

Theoretical Neuroscience

Pei Sun, Yang Tyan

Department of Psychology, Tsinghua University

peisun@tsinghua.edu.cn

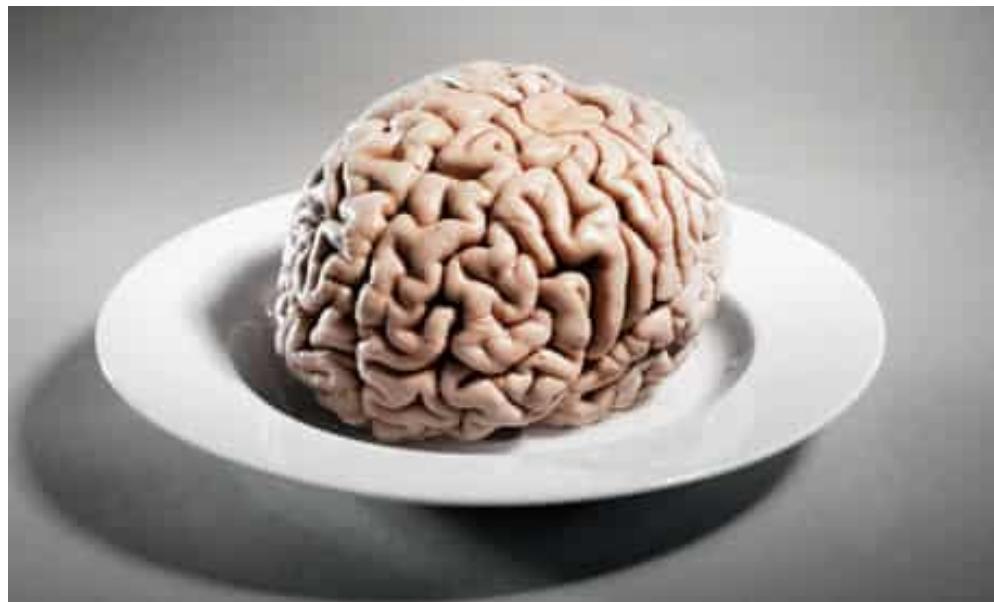
How many neurons are there in the brain?

Outlines

How many neurons are there in the brain?

What is the connectome project?

How many neurons are there in the brain?



<https://www.theguardian.com/science/blog/2012/feb/28/how-many-neurons-human-brain>

**It was a question that scientists thought
they had nailed – and the answer was
100 billion (give or take).**

<https://www.theguardian.com/science/blog/2012/feb/28/how-many-neurons-human-brain>

But when a researcher in Brazil called Dr Suzana Herculano-Houzel started digging, she discovered that no one in the field could actually remember where the 100bn figure had come from – let alone how it had been arrived at. So she set about discovering the true figure (HT to the excellent Nature neuroscience podcast NeuroPod).

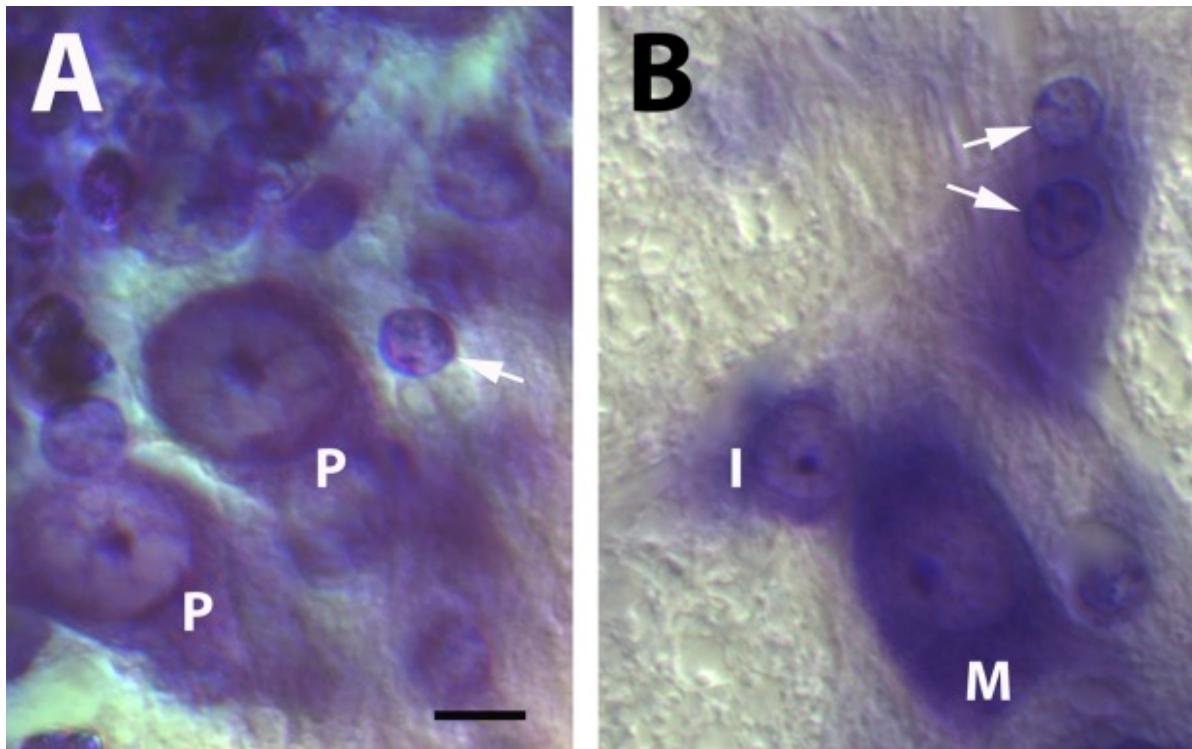


Review |  Free Access

The search for true numbers of neurons and glial cells in the human brain: A review of 150 years of cell counting

Christopher S. von Bartheld✉, Jami Bahney, Suzana Herculano-Houzel

First published: 17 May 2016 | <https://doi.org/10.1002/cne.24040> | Cited by: 27



Photomicrographs of Nissl - stained neurons and glial cells.

A: Purkinje cells (P) and granule cells (arrow) in the cerebellum of an adult mouse brain.

B: Motoneuron (M), interneuron (I), and glial cells (arrows) in the trochlear nucleus of an adult mouse brain.

有关人类大脑新皮层的统计数据

Table 1. Basic Statistics of Human Neocortex

Property	Value	Source
Surface area (mm ²)	190,000	[7, 24]
Thickness (mm)	2.5	[6, 25]
Glucose Consumption (μmol/g/min)	0.40	[10–12]
Glia/mm ³	38,000	[7]
Neurons/mm ³	40,000	[6, 26]
Synapses/mm ³	7×10^8	[5, 27, 28]
Axon Length m/mm ³	4,000	[5]
Average Axon Diameter (μm)	0.3	[5]
Dendrite Length m/mm ³	400	[5]
Average Dendrite Diameter (μm)	0.9	[5]

$$190,000 \times 2.5 \times 40,000 = 19,000,000,000$$

19 Billion

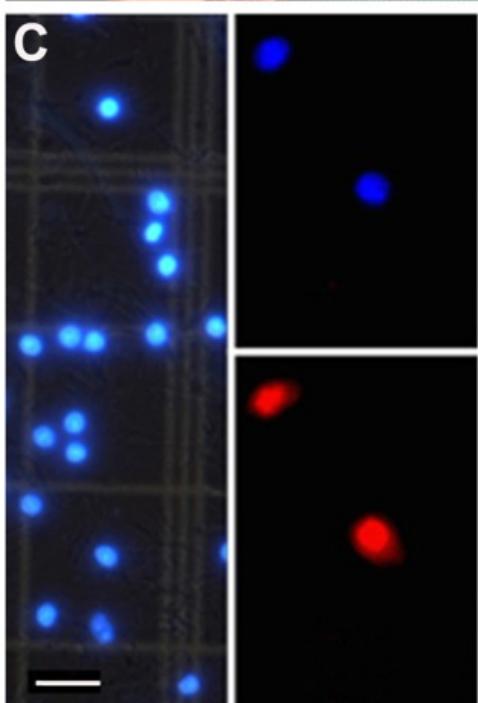
What are the challenges?

TABLE 1.

Estimates of Numbers of Neurons (N), Non-neurons (nN), and Glia (G) in Human Cerebral Cortex (in billion)

Author	Year	N One side	N Total	nN Total	G Total
Meynert	1868/1872	0.612	1.224		
Donaldson	1895		1.200		
Thompson	1899		9.282		
Berger	1921		5.512		
von Economo & Koskinas	1925		14		
von Economo	1926		13.653		
Agduhr	1941		5.0		
Shariff	1953		6.9		
Sholl	1956	5.000	10.000		
Haug & Rebhan	1956		16.5		
Haug	1959	8.200	16.400		
Pakkenberg	1966		2.6		
Gallatz et al.	1982		10.030		
Haug	1985		13.8 ± 2.4		
Haug	1987		10-19		
Pakkenberg et al.	1989		~20		
Braendgaard et al.	1990	13.7	[27.4]		
Pakkenberg	1992		25.1		
Jensen & Pakkenberg	1993		23.2		
Pakkenberg	1993		22.1		
Regeur et al.	1994		18.1		
Pakkenberg & Gundersen	1997		19.3-22.8 [range: 14.7 - 32.0]		
Gredal et al.	2000		22.3		
Pakkenberg et al.	2003		19.3-22.8		39
Pelvig et al.	2003		21.2		29.1
Koch	2004		20		
Pedersen et al.	2005		18.8		
Pelvig et al.	2008		21.4-26.3		27.9-38.9
Azevedo et al.	2009	6.18	12.36		
Azevedo et al.	2009		[16.34]	[60.84]	
Lyck et al.	2009		[15-19.7]	[35.4-40.6]	[18.5-20.3]
Karlsen & Pakkenberg	2011		17.9		18.2
Andrade-Moraes et al.	2013		[12.7]	[54.9]	

Cortex comprises only gray matter, but does not include white matter (WM), unless specifically indicated with brackets to include white matter (WM).

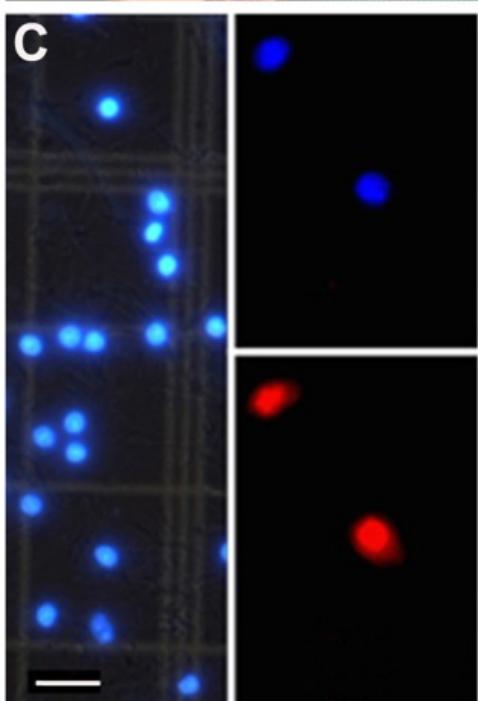


1. Fix brain tissue → 2. Homogenize
 - ↓
 3. Collect homogenate → 4. Centrifuge 10'
 - ↓
 5. Remove supernatant → 6. Add DAPI: count nuclei in supernatant
 - ↓
 7. Suspend nuclei in exact volume (Vol) of DAPI, PBS:
isotropic suspension
 - ↓
 8. Count aliquots:
total cell number = dens nuclei x Vol
 9. Stain sample for NeuN
 10. Count aliquots: determine Fr NeuN+
- Total neuron number = Fr NeuN+ x Total cell number**



“brain soup”

von Bartheld, Bahney, Herculano-Houzel. *J Comp Neurol*. 2016.



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87 Billion

von Bartheld, Bahney, Herculano-Houzel. *J Comp Neurol*. 2016.

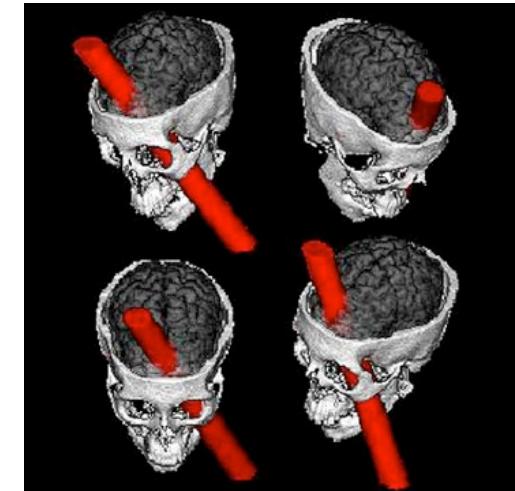
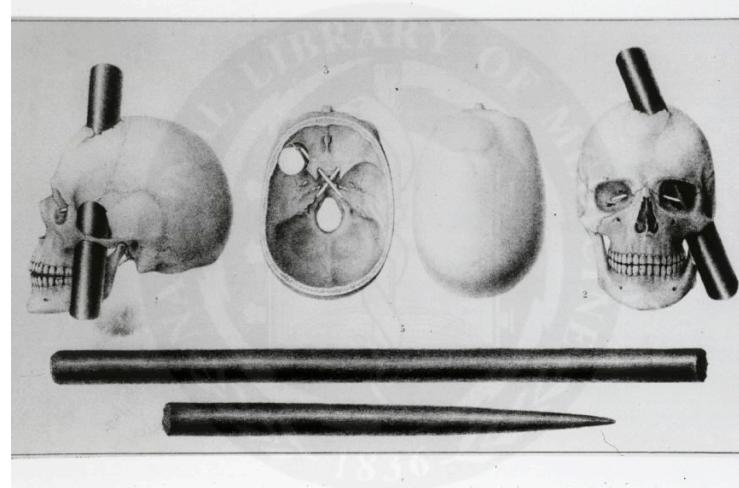
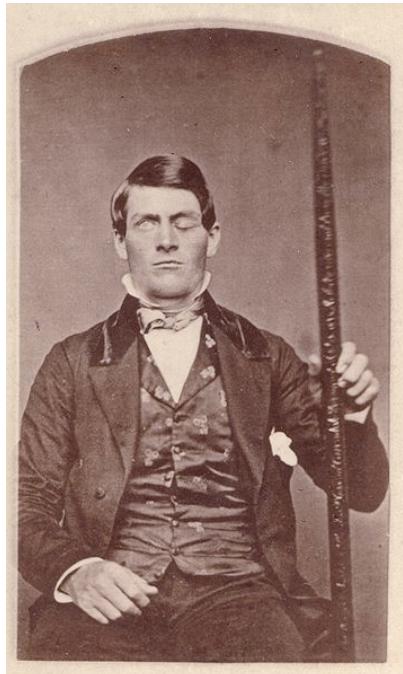
**How such a huge number of neurons work
together?**

Phineas P. Gage

(July 9, 1823 – May 21, 1860)

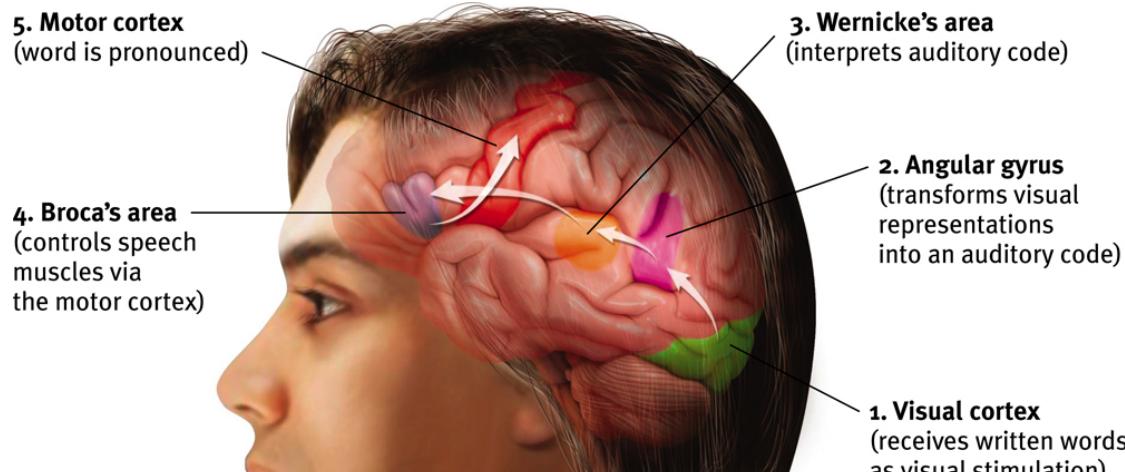
American Crowbar Case

Stimulates discussion on “brain localization” in the 19th century

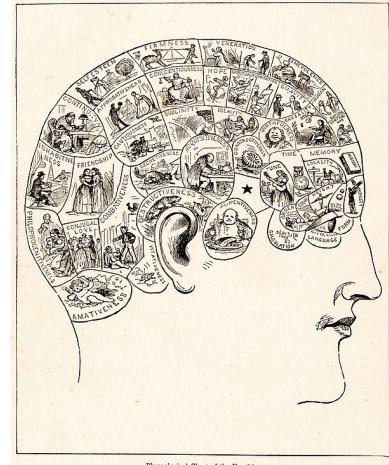


Language

Aphasia is an impairment of language, usually caused by left hemisphere damage either to **Broca's area** 布洛卡区 (impaired speaking, 1861) or to **Wernicke's area** 维尔尼克区 (impaired understanding, 1874).



Phrenology (颅相学)

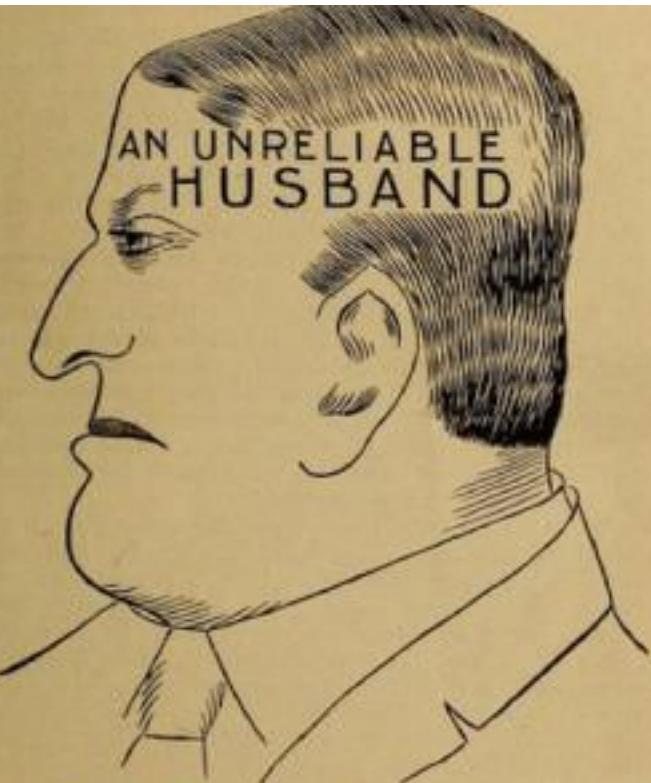


Phrenology (from Ancient Greek φρήν (phrēn), meaning 'mind', and λόγος (logos), meaning 'knowledge') is a pseudomedicine primarily focused on measurements of the human skull, based on the concept that the brain is the organ of the mind, and that certain brain areas have localized, specific functions or modules.

Phrenology (颅相学)

Developed by German physician Franz Joseph Gall in 1796, the discipline was very popular in the 19th century, especially from about 1810 until 1840. The principal British center for phrenology was Edinburgh, where the Edinburgh Phrenological Society was established in 1820.

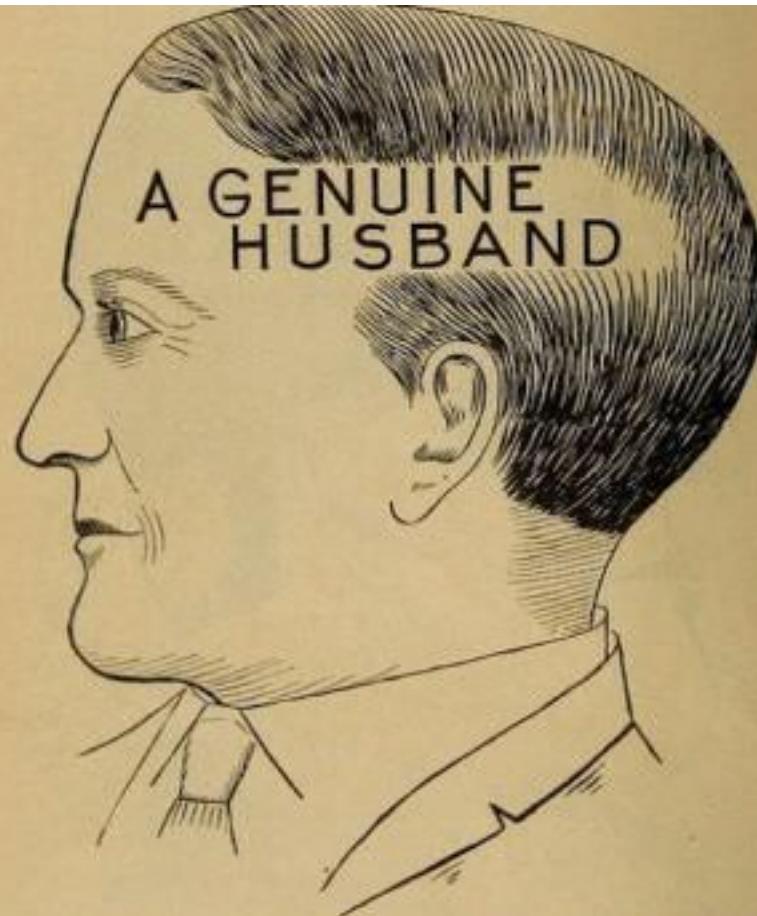
Gall's assumption that character, thoughts, and emotions are located in specific parts of the brain is considered an important historical advance toward neuropsychology.



The reason this man is an unreliable husband is because he is very weak in Conjugality and Parental Love and exceedingly strong in Amativeness. Young ladies, beware of such men as husbands.

BIGAMY.

Ridicule comes directly from Amativeness. Con-



Young ladies, indelibly fix this shape of head in your

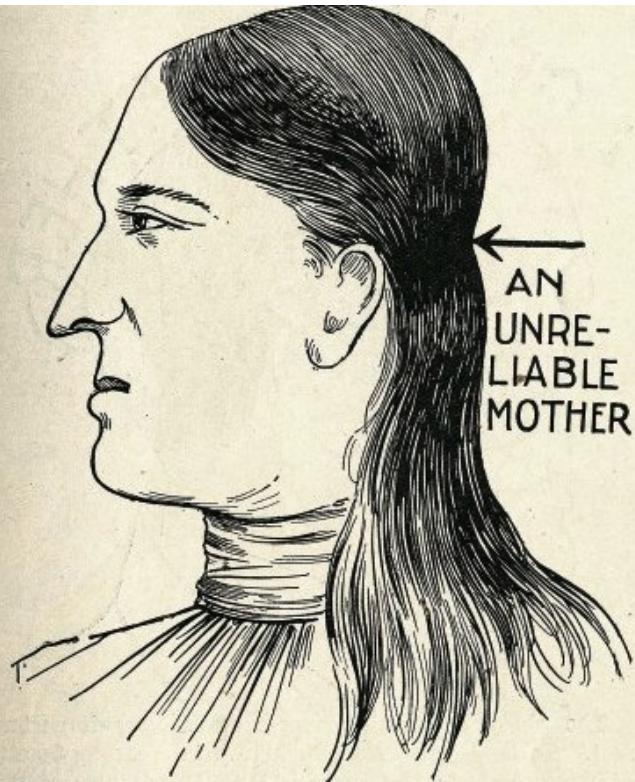


A GENUINE MOTHER.

We affirm in the most absolute manner that words can be used that mother love is located exactly where this backhead projects most. To be a true, natural mother is to have this faculty highly developed. Young men, fix this picture in your minds.

MOTHER LOVE.

Mother love is nothing more nor less than the faculty of Parental Love. It all comes from this one faculty.



This is a striking illustration. It will pay all to remember this head formation and especially all men who would select wives who will make good mothers.

SLOVENLINESS.

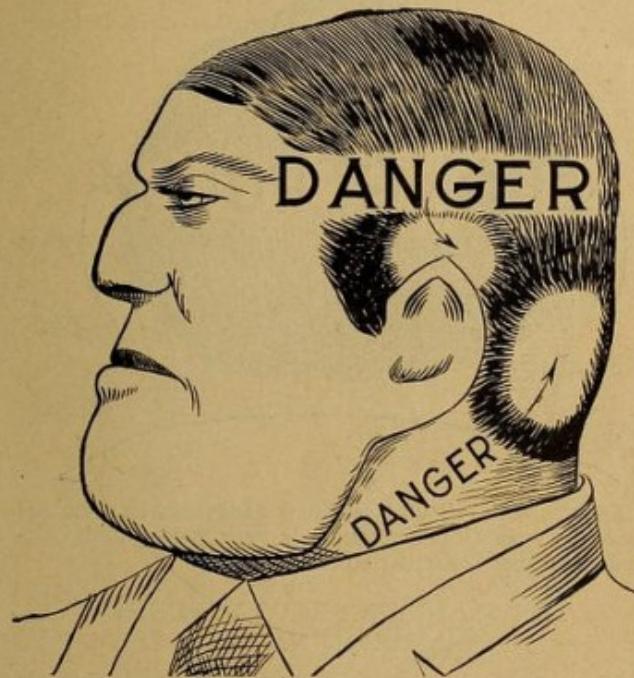
Why is one slovenly? Because his faculties of Ideality, Order, Self-esteem and Approbateness are weak. Positively nothing more true.

ANOTHER KIND OF CRYING.

There are selfish children who seem to cry but do not. They use the cry as a means to an end. This should not be termed crying, but calling, bawling, howling, screeching.



Always look for sentiment in these two regions of the head. There are no other sentiments and no other places to look for them. We say this with absolute certainty.



We wish to emphasize in the most absolute way the fact that so far as a human being is concerned all danger lies in these two faculties. They are easily located and should be understood by every man, woman and child. Be on guard against the danger in such men and women.

Discussion

Do you think the shapes of faces (physiognomy, 面相学)
carry information about personality?



Function vs. Structure

For any system, philosophy

Discussion

What the brain can do?

What the brain can not do?

Function vs. Structure

Connectome

Genome

The Human Genome Project

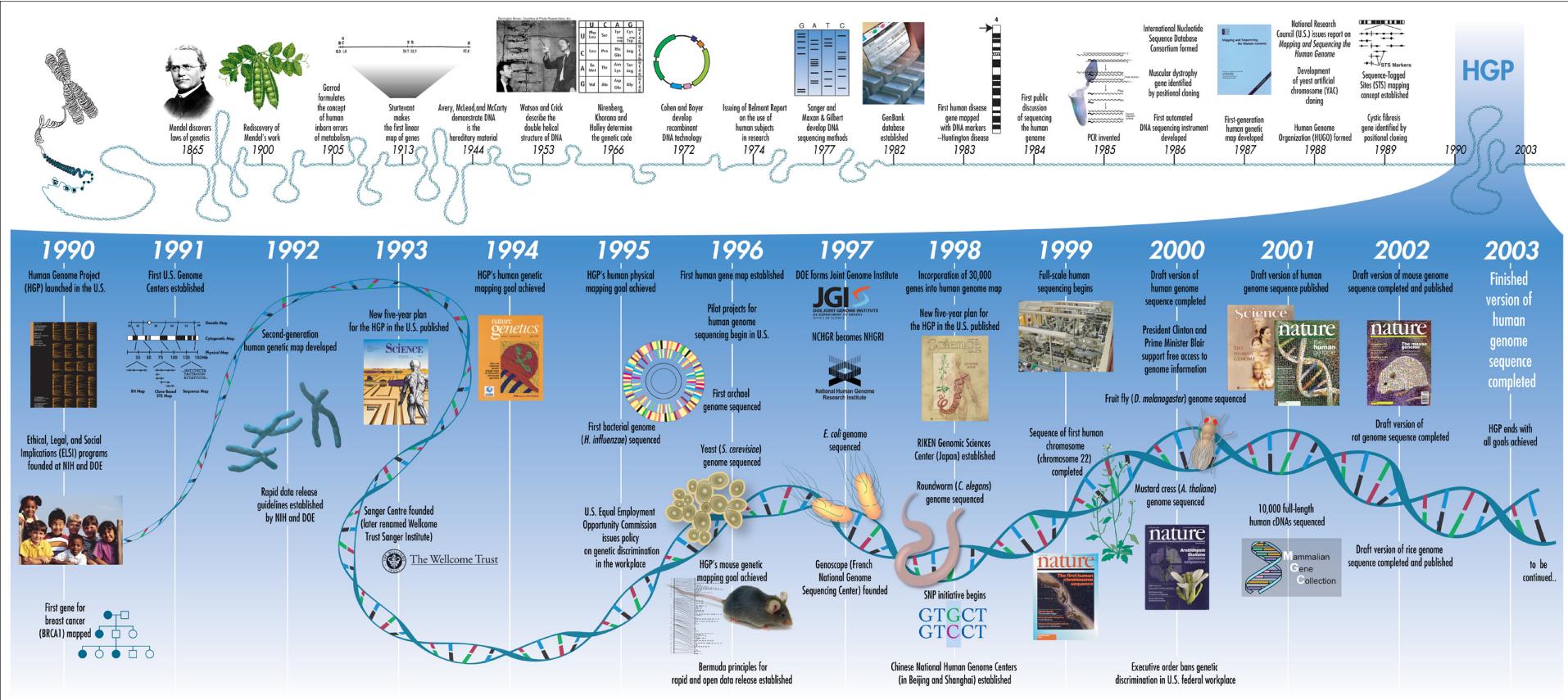
THE HUMAN GENOME

3 billion bases 30,000 genes

<http://www.genome.gov/>

Partnership for Minority Advancement
in the Biomolecular Sciences

Genome



Proteomics?

连接组学（connectomics）

连接组学的研究目标是理解和分析全脑的神经连接。

连接组学有可能会成为一个全新的、独立的研究领域，因为传统意义上的神经科学主要集中在单个或一些神经元的结构和功能上，而连接组学是在更大范围和尺度上，对于整体神经网络的探索。

在某种程度上，连接组学与神经科学的关系，类似于基因组学和基因学的关系

Where did the researchers start?

Review



Cite this article: Emmons SW. 2015 The beginning of connectomics: a commentary on White *et al.* (1986) 'The structure of the nervous system of the nematode *Caenorhabditis elegans*'. *Phil. Trans. R. Soc. B* **370**: 20140309.

<http://dx.doi.org/10.1098/rstb.2014.0309>

One contribution of 18 to a theme issue 'Celebrating 350 years of *Philosophical Transactions*: life sciences papers'.

Subject Areas:

The beginning of connectomics: a commentary on White *et al.* (1986) 'The structure of the nervous system of the nematode *Caenorhabditis elegans*'

Scott W. Emmons

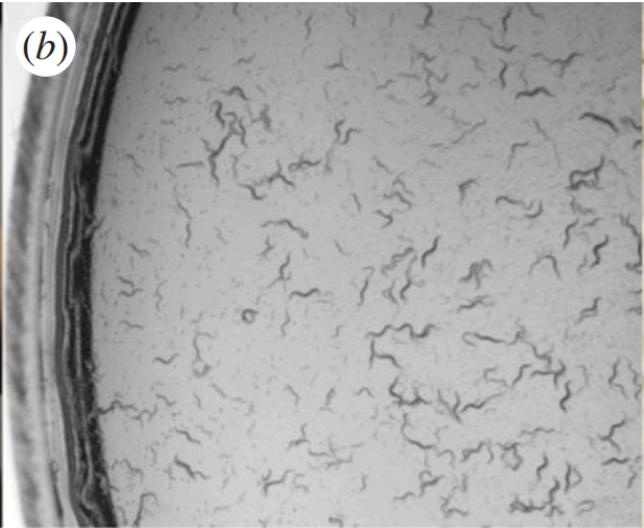
Department of Genetics, Albert Einstein College of Medicine, Bronx, NY, USA

The article 'Structure of the nervous system of the nematode *Caenorhabditis elegans*' (aka 'The mind of a worm') by White *et al.*, published for the first time the complete set of synaptic connections in the nervous system of an animal. The work was carried out as part of a programme to begin to understand how genes determine the structure of a nervous system and how a nervous system creates behaviour. It became a major stimulus to the field of *C. elegans* research, which has since contributed insights into all areas of biology. Twenty-six years elapsed before developments, notably more powerful computers, made new studies of this kind possible. It is hoped that one day knowledge of synaptic structure, the *connectome*, together with results of many other investigations, will lead to an understanding of the human brain. This commentary was written to celebrate the 350th anniversary of the journal *Philosophical Transactions of the Royal Society*.

‘The mind of a worm’

How genes determine the structure of a nervous system and how a nervous system creates behaviour.

The landmark paper, ‘The structure of the nervous system of the nematode *Caenorhabditis elegans*’ described for the first time the map of the entire nervous system of an animal.



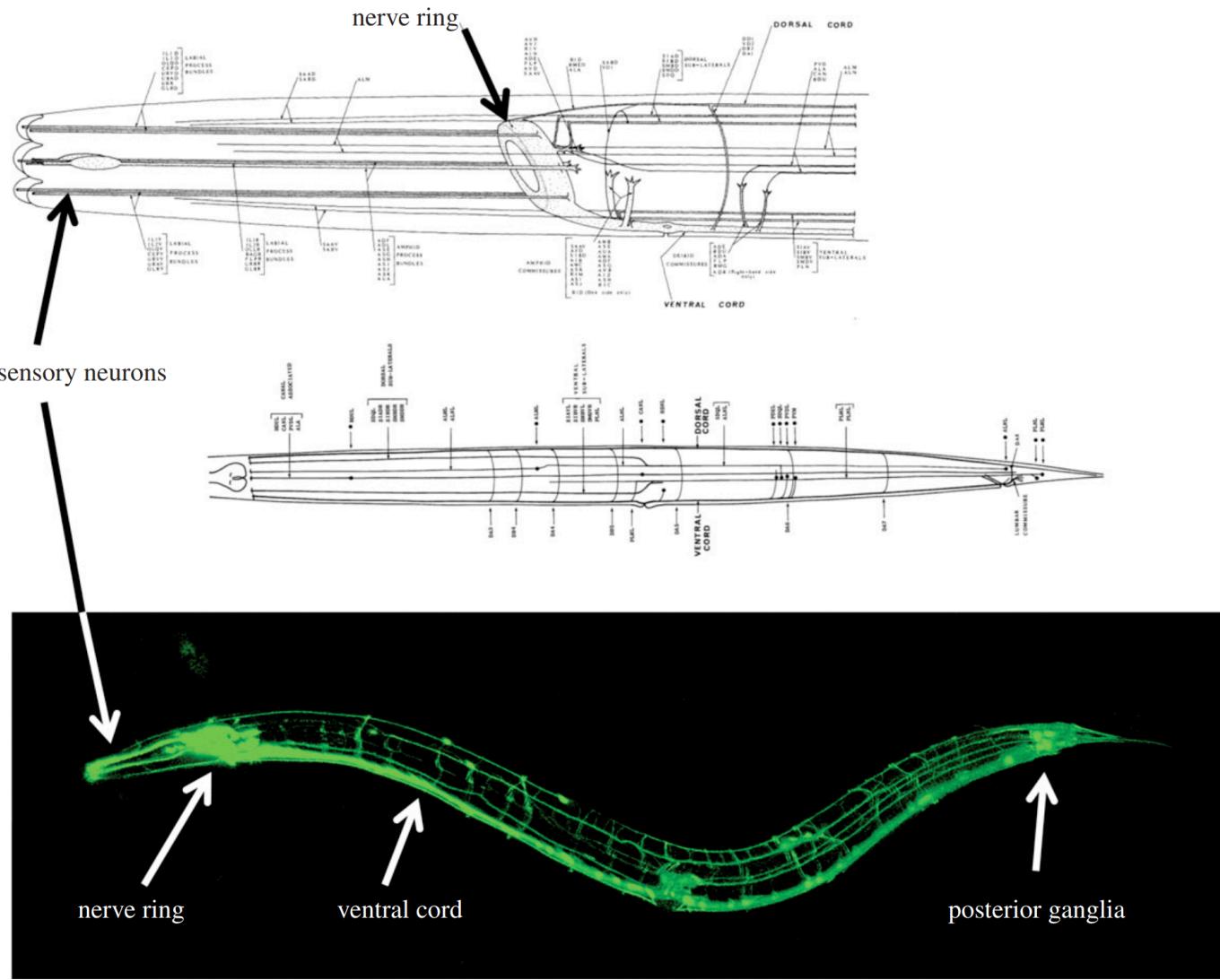
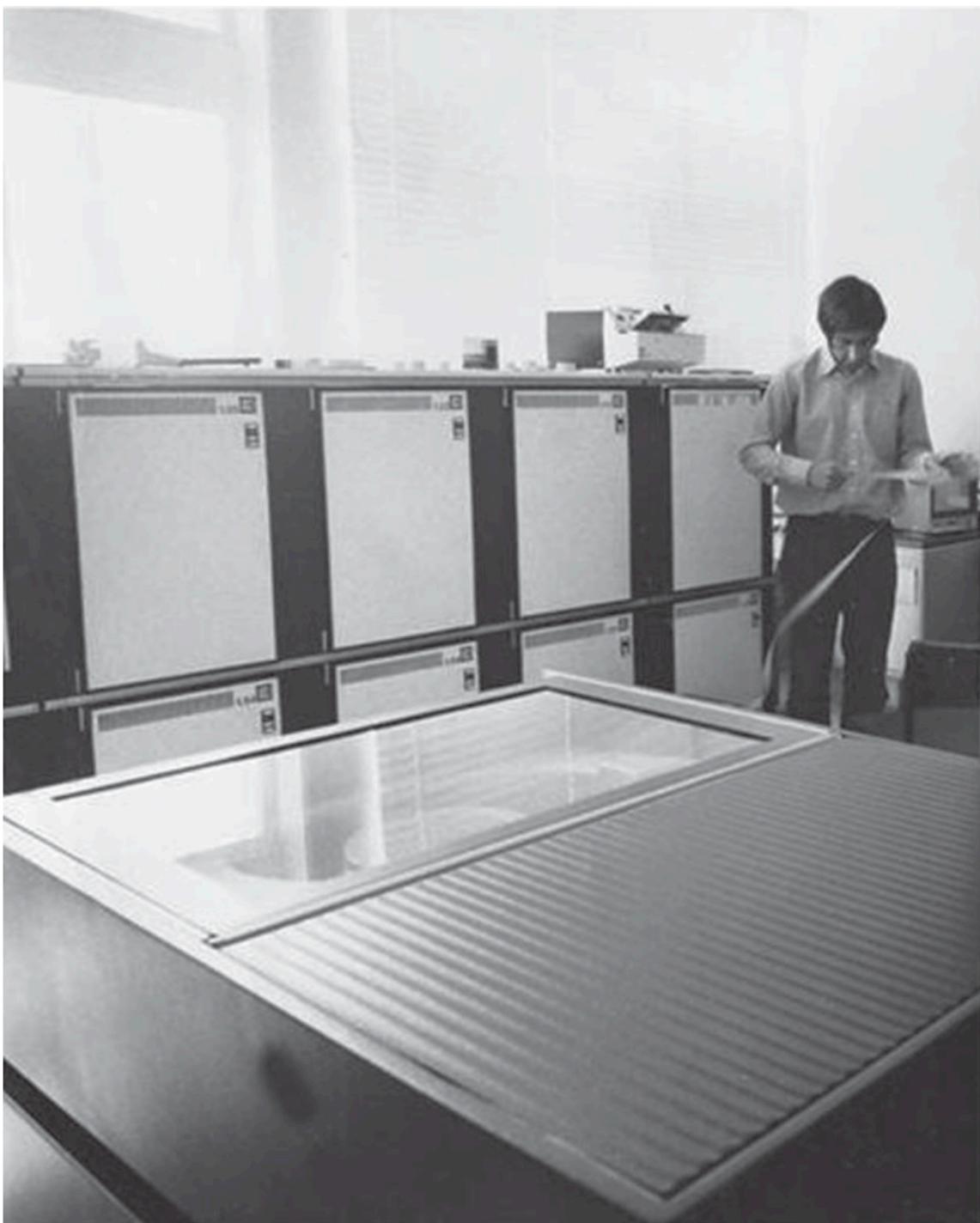
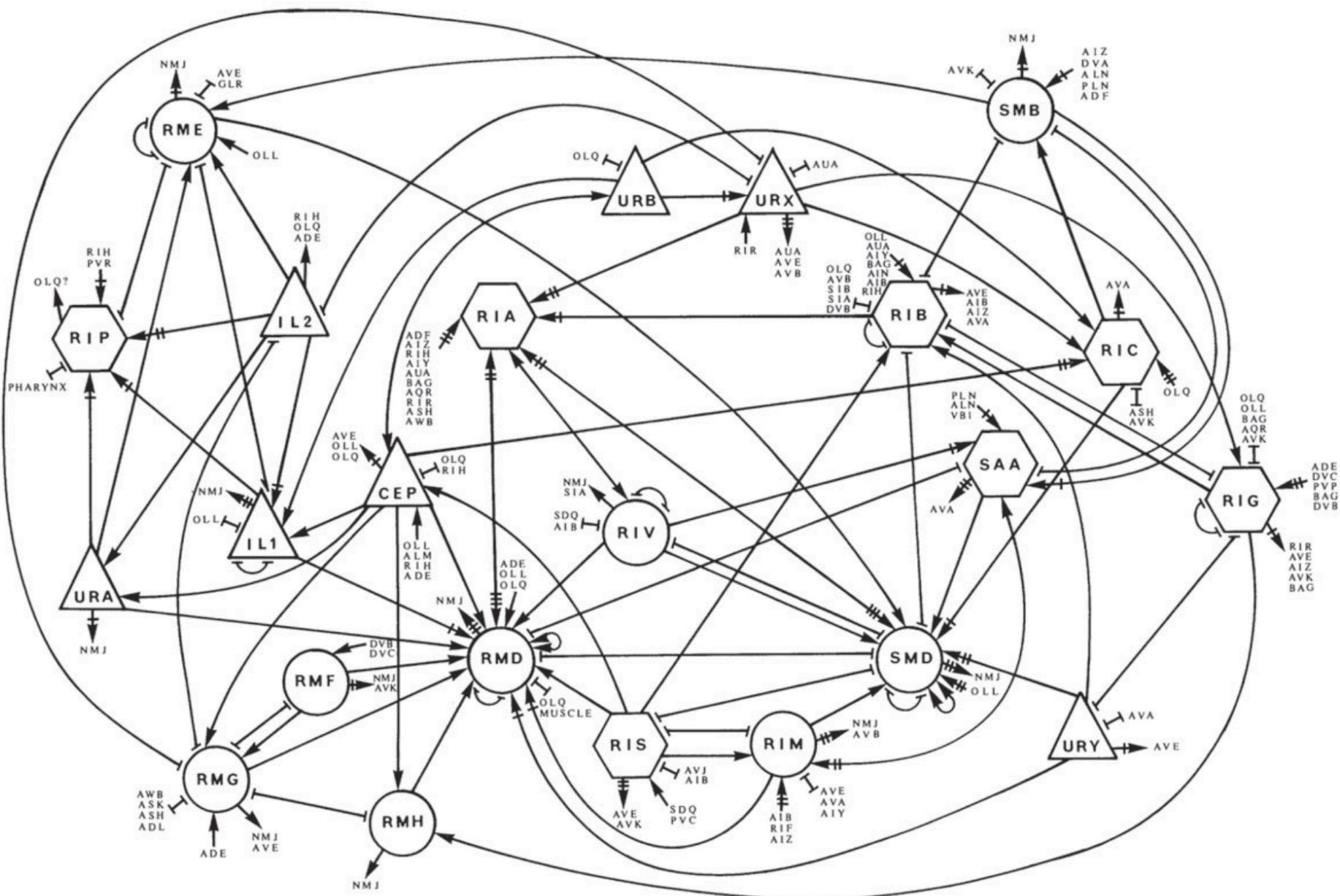


Figure 2. The *C. elegans* nervous system. Top: diagrams from 'The mind of a worm' [1, figs 6 and 7]. Bottom: a worm expressing the fluorescent protein GFP in its entire nervous system. The hermaphrodite nervous system contains precisely 302 neurons, the male, 383 (46% of its somatic nuclei). The nerve ring surrounding the pharynx contains complex circuitry governing most aspects of behaviour. This is the closest thing the worm has to a 'brain'. The ventral nerve cord contains motor-neurons that govern undulatory locomotion. Many sensory neurons have endings arrayed around the mouth. The extra male neurons are mostly situated in the tail where they form the circuits for mating. (Picture from Hang Ung, Jean-Louis Bessereau laboratory, France, with permission.)







But there was one big problem. At 450 pages, the manuscript was 350 pages over their maximum! It would cost the society above £17 000 to publish and would increase each subscriber's subscription cost by 25%! Eventually, funds were found and publication went ahead, but it had to wait a year and added a volume to the 1986 output. The authors were amused when they found the editors had used 'The mind of a worm' as the running head.

连接组学 (connectomics)

线虫大约有300个神经元，是连接组学的理想模型动物。实际上，这项工作从1970年代就已经开始，研究者们付出了极大的努力来完成这项工作，花费了将近十年的时间。

由于大鼠约有7500万个神经元，人类约有1000亿个神经元，要探索这些更为复杂和发达大脑的连接组学，就必须发展出新的研究方法和技术。

连接组学 (connectomics)

连接组学有不同的空间尺度，主要可以分为微观 (microscale)、介观 (mesoscale) 和宏观 (Macroscale) 三个水平，对应分别是突触 (synapse)、细胞和脑区间的神经连接，常用影像技术分别为电子显微镜、光学显微镜 (light microscopy) 和磁共振成像 (magnetic resonance imaging, MRI)。

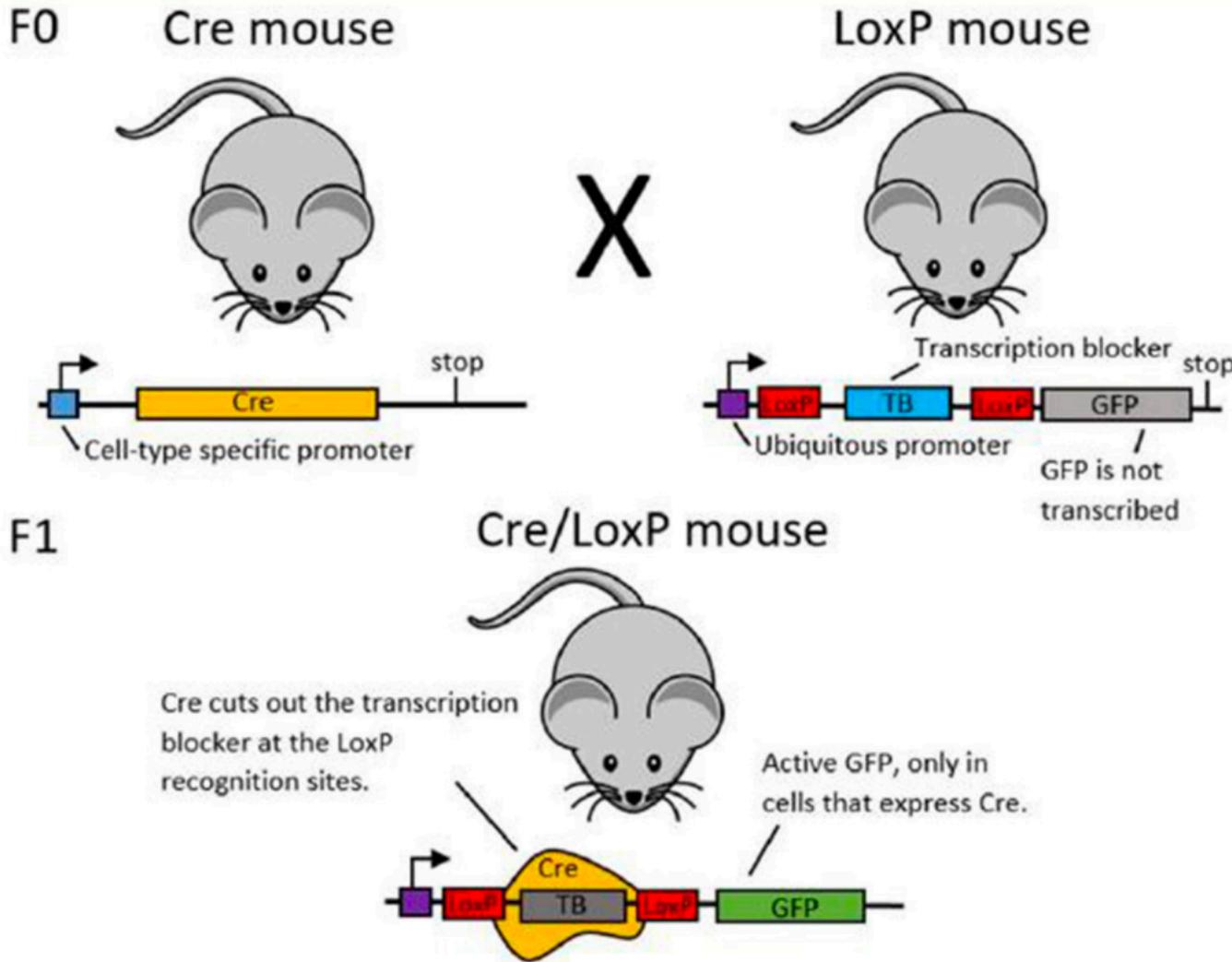


FIGURE 3 | Schematic illustration of the Cre/LoxP system. In the F0 generation, mouse line 1 (**left**) expresses Cre under a cell-type specific promoter. Mouse line 2 (**right**) expresses the labeling protein (here: GFP), but has an upstream transcription blocker, which prevents transcription of GFP. When these two mouse lines are crossed, some offspring will have both the Cre DNA and the LoxP-TB-LoxP-GFP DNA. In these animals, Cre is expressed only in the desired cell type and in this cell type, Cre cuts out the transcription blocker at the LoxP sites. This enables GFP transcription and thereby cell-type specific labeling.

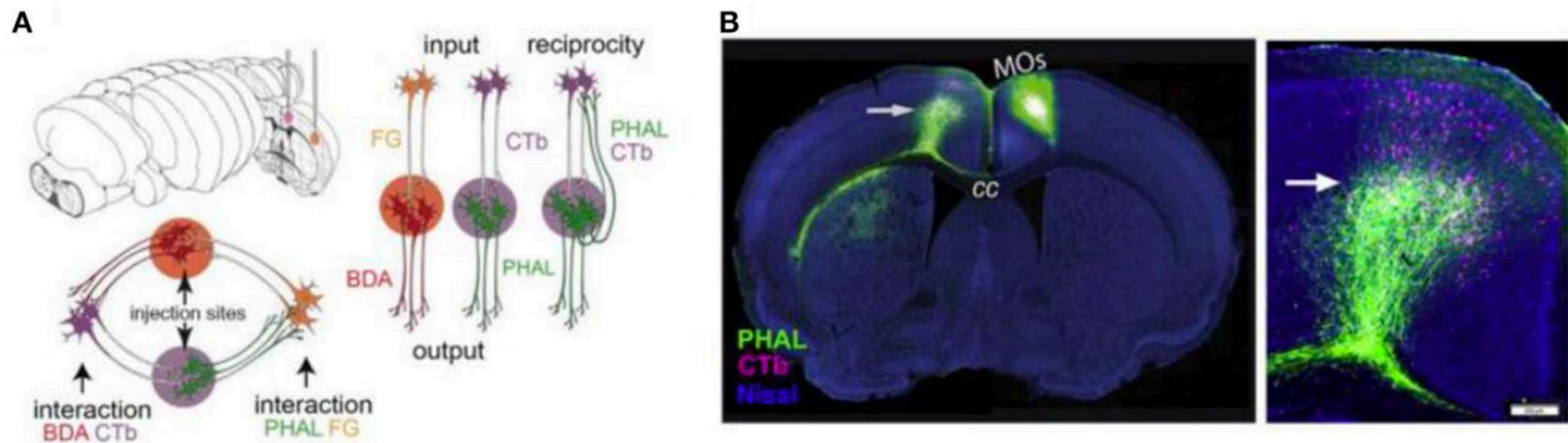


FIGURE 4 | Projection tracing using double co-injection labeling. (A) Schematic illustration of how four different types of pathways can be labeled using four different tracers in two injection sites. PHAL, *Phaseolus vulgaris* leucoagglutinin; BDA, biotinylated dextran amine; CTb, cholera toxin subunit b; FG, Fluorogold. **(B)** Example fluorescence images showing one injection into a secondary somatomotor area (Mos) (left) and an area both providing input (cells labeled in pink) and receiving output (cells labeled in green) from this area in the opposite hemisphere (right). Adapted from Zingg et al. (2014, p. 1098). Copyright 2014 by Elsevier Inc.

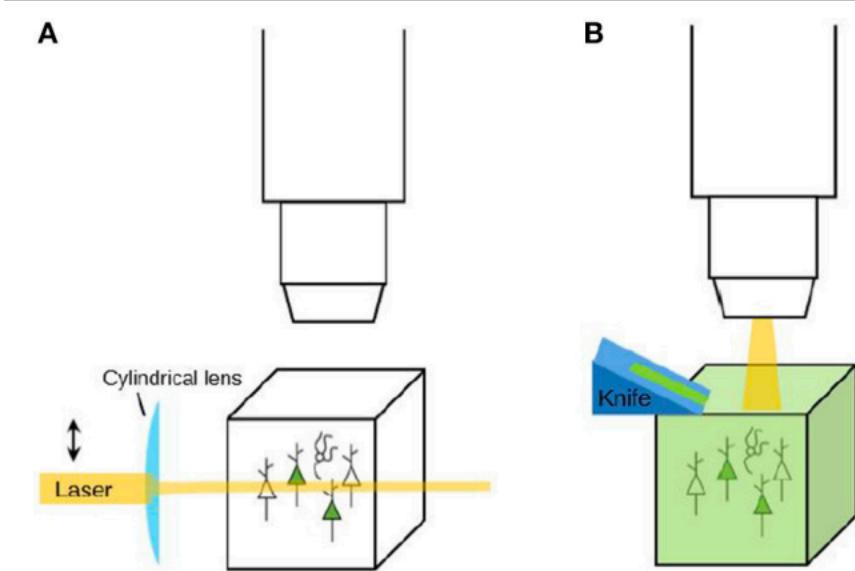


FIGURE 5 | Imaging strategies: clearing and optical sectioning vs. physical sectioning (tomography). **(A)** When a labeled tissue is cleared, there is no need to cut the tissue. Instead, tools like light-sheet microscopy (as depicted here) can be used to selectively illuminate the tissue and thereby perform *optical sectioning*. **(B)** When a tissue is not cleared, it needs to be sectioned in order to reveal the labeling that is present in deeper layers. A tomography system is a system that automatically sections and images a tissue. Sectioning may be performed before imaging (the resulting tissue ribbons can then subsequently be imaged) or after imaging (to reveal a new layer of tissue for imaging).

Brainbow

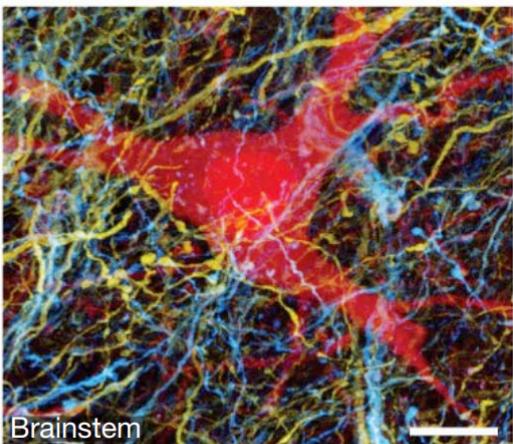
ARTICLE

Transgenic strategies for combinatorial expression of fluorescent proteins in the nervous system

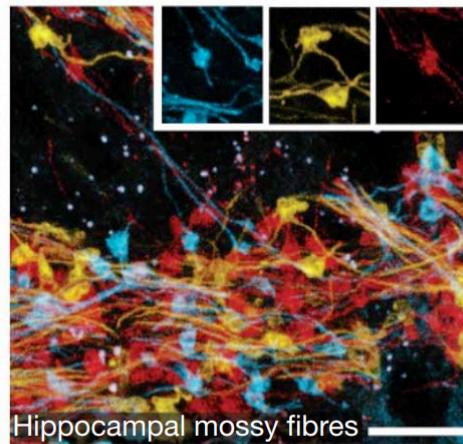
Jean Livet¹, Tammy A. Weissman¹, Hyuno Kang¹, Ryan W. Draft¹, Ju Lu¹, Robyn A. Bennis¹, Joshua R. Sanes¹ & Jeff W. Lichtman¹

Detailed analysis of neuronal network architecture requires the development of new methods. Here we present strategies to visualize synaptic circuits by genetically labelling neurons with multiple, distinct colours. In *Brainbow* transgenes, Cre/lox recombination is used to create a stochastic choice of expression between three or more fluorescent proteins (XFPs). Integration of tandem *Brainbow* copies in transgenic mice yielded combinatorial XFP expression, and thus many colours, thereby providing a way to distinguish adjacent neurons and visualize other cellular interactions. As a demonstration, we reconstructed hundreds of neighbouring axons and multiple synaptic contacts in one small volume of a cerebellar lobe exhibiting approximately 90 colours. The expression in some lines also allowed us to map glial territories and follow glial cells and neurons over time *in vivo*. The ability of the *Brainbow* system to label uniquely many individual cells within a population may facilitate the analysis of neuronal circuitry on a large scale.

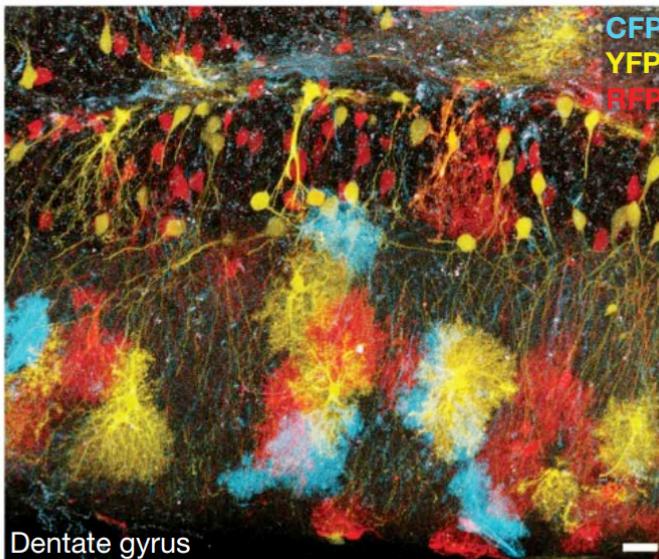
a Brainbow-1.0



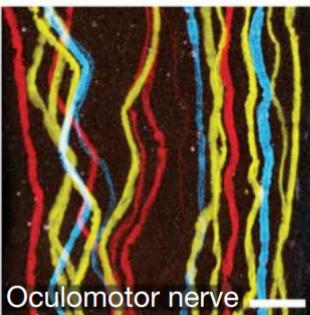
b Brainbow-1.1



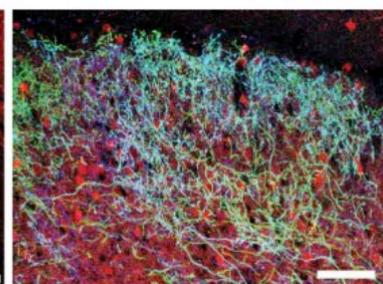
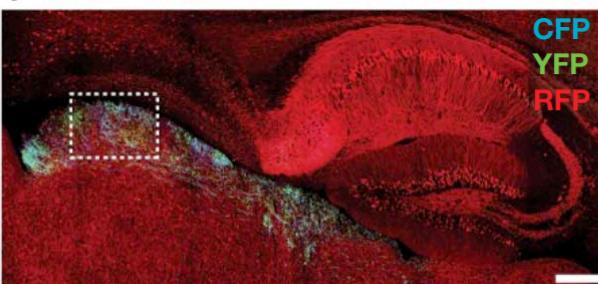
c Brainbow-2.0

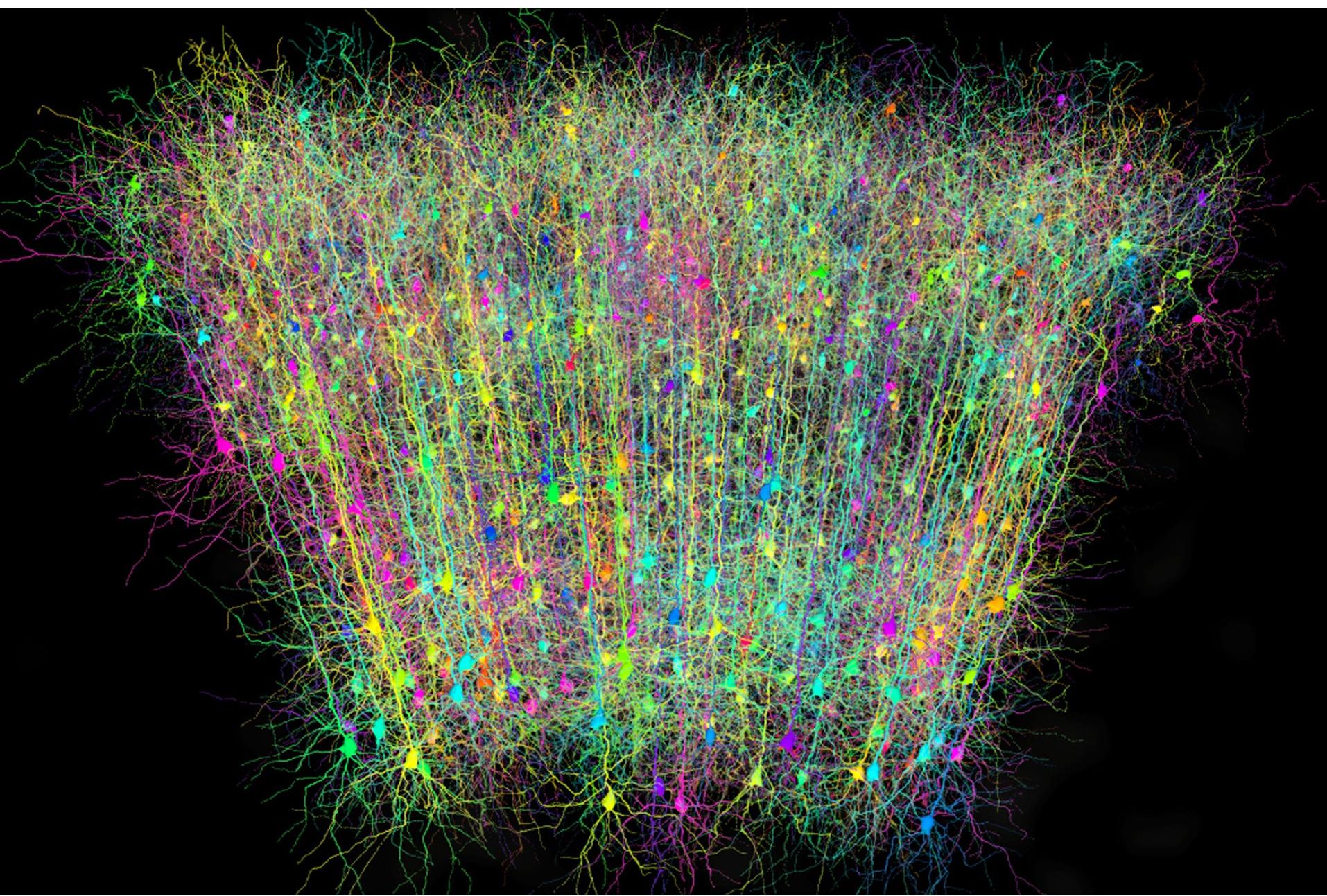


d Brainbow-2.1



e Restriction of recombination





**Record function and describe structure
together**

Network anatomy and *in vivo* physiology of visual cortical neurons

Davi D. Bock^{1,2}, Wei-Chung Allen Lee^{1,2}, Aaron M. Kerlin¹, Mark L. Andermann¹, Greg Hood³, Arthur W. Wetzel³, Sergey Yurgenson¹, Edward R. Soucy², Hyon Suk Kim^{1,2} & R. Clay Reid^{1,2}

In the cerebral cortex, local circuits consist of tens of thousands of neurons, each of which makes thousands of synaptic connections. Perhaps the biggest impediment to understanding these networks is that we have no wiring diagrams of their interconnections. Even if we had a partial or complete wiring diagram, however, understanding the network would also require information about each neuron's function. Here we show that the relationship between structure and function can be studied in the cortex with a combination of *in vivo* physiology and network anatomy. We used two-photon calcium imaging to characterize a functional property—the preferred stimulus orientation—of a group of neurons in the mouse primary visual cortex. Large-scale electron microscopy of serial thin sections was then used to trace a portion of these neurons' local network. Consistent with a prediction from recent physiological experiments, inhibitory interneurons received convergent anatomical input from nearby excitatory neurons with a broad range of preferred orientations, although weak biases could not be rejected.

Wiring specificity in the direction-selectivity circuit of the retina

Kevin L. Briggman¹, Moritz Helmstaedter¹ & Winfried Denk¹

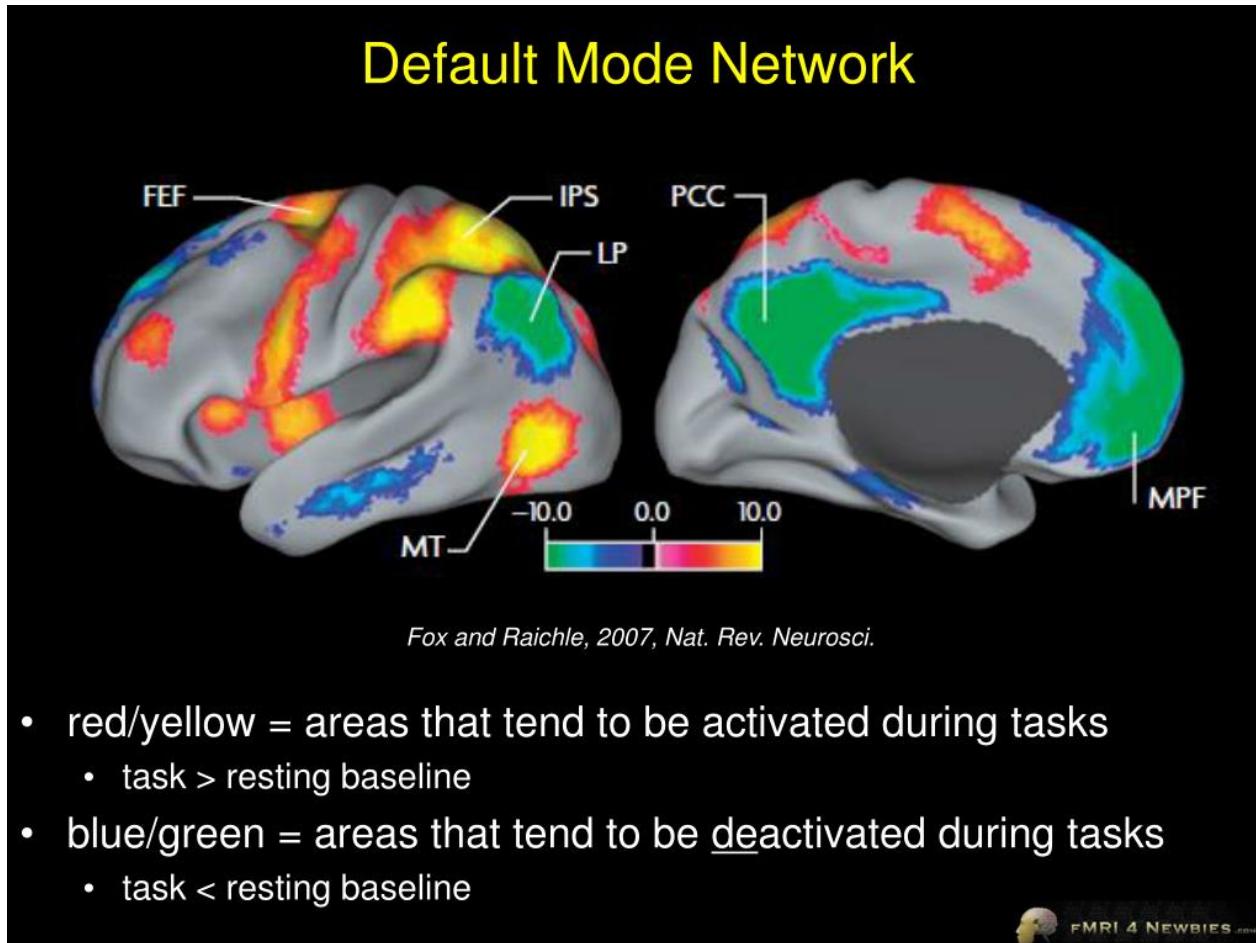
The proper connectivity between neurons is essential for the implementation of the algorithms used in neural computations, such as the detection of directed motion by the retina. The analysis of neuronal connectivity is possible with electron microscopy, but technological limitations have impeded the acquisition of high-resolution data on a large enough scale. Here we show, using serial block-face electron microscopy and two-photon calcium imaging, that the dendrites of mouse starburst amacrine cells make highly specific synapses with direction-selective ganglion cells depending on the ganglion cell's preferred direction. Our findings indicate that a structural (wiring) asymmetry contributes to the computation of direction selectivity. The nature of this asymmetry supports some models of direction selectivity and rules out others. It also puts constraints on the developmental mechanisms behind the formation of synaptic connections. Our study demonstrates how otherwise intractable neurobiological questions can be addressed by combining functional imaging with the analysis of neuronal connectivity using large-scale electron microscopy.

Diffusion-weighted imaging

A



Default mode network



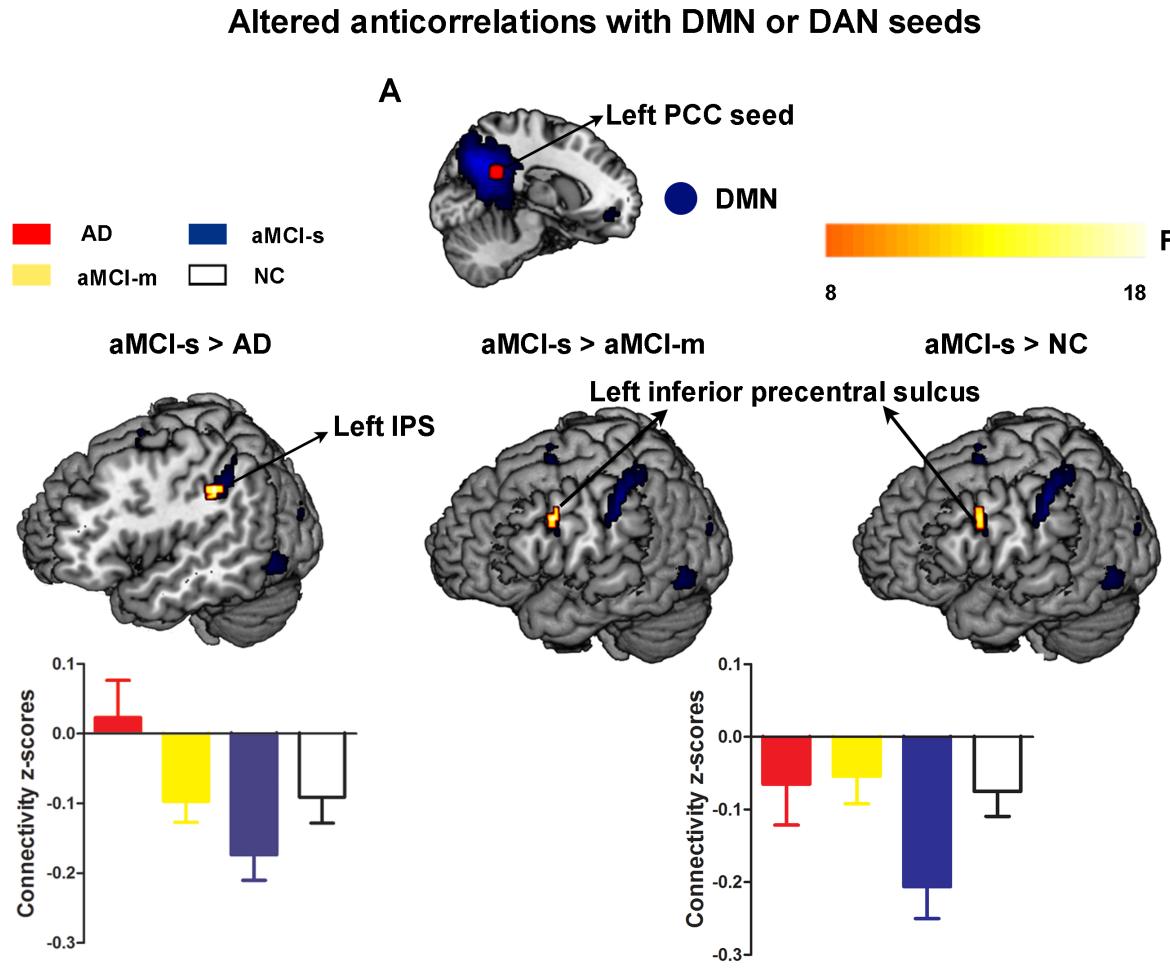
- red/yellow = areas that tend to be activated during tasks
 - task > resting baseline
- blue/green = areas that tend to be deactivated during tasks
 - task < resting baseline

Functional connectome fingerprinting: identifying individuals using patterns of brain connectivity

Emily S Finn^{1,7}, Xilin Shen^{2,7}, Dustin Scheinost², Monica D Rosenberg³, Jessica Huang², Marvin M Chun^{1,3,4}, Xenophon Papademetris^{2,5} & R Todd Constable^{1,2,6}

Functional magnetic resonance imaging (fMRI) studies typically collapse data from many subjects, but brain functional organization varies between individuals. Here we establish that this individual variability is both robust and reliable, using data from the Human Connectome Project to demonstrate that functional connectivity profiles act as a ‘fingerprint’ that can accurately identify subjects from a large group. Identification was successful across scan sessions and even between task and rest conditions, indicating that an individual’s connectivity profile is intrinsic, and can be used to distinguish that individual regardless of how the brain is engaged during imaging. Characteristic connectivity patterns were distributed throughout the brain, but the frontoparietal network emerged as most distinctive. Furthermore, we show that connectivity profiles predict levels of fluid intelligence: the same networks that were most discriminating of individuals were also most predictive of cognitive behavior. Results indicate the potential to draw inferences about single subjects on the basis of functional connectivity fMRI.

Dysfunctional interactions between the default mode network and dorsal attention network in subtypes of amnestic mild cognitive impairment



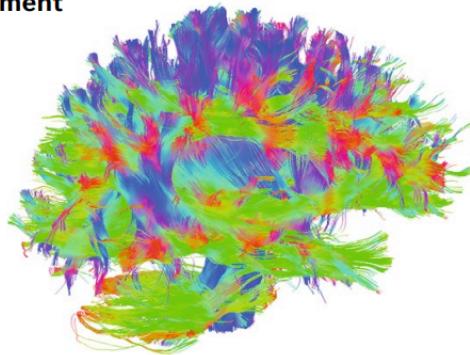
The physics of brain network structure, function and control

Christopher W. Lynn¹ and Danielle S. Bassett^{1,2,3,4,5*}

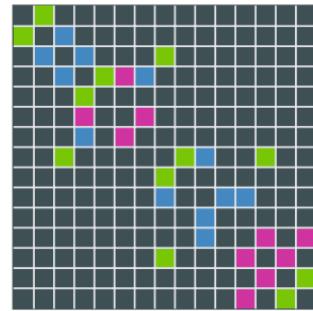
Abstract | The brain is characterized by heterogeneous patterns of structural connections supporting unparalleled feats of cognition and a wide range of behaviours. New non-invasive imaging techniques now allow comprehensive mapping of these patterns. However, a fundamental challenge remains to understand how the brain's structural wiring supports cognitive processes, with major implications for personalized mental health treatments. Here, we review recent efforts to meet this challenge, drawing on physics intuitions, models and theories, spanning the domains of statistical mechanics, information theory, dynamical systems and control. We first describe the organizing principles of brain network architecture instantiated in structural wiring under constraints of spatial embedding and energy minimization. We then survey models of brain network function that stipulate how neural activity propagates along structural connections. Finally, we discuss perturbative experiments and models for brain network control; these use the physics of signal transmission along structural connections to infer intrinsic control processes that support goal-directed behaviour and to inform stimulation-based therapies for neurological and psychiatric disease. Throughout, we highlight open questions that invite the creative efforts of pioneering physicists.

Measuring and modelling brain network structure

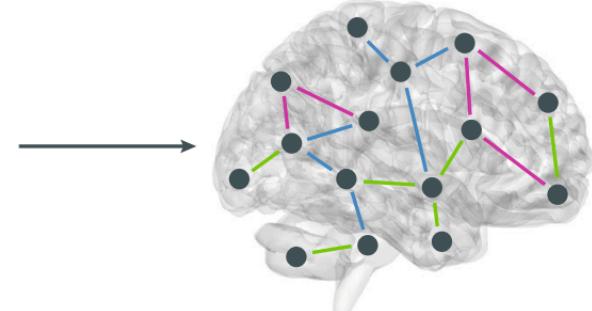
a Measurement



Example: white matter tracts (via diffusion tensor imaging)



Adjacency matrix

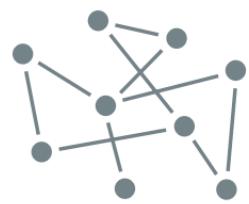


Structural brain network

b Modelling

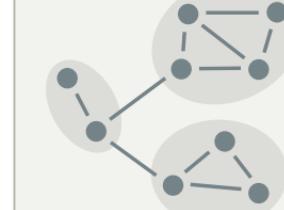
Network type

Random
(no structure)



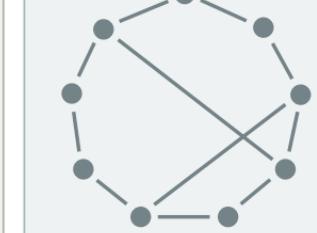
Erdős–Rényi

Community structure



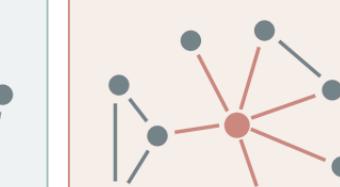
Stochastic block

Small-world
(efficient communication)



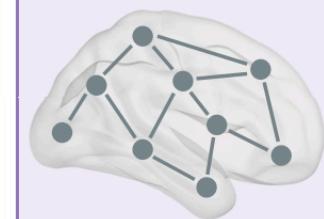
Watts–Strogatz

Hub structure
(heavy-tailed degree distribution)



Barabási–Albert

Spatially embedded

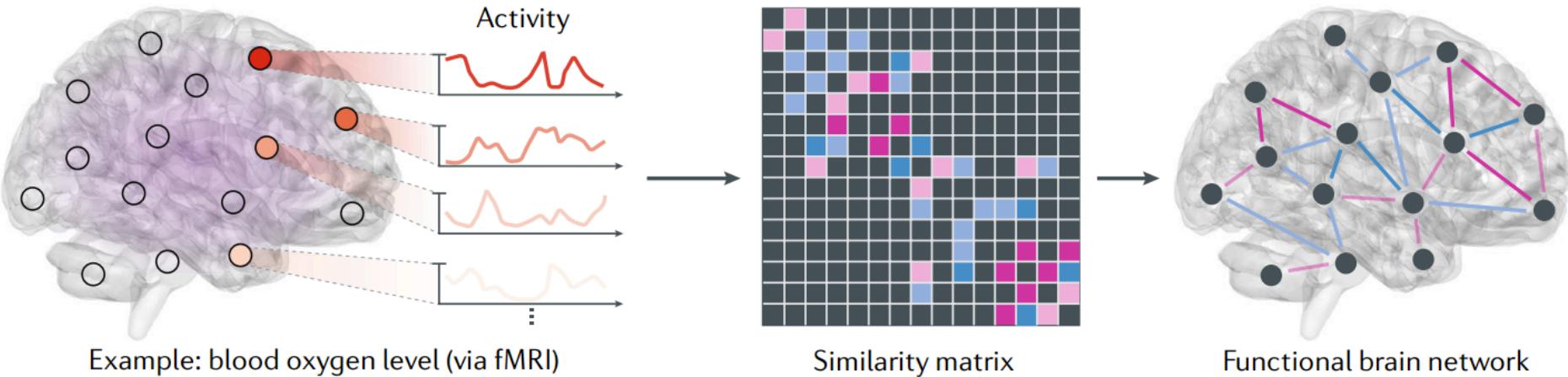


Spatial model

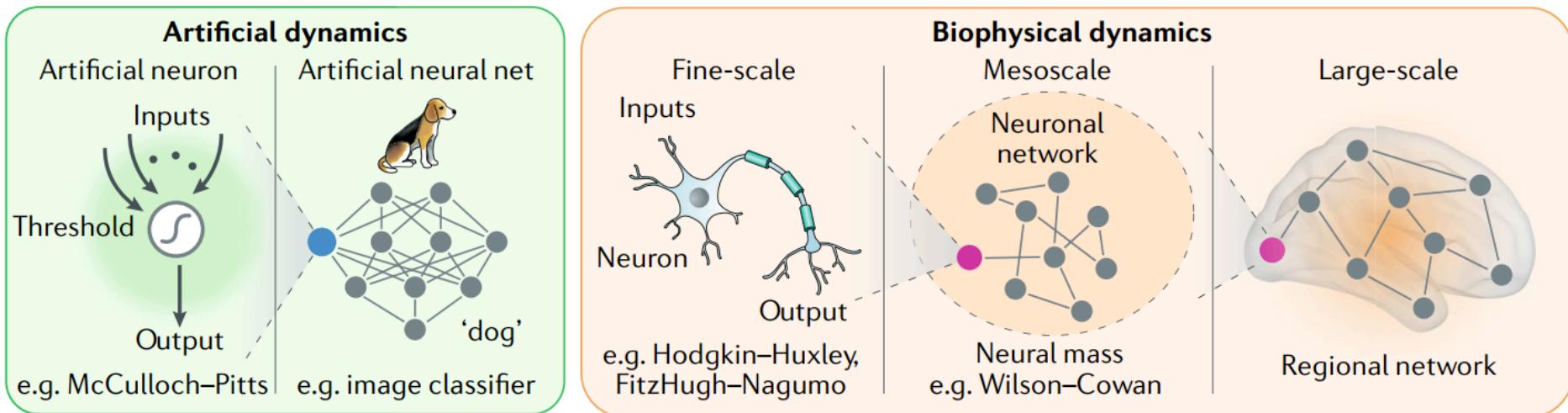
Generative model

Measuring and modelling brain network function

a Measurement



b Modelling



Thank you!