NEUROSCIENCE

The Neuron Doctrine, Redux

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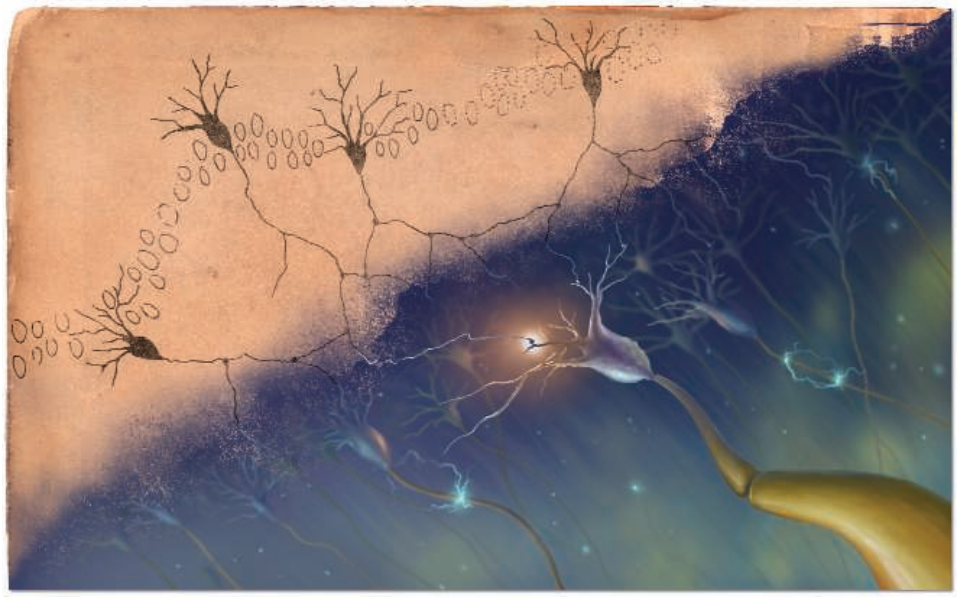
After a century, neuroscientists are rethinking the Neuron Doctrine, the fundamental principle of neuroscience. This proposition, developed primarily by the great Spanish anatomist and Nobel laureate Santiago Ramón y Cajal, holds that a neuron is an anatomically and functionally distinct cellular unit that arises through differentiation of a precursor neuroblast cell. In principle, part of this tenet has held up, but technology and research have extended our knowledge far beyond this simple description. What has evolved is a modern view of the neuron that allows a more broad and intricate perspective of how information is processed in the nervous system. One hundred years since its inception, an examination of the Doctrine indicates that it no longer encompasses important aspects of neuron function. If we are to understand complex, higher level neuronal processes, such as brain function, we need to explore beyond the limits of the Neuron Doctrine.

In the early 20th century, the nervous system was thought to function as a web of interconnected nerve fibers. The cytoplasm and nervous impulses were thought to flow freely in any direction through the network of fibers. But it was Cajal who envisioned the neuron as an individual functional unit, polarized such that signals are received through its rootlike dendrites and transmitted through its long axonal process. He posited that although an axon terminates adjacent to a dendrite of the next neuron (see the figure), the cleft between them would act as a synaptic switch regulating information flow through neural circuits. The synaptic cleft went unseen until a half-century later, when in 1954 the electron microscope provided convincing evidence that essentially refuted the earlier "reticular" view of a nerve fiber web (*1*).



Information processing, past and present. The Neuron Doctrine transformed the 19th-century view of the nervous system which saw the brain as a network of interconnected nerve fibers (**upper left**). A century later, the modern view (**lower right**) holds the neuron as a discrete cell that processes information in more ways than original envisaged: Intercellular communication by gap junctions, slow electrical potentials, action potentials initiated in dendrites, neuromodulatory effects, extrasynaptic release of neurotransmitters, and information flow between neurons and glia all contribute to information processing.

At the same time, physiological studies established that conduction of electrical activity along the neuronal axon involved brief, all-or-nothing, propagated changes in membrane potential called action potentials. It was thus often assumed that neuronal activity was correspondingly all-or-nothing and that action potentials spread over all parts of a neuron. The neuron was regarded as a single functional unit: It either was active and "firing" or was not.

This dogma began to erode with the advent of microelectrodes that could be inserted into neurons to record electrical signals. In 1959, it was realized that much of the information processing by neurons involves electrical events that are graded in amplitude and decay over distance, rather than all-or-nothing electrical spikes that propagate regeneratively (*2*). It was also determined that evoked electrical responses often occur on a background of spontaneous changes in membrane potential (i.e., produced without input from other neurons) and that some parts of the neuron are incapable of producing all-or-nothing action potentials (*3*). Today, it is apparent that information processing in the nervous system must operate beyond the limits of the Neuron Doctrine as it was conceived. This has evolved from detailed information gained from techniques developed in the past 50 years—notably single-channel recording, live cell imaging, and molecular biology.

Although Cajal wisely considered that "neuronal discontinuity… could sustain some exceptions" to the Doctrine's definition (*4*), he could not have foreseen the presence and role of neuronal gap junctions as one of these exceptions. These assemblages of protein pores form small aqueous channels of limited selectivity that connect neurons, providing cytoplasmic continuity (*5*). We now know that gap junctions are widespread in the mammalian nervous system (*5*) and function to synchronize neuronal firing. They constitute electrical synapses that couple groups of cells into functional syncytia—in this

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sense, the reticular concept, reinvoked. Electrical transmission through gap junctions was initially considered primitive and likely incapable of the subtleties of chemical transmission through axon-dendrite synapses (early studies showed that synapses with gap junctions between the axon of one neuron and the cell soma of another neuron also have regions resembling the active zones of chemical synapses, although there is no chemically mediated signal transmission and response). Although gap junctions can behave as simple electrical resistances between connected cells, an electrical impulse in one cell by no means inevitably propagates to the other cells with which it shares gap junctions. In fact, a channel within a gap junction is not necessarily open, and an entire gap junction may not transmit electrical current until it is appropriately modified in response to transmission from chemical synapses of the same, "presynaptic" neuron. This modulation of channels provides electrical synapses at gap junctions with the plasticity long considered an exclusive province of chemical synapses at axon-dendrite junctions (*6*). Furthermore, gap junctions have been described between neurons and non-neuronal cells such as astrocytes (*7*), a somewhat controversial finding not conceived in the original Neuron Doctrine.

Fifty years ago, neuroscientists also did not realize that a plethora of neuromodulatory substances, such as amines and neuropeptides, can reconfigure neuronal circuits into different patterns of functional connection, capable of a variety of activity patterns (*8*). Almost all neurons and synapses are subject to such neuromodulation, which acts to remodel neuron behavior and circuitry within minutes and hours rather than on the millisecond time scale typical of electrical impulse transmission. Many behaviors, including learning and memory, sexual cycles, mood, and sleep, occur over much slower time scales relative to processes such as reflexes or sensory and motor function. In addition, neuromodulatory substances can act at multiple sites on the neuron, including the axon. For example, some crab (*9*) and lobster (*10*) axons have receptors to amines such as dopamine, serotonin, and octopamine. When these amines are applied to the axons, these areas can spontaneously initiate action potentials in a nonclassical mode of integration.

Research during the past 10 years has shown that in many neurons, action potentials can travel backward from the axon and soma regions into the dendrites (*11*). Moreover, under certain conditions action potentials can be initiated in dendrites, remaining local or sometimes propagating into the soma to initiate single or multiple spikes of activity in the axon (*12*). The functional complexity of dendrites and the roles they play in synaptic integration and plasticity are well beyond what could have been deduced from Cajal's anatomy or from later somatic recordings (*2*). Dendrites contain a mosaic of voltage-gated ion channels (*13*). The types, densities, and properties of these channels are very diverse among classes of neurons (and even within a single class), and these channels regulate, on wide-ranging time scales, how a neuron responds to the thousands of incoming synaptic events that impinge on its dendrites. Important questions for the future will be how the spatial distributions of individual ion channels in dendrites are established, how this localization changes in response to incoming synaptic inputs and output firing patterns (*14*), and how the channels dynamically regulate excitability during different behavioral states.

Cajal was also careful to distinguish neurons from the many other cells in nervous tissue. The function, origin, and diversity of non-neuronal cells eluded Cajal, because a staining method, which revealed neuronal structure with brilliant clarity, left major classes of non-neuronal cells invisible (including microglia and oligodendrocytes). It is ironic that today we understand that the fundamental tenet of the Neuron Doctrine—polarized communication between neurons by action potentials—is heavily influenced by non-neuronal cells. These are the constituents of the nervous system that form the myelin sheath around axons and organize ion channels into periodic clusters along the axon, features that facilitate action potential propagation (*15*).

Myelinating glia do not fire action potentials, but they can detect impulses in axons through membrane receptors that bind signaling molecules. These include ATP (*16*) and adenosine (*17*) that are released along the axon and also potassium that is released during intense neural activity. This axon-glial communication violates the Neuron Doctrine in two ways. Information is communicated between cells at sites far removed from chemical synapses, and it propagates in a transduced form through cells that are not neurons (*18*). In response to neural firing, glia communicate with other glia by chemical signaling and gap junctions rather than by electrical impulses (*18*). Unexpectedly, chemical synapses have recently been detected between neurons and a class of glia (oligodendrocyte precursor cells) (*19*), undermining a defining feature of neurons. However, the functional importance of this neuron-glia interaction is unknown. We now know that during vertebrate embryonic development, glia can give birth to neurons (*20*), challenging Cajal's conclusion that neurons develop only from neuroblasts.

Astrocytes are now known to communicate among themselves by means of glial transmitters and neuromodulators as well as by gap junctions (*18*). Moreover, astrocytes can detect neurotransmitters that are released from neuronal chemical synapses (*21*). These transmitters are delivered via synaptic vesicles into the synaptic cleft and diffuse to perisynaptic astrocytes. Additionally, neurotransmitters can be released outside the synapse and detected by perisynaptic glia (*22*, *23*). In response, astrocytes can regulate communication between neurons by modifying synaptic transmission through the release of neurotransmitters and neuromodulators (*18*). Thus, there may be a parallel system of information processing that interacts with neuronal communication but propagates over much slower time scales through a functionally reticular network of non-neuronal cells. This functional reticulum results from gap junction coupling and the omnidirectional communication that is mediated by chemical messengers released from astrocytes over much slower time scales. Such may be the case in the human brain.

Obviously, although neurons are indeed anatomically discrete units, they are not the single functional units in the sense envisioned by early proponents of the Neuron Doctrine. And the simplistic and static connectivity patterns described by Cajal and other cellular neuroanatomists must be revised in light of new information. The differences in specific membrane and cellular properties among cell bodies, axons, and dendrites, and even between different areas along dendrites, are far more extensive and sophisticated than would have been imagined nearly 50 years ago. Absolutely unforeseen a century ago is the active participation of non-neuronal constituents of the nervous system. A Neuron Doctrine reexamined hence provides a renewed perspective to ask many intriguing questions, particularly those about the human brain. For example, what features of the human brain account for our level of behavioral complexity? It is doubtful that the answer emerges from knowing the sheer number of cells, or the properties of synapses, or the identity of neurotransmitters and modulators. Such features are shared by many animals, especially vertebrates. There are, however, fundamental differences in electroencephalograms across the evolutionary spectrum—that is, in the electric field potentials arising from assemblies of functioning neurons. This suggests that the complexity of the human brain and likely other regions of the nervous system derive from some organizational features that make use of the permutations of scores of

integrative variables and thousands or millions of connectivity variables (*24*) and perhaps integrative emergents yet to be discovered. The answers extend well beyond explanation by the neuron acting as a single functional unit.

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STRUCTURAL BIOLOGY

A Ribosomal Coup: *E. coli* at Last!

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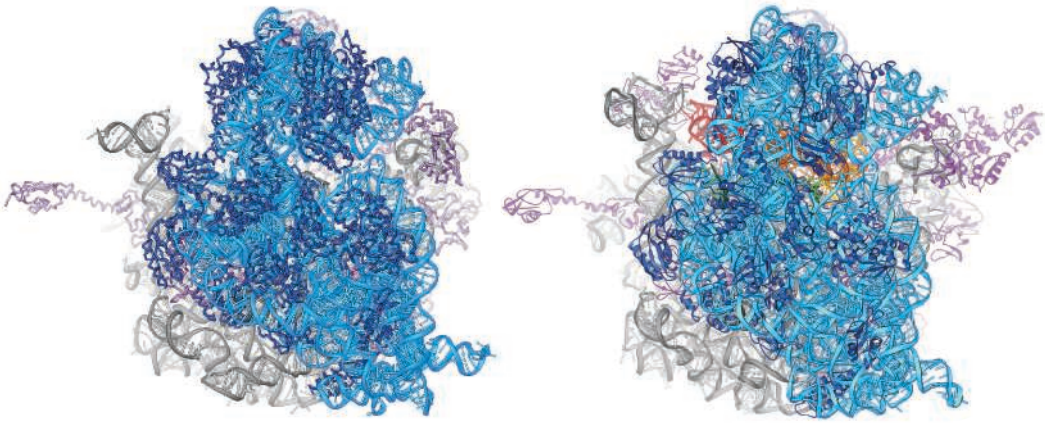
On page 827 in this week's issue, Schuwirth *et al.* (*1*) report an atomic resolution (3.5 Å) crystal structure for the 70*S* ribosome from the bacterium *Escherichia coli* (see the figure). More accurately, they report the atomic resolution for two such structures, because there are two, nonequivalent copies of the 70*S* ribosome per asymmetric unit in the crystals they have analyzed. The ribosome is the ribonucleoprotein enzyme that catalyzes messenger RNA–directed protein synthesis in all organisms, and the 70*S* ribosome, which is a 1:1 complex of a large and a small ribosomal subunit, is the particle that synthesizes proteins in prokaryotes. Because this enzyme plays a central role in gene expression, its structure has long been sought by molecular biologists.

The structures reported by Schuwirth *et al.* are by no means the first ribosomal crystal structures to appear. We already have a 2.4 Å resolution crystal structure for the large ribosomal subunit from *Haloarcula marismortui* (*2*), and a 3.1 Å resolution structure for the large ribosomal subunit from *Deinococcus radiodurans* (*3*). Two versions of the structure of the small ribosomal subunit from *Thermus thermophilus* have appeared, one at a resolution of 3.0 Å (*4*), and the other at a slightly lower resolution (*5, 6*). In addition, there is a structure for the 70*S* ribosome from *T. thermophilus* determined at 5.5 Å (*7*). Our sense of *déjà vu* is heightened by the impression that these new structures look very much like those that have appeared before (see the figure). Thus, we might wonder why these new structures should be considered noteworthy (which they are).

There are three reasons why these structures deserve attention. First, the structures that Schuwirth and colleagues have solved are that of the ribosome from *E. coli*. Since 1960, the *E. coli* ribosome has been the ribosome of choice for biochemists and molecular biologists; for no other ribosome is the information more complete. Observations made with the *E. coli* ribosome have been extensively used to interpret all the ribosome structures published previously, all of which came from other organisms. The argument has been that the extensive sequence homology that exists between ribosomes from different species justifies such cross-species comparisons. However, at some level, observations made on ribosomes from a mesophilic eubacterium like *E. coli* cannot be valid for ribosomes obtained from an extreme archaeal halophile like *H. marismortui*, or from an extreme eubacterial thermophile like *T. thermophilus.* These concerns can now be directly addressed.

Second, Schuwirth *et al.* are not the first investigators to attempt the crystallization of ribosomes from *E. coli*. For decades, laboratories all over the world have tried to obtain such crystals because of the obvious importance of the structures that might emerge from them. Schuwirth *et al.* are the first to obtain ribosomal crystals from this species that were worth analyzing, and that in itself is a coup. It should also be noted that the asymmetric unit of the crystals they have solved is gigantic; it contains roughly 5 megadaltons of macromolecular material. Determining structures this large is not trivial, even when much is known about them already, as was the case here.

Third, there is the matter of resolution. The resolution of the best 70*S* structure pub-



Structures of the 70*S* ribosome from two prokaryotes. (**Left**) *E. coli.* ribosome at 3.5 Å resolution [from (*1*)]. (**Right**) *T. thermophilus* ribosome at 5.5 Å resolution [from (*7, 9*)]. Both are oriented such that the small subunit [ribosomal RNA (light blue) and protein (dark blue)] is in the front.

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