Aqp3) associated with progenitor cells, and fewer and less proliferative Krt5⁺ progenitor cells. These defects, as well as impaired branching morphogenesis, could be rescued by addition of the cholinergic agonist carbachol. Other studies in glandular epithelial cell lines showed that cholinergic stimulation triggers the release of heparin-binding epidermal growth factor (HBEGF), which binds to the EGF receptor (*2*). Knox *et al*. also found that addition of HBEGF rescued branching morphogenesis in denervated gland explants. The data support a model in which cholinergic stimulation maintains a population of proliferative Krt5⁺ progenitors in the embryonic submandibular gland in an EGF-dependent manner.

Coordinated development of the peripheral nervous system and branched epithelial organs containing progenitor cells is also implicated in other systems, including the lung, mammary gland, and prostate. For example, stimulation of mouse embryonic lung explants with nicotine increases branching (*3*). Knox *et al*. show that inhibiting acetylcholine stimulation of explanted developing mouse ventral prostate gland leads to decreased expression of markers associated with Krt5⁺ epithelial progenitor cells. Krt5⁺ basal stem cells of adult mouse and human airways express the nerve growth factor receptor NGFR (p75/Tnfrsf16) (*4*), but its function is not yet clear. As well, this

receptor is expressed on Krt5⁺ basal epithelial cells of human epidermis (*5*), but to date no function has been attributed to the NGFR in epithelial development. How both nerves and growth factors coordinately interact with the salivary gland epithelium during development can now be investigated with a combination of pharmacological and genetic tools and different injury-repair models in vivo and in organ culture.

The concept of nerve-dependent organ regeneration has long been studied in the salamander, an organism with a remarkable capacity for regeneration (amputated limbs fully regenerate within several weeks). Interestingly, this process does not occur if the nerve supplying the stump is severed (*6*). But unlike the mammalian salivary gland, salamander limb regeneration is not mediated by cholinergic stimulation (*7*), but by a growth factor produced locally by the nerve (*8*).

An important question is whether the nerve–progenitor cell interaction observed by Knox *et al*. has therapeutic potential. One side effect of radiation therapy for head and neck tumors is irreparable damage to the salivary glands (*9*). This can cause xerostomia (dry mouth), a condition that severely limits postradiation quality of life and is presently incurable. However, not all damage to salivary glands is irreversible. For example, ligation of the salivary duct

reverses gland atrophy in human patients as long as the autonomic innervation is intact (*10*). Thus, it might be possible to stimulate gland regeneration after irradiation by local pharmacological cholinergic stimulation of the epithelium (which harbors progenitor cells) or by promoting neurogenesis. Toward testing this hypothesis, Knox *et al*. cultured denervated lobules of adult mouse submandibular glands in the presence of the cholinergic agonist carbachol and found increased expression of Krt5 and Krt14, markers of progenitor cells. Determining whether this approach promotes organ growth and salivation in vivo will be an important next step.

References
1. S. M. Knox *et al*., *Science* **329**, 1645 (2010).
2. N. Prenzel *et al*., *Nature* **402**, 884 (1999).
3. C. Wongtrakool, S. Roser-Page, H. N. Rivera, J. Roman, *Am. J. Physiol. Lung Cell. Mol. Physiol.* **293**, L611 (2007).
4. J. R. Rock *et al*., *Proc. Natl. Acad. Sci. U.S.A.* **106**, 12771 (2009).
5. Y. C. Dou, L. Hagströmer, L. Emtestam, O. Johansson, *Arch. Dermatol. Res.* **298**, 31 (2006).
6. M. Singer, L. Craven, *J. Exp. Zool.* **108**, 279 (1948).
7. D. B. Drachman, M. Singer, *Exp. Neurol.* **32**, 1 (1971).
8. A. Kumar, J. W. Godwin, P. B. Gates, A. A. Garza-Garcia, J. P. Brockes, *Science* **318**, 772 (2007).
9. S. A. Bhide, A. B. Miah, K. J. Harrington, K. L. Newbold, C. M. Nutting, *Clin. Oncol. (R. Coll. Radiol.)* **21**, 737 (2009).
10. G. B. Proctor, G. H. Carpenter, *Auton. Neurosci.* **133**, 3 (2007).

10.1126/science.1196016

NEUROSCIENCE

Dendrites Do It in Sequences

Alain Destexhe

Brain cells are exquisitely sensitive to the pattern of inputs on their dendritic branches.

Nearly 50 years ago, Wilfrid Rall, one of the founders of computational neuroscience, proposed a mechanism to help answer one of the most basic questions in neuroscience: How do neurons detect, organize, and process the numerous chemical and electrical signals that flow from the thousands of synapses arrayed along their branching fibers (dendrites)? He proposed that the sequence in which synapses activate—specifically, whether the activation moves progressively in, along the branch toward the central cell body (soma); or out, away from the soma—could determine how

the soma responds (*1*). A pattern that moved in one direction, for instance, could cause a neuron to "fire," or produce an electrical spike, whereas the soma would remain quiet if the pattern of activation moved in the opposite direction.
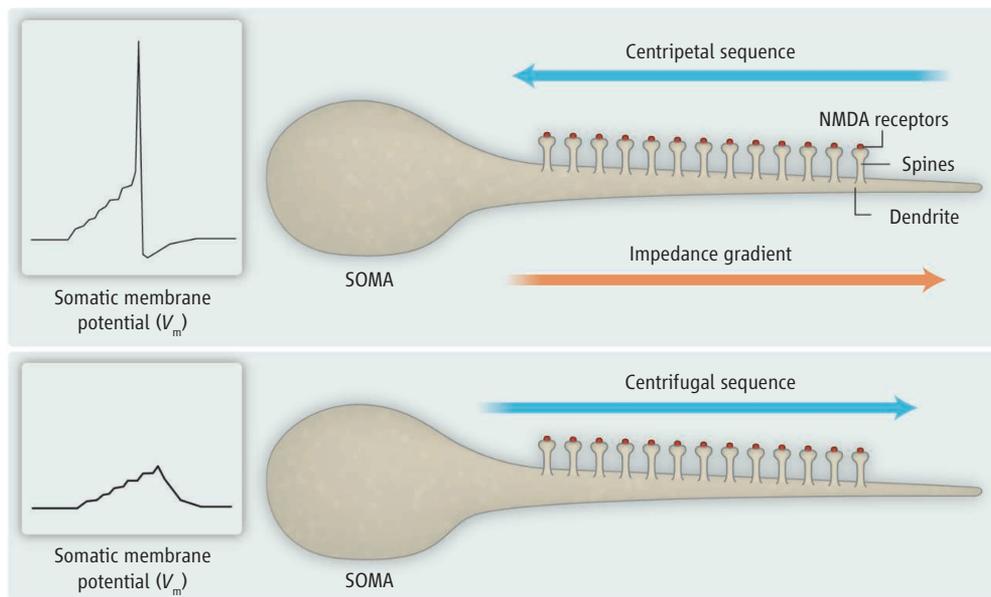
Over the past few decades, researchers have shown that individual neurons and populations process precisely timed sequences of inputs (*2*, *3*), and that "directional selectivity" appears to play a role in the sensory cortex, the part of the brain that processes information from the eyes, ears, and other sense organs (*4*). Investigators, however, had not been able to show experimentally that activating synapses in a centrifugal sequence (outward from the soma) caused a different neuronal response than activating the synapses in a centripetal

(inward) sequence. Branco *et al*. (*5*) do just that in a technically impressive set of experiments described on page 1671 of this issue. Using slices of brain tissue from rats and a laser-based technique that enables stimulation of selected parts of a dendrite, they confirm Rall's idea but also reveal some surprising aspects of how directional selectivity works.

Branco *et al*. studied cortical pyramidal neurons, which are among the largest brain cells and have several kinds of dendrites, each of which carries thousands of spines, or protrusions that receive input from a synapse. Using a technique called two-photon guided laser photostimulation, they were able to release glutamate, an abundant neurotransmitter, directly onto certain spines. This enabled them to precisely control stimulation

Unité de Neurosciences, Information & Complexité, CNRS, 91198 Gif-sur-Yvette, France. E-mail: destexhe@unic.cnrs-gif.fr

In and out. Neurons are sensitive to the sequence of activation of their synaptic inputs. Activating excitatory inputs on dendritic spines in a centripetal (**top**) or centrifugal (**bottom**) direction leads to different responses by the soma. One reason for this difference is that the thickness of dendrites tapers away from the soma, producing a gradient in electrical impedance. Activation sequences move with or against this gradient, and some patterns activate NMDA receptors (red), which in turn greatly amplify the excitatory signal.

in both space and time, thereby creating complex patterns of synaptic activation.

Their main finding is that the dendrites of cortical pyramidal neurons are exquisitely sensitive to the sequence of excitatory inputs, and that—as Rall predicted—centripetal activation leads to the strongest somatic response (see the figure). Rall's mechanism was not entirely correct, however, although he was remarkably forward-looking. He had postulated that directional sensitivity was entirely due to a "passive" mechanism related to the impedance, or resistance, that electrical signals encounter as they travel along a dendrite. Dendrites taper and become thinner farther from the soma. This tapering results in an "impedance gradient," with signals originating in distal dendrites (those farthest from the soma) experiencing greater impedance than those from proximal (closer) dendrites. As a result, Rall suggested that a dendrite can be seen as a "delay line" that is naturally sensitive to the location of the sequence of inputs. The main drawback of this passive mechanism, however, is that it requires very long dendrites to work efficiently. This is a problem in cortical pyramidal cells, in which up to 90% of inputs are on one type of dendrite: basal dendrites (6), which are too short for Rall's mechanism to work in a convincing way.

Branco et al. discovered an elegant alternative: Directional selectivity strongly depends on electrical voltage changes induced by synaptic activation. The authors demonstrated that a key player in sequence selectivity is the N-methyl-D-aspartate (NMDA) receptor, which is activated by glutamate. The receptor is voltage-dependent—it opens and closes an ion channel depending on the voltage dif-

ference between the interior and exterior of the cell (membrane potential)—and activates at depolarized, or more positive, membrane potentials (7, 8). Under the right circumstances, NMDA receptors can amplify the sensitivity of a dendrite to certain sequences of synaptic activation. A sequence "favored" by the impedance gradient, for instance, also activates NMDA receptors; in contrast, inputs acting against the gradient do not achieve sufficient depolarization to activate the receptor. Indeed, blocking NMDA receptors eliminates a large part of the difference between centripetal and centrifugal activations.

The most surprising result of Branco et al., however, is that dendrites are not only sensitive to ordered sequences of activation on a single dendrite segment, but are also sensitive to input sequences arranged randomly, or even distributed on different dendritic branches. The authors tested many spatiotemporal patterns of spine activation and found that sequences evoking the strongest responses always corresponded to waves of centripetal activation, even when the activity was occurring on different branches. They used computational models to explore this mechanism, and found that the two basic ingredients— the impedance gradient and amplification by NMDA receptors—are sufficient to reproduce the experimental findings. This shows that the neuron can be highly selective, responding only to very specific sequences of activation from a given dendritic morphology and distribution of synaptic contacts. This mechanism may be valid for many regions of the brain, because an impedance gradient and NMDA receptors are present in most neurons.

Still, many questions remain. One is whether these results can be validated in

vivo. In particular, researchers need to test whether directional sensitivity occurs with high levels of background synaptic activity, as in animals that are awake (9). Whether directional selectivity observed in the visual cortex has anything to do with the selectivity found by Branco et al. also needs to be addressed. Do dendrites process sequences involving both excitatory and inhibitory inputs? Investigators have shown that, in awake animals, some synapses inhibit a neuron's activity by influencing its action potential—the momentary change in the electrical potential on the cell's surface that occurs when it is stimulated; these inhibitory inputs may perhaps play a bigger role than excitatory inputs (9). Finally, it is not clear how dendritic selectivity operates under the highly nonlinear nature of voltage-dependent conductances (10). Could neurons regulate their intrinsic properties, position of inputs, and morphology to optimize selectivity to specific and preestablished sequences of inputs? How? Answering such questions will require new theoretical paradigms and experimental techniques.

References and Notes
1. W. Rall, in *Neural Theory and Modeling*, R. Reiss, Ed. (Stanford Univ. Press, Stanford, CA, 1964), pp. 73–97.
2. B. L. Strehler, R. Lestienne, *Proc. Natl. Acad. Sci. U.S.A.* **83**, 9812 (1986).
3. M. Abeles, *Corticonics: Neuronal Circuits of the Cerebral Cortex* (Cambridge Univ. Press, Cambridge, 1991).
4. D. Ferster, K. D. Miller, *Annu. Rev. Neurosci.* **23**, 441 (2000).
5. T. Branco, B. A. Clark, M. Häusser, *Science* **329**, 1671 (2010); published online 12 August 2010 (10.1126/science.1189664).
6. V. Braitenberg, A. Schüz, *Cortex: Statistics and Geometry of Neuronal Connectivity* (Springer-Verlag, Berlin, ed. 2, 1998).
7. L. Nowak, P. Bregestovski, P. Ascher, A. Herbet, A. Prochiantz, *Nature* **307**, 462 (1984).
8. C. E. Jahr, C. F. Stevens, *J. Neurosci.* **10**, 3178 (1990).
9. M. Rudolph, M. Pospischil, I. Timofeev, A. Destexhe, *J. Neurosci.* **27**, 5280 (2007).
10. R. R. Llinás, *Science* **242**, 1654 (1988).
11. Supported by CNRS, l'Agence Nationale de la Recherche, and the European Community (FACETS project).