Cortical Systems: Interneurons and GABAergic inhibition

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Neuronal activity is determined by excitation and inhibition
Outline

- What is synaptic inhibition?
- What cells mediate inhibition?
  - Diversity of cortical interneurons
  - Interneuron development
- Functional considerations of GABAergic inhibition in the neocortex
Cortical inhibition is primarily mediated by GABA (gamma-amino butyric acid)

Glutamate

\[ \text{H}_2\text{N} - \text{C} - \text{O} - \text{OH} \]

GABA

\[ \text{H}_2\text{N} - \text{C} - \text{O} - \text{OH} \]

Glutamic Acid Decarboxylase (GAD)

GAD1 (GAD67) – somatodendritic, GAD2 (GAD65) - axonal
GABA-A receptors are ionotropic ligand-gated channels

Jacob et al. 2008
**GABA-B receptors are metabotropic G-protein-coupled**

![Diagram of GABA-B receptors and their interactions](Image)

*Benarroch 2012*
GABAergic inhibition has slow and fast components
What cells are responsible for evoking balanced excitation and inhibition?
Excitatory neuron morphology

A

Apical dendrite

Pyramidal soma

Basilar dendrites

C

Spiny dendrites

Apical tuft

D
Inhibitory interneuron morphology

Local processes

Aspiny

Round soma

Basket Cell

Martinotti Cell

Bitufted Cell

Small Basket Cell

Nest Basket Cell

Axon Shell Distance (µm)

Axon Segment Length (µm)

Interneuron Type

BC NBC SBC

*
Morphological diversity of interneurons

Huang et al. 2007
Basket Neurons are a Major Inhibitory Cell Type in the Cerebral Cortex

Figure 4. Camera lucida drawing of a basket cell from the junction of layers IV and V in the motor cortex of a human infant. Courtesy of Marin-Padilla (1969).
Basket Cells Collectively Form Pericellular Nests

Figure 2. Figures 361 (upper) and 362 (lower) of Ramón y Cajal (1911) showing what he called pericellular nests about the somata of pyramidal neurons in the deep part of layer III of a human infant. In the lower figure, "a" is identified as "afferent fibers."
Chandelier Cells form “candles” of synaptic connections on the Axon Initial Segments of Pyramidal Neurons
Examples of Chandelier Cells

Valverde, 1985

Figure 12. Examples of chandelier cells in diverse mammalian species and cortical areas. Reproduced from Valverde (1983) with permission.
Neurogliaform Cells

Dense local axonal projections

Figure 4. Camera lucida drawings of spiderweb cells from layer IV of the squirrel monkey somatosensory cortex. From material illustrated in Jones (1975). Only the parts of the axon found in one section are drawn; the axonal plexus formed by each cell usually extends through three or four 100-μm-thick sections. Lower cell in (B) has axon omitted to show dendritic field. Bars represent 100 μm and 1 mm (inset).
Martinotti cells target dendrites

Wang et al. 2004
Interneurons establish innervation domains on target dendrites ad somata
**Electrical diversity of interneurons**

AHP

Firing rate

AP duration

A

Firing rate adaptation

C

Spike Frequency (Hz)

Time (ms)
Interactions between intrinsic properties and synaptic properties
Simplified Example of Differences in Lamination Patterns Between Cortical Areas
Synaptic connectivity by layer

Pyramidal Neurons

Fast Spiking Inhibitory Neurons

C
D

excitatory input strength

inhibitory input strength

layer 2/3 4 5a 5b 6

layer 1 2/3 4 5a 5b 6
Synaptic connectivity by layer

Chandelier Cells

Excitatory input strength

Inhibitory input strength

Neurogliaform Cells

Excitatory input strength

Inhibitory input strength
Feed-forward flows of excitation and inhibition

Layer 2/3

Layer 4

Thalamus
Chemical composition of cortical GABAergic population.

Uematsu M et al. 2007
Three groups of interneurons account for nearly 100% of neocortical GABAergic neurons.

Neocortical GABAergic Interneurons

- **PV**: 40% [1]
  - Basket FS Cells
  - Chandelier FS Cells
  - Variations in:
    - Somatodendritic size
    - Axonal projection
    - Excitatory input dynamics
    - Intrinsic firing pattern
    - Molecular expression
  - MB? [2]
  - Martinotti cells
    - CR+ / CR-
    - Layer IV
    - Layer V

- **SST**: 30% [1]
  - X94 (NFS1)
  - Other? [3]
  - Layer IV
  - Layer V

- **5HT3aR**: 30% [1]
  - VIP 40% [4]
  - non-VIP 60% [4]
  - Reelin (SST-): 80% [6]
    - Unknown markers
    - LS1 (NGF)
    - LS2
    - bNA1
    - dB [5]
    - vLB [5]
    - mLS [7]
Interneurons arise from the ganglionic eminences

- Differentiated from sub-pallium
- MGE, CGE -> neocortex and hippocampus
- LGE -> olfactory bulb and striatum
What is the function of GABAergic inhibition?
Somatic inhibition enforces spike timing fidelity

Pouille and Scanziani 2001
Inhibition and sensory processing: Somatosensory (barrel) cortex
Feed-forward inhibition in somatosensory (barrel) cortex

Wilent and Contreras 2004
Fine timing of excitation and inhibition shapes response precision

Higley and Contreras 2006
Direction selectivity in barrel cortex

(a) Experimental setup and evoked responses

(b) Direction selectivity

(c) Synaptic response (Avg n = 47)

Wilent and Contreras 2005
Direction selectivity in barrel cortex

Wilent and Contreras 2005
GABA and rhythmogenesis

Cardin et al. 2009
**Inhibitory synapses target postsynaptic dendrites**

- 15% of synaptic terminals synthesize GABA
- 84% of GABAergic synapses are on dendrites (58% shafts, 26% spines)
- 15% of GABAergic synapses are on somata or initial segments

*Somogyi, 1989*

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*Halasy...Somogyi 1996*
What is the function of dendritic inhibition?
Expression of ChR2 in SOM-INs

- Acute PFC Slices, P30-50, 25°C
- 473nm, 3-5ms, 1-2mW through obj.

Chiu, Lur et al., Science, 2013
2-photon microscopic imaging of dendritic Ca$^{2+}$ signals

Alexa-594 (10µM)  Fluo 5F (300µM)
Combining 2p imaging and optogenetics

Image pyramidal neuron spines, stimulate SOM-INs with ChR2

- Image
- Current Pulse (AP) 2 nA, 1 ms pulse through recording pipette
- Laser Pulse (IPSP) 1-2 mW, 5 ms pulse through microscope objective

\[ \Delta G/G_{sat} = 4 \text{ mV} \]
\[ \Delta G/G_{sat} = 10 \text{ ms} \]

Chiu, Lur et al., Science, 2013
SOM-IN-evoked inhibition along a dendritic branch

Inhibition is compartmentalized in spines!

Chiu, Lur et al., Science, 2013
Compartmentalized inhibition of glutamatergic inputs

Dual glutamate & GABA uncaging

CDNI-Glu + RuBi-GABA

ΔCa$^{2+}$

uEPSP

Chiu, Lur et al., Science, 2013
A “balance” of glutamatergic excitation and GABAergic inhibition
Cellular mechanisms for regulating synaptic strength
Does activation of NMDARs alter GABAergic inhibition?

- “Chemical” induction (20 μM NMDA)
- Acute slices from prefrontal cortex
- Whole-cell Vclamp recordings
- Room Temp.
NMDAR activation potentiates dendritic inhibition

Chiu and Higley, in prep.
iLTP requires postsynaptic Ca2+

Chiu and Higley, in prep.
The locus of iLTP is postsynaptic

Chiu and Higley, in prep.
iLTP involves membrane insertion of GABARs

Non-Stationary Fluctuation Analysis:

Chiu and Higley, in prep.
iLTP requires activation of CaMKIIa

Chiu and Higley, in prep.
Activation of CaMKIIa potentiates dendritic inhibition

Chiu and Higley, in prep.
Synaptic triggering of iLTP

Theta-burst + Depolarization

Chiu and Higley, in prep.
Composition and modulation of $\text{GABA}_A$ receptors

$\text{GABA}_A$ receptor

$(2\alpha 2\beta 1\gamma)$

CaMKII$\alpha$ phosphorylation sites

- $\alpha 1$: unknown
- $\beta 1$: S384*, S409
  * $\beta 2$: S410
  * $\beta 3$: S383, S409
- $\gamma 2$: S348, T350 (L:S343)

* Linked to iLTP (Petrini et al. 2014, He et al. 2015)
SOM- and PV- synapses utilize distinct GABAR subunits

Chiu and Higley, in prep.
Role of beta subunits in iLTP

β2 or β3 f/f

AAV-Cre-GFP
5-6 weeks

GABA Uncaging
473 nm
Rubli-GABA

W.T.
β3 -/-
β2 -/-

baseline
20 min. post-NMDA

IPSC\text{post}/IPSC\text{pre}

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50 pA
100 ms
...do NMDARs control an E/I balance?
Testing the balance of E and I

E/I balance diverges at the subcellular scale!