Playing Piano
(without the brain):
Movement and timing through cerebellar circuits.

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$10^{10}$ people on Earth
$10^{11}$ neurons in the human brain

$10^5$ synapses on a Purkinje cell
Cerebellum plays a function in posture, fine motor control, and programming.

- 50% of cells in the brain.
- Cerebellar damage:
  - *Hypotonia* reduced resistance (pendulum).
  - *Astasia* inability to stand or walk (spread).
  - *Ataxia* irregular rhythmic movement (up-down).
  - *Intention tremor* antagonist muscle control error.
Intention tremor.

What is it in the structure of the cerebellum that allows us to control fine movements?
Cerebellum is compartmentalized functionally.

- Deep nuclei -> superior cerebellar peduncle.
- Vesticular input -> flocculonodular lobe -> vestibular nuclei (smooth eye track pursuit).
- Spinal chord -> vermis fastigial nucleus -> red nucleus descending tract (dorsal tract passive feedback, ventral tract active efference copy, ipsilateral, deep nuclei somatotopic).
- Cortical areas -> pons -> lateral cerebellum -> dentate -> motor prefrontal cortices.

GCL: 100 billion granule cells, Golgi interneurons.  
PCL: 70um Purkinje cells, -> deep nuclei, vestibular.  
ML: Stellate, basket, parallel fibers relay perp to PCd.
Parallel fibers are axons of granule cells, project to Purkinje cells in molecular layer

Optican, 1998
Parallel fibers are axons of granule cells, project to Purkinje cells in molecular layer

Optican, 1998

Input – Error
(one-to-one PC)
(Ca -> complex)
(IO synchrony)

Input – Motor
(convergence)
(simple spikes)
(glomerulus)

Parallel fibers are axons of granule cells, project to Purkinje cells in molecular layer

Optican, 1998

Output
(inhibit mod)
(negative feedback)
(IO gap junctions)
Parallel fibers are axons of granule cells, project to Purkinje cells in molecular layer

Both excitatory and inhibitory connections converge at cerebellar cortex and deep nuclei.
Cognitive functions of cerebellum.

- Lateral lesions interfere with (subjective) serial timing (duration and speed judgments), not only irregular tapping.
- Right lateral cerebellar activation in word association vs reading aloud task.
- Greater activation when solving peg puzzle vs simply moving pegs in a board.
- Anticipatory postural adjustments require efference copy of intended movement.

One of multiple sites of cerebellar motor learning.

- Climbing fiber (CF) induced long term depression LTD of parallel fiber to Purkinje synapses.
- Motor system as cerebellum implemented internal (inverse dynamics) model.

Gilbert, Thach, 1977
Cerebellar cortex can be thought of as an adaptive filter (Marr-Albus-Ito).

Lepora et al., 2010.

Computational simulation (10,000 grc, 20 PC) of cerebellar circuits recapitulate behavior.

Medina, Mauk, 2000
Eyeblink conditioning repeatedly pairs tone CS to air puff US, to enable learning of blink CR.

Woodruff-Pak, 2002.

Learned to blink regardless of context.

Cerebellar interpositus nuclei are necessary for learning of classical eyeblink conditioning.

Kim, Thompson, 1997
Cerebellar cortex lesions show shortened eyelid responses to 150ms and 750ms CS-US pairs.

Perrett, Ruiz, Mauk 1992

Effect on lesion on eyelid response depends on duration of CS-ISI and extent of lesion.
Single CS-ISI learning deficits are inconsistent with disruption of auditory discrimination.

Summary of cerebellar action.

- The cerebellum is necessary for everyday movements like saccades (vermis) & pointing.
- Purkinje cells and deep nuclei cells fire during voluntary movements in feedforward control.
- Cerebellum has an internal model of limb structure that anticipates forces in movement.
- Altering strength of particular parallel fiber to Purkinje cell synapses reduces motor error, and may allow for accurate learning (piano).
Can we link behavior and physiology to circuits and molecules?

**Evidence for depolarizing GABA action in mature neurons**

- **Zhang and Jackson 1995**
  - Posterior pituitary

- **Szabadics et al. 2006**
  - Axo-axonic cells cerebral cortex

- **Choi et al. 2008**
  - Suprachiasmatic nucleus

- **Turecek and Trussell 2002**
  - Calyx of Held terminal
Evidence for *excitatory* GABA action in mature neurons

- Turecek & Trussell 2002, excitatory glycine and GABA currents at calyx of Held MNTB.
- Szabadics et al. 2006, excitatory action of GABA released by axo-axonic cortical cells, absence of KCC2 transporter in axons.
- Stell et al. 2007, bursts of EPSCs recorded in Purkinje cells and molecular layer interneurons evoked by GABA$_A$ agonist.

GABA$_A$R activation excites presynaptic input to Purkinje cells and MLIs

Stell, Marty 2007

Pugh, Jahr 2011
Where do we come from?
Where are we going?

Calcium transients
Fiber volleys

Local perfusion of calcium indicator limited to molecular layer and parallel fibers

Oregon Green BAPTA-1 AM -> transverse slice

Luo, Dellal, Otis, 2012
Regehr and Atluri, 1997
Presynaptic calcium transients are potentiated by GABA$_A$R agonist muscimol

Luo, Dellal, Otis, 2012
Review Questions

• 1. Which of the following is NOT a disease caused by cerebellar dysfunction?
  – A. hypotonia: lack of muscle resistance
  – B. ataxic body movements
  – C. autism: verbal impairments and repetitive behavior
  – D. intention tremors

• 2. Which of the following forms the output of the cerebellar cortex circuit?
  – A. deep nuclei
  – B. parallel fibers
  – C. granule cells
  – D. Purkinje cells

• 3. We trained a rabbit to anticipate an airpuff (US) by blinking in response to a tone (CS). What happens if on the next day we present the same tone (CS) only (without US) in a new room that smells like pineapple but with lights dimmed?
  – A. the rabbit will cease to blink to the tone
  – B. the rabbit will blink continue to blink to the tone
  – C. the rabbit will not blink initially but will gradually regain blinking to the tone
  – D. the rabbit will move around when tone occurs but when she realizes there's no airpuff she will stop moving around
Is GABA-mediated excitation working at single parallel fibers? Or recruiting fibers?

Low Stimulation

Low Stim and GABA activation

High Stimulation

High Stim and GABA activation

GABA\(_A\)R activation brings additional fibers closer to threshold for spike generation
NKCC1 transporter is the chloride accumulating transporter

Payne et al. 2003

Muscimol-induced increase in calcium transient depends on the chloride gradient
Compound action potential (fiber volley) amplitude is increased by uncaging GABA

GABA$_A$R activation increases fiber volley amplitude and shortens latency

Dellal, Luo, Otis, 2011
Conduction velocity measurements using fiber volleys in a transverse slice

GABA$_{A}$R activation increases fiber volley conduction velocity
GABA-mediated excitation of parallel fibers is independent of granule cell

GABA<sub>A</sub> receptors can be categorized into two major types

- extrasynaptic
- high affinity
- tonic inhibition
- α(4/6) β δ

Granule cells: α<sub>1/6</sub> β<sub>2/3</sub> δ/γ<sub>2</sub>

- synaptic
- low affinity
- phasic inhibition
- α(1/2/3/5) β γ
Blocking GABA reuptake prolongs parallel fiber excitation

Endogenous GABA is sufficient to increase parallel fiber excitability
Endogenous GABA is sufficient to increase parallel fiber excitability

![Graph showing PTX-sensitive fraction over time with different conditions]

Granule cell model.

5.8 μm diameter granule cell, 0.3 μm diameter axon, 70 μm ascending branch, 0.5 mm fiber after T-junction segment simulated.

Hodgkin-Huxley dynamics (from Walther), K-A channels, K-mixed-ion leak, chloride leak to maintain Erev-Cl, GABARs high conductance chloride leak.

At 303 K, [Cl] out = 130 mM, a change in reversal potential for [Cl] of 5 mV results from [Cl] in rise from 10.8 to 13 mM (10 mV -> [Cl] in = 15.8 mM).
Granule cell compartmental model

5.8 μm diameter granule cell
0.3 μm diameter axon
Mammalian Hodgkin-Huxley (Wang 2003)
10 mV Erev change from 5mM [Cl] rise

Model recapitulates effect of GABA_A R activation on parallel fiber excitability

A

stimulate record AP

gGABA_A R

B

Dellal, Luo, Otis, 2011

GABA

C

D

Control

Threshold change

Injected Current (pA)

Spike variability

Threshold change

Intracellular Potential of GABA-gated Chloride (mV)
GABA$_A$R activation decreases threshold for orthodromic spike initiation

Local GABAR activation is sufficient for lowering fiber volley spike threshold.
Local GABAR activation in molecular layer decreases GRC AP threshold.

Conduction velocity is increased by local molecular layer GABAR activation.
GABA effect on conduction velocity depends on sodium channel inactivation

GABA$_A$R activation brings axon closer to threshold for spiking at low conductances.
Spike failures (e.g. near the T) can occur near 100x the tonic conductance.

Can GABA excitation relieve spike failures at T-junction? My cat says yes.
Spike failures (e.g. near the T) can occur near 100x the tonic conductance.

What it means to be fast? 10% increase from 200 µm/ms => 2 ms
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Effect of GABAR on velocity depends on sodium channel inactivation.
GABAR activation in ML decreases GRC AP threshold and initiation time.