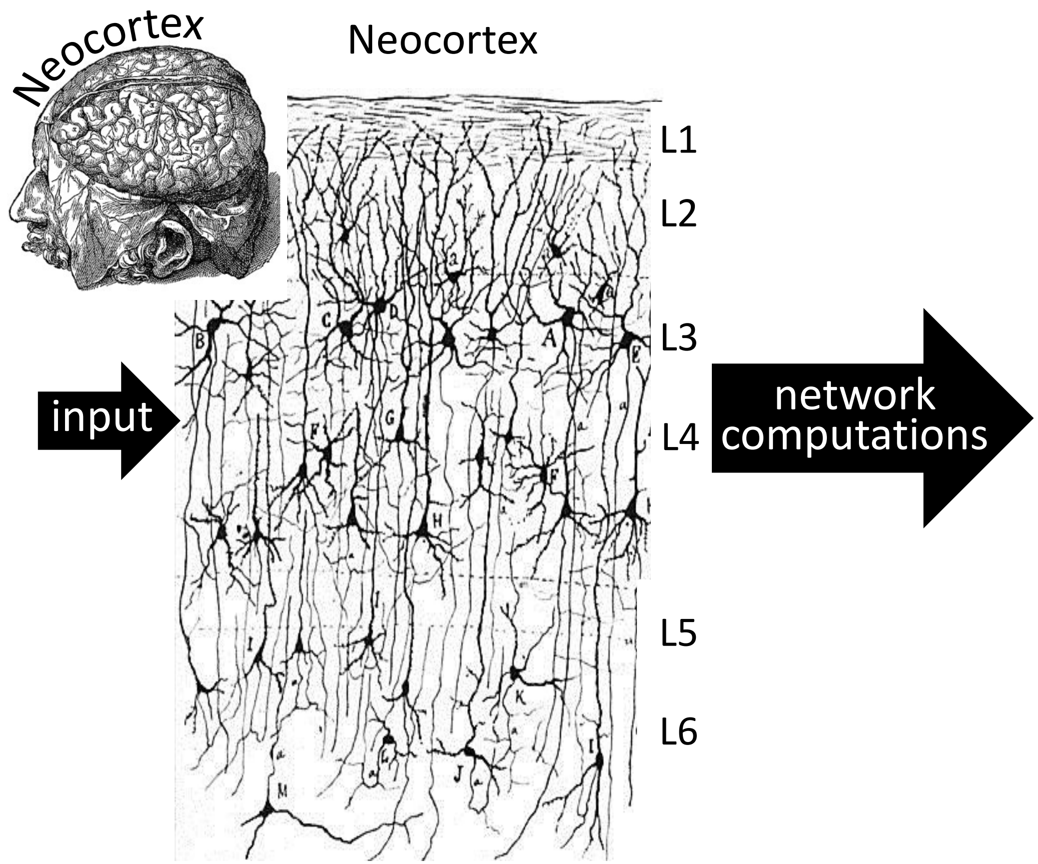
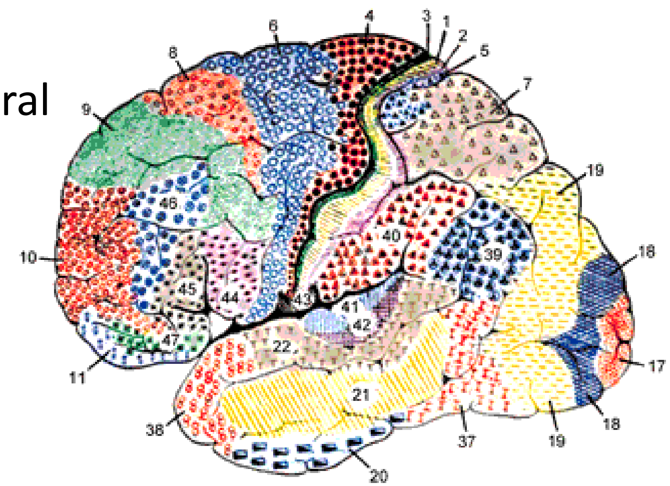
Cortical Layers:
What are they good for?

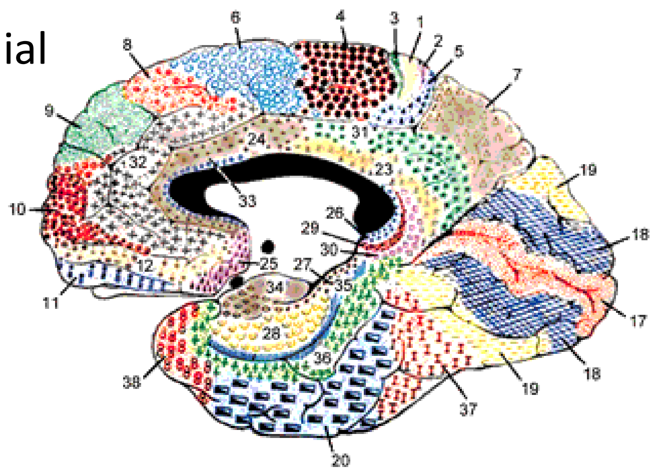

Neocortex
Neocortex
L1
L2
L3
L4
L5
L6
input
network computations

Brodmann Map of Cortical Areas

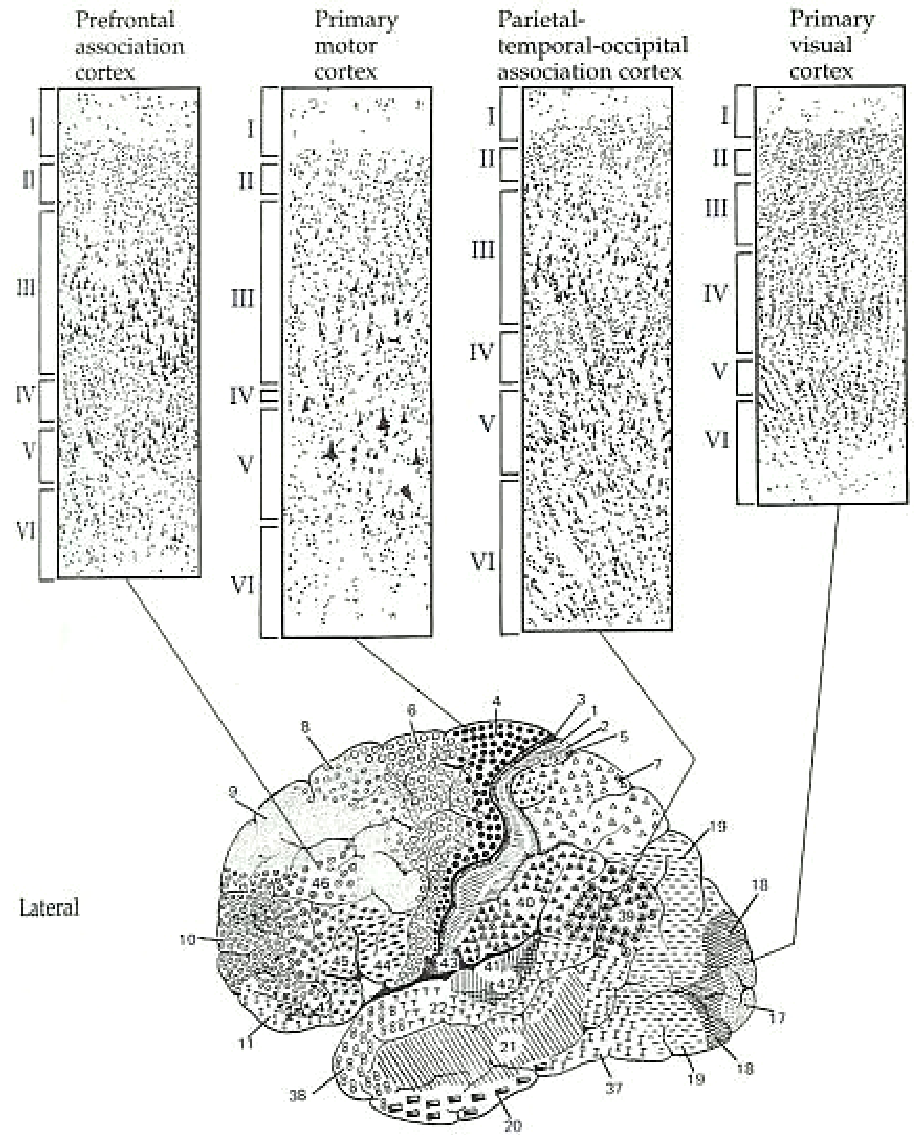
lateral



medial

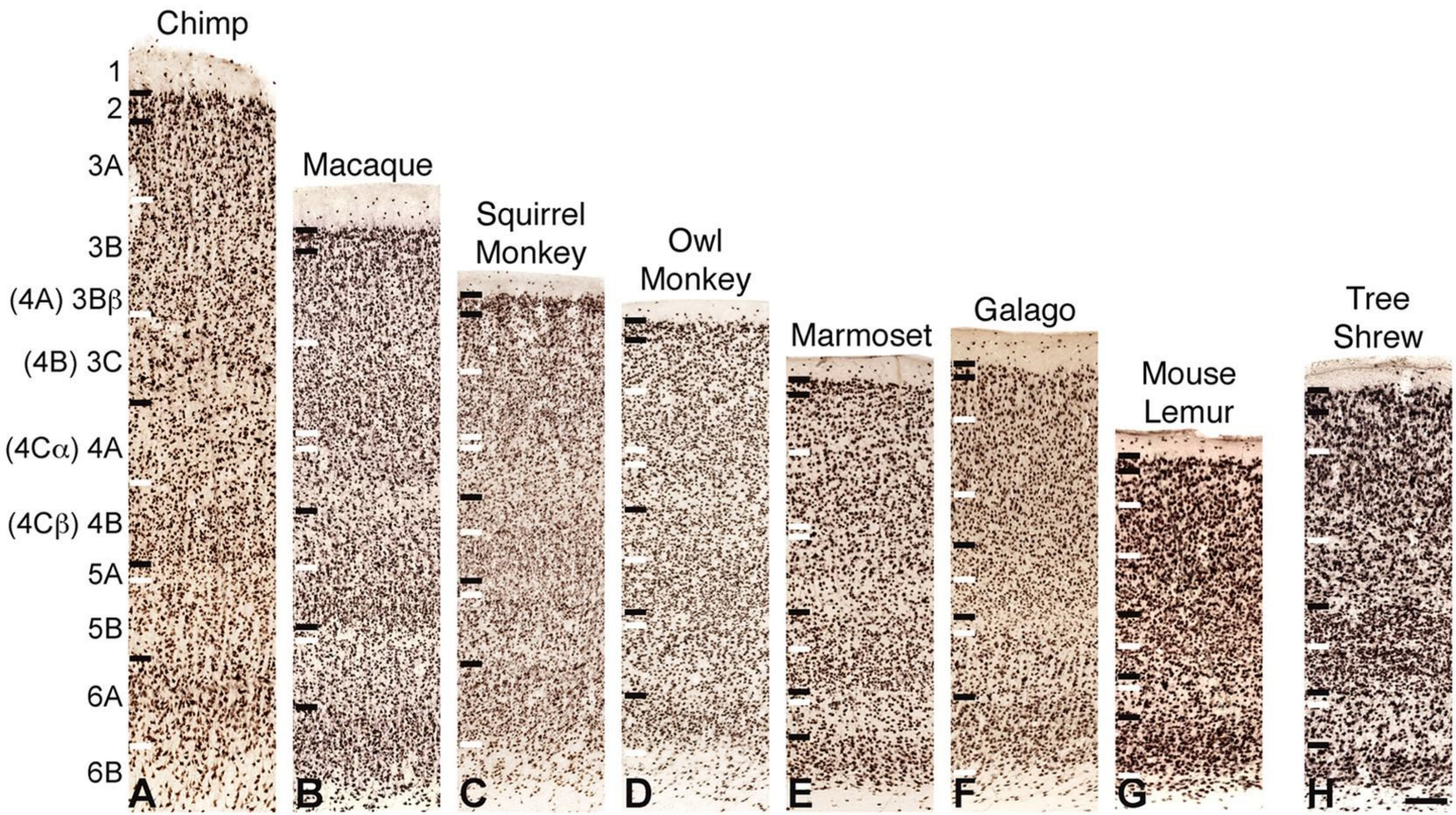


44 areas, numbered in order of samples taken from monkey brain



Brodmann, 1908.

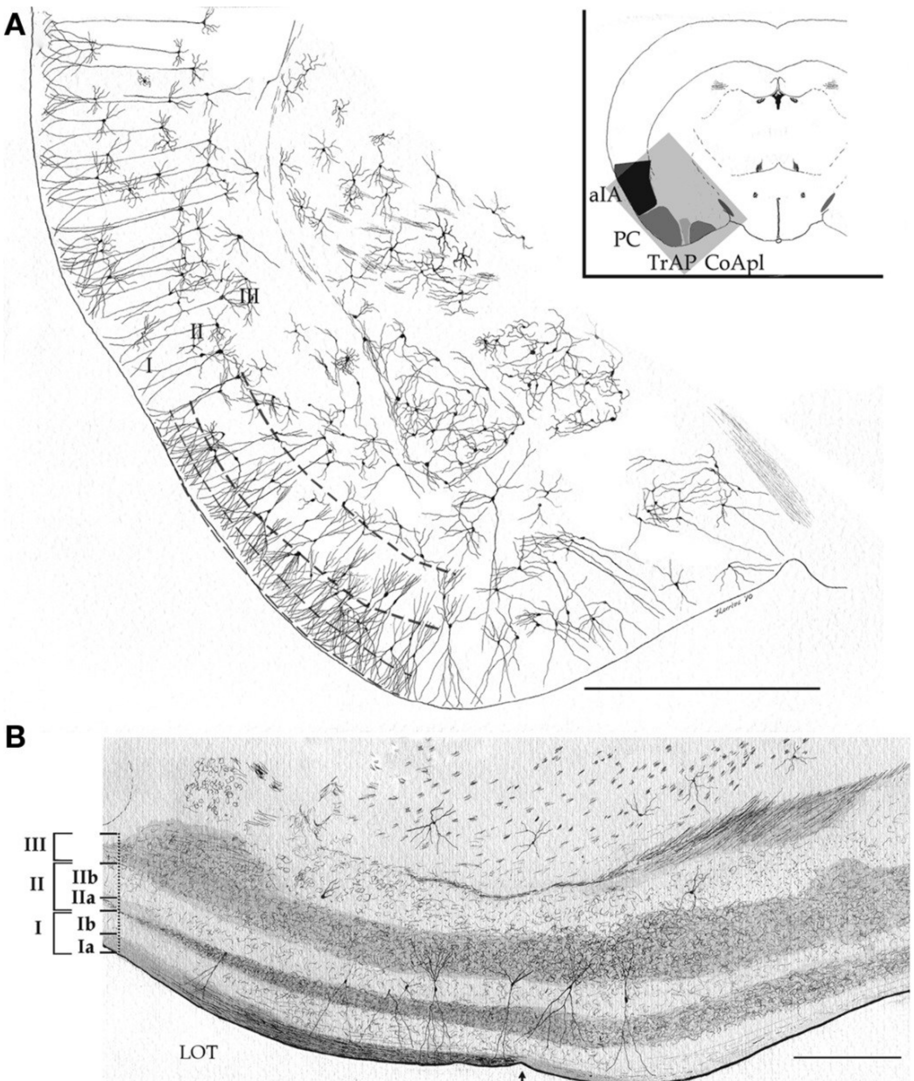
Primary visual cortex lamination across species



Balaram & Kaas 2014 Front Neuroanat

Cortical lamination: not always a six-layered structure (archicortex)

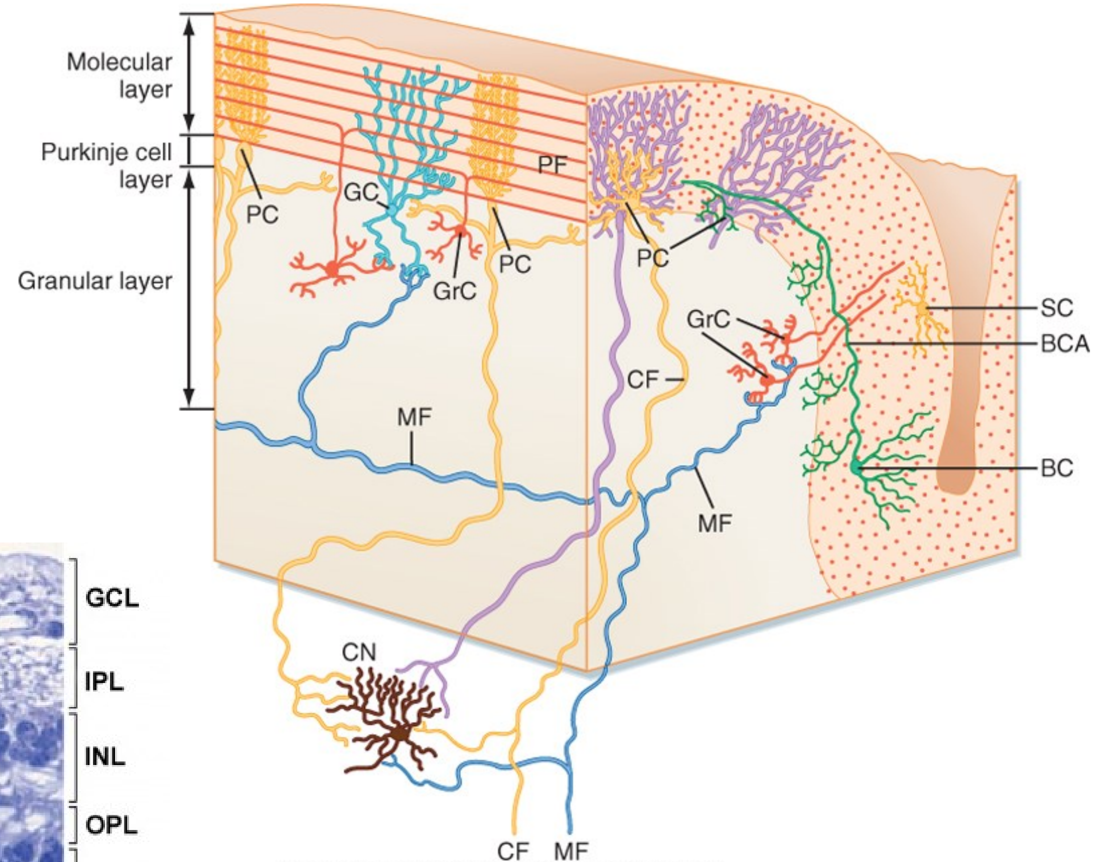
e.g. Piriform, entorhinal



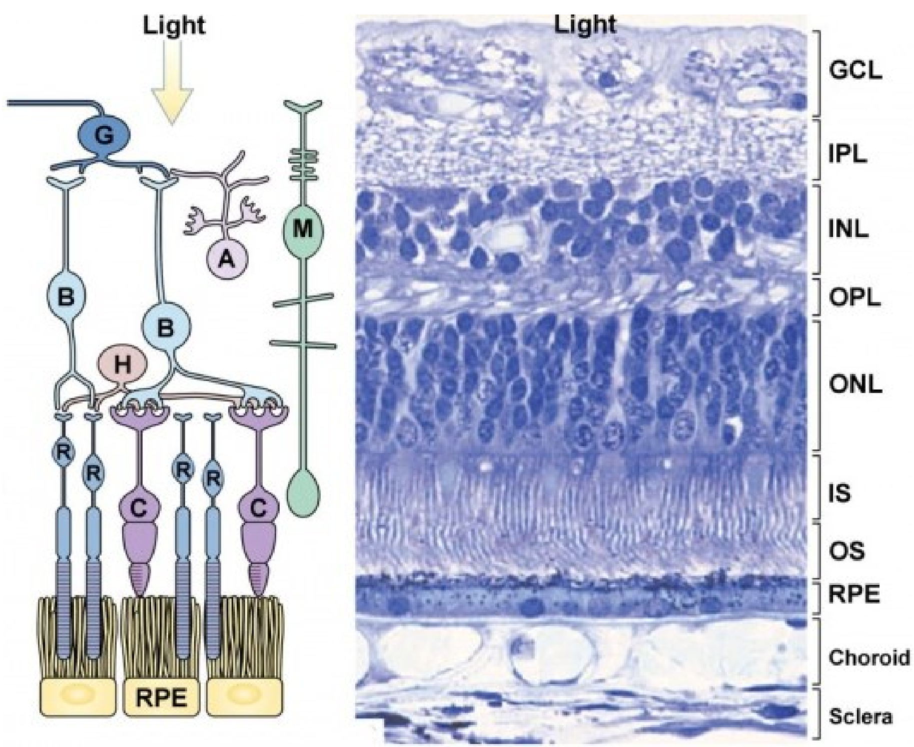
Larriva-Sahd 2010 Front Neuroanat

Other layered structures in the brain

Cerebellum

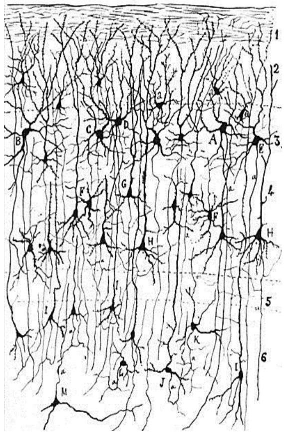


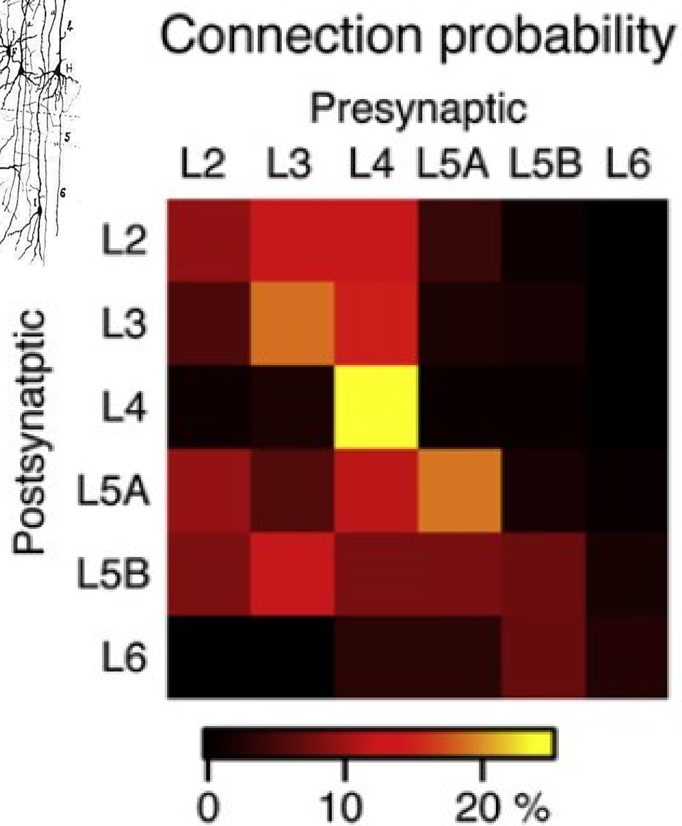
Retina



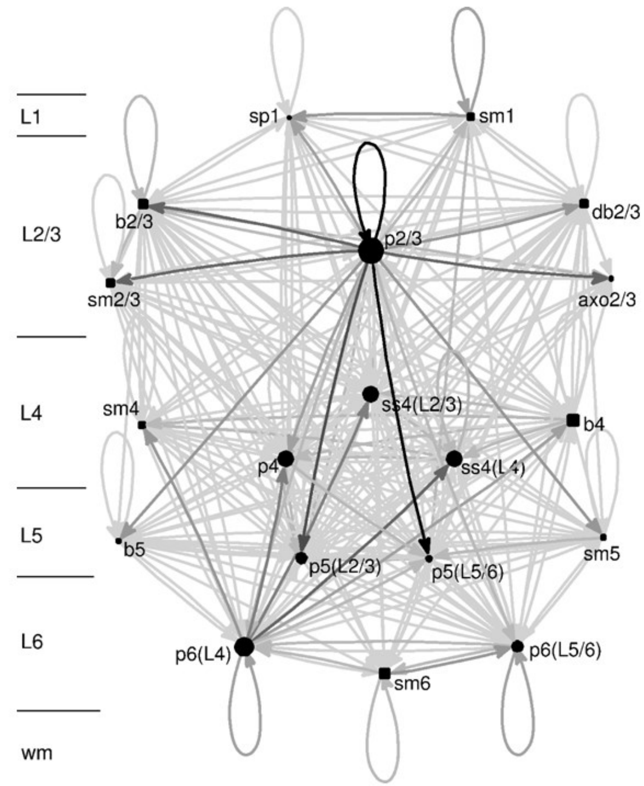
Complexity of connectivity in a cortical column

- Paired-recordings and anatomical reconstructions generate statistics of cortical connectivity



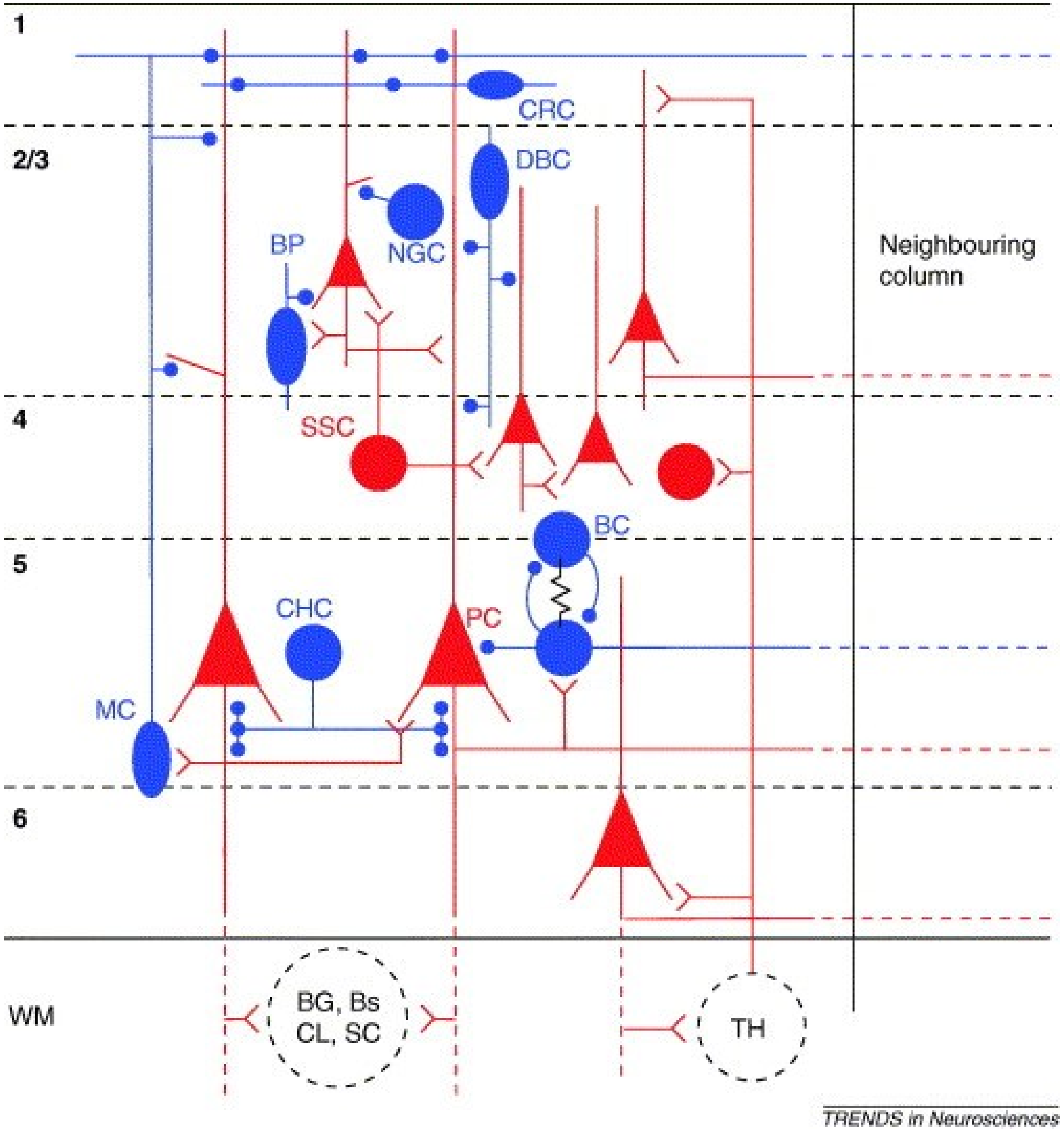


Lefort et al. 2009

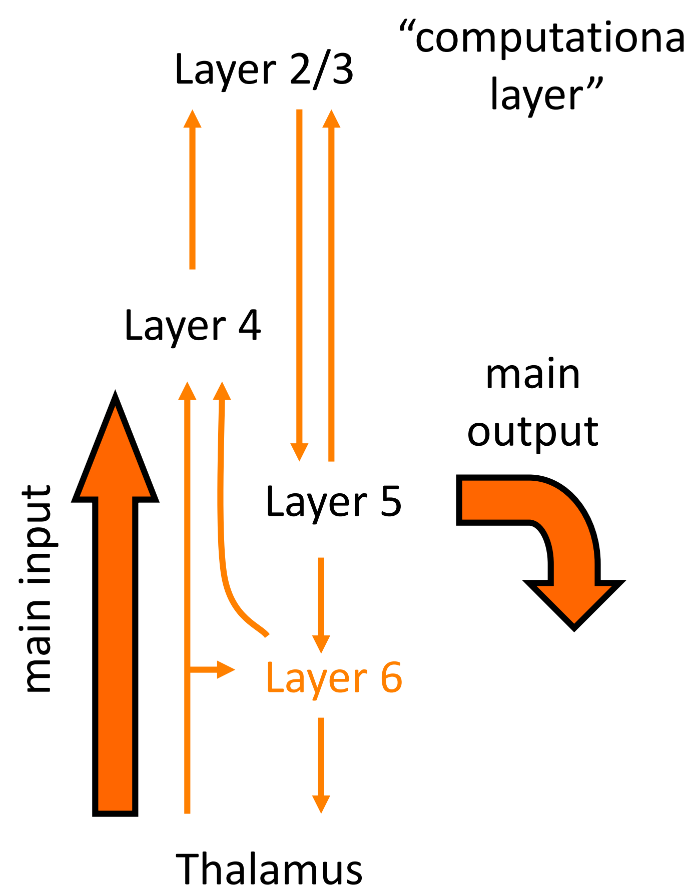


T. Binzegger et al. / Neural Networks 22 (2009) 1071–1078

Information flow in neocortical microcircuits



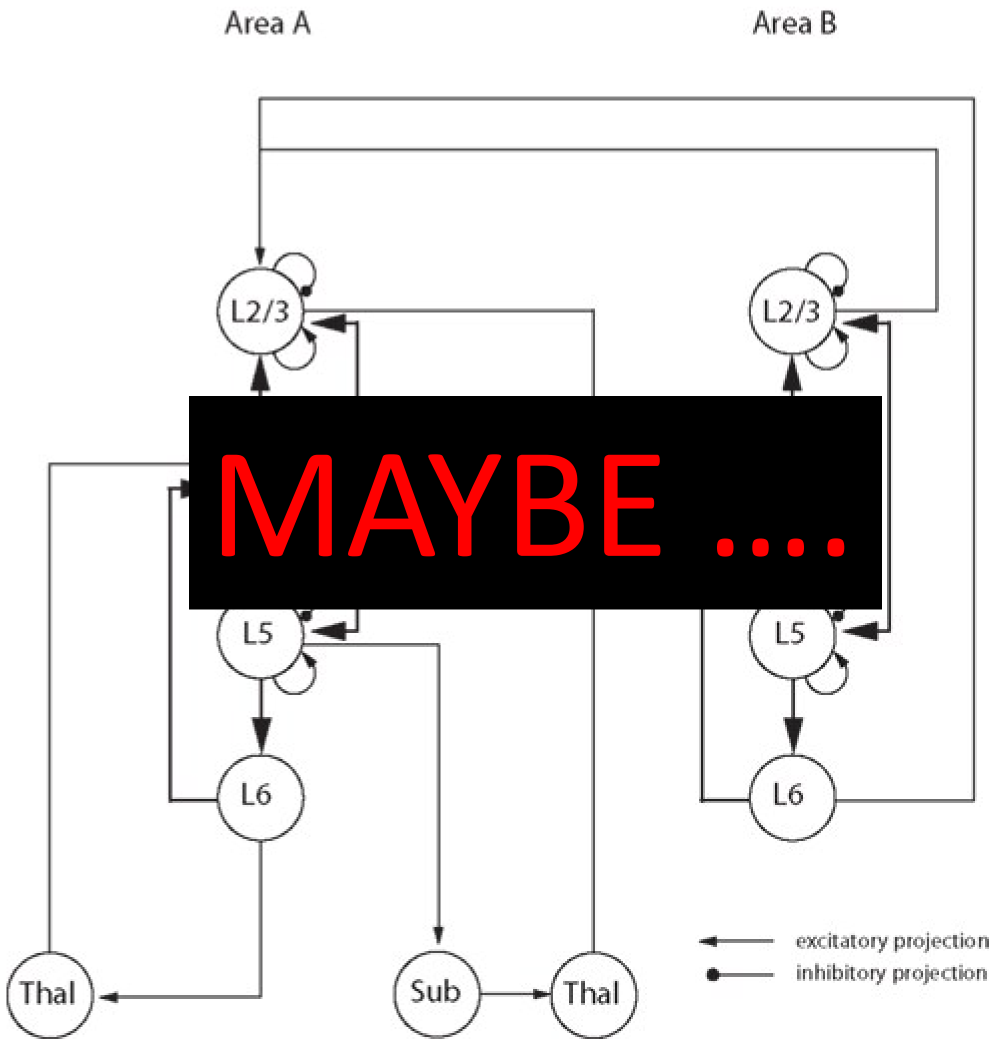
Simplified version



e - excitatory, i - inhibitory

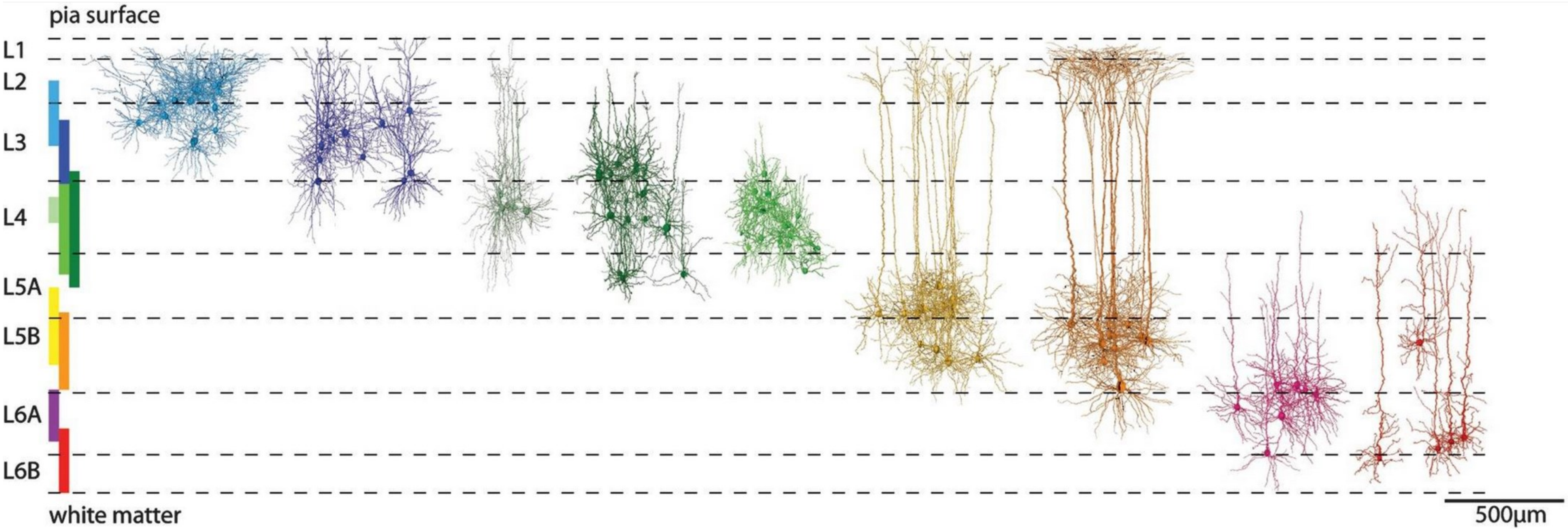
Grillner et al TINS 2005

The canonical cortical circuit



DaCosta & Martin, 2010

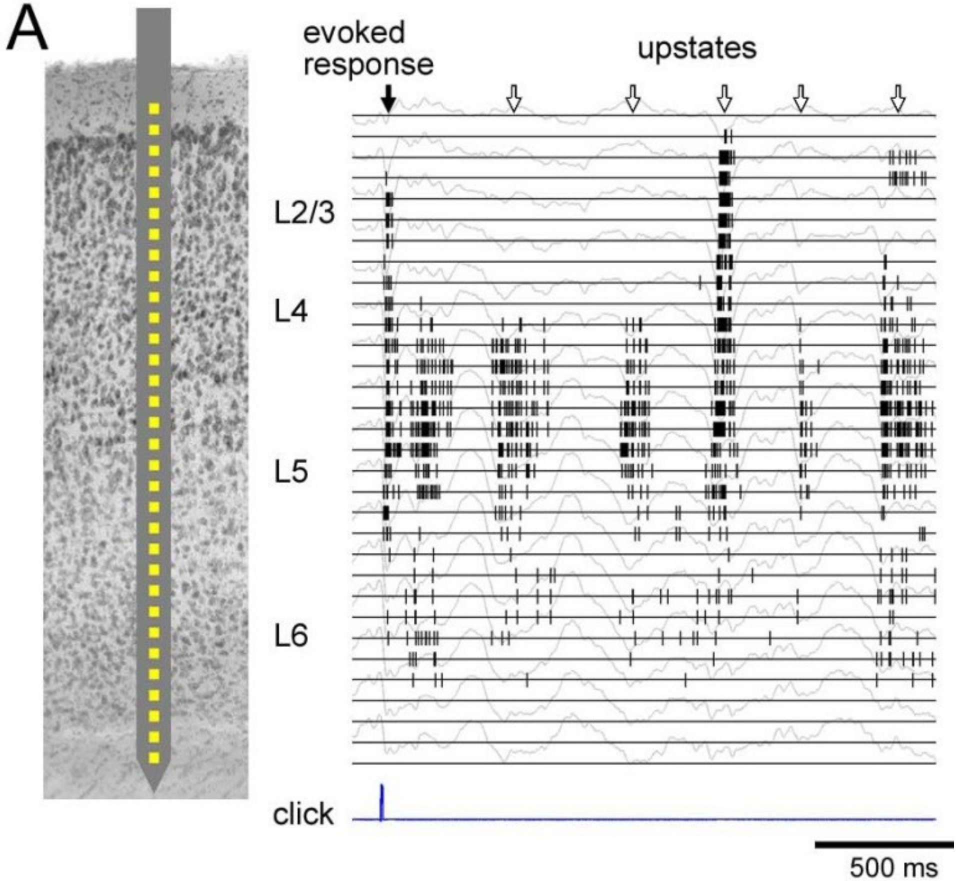
Excitatory cell types across layers (rat S1 cortex)



Canonical models do not capture the diversity of excitatory cell classes

Oberlaender et al., Cereb Cortex. Oct 2012; 22(10): 2375–2391.

Coding strategies of different cortical layers



Sakata & Harris, Neuron 2010

Canonical models do not capture the diversity of firing rates and selectivities

Why is the cortex layered?

Do different layers have distinct functions?

Is this the right question?

Alternative view:

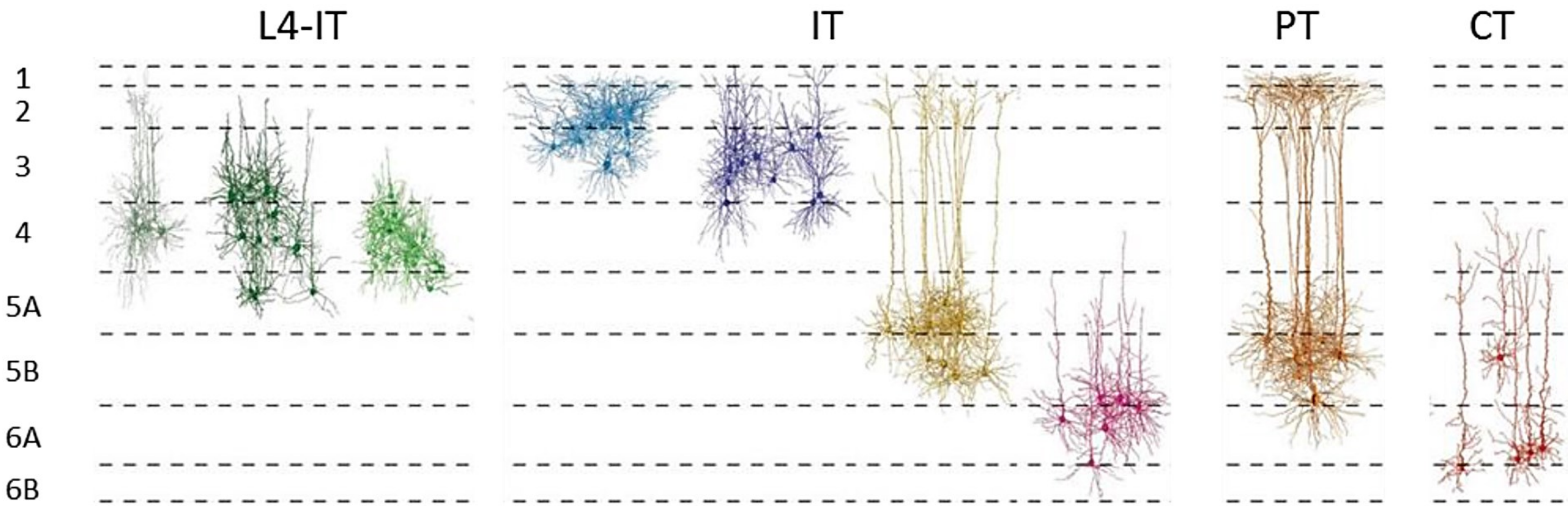
- When thinking about layers, we should really be thinking about cell classes

- A cells class may be defined by its *input connectome* and *output projectome (*and some other properties*)*

- The job of different classes is to (i) make associations between different types of information available in each cortical column and/or (ii) route/gate different streams of information

- Layers are convenient way of organising inputs and outputs of distinct cell classes

Excitatory cell types across layers (rat S1 cortex)



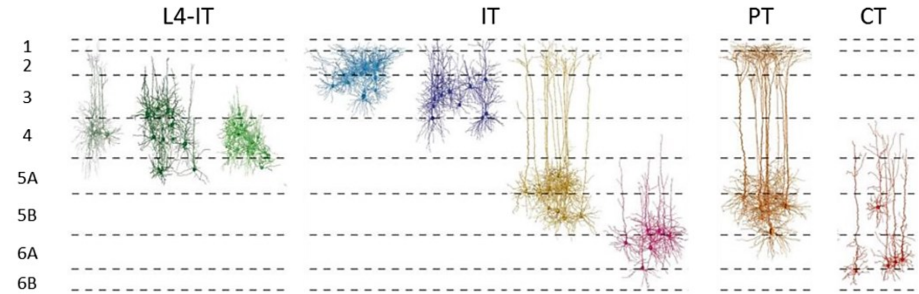
INTRATELENCEPHALIC (**IT**) | PYRAMIDAL TRACT (**PT**) | CORTICOTHALAMIC (**CT**)

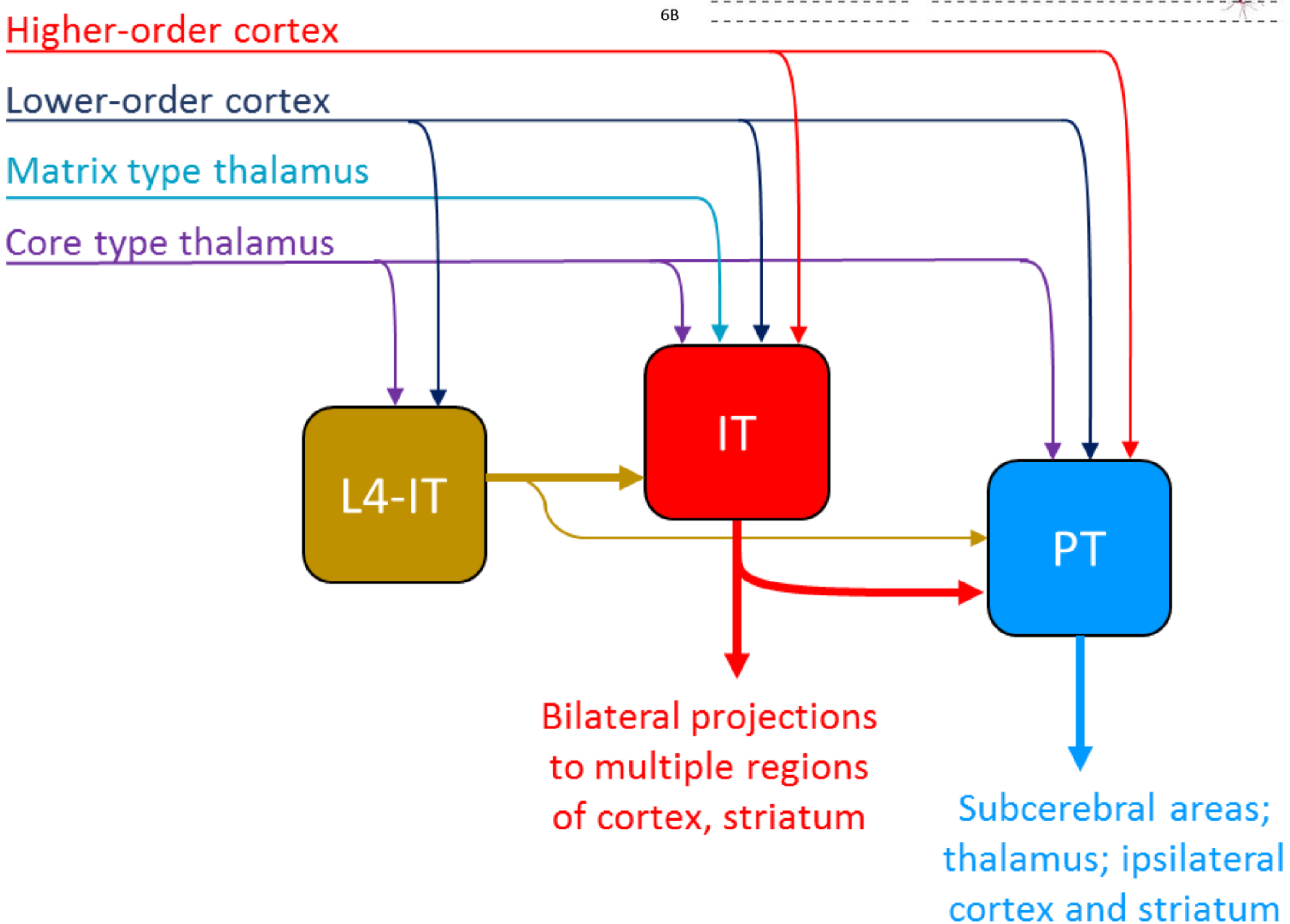
From Cereb Cortex. Oct 2012; 22(10): 2375–2391.

Modified by Harris and Shepherd, 2014

Hodology* of main excitatory cortical classes

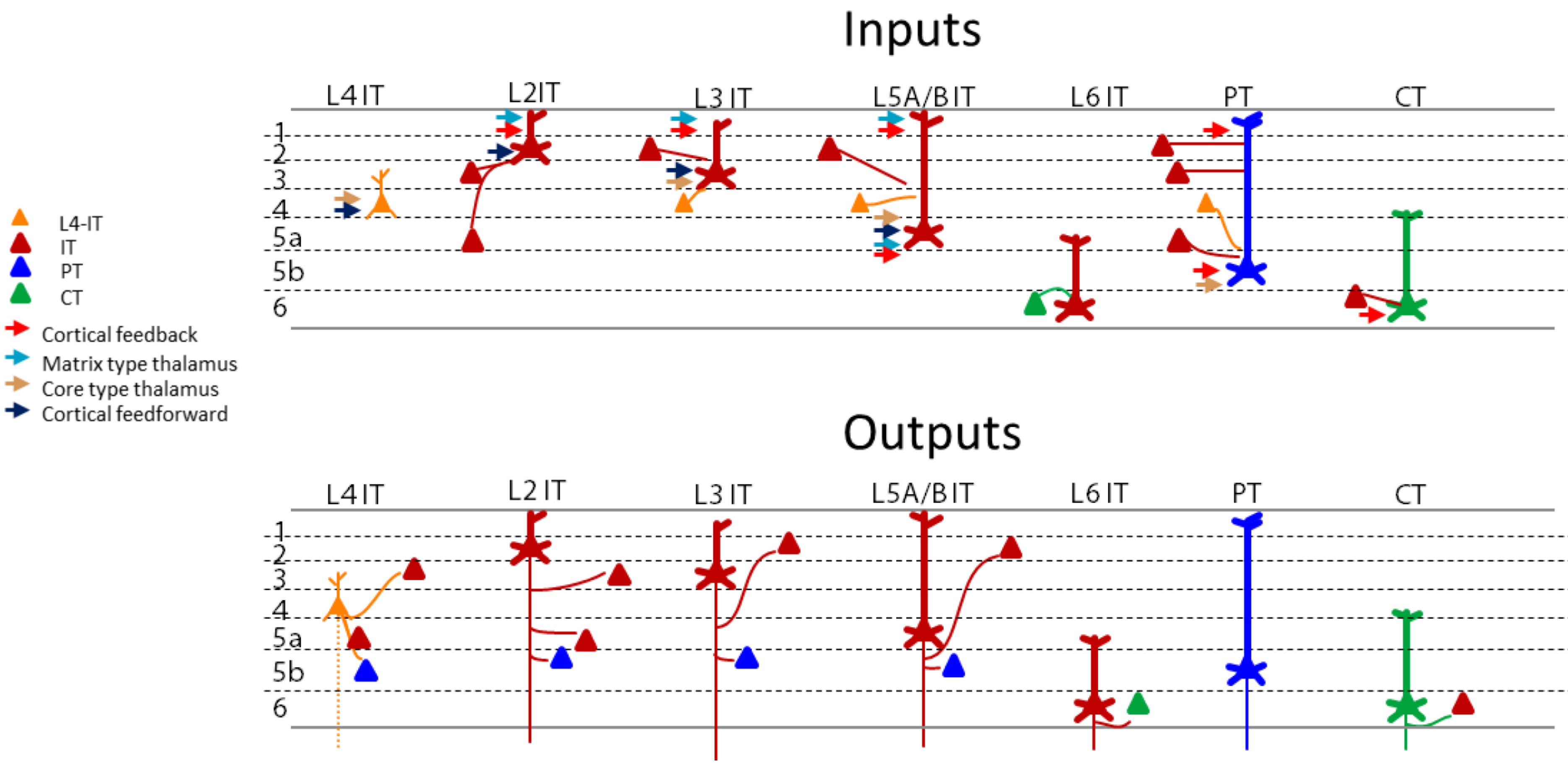
** the patterns/rules of connectivity*





Harris and Shepherd, 2014, review

Putative hodology of main excitatory cell classes



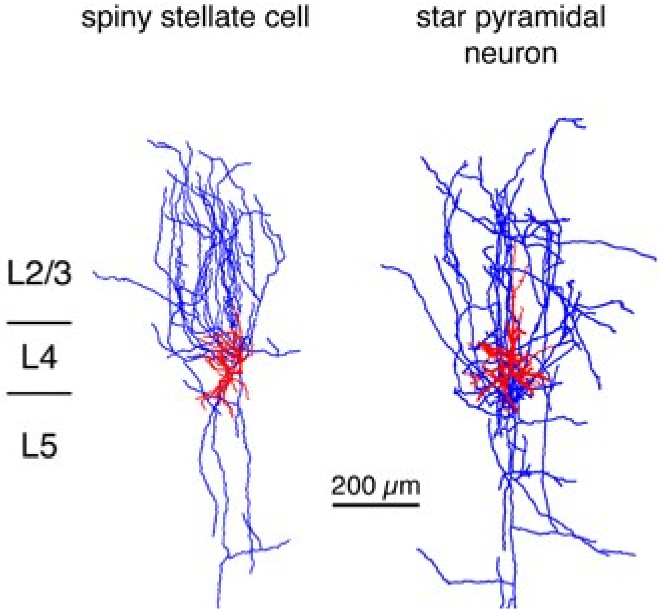
Harris and Shepherd, 2014, review,

	IT	IT	PT	CT
	L4	Other layers		
Genes	Rorb, Satb2	Satb2	Fezf2, Ctip2	Tbr1
Inputs from local E cell classes	Few	Many, incl. L4 IT and other IT.	Many, mainly from IT	Few, mainly deep-layer (L5B/6) IT
Outputs to other local cell classes	Mainly IT, mainly L3. Sometimes PT.	IT (but not L4-IT), PT, CT	Few	Some interconnectivity with IT, possibly PT
Long-range inputs	Thalamus, lower-order cortex	Thlalamus, higher and lower-order cortex	Thalamus, higher and lower-order cortex	Higher order cortex
Long-range outputs	Few	Many, but only within telencephalon (neocortex, striatum); the only ECs sending callosal/commisural projections	Many, to multiple subcortical and subcerebral regions (brainstem, tectum, spinal cord, thalamus, basal ganglia)	Thalamus. The only ECs to excite reticular nucleus. The only ECs without longer-range corticocortical axons.
Morphology/Layers	L4 pyramidal/stellate	L2/3, 5A, 5B, 6; pyramidal	L5B, thick tufted pyramidal	L6, pyramidal
Intrinsic physiology	Regular spiking or bursting	Hyperpolarized (L2/3), little h-current, spike train adaption	Depolarized, strong h-current, little adaptation, bursting (subset)	Regular spiking
In vivo activity	Fast sensory response	Sparse firing/code	Dense code	Very sparse

IT: intratelencephalic neurons; **PT:** pyramidal tract neurons of L5B; **CT:** corticothalamic neurons of L6.

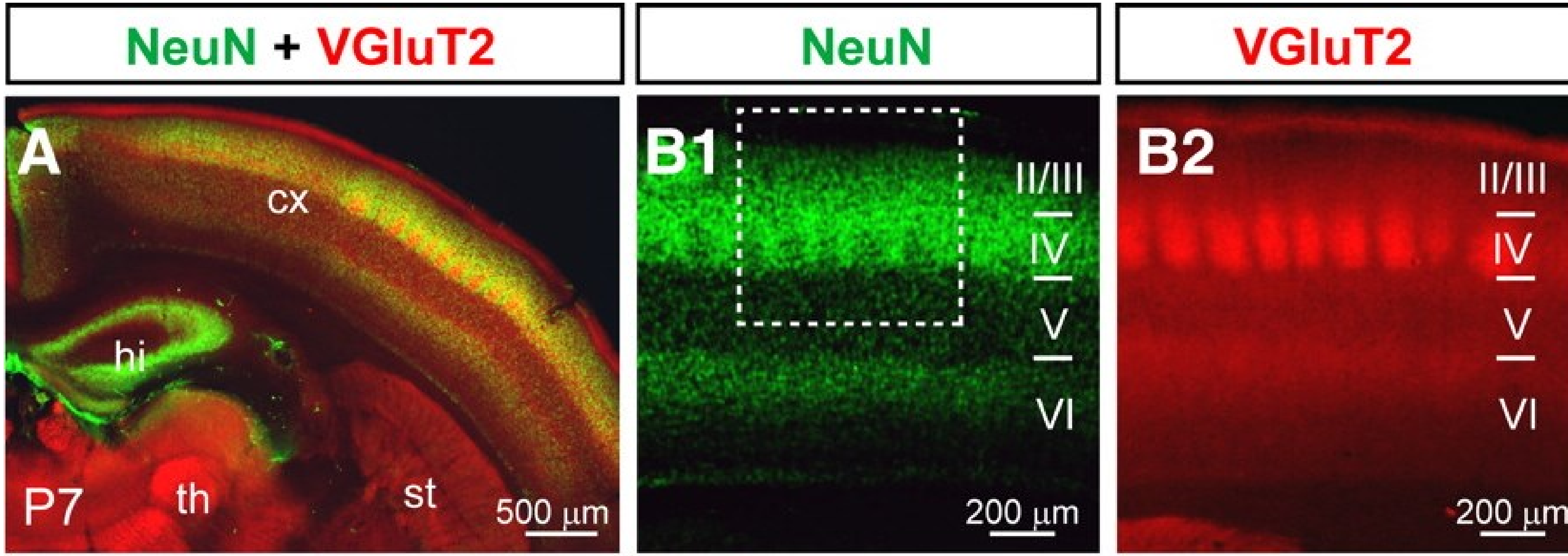
Layer 4

- Main input layer
- Specialised for each sensory modality
- Contains L4 IT neurons that project within a column
- Creation of new sensory response properties

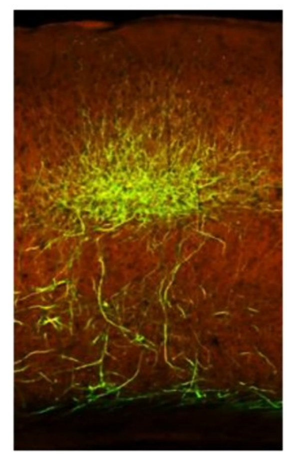


Layer 4: input specialisations

VGLUT2 immunoreactivity (TC axons)



Belester-Rosado et al 2010 J Neurosci



Layer 4: input specialisations

Primary visual cortex

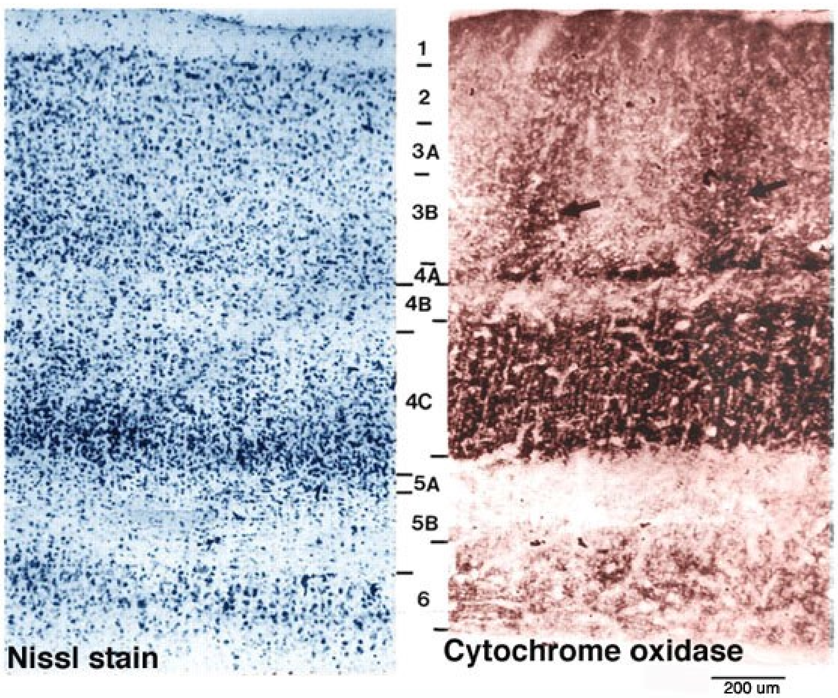
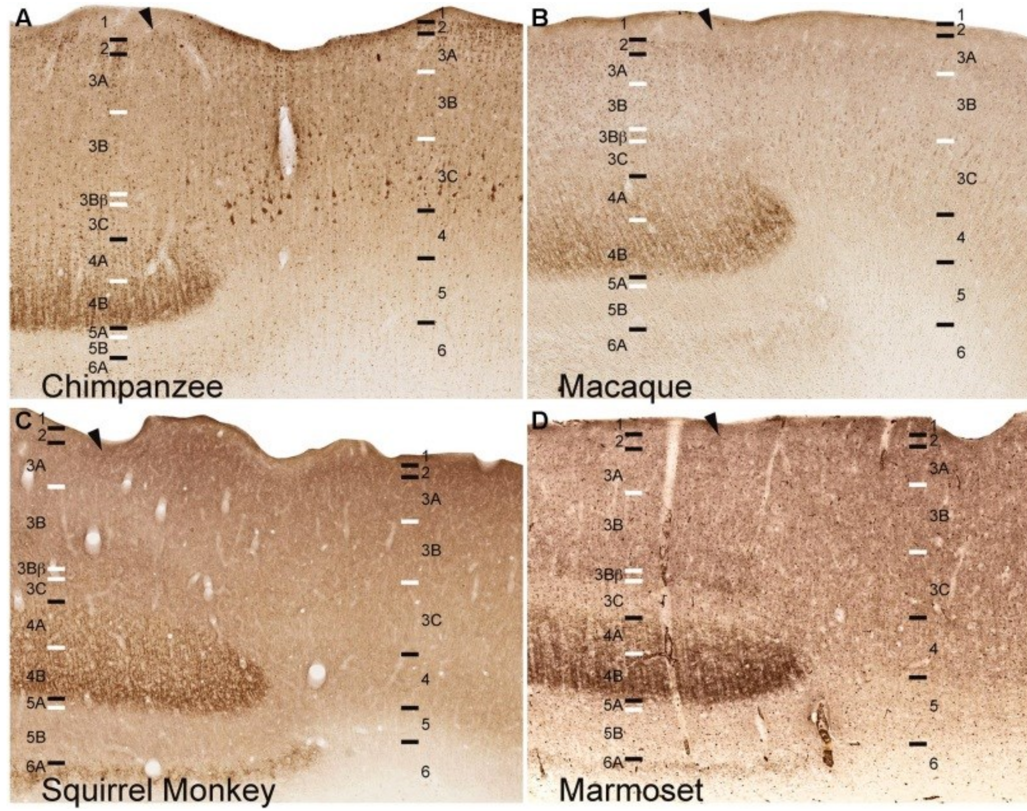


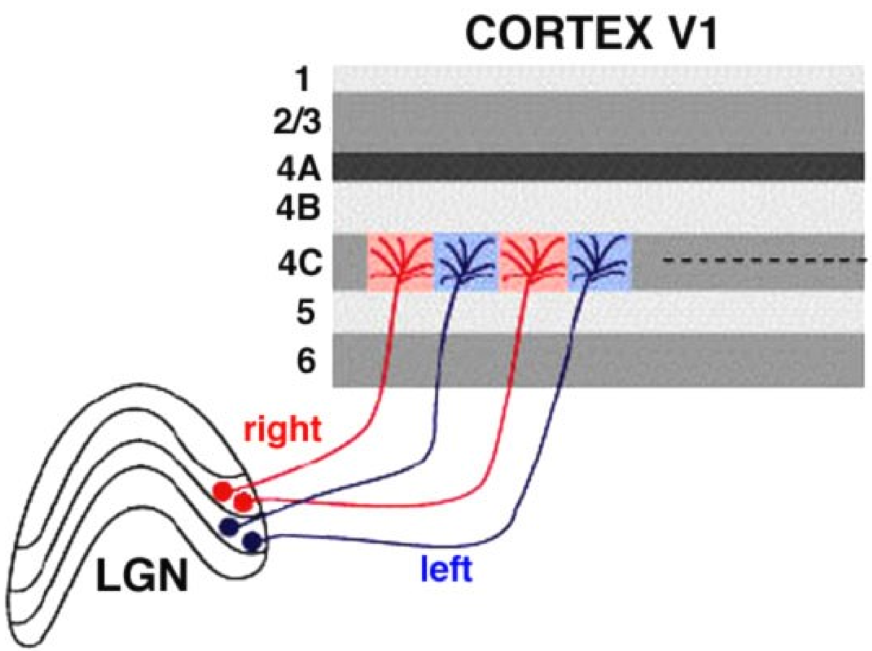
Figure 10. Nissl (left) and cytochrome oxidase (right) labeled cross sections of the visual cortex of a macaque monkey, showing the individual layers.

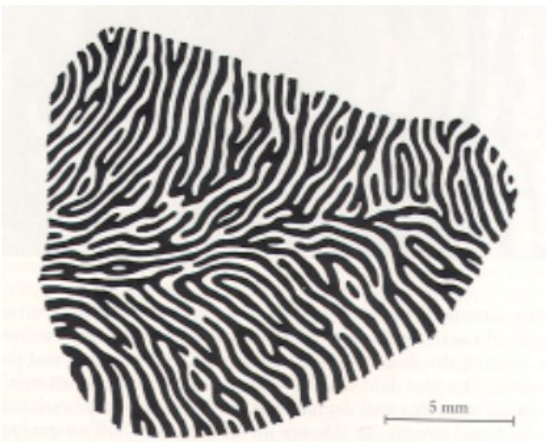


VGLUT2 immunoreactivity (TC axons)

Balaram & Kaas 2014 Front Neuroana

Ocular dominance visual cortex





Reconstruction of OD columns from area 17 of cat cortex

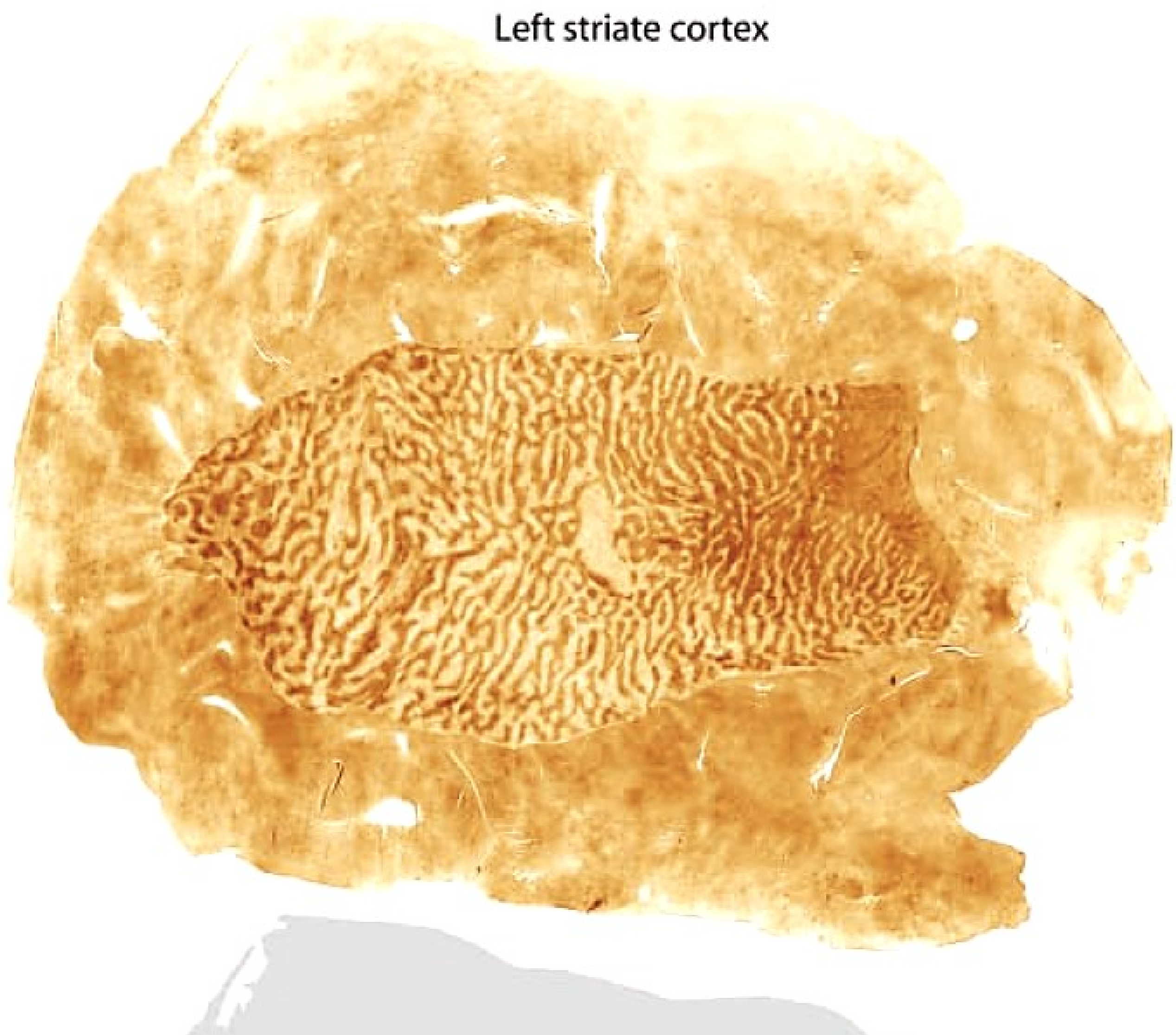
Cells in layers 4Cα and 4Cβ are driven by the input from one eye only.

If a particular cells receives input from the right eye, cells above and below it will also receive input from the right eye.

However, adjacent cells either side will receive their input from the opposite eye.

The pattern of alternate eyes driving all the cells within a penetration perpendicular to the surface of the brain is referred to as **ocular dominance columns**.

Organisation of ocular dominance in layer 4C in human V1

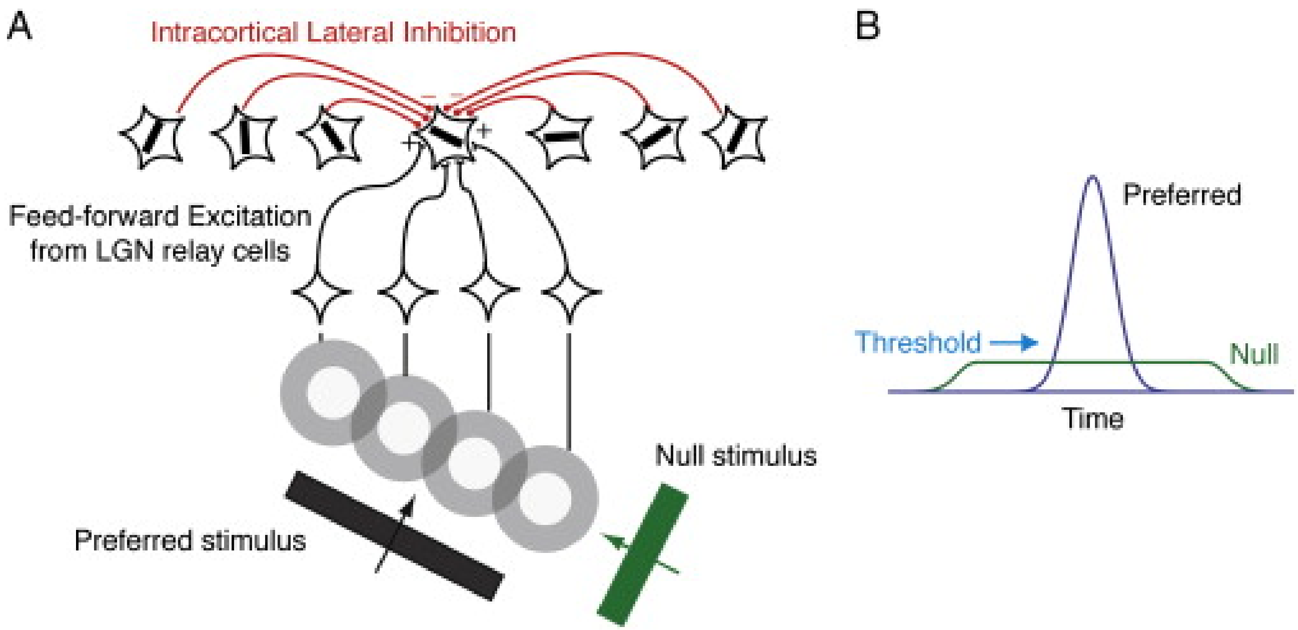


Cytochrome oxidase staining in **layer 4C** of human brain

Horton & Adams 2005

Layer 4

- Building new response properties via convergence/selection of TC inputs



Lateral geniculate nucleus (LGN)

ON cell

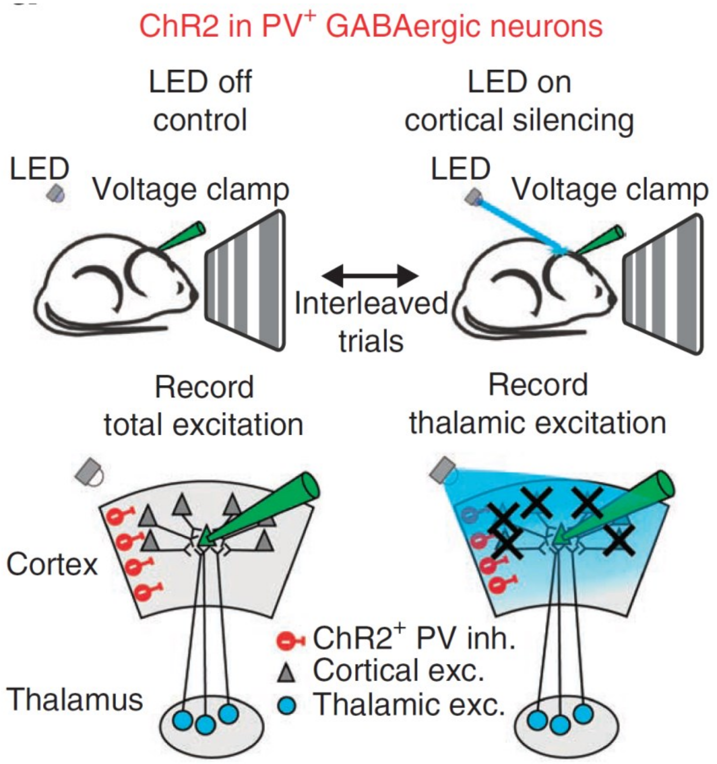
Primary visual cortex (V1)

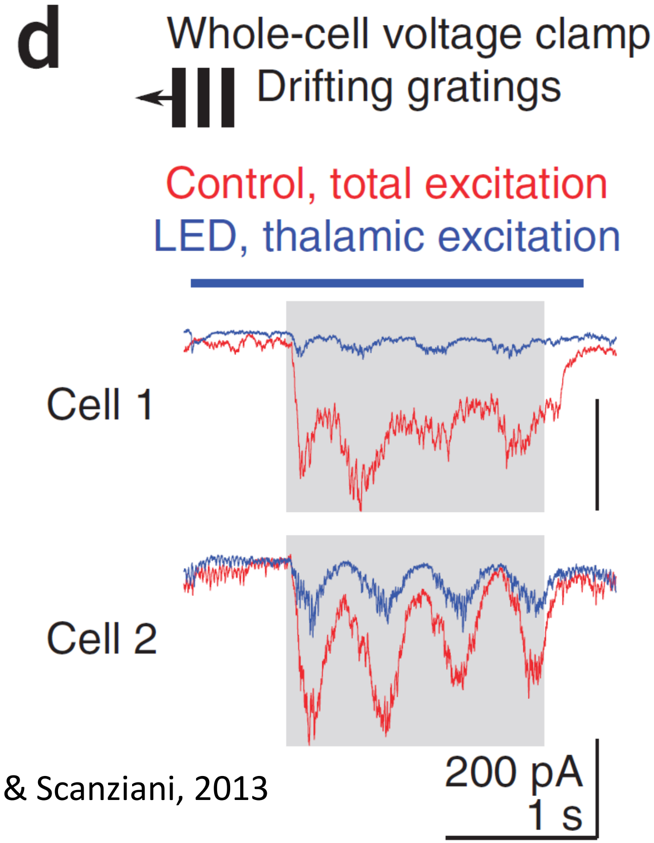
Orientation-preferring cell

Hubel and Wiesel

Layer 4

- Amplification of sparse and weak sensory input





Lien & Scanziani, 2013

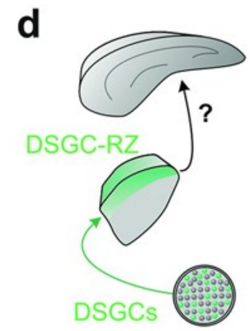
BUT…not all input from the primary thalamus terminates in L4!!

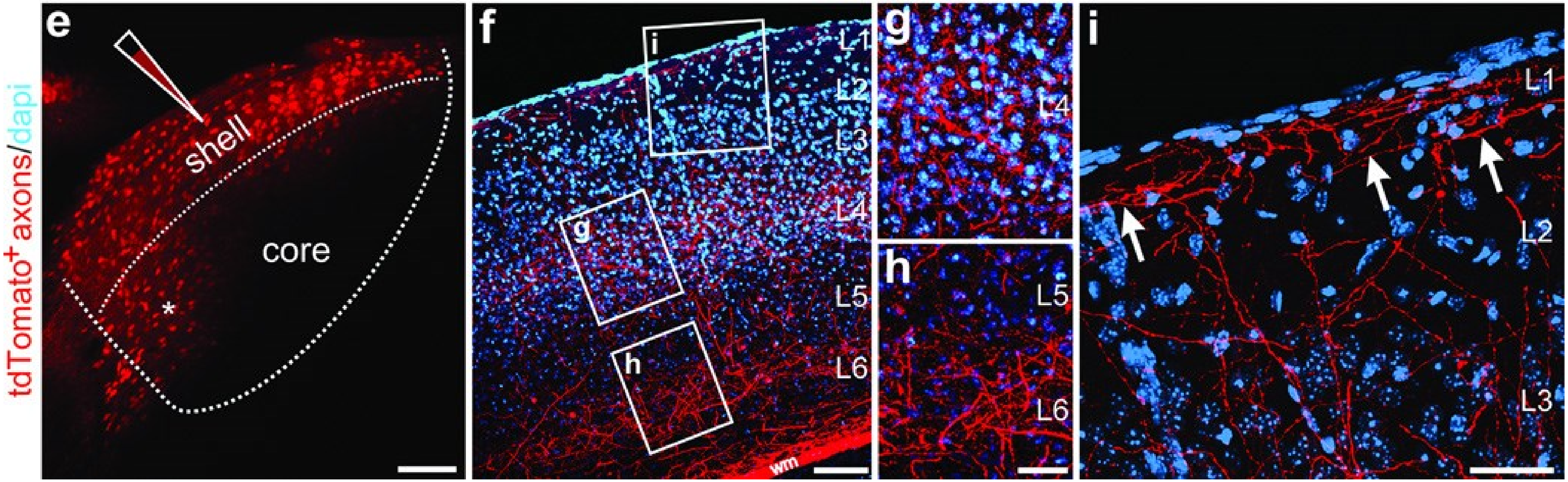
2 examples:

(i) LGN → L1/2/3 of mouse V1

(ii) VPN → L5 of mouse S1

Even projections from the primary thalamus can target superficial layers including L1





Cruz-Martin et al., Nature. Mar 20, 2014; 507(7492): 358–361.

BUT…not all input from the primary thalamus terminates in L4!!

2 examples:
(i) LGN → L1/2/3 of mouse V1
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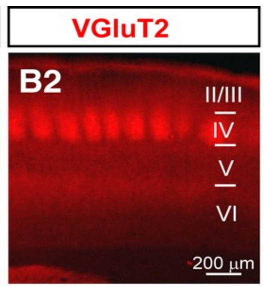
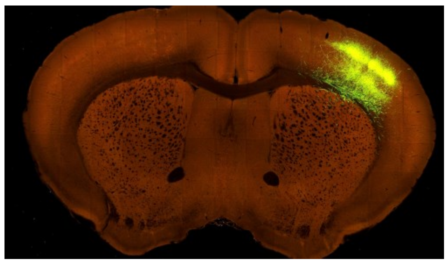
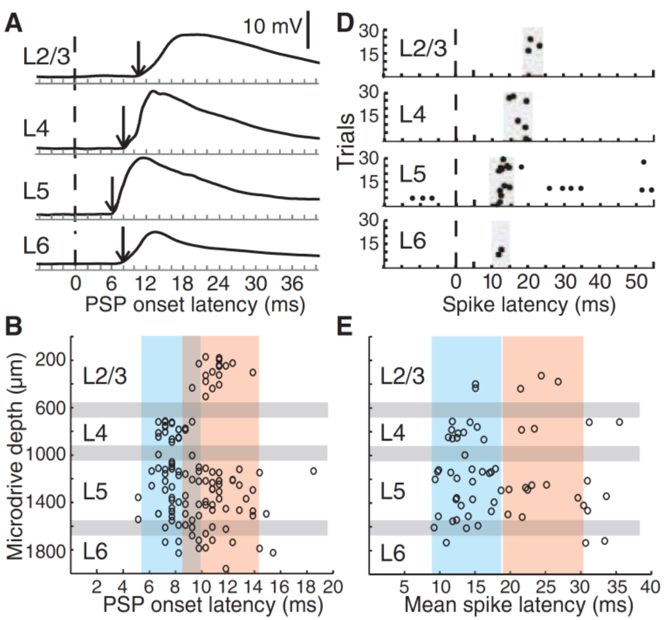
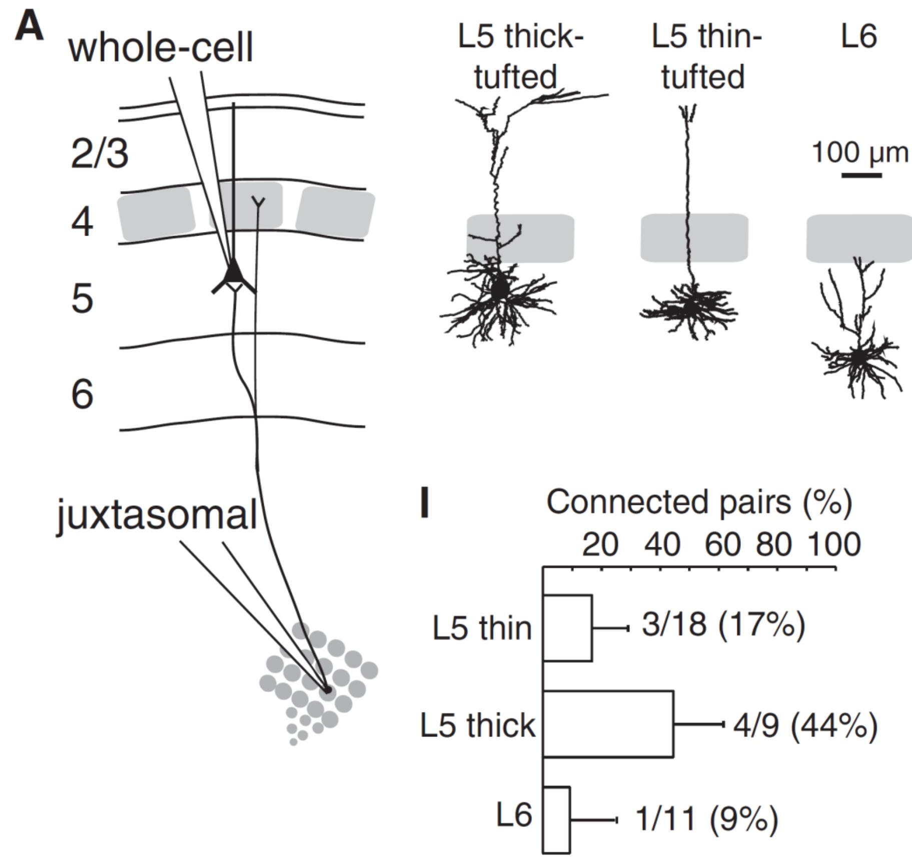








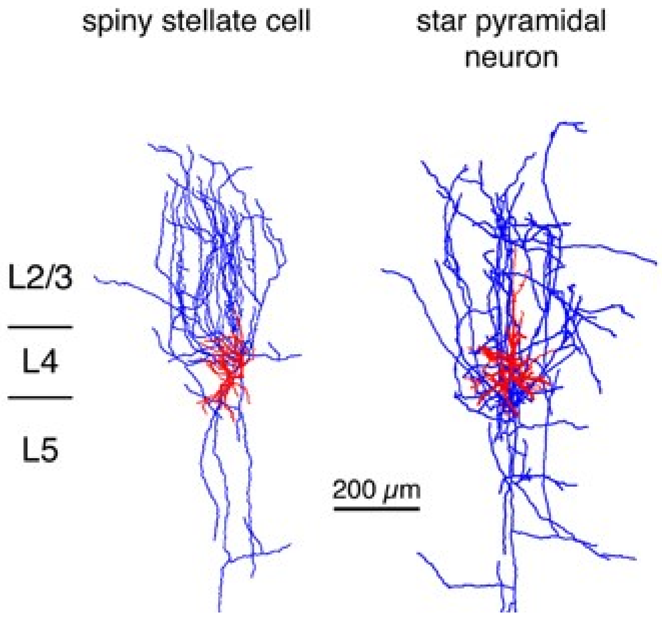
Constantinople & Bruno, 2013, Science

Layer 4

- Main input layer
- Specialised for each sensory modality **only in primary cortex (not a trend across cortex)**
- Contains L4 IT neurons that project within a column
- Creation of new sensory response properties

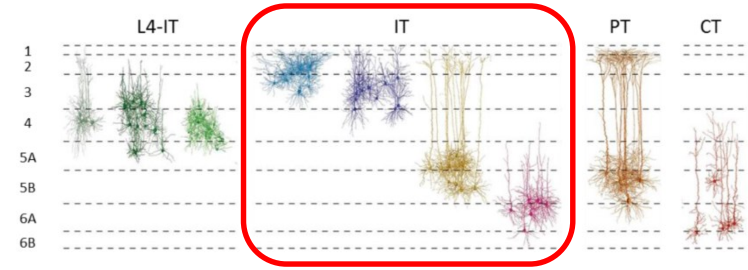
(e.g. orientation tuning in visual cortex but not always)

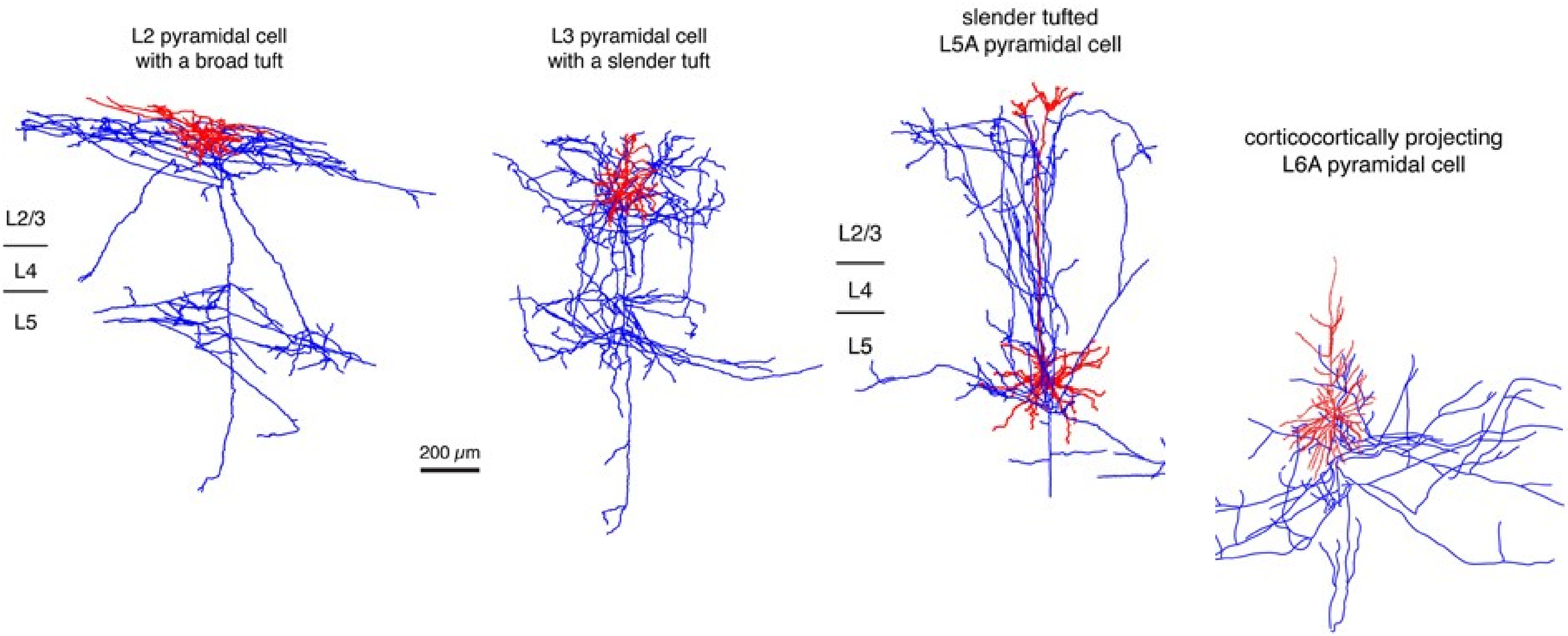
- Amplification of sparse sensory input
- But it is not unique in receiving primary sensory input
- Re-coding of peripheral sensory information into a cortical code



Intratelencephalic (IT) projection neurons

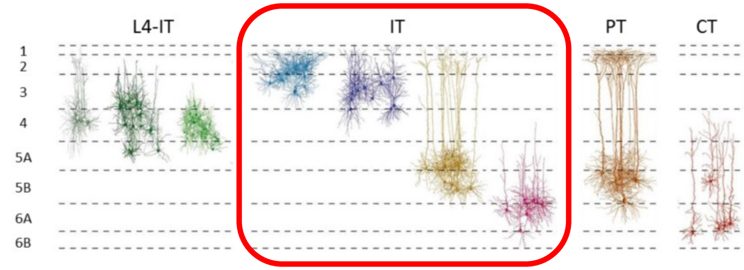
- Pyramidal cells (L2/3/5/6)
- Extensive local projections

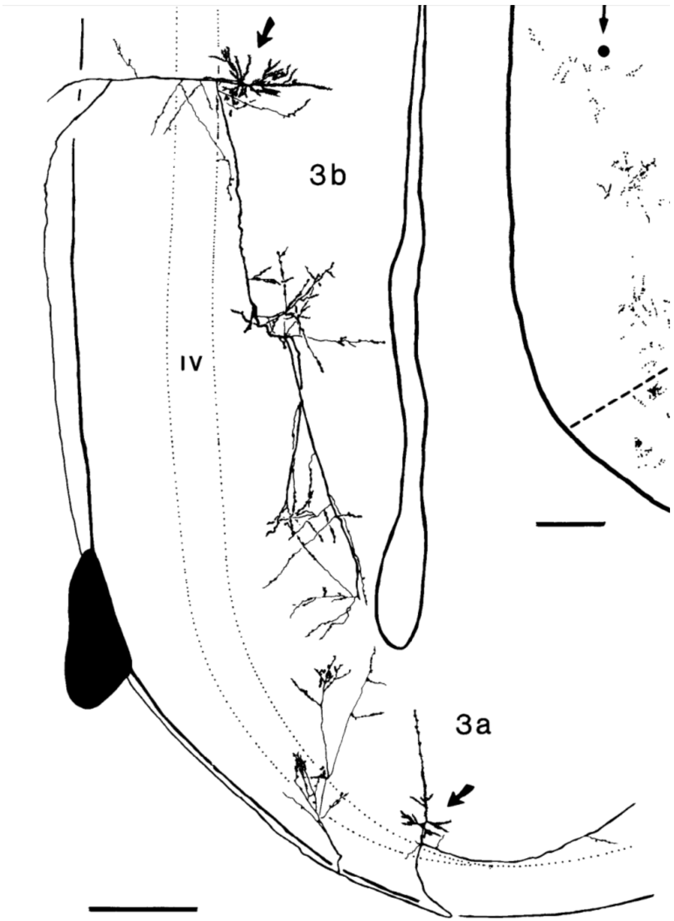




Intratelencephalic (IT) projection neurons

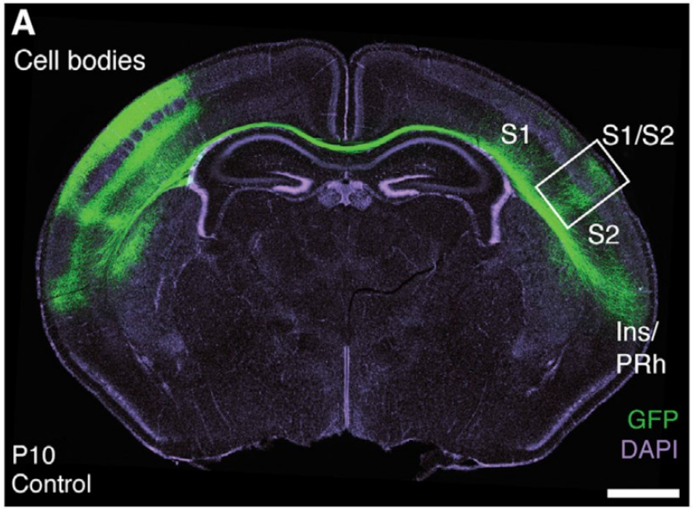
- Pyramidal cells (L2/3/5/6)
- Intrahemispheric, striatal and callosal projections

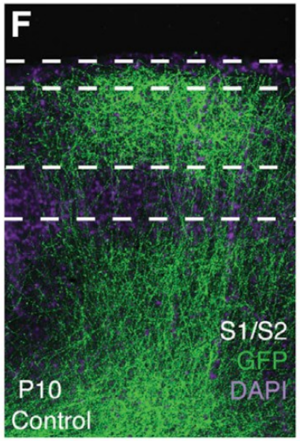




Long-Range Focal Collateralization of Axons Arising from Corticocortical Cells in Monkey Sensory-Motor Cortex

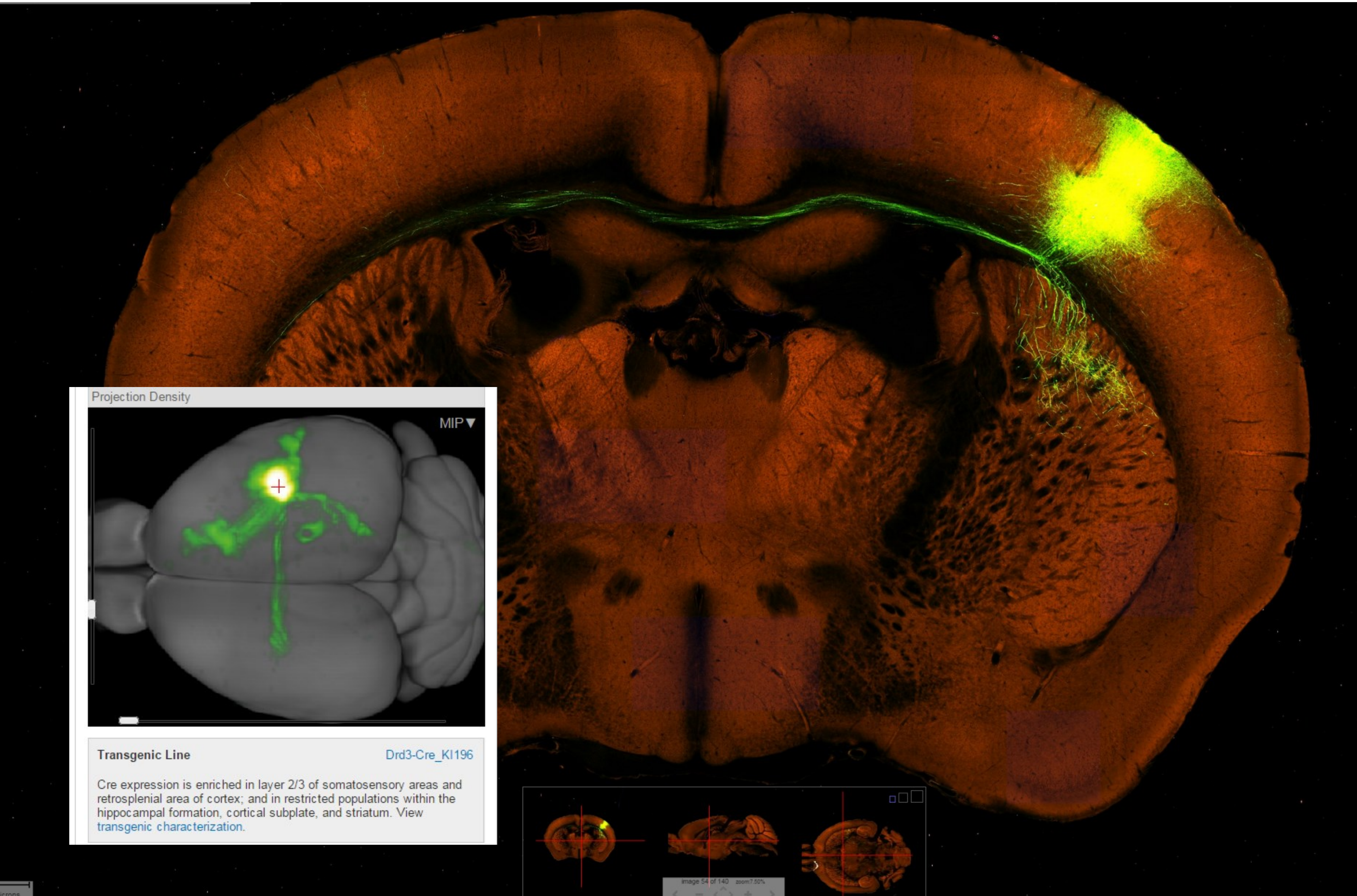
DeFelipe & Jones, 1986, JN





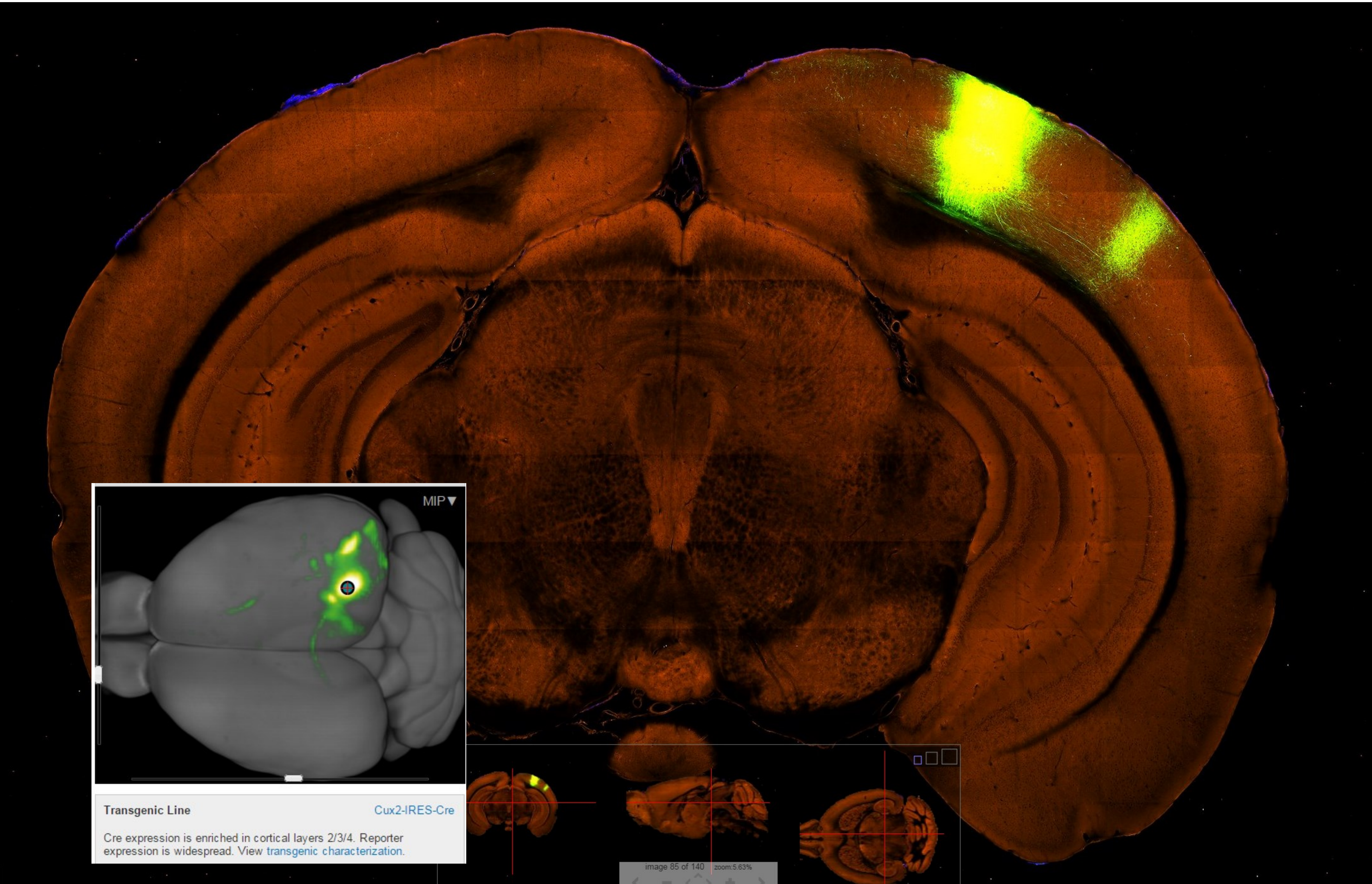
Suarez and Richards, 2014
Neuron

IT cells

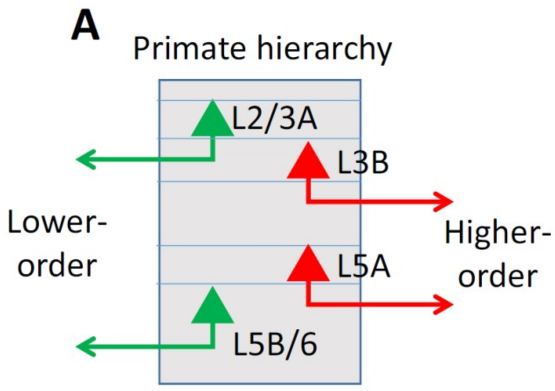


Projection Density
MIP▼
Transgenic Line
Drd3-Cre_KI196
Cre expression is enriched in layer 2/3 of somatosensory areas and retrosplenial area of cortex; and in restricted populations within the hippocampal formation, cortical subplate, and striatum. View transgenic characterization.

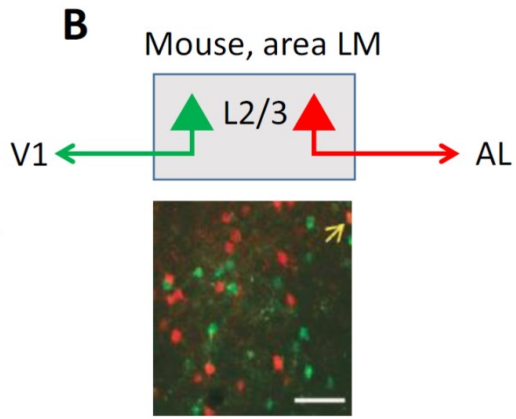
IT cells

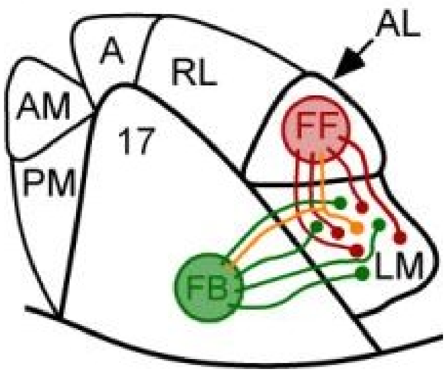

MIP▼
Transgenic Line
Cux2-IRES-Cre
Cre expression is enriched in cortical layers 2/3/4. Reporter expression is widespread. View transgenic characterization.
image 85 of 140
zoom:5.63%

How many classes of IT projection neurons?



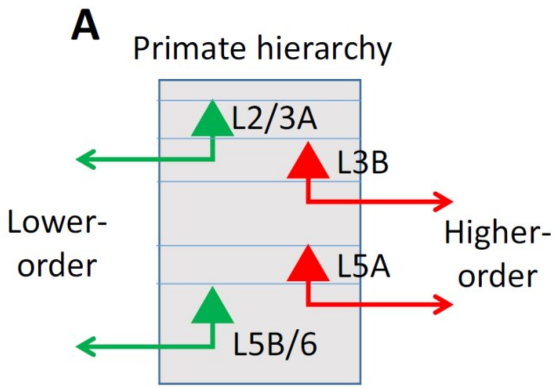
From Nassi & Callaway



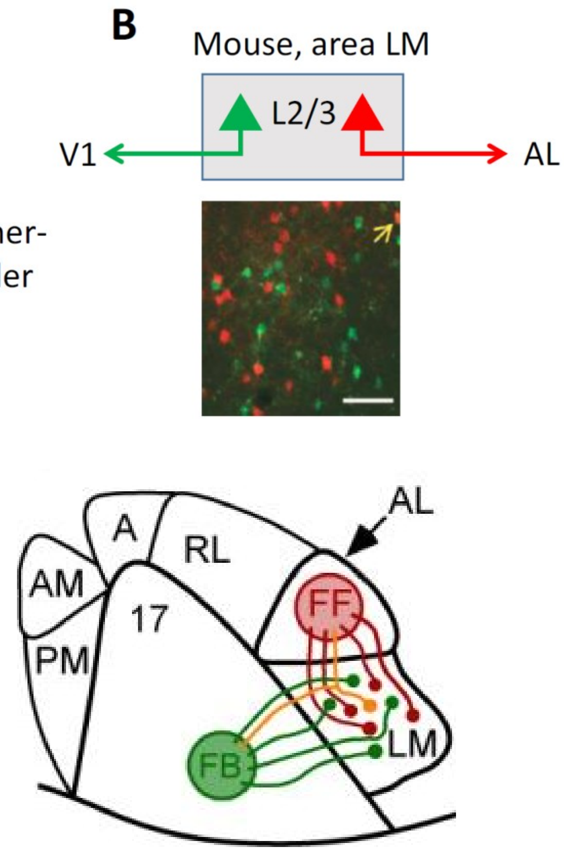


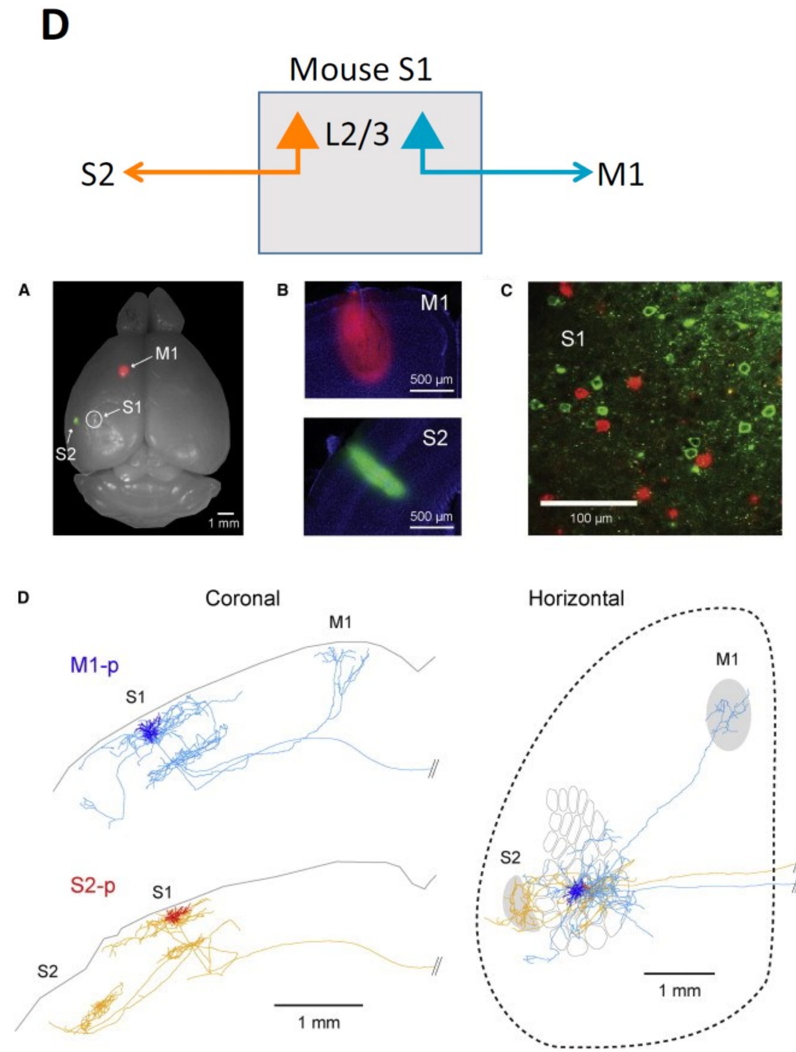
Adapted from Harris and Shepherd, 2014, review

How many classes of IT projection neurons?



From Nassi & Callaway

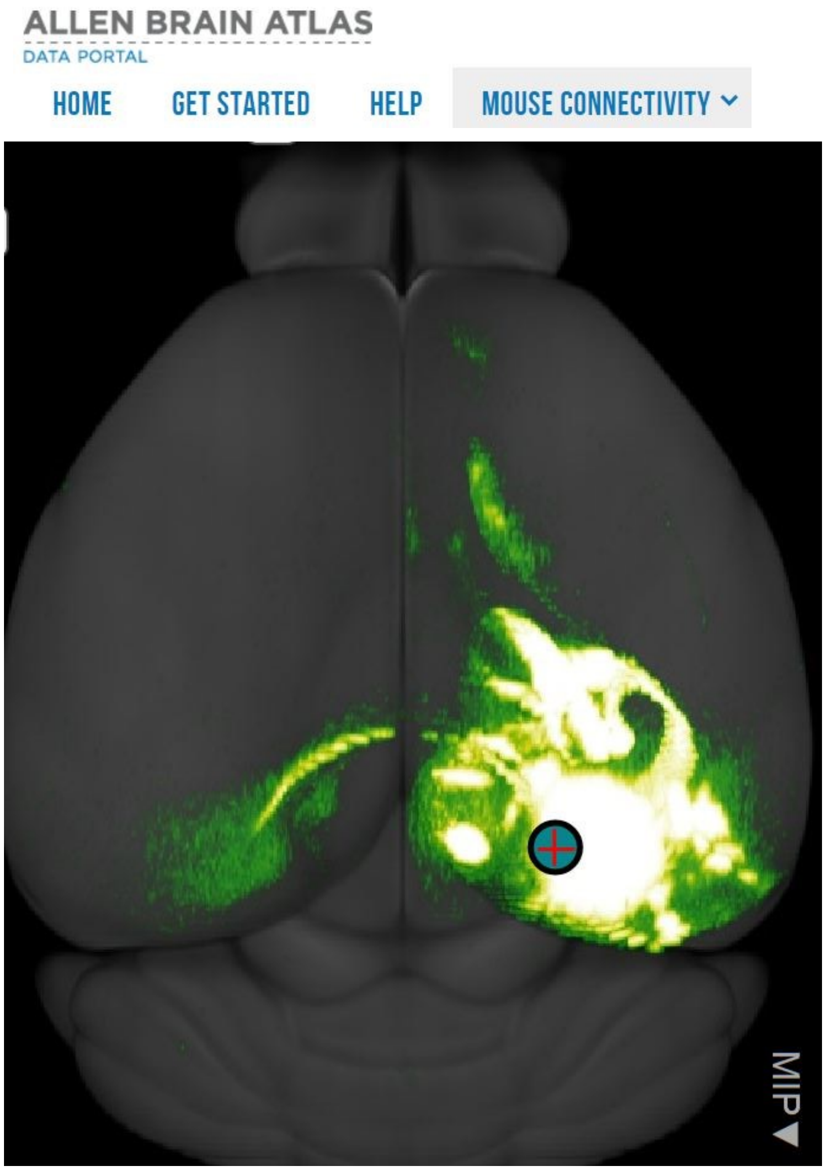




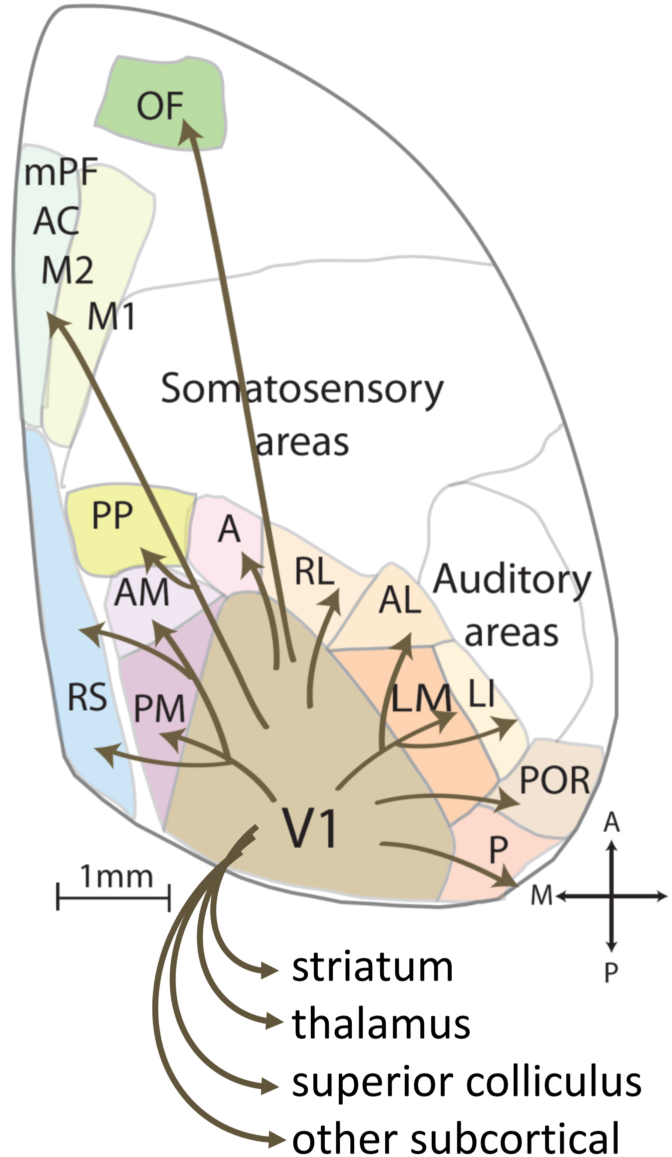
Yamashita & Petersen, Neuron, 2013

Adapted from Harris and Shepherd, 2014, review

Case study: How many types of IT projection neurons are there in V1?

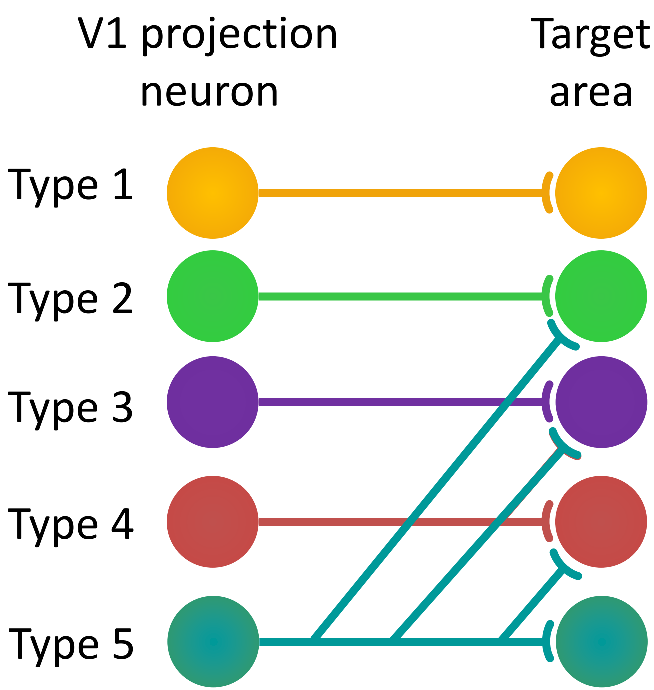


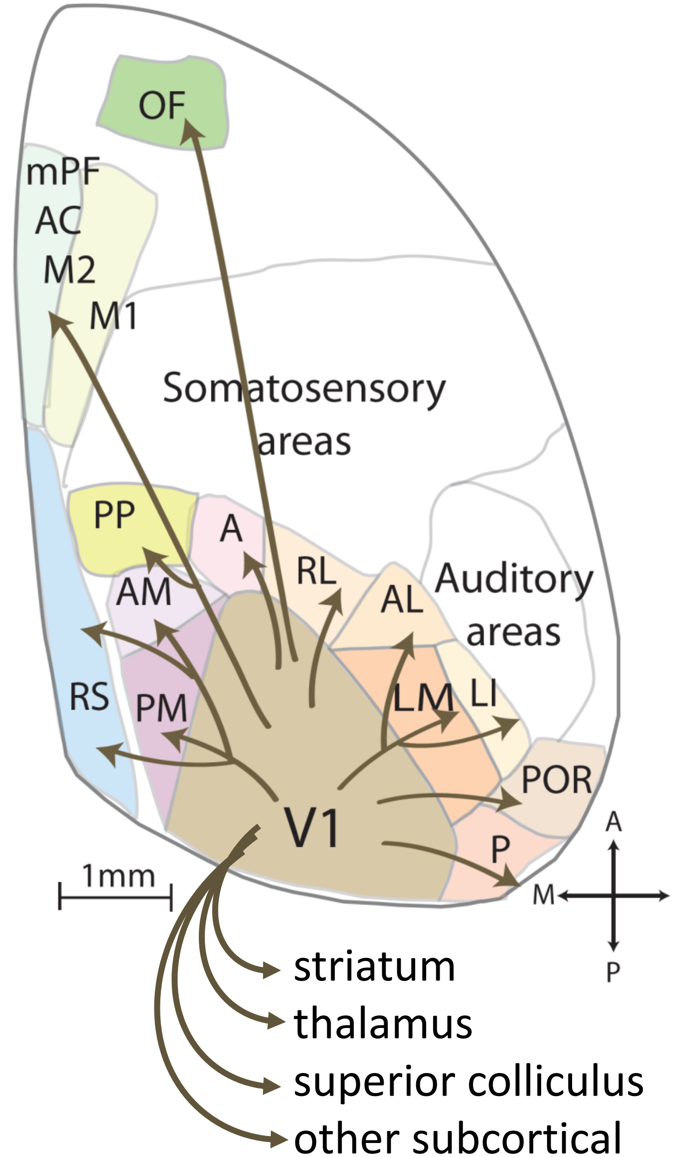
http://connectivity.brain-map.org/



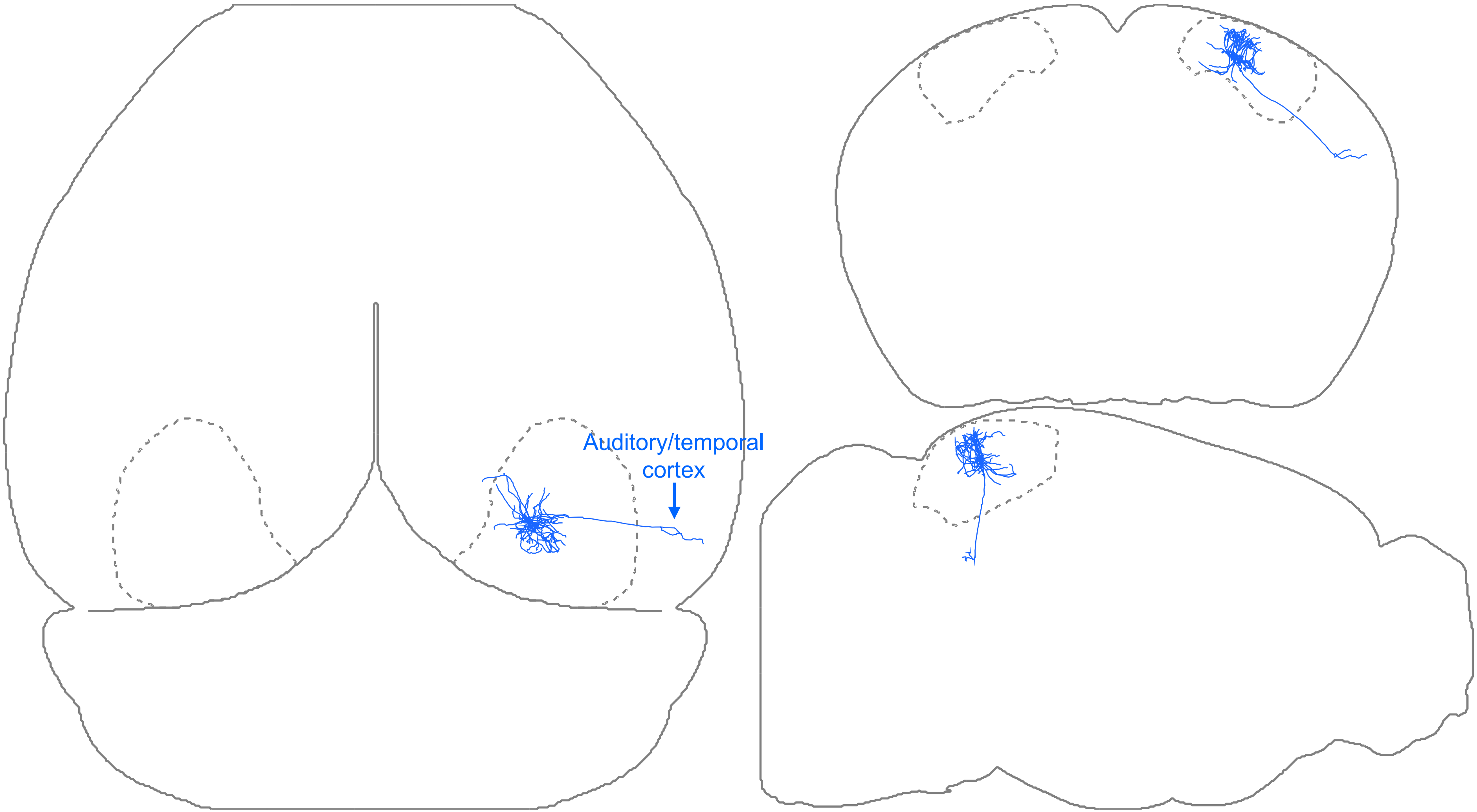
Understanding the logic of cortical projections

- How many projection classes?

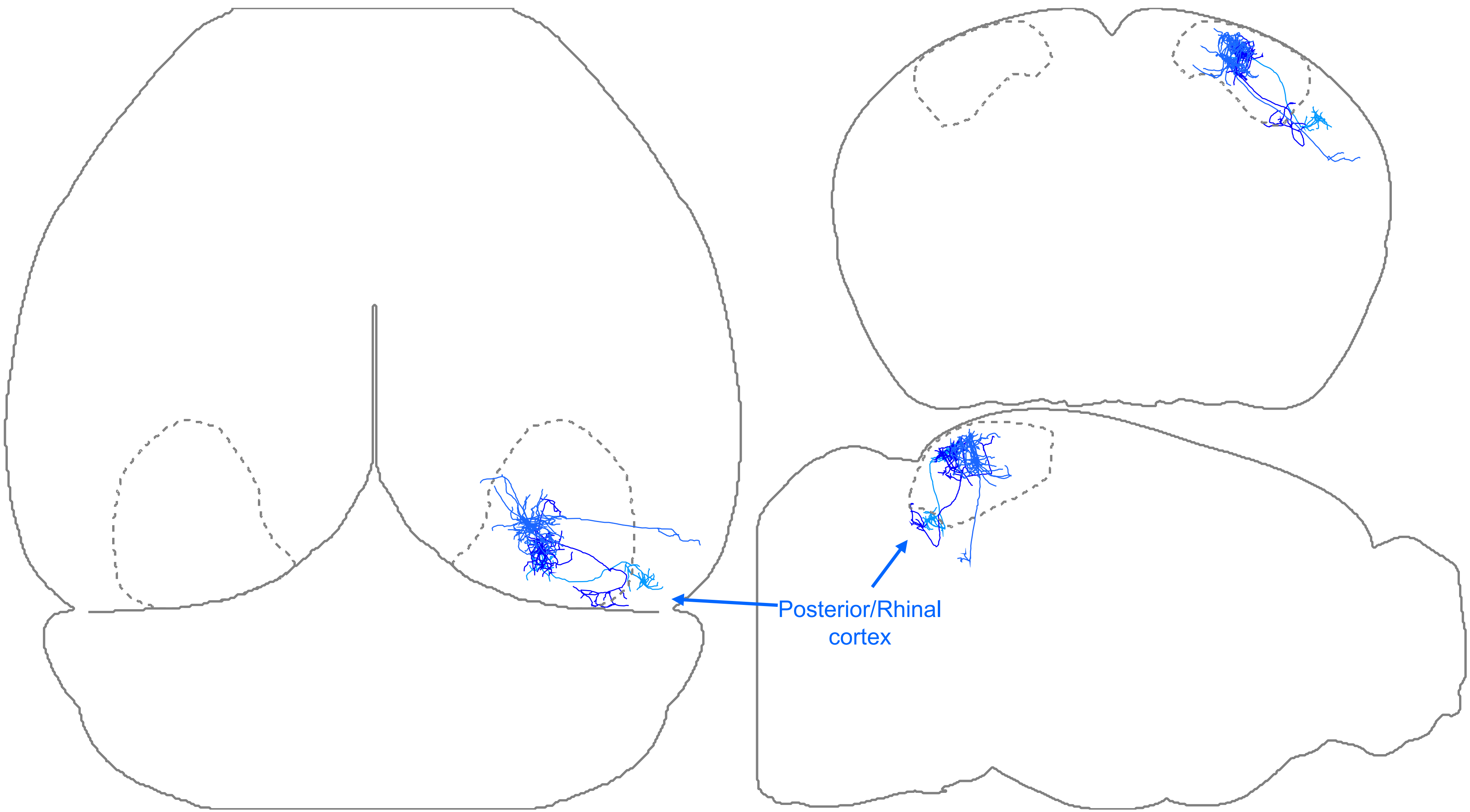




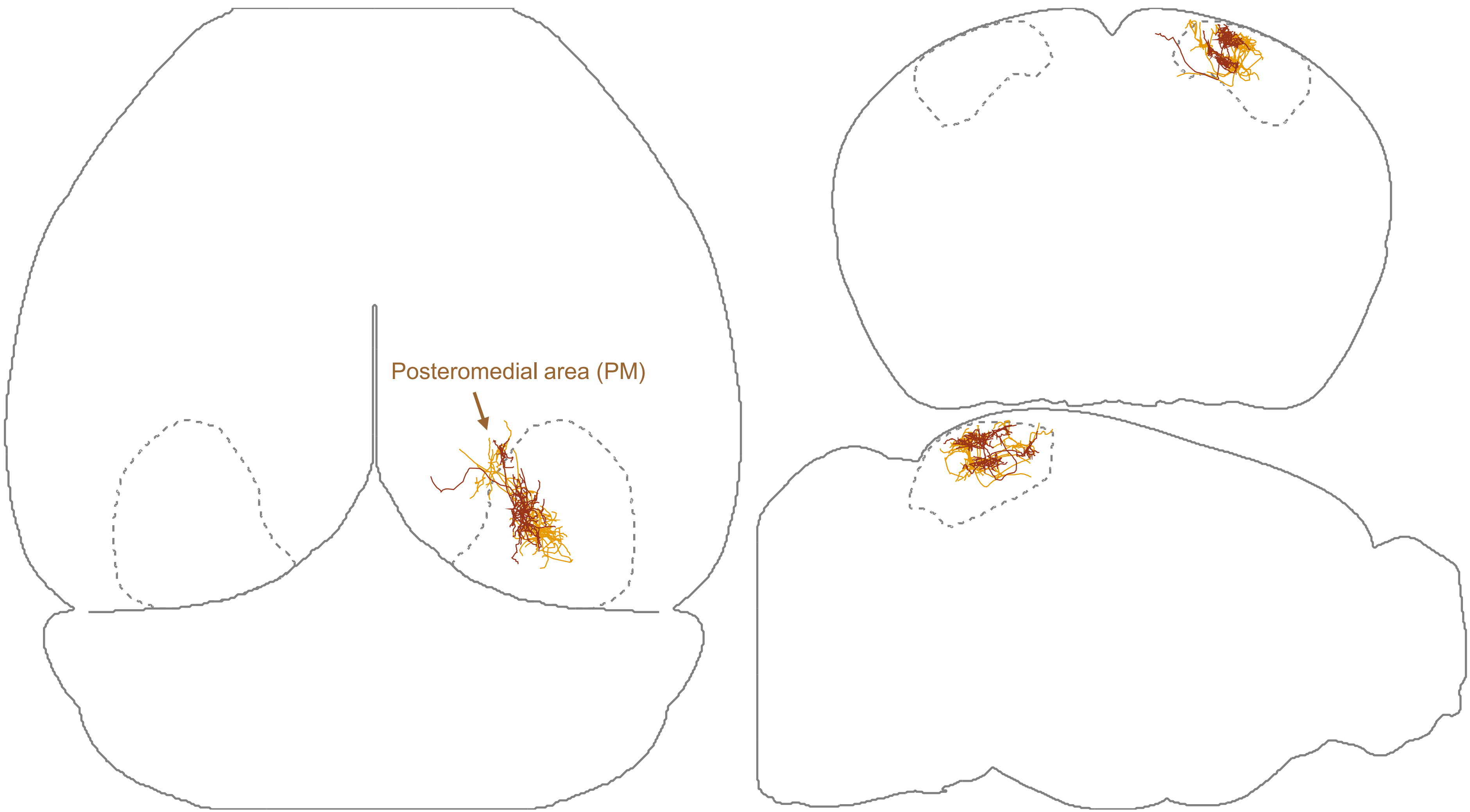
Example L2/3 projection neurons



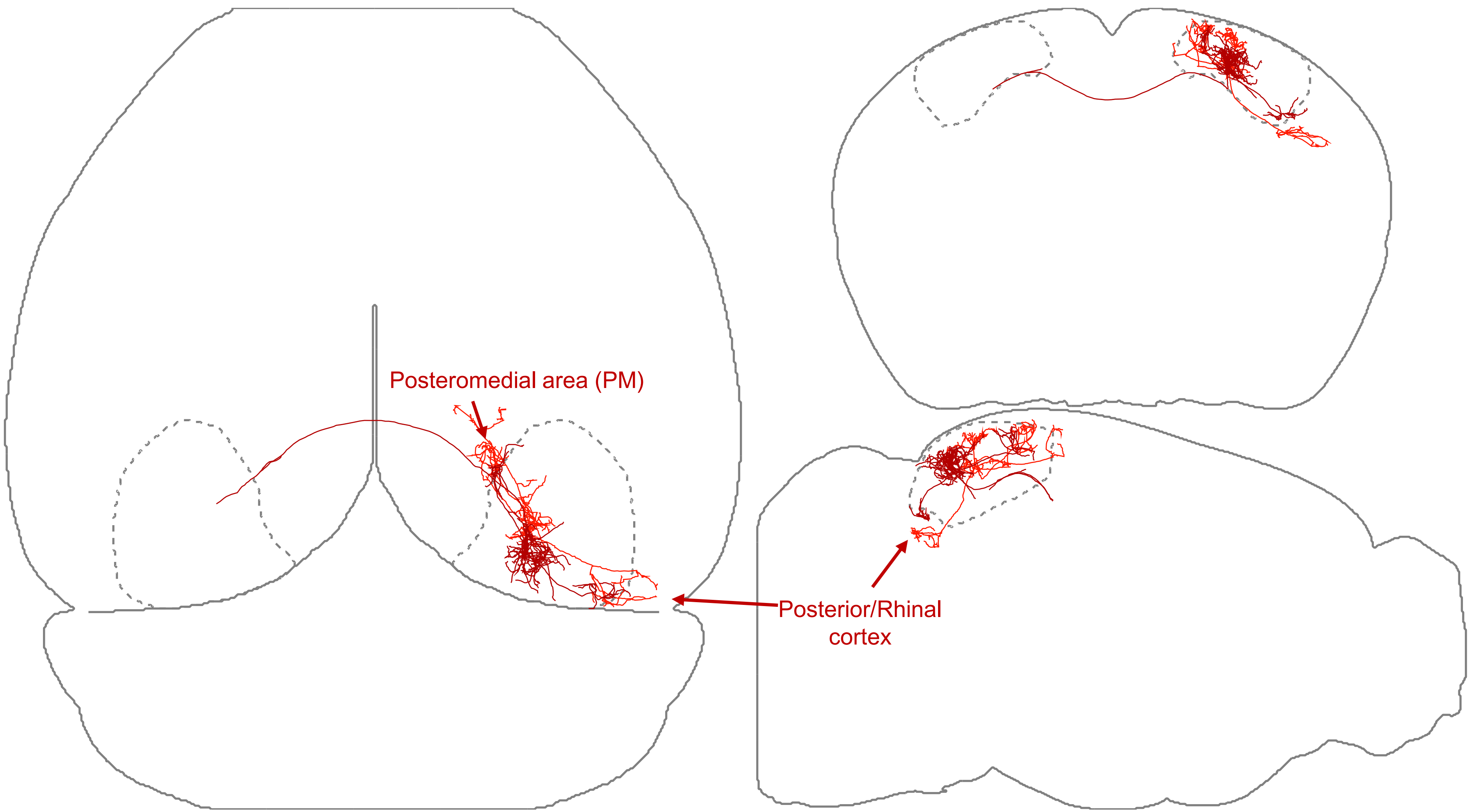
Example L2/3 projection neurons



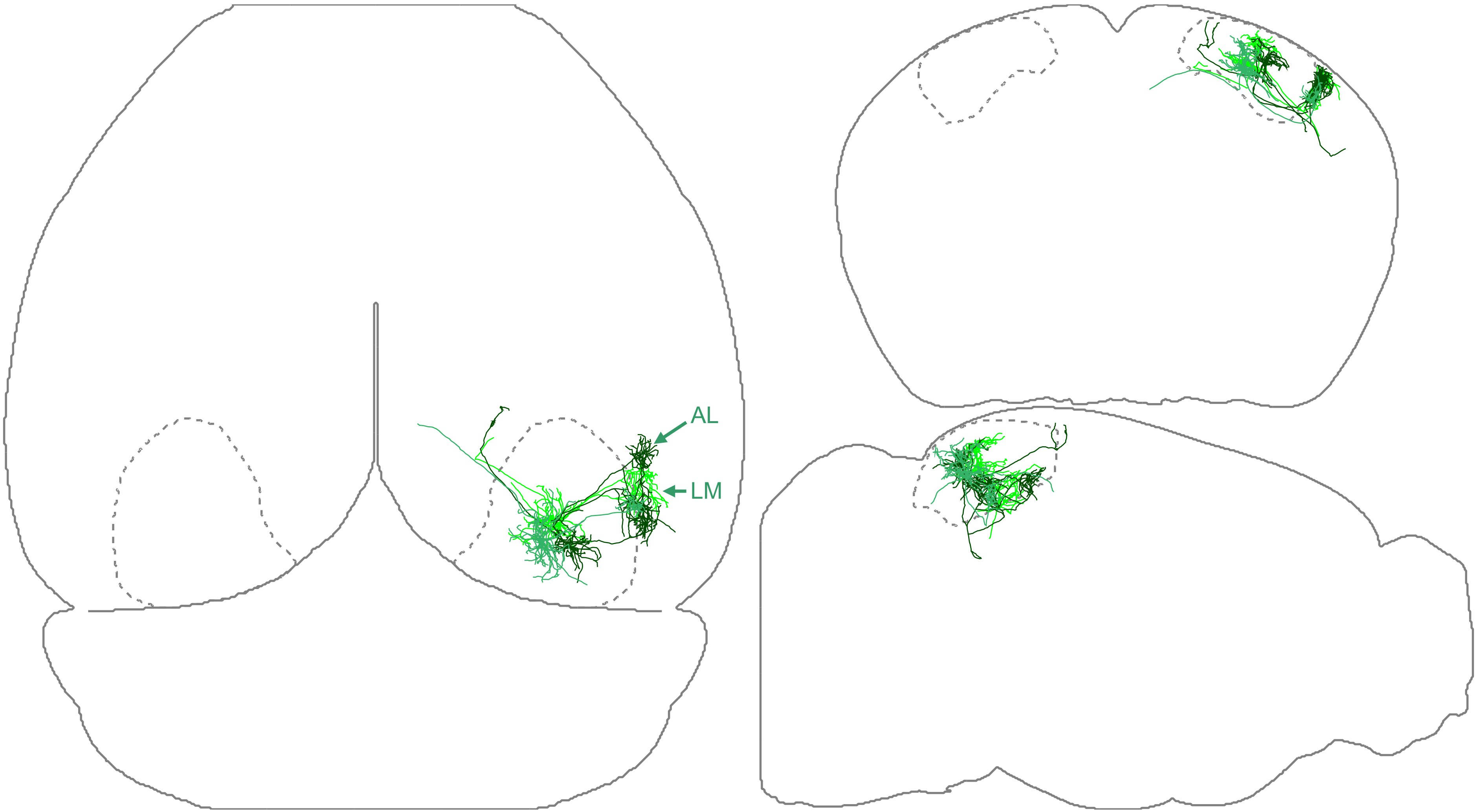
Example L2/3 projection neurons



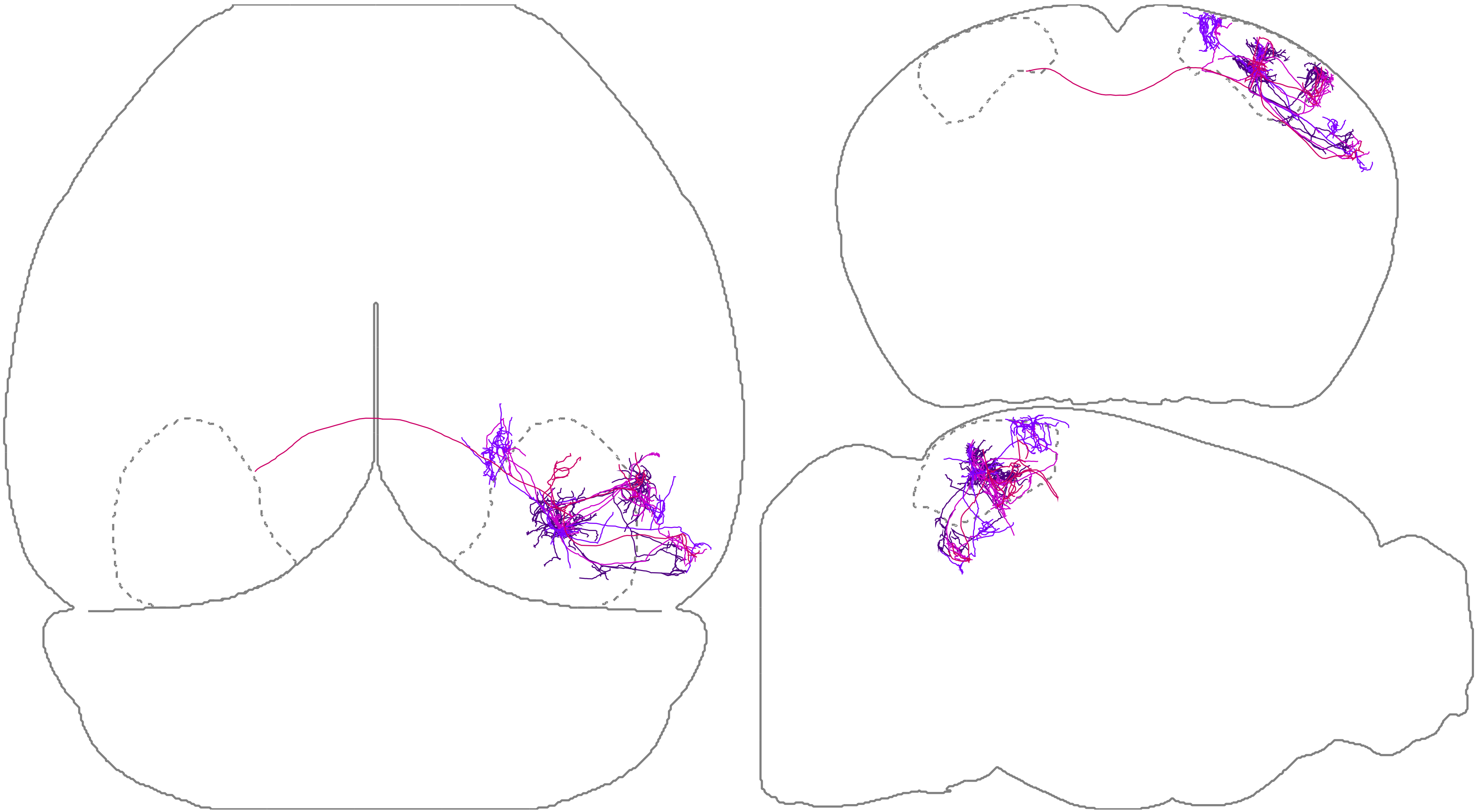
Example L2/3 projection neurons



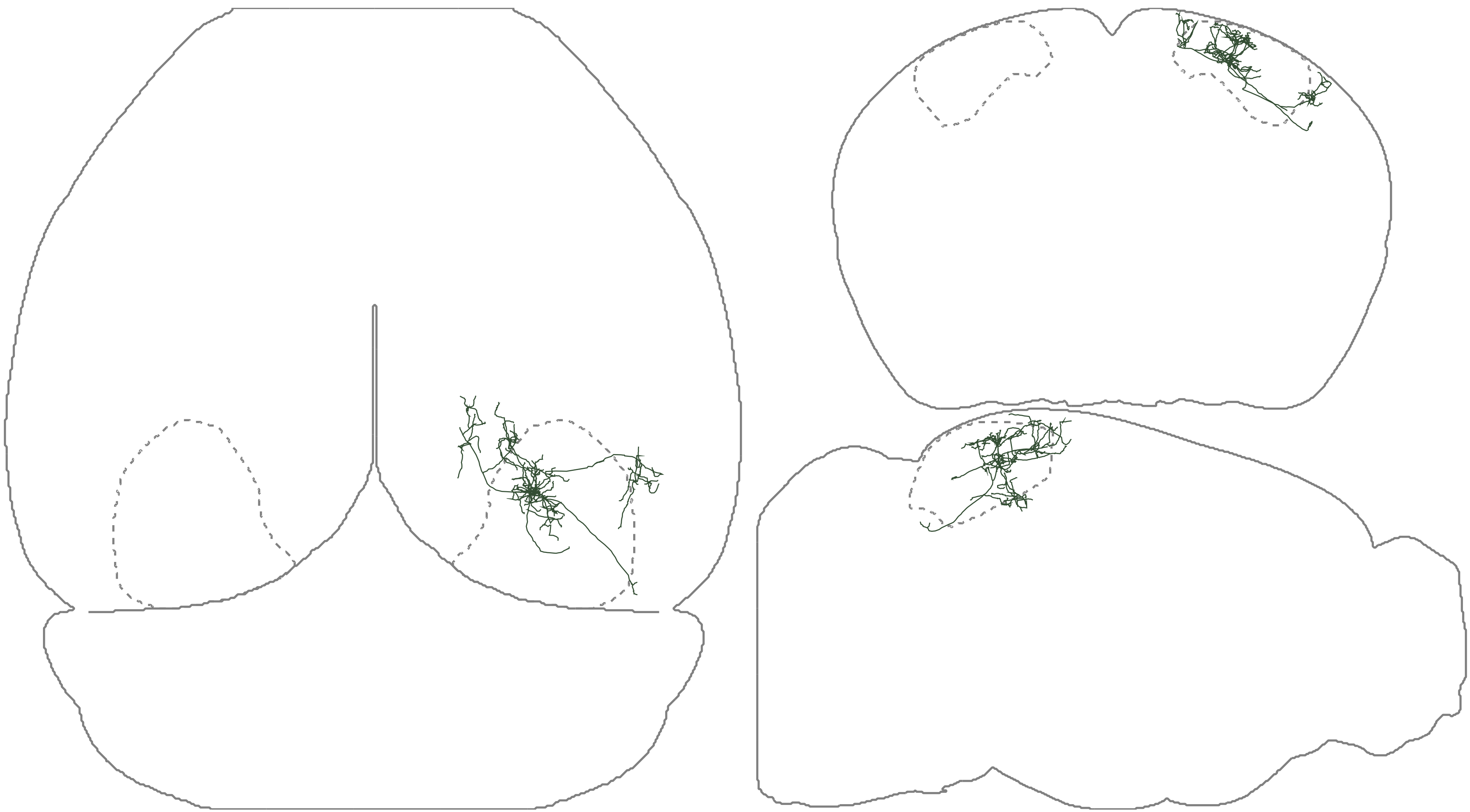
Example L2/3 projection neurons



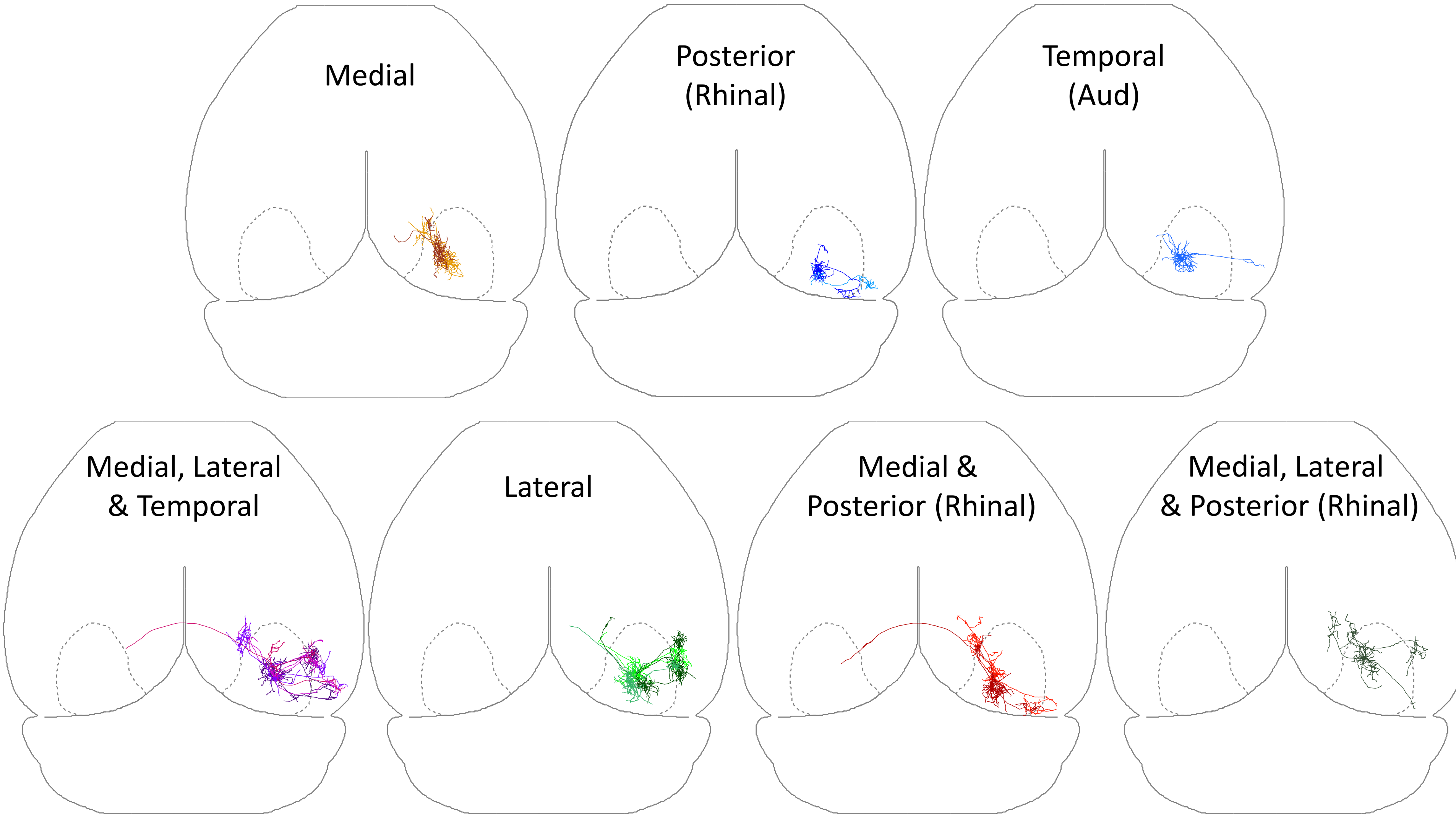
Example L2/3 projection neurons



Example L2/3 projection neurons



IT cells: highly diverse projections of individual neurons

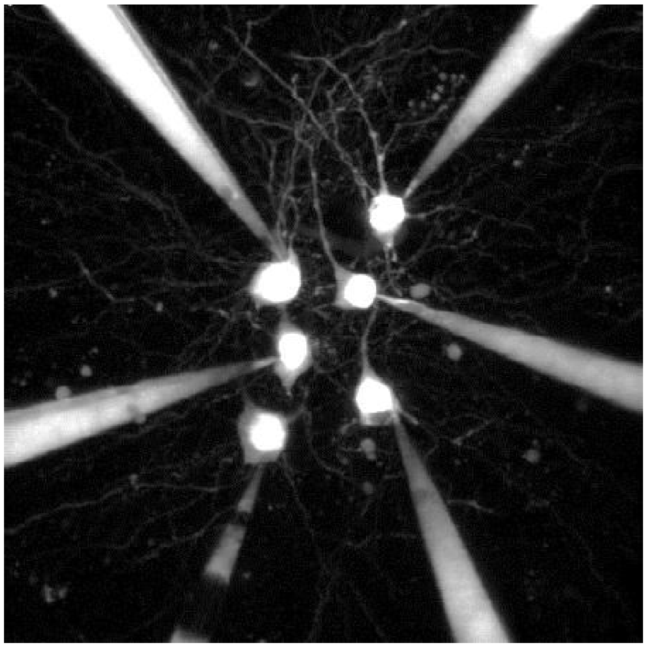


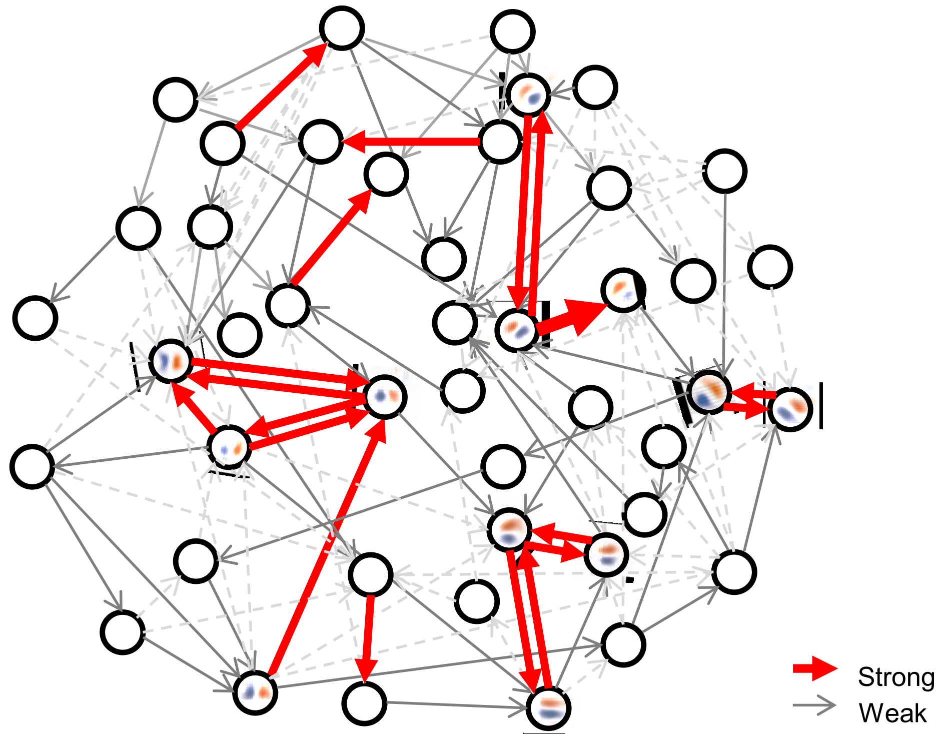
Layer 2 & 3 IT pyramidal cells

- pyramidal cells and most interneuron types
- Project strongly primarily to L5 and to other cortical areas (also striatum)
- Highly diverse projections of individual neurons
- Sensory feature specific subnetworks (selective amplification/pattern completion)
- Contextual integration of information across the same sensory modality
- Strong lateral/surround inhibition
- Sparse coding

Layer 2 & 3 IT pyramidal cells

- Sensory feature specific subnetworks (selective amplification / pattern completion)



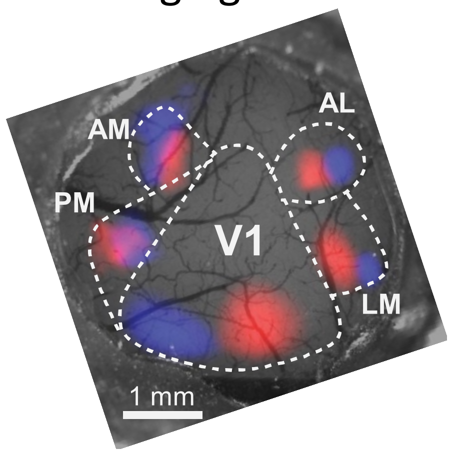


Layer 2/3 pyramidal cell network

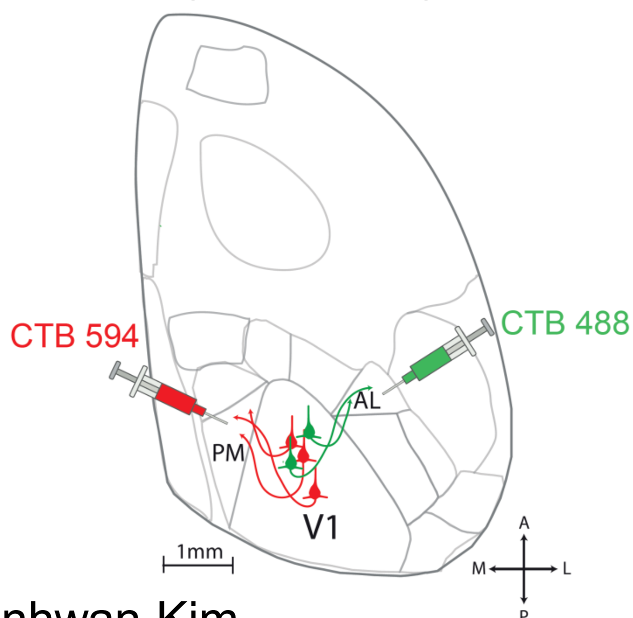
How does the local connectivity of IT cells relate to their long-range targets?

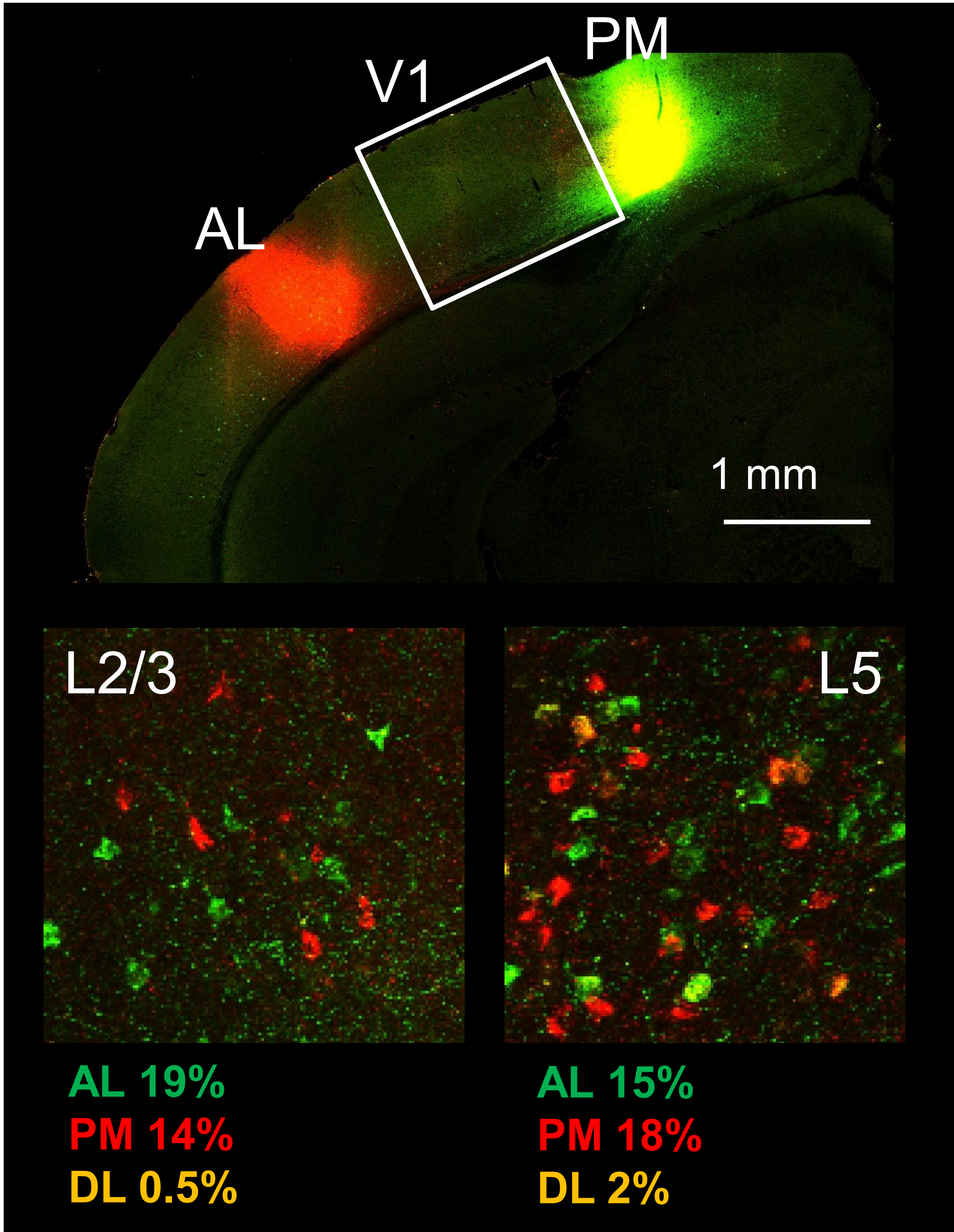
AL and PM projecting neurons are largely non-overlapping populations

Intrinsic imaging of visual areas



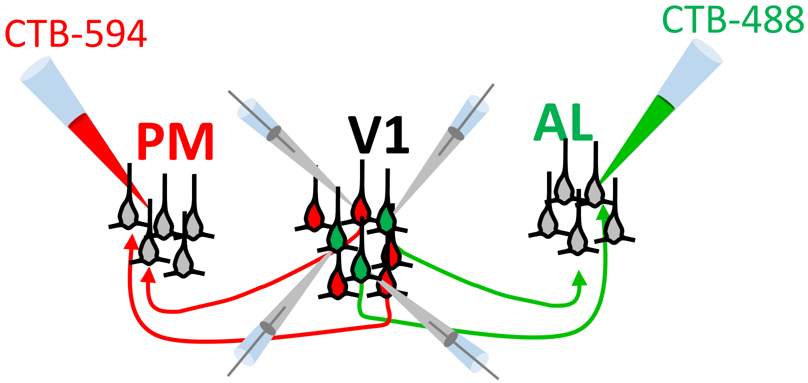
Retrograde tracing

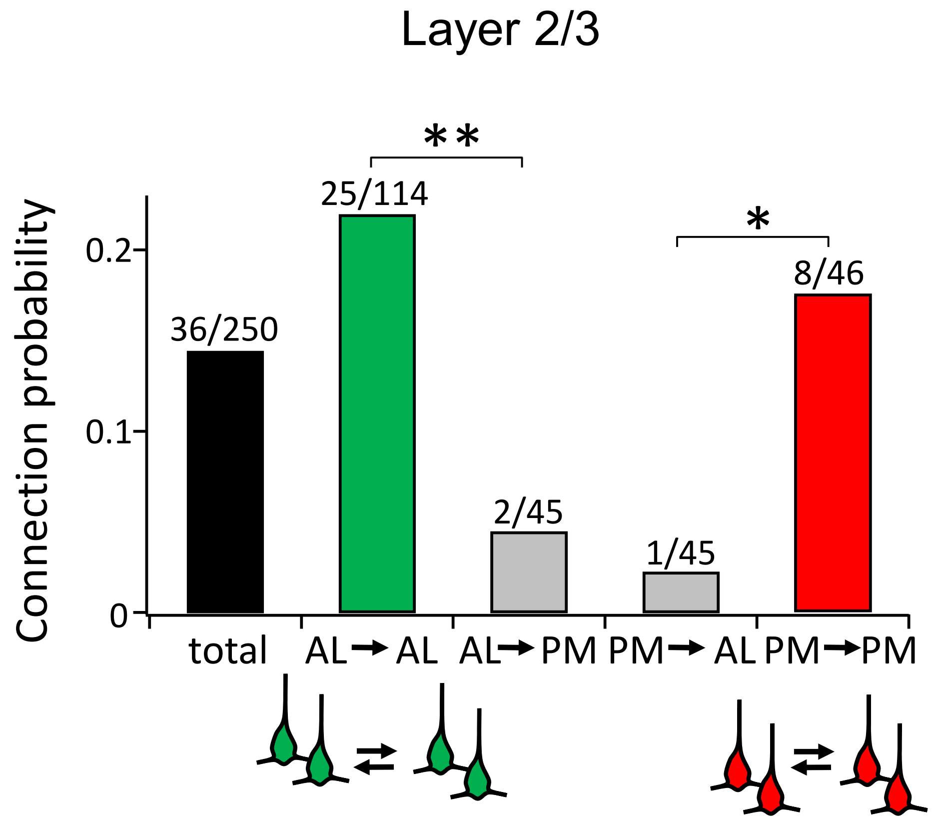




Meanhwan Kim

Local connectivity of L2/3 projection neurons is target-specific

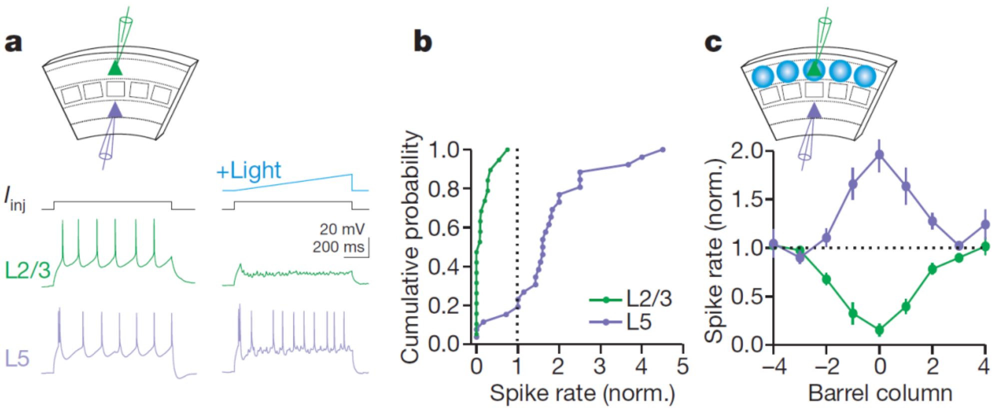


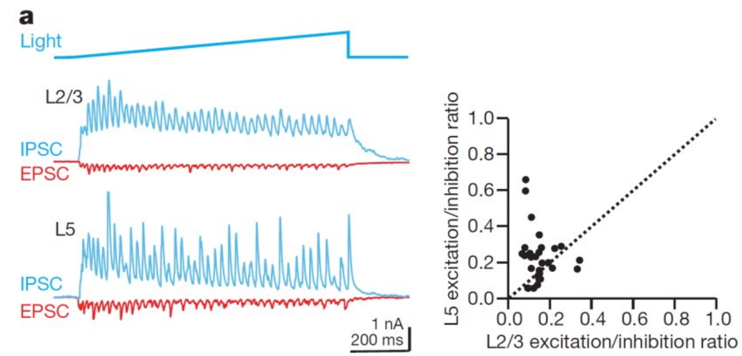


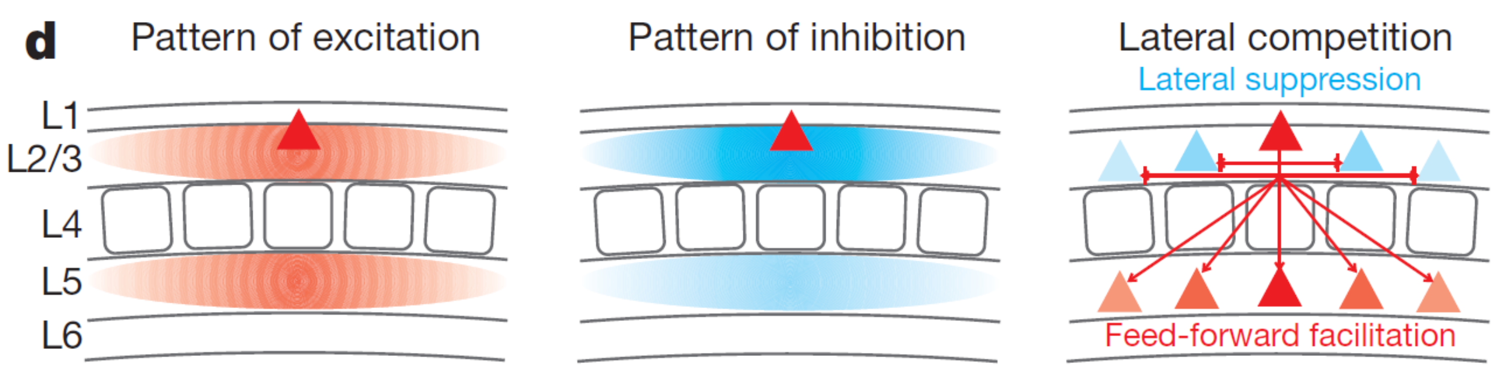
Meanhwan Kim

Layer 2 & 3 IT PCs: Contextual Interactions and Surround Inhibition

- Whole-cell recordings form L2/3 IT pyramidal cells
- Strong lateral/surround inhibition



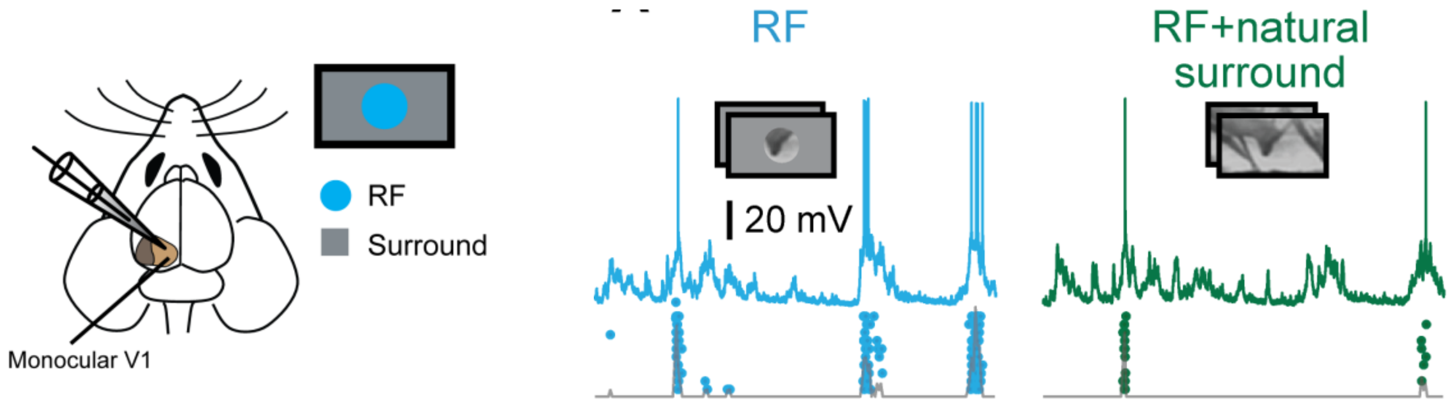




Adesnik &Scanziani 2010 Nature

Layer 2 & 3: Contextual Interactions and Surround Inhibition

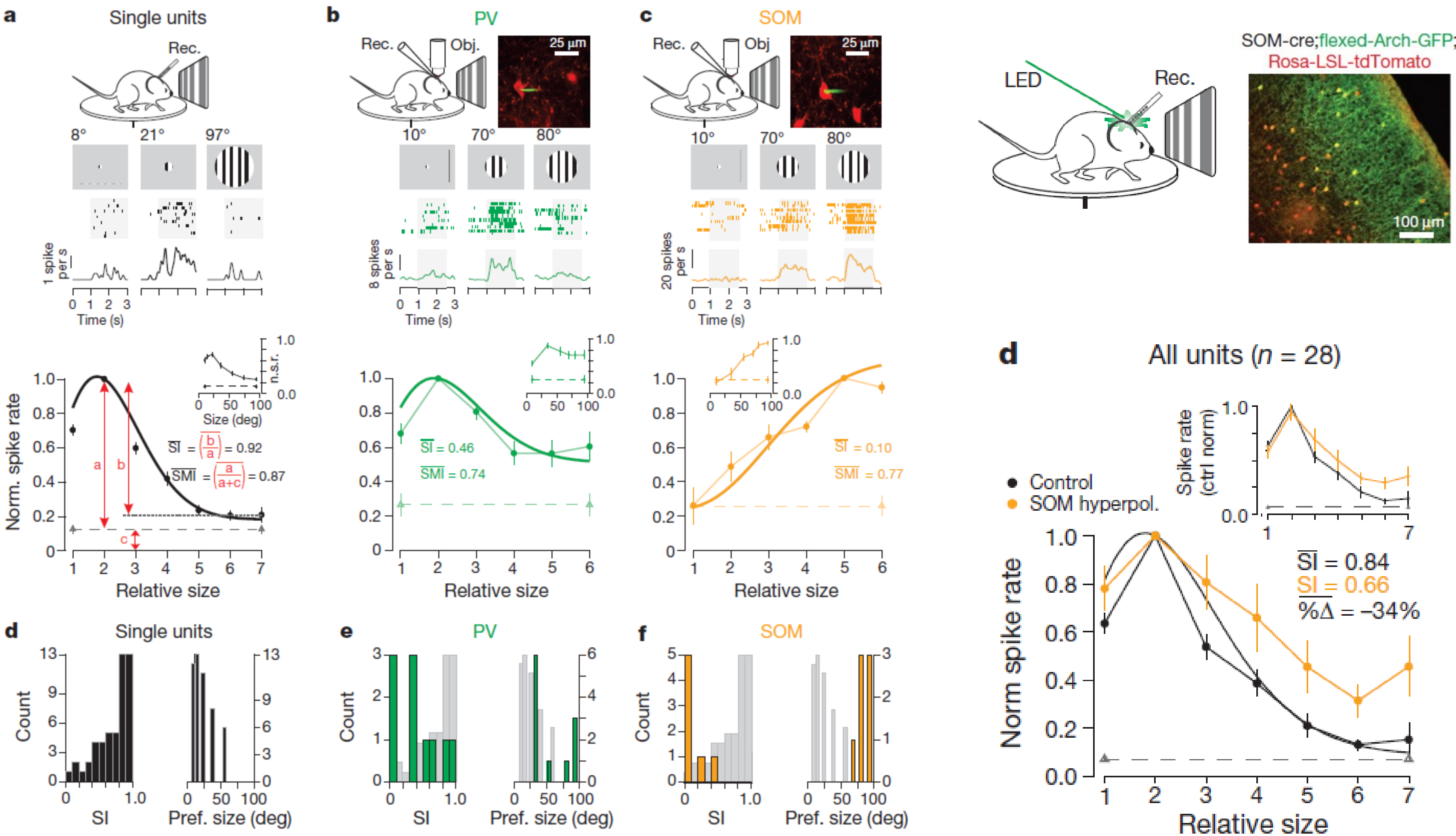
- Whole-cell recordings form L2/3 IT pyramidal cells
- Strong lateral/surround inhibition



Pecka et al., 2014, Neuron

Somatostatin-positive interneurons (SOM or SST)

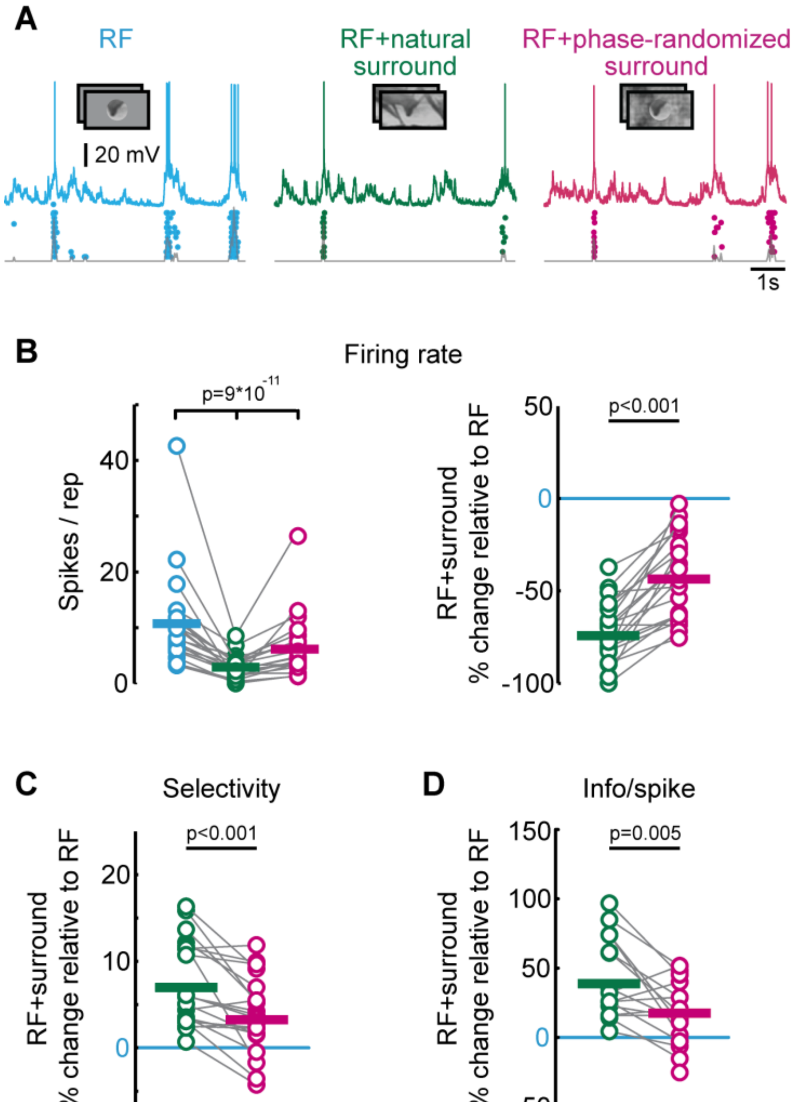
L 2/3 SOM cells may play a role in surround suppression (as might PV cells…)



Adesnik et al., 2013, Nature

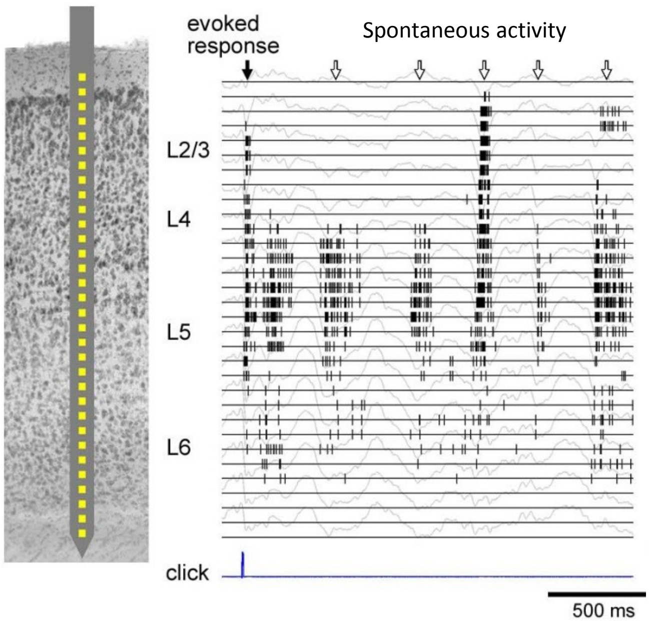
Layer 2 & 3: Contextual Interactions and Surround Inhibition

- Surround suppression changes selectivity and depends on context

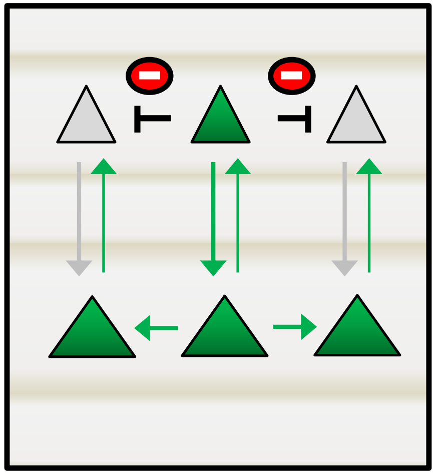


Pecka et al., 2014, Neuron

Sparse coding in L2/3 (and L5) IT cells

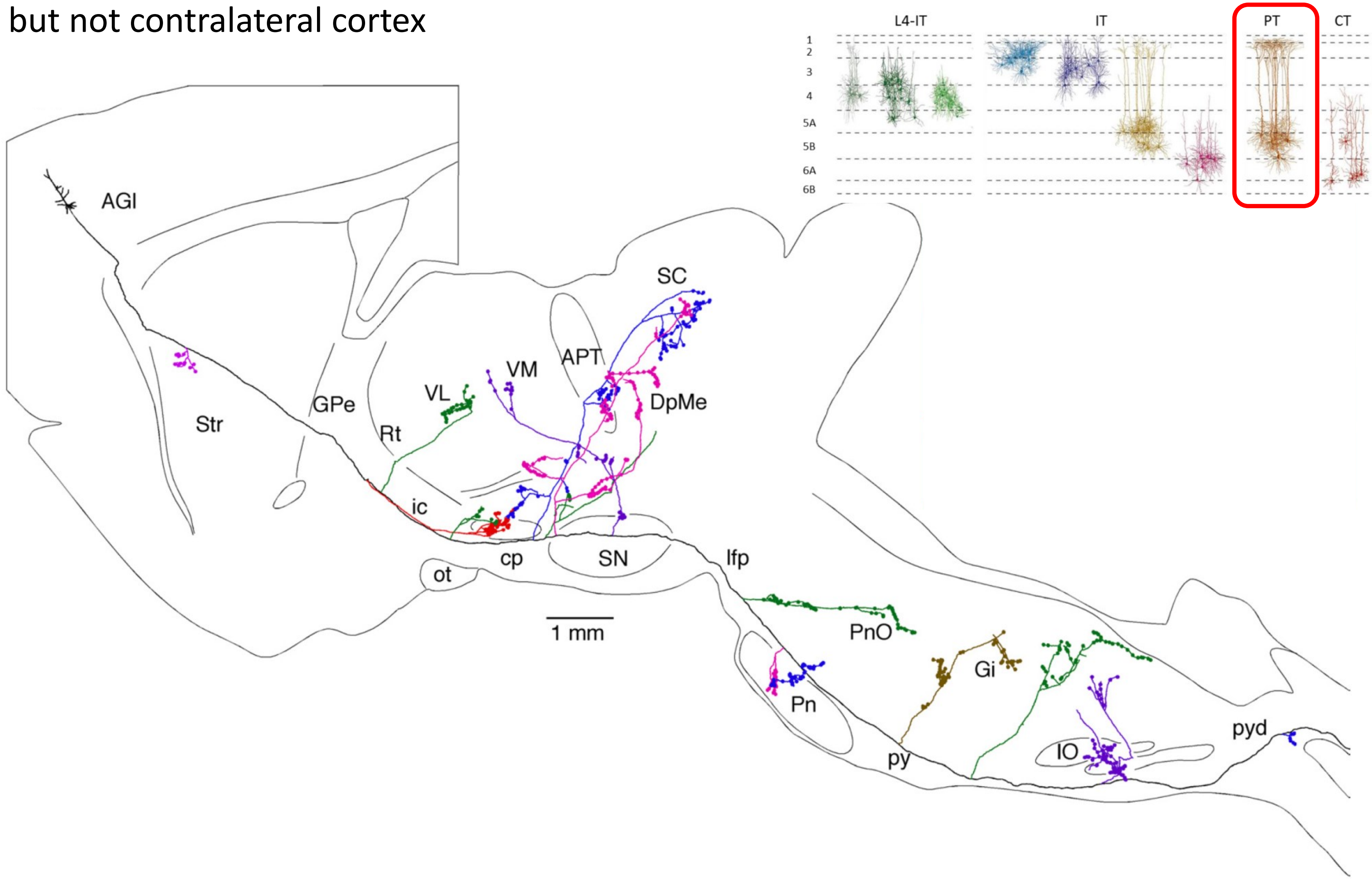


Sakata & Harris, Neuron 2010



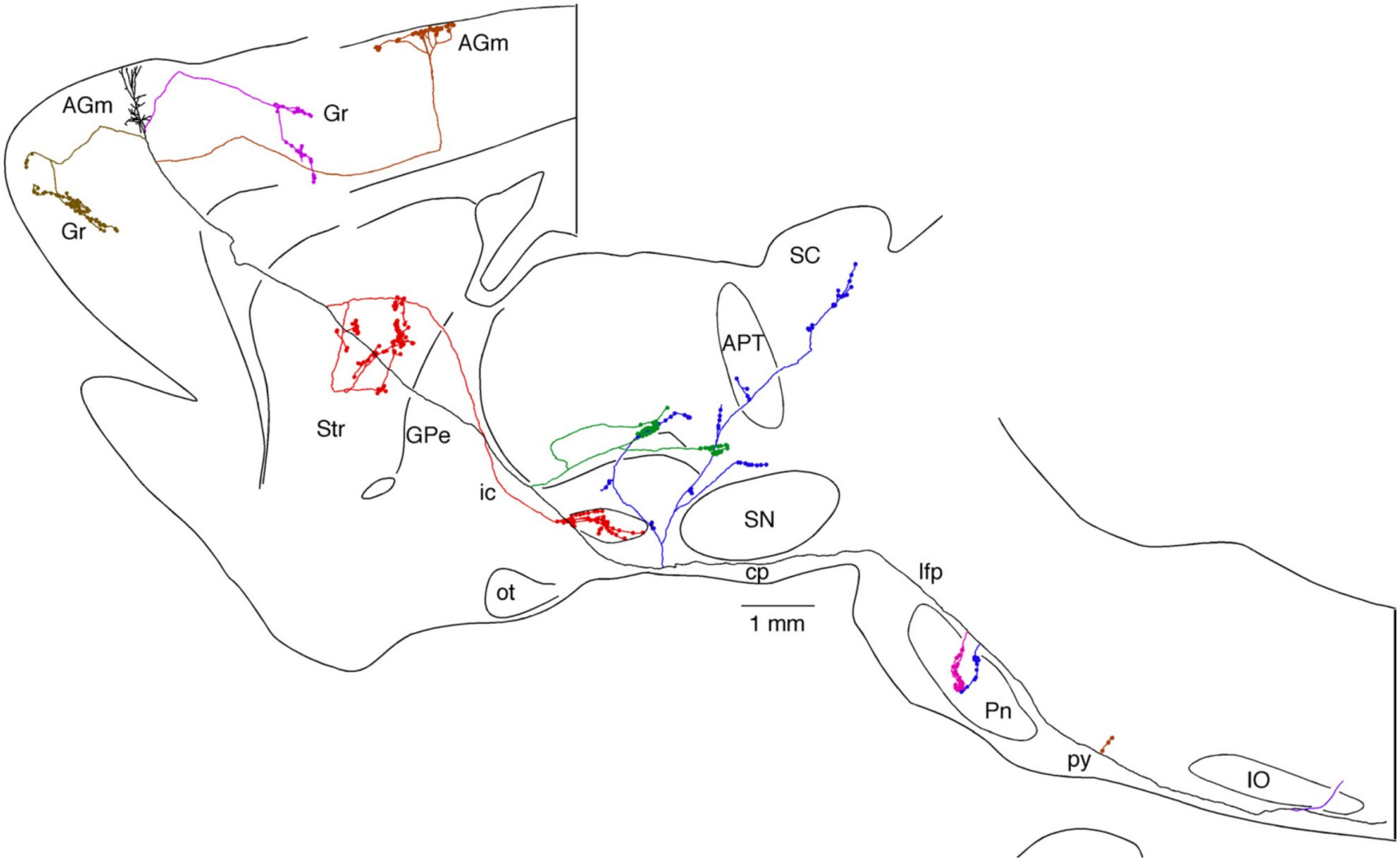
Layer 5: PT cells

- Pyramidal cells (subcerebral projections neurons, e.g. brainstem, superior colliculus)
- Typically project to motor-related structures, and ipsilateral cortex, striatum, thalamus, but not contralateral cortex



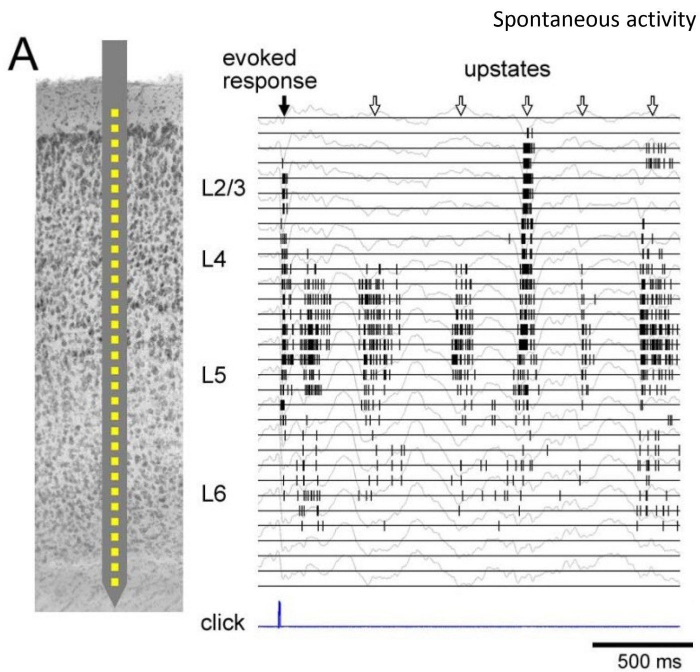
Kita and Kita, 2012, JN

Layer 5: PT cells

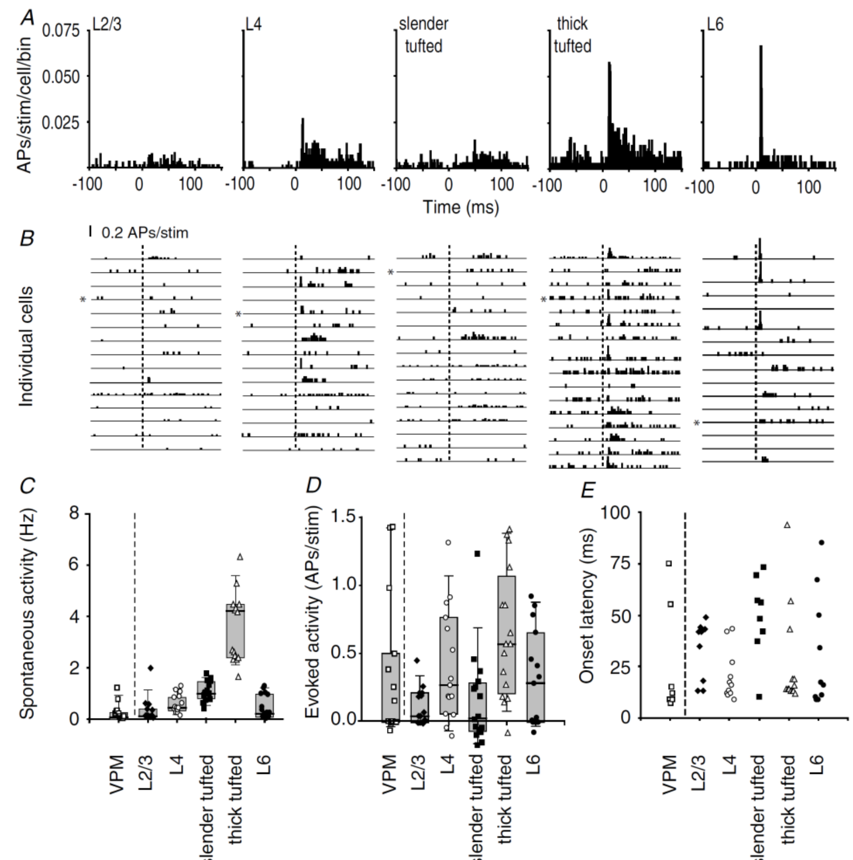


Kita and Kita, 2012, JN

Coding strategies of different cortical layers

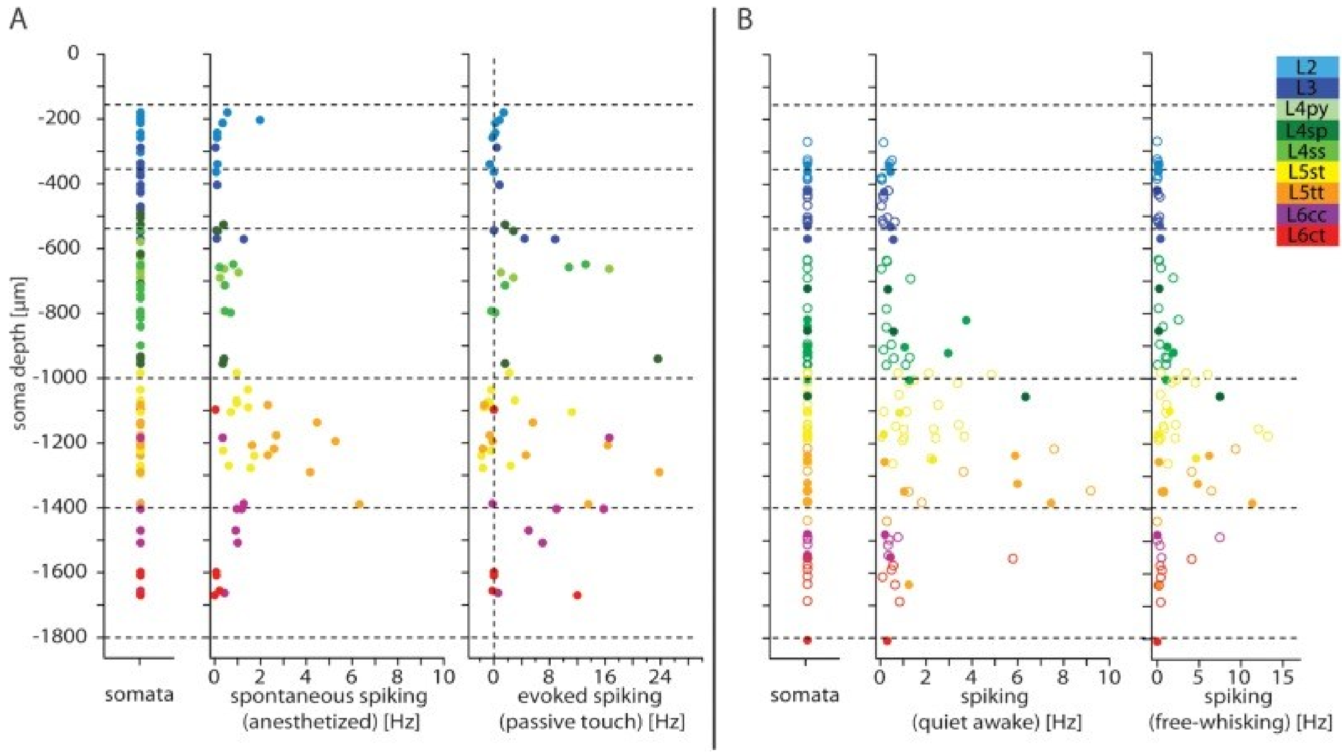


Sakata & Harris, Neuron 2010



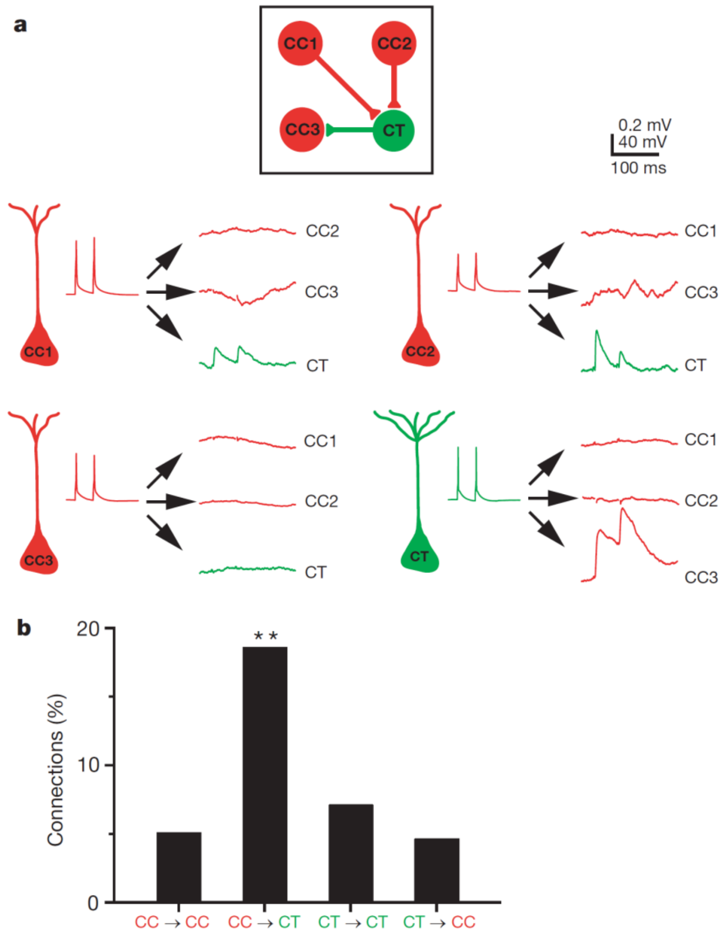
De Kock et al, 2007

IT versus PTs neurons: Firing rate differences



Cereb Cortex. Oct 2012; 22(10): 2375–2391.

IT versus PTs neurons: Local connectivity differences



Brown & Hestrin 2009 Nature

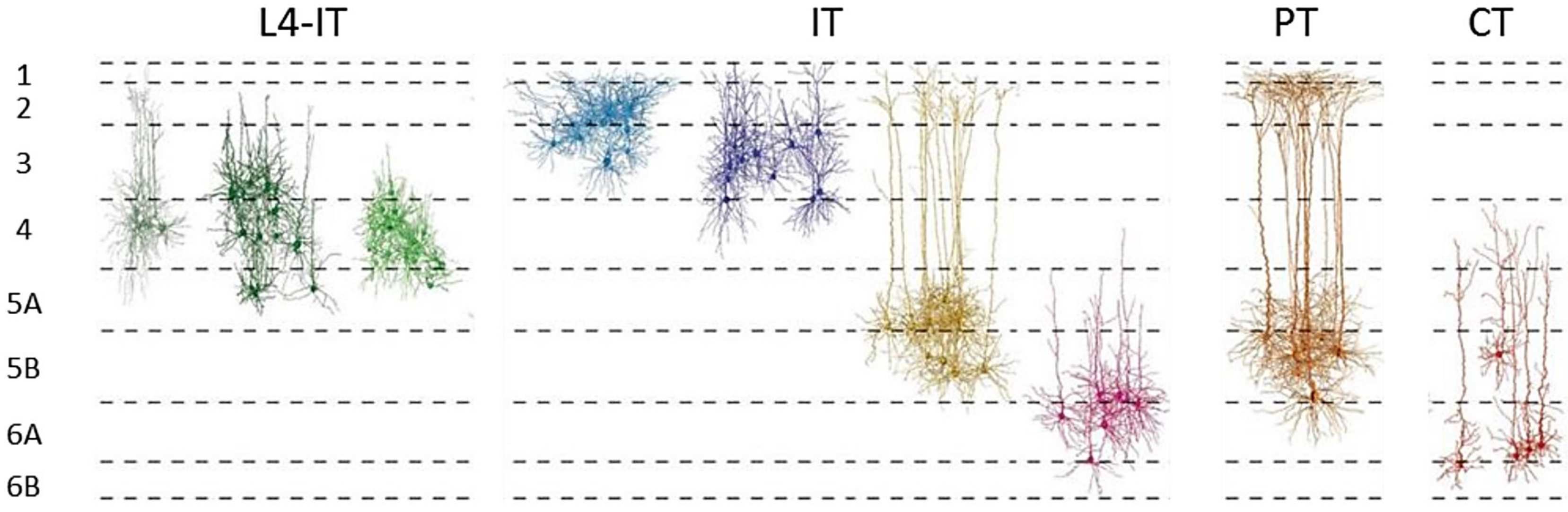
Layer 5: PT cells

- Pyramidal cells
- Typically project to motor-related structures, and ipsilateral cortex, striatum, thalamus, but not contralateral cortex
- High firing rates, non-adapting, sometimes bursting
- PT cells integrate cortical and TC inputs and broadcast them to subcerebral structures

What happens in Layer 1 (superficial layer 2)?

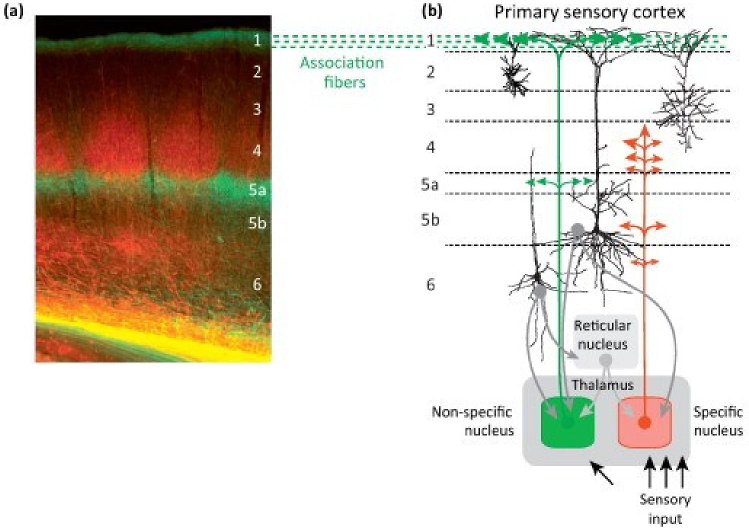
- Very few cell bodies
- Dendrites of pyramidal cells whose somata reside in layers 2-6.
- Only a few Interneurons (mostly 5HT3R positive neurogliaform cells)
- But axons of other interneurons (VIP, SST, PV)
- Many cortical and thalamic long-range inputs

→ Main site of association in the cortex
→ integration of feedforward and feedback/top-down inputs



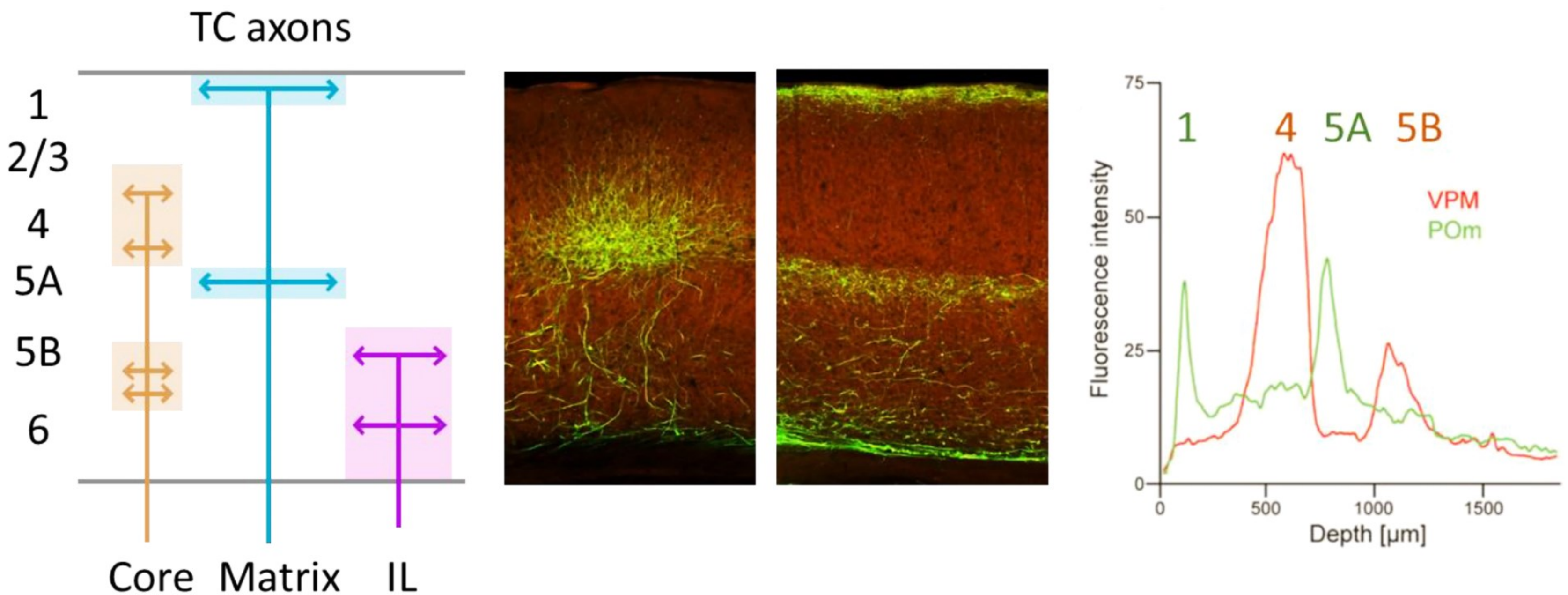
What kind of inputs innervate L1?

Thalamocortical (higher-order/matrix thalamus)
→ Contextual information within the same sensory modality



Larkum, 2013, TINS

Laminar profile of thalamocortical (TC) input types

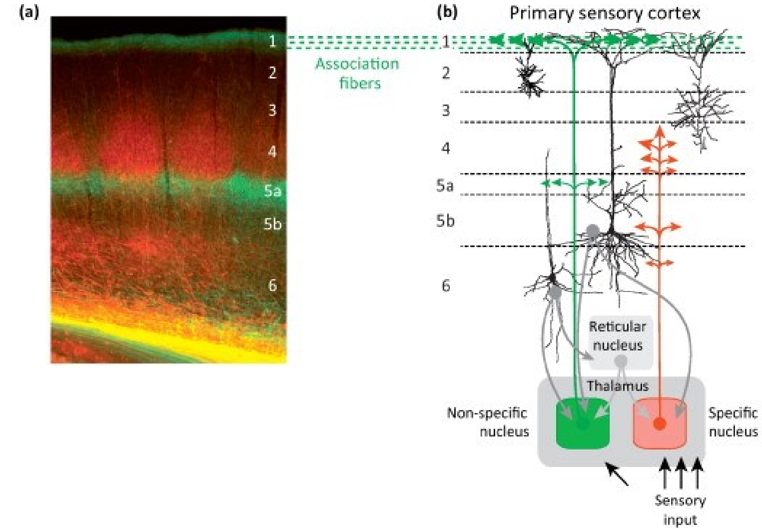


Harris and Shepherd, 2014, review, unpublished

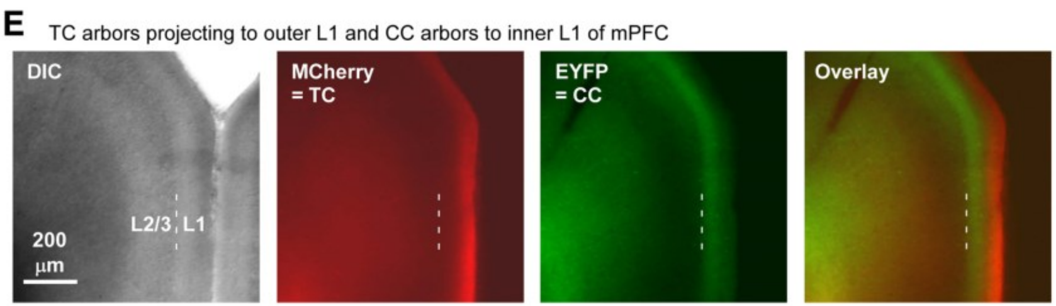
What kind of inputs innervate L1?

Thalamocortical (higher-order/matrix thalamus)
→ Contextual information within the same sensory modality

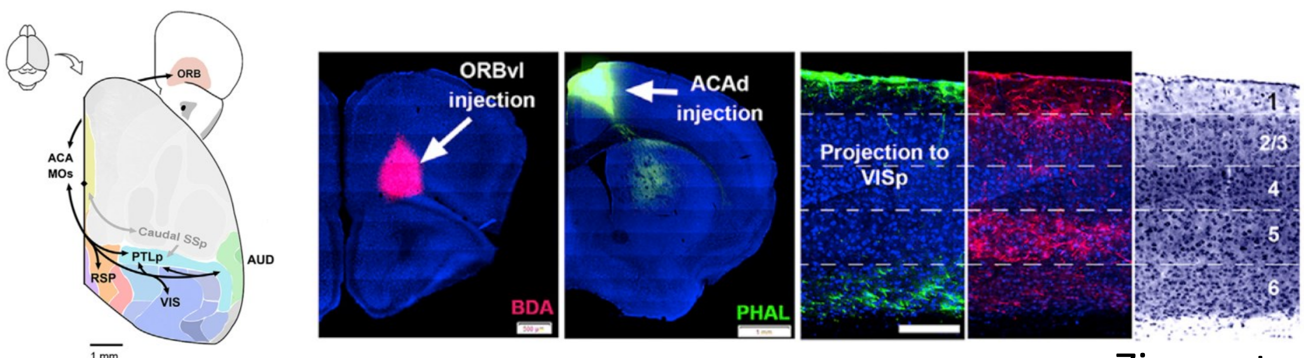
Corticocortical
→ contextual information from the same or different modalities



Larkum, 2013, TINS



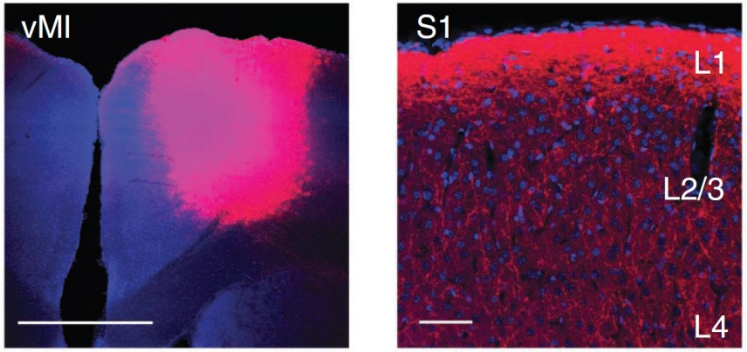
Cruikshank et al., 2012 JN



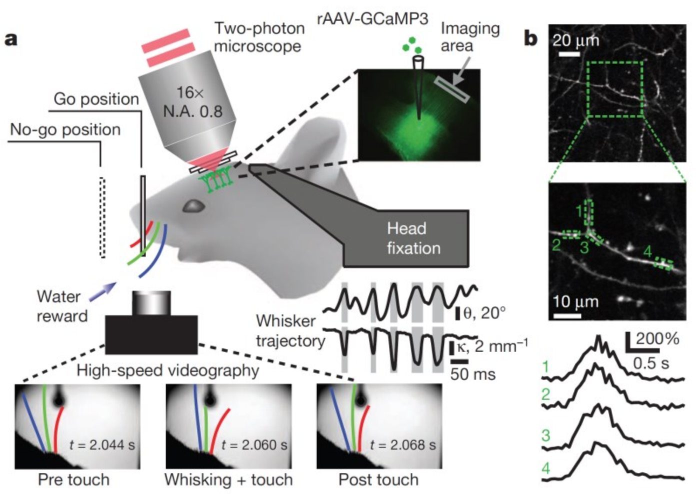
Zingg et al 2014 Cell

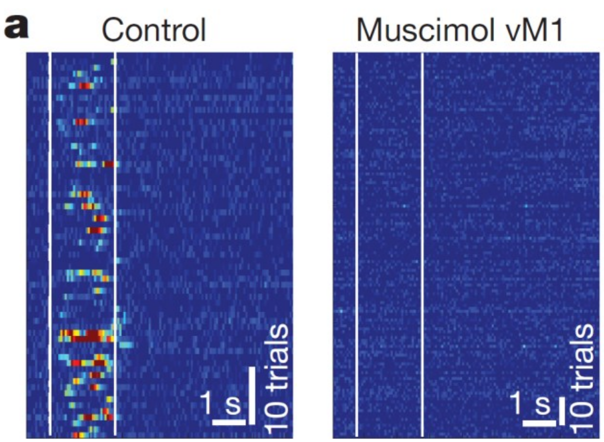
<u>Case study</u>: response modulation by dendritic plateau potentials in barrel cortex during active sensation

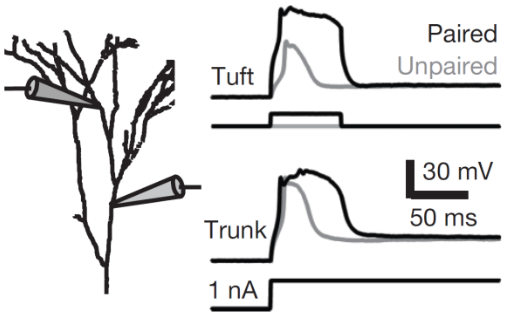
- Long-range Input from vM1 to layer 1 of vS1



- L5 pyr cell tufts exhibit calcium touch-related plateau potentials, dependent on vM1 input

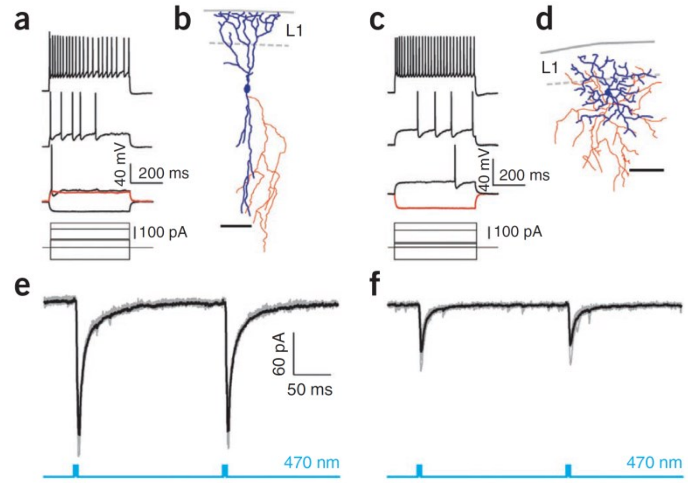


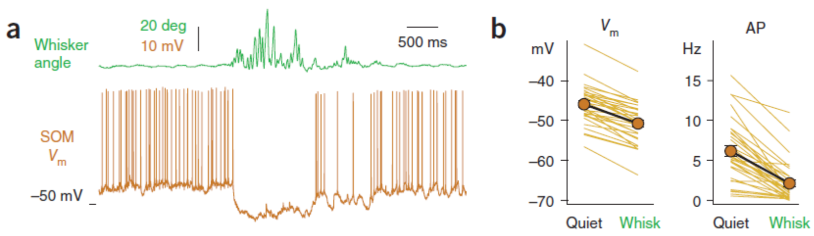




Lee & Rudy 2013 Nat Neurosci; Xu & Magee 2012, Nature

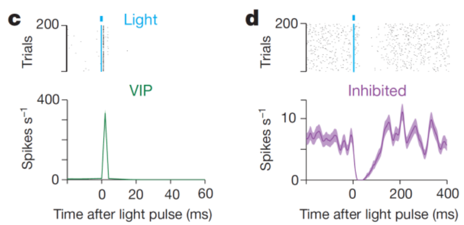
- VIP interneurons receive strongest input from vM1





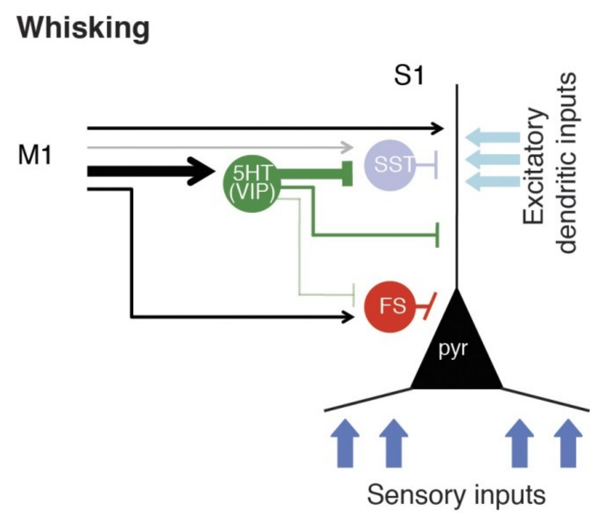
Gentet et al., 2013, Nat Neurosci

Chr2 in VIP cells

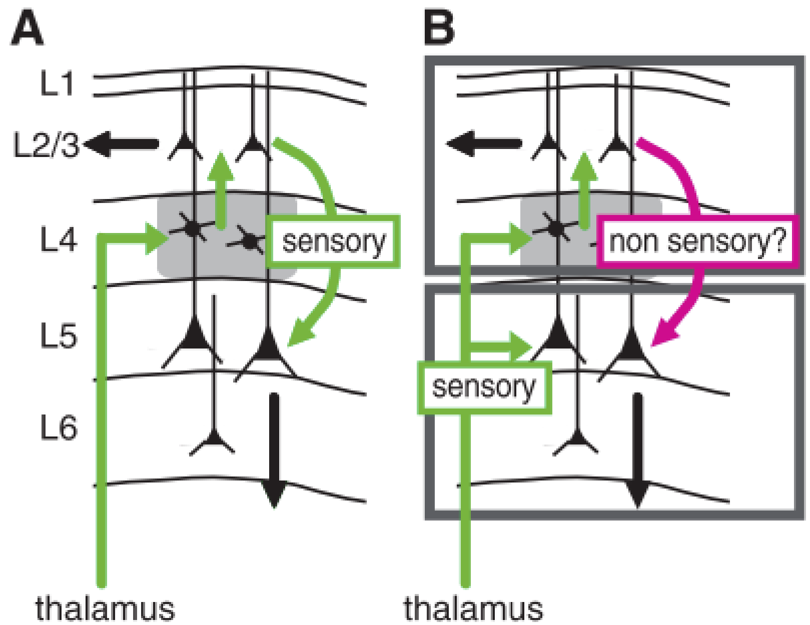


Pi et al., 2013, Nature

- SST neurons suppressed by whisking and touch
- VIP interneurons most strongly inhibit SST interneurons that inhibit pyr cell dendrites
  → Disinhibition of pyramidal cell dendrites

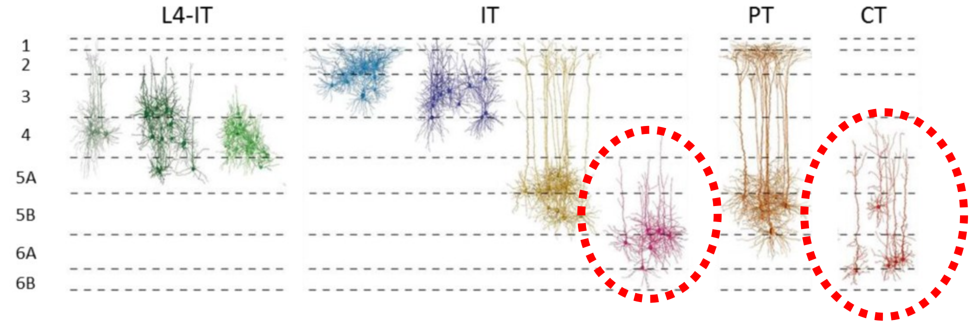


- Inhibitory (VIP) interneuron control of the influence of long-range signals
  → role in active sensing

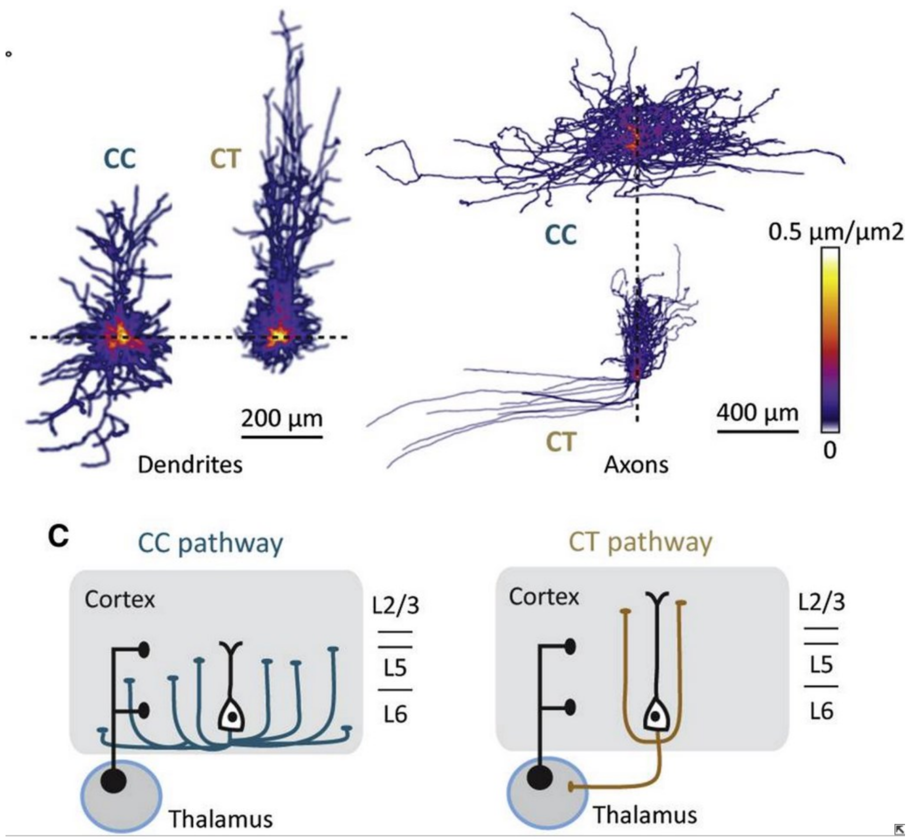


Constantinople & Bruno, 2013, Science
Larkum, 2013, TINS

The mysterious layer 6



- at least two classes of projection neuron
- - IT and CT
- in mouse V1, CTs receive more input from higher order cortical areas
- CT only cell class that projects to reticular nucleus of the thalamus (RTN) and the core thalamus
- intracolumnar inhibition?



Velez-Fort et al., 2014 Neuron

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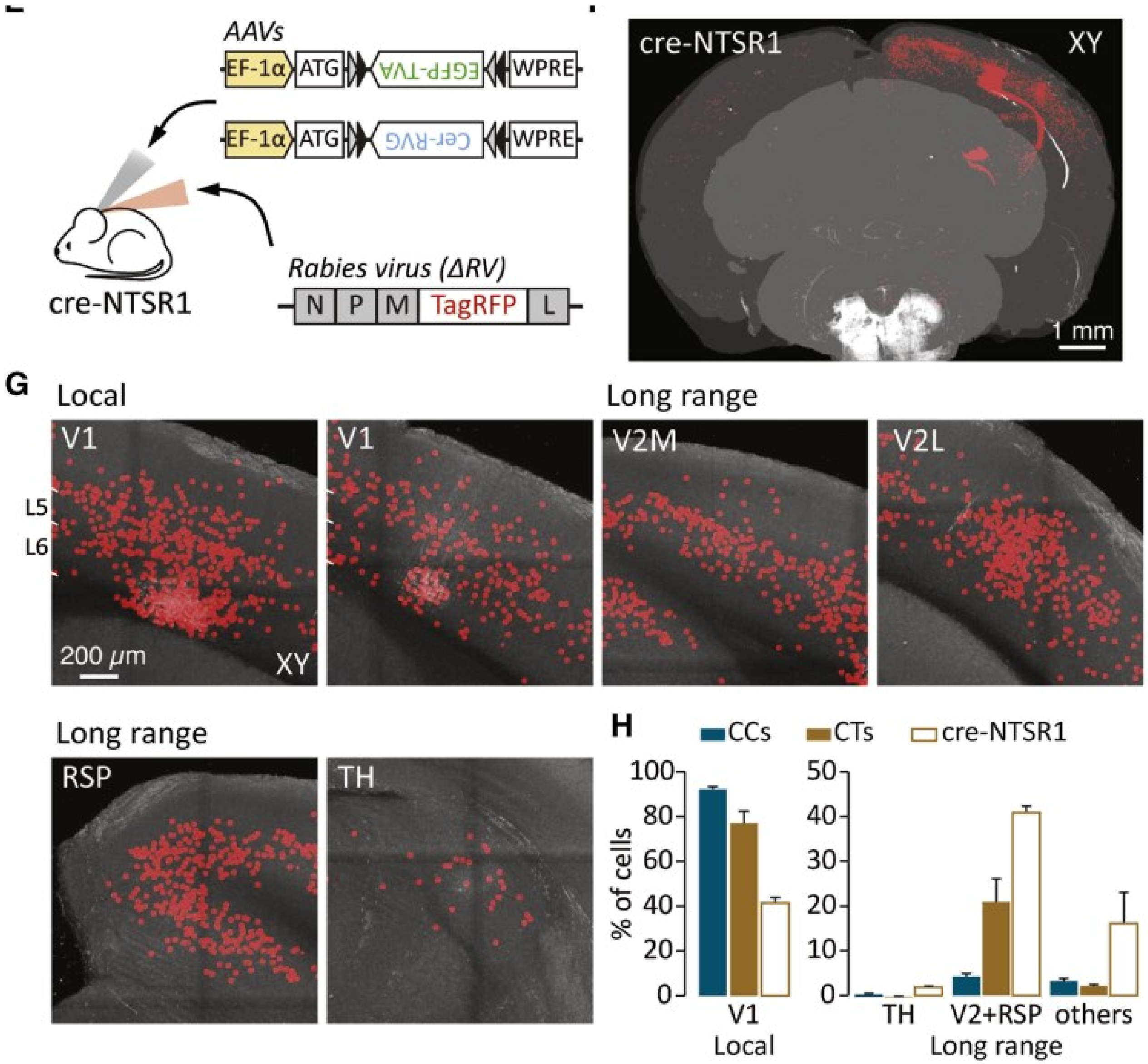
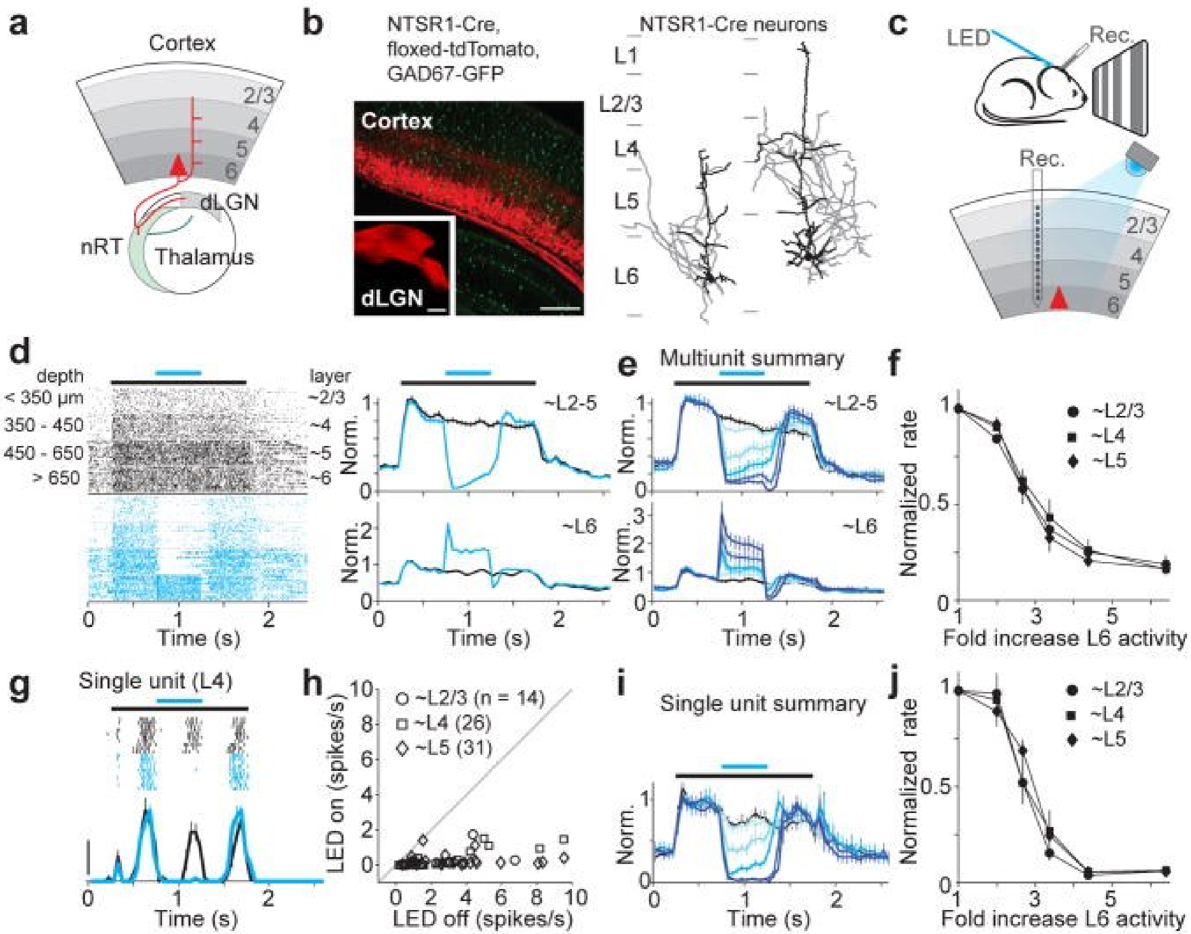


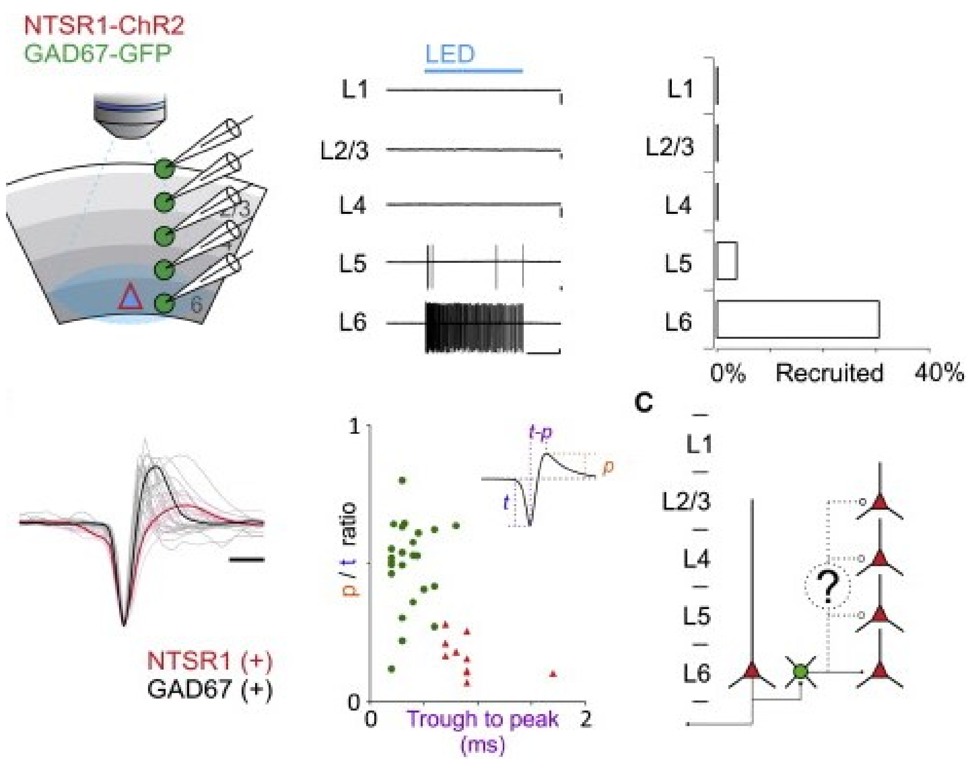
Velez-Fort et al., 2014 Neuron

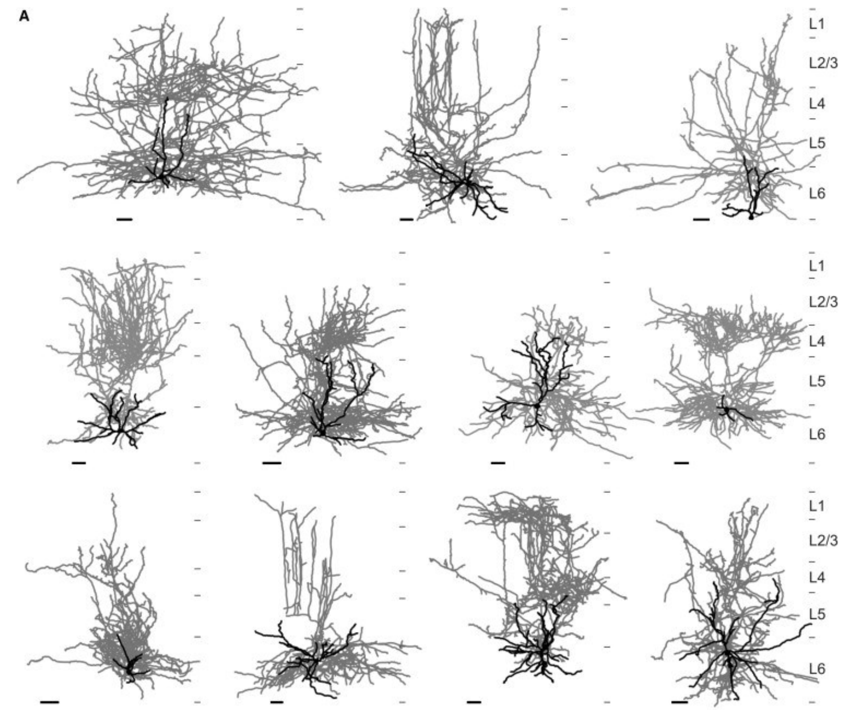
In mouse V1, L6 CTs mediate intracolumnar inhibition



Olsen et al, 2012 Nature

L6 intracolumnar inhibition mediated by L6 interneurons projecting to all layers



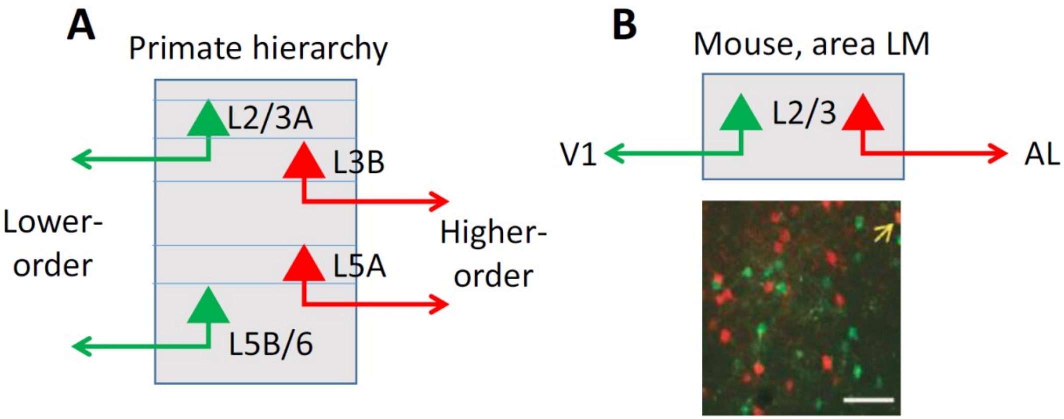


Bortone et al, 2014 Neuron

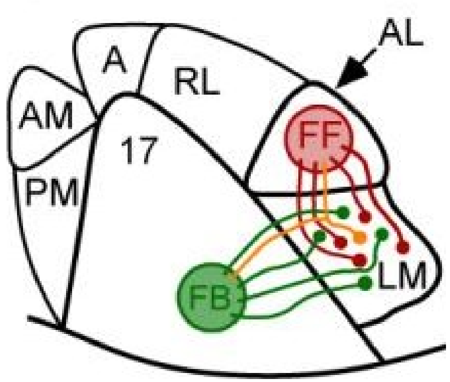
Open questions

- How many cell classes and subclasses?
- Genetic versus activity dependent determinism?
- What does L2/3 actually do?
- Do different L1 inputs target different cell types?

How many classes of IT projection neurons?



From Nassi & Callaway



Adapted from Harris and Shepherd, 2014, review, unpublished

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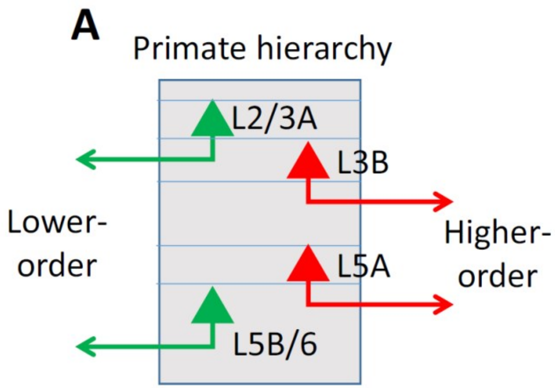

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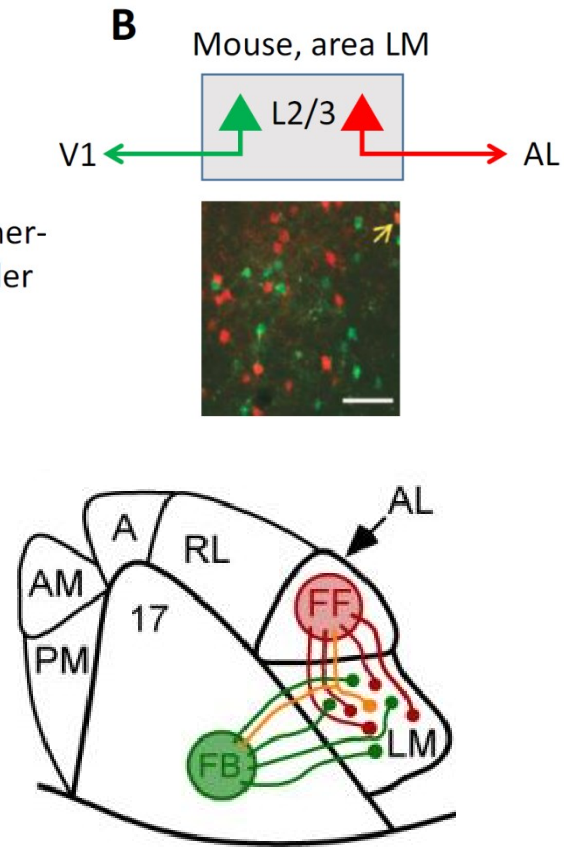
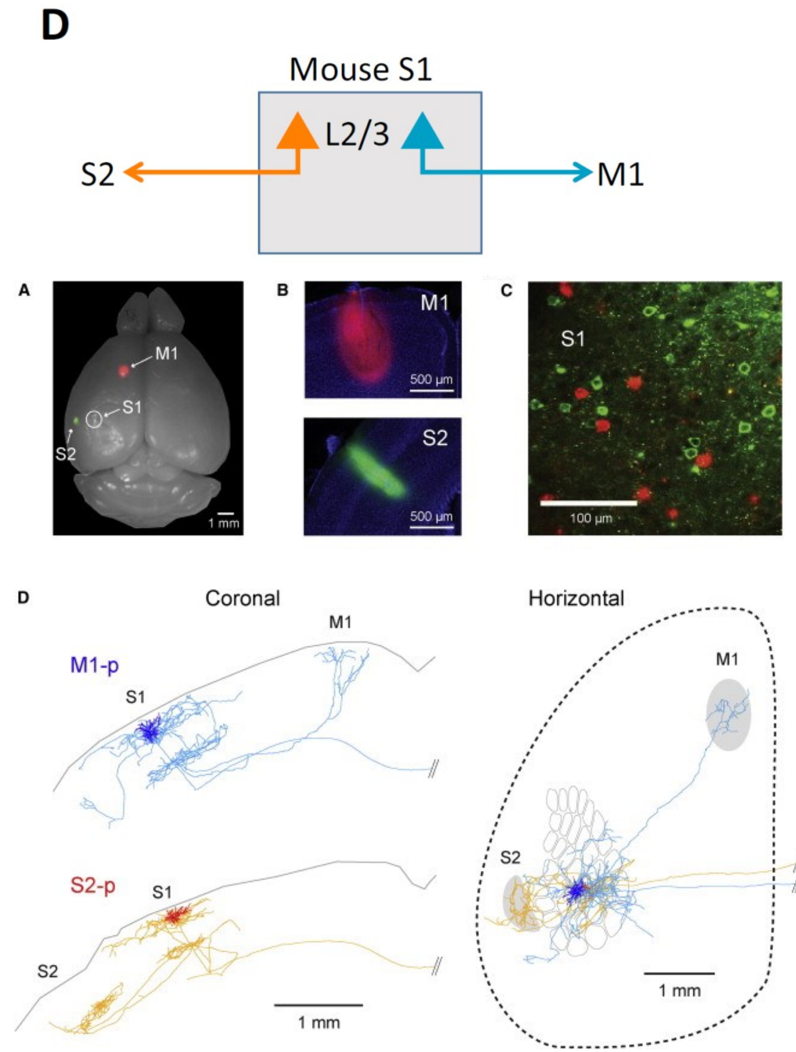


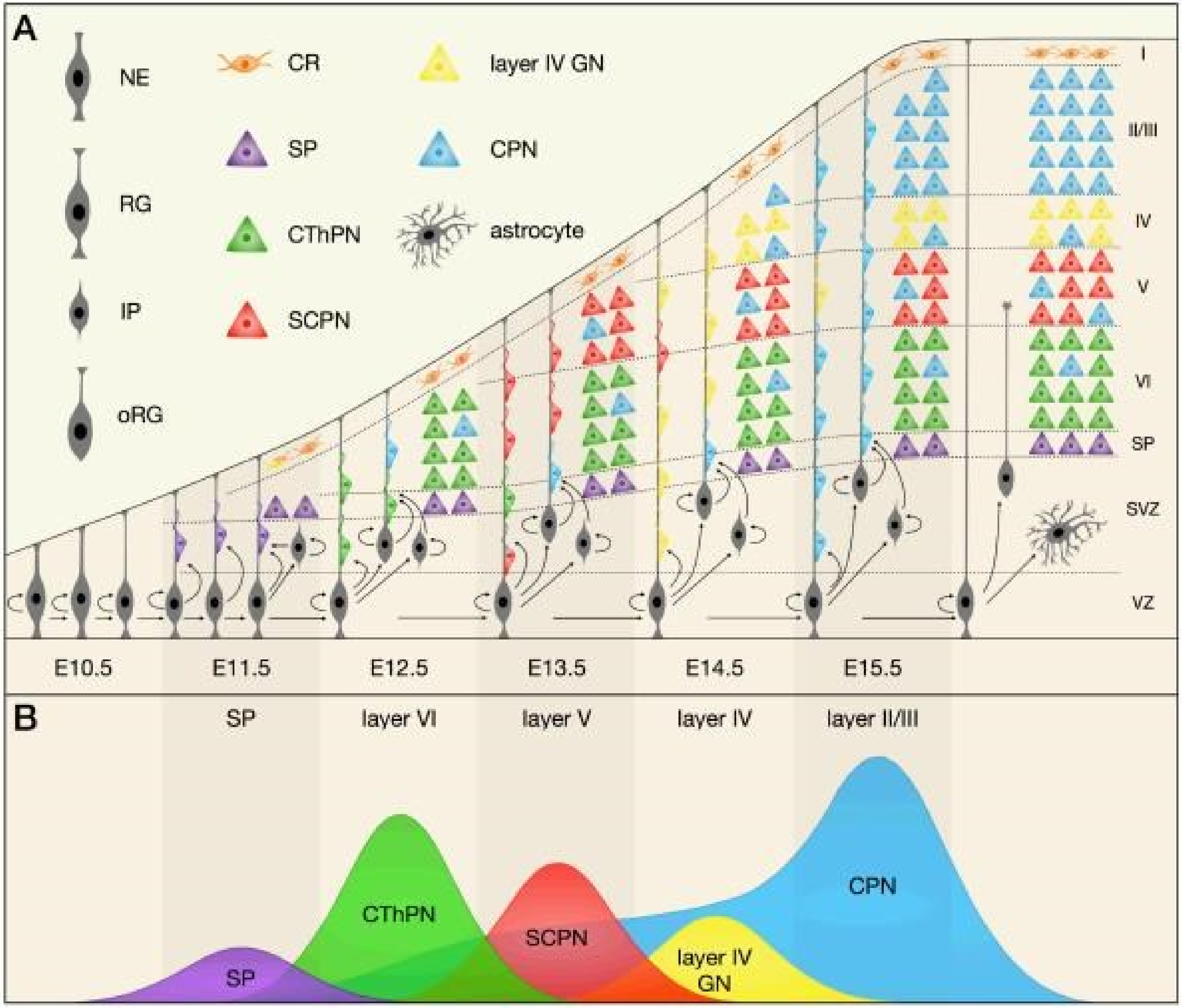


Yamashita & Petersen, Neuron, 2013

Adapted from Harris and Shepherd, 2014, review, unpublished

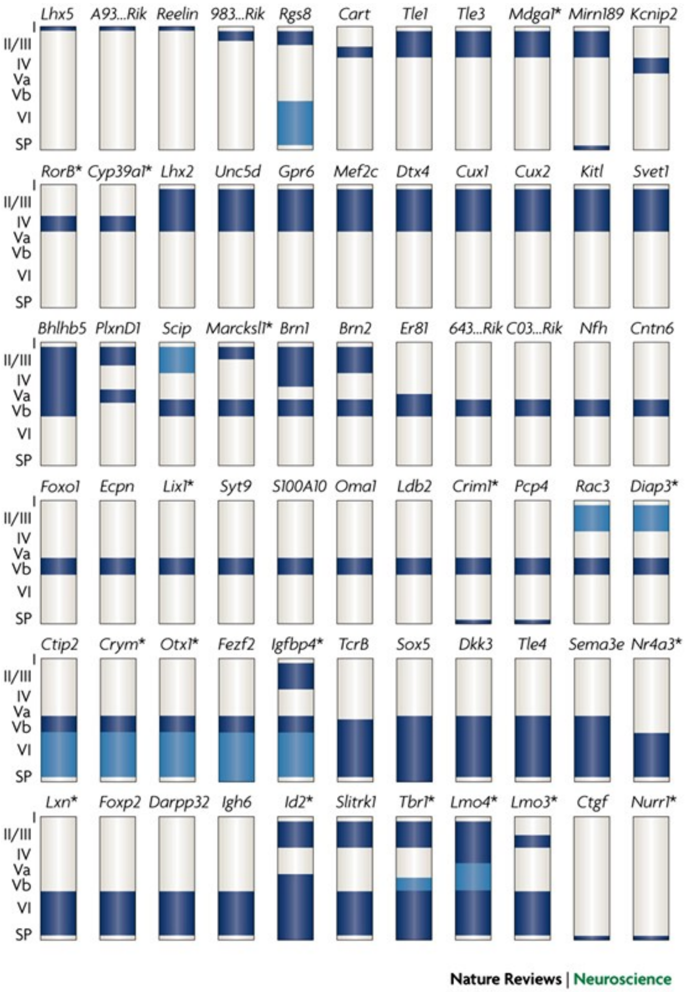
Genetic determinism of cell classes

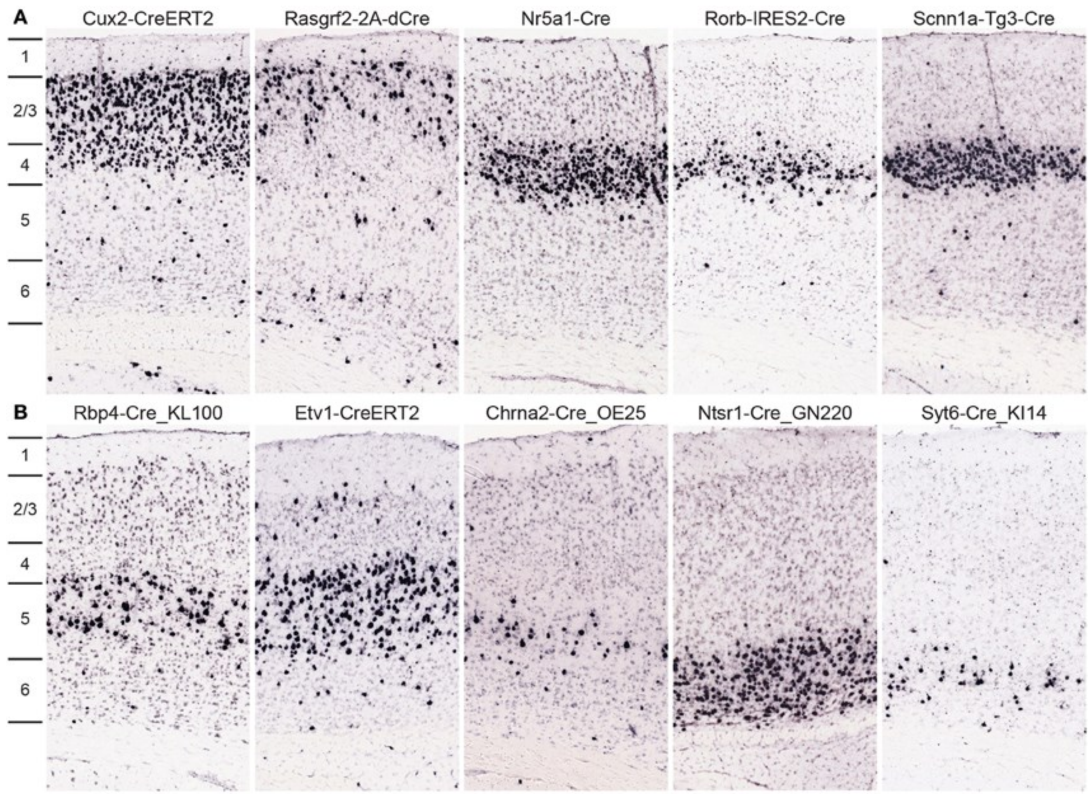
Development of cortical lamination



Nat Rev Neurosci. Nov 2013; 14(11): 10.1038/nrn3586

Molecular markers of cortical layers





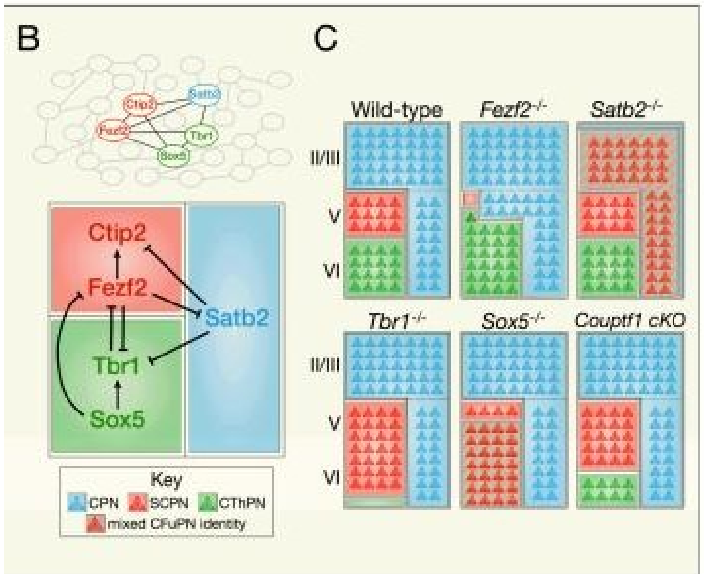
Harris et al , 2014

Molyneaux et al., 2007

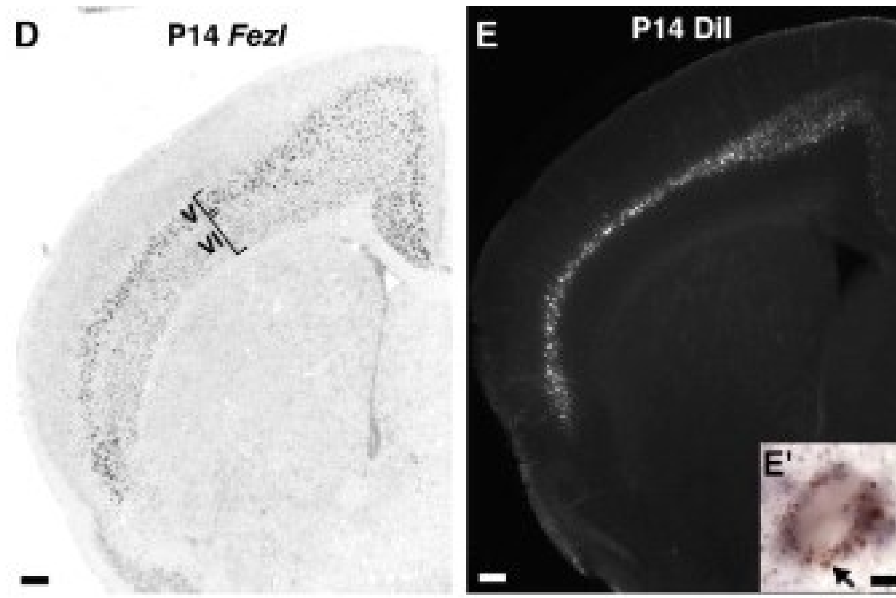
Allen Institute mission: functionally characterise cells in these Cre-lines!

e.g. Fezf2 specifies PT projection neurons

Mutually suppressive transcription factors

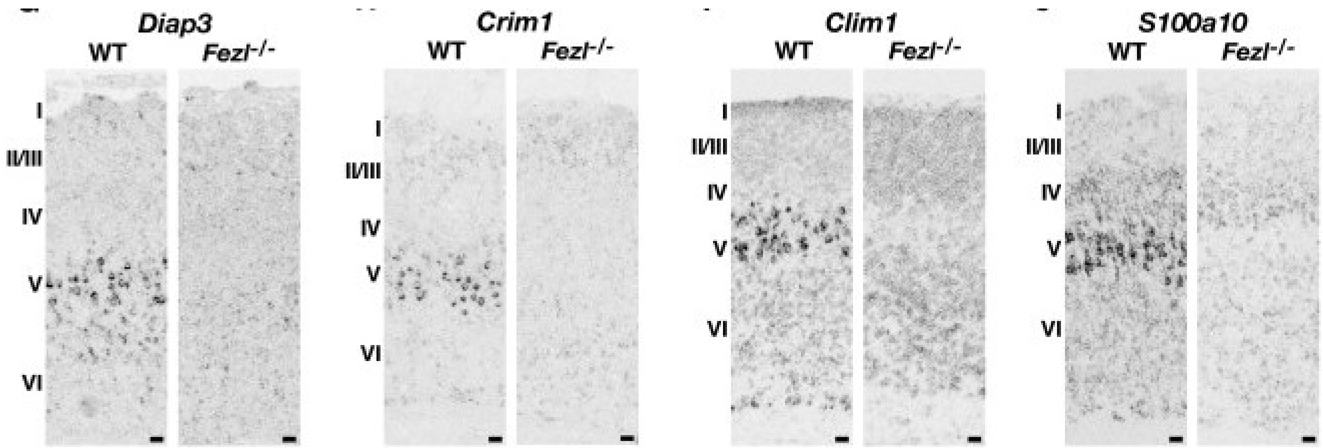


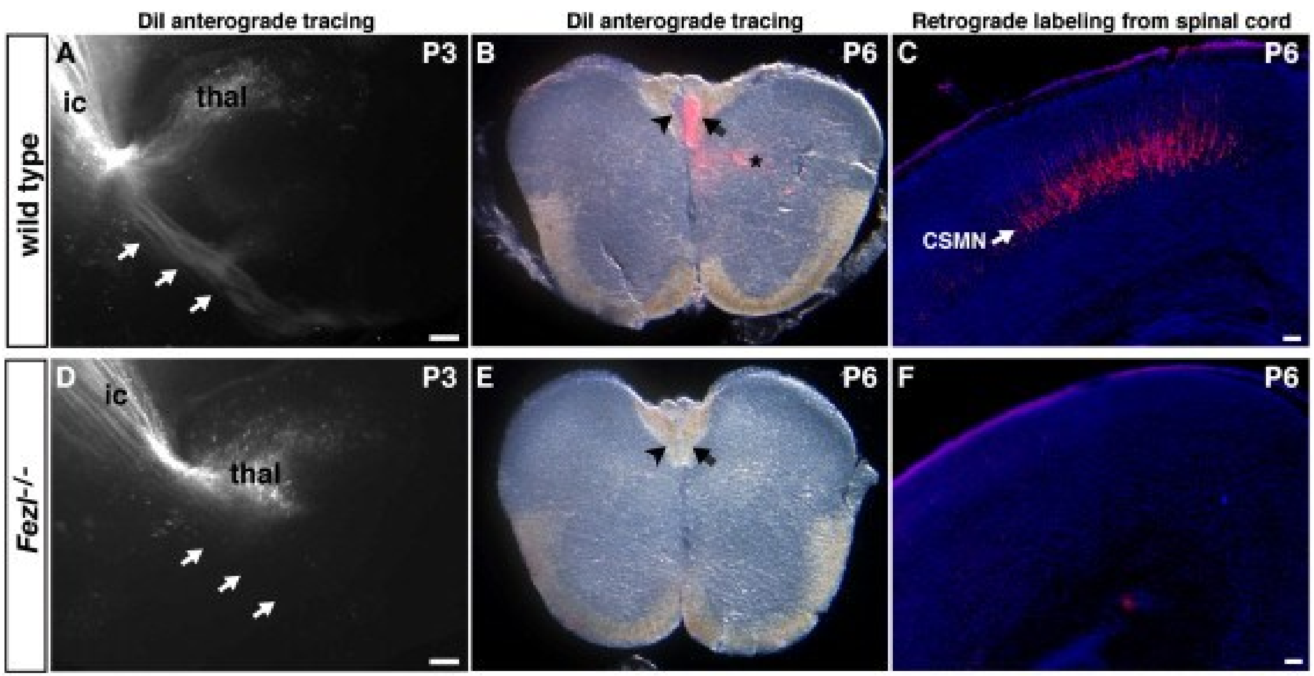
Nat Rev Neurosci. Nov 2013; 14(11): 10.1038/nrn3586



Molyneaux et al., 2005, Neuron

e.g. Fezf2 specifies PT projection neurons

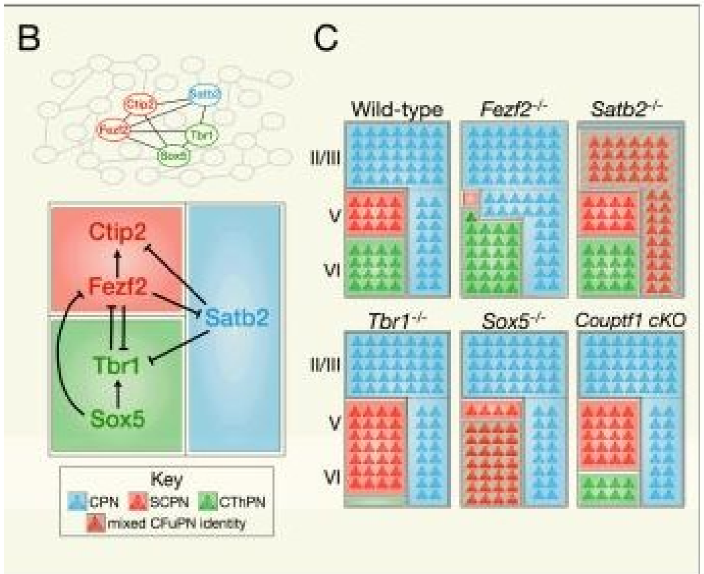




Molyneaux et al., 2005, Neuron

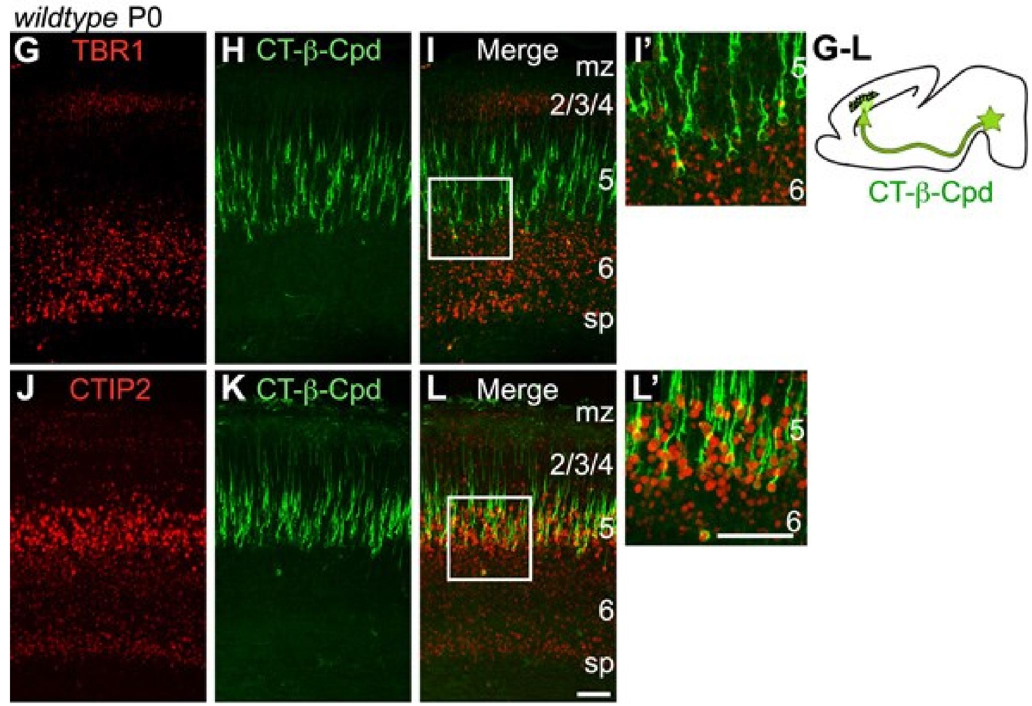
e.g. Fezf2/Ctip2 specify PT projection neurons

Mutually suppressive transcription factors



Nat Rev Neurosci. Nov 2013; 14(11): 10.1038/nrn3586

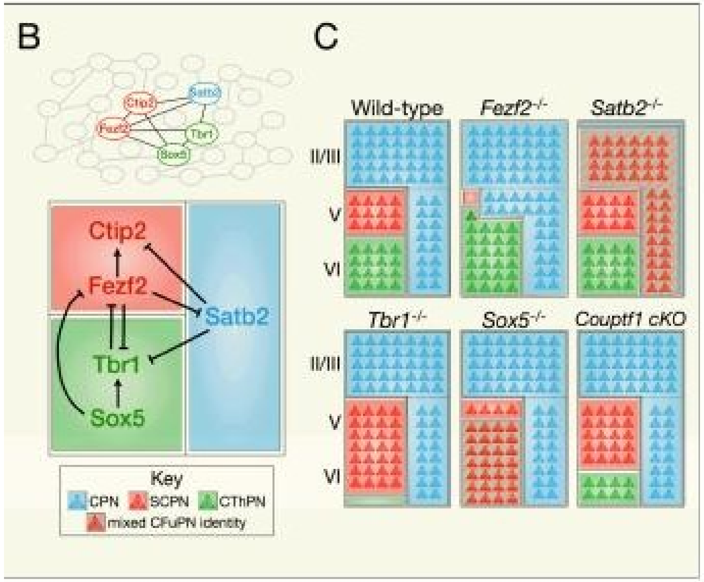
Ctip2 expressed in PT projection neurons



McKenna et al., 2011 JN

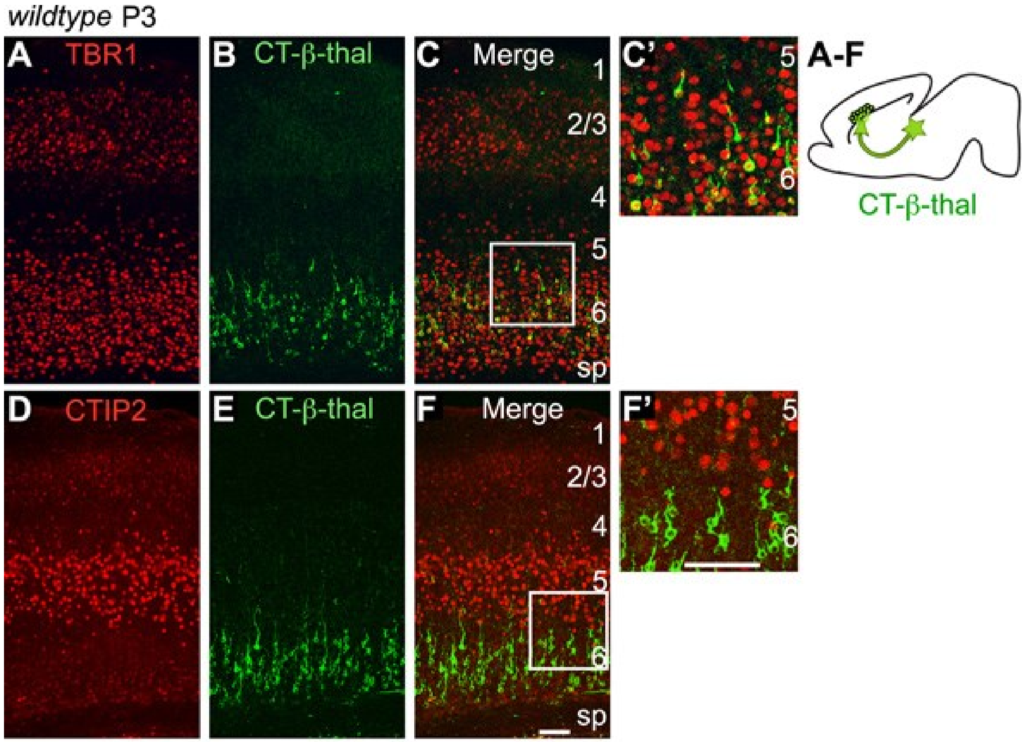
e.g. Tbr1 specifies CT projection neurons

Mutually suppressive transcription factors



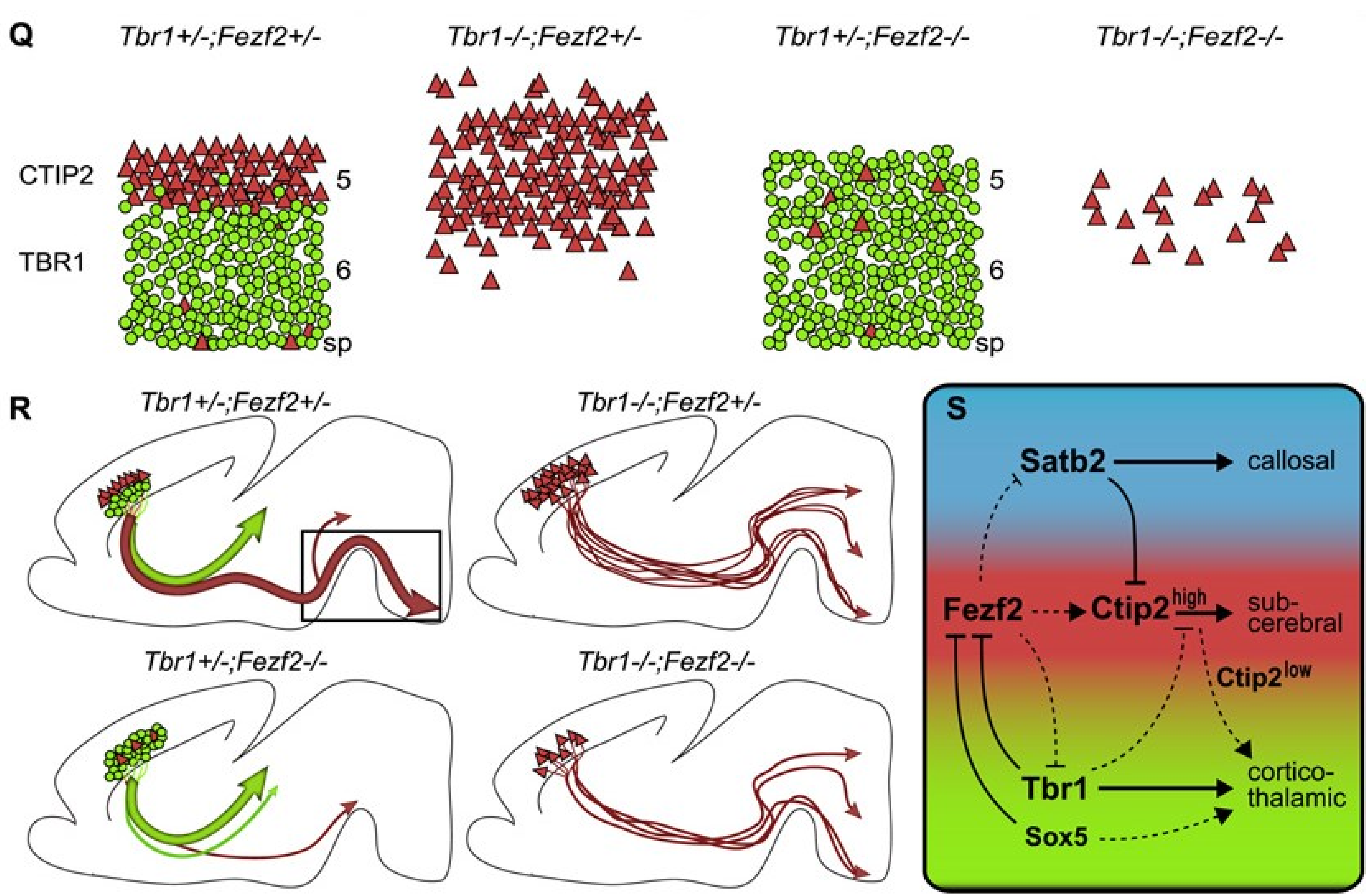
Nat Rev Neurosci. Nov 2013; 14(11): 10.1038/nrn3586

Tbr1 expressed in CT projection neurons



McKenna et al., 2011 JN

CT versus PT specification



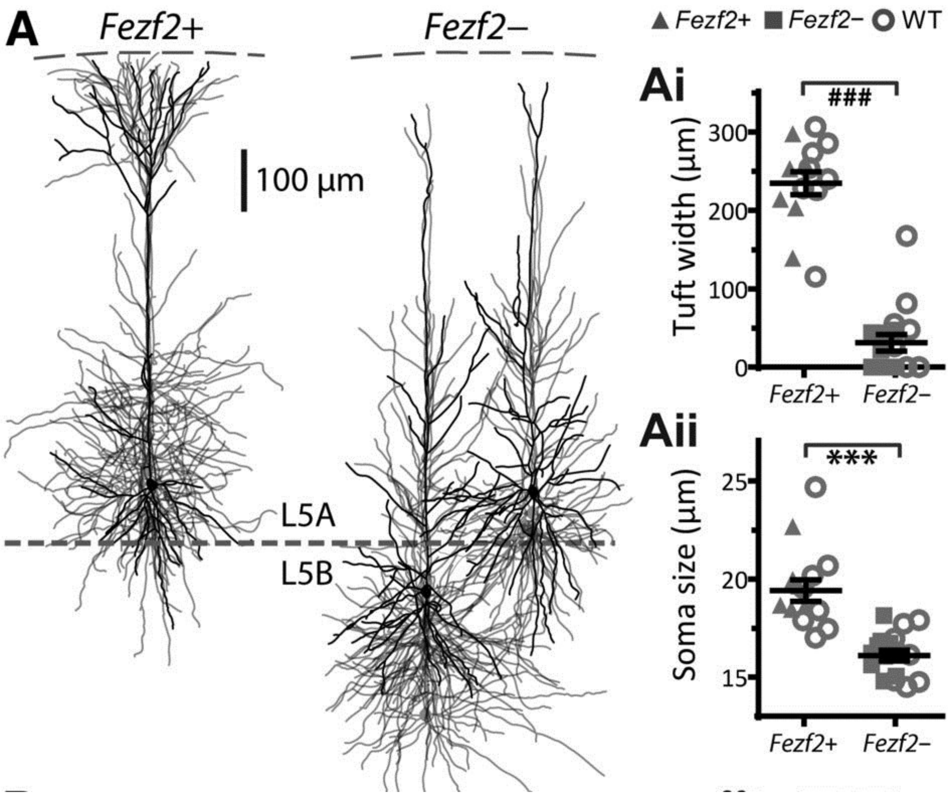
McKenna et al., 2011 JN

	IT	IT	PT	CT
	L4	Other layers		
Genes	Rorb, Satb2	Satb2	Fezf2, Ctip2	Tbr1
Inputs from local E cell classes	Few	Many, incl. L4 IT and other IT.	Many, mainly from IT	Few, mainly deep-layer (L5B/6) IT
Outputs to other local cell classes	Mainly IT, mainly L3. Sometimes PT.	IT (but not L4-IT), PT, CT	Few	Some interconnectivity with IT, possibly PT
Long-range inputs	Thalamus, lower-order cortex	Thlalamus, higher and lower-order cortex	Thalamus, higher and lower-order cortex	Higher order cortex
Long-range outputs	Few	Many, but only within telencephalon (neocortex, striatum); the only ECs sending callosal/commisural projections	Many, to multiple subcortical and subcerebral regions (brainstem, tectum, spinal cord, thalamus, basal ganglia)	Thalamus. The only ECs to excite reticular nucleus. The only ECs without longer-range corticocortical axons.
Morphology/Layers	L4 pyramidal/stellate	L2/3, 5A, 5B, 6; pyramidal	L5B, thick tufted pyramidal	L6, pyramidal
Intrinsic physiology	Regular spiking or bursting	Hyperpolarized (L2/3), little h-current, spike train adaption	Depolarized, strong h-current, little adaptation, bursting (subset)	Regular spiking
In vivo activity	Fast sensory response	Sparse firing/code	Dense code	Very sparse

IT: intratelencephalic neurons; **PT:** pyramidal tract neurons of L5B; **CT:** corticothalamic neurons of L6.

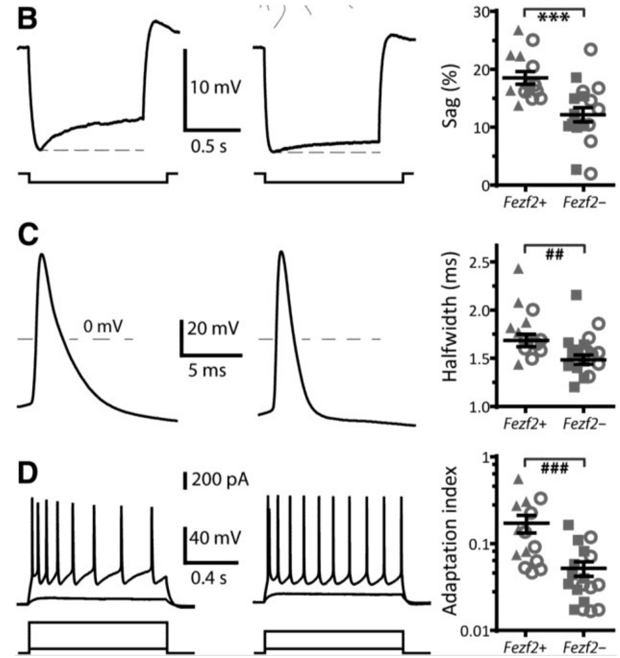
Unique electrophysiological and morphological properties of L5 Fezf2+ IT pyramidal cells

Morphology

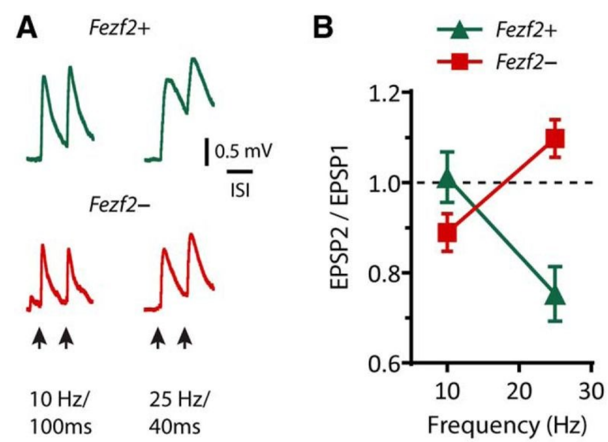


Tantirigama M L S et al. J. Neurosci. 2014;34:4303-4308

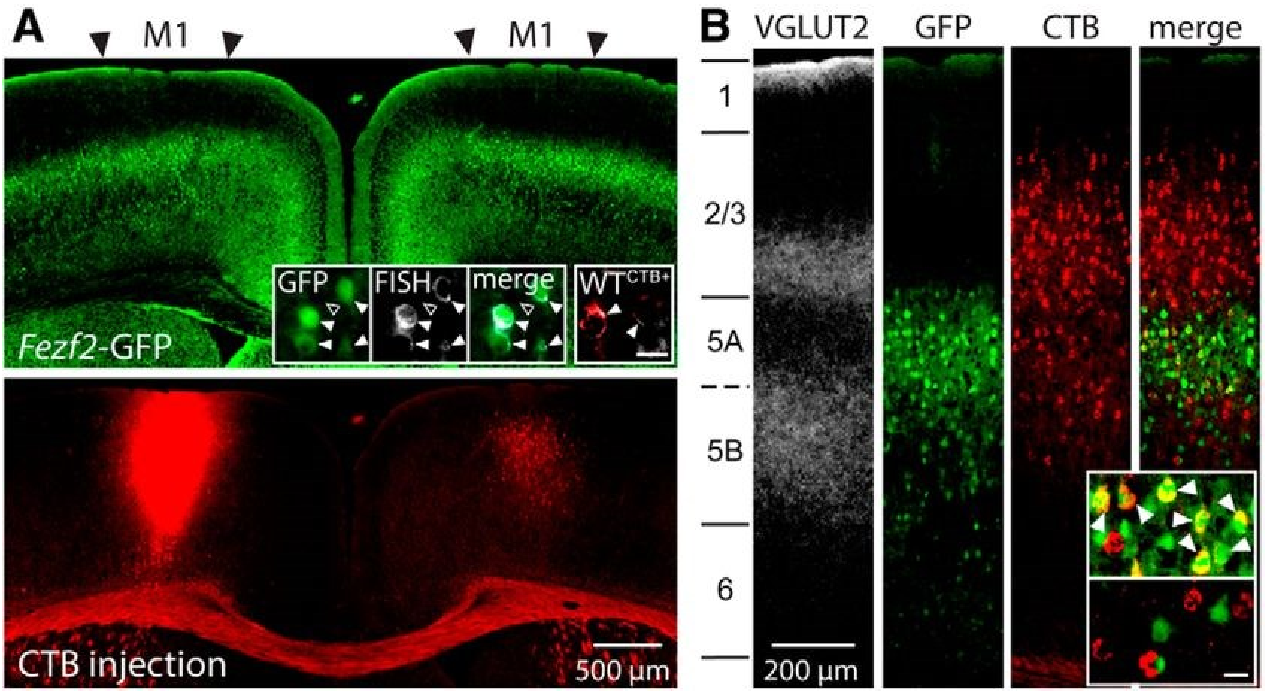
Electrophysiological properties



Short-term plasticity



Identification of Fezf2+ IT-PNs in mature M1.



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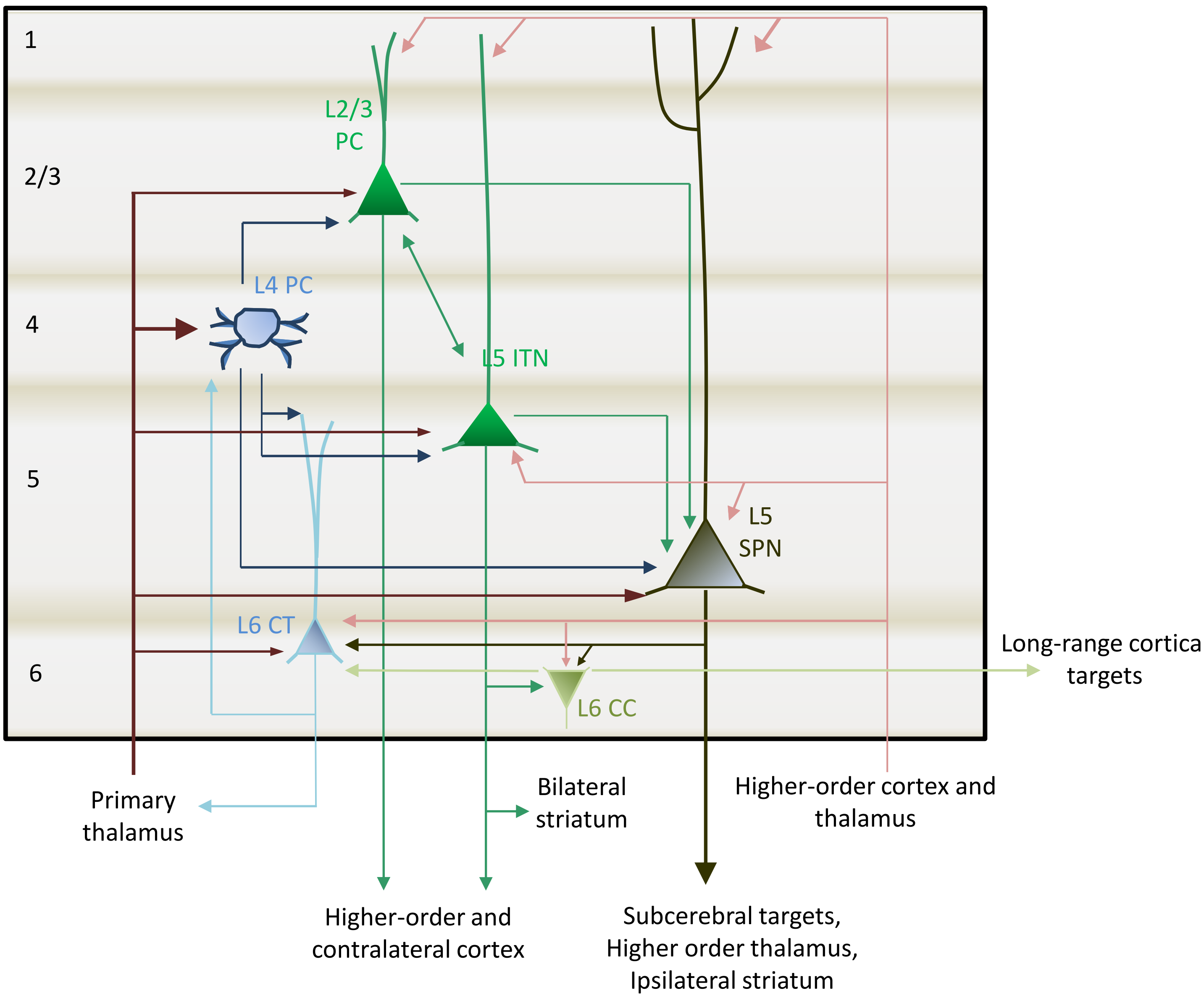
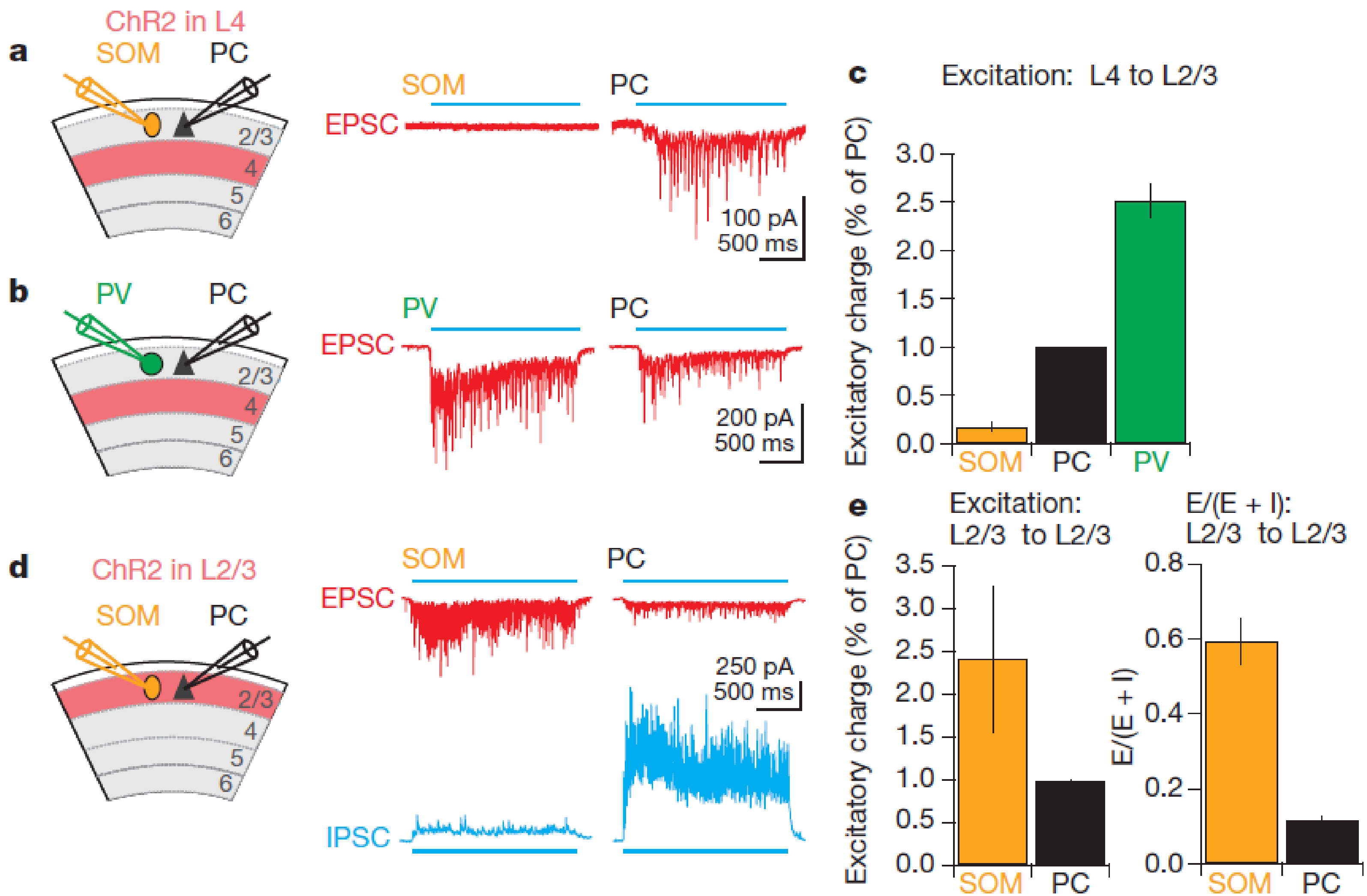
Hodology of main excitatory cortical classes



Somatostatin-positive interneurons (SOM or SST)

L 2/3 SOM cells receive much more excitatory input from layer 2/3 than from layer 4



Adesnik et al., 2013, Nature

Layer 1; ; top-down inputs – bistratified? Function?; interneurons; fear conditioning; dendrtitic spikes and plasticity
Layer 2/3; cell types; sparse activity (Sakata & Harris); pyramidal cells; specific connectivity; memory; lateral suppression (Adesnik); local feature specific amplification?
Layer 4; cell types; stellates and pyramidal; primary input layer; lacking in M1 and some other cortices; highly specialised in some cortical areas; segregate different types of information; feedforward inhibition; amplification?
Layer 5a – similar to layer 2/3
Layer 5b – dense coding, lateral spread of activity; more exuberant input connectivity; different target strctures than layer 2/3 and 5a.
Layer 6 – the deep mysterious layer. Two types; CT and CCs. Unknown role, but differences in and input and output connectivity for the two types; intracolumnar suppression?

Convergence onto PT, PT non-adapting, burst.
Bruno – input to L5 cells