Dendritic Spines: Similarities with Protrusions and Adhesions in Migrating Cells

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Abstract: Dendritic spines are specialized, micron-sized post-synaptic compartments that support synaptic function. These actin-based protrusions push the post-synaptic membrane, establish contact with the presynaptic membrane and undergo dynamic changes in morphology during development, as well as in response to synaptic neurotransmission. These processes are propelled by active remodeling of the actin cytoskeleton, which includes polymerization, filament disassembly, and organization of the actin in supramolecular arrays, such as branched networks or bundles. Dendritic spines contain a plethora of adhesion and synaptic receptors, signaling, and cytoskeletal proteins that regulate their formation, maturation and removal. Whereas many of the molecules involved in dendritic spine formation have been identified, their actual roles in spine formation, removal and maturation are not well understood. Using parallels between migrating fibroblasts and dendritic spines, we point to potential mechanisms and approaches for understanding spine development and dynamics.

INTRODUCTION

Dendritic spines are small protrusions that decorate the dendrites of Purkinje neurons in the cerebellum and pyramidal neurons in the cortex and hippocampus [1, 2]. Dendritic spines function as specialized post-synaptic structures that support excitatory neurotransmission [2-4]. They contain ion channels and adhesive receptors, as well as a multitude of signaling intermediates and cytoskeletal components [5, 6]. These molecules are essential for transmission of synaptic input and also support long-term responses to stimulation, which are central for learning and memory.

Dendritic spines adopt varied morphologies, from long, filopodia-like to short and stubby, and have a well-defined life cycle (Fig. **1A**). During spinogenesis, dendritic spines appear as immature precursors, which are usually long and thin (Fig. **1A**, *left*). A fraction of these undergo maturation, becoming shorter, thicker and wider, i.e. mushroom-shaped or stubby; and those spine precursors that are not innervated tend to turn over, undergoing cycles of growth and shrinkage (Fig. **1A**, *middle and right*) [1, 7-9]. Morphological maturation of spines can be induced by physical contact with an axon and associated with synaptic stimuli. For example, mature spines of pyramidal cells are stabilized by synaptic input; but removal of afferent input, such as whisker trimming, results in the selective spines loss [10, 11]. On the other hand, dendritic spines on Purkinje cells of the cerebellum form and stabilize in the absence of afferent input [12].

The increase in contact area with the presynaptic terminal correlates with synaptic strength, which contributes to long-term potentiation (LTP) by increasing synaptic receptor density at the synaptic cleft. Electrophysiologic studies show that the bulbous head morphology of the mature spine is better suited to receive and propagate neuronal signals than the thin structure of the immature spine [13]. Synaptic input itself may also induce such an increase in surface contact area [14, 15].

Actin is a major component of dendritic spines. Its polymerization and organization dictate the size, motility, and morphology of the spines and has a profound impact on synaptic transmission [2, 16]. For example, inhibition of actin polymerization or depolymerization using chemical inhibitors disrupts LTP [17]. Furthermore, LTP induction causes an increase in F-actin which may underlie the structural enlargement of spine heads [18]. One mechanism is the recruitment or activation of actin regulators. For example, the actin-binding protein profilin, is targeted to spine heads in response to postsynaptic glutamate receptor activation; this increases the pool of actin monomers available for filament assembly. Profilin enrichment in spine heads also inhibits spine motility and promotes maturation [19].

The organization of actin in spines is tightly controlled by a multitude of signaling proteins. Interestingly, some diseases characterized by cognitive decline or impairment, such as non-syndromic mental retardation, schizophrenia, Down's syndrome or Alzheimer's disease, display abnormal spine morphology and/or a decreased number of dendritic spines as a result of alterations in actin regulatory molecules. For example, long tortuous spines lacking a bulbous head and dendrites lacking spines have been described in individuals with non-syndromic mental retardation, schizophrenia, and Down's syndrome [20]. Genomic mutations of different modulators, activators and effectors of Rho GTPases involved in actin reorganization have been linked to families with a high incidence of non-syndromic mental retardation [21-24]. Also, the beta-amyloid oligomers that cause inflammatory damage to the brain in Alzheimer's disease also alter the function of key Rho GTPases that regulate actin organization, causing long-term disassembly of the synaptic

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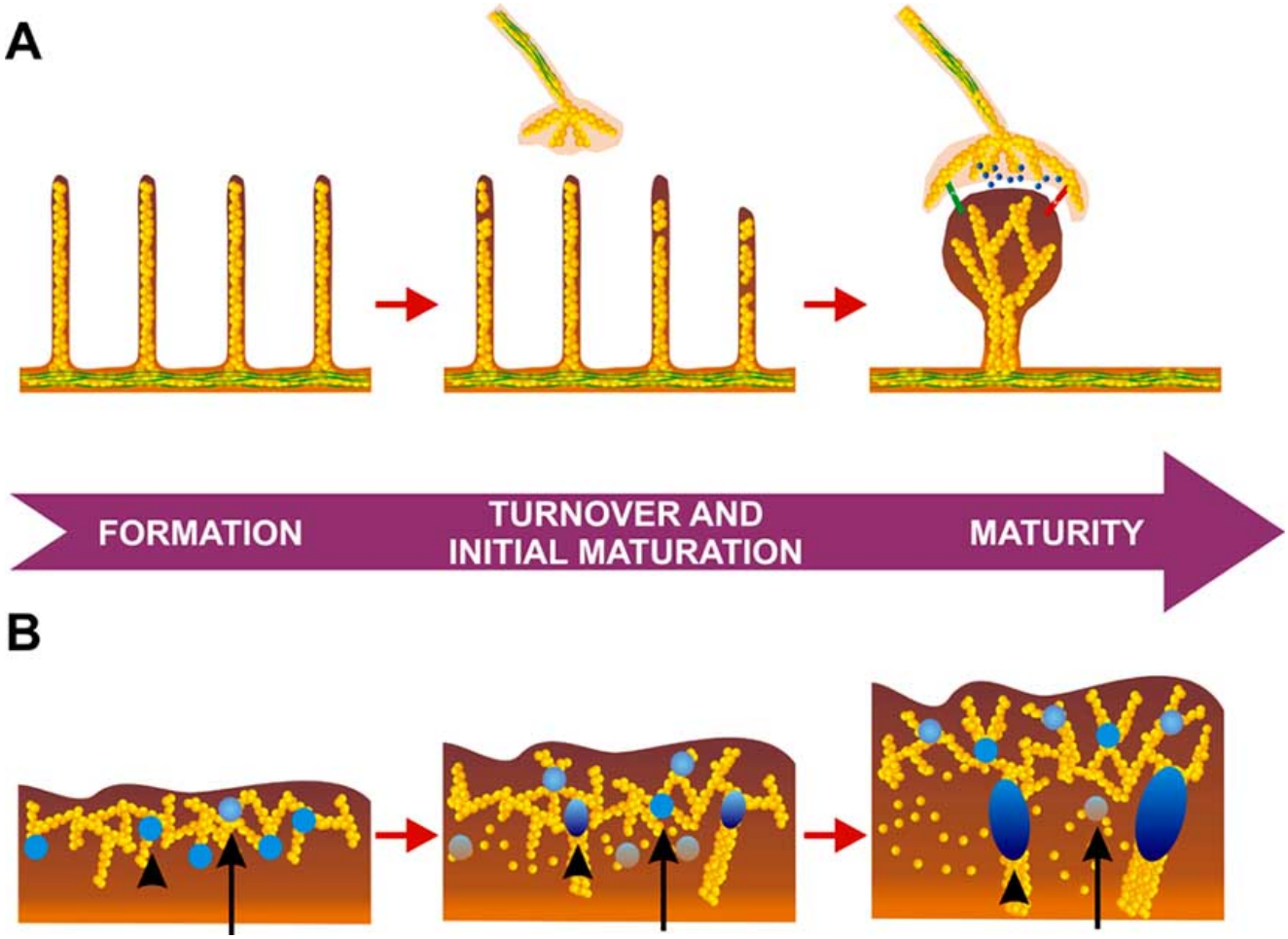

Fig. (1). Formation and evolution over time of dendritic spines and adhesions in migrating cells.
(**A**) Dendritic spine formation. *Left*, immature spine precursors form along the dendritic shaft, driven by actin polymerization. *Middle*, pre-synaptic contact and/or neurotransmitter secretion stabilizes an immature dendritic spine, whereas immature precursors that are not contacted by pre-synaptic portals disassemble (represented by breaking actin filaments in protrusions). *Right*, stable contact with a pre-synaptic terminal induces active remodeling of the post-synaptic terminal, which becomes shorter and wider. This process is driven by the combination of synaptic input (dark blue spheres) and adhesive signaling (green-red receptor pairs). The unselected precursors are reabsorbed in the dendritic shaft. A single actin filament in each protrusion is shown for simplicity.
(**B**) Adhesion assembly, maturation and turnover in migrating cells. *Left*, nascent adhesions form inside the branched actin network at the leading edge (indicated by arrowhead and arrow). *Middle*, as the protrusion advances, some adhesions elongate centripetally as the actin filaments with which they associate become larger and thicker (arrowhead); newly formed adhesions are stable as long as they are associated to the branched actin network (arrow). *Right*, maturing adhesions (arrowhead) continue growing as the actin bundles become thicker and more stable. Adhesions not associated with growing actin bundles turn over and disappear as the branched actin network moves past them (arrow).

actin filaments and cognitive decline [25, 26]. Thus, proper regulation of the actin cytoskeleton is crucial for the morphological plasticity of the spine and provides a mechanistic link to cognitive function.

Adhesion is another critical component of dendritic spines. In general, adhesion provides anchoring, traction and communication with the cellular environment to optimize cell behavior, or to ensure a specialized response, such as immune activation, or transmission of synaptic input [27]. From this point of view, dendritic spines comprise the post-synaptic half of a highly specialized cell-cell adhesion structure that forms between pre-synaptic and post-synaptic terminals. Several families of adhesion receptors are found in dendritic spines, including integrins [28, 29], cadherins [30, 31], neurexins/neuroligins [32, 33], Eph receptors [34] and other families of specific neuronal receptors, such as SynCAMs and SALMs [35, 36]. These receptors are involved in both spinogenesis and synaptogenesis [37-39].

A common property of adhesion receptors is that ligand binding induces the formation of supramolecular complexes that contain signaling adaptors and cytoskeletal molecules [40]. These "adhesions" are signaling centers that provide anchorage and traction for the organization of the actin cytoskeleton, which drives protrusion, adhesion modulation, and also controls gene expression [29, 41, 42]. Thus, actin and adhesion are critical components not only in a variety of cell types and processes, e.g. migratory lamellipodia and filopodia in motile cells, growth cones in neurons, cell-matrix adhesions and cell-cell junctions in epithelial cells, but also in dendritic spine formation [43-47]. Furthermore,

many regulators of both actin and adhesion are common throughout the different cellular systems. This striking resemblance is clear at a molecular level but has not been exploited explicitly and aggressively to develop insights into dendritic spine formation and structure and synaptic function.

In this mini-review, we discuss what is known about the function of actin and adhesion in non-neuronal systems and its implications and parallels for dendritic spine formation and organization. We highlight the critical role of the actin cytoskeleton and its regulators in the development, removal and maintenance of dendritic spines, pointing out the common players and their spatiotemporal regulation. Since other reviews in this volume are specifically devoted to the detailed description of some of the cytoskeletal and regulatory molecules in the synapse, we will not address their molecular characterization, but rather focus on their role in the morphological and compositional changes that take place during the life time of dendritic spines.

SPINOGENESIS, LIKE PROTRUSION AND ADHESION, IS DRIVEN BY ACTIN POLYMERIZATION

Two hypotheses have been postulated to explain initial spinogenesis [1]. One hypothesis proposes that contact of a pre-synaptic terminal with the shaft of the post-synaptic membrane induces the formation of a protrusion. Conversely, another hypothesis proposes the spontaneous initial formation of multiple immature dendritic protrusions, followed by contact with presynaptic terminals, which induces their maturation. Immature dendritic protrusions seem to have an active function in this process; their motion in time-lapse movies suggests they may play an exploratory role, cycling between protrusive elongation and retraction until physical contact with a pre-synaptic terminal is made [48, 49].

Immature spines (or dendritic spine precursors) are usually long, thin actin rich protrusions. Actin polymerization, which creates protrusions in migrating cells and growth cones, is likely to drive the initial emergence of immature dendritic precursors as well. There are two main modes of actin polymerization: a linear mode that is propelled by formins (e.g. mDia1, 2 and 3) (Fig. **2**); and a branched mode nucleated by the Arp2/3 complex, which binds to the side of an actin filament and promotes growth of another actin filament at a 70° angle (Fig. **2**) [50].

The thin, linear shape of dendritic precursors suggests the involvement of mechanisms used to generate filopodia in other cell types; however, it seems clear that these precursors are not identical to filopodia. They do not contain typical filopodial markers such as fascin, which bundles F-actin into tight parallel arrays [16]. Rather, barbed ends of F-actin are seen at the base of dendritic protrusions in addition to their tips, suggesting the existence of anti-parallel arrangements of actin filaments in immature spine precursors [51]. Also, Cdc42, which generates filopodia in migrating cells *via* activation of the formin mDia3, does not produce an increase in dendritic spine precursors [51]. Rather, expression of a constitutively active mutant of Cdc42 promotes spine head formation, causing an increase in the number of mushroom-shaped and stubby spines [51]. RNAi inhibition or a dominant negative form of Cdc42 inhibits dendritic spine and synapse formation [52], suggesting that Cdc42 is necessary for maturation; but its activation is not sufficient to induce the initial outgrowth of spine precursors from the shaft of dendrites. Interestingly, a similar GTPase/formin tandem, Rif/mDia2 may fulfill this role in hippocampal neurons; exogenous expression of either Rif or mDia2 promotes formation of long and thin dendritic spines (Fig. **2**) [51].

Arp2/3, which produces branched actin, also localizes to dendritic precursors and is involved in dendritic spine formation. RNAi knockdown of the Arp2/3 complex or its upstream activator N-WASP inhibited spine and synapse formation, as shown by a decrease in the total number of dendritic spines and synapses [52]. Similar results were observed in hippocampal sections from mice deficient for WAVE-1, another upstream activator of Arp2/3 [53]. This study also revealed altered neuritogenesis and field excitatory post-synaptic potential (fEPSP) in WAVE-1-deficient mice [53]. Several other studies have ascribed an important role to the small GTPase Rac and its downstream effectors in dendritic spine formation [54-56].

The complementary function of the Arp2/3 complex and formins in the formation of immature spine precursors can be inferred from studies in motile cells, in which actin polymerization drives formation of filopodia and advancing protrusions. Filopodia are generated by formin-driven actin polymerization into thin parallel filaments. Close to the leading edge of the protrusion, actin is organized in a branched network nucleated by the Arp2/3 complex [44, 50]. Formins also participate in this process by inducing polymerization at the growing (barbed) ends of these branches [57]. Often, advancing protrusions contain embedded filopodia that emanate from Arp2/3-dependent branching points [58], suggesting that Arp2/3 may also participate in filopodia formation. Translating these observations to immature spine formation suggests that activation of the Arp2/3 complex in the dendritic shaft could generate a branching point, which could be subsequently extended by the action of mDia2 or Rif/mDia3, resulting in linear actin arrays typical of immature spine precursors. However, the localization of barbed ends and the Arp2/3 complex at both the tip and the base of the spine [51] suggests that actin polymerization is active at both locations, where they generate antiparallel arrays of actin filaments. Also, the localized activity of ADF/cofilin, which severs actin filaments, could generate new barbed-ends within the spine.

The role of adhesion in initial spinogenesis may also parallel its role in migrating cells. As motile cells extend new protrusions, they attach to the substratum *via* small adhesions that form within the protrusion. These adhesions provide traction through their linkage to the actin cytoskeleton (Fig. **1B**, *left*) [59], and they accumulate regulatory proteins that control actin polymerization, reorganization and adhesive strength. A complex network of signaling pathways originating in adhesions converge on Rac [60-63], which triggers actin polymerization through binding to downstream effectors, e.g., the WAVE/Scar family, which in turn activate the Arp2/3 complex (Fig. **2**) [64]. Other adhesion-related signaling proteins, such as FAK (Focal Adhesion Kinase) are also essential for dendritic spine formation [65]. FAK also

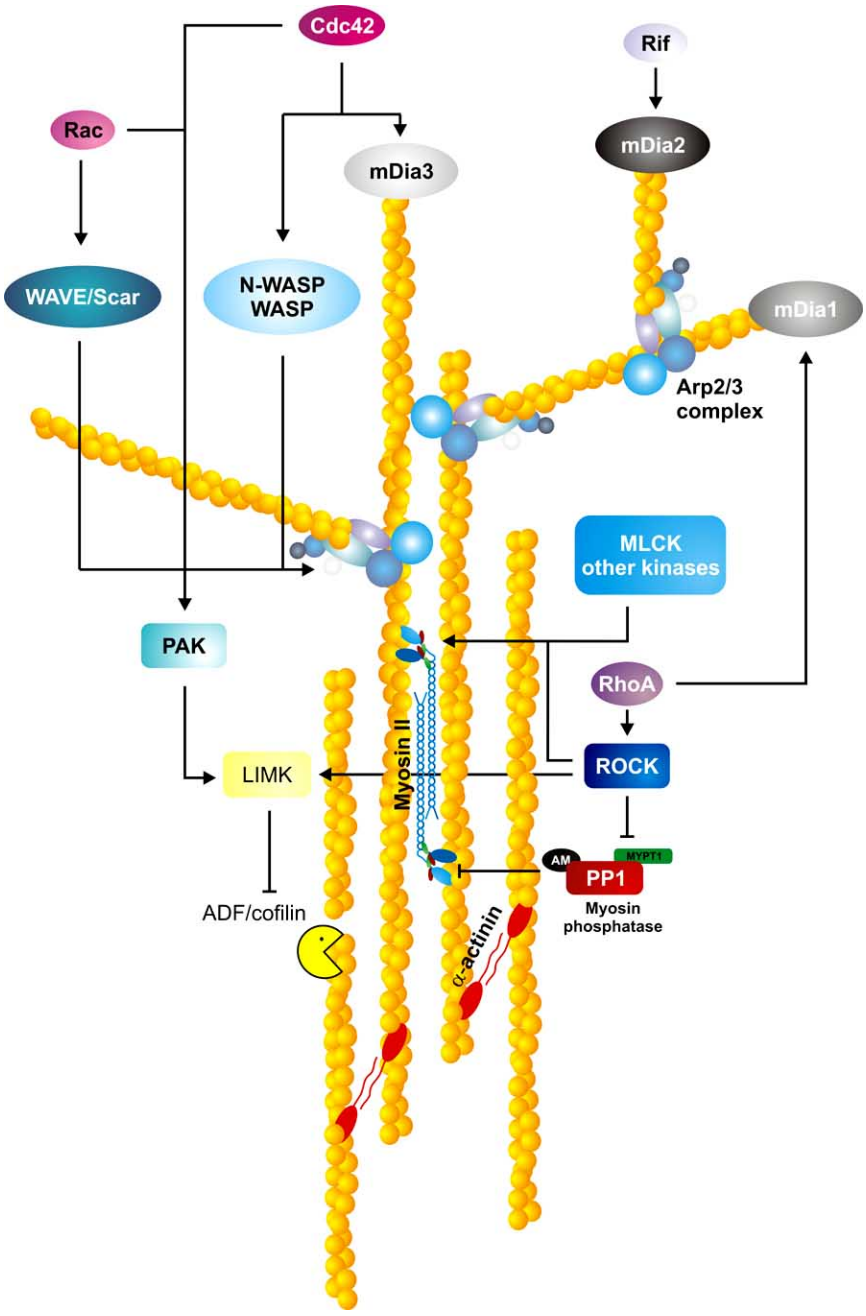

Fig. (2). Mechanisms of actin regulation in dendritic spines.
The cartoon depicts the main molecules that control actin polymerization and organization in dendritic spines. Actin polymers are represented as coiled chains of yellow beads. The regulatory molecules include: 1) the Arp2/3 complex, which binds to the side of a pre-existing actin filament and promotes formation of a branched actin filament. Arp2/3 is activated by N-WASP/WASP under the control of the small GTPase Cdc42, and WAVE, which is activated by the small GTPase Rac. 2) formins, including mDia1 (activated by the small GTPase RhoA), mDia2 (small GTPase Rif) and mDia3 (Cdc42), which bind to the barbed (polymerizing) end of the actin filament and promote processive incorporation of actin monomers. 3) actin cross-linkers such as α-actinin and myosin II. Myosin II activity and assembly are controlled through phosphorylation. Kinases like ROCK and MLCK can activate myosin II. ROCK is controlled by RhoA, and also inhibits the phosphatase that dephosphorylates myosin II. Finally, ADF/ cofilin (yellow pac-man) severs actin filaments. It is inhibited by LIMK phosphorylation, which in turn is activated by phosphorylation *via* ROCK and PAK, which is regulated by Rac and Cdc42.

modulates the function of the actin cross-linker α-actinin [66], suggesting that this pathway might be important in actin bundling during initial spinogenesis.

In addition to its role in generating signals that regulate actin, adhesion also fulfills an exploratory role. Migrating cells use filopodia and nascent adhesions as small chemo- and mechano-sensitive devices to guide cell migration [67, 68]. Similarly, immature dendritic spines seek presynaptic terminals to undergo stabilization. This process is likely to involve chemotactic, chemorepellent and/or mechanotactic

signals emanating from the pre-synaptic terminal or the microenvironment of the protrusion, which is stabilized by adhesion to the pre-synaptic terminal. Once contacted, actin organization, contraction, and adhesion mediated signaling could drive subsequent spine maturation as these adhesions do in other cell types.

ADHESIVE SIGNALING AND ACTIN DEPOLYMERIZATION REGULATE ADHESION AND TURNOVER, AND DENDRITIC SPINE REMOVAL

More than a hundred years ago, Ramon y Cajal reported that the processes of the pyramidal neurons of newborns contained more protrusions than later in development [69]. This early observation suggested that synaptic connectivity is fine-tuned through the disassembly of unused or defective spines [70]. Later studies confirmed that the initial proliferation of spines is followed by a marked decrease in their number at later developmental stages [71, 72].

In one model, the removal of immature spine precursors is caused by the lack of contact and/or pre-synaptic input; accordingly, those precursors not making contact with pre-synaptic structures would be reabsorbed into the dendritic shaft, whereas those that establish contact with pre-synaptic terminals would evolve into mature spines. A separate population of mature spines is selectively eliminated during functional rewiring of neural circuits in response to sensory experience [73].

Turnover of immature spine precursors or selective elimination of mature, innervated spines is probably linked to actin filament disassembly, or a contraction-induced retraction of actin filaments back into the dendritic shaft. Filament disassembly is more likely. Contraction requires activation of proteins like non-muscle myosin II (NM II), and the present evidence suggests that NM II activation induces maturation of precursors into dendritic spines (see below) [74, 75]. However, some synapses can survive active actin disassembly; for example, actin depolymerization induces a significant, but not complete elimination of synapses when cells are treated with the actin polymerization inhibitor latrunculin A [76]. Actin filament disassembly can occur *via* two complementary mechanisms: an increase in barbed end capping, which would block actin polymerization, and actin depolymerization, mediated by filament-severing proteins, such as gelsolin or ADF/cofilin [50]. Gelsolin is a dual-function, calcium-sensitive actin filament-severing protein that also caps the newly formed barbed ends, impeding further polymerization [77]. Gelsolin-null neurons contain numerous spines that are not stabilized by synaptic stimulation, implicating gelsolin in activity-induced spine maturation and removal of unstable, immature precursors [78].

The other severing protein, ADF/cofilin, is required for actin depolymerization in protrusions of migrating cells [79]. Expression of an active mutant of cofilin, S3A, induces accumulation of branched actin, suggesting that the increased treadmilling of actin monomers and creation of new barbed ends supersede its filament-severing activity [80]. In hippocampal neurons, cofilin activity is required for the spine shrinkage observed during long-term depression (LTD), which is the activity-dependent elimination of synaptic connections [81]. Consistently, RNAi-mediated cofilin inhibition induced longer dendritic protrusions [51]. Expression of a constitutively active cofilin mutant significantly decreased the area of the spine head, but did not lead to its disappearance [82]. These results can be explained by the dual function of cofilin. On one hand, it severs actin filaments; but it also provides the actin monomers that are recycled into *de novo* polymerization at the barbed end, *via* treadmilling [50, 79]. Therefore, the activation and inactivation of cofilin is a key regulatory step in maintaining an adequate balance of actin depolymerization and polymerization and acts in concert with capping factors. The key role of cofilin in actin function is further supported by studies of its regulation. LIMK is activated by Rho-associated kinase (ROCK) and p21-associated kinase (PAK) [83, 84], which are under the control of the small GTPases RhoA and Rac/Cdc42, respectively (Fig. **2**]. LIMK phosphorylates cofilin and inhibits its binding to actin filaments, thus preventing filament severing [85, 86]. Consistent with this, altered cofilin phosphorylation, abnormal spine morphology and synaptic function are observed in LIMK1-deficient mice [87, 88]. Interestingly, a loss-of-function mutation in LIMK1 is implicated in the cognitive deficit associated with Williams' syndrome [89].

Adhesion formation in protrusions is linked to polymerized actin; adhesions disassemble or mature when and where branched actin undergoes depolymerization or reorganization, respectively (Fig. **1B**, *middle*) [90, 91]. This constitutes a putative feedback loop: polymerized actin provides a physical scaffold for the formation of adhesions, which in turn generate Rac-dependent signals that promote actin polymerization and inhibit filament severing. In a similar manner, filament disassembly in immature spine precursors would disrupt adhesion, also suggesting that adhesion to the pre-synaptic terminal may induce spine maturation by inhibiting filament disassembly.

In summary, the removal of immature spine precursors during development involves actin filament disassembly, presumably through a combination of actin depolymerization and inhibition of actin polymerization; the resulting actin monomers treadmill and are used to generate new dendritic precursors during the maturation of a subpopulation of dendritic spines.

MYOSIN II IN ACTIN ORGANIZATION DURING DENDRITIC SPINE MATURATION

Maturing spines undergo dramatic morphological changes, including shortening, formation of a neck, widening of the head and organization of the post-synaptic density (PSD), which is an accumulation of synaptic and adhesion receptors, signaling adaptors and cytoskeletal proteins [5, 6, 92, 93]. The PSD itself undergoes rapid morphology fluctuations in response to synaptic activity, and also widens concomitantly with expansion of the spine head during its maturation [94].

Non-muscle myosin II (NM II) is a key contractile protein that organizes and contracts actin in migrating cells. It regulates front-back polarity and modulates adhesion organization, inducing maturation [95, 96]. It is likely that it plays an analogous role in spine and PSD organization.

NM II is a hexameric complex formed by two heavy chains (NMHC-II), two regulatory light chains (RLC), and two essential light chains (ELC). NM II binds to actin filaments and promotes their bundling; it also mediates filament

contraction through ATP hydrolysis. The three isoforms of NMHC-II, NMHC II-A, II-B and II-C, are encoded by three genes, *Myh9*, *Myh10* and *Myh14*, respectively [97]. Of these, NMHC II-B, is the most prominently expressed in neurons [98, 99]. It plays a pivotal role in growth cone dynamics and in the development of the CNS. Mice ablated for NM II-B exhibit profound developmental defects, including hydrocephalus [100]. NM II-B down-regulation inhibits dendritic spine maturation. RNAi targeting of NM II-B in *in vitro* cultured hippocampal neurons or acute treatment with the NM II inhibitor blebbistatin drastically reduced the number of mature spines and synapses [74, 75].

Spine Shortening: Role of Myosin II

Spine shortening occurs concomitant with a dramatic reorganization of the actin and is likely mediated by NM II, which induces actin contraction and reorganization. NM II activation inhibits protrusion in motile cells and causes retraction of the leading edge. It also promotes adhesion maturation and actin filament thickening (Fig. **1B**, *right*) [91, 95]. In epithelial cells, NM II promotes the consolidation of the cell-cell junction, by generating contractile actin bundles parallel to the plasma membrane, increasing the contact surface between cells (contact compaction), and inducing cadherin clustering [101, 102]

NM II-B-mediated spine shortening is likely related to its contractile activity, exerting force that would pull on the actin filaments tethered to the tip of the spine or the PSD, causing spine retraction and compaction of the material inside the spine (Fig. **3**). In addition, data from epithelial cell studies suggest that NM II-driven contraction may enhance adhesive strength between pre- and post-synaptic terminals by promoting clustering of adhesion receptors, e.g. cadherins [101].

NM II function is regulated by phosphorylation of the RLC; therefore phosphorylated RLC is a marker for active NM II. Phosphorylated RLC localizes to dendritic spines, and a phosphomimetic form of RLC induces dendritic spine formation [75]. In addition, adhesion and LTP induction activate multiple signaling pathways, including RhoA/ROCK [103-105], which increase the level of RLC phosphorylation in fibroblasts [106].

Formation of a Spine Neck

The spine neck is thought to be an important geometrical feature of mature spines by serving to confine neurotransmission to the spine and blocking diffusion of the signal into the shaft and adjacent spines [5].

NM II participates in actin bundles of different geometries. In migrating cells, it mainly forms thick linear actomyosin bundles [107]; but in dividing cells it is involved in the formation of the contractile ring during cytokinesis [108]. Also, NM II activation at cell-cell junctions promotes the compaction of the contact [101]. Interestingly, similar phenomena are observed at a multi-cellular level, in which coordinated cohorts of cells integrate their contractile activities: an outstanding example is the "purse-string" model of epithelial dorsal closure, which is driven by NM II activation [109].

Analogously, NM II could mediate the formation of a small contractile ring-like structure that constricts the contact area of the spine with the dendritic shaft. Alternatively, the spine neck can be comprised of linear actin bundles generated during the formation of the immature spine precursor that does not undergo complete retraction. Both these possibilities are shown in Fig. (**3**).

Spine Head Expansion

During maturation, the tip of the dendritic spine expands to provide a larger surface area of interaction with the presynaptic terminal; this is a hallmark of activity-induced plasticity. There are at least two coordinated mechanisms for controlling spine head expansion. One is an increase in membrane surface area, which is mediated by increased targeted delivery of vesicles under the control Rab/Arf family of GTPases like Arf6 [15]. The other is the reorganization of the actin cytoskeleton, in which branched actin filaments replace the linear arrays observed in immature spine precursors. In this manner, actin branching at the tip of the spine potentially sustains the increase in volume and surface area, much like the extension of a protrusion in migrating cells.

The morphological changes that take place during spine maturation can be integrated into a model in which the local activation of Rac and Arp2/3 (and local inactivation of RhoA) at the tip of the spine supports the formation of a branched actin network that expands the head. In migrating cells, there is evidence that the activation of Rac and Rho is spatially and temporally segregated. Rac is active at the protruding edges of migrating cells, where it triggers dendritic actin formation [61, 90, 91]. In addition, Rac signaling suppresses RhoA activation [110]. On the other hand, RhoA is more active in the more posterior part of the protrusion and the center of the cell, where it induces thick actomyosin filaments, stable adhesions and inhibition of Rac activation [107, 110].

Similarly, Rac activation closer to the synaptic cleft would promote branched actin to widen the spine head (Fig. **3**, insert), whereas activation of RhoA closer to the dendrite shaft would promote bundling of actin tethered to the PSD, possibly by forming an actomyosin cup, or pedestal (Fig. **3**). Supporting this model, it has been proposed that Arf6, which regulates vesicle trafficking and provides membrane for membrane expansion during spine widening, creates sites for targeting of Rac to the membrane [111].

CONCLUDING REMARKS

In this review, we have used insight from studies on adhesion and protrusion in migrating cells as a model for dendritic spine and PSD organization. In migrating cells, protrusions form using two actin regulators, Arp2/3 and formins, and adhesion maturation is determined by the organization of the actin cytoskeleton. Both of these are regulated by signals emanating from adhesions. Finally, the formation of epithelial adherens junctions is also mediated by actin and is accompanied by the cessation of Arp2/3 activity and stimulation of actomyosin contraction as the junctions form. Emerging evidence suggests that dendritic spine maturation is similarly mediated by actin organization and driven by contact with the pre-synaptic terminal. Thus, actin polymerization and the organization of the actin cytoskeleton remains a cen-

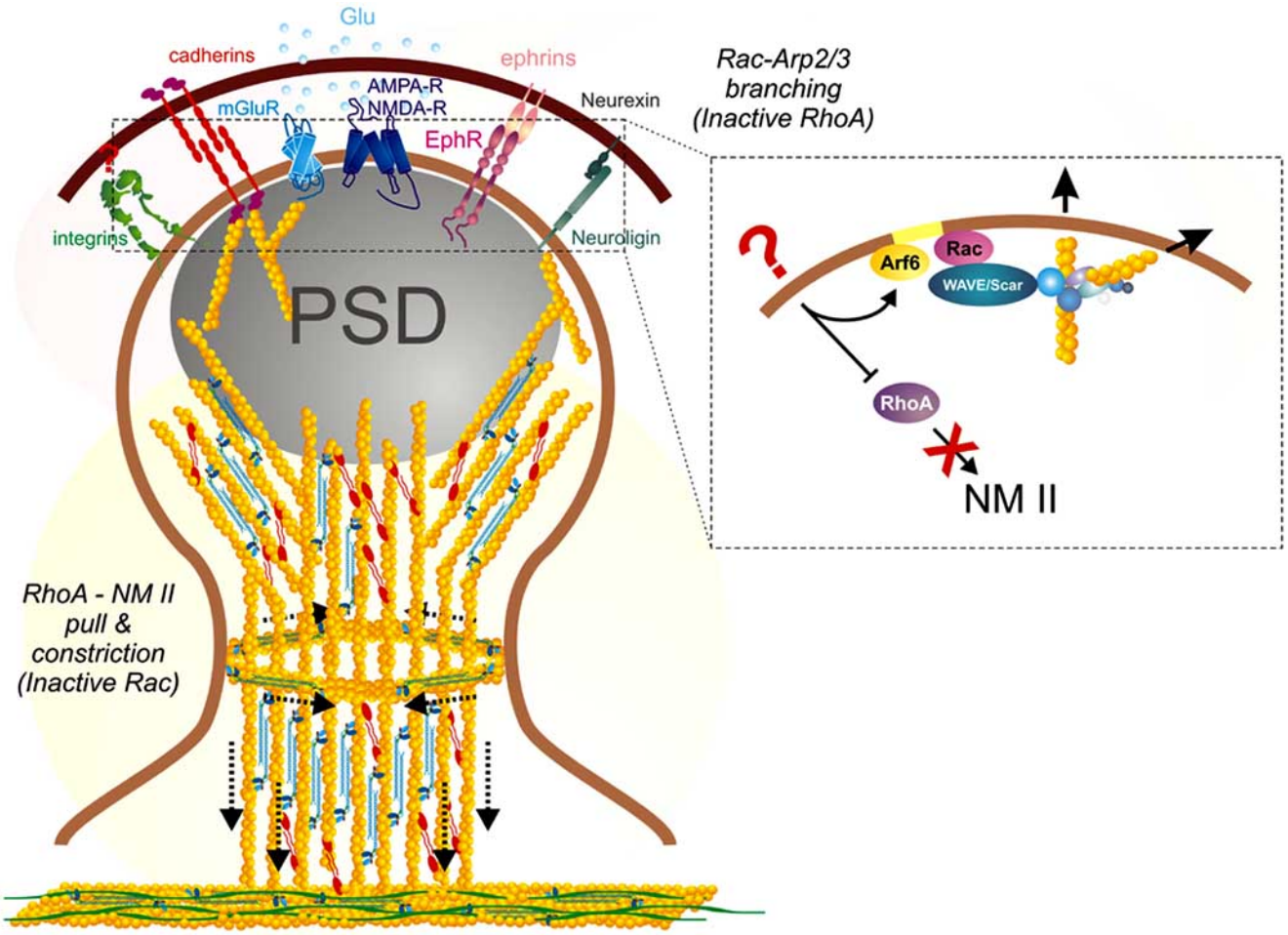

Fig. (3). Hypothetical model of dendritic spine organization.
The cartoon represents activation of NM II (blue) at the base of the spine, which triggers retraction of the spine by pulling the actin filaments tethered to the PSD, and/or constriction of the spine neck. These movements are represented by dashed arrows. Other cross-linkers, e.g., α–actinin (shown in red), also mediate actin bundling in the spine. NM II is also found in the PSD, and controls its integrity. At the tip of the spine, activation of adhesive molecules (integrins, cadherins, neurexins/ neuroligins, Eph receptors and others) or synaptic receptors (metabotropic Glu and AMPA/NMDA receptors) associated to the PSD trigger the local activation of Rac and branched actin growth by the Arp2/3 complex to support spine widening, as well as RhoA inactivation. Rac activation could be supported by the translocation of membrane domains (shown in yellow) required for spine membrane expansion under the control of the small GTPase Arf6.

terpiece of these processes, which share many common regulatory elements.

Despite the different repertoire of receptors between fibroblasts and neurons, most of the signaling pathways originate with membrane receptors and converge on the regulation of adhesion and the actin cytoskeleton through Rho GTPases. The regulators that control actin polymerization and filament disassembly downstream of the GTPases are also the same (formins and the Arp2/3 complex, and cofilin, respectively). Finally, actin cross-linkers and contractile proteins, like NM II, play similar roles in the two processes, facilitating actin reorganization and reshaping of the stable structure through actin bundling and/or contraction.

The discovery that some mental retardations are accompanied by altered Rho GTPase regulation and abnormal morphology of dendritic spines highlights the importance of understanding how the actin cytoskeleton regulates the morphological changes that dendritic spines undergo upon activation. It also points to therapeutic targets using gene-based therapy and interventions directed at neuron specific isoforms of key adhesion and actin related molecules for the treatment of diseases with cognitive decline, such as Alzheimer's or Parkison's disease, senile dementia, or congenital and non-syndromic mental retardation.

While some clear parallels exist, many aspects of dendritic spine and PSD development remain unstudied; hopefully this discussion will provide one blueprint for a useful approach.

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