Review

CellPress

Molecular mechanisms of activity-dependent changes in dendritic morphology: role of RGK proteins

Amy E. Ghiretti and Suzanne Paradis

Department of Biology, National Center for Behavioral Genomics, and Volen Center for Complex Systems, Brandeis University, Waltham, MA 02454, USA

The nervous system has the amazing capacity to transform sensory experience from the environment into changes in neuronal activity that, in turn, cause long-lasting alterations in neuronal morphology. Recent findings indicate that, surprisingly, sensory experience concurrently activates molecular signaling pathways that both promote and inhibit dendritic complexity. Historically, a number of positive regulators of activity-dependent dendritic complexity have been described, whereas the list of identified negative regulators of this process is much shorter. In recent years, there has been an emerging appreciation of the importance of the Rad/Rem/Rem2/Gem/Kir (RGK) GTPases as mediators of activity-dependent structural plasticity. In the following review, we discuss the traditional view of RGK proteins, as well as our evolving understanding of the role of these proteins in instructing structural plasticity.

Activity-dependent regulation of neuronal plasticity

An essential property of the central nervous system (CNS) is the ability to respond to sensory input with corresponding changes to neuronal structure and function. At the behavioral level, this plasticity allows an organism to respond to a changing environment appropriately in order to survive. At the level of neuronal networks, this sensory experience is reflected in changes in neuronal activity that, in turn, mediate structural plasticity: experience-dependent alterations in various neuronal processes, including synaptic function and neuronal morphology [1,2]. It is well established that sensory input during critical periods of development has a profound effect on neuronal networks, as illustrated by classic experiments exploring the development of ocular dominance and receptive fields in the visual system [3]. However, over the past 20 years or so, it has become abundantly clear that neuronal architecture remains plastic throughout development and into adulthood, with acute changes in sensory experience and neuronal activity continuing to elicit profound effects on synaptic plasticity and neuronal morphology [4].

Corresponding author: Paradis, S. (paradis@brandeis.edu).
Keywords: activity; dendrite; plasticity; RGK.

0166-2236/
 http://dx.doi.org/10.1016/j.tins.2014.05.003

One of the major consequences of increased activity in the nervous system is the increased transcription of the so-called activity-regulated genes, which occurs in response to extracellular stimuli such as growth factors or, in the case of neurons, depolarization that opens ion channels that allow calcium (Ca^{2+}) influx [1,2]. These extracellular events activate intracellular signal transduction pathways, many of which regulate transcription factors, which in turn cause changes in the expression of their downstream target genes [5]. Activity-regulated genes have been extensively implicated in various neuronal processes, including cell survival, regulation of dendrite morphogenesis, and synaptic plasticity [2,6].

Although many activity-regulated genes have been identified as positive regulators of neuronal structure and function, it is important to note that the upregulation of genes that restrict neuronal growth, synapse development, or synaptic transmission is equally important to maintain neuronal function in an appropriate physiological range in response to increased network activity [7] (Figure 1). For example, the activity-regulated genes *Arc* and *Mef2* limit the strength and formation, respectively, of excitatory synapses following increased activity [8–10]. In neurons, calcium-dependent signaling pathways are triggered by neuronal depolarization primarily via calcium entry into a neuron through NMDA receptors, or P/Q or L-type voltage-gated calcium channels (L-VGCCs) [11–13]. Interestingly, expression of some activity-dependent genes is dependent on calcium entry via only one of these sources (e.g., calcium entry through L-VGCCs but not NMDA receptors), suggesting that specific signal transduction pathways are activated in response to particular neuronal stimuli [11,14]. In general, activity-regulated genes are well poised to link changes in sensory experience to changes in neuronal structure and function.

Activity-dependent regulation of dendritic morphology

Structurally, one of the most salient aspects of neurons is their polarized morphology. Neurons are typically comprised of a cell body and an axon, through which they transmit information to other neurons, and a dendritic arbor, where input from other neurons is primarily received [15]. This dendritic arbor is usually highly branched, with the degree of complexity (a term that describes both the length of dendrites and the degree of

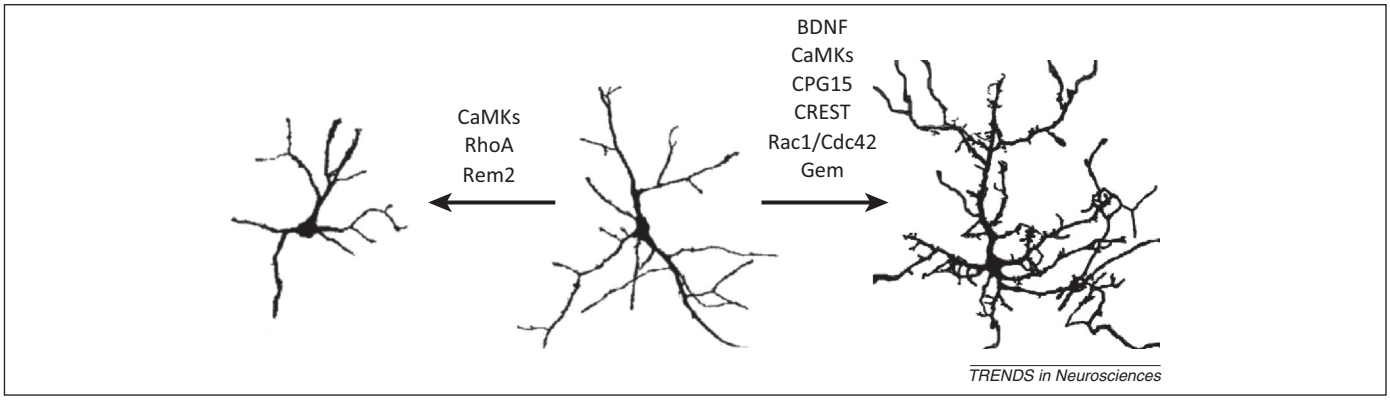

Figure 1. Positive (right) and negative (left) regulators of activity-dependent changes in dendritic morphology. Neuron images are five DIV cultured rat cortical neurons transfected with a green fluorescent protein (GFP)-expressing plasmid and treated with nifedipine (left), untreated (center), or treated with potassium chloride (right) (for further details, see [14]). Although the net effect of increased activity is increased complexity (right), a number of molecules that either enhance or inhibit the dendritic arbor are upregulated and contribute to the net change. Abbreviations: BDNF, brain-derived neurotrophic factor; CaMKs, Ca^{2+}/calmodulin-dependent protein kinases; CPG15, candidate plasticity gene 15; CREST, calcium responsive transactivator; DIV, day *in vitro*.

branching of the arbor) playing a major part in the function of the neuron. The dendritic morphology of a given neuron determines the connections that neuron will make, and neurons with distinct morphologies often serve different functions in neural circuits [16]. For example, pyramidal cells in the mammalian cortex and hippocampus are easily identified by their distinct apical and basal dendritic arborizations. Local interneurons (which have their own distinctive morphologies) and projections from other brain regions will target specific areas of the pyramidal neuron dendritic arbor, soma, or axon initial segment, and the proper integration of these multiple inputs is essential for proper circuit integration and, ultimately, function [17].

Dendritic morphology is highly subject to regulation by changes in neuronal activity. In general, the net effect of increased neuronal activity is an enhancement of dendritic complexity [18,19]. An elegant example of this comes from the optic tectum of *Xenopus laevis* tadpoles, where increased activity in the form of 4 hours of visual experience leads to an increase in dendritic complexity of tectal projection neurons *in vivo*; application of the NMDA receptor antagonist 2-amino-5-phosphonopentanoate (APV) suppresses the effect of visual experience on dendritic complexity, firmly demonstrating the role of activity in this process [18].

Much of our understanding of the molecular mechanisms that regulate activity-dependent cellular processes has come from studies using depolarization of neurons in culture with potassium chloride (KCl) [5], which results in an increase in dendritic complexity [14,19–21]. Although not a perfect mimic of neuronal activity *in vivo*, KCl-mediated neuronal depolarization causes physiologically relevant calcium influx into neurons [22–24]. Further, co-treatment of cultures with KCl and either APV or the L-VGCC blocker nifedipine attenuates depolarization-dependent increases in dendritic complexity, suggesting that calcium entry from multiple sources contributes to this net effect [14,19,20]. The identification and characterization of molecules that transduce this increase in calcium entry into a corresponding increase in dendritic morphology remains an area of intense research.

Importantly, it has become clear that activity regulates the function of both positive and negative mediators of dendritic complexity; it is the integration of these opposing signals that ultimately results in the proper dendritic morphology [14,18]. Presumably, the CNS evolved such that both positive and negative regulators of dendritic arborization are activated in order to ensure that growth-promoting processes do not proceed unchecked. A similar mechanism maintains homeostasis and allows for synaptic plasticity in the face of changing network activity, providing gain control such that signals are propagated successfully throughout a neuronal network [7]. This type of regulation is also present in many other tissue systems within an organism (e.g., immune system homeostasis) [25,26], where a balance between positive and negative signal transduction networks achieves the final outcome.

Molecules that regulate dendritic morphology

The development of the dendritic arbor is a highly dynamic yet carefully controlled process consisting of an early period of dynamic extension and retraction, followed by subsequent stabilization, pruning, and maturation of the arbor [27]. Studies in both invertebrate and vertebrate model organisms demonstrate that the ultimate morphology of a neuron is regulated by a variety of both intrinsic and extrinsic factors [28,29] (Figure 2). Within a given neuron, a number of molecules have been identified that help to shape the dendritic arbor in several different ways. Many genes, including transcription factors such as CUT, Abrupt, NeuroD, and calcium responsive transactivator (CREST), instruct dendritic morphology by initiating changes in gene expression either during development or in response to changes in neuronal activity [1,28]. Others, such as the Rho GTPases, interact directly with the actin cytoskeleton to effect changes in the cytoskeletal organization of dendrites [30]. In addition, secreted proteins such as neurotrophins [e.g., nerve growth factor (NGF) and brain-derived neurotrophic factor (BDNF)] also play a part in shaping dendritic morphology [31].

Interestingly, a number of molecules have been shown to preferentially affect either dendritic length or branching, whereas others mediate both components of dendritic complexity. The apparent selectivity of some molecules for mediating particular aspects of dendritic morphology adds a further layer of intricacy to the regulation of the dendritic