Development and Regulation of Dendritic Spine Synapses

Dendritic spines are small protrusions from neuronal dendrites that form the postsynaptic component of most excitatory synapses in the brain. They play critical roles in synaptic transmission and plasticity. Recent advances in imaging and molecular technologies reveal that spines are complex, dynamic structures that contain a dense array of cytoskeletal, transmembrane, and scaffolding molecules. Several neurological and psychiatric disorders exhibit dendritic spine abnormalities.

Dendritic Spines: What Are They? What Are They For?

Dendritic spines are micron-sized protrusions of the dendritic membrane that serve as the postsynaptic component for the vast majority of central nervous system excitatory synapses. They are found on excitatory and inhibitory neurons including glutamatergic pyramidal neurons of the neocortex and hippocampus as well as GABAergic cerebellar Purkinje neurons and medium-sized projection neurons of the striatum. Although their existence has been known for over a century (89), their ultrastructural and molecular organizations have only recently begun to be elucidated due to advances in microscopy and molecular approaches. The number and shape of dendritic spines are varied and highly mutable on time scales ranging from seconds to days, and morphological differences between spines are known to reflect functional differences (42). Furthermore, spine numbers and shape are regulated by both physiological and pathological events.

Most mature spines have a club-like morphology, with variably-shaped bulbous tips, ~0.5–2 μm in diameter, connected to the parent dendrite by thin stalks 0.04–1 μm long (41). Spine density ranges from 1 to 10 spines per micrometer length of dendrite, depending on neuronal cell type and maturational stage. By electron microscopy, the ultrastructure of dendritic spines is characterized by a conspicuous postsynaptic density (PSD). The PSD is a compact matrix that lies just beneath the postsynaptic membrane. It organizes the receptors and signaling molecules that are positioned across the synaptic cleft from clusters of presynaptic neurotransmitter vesicles (98).

The shape and stability/motility of spines are determined by a cytoskeleton composed mostly of filamentous actin (F-actin). Spines are nearly completely devoid of the intermediate filaments and microtubules that are plentiful in the dendritic shaft (48). Despite their modest size, most spines contain at least some form of smooth endoplasmic reticulum, which in the largest spines takes the form of a specialized organelle called the “spine apparatus” (34). Endocytic machinery is also identified in spines, within specialized subdomains (88, 102). In addition, polyribosomes and protein translational machinery are often anchored at the base of spines (80). Thus increasing evidence suggests that individual spines represent partially autonomous compartments, having their own regulated membrane-trafficking events that shutle components into and out of the spine membrane. Together, these specialized molecular assemblies determine spine shape and, most importantly, enable the postsynaptic neuron to respond biochemically to glutamate or other transmembrane signals (44, 100, 127). Proteomic approaches have begun to unravel the molecular composition of isolated PSD fractions and glutamate receptor complexes and have uncovered a remarkable degree of complexity (48, 83, 116, 124).

Although it appears that all spines share several core components, at least some of the apparent molecular complexity probably reflects the fact that individual spine synapses are heterogeneous, exhibiting variable levels of important signaling molecules that can fine-tune individual synaptic responses. Despite advances in our understanding of the molecular composition of spines, their function is not entirely understood. For example, why do some synapses occur on spines and others (“shaft synapses”) directly on the dendrite? One widely accepted explanation is that spines provide biochemical compartmentalization (76). The narrow neck of the spine creates a spatially isolated compartment where biochemical signals can rise and fall without spreading to neighboring synapses along the parent dendrite, thus allowing the isolation and/or amplification of incoming signals. Similarly, a spine compartment may help confine membrane trafficking to a localized region. Such restriction of molecular signals to one spine may contribute to the phenomenon of “input specificity,” allowing a given set of nerve terminals to induce changes only at those synapses that are specific to their postsynaptic contacts and not at other synapses on the same neuron that are driven by different axons (64).
Spine Development

During early synaptogenesis, dendritic shafts are covered with filopodia, which are long, narrow protrusions that are more transient and contain less F-actin than spines. Neuronal growth cones also display numerous filopodia, but these seem to be somehow distinct. Growth cone filopodia are involved in activity-independent dendritic growth and branching, whereas the filopodia that protrude from the dendrite shaft are involved in activity-dependent synaptogenesis (87). Dendritic filopodia transiently extend and retract from the dendritic shaft with an average lifetime of ~10 min (129). Filopodia probably function to maximize the chance encounter between a developing axon and a target dendrite. Once contact is made—either physically via cell-cell adhesions or chemically via locally released signals—a synapse can be initiated and proceed through appropriate maturational steps. Such maturation requires intricate crosstalk between the nascent presynaptic and postsynaptic parts of the synapse (31).

As synapse formation progresses over the course of several days, the numerous dendritic filopodia are gradually replaced by spines (67). It appears that many, but not all, spine synapses result when synapses initially form on filopodia, which then "convert" directly into spines. However, other synapses form directly on the dendritic shaft, followed by the gradual emergence of spines at the site of contact (24). Laboratory observations suggest that both mechanisms can and do occur even within a single neuron (22). It should be noted, however, that not all filopodia are transformed into spines (129). Filopodia are also found on nonspiny neurons during early synaptogenesis (121). Thus multiple factors, including cell-recognition molecules and downstream signals, orchestrate both synapse identity and synapse shape during neuronal maturation.

Regulation of Spine Development

The exquisite molecular organization of the pre- and postsynaptic apparatus requires positional information to coordinate the correct placement of pre- and postsynaptic elements. Once synapses have appeared, the next steps are to ensure synaptic specificity and to strengthen or weaken synaptic connections (126), processes that are believed to be at the core of learning and memory. The formation of synapses and their neurotransmitter specificity are controlled by the expression of adhesion and signaling molecules, including several classes of neural cell adhesion molecule (NCAM), N-cadherins, protocadherins, neurexins with their neurelinins, Eph receptors with their ephrin ligands, and extracellularly secreted molecules like proteoglycans (107, 117). The formation of cell-cell contacts at synapses is similar in basic principles to formation of junctions between nonneuronal cells. However, the polarized, asymmetric nature of pre- and postsynaptic elements confers unique demands on synaptic junctions. Identification of the key signals that establish initial contact is a rapidly growing area of investigation. Much of the remarkable diversity in synaptic function that varies across brain region, neuron type, maturational state, and activity level may stem from diversity in the molecules that govern synaptogenesis.

Cadherins are a family of calcium-dependent homotypic cell-adhesion molecules (CAMs) that are found in nearly all cells; neurons express an isoform known as N-cadherin (107). As synapses are initiated, N-cadherin begins to cluster on both pre- and postsynaptic sides of the nascent synapse. This clustering and adhesion is required to establish strong, stable junctions. In older synapses, N-cadherin clusters are mainly concentrated lateral to the PSD (106, 107, 113). To establish strong adhesion, N-cadherins must link to the cytoskeleton by binding to β-catenin, which in turns binds to α-catenin, which binds directly to F-actin. The cadherin/β-catenin complex also bridges to the actin cytoskeleton indirectly via actin-binding proteins such as α-actinin or profilin (74). The expression of a dominant-negative truncated N-cadherin, lacking parts of its extracellular domain, leads to major morphological and functional perturbation of spines and synapses in hippocampal neurons (111). These results are supported by studies with α-catenin-deficient neurons that suggested that α-catenin is required for both N-cadherin-mediated adhesion and the shortening of spines and the maturation of synapses (1). Importantly, depolarization-induced redistribution and association of β-catenin with N-cadherin indicates an active involvement of β-catenin in N-cadherin-mediated synaptic adhesion (71). β-Catenin’s phosphorylation state regulates its redistribution between the dendritic spine or the shaft compartment: a single point mutation (Y654F) that prevents phosphorylation by a tyrosine kinase concentrates β-catenin in spines, whereas a single point mutation that mimics phosphorylation (Y654E) accumulates β-catenin in dendritic shafts (71). Other catenins known as δ-catenins also participate in spine regulation (57). They not only bind to classical cadherins but also interact directly with cortactin in a tyrosine phosphorylation-dependent manner (65).

The protocadherins represent the largest subgroup of the cadherin superfamily, and they display numerous splice variants that could potentially encode aspects of synaptic diversity (55). Protocadherin extracellular domains exhibit characteristics of calcium-dependent adhesion molecules, but their cytoplasmic domains are different from those of classical cadherins. The α- and γ-protocadherin proteins are enriched in neurons and targeted to synapses (86). Synapse number is reduced in the spinal cord of mice...
lacking protocadherin-γ proteins, suggesting that they may be essential for the assembly of certain synapse populations (119).

Other candidates for mediating synapse selectivity include several members of the immunoglobulin superfamily of CAMs, which, like the cadherins, work through homotypic association of their extracellular domains to join adjacent cells. CAMs work in concert dynamically together with integrins to mediate and regulate adhesion across the synapse (4, 5, 12). Such immunoglobulin family members include, among others, NCAM, synaptic cell adhesion molecule, and nectin, and these are known in many cases to affect dendritic spine shape and development (see Ref. 117 for review).

The neurexin-neuroligin system has received recent attention as a major regulator of synapse and spine formation (95). These molecules work transsynaptically in pairs at both glutamatergic and GABAergic synapses, generally with neurexin on the presynaptic side inducing postsynaptic differentiation and neuroligin on the postsynaptic side inducing presynaptic differentiation (33).

One crucial aspect of synapse formation is whether a nascent synapse will develop into an excitatory or inhibitory contact. The tight balance between excitatory vs. inhibitory synapses regulates the overall excitability of the neuron and is thus critical for normal circuit establishment and firing pattern. The importance of the neuroligin family in this function was demonstrated using RNA interference directed against the neuroligin isoforms NL-1, -2, and -3, which showed that decreased expression of neuroligin resulted in a loss of both excitatory glutamatergic and inhibitory GABAergic synapses in cultured rat hippocampal neurons. The loss of glutamatergic synapses was accompanied by a dramatic decrease in dendritic spines. However, the loss of inhibitory synapse function was more predominant, resulting in a shift in the balance of excitability (14). In a complementary study it was shown that β-neurexin expressed on the surface of nonneural cells or attached to synthetic beads was by itself sufficient to induce the formation of postsynaptic specializations in contacting dendrites (33).

Initially it was thought that the localization of NLs, rather than their isoform, was the key determinant of synapse phenotype (59). However, it was recently shown that neuroligin splice variants can display distinct neurexin binding preferences and thereby induce distinct effects on synapse formation (9). In addition, association with postsynaptic scaffolding proteins, such as PSD-95, regulates the distribution of NLs and their synaptogenic activity. Whereas NL-2 is usually prevalent at inhibitory synapses, an increase in PSD-95 levels through overexpression sequesters NL-2 at excitatory postsynaptic sites (114), thereby reducing its distribution to inhibitory synapses. This results in an increased ratio of excitatory to inhibitory synaptic contacts (59, 60).

Extracellularly secreted molecules can also influence spine development. Syndecan-2, a heparin sulfate proteoglycan, was one of the first molecules identified that can trigger spine formation (23). Syndecan-2 accumulates on spines beginning at the onset of spine formation, and spine-like structures lacking synaptic contacts can be induced when syndecan-2 is overexpressed in neurons before spinogenesis (23). Brain-derived neurotrophic factor or stimulation of its receptor, TrkB, increases Purkinje and hippocampal cell spine density (53, 99). Soluble glia-derived signals are also important in synapse formation and maturation. One such signal secreted by astrocytes is cholesterol, which becomes bound to apoE-containing lipoproteins (68). In addition, thrombospondins released by astrocytes have been identified as participating in spinogenesis (16), and tissue plasminogen activator regulates spine pruning (66).

Intracellular components of normal spines can also sometimes trigger formation of spine-like protrusions in neurons that normally lack spines. For example, overexpression of the glutamate receptor 2 (GluR2) subunit of AMPA receptors induces spine formation in GABA-releasing interneurons, which normally lack spines (82). Also, overexpression of Shank, a scaffolding molecule that links up to the actin cytoskeleton, can trigger spine formation in such aspiny inhibitory neurons (92), whereas in spiny excitatory neurons, where it is expressed naturally, Shank overexpression induces spine enlargement (94). Such studies demonstrate that certain synaptic molecules can recruit the necessary cytoskeletal and membrane components that enable the postsynaptic membrane to reorganize and protrude into a spine-like structure.

Interactions between the membrane-bound ligands ephrins and their tyrosine kinase receptors, Ephs, can also strongly alter spine morphology. Neurons from mice lacking the three receptors EphB1, B2, and B3 fail to form mature spines in vitro, retaining immature filopodia and develop abnormal spines in vivo (43). The binding of ephrin B to EphB triggers various signal transduction pathways involving Rho-family GTPases (see below).

In summary, dendritic development and differentiation are regulated by a combination of neurotrophin, cell adhesion, and neural and glial activities. However,
research continues to uncover new and unexpected pathways that affect spine development. For example, recent studies (46) implicate a role for the origin recognition complex, the core machinery for initiating DNA replication, in the regulation of dendrite and spine morphology.

**Spines are Dynamic Structures**

Dendritic spines are highly dynamic in size and shape (8), and it is increasingly apparent that wholesale turnover of spines is a normal part of brain physiology. Spine density reaches its maximum level during late development when synaptic plasticity is at its height and then decreases to a relatively stable level throughout adulthood (126). There are regional differences in the timing of spineogenesis. In rodent hippocampus, spine formation peaks during the third postnatal week, then spine numbers remain relatively constant in this region. In contrast, in mouse sensory cortex spinogenesis occurs at a steady decline from adolescence through late adulthood. Interestingly, experience plays an important role in this normal loss of spines, inasmuch as it can be prevented by sensory deprivation (131).

Studies using multiphoton microscopy over days to months in living mice have elegantly confirmed the long-held belief that spines and their synapses can form and retract throughout adulthood (36, 112). The mature brain retains the capacity to form new synapses and thereby remodel its circuitry throughout life. The degree of spine turnover varies with age and possibly also with brain region. Studies in somatosensory cortex by Svoboda and colleagues (112) indicated that a significant fraction of spines turn over with a half-life of hours to days, whereas studies by Gan and coworkers (36) in visual cortex suggested that the vast majority of spines are stable over several months. It is reasonable to imagine that different brain circuits turn over their spines and synapses with different frequencies, perhaps reflecting a need to retain stable circuits for certain brain functions, such as motor patterns or other forms of persistent memory, but to allow continual remodeling of circuits elsewhere, for example to acquire and process new information. However, it remains to be determined whether such variation in spine stability truly exists. The reported differences in spine turnover in vivo might simply reflect methodological differences among laboratories. Recently, it was reported that in adult mice (>4 mo of age), only 3–5% of spines turn over within 2 wk, regardless of which cortical region was examined (130).

Although many spines can persist for days to months, their size and shape continually shift on a time scale of seconds to minutes (20, 26). The functional significance of this “morphing” of spine shape is a topic of investigation (39). Transmembrane signals and regulatory molecules can alter morphing behavior (10, 27). It is likely that the ability to rapidly change the biochemical compartmentalization allows for the adjustment of signaling properties of the synapse (7). Alternatively, such morphing may be the consequence of postsynaptic membrane trafficking. Active exocytosis and endocytosis, which continually remodel the synaptic membrane, mediate receptor turnover and thereby synapse strength (85). Changes in spine shape and number are observed following high-frequency synaptic activity, behavioral stimuli, or in response to endogenous hormonal cycles (39, 62). Thus both wholesale spine turnover and morphological changes in existing spines are important for establishing and modulating synaptic circuits during development and synaptic plasticity.

**Regulation of Spine Motility and Stabilization**

Actin filaments provide the main foundation for spine shape, motility, and stability. Several actin-regulatory pathways activated by transmembrane signals are now known to influence the properties of dendritic spines (FIGURE 1). In all cells a variety of actin-binding and -regulatory proteins help determine the degree of actin polymerization and thus the equilibrium between the two forms of actin: G-actin monomers and F-actin filaments. Actin filaments have two ends: a fast-growing barbed end and a slow-growing pointed end. Actin monomers are primarily added to the barbed ends in cells unless the filament ends are capped. The importance of F-actin in maintaining spine shape was demonstrated by the fact that the actin-stabilizing compound jasplakinolide was sufficient to prevent excitotoxic spine collapse in neuronal cultures (40), Star et al. (103), using fluorescence recovery after photobleaching (FRAP), discovered that 85% of actin in the spine is dynamic, with an average half-life of 44 s, and was independent of the size of the spines. However, such rapid turnover is somewhat at odds with the concept of stable actin filaments maintaining postsynaptic components and spine integrity. Low concentrations of the actin assembly inhibitor cytochalasin D, which caps and stabilizes the barbed end of actin filaments, causes spines to round up and cease morphing but not to collapse altogether (21, 26), thus implying that some pools of spine actin turn over more slowly than others. Finally, new approaches, such as intracellular and extracellular application of rhodamine-phalloidin (61) or analysis of actin polymerization based on fluorescence resonance energy transfer (FRET) probes (78), have demonstrated that activity-dependent modifications of spine shape correlate with the degree of actin polymerization.

Given that actin is at the crux of spine number, shape, and motility, the mechanisms that regulate actin in spines have become the topic of intense inves-
FIGURE 1. Some important components of dendritic spines
Spines are small membrane protrusions at synaptic junctions that use the excitatory neurotransmitter glutamate, which is released from synaptic vesicles clustered in the presynaptic terminal. Across from these glutamate release sites, α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid (AMPA) and N-methyl-D-aspartate (NMDA) subtypes of glutamate receptors are clustered at the postsynaptic active zone within a dense matrix called the postsynaptic density (PSD; pink). Beyond the PSD lie subregions of spine membrane that contain G protein-coupled glutamate receptors (mGluR) and endocytic zones for recycling of membrane proteins. Receptors, in turn, connect to scaffolding molecules, such as PSD-95, which recruit signaling complexes (e.g., regulators of RhoGTPases, or protein kinases). Actin filaments provide the main structural basis for spine shape. Via a network of protein interactions, actin filaments indirectly link up with the neurotransmitter receptors and other transmembrane proteins that regulate spine shape and development, including Eph receptors, cadherins, and neuroligins. Actin-regulatory molecules such as profilin, drebrin, cofilin, and gelsolin control the extent and rate of actin polymerization. These, in turn, are regulated by signaling cascades through engagement of the transmembrane receptors.
tigation. Actin depolymerization factor (ADF)/cofilin severs actin filaments without capping, thus promoting actin turnover. Phosphorylation of ADF/cofilin by LIM kinase inhibits this function (3, 70, 123). A role has been implicated for ADF/cofilin in spine-shape regulation during long-term potentiation and depression (28, 128). In addition, ADF/cofilin activity, spine structure, and long-term potentiation are all altered in LIM kinase knockout mice (69). Such mice are one model for Williams syndrome, a genetic disorder in which cognition is severely altered and spine synapses show structural abnormalities (45). Gelsolin also severs actin filaments but caps the barbed ends, thereby preventing further polymerization of the newly exposed barbed ends (17). Neurons from gelsolin knockout mice exhibit altered N-methyl-D-aspartate (NMDA) channel currents (29) and impaired activity-dependent actin stabilization in spines (103). These data suggest that gelsolin is involved in NMDA receptor-mediated F-actin function.

**Spines and Rho-Family GTPases**

Members of the Rho-family GTPases, a subgroup of the Ras superfamily of GTPases, are key regulators of the actin cytoskeleton (37, 38). These proteins function as molecular switches that cycle between an inactive GDP-bound form and an active GTP-bound form. Guanine nucleotide exchange factors (GEFs) stimulate the interaction of G proteins with their effectors by catalyzing GDP-GTP exchange, whereas GTPase-activating proteins (GAPs) inhibit effector activity by stimulating the hydrolysis of GTP to GDP (73). The GDP-bound, inactive form of the GTPases is retained in the cytoplasm by GDP-dissociation inhibitors.

The Rho GTPases RhoA, Rac, and Cdc42 have all been implicated in dendritic spine formation and shape (75). As with other cellular functions, Rac and Rho appear to act in opposite fashions in regulating spine numbers (108). Constitutively active RhoA decreases both spine density and length, whereas its inhibition by C3 exoenzyme increases them. In contrast, neurons expressing constitutively active Rac 1 tend to form numerous small spines. Such spines are often engaged in multiple synaptic contacts, which is otherwise rare in normal animals (63). Dominant negative Rac causes a reduction in spine density, similar to constitutively active RhoA, in rat and mouse hippocampal slices (72, 108). Blockade of Rac activity seems also to transform the remaining spines into long, thin, filopodia-like protrusions (109). Rnd1, a constitutively active Rho family GTPase that, during synaptogenesis, is strongly localized in spines of primary hippocampal neurons, induces a similar spine-neck elongation when overexpressed and an overall decrease in spine density and width when suppressed (50). Rnd1 acts as an endogenous antagonist for RhoA through association with p190RhoGAP or direct inhibition of Rho kinase, one of the effectors of RhoA (120). The role of Cdc42 in spine morphogenesis is presently less well defined. Neither its constitutively active nor dominant negative forms significantly altered spine density or length when overexpressed in 2-wk-old neurons cultured from newborn mice (108). However, other recent findings (49) do implicate a role for Cdc42 in spine development.

**Effectors of Spine Changes**

Some of the signaling cascades involving Rho-family GTPases and their multiple GEFs or GAPs have begun to be elucidated. Kalirin-7, a GEF for Rac1, mediates EphB effects on spine maturation by activating Rac1 and its downstream effector p21-activated kinase (PAK) (84). PAK is a protein kinase that, like LIM kinase described above, can specifically phosphorylate and thereby inactivate ADF/cofilin, thereby altering spine actin turnover. An alternative pathway recently identified for EphB signaling suggests a model in which activated EphB forms a complex with intersectin, a specific GEF for Cdc42, together with the Wiskott-Aldrich syndrome protein (N-WASP), one of the effectors of Cdc42. Such a complex triggers actin polymerization via the Arp2/3 complex, which is proposed to result in expansion of dendritic spine heads (49). Pix, a GEF for Rac, together with its binding partners G protein-coupled receptor kinase-interacting protein 1 (GIT1), Rac, and the Rac effector PAK also regulate dendritic spine number and morphology (125). The myosin II regulatory light chain (MLC) is also downstream of the GIT1/Pix/Rac/PAK pathway. MLC activation stimulates dendritic spine and synapse formation, similar to Rac or PAK activation (125).

Another type of downstream effector of both Rac and Cdc42 is the insulin receptor substrate 53 (IRS53), which is highly expressed in the PSD (15). IRS53 interacts with WAVE 2, a member of the WASP family. Overexpression of IRS53 in cultured neurons increases the number of dendritic spines, without affecting their length and width; conversely, knockdown of IRS53 reduces density, length, and width of spines (15). Similarly, Abl-interactor (Abi) family of adaptor proteins are Rac effectors involved in regulating dendritic spine morphology. Loss of Abi2 results in a reduction of the length and density of dendritic spines, as well as impaired learning and memory (35). These deficits may be explained by the interaction of Abi2 with WAVE 2 and the Nck-associated proteins Nap1/Nap125, ultimately resulting in reorganization of spine F-actin and consequent alteration in synapse function.

Another way in which the Rho-family GTPases can affect the actin cytoskeleton is via the regulation of signaling phospholipids, such as phosphatidylinositol-4,5-bisphosphate (PIP2) (118). PIP2 stimulates actin
polymerization in several ways, such as removing gelsolin and capping protein from actin filaments (52) or activating regulatory proteins like profilin (122). Although roles in spines for PIP2 and other phospholipids have not been examined in detail, a significant function seems likely, since recent studies demonstrate that the protein myristoylated alanine-rich C-kinase substrate (MARCKS), which binds and sequesters PIP2 in a phosphorylation-dependent manner, has rapid and dramatic effects on spine shape and stability (10). MARCKS also binds directly to actin filaments and the small second messenger CaC2(calmodulin and thus may be a key mediator of both PKC and calmodulin-dependent spine functions.

In addition to the Rho family, two other small GTPases, Ras and Rap, are important in the context of dendritic spines, where they interact with PSD-95 through the signaling molecules synaptic GAP (SynGAP) (13) and spine-associated RapGAP (SPAR) (81), respectively. SynGAP is a GAP of Ras. Neurons cultured from SynGAP-deficient embryos form spines and synapses earlier, and they become larger than those in wild-type neurons (115). SPAR also causes enlargement of spine heads; such spines often exhibit an irregular morphology and bear multiple synapses (81).

Other significant players that influence and/or regulate spine shape and stability include cytoskeletal proteins such as profilin, cortactin, drebrin, and Homer, which have been discussed in a recent review (56), as well as neurabin and spinophilin, which are two actin-binding proteins (93, 110). In addition, the loss of myosin Va (as seen in the dilute mouse) (18, 105) or myosin VI (as in the Snell’s waltzer mouse) (79) alters clathrin-mediated endocytosis of α-amino-3-hydroxy-5-methyl-4-isoxazolepropionic acid receptors (AMPARs), resulting in changes in synapse number and dendritic spine length.

Dendritic Spine Pathology in Neurological Disorders

Myriad psychiatric and neurological diseases are characterized by spine loss or morphological disruption (6). Spines become malformed or lost in epilepsy, stroke, trauma, schizophrenia, dementia, major depression, normal aging, and chronic substance abuse (25, 30, 76, 104). Because spine morphology is linked to synaptic function, altered spines in disease states are likely to have diverse functional effects. Cognitive deficits in neurological disorders are frequently characterized by aberrant spine structure and plasticity that are restricted to the critical brain regions affected by the disease. For example, schizophrenic patients exhibit decreased spine density in prefrontal cortical pyramidal neurons (30), and loss of spine density is observed in hippocampus in both human epilepsy (104) and animal models (51, 54). In contrast, chronic administration of cocaine and amphetamine produce increased spine density in nucleus accumbens, a region tied to addiction (77, 91). Chronic neurodegenerative diseases such as Alzheimer disease and Parkinson disease often include spine loss or other dendritic changes in the early stages of disease progression (97). Although it is not known whether spine abnormalities are a direct cause of cognitive deficits or are secondary to some other event, a molecular understanding of spine function will shed light on the implications of these region-specific defects.

Several hereditary forms of mental retardation, both syndromic and nonsyndromic varieties, are attributable to defects in expression of proteins that regulate spine development and maintenance. One of the earliest of such disorders to be linked to spine abnormalities was fragile X syndrome, wherein the fragile X mental retardation protein (FMRP) fails to express properly due to a nucleotide expansion in noncoding regions of the gene (19). Fragile X syndrome patients and FMRP knockout mice both show reduced numbers of spines, with those remaining being long and spindly in shape. FMRP is involved in regulating protein translation locally at synapses and thereby affects a number of specific target molecules, including some involved in cytoskeletal function (90); for review, see Ref. 2. The FMRP protein has been shown to interact with cytoplasmic fragile X mental retardation protein interacting (CYFIP)1/2, effectors of Rac1, resulting in thin and elongated dendritic spines with small synaptic contacts (11, 96). Forms of nonsyndromic X-linked mental retardation have similarly proved to involve defects in genes that encode various actin-regulatory pathways. Examples include Rho-linked genes such as the Rho-GEF α-Pix, the Rac effector and LIM kinase activator Pak3, and the Rho-GAP oligophrenin 1 (see Refs. 10a and 32 for a recent review).

Perspectives

New molecular and imaging methods have rapidly expanded our knowledge of the composition of the spine. We now appreciate that hundreds of different molecules reside within this tiny compartment. Attention is now turning beyond cataloging these numerous proteins and signal-transduction pathways toward understanding how such diversity contributes to morphological and functional heterogeneity of synapses (47, 58, 101). In addition, many other questions remain: What changes do mature and well established spines undergo during learning, memory loss, or cognitive malfunction? What mechanisms govern spine disassembly? What is the relationship between the plasticity of spines and the movements of proteins and membranes into and out of the spine compartment? Does the morphing of spine heads relate to dendritic spine function or turnover? What is the rela-
tionship among presynaptic and postsynaptic compartments and their surrounding glial cells at both emerging and established synapses? Answers to these and other questions will continue to inform our understanding of neural physiology and neural disease as spines begin to yield their secrets.

References
43. Henkemeyer M, Itis OS, Ngo M, Hickmott PW, and Ethell IM. Multiple EphB receptor tyrosine kinases shape dendritic


107. Takeichi M and Abe K. Synaptic contact dynamics
106. Takeichi M. Cadherin cell adhesion receptors as a
109. Tashiro A and Yuste R. Regulation of dendritic
108. Tashiro A, Minden A, and Yuste R. Regulation of
102. Song I and Huganir RL. Regulation of AMPA
103. Star EN, Kwiatkowski DJ, and Murthy VN. Rapid
105. Takagishi Y, Oda S, Hayasaka S, Dekker-Ohno K,
111. Togashi H, Abe K, Mizoguchi A, Takeoka K, Chisaka O, and Takeichi M. Cadherin regulates
120. Wennerberg K, Forget MA, Ellerbroek SM, Arthur
WT, Burridge K, Settleman J, Der CJ, and Hansen
SH. Rnd proteins function as RhoA antagonists by
114. Varoqueaux F, Jamain S, and Brose N. Neuroligin
118. Weernink PA, Meletiadis K, Hommeltenberg S,
Hinz M, Ishihara H, Schmidt M, and Jakobs KH.
Activation of type I phosphatidylinositol 4-phos-
110. Terry-Lorenzo RT, Roadcap DW, Otsuka T, Blangried TA, Zamarano PL, Gamme CC, Shenolkar S, and Ehlers MD. Neurabin/protein
phosphatase-1 complex regulates dendritic spine
morphogenesis and maturation. Mol Biol Cell 16: