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Connectomics and other novel methods for examining neural systems☆

*Richard J. Wurtman**

Department of Brain and Cognitive Sciences, Massachusetts Institute of Technology, Cambridge, MA 02139, USA

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ABSTRACT

Novel approaches for studying the brain and relating its activities to mental phenomena have come into use during the past decade (Bargmann, 2015). These include both new laboratory methods – involving, among others, generation of isolated cells which retain neuronal characteristics in vivo; the selective stimulation of neurons by light in vivo; and direct electrical stimulation of specific brain regions to restore a system's balance of excitation and inhibition – and a new organizing principle, "connectomics", which recognizes that networks, and not simply a key nucleus or region, underlie most brain functions and malfunctions. Its application has already improved our comprehension of how the brain normally functions and our ability to help patients with such poorly treated neurologic and psychiatric diseases as Alzheimer's disease.

1. Introduction

Perhaps the most important recent advance in understanding the brain's operations has been the recognition that the brain units, which process most of the information that it receives and which generate most of its outputs, are not single nuclei, or even regions. Rather, they are specific multisynaptic circuits, or networks, involving multiple regions, nuclei, and tracts. Moreover, a single such component can also participate in additional, functionally distinct circuits. It is now possible to map these networks using sophisticated "connectomics" tools, described below, which integrate neuroanatomic, electroencephalographic, and imaging data, and major resources are becoming available to support work in this field, e.g. a large-scale NIH commitment, the Human Connectome Project [1]. Some such networks under vigorous exploration include the "default network" – which integrates brain function in the absence of major sensory inputs [2] – and the circuits that underlie decision-making, circadian rhythms, learning and memory, motor coordination, and emotional responses.

Important new laboratory technologies have also improved our ability to measure and to modify brain operations [3]. These include, among others:

Methods for producing, from fibroblasts, specific types of neurons which retain characteristic properties after implantation into brains;

Methods for conducting complex genetic analyses capable of implicating multiple genes (and the biochemical processes their protein products mediate) in the etiologies of neurobehavioral diseases (e.g., excessive synaptic pruning in schizophrenia and Alzheimer's disease [4,5]);

Optogenetic methods for selectively activating particular neuronal populations in heterogeneous fields, by first tagging neurons with a photosensitive pigment and then shining laser light on them [6,7];

A method (DBS; deep brain stimulation) for restoring balance within a brain system that malfunctions because one component is disturbed, by targeted stimulation of a different, counterbalancing component (for example, in patients with Parkinson's disease, reducing the movement-inhibiting output

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* Tel.: +1 617 686 8181.
E-mail address: dick@mit.edu.

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from the subthalamic nuclei to compensate for the loss of movement-promoting dopaminergic neurons [8]).

1.1. *Connectomics*

The chief goals of the new field of "connectomics" [9,10] have been described as illumination of the structural and functional connectivity patterns of human brain regions, and, especially, of the interactions among the components of neural networks. A full understanding of these patterns and their interactions will surely require detailed knowledge of the types of neurons present in each component. Difficulties abound in accumulating this knowledge; however, new technologies discussed below are helping.

Connectomics utilizes an array of techniques for providing data on the components of neuronal networks in living subjects and the interactions of these components. These include, principally, imaging studies (e.g., structural MRI; high angle resolution diffusion imaging; functional connectivity MRI [fcMRI]) and neurophysiologic measurements (EEG and MED). Data thus obtained are subjected to sophisticated mathematical analyses, based on graph theory and basal network analysis [11,12].

Graph theory is the branch of mathematics that studies the properties of graphs, which are mathematical structures that model the relations between pairs of objects, for example in a *network*. The objects themselves are sometimes physical, or biological, or social, or even units of information. A *graph* is made up of *vertices*, *nodes*, or *points* which are connected by *edges*, *arcs*, or *lines*; the former are usually indicated by dots or circles, two of which are connected by a line.

For example, a *graph* representing a human brain might show N nodes, each represented by a circle that indicates a region or a nucleus, and some of the pairs of *nodes* would be connected by lines, representing tracts. Each such tract might contain a characteristic number of axons, generating many synapses. A *node* called the substantia nigra would generate an *edge* or *line* called the nigrostriatal tract, and axons in that tract would synapse on the neurons of another *node*, the corpus striatum. The strength of communications between the transmitting and receiving *nodes* (as calculated using electroencephalographic measurements) might be strong or weak depending in part on the number of axons in the nigrostriatal tract and the total number of synapses they generate. Graph theory might be used to quantify the strength of communications in a disease, e.g. Alzheimer's disease, and to follow its changes as synapses are lost or possibly regained after an effective treatment.

The connectomics approach postulates that many if not most neurobehavioral syndromes are the consequence of neurological abnormalities that are highly distributed, in networks and systems, and not resident within single populations of neurons. Hence, "… studying brain parts in isolation will be insufficient to account for brain alterations associated with mental disorders [9]". At a micro-level, connectomics also, as discussed below, seeks to identify the populations of postsynaptic neurons with which presynaptic terminals in a brain region make functional contacts. Utilized for this purpose are specialized modifications of electron- and light- microscopic techniques; single-cell recordings; novel chemical tracing methods [13]; and neurophysiologic studies, e.g., based on optogenetics, [7].

Two major unsolved problems have motivated the rise of connectomics: the first is the disparity between our assumption that a given neuron may form synapses with perhaps a thousand or more postsynaptic neurons and the reality that only a tiny minority of such contacts are usually identifiable using available methods. The second is that the perception, described above, that attempts to treat complex cognitive disturbances – like those in schizophrenia or Alzheimer's disease – by targeting only a single anatomically or chemically defined neuronal population has not worked as well as it might, perhaps because the disease process involves networks that can encompass multiple brain structures, cell types, and neurotransmitters.

1.2. *Complete Identification of the Neurons and Synapses in a Brain Region*

When an axon from what will become a potential neuron enters a brain microregion, will it form synapses with the spines on most nearby dendrites? Or only on those of a relatively few neurons which it chemically targets? If it initially makes synapses with most nearby neurons and if most of these survive the pruning process, how might we distinguish among the various populations of the postsynaptic neurons so that we can relate them to the region's functional outputs? The prevailing theory of synapse formation, "Peters' Rule" [14] – which was based on data obtained by counting "potential synapses", i.e. proximities between axons and dendrites that were small enough to be bridged by a spine [15] – has held that "…once afferents reach their specific destinations they seek to synapse with all of the postsynaptic elements that are capable of forming synaptic junctions with them." But recent connectomics-based studies using the newer technique of manual and automated serial section transmission electron microscopy (ssTEM) have demonstrated that the proportion of axodendritic "touches" in rat hippocampus which ultimately becomes synapses is more like 20%, – still a very large number to identify using, for example, recordings of action potentials or the newer optogenetic methods [6,7,16]. Until the search process can be effectively automated – as was critically important in the Human Genome Project – its success must surely require the participation of very many laboratories.

How many distinct cortical regions with clear boundaries – included, with brain nuclei, among the *nodes*, in connectomics terminology – exist in human brain? A recently published study [17], based in part on MRI data from 210 healthy young adults, suggests that there are at least 180 such regions, each of which may exhibit characteristic differences in cortical thickness; functions; connectivity patterns; levels of myelin; and the topographic organization of the neurons. The sizes of particular regions apparently vary from person to person, raising the possibility that functions mediated by a given region might exhibit parallel variations.

1.3. *Abnormalities of Neural Networks Shown by Connectomic Studies: Alzheimer's Disease*

The quantitative analysis of EEG data, using newer analytic techniques including graph theory [11,12], demonstrates characteristic abnormalities in networks within brains of

people suffering from Alzheimer's disease – for example, a slowing in background EEG activity, which correlates with decreased performance in memory tasks [12,17–19]. Changes are also observed in functional connectivities, as illustrated by the coupling or statistical interdependence of the EEG patterns of paired brain regions. In normal brains, adjacent loci from which EEG data are collected tend to exhibit interconnectedness and a tendency toward parallel variations, and signals may tend to travel through the brain along either a relatively long, tract-mediated path ["global integration"] [19] or a shorter path ["small-world configuration" of local-circuit clustering] [20]. Both types of pathways may be impaired in Alzheimer's disease. Since interconnectedness and path length can now be quantified, as discussed above, their assessment allows examination of possible effects of putative AD treatments. For example, brain networks were found to be preserved when patients with early AD were given a treatment thought to increase synaptogenesis [19]. Presumably, this decreased the "disconnectedness" of diseased brain regions by increasing the ability of surviving axons to transmit information to postsynaptic neurons.

2. Conclusions

Deciphering the vast numbers of neurons, the much greater numbers of synapses, and their connection patterns, in networks, within the functioning human brain will surely occupy neuroscientists and neurologists for many years to come. Each major improvement in knowledge has the potential to yield major benefits to patients with complex neurobehavioral disorders which involve multiple brain nuclei and tracts and which presently are poorly treated. Such advances continue a tradition established more than a century ago with the articulation of the "neuron doctrine" and typified by the rational development in the 20th century of therapeutic agents that act at specific synapses to promote or suppress neurotransmission.

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Conflict of Interest

None.

REFERENCES

[1] Van Essen DC, Smith SM, Barch DM, Behrens TE, Yacoub E, Ugurbil K, et al. The WU-Minn Human Connectome Project: an overview. NeuroImage 2013;80:62–79.
[2] Buckner RL, Andrews-Hanna JR. The brain's default network: anatomy, function, and relevance to disease. Ann N Y Acad Sci 2008;1124:1–37.
[3] Bargmann CI. How the new neuroscience will advance medicine. JAMA 2015;314:221–2.
[4] Fornito A, Zalesky A, Pantelis C, Bullmore ET. Schizophrenia, neuroimaging, and connectomics. NeuroImage 2011;62:2296–314.
[5] Fornito A, Zalesky A, Breakspear M. The connectomics of brain disorders. Nat Rev Neurosci 2015;16:159–72.
[6] Cardin JA, Carlén M, Meletis K, Knoblich U, Zhang F, Deisseroth K, et al. Targeted optogenetic stimulation and recording of neurons in vivo using cell-type-specific expression of Channelrhodopsin. Nat Protoc 2010;5:247–54.
[7] Deisseroth K. Circuit dynamics of adaptive and maladaptive behavior. Nature 2014:309–17.
[8] DeLong MR, Benabid AL. Discovery of high-frequency deep brain stimulation for treatment of Parkinson disease: 2014 Lasker Award. JAMA 2014;312:1093–4.
[9] Sporns O. Networks of the brain. 1st ed. Cambridge MA: MIT Press; 2011.
[10] Hagmann P. From diffusion MRI to brain connectomics. Lausanne: Signal Processing Institute. Ecole Polytechnique Federale de Lausanne (EPFL); 2005 127.
[11] He Y, Evans A. Graph theoretical modeling of brain connectivity. Curr Opin Neurol 2010;23:341–50.
[12] Xie T, He Y. Mapping the Alzheimer's brain with connectomics. Front Psychiatry 2012. http://dx.doi.org/10.3389/fpsyt.2011.00077.
[13] Hagmann P, Cammoun L, Gigandet X, Gerhard S, Grant PE, Wedeen V, et al. MR connectomics: principles and challenges. J Neurosci Methods 2010;194:34–45.
[14] Peters A, Feldman ML. The projection of the lateral geniculate nucleus to area 17 of the rat cerebral cortex. 1. General description. J Neurocytol 1976;5:63–84.
[15] Mischenko Y, Hu T, Spacek J, Mendenhall J, Harris KM, Chklovskii DB. Ultrastructural analysis of hippocampal neuropil from the connectomics perspective. Neuron 2010;67:1009–20.
[16] Alivasatos AP, Chun M, Church GM, Greenspan RJ, Roukes ML, Yuste R. The brain activity map project and the challenge of functional connectomics. Neuron 2012;74:970–4.
[17] Glasser MF, Coalson TS, Robinson EC, Hacker CD, Harwell J, Yacoub E, et al. A multi-modal parcellation of human cerebral cortex. Nature 2016. http://dx.doi.org/10.1038/nature 18933.
[18] Jeong J. EEG dynamics in patients with Alzheimer's disease. Clin Neurophysiol 2004;115:1490–505.
[19] Fornito A, Bullmore ET. Connectomics: a new paradigm for understanding brain disease. Eur Neuropsychopharmacol 2014. http://dx.doi.org/10.1016/j.euroneuro.2014.02.011.
[20] de Waal H, Stam CJ, Lansbergen MM, Wieggers RL, Kamphuis PJ, Scheltens P, et al. The effect of souvenaid on functional brain network organization in patients with mild Alzheimer's disease: a randomized controlled study. PLoS One 2014;9:e86558.