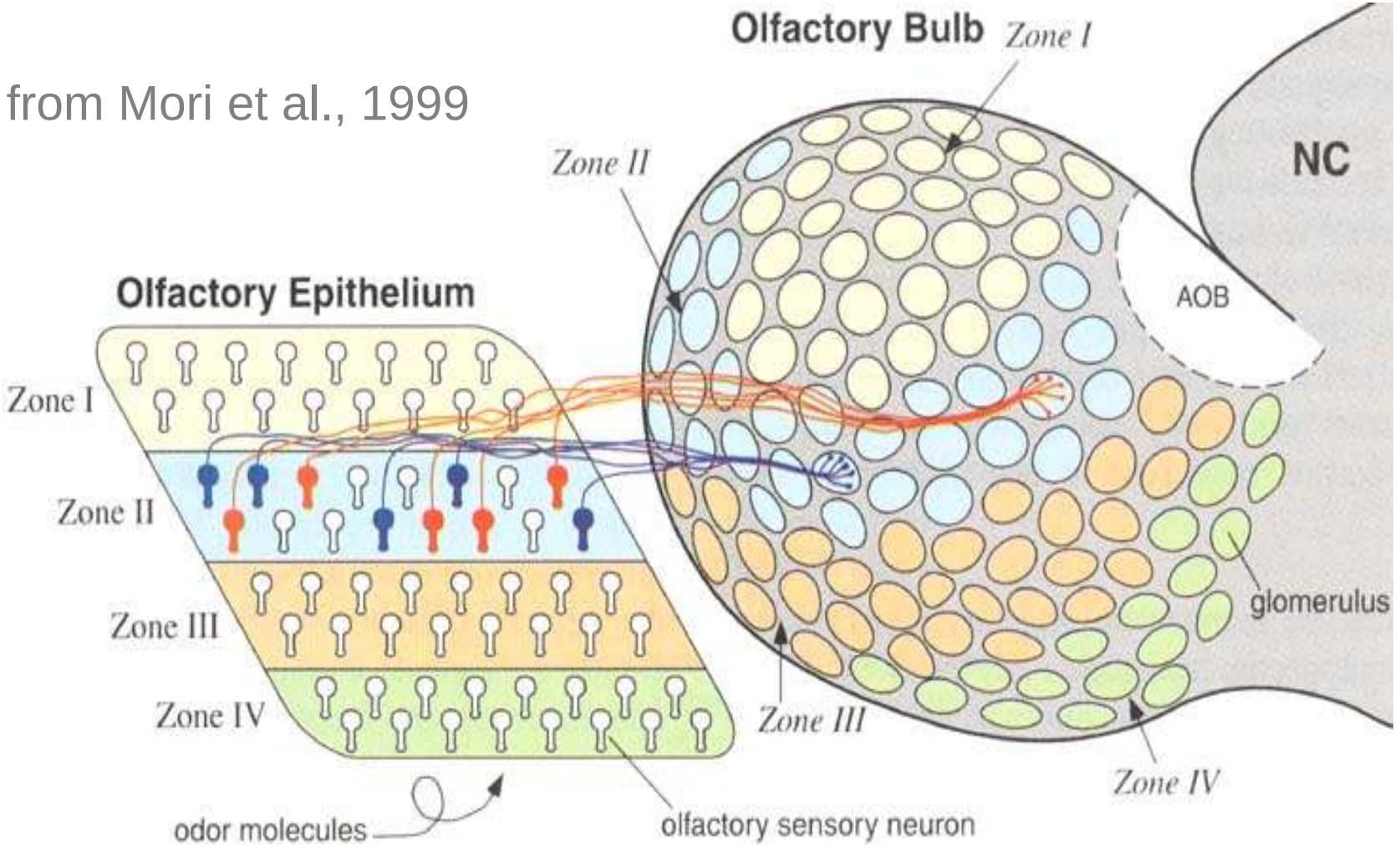
The sense of smell is mediated through the stimulation of olfactory receptors neurons (ORNs) by volatile chemicals. ORNs are contained in a neuroepithelium located at the top of the nasal cavity. Afferent information from these receptors is carried to the olfactory bulbs by the olfactory nerve, the first cranial nerve.

-Reproduction/maternal functions

-Recognition of predators and prey, food selection

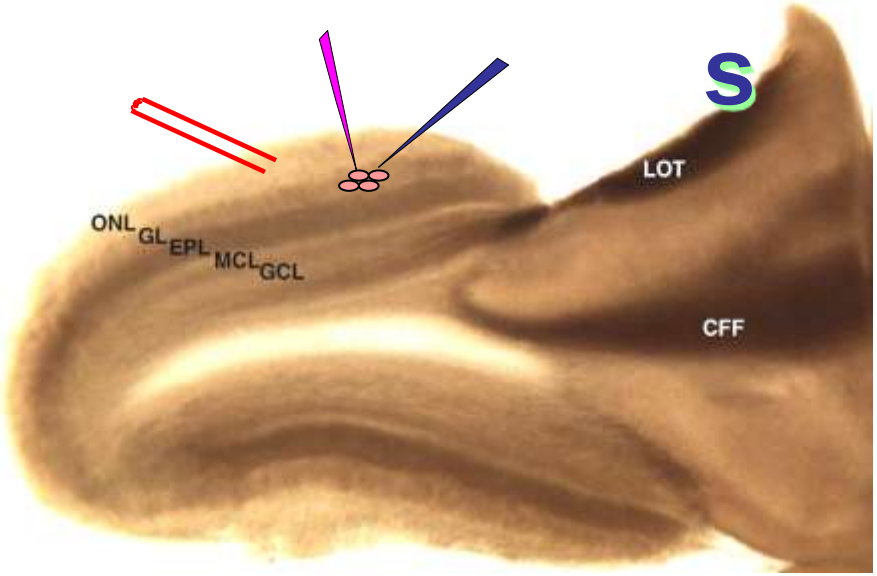
Projections from olfactory receptor neurons to the bulb

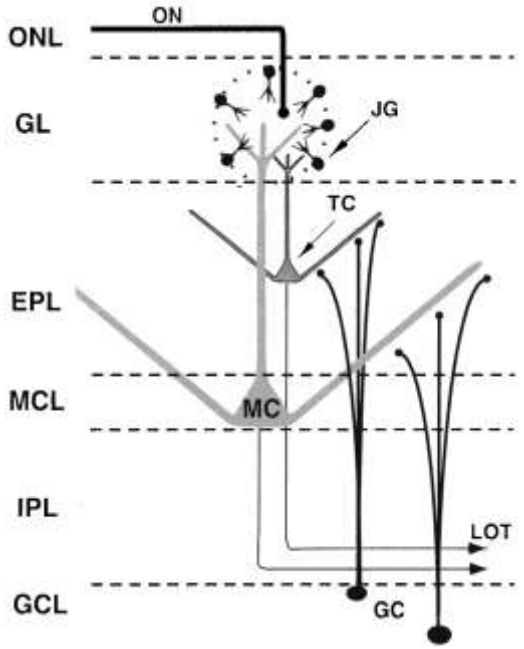
from Mori et al., 1999

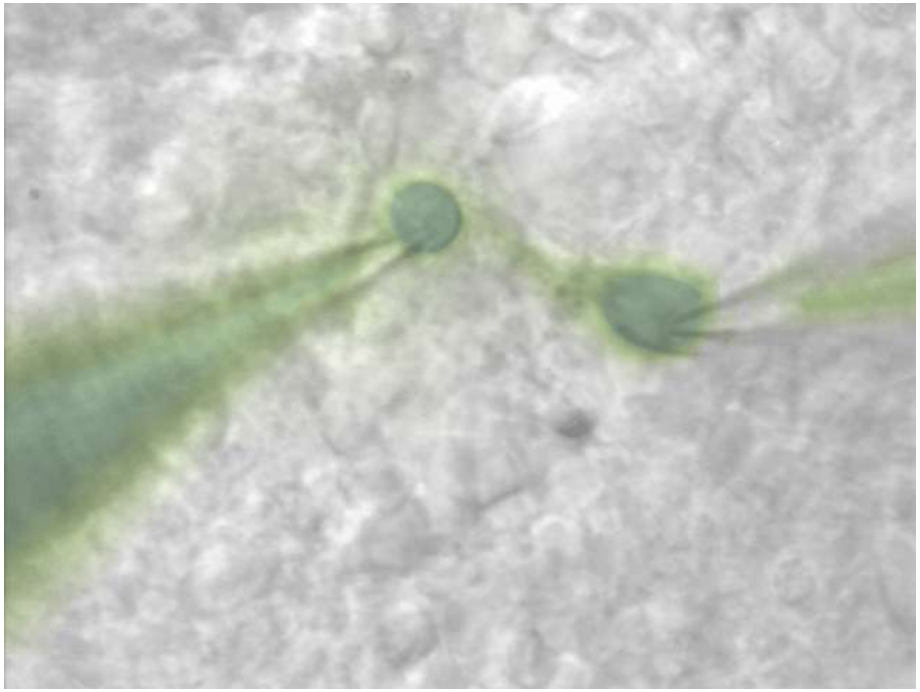


~1,000 olfactory receptor genes, ~1,000-2,000 glomeruli
~10,000 olfactory receptor neurons express the same receptor
Olfactory receptor neurons of the same type targets one or few, topographically-fixed glomeruli on each side of the main olfactory bulb

Methods







⇒ Olfactory bulb slices (400 µm thick) were cut from young rats (21- 30 days).

⇒ Single and paired whole-cell current and voltage clamp recordings were made at 30°C; drugs applied by perfusion.

⇒ Recorded neurons filled with Lucifer Yellow and biocytin and their morphology was reconstructed.

⇒ Olfactory nerve (ON) electrical stimulation.

The Olfactory Bulb Network

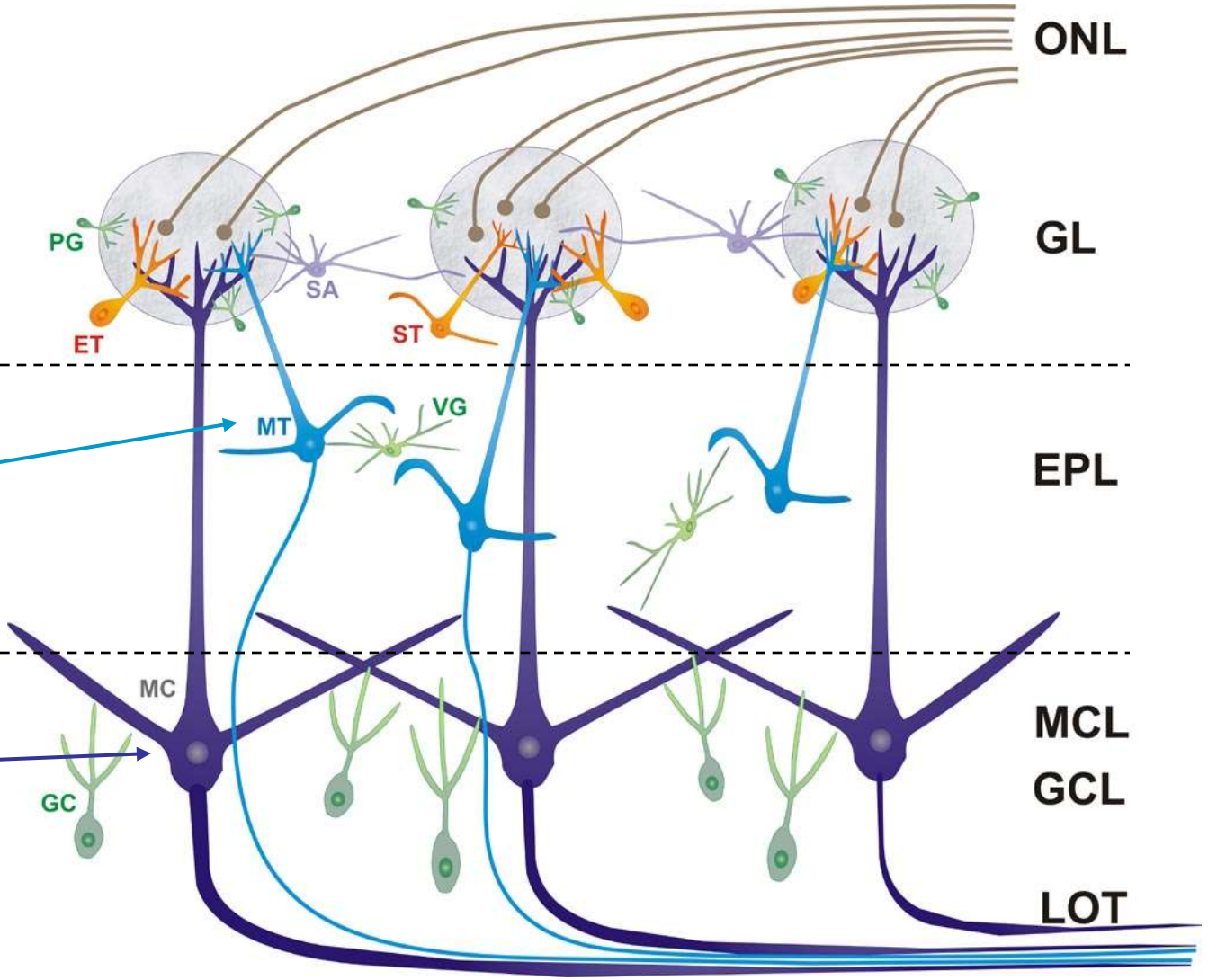
Each glomerulus is innervated by:

10,000 olfactory axons,

1000 juxtaglomerular cells

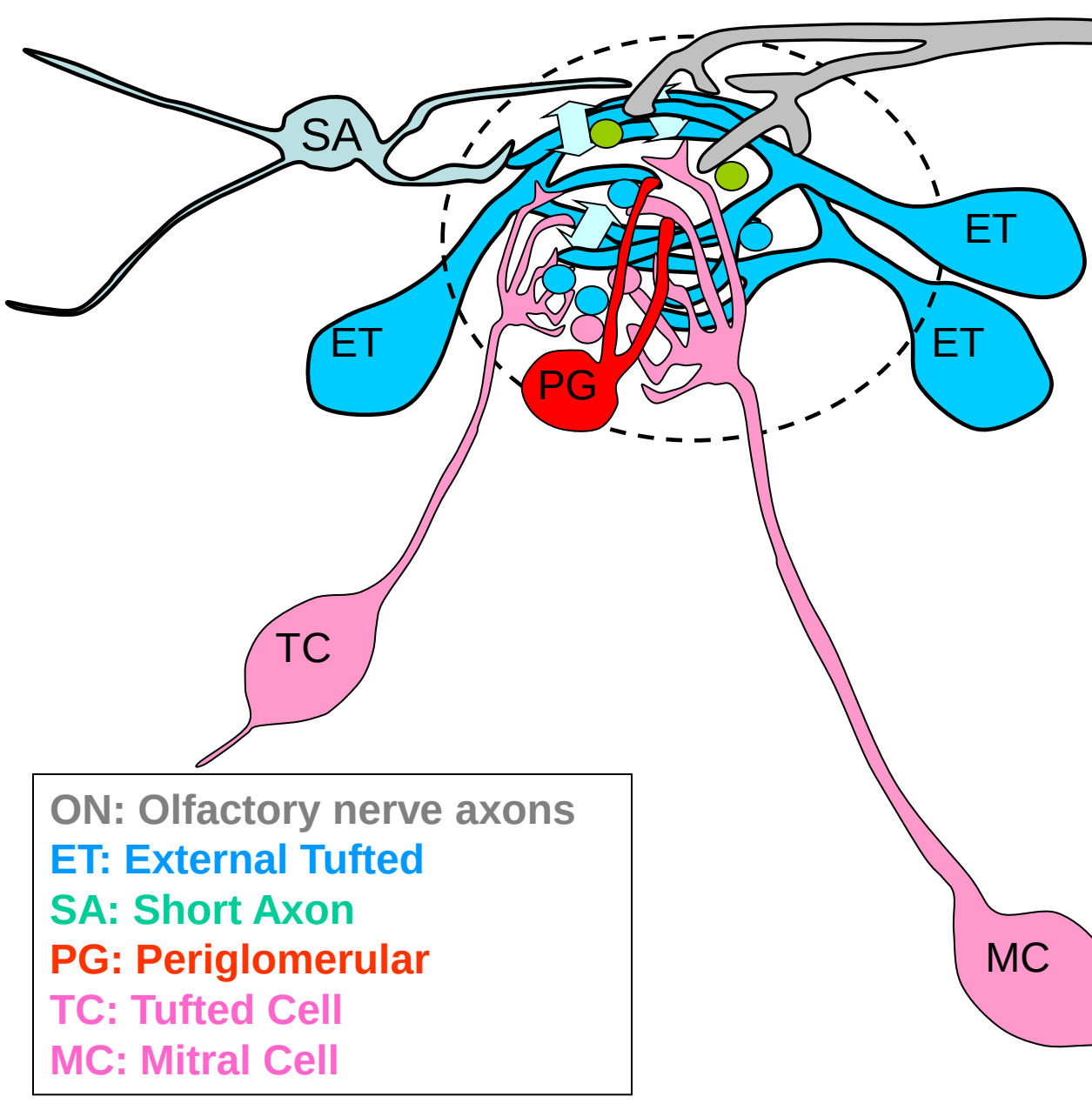
50 tufted cells,

25 mitral cells,

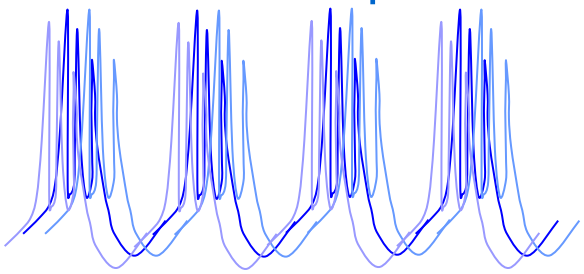


Thus, juxtaglomerular cells outnumber mitral/tufted cells by 20-40x

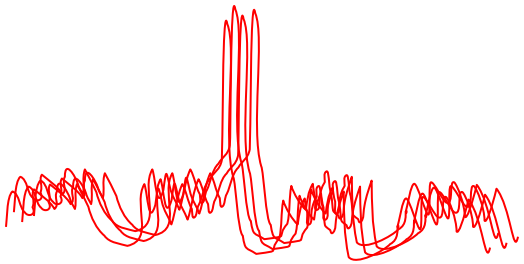
ET cells may represent the rhythm generator of the glomerular circuit



Bursts of spikes



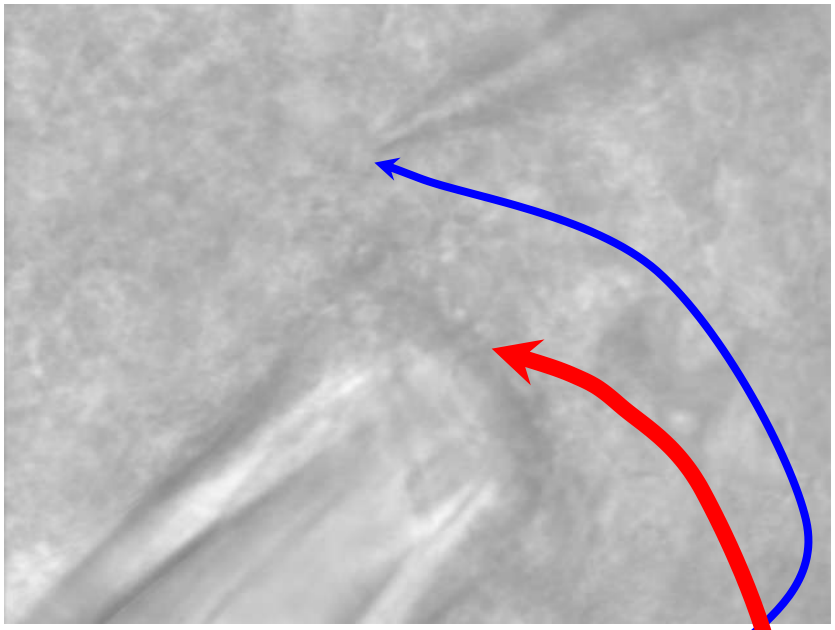
Bursts of EPSPs

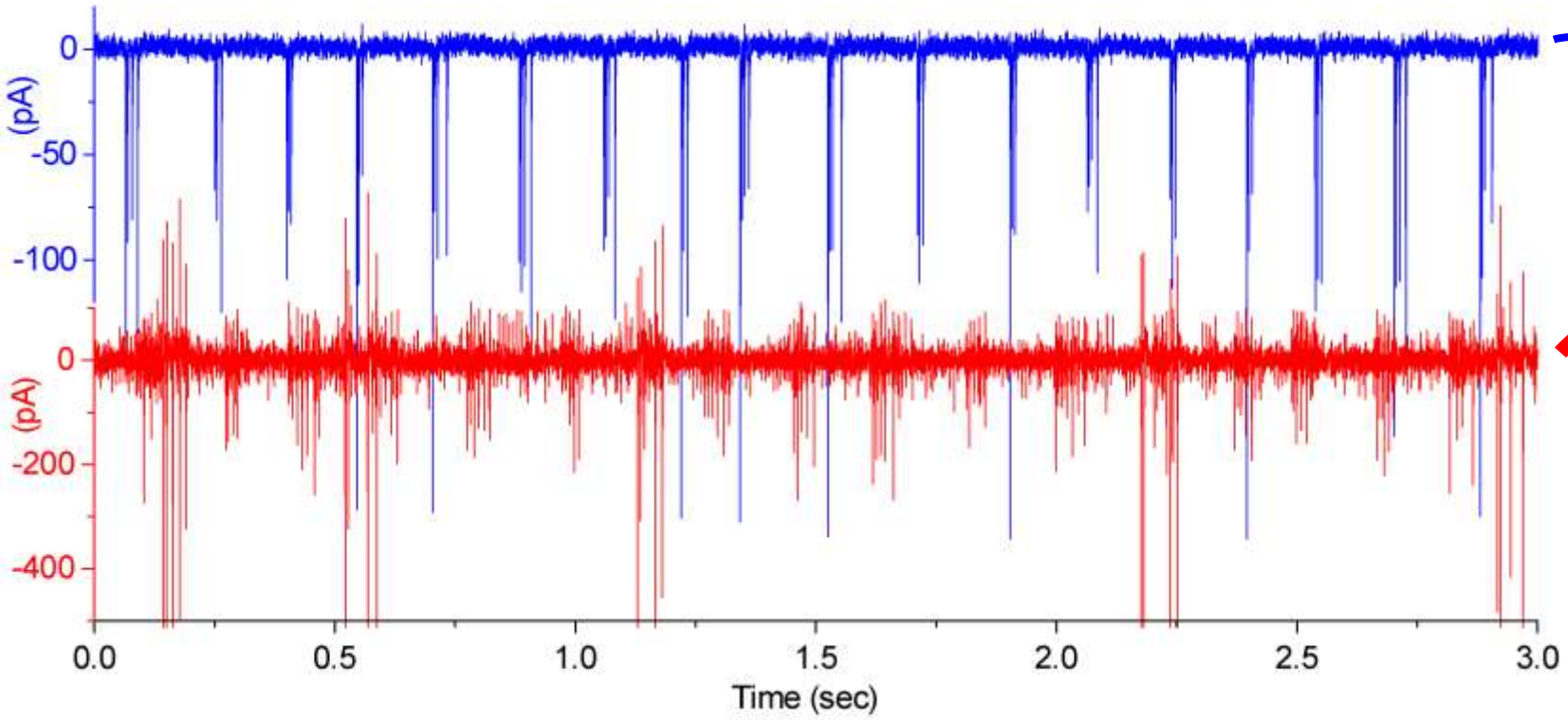


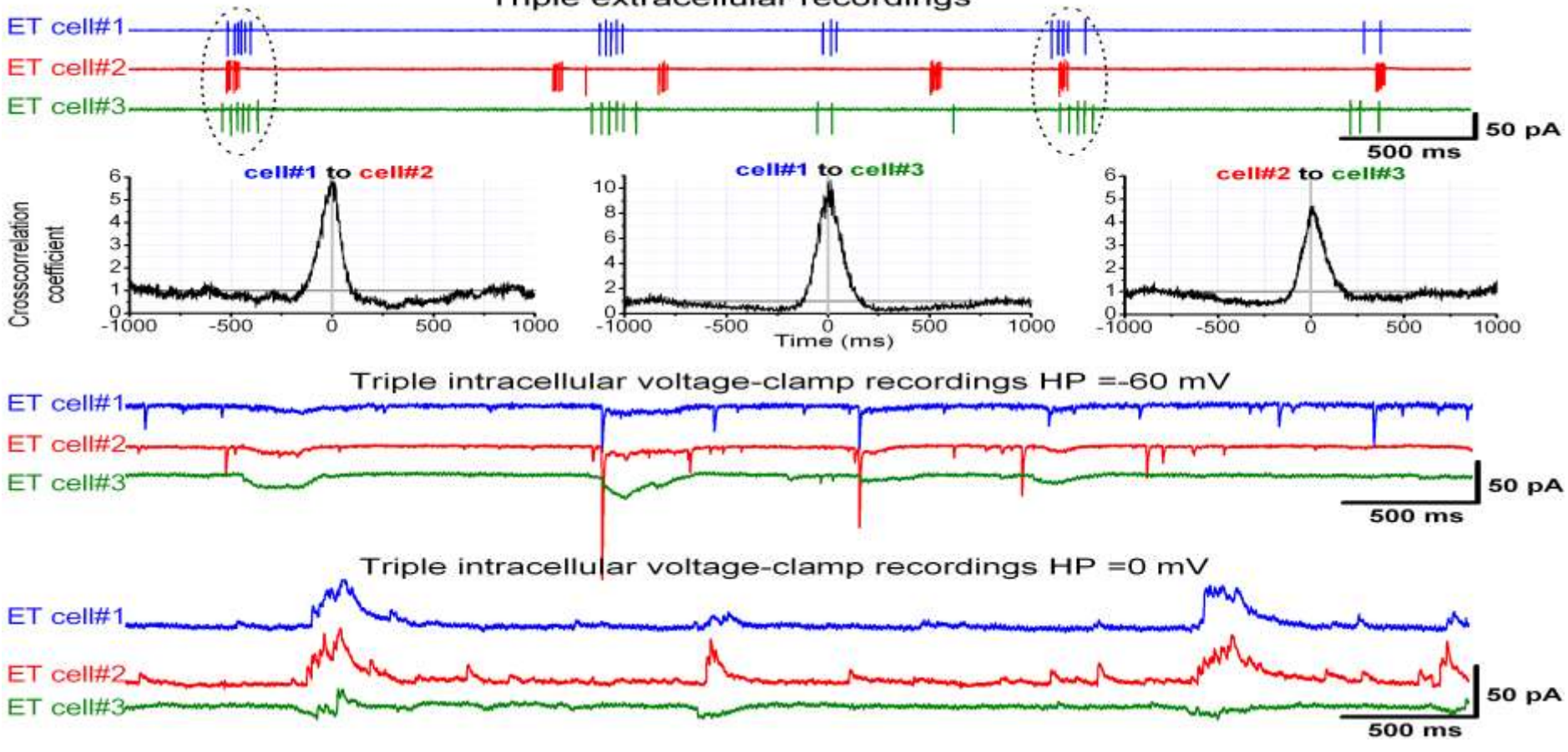
Long-lasting depolarization

ON: Olfactory nerve axons
ET: External Tufted
SA: Short Axon
PG: Periglomerular
TC: Tufted Cell
MC: Mitral Cell

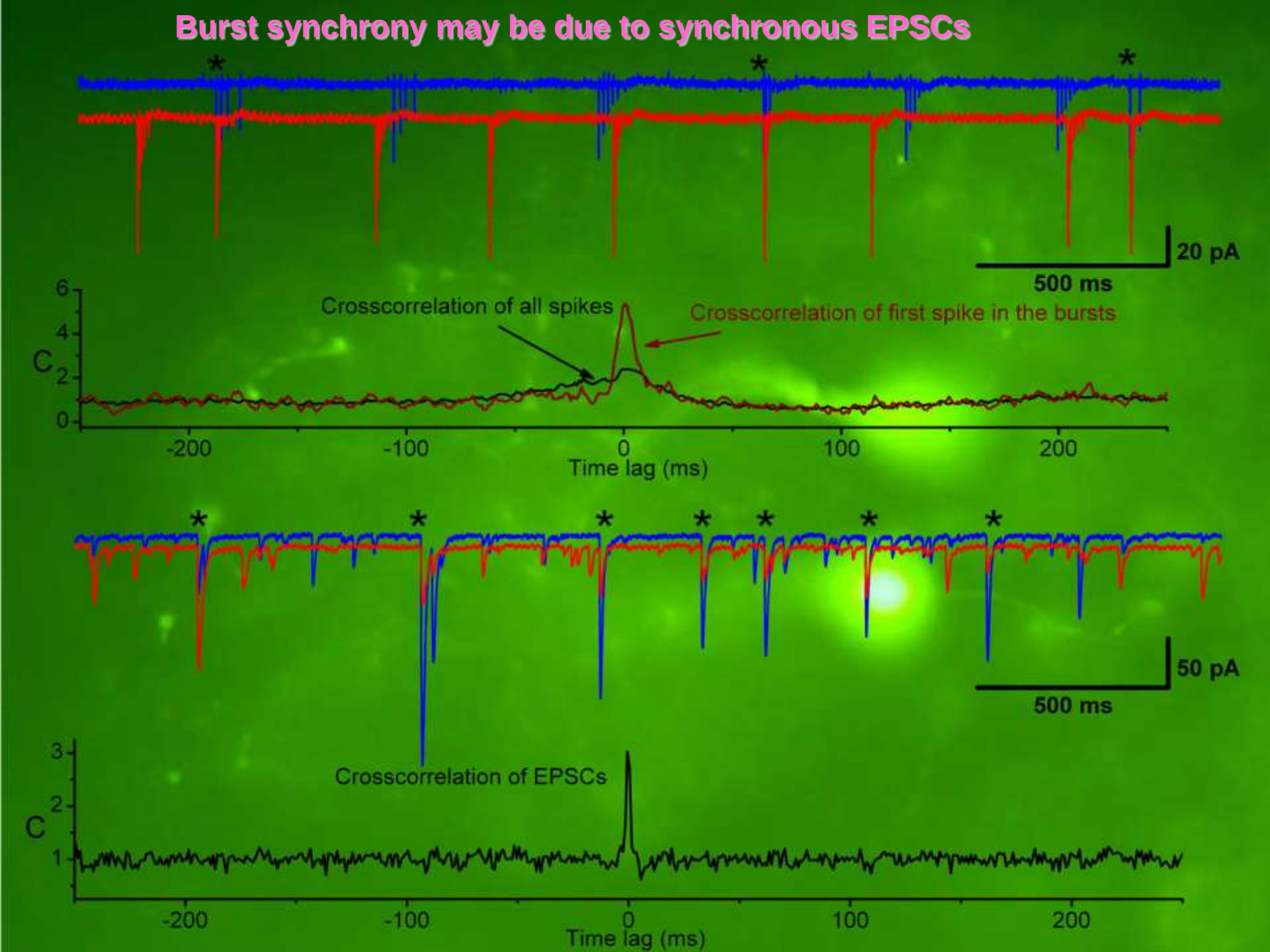
Dual multi unit – single unit recordings indicate that JG neurons are entrained to a ~5 Hz theta rhythm generated by rhythmically bursting ET cells



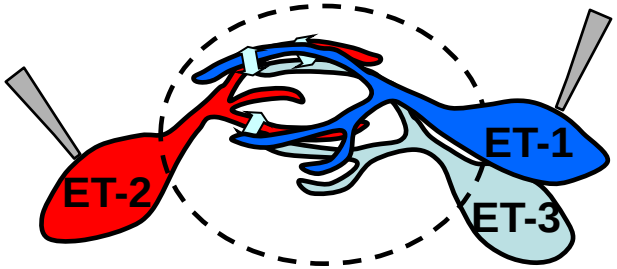


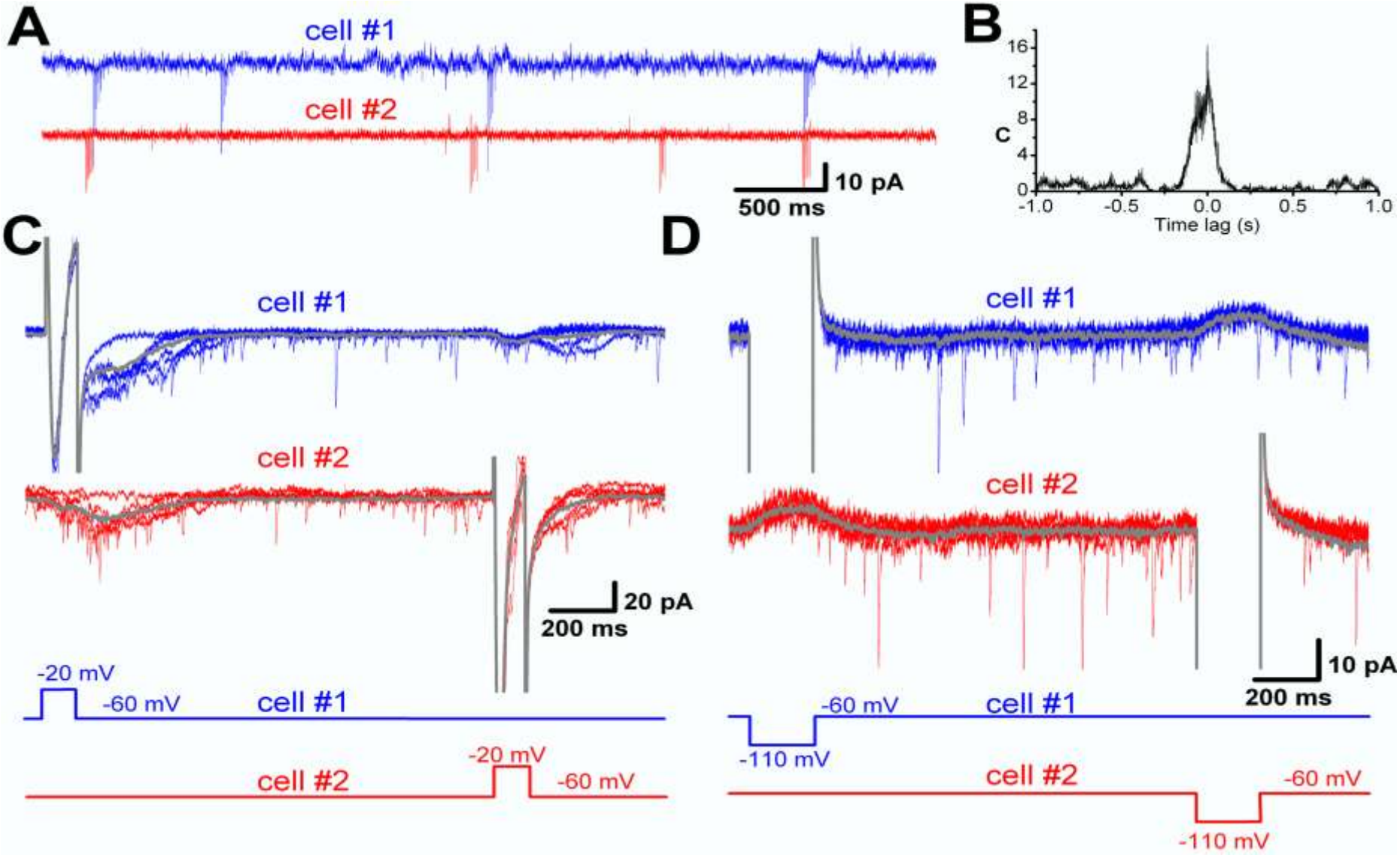

Triple extracellular recordings
ET cell#1
ET cell#2
ET cell#3
50 pA
500 ms
cell#1 to cell#2
cell#1 to cell#3
cell#2 to cell#3
Crosscorrelation coefficient
Time (ms)
Triple intracellular voltage-clamp recordings HP =-60 mV
ET cell#1
ET cell#2
ET cell#3
50 pA
500 ms
Triple intracellular voltage-clamp recordings HP =0 mV
ET cell#1
ET cell#2
ET cell#3
50 pA
500 ms

Burst synchrony may be due to synchronous EPSCs

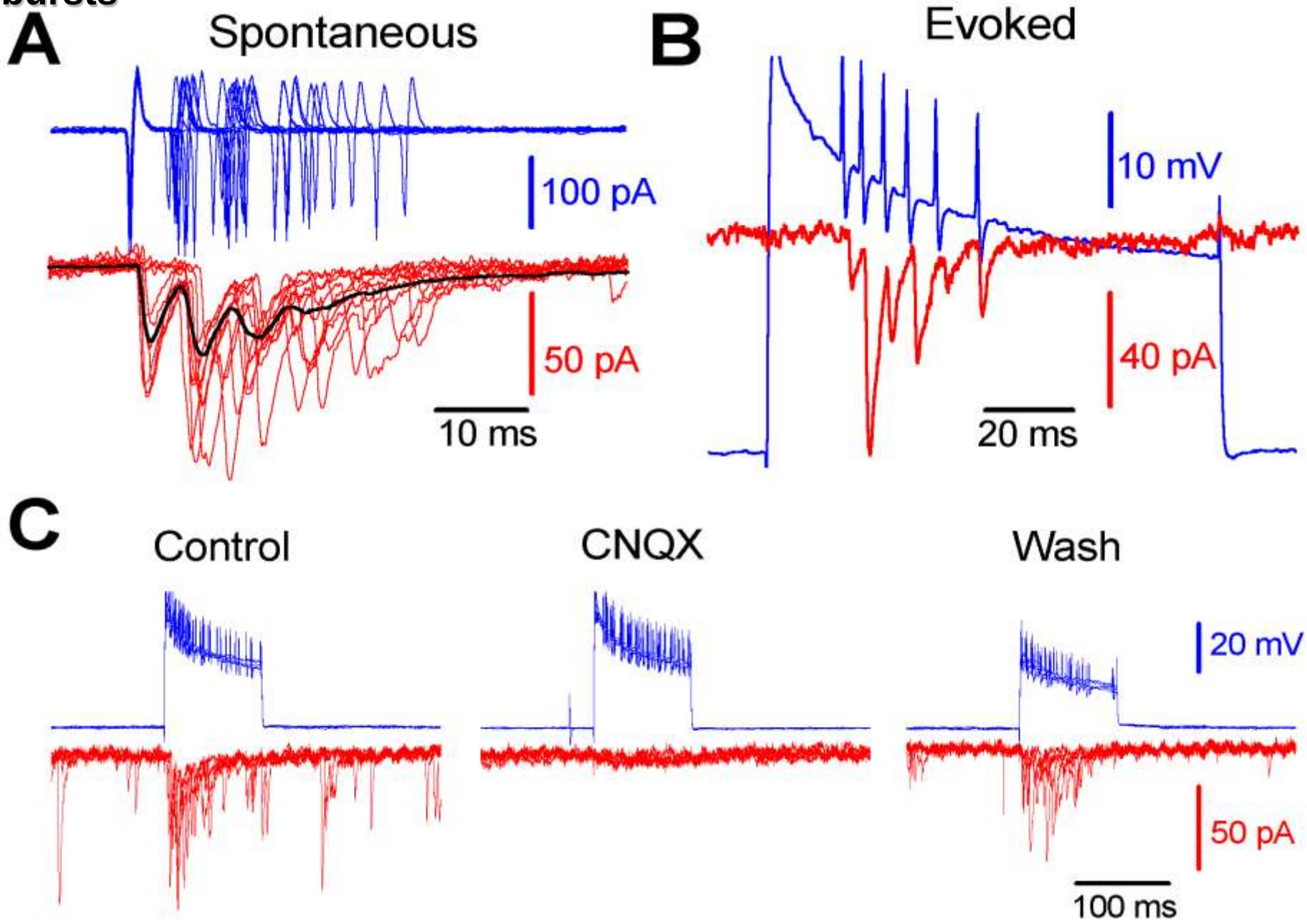


ET cells communicate via synaptic and gap junction currents

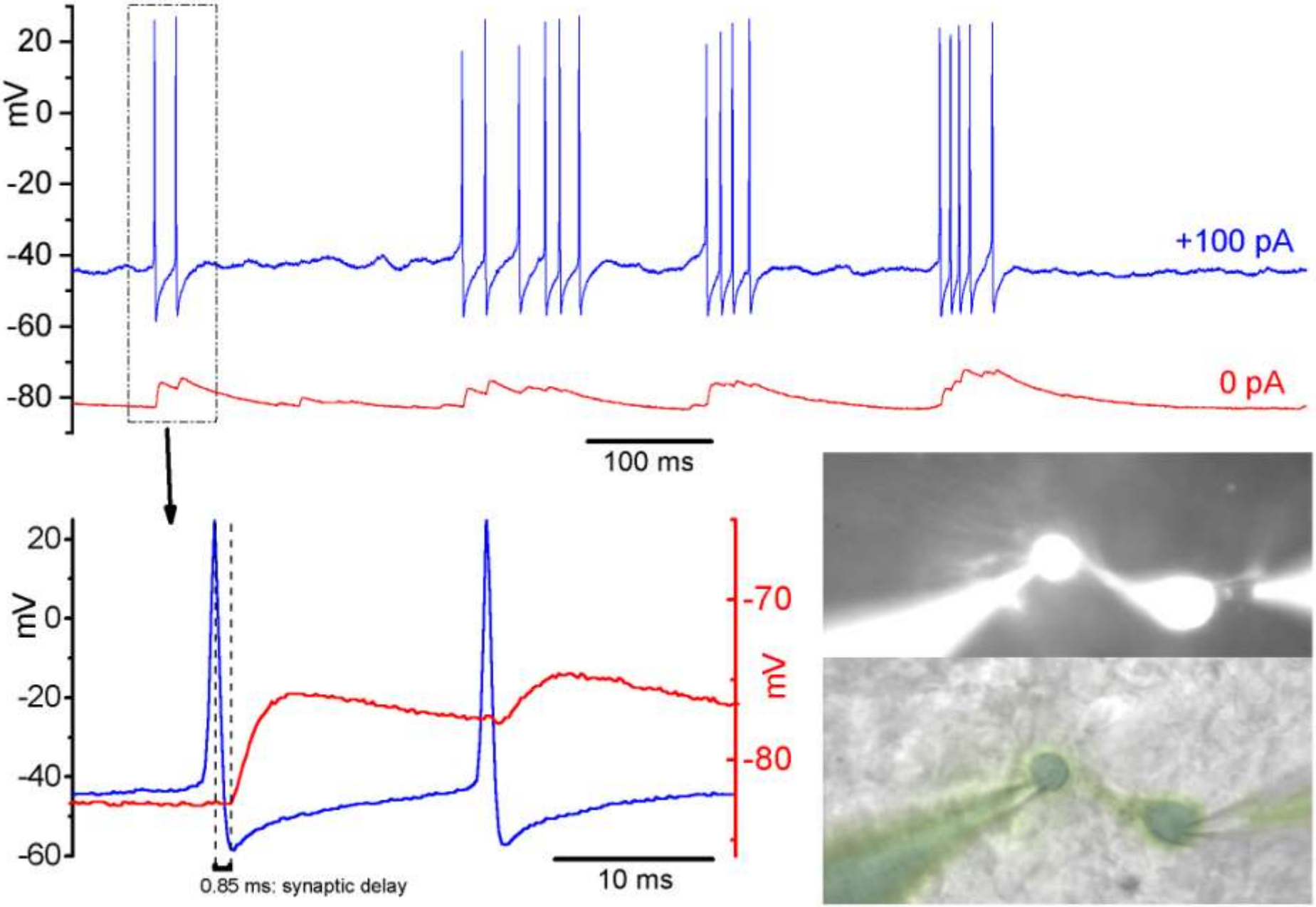




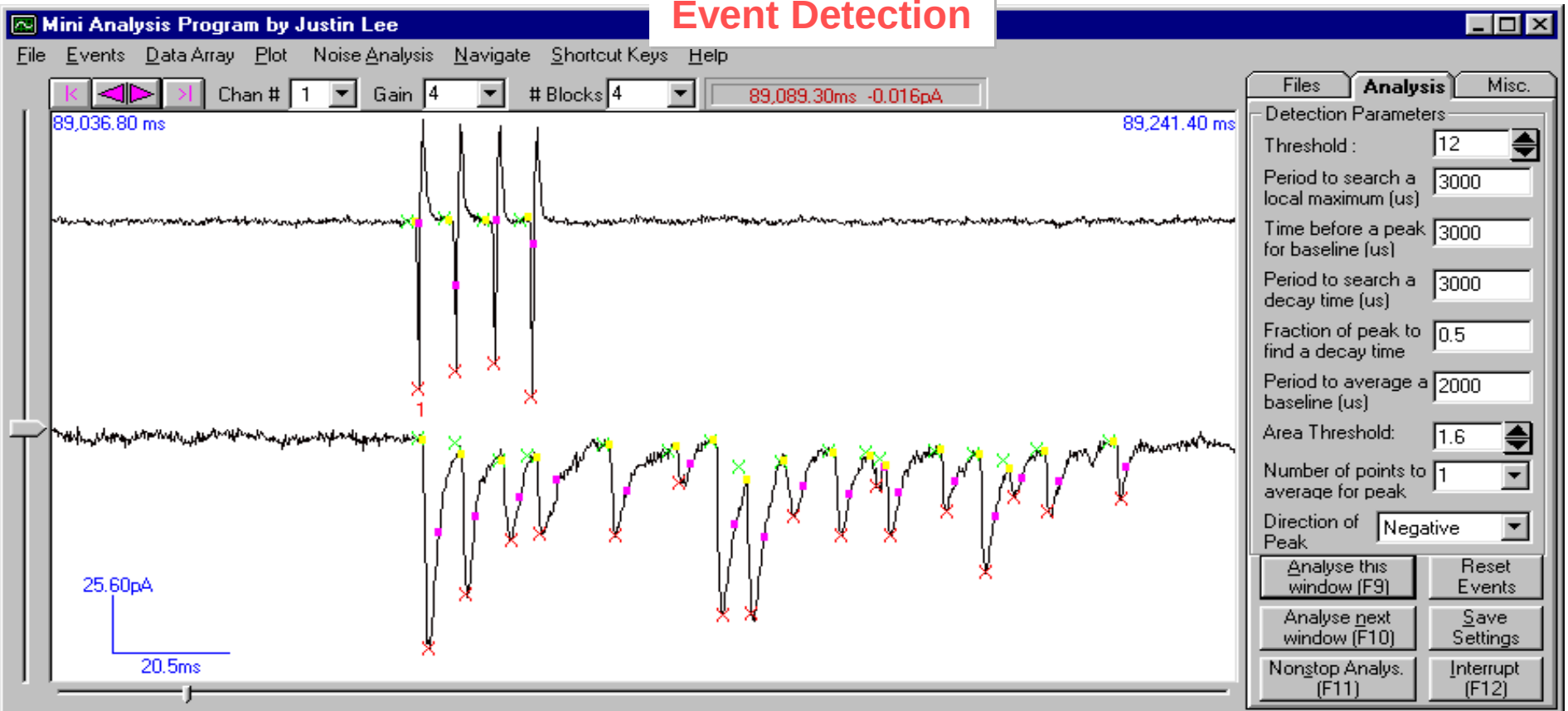
ET cells provide monosynaptic excitatory input to PG/SA cells: Evoked bursts



Direct synaptic coupling between an ET cell and a PG cell

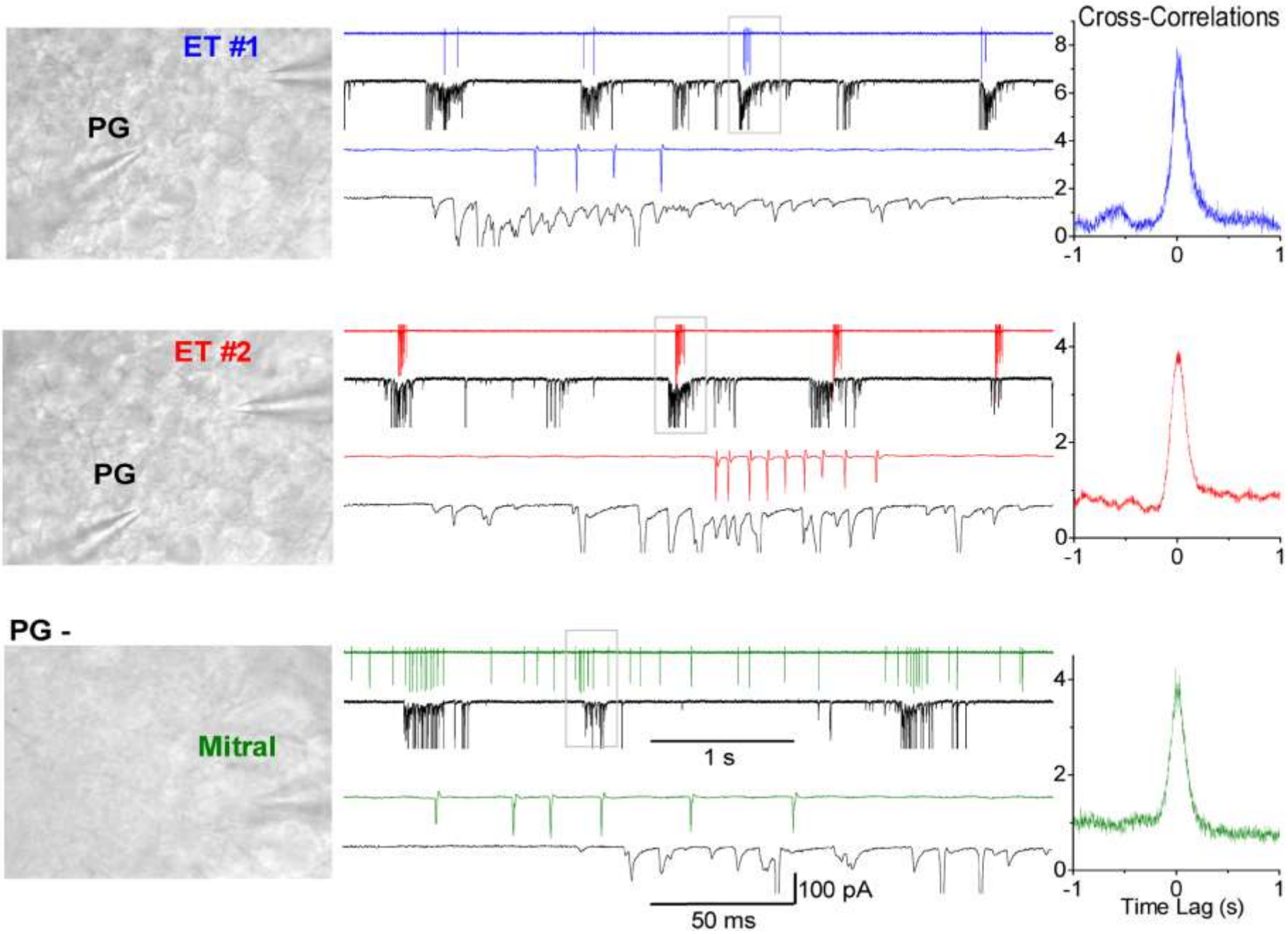


Event Detection

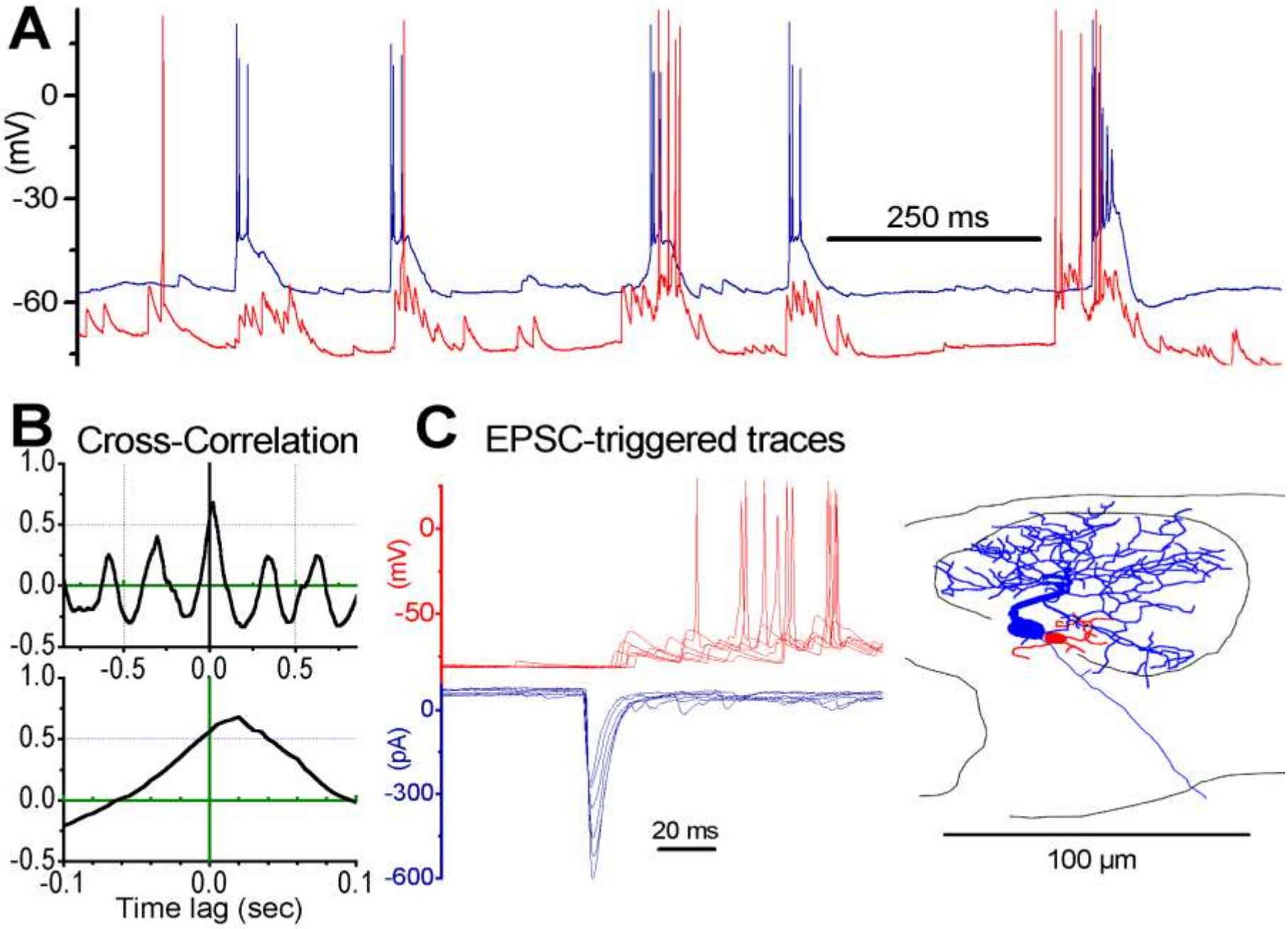


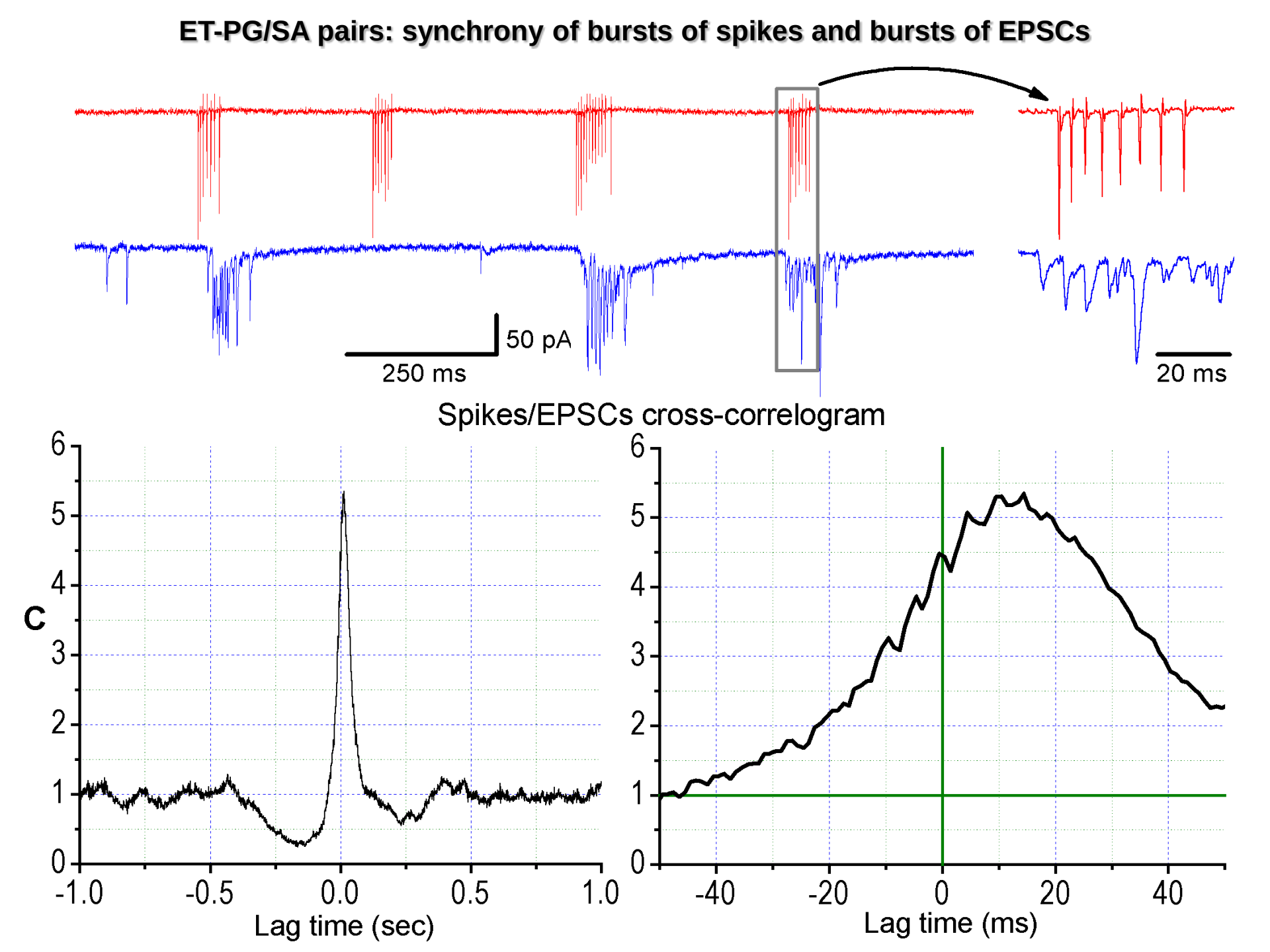
21	Time (ms)	Amplitude	Rise (ms)	Decay (ms)	Area	Baseline	Noise	Group	Channel	10-90Rise	HalfWidth	Rise50	Peak Dir	Burst#	BurstE#
1	89,100.20	100.33	0.60	0.20	33.94	-0.67	1.92	1	0	0.35	0.27	0.20	-1	0	0
2	89,106.60	91.62	1.20	0.20	32.18	1.13	1.13	0	0	0.31	0.31	0.20	-1	0	0
3	89,113.40	85.92	1.40	0.20	33.54	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

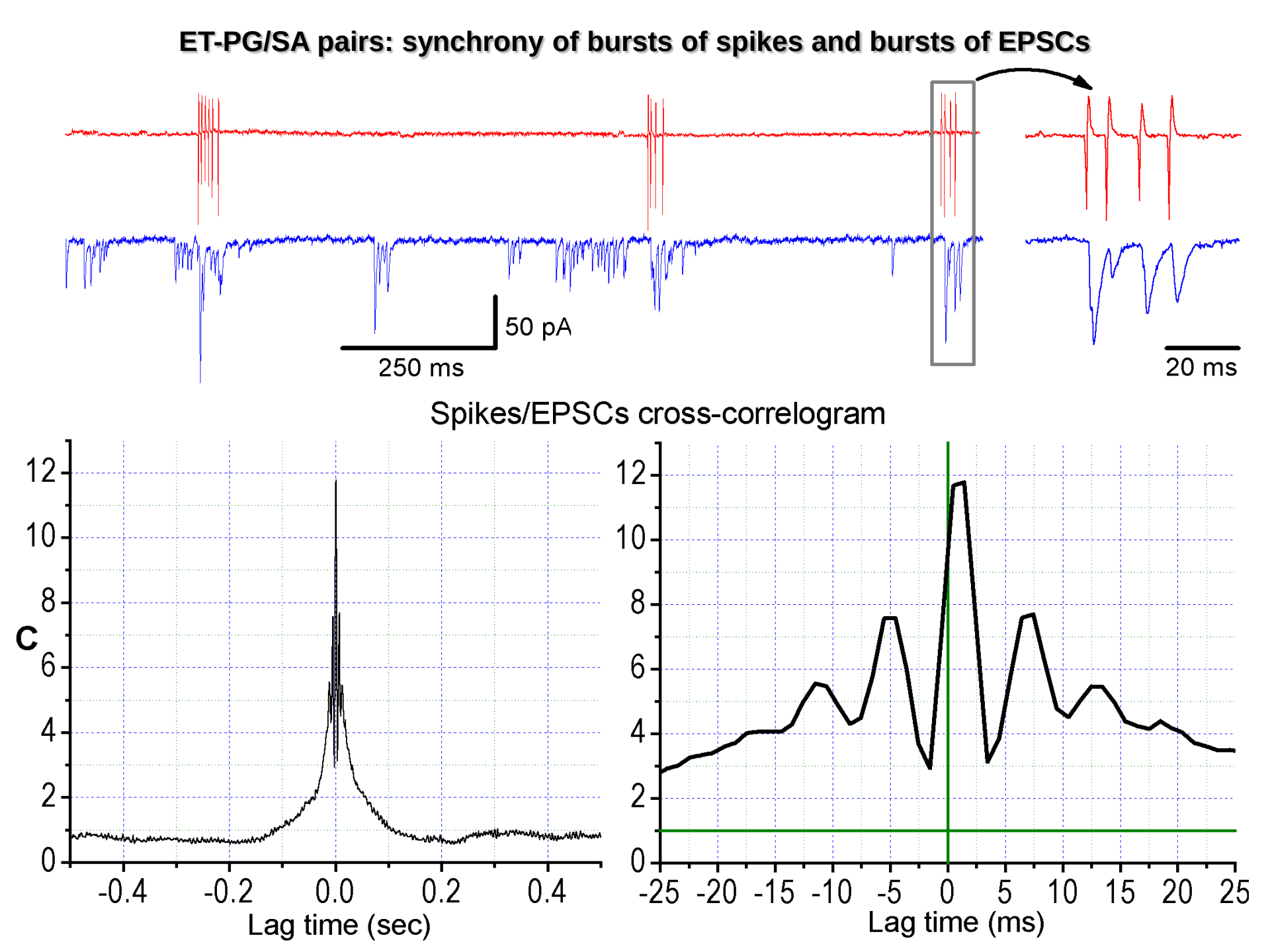
Mitral & ET cell firings are correlated with the bursts of EPSCs in PG/SA cells



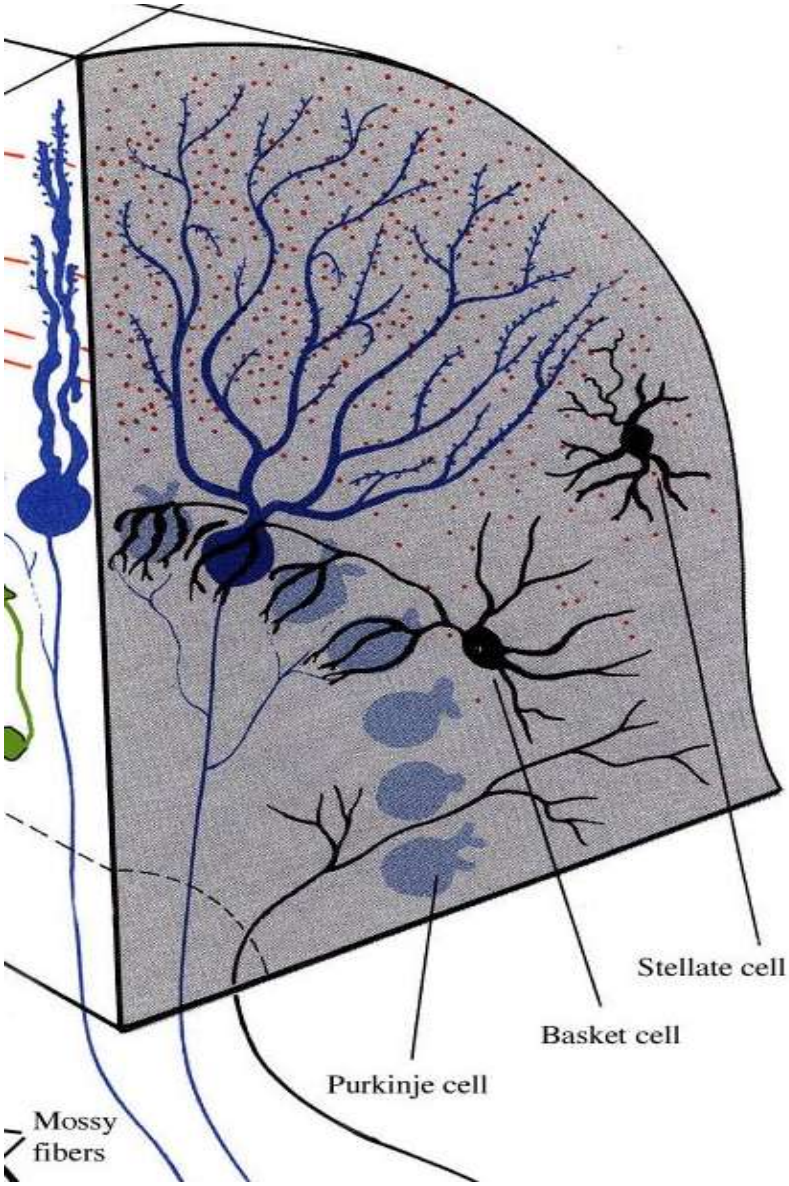
Spontaneous activity among ET-PG/SA cells of the same glomerulus is synchronous

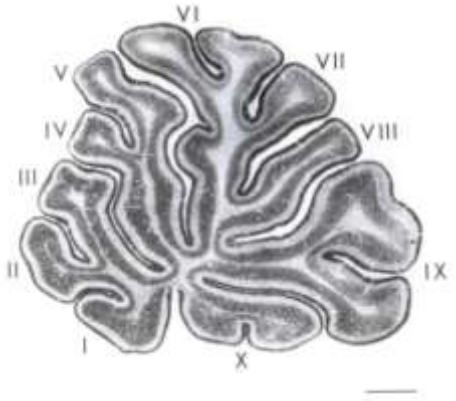



ET-PG/SA pairs: synchrony of bursts of spikes and bursts of EPSCs
50 pA
250 ms
20 ms
Spikes/EPSCs cross-correlogram
C
6
5
4
3
2
1
0
-1.0
-0.5
0.0
0.5
1.0
Lag time (sec)
-40
-20
0
20
40
Lag time (ms)

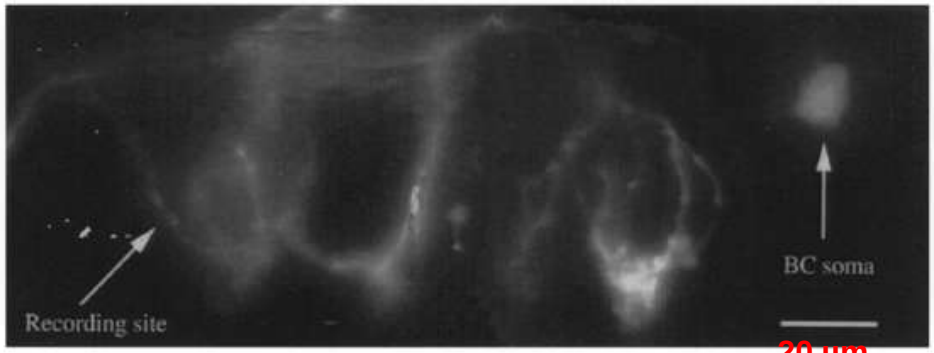

ET-PG/SA pairs: synchrony of bursts of spikes and bursts of EPSCs
50 pA
250 ms
20 ms
Spikes/EPSCs cross-correlogram
C
12
10
8
6
4
2
0
-0.4
-0.2
0.0
0.2
0.4
Lag time (sec)
-25
-20
-15
-10
-5
0
5
10
15
20
25
Lag time (ms)

Our results prove for the first time that PCs are synchronized by a common inhibitory input originating from an unidentified cell type

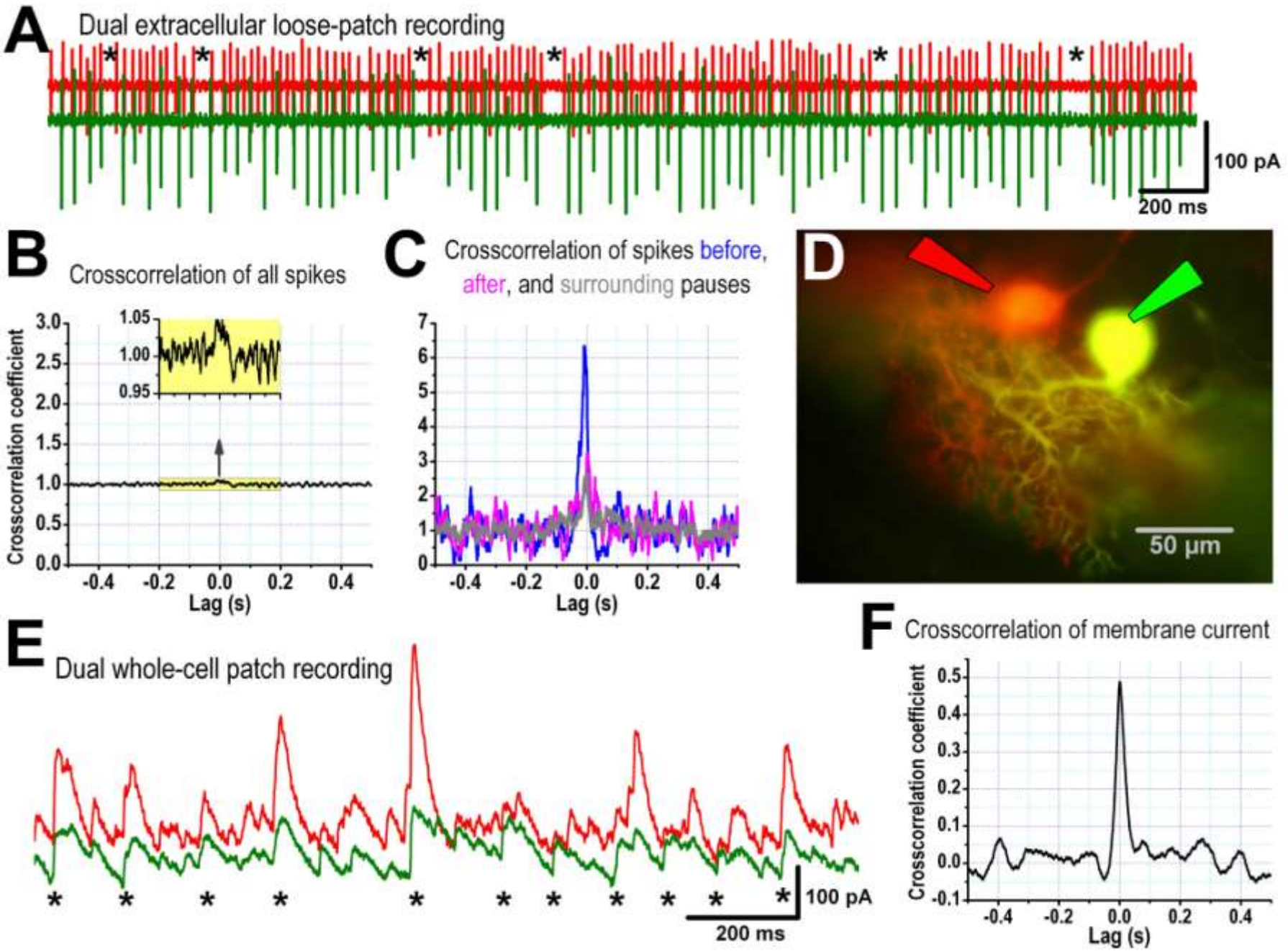




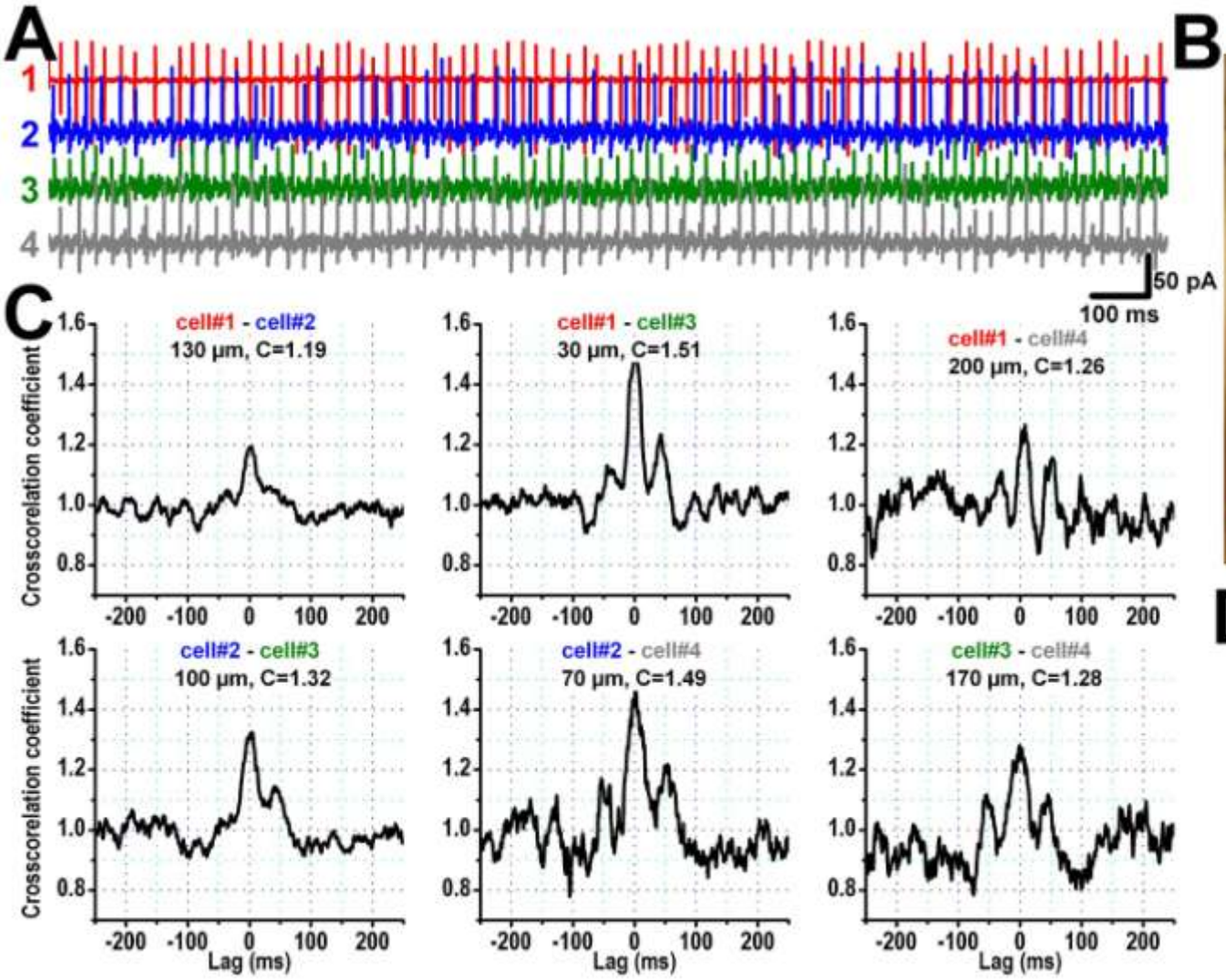
The source of inhibition could be basket cells because presynaptic axonal recording with a patch pipette containing a fluorescent dye has revealed that one basket cell can form at least 4 pericellular baskets and pinceau structures around adjacent PCs (Southan et al. 2000).



Purkinje cells are synchronized by inhibitory input



Synchrony decreases as a function of distance



Impaling oocytes with patch pipette electrodes