Role of NCAM in Spine Dynamics and Synaptogenesis

D. Muller, P. Mendez, M. DeRoo, P. Klauser, S. Steen, and L. Poglia

Introduction

Synapse formation and remodeling are one of the key mechanisms that regulate the development and function of synaptic networks. Recent evidence from in vivo imaging experiments has shown that synapse formation and elimination represent a major process contributing to the build-up and shape of brain networks [1–3]. Interestingly, this process remains active throughout life, although at a significantly lower level [4], it can be reactivated following injury or brain lesions [5], and it possibly underlies or is associated with cognitive functions and learning mechanisms [6]. Understanding how two partners establish and maintain a stable and functional synaptic connection between them has therefore become a major issue. The importance of this question is further strengthened by accumulating evidence suggesting that many of the cognitive or psychiatric disorders that affect human behavior, such as autism, mental retardation, or even schizophrenia, could involve alterations of the mechanisms regulating synapse formation in the brain [7].

At the molecular level, the number of proteins, enzymes, or messengers identified as contributing to the formation of a functional synapse has exploded over the last years. On the postsynaptic side, more than hundred molecules, including receptors, scaffold proteins, protein kinases, adhesion molecules, cytoskeletal components, or intracellular messengers, appear to participate one way or the other in the organization, function, or plasticity of the postsynaptic density. Similarly, the complexity and properties of the machinery responsible for transmitter release and the fusion of docked synaptic vesicles become progressively better and better elucidated. The challenge to establish a synaptic contact is to coordinate the interactions required between the two partners for the formation and differentiation of these specialized structures. Much recent evidence indicates that adhesion molecules

D. Muller (✉)
Department of Neuroscience, University of Geneva Medical Center, 1,
M. Servet, 1211, Geneva 4, Switzerland
e-mail: Dominique.Muller@medecine.unige.ch

V. Berezin (ed.), *Structure and Function of the Neural Cell Adhesion Molecule NCAM*, *Advances in Experimental Medicine and Biology* 663, DOI 10.1007/978-1-4419-1170-4_16,

play an important role in these mechanisms and analyses of their contribution to synaptogenesis have been the subject of several recent reviews [8–12]. Here, we focus on NCAM and discuss recent advances in our understanding of the mechanisms of excitatory synapse formation, as revealed by repetitive confocal imaging, and the role of NCAM in this process.

Adhesion Molecules and Synaptogenesis

Formation of a synaptic contact requires a complex coordinated interaction between pre- and postsynaptic structures leading to the differentiation of highly specialized and regulated structures [13]. At the presynaptic level, synaptic vesicles have to accumulate in the region of contact and become organized in a meshwork able to sustain vesicle fusion at high speed. This implicates various protein complexes, but also channels and signaling pathways. Postsynaptically, at excitatory synapses, a major step involves the formation of a protrusion, the differentiation into a dendritic spine and the expression of a postsynaptic density facing the release site. More than one hundred different proteins organized into complexes have been identified to contribute to these events [7]. In general, pre- and postsynaptic protein complexes are present in neurons and axons before the contact is made and they accumulate in mobile transport packets that are recruited to sites of contact [14, 15]. Once expressed at the synapse, these proteins still undergo a continuous turnover that may even be quite fast as demonstrated for example for glutamate receptors [16] or PSD-95 [17].

Several steps in the process of synapse formation can now be identified and most of these have been found in recent years to involve a contribution of adhesion molecules. A first step at excitatory synapses is to form a contact with an adequate partner on a dendritic spine. Early work in hippocampal neurons has suggested that this process could be initiated by the postsynaptic partner through the growth of a highly motile filopodium, which then stabilizes as a dendritic spine when it gets in contact with the right axon [18]. More recent 2-photon video time lapse analyzes have however shown that dendritic spines may also simply pop out from the dendrite within a few minutes without initial formation of a filopodium [19, 20]. It remains unclear in this case whether a presynaptic partner is already present to initiate the growth of the spine. However, as will be discussed later, the observation that newly formed spines usually lack postsynaptic densities until a few hours after appearance suggests that they might initially grow without identified partner [21, 22]. Expression of adhesion molecules on the growing structures could be important during this step for the matching of pre- and postsynaptic components. Molecules such as cadherins, nectins or neurofascin have been proposed to exhibit such recognition functions, although the evidence remains sparse for excitatory synapses [23]. A second important step is the differentiation of pre- and postsynaptic structures in order to make functional and morphologically mature synapses. This in particular implies the accumulation of release-competent vesicles and proper

receptors at the site of contact. Neurexin and neuroligins have been shown in this respect to be unique among synaptogenetic factors because each of them has the capacity to induce the differentiation of a complementary hemi-synapse on the corresponding partner [10]. Neuroligin expression can induce presynaptic differentiation of nearby axons, while neurexin stimulates the aggregation of postsynaptic components such as receptors or PSD proteins. While the example of neuroligins is certainly the most impressive, it is certainly not the only one and, as will be discussed later, the other adhesion molecules and in particular NCAM and PSA-NCAM might also contribute to bring specific molecular components to the synapse. Finally, a last important step might be the long-term stabilization of the newly formed synapse. Evidence from recent in vivo and in vitro work does indeed indicate that while some spines are definitely highly stables, there are others that are essentially transient [2]. It is very likely that this process could be directly related to activity or even also to synaptic plasticity. Molecules such as BDNF, but also ephrins, cadherins, PSD-95 or even NCAM have been shown to participate to forms of activity-dependent plasticity and could play a role in these aspects of synapse maturation [11, 24–26]. Together, these results point clearly to the notion that the same adhesion molecule may in fact contribute to various aspects of the mechanisms that are required to form a functional and mature synapse and that several adhesion molecules may very well have overlapping functions.

Role of NCAM and PSA-NCAM in Synaptic Function and Plasticity

An important step in our understanding of NCAM function in the brain has come from studies of transgenic mice deficient in NCAM, which showed deficits in learning and memory [27]. Consistent with this observation, numerous subsequent studies confirmed a role of NCAM and more specifically PSA-NCAM in mechanisms of synaptic plasticity such as long-term potentiation, an increase in synaptic strength considered as one of the physiological bases for information storage by a synaptic network. In the CA1 region of the hippocampus, interference with NCAM through specific antibodies or suppression of PSA from NCAM by the enzyme Endo-N or simply suppression of NCAM all resulted in a reduced or even abolished LTP [28–30]. This effect occurred without noticeable changes in the properties of synaptic transmission. Similar results were then further observed in conditional NCAM knockout mice [31], an experiment which allowed to exclude possibilities of developmental anomalies as a possible cause of the defect. Additionally, transgenic mice deficient in the sialyltransferase enzyme responsible for adding PSA to NCAM in adult mice (ST8SialV/PST-1) also showed impaired LTP and they were also selectively deficient in spatial learning tasks and contextual fear conditioning, which depend upon hippocampal plasticity [32]. Interestingly however, while NCAM knockout mice are impaired in cued fear conditioning experiments, a behavior, which depends on synaptic plasticity induced in the amygdala, ST8SialV/PST-1

deficient mice did not show any deficit, suggesting that different brain regions may differentially require PSA or NCAM for plasticity [33] A. Consistent with this interpretation, LTP experiments also suggest that the relative contribution of PSA-NCAM or NCAM to synaptic plasticity may vary as a function of the region analyzed. In the CA3 region of the hippocampus, where LTP properties clearly differ from those expressed in the CA1 region, but also in the dentate gyrus, NCAM seems to be more important than PSA-NCAM since LTP is present in ST8SialV/PST-1 deficient mice, but abolished in NCAM deficient mice [34]. Although it is therefore likely that NCAM and PSA-NCAM may play different roles at specific synapses, these results clearly point to the important regulatory function of PSA-NCAM and NCAM in the control of synaptic strength.

The mechanisms through which this might be achieved remain however unclear. Several hypotheses have been considered which mainly go along two main, non-exclusive lines of thinking. On the one hand, the ratio of PSA-NCAM to NCAM could modulate the adhesion properties between pre- and postsynaptic membrane and thus allow dynamic changes to take place [35]. In the case of LTP, it is now well accepted that a major contribution to the synaptic enhancement involves the expression of new AMPA receptors [16]. Associated to this, LTP in the CA1 region probably requires a remodeling of the synaptic structures, which includes a reorganization of the postsynaptic density and an enlargement of the postsynaptic spine head [36–39]. Variations in the level of expression of PSA-NCAM and changes in the ratio of PSA-NCAM to NCAM, as it occurs under conditions of increased activity [29, 40], could provide the structural plasticity required for these events. A second possibility however is that the ratio of PSA-NCAM to NCAM modifies homo- and heterophillic interactions and thereby affects NCAM signaling [41–43]. Evidence from a number of studies indicates that NCAM may indeed participate in a broad range of biological processes through activation or regulation of various molecular pathways [43–45]. These include growth factor receptors such as FGF receptors, which undergo dimerization and autophosphorylation upon NCAM homophillic binding [46, 47] [43, 48], but also neurotrophin receptors and in particular the BDNF TrkB or P75 receptors [49, 50], which play an important role for properties of synaptic plasticity [51], or direct interactions with either glutamate receptors, cytoskeletal proteins such as spectrin, or the Fyn/MAP kinase cascade [44] which are also critical for the remodeling of postsynaptic structures. Taken together, these results suggest that the ratio of PSA-NCAM to NCAM could represent an interesting system for setting the level of signaling in pathways involved in properties of synaptic plasticity, thus allowing to link activity with capacity for adaptation.

Role of PSA-NCAM in Synaptogenesis

In addition to their roles in regulation of the function and strength of synapses, several lines of evidence suggest that NCAM and PSA-NCAM could also participate in mechanisms of synapse formation. In cell cultures, NCAM accumulate rapidly, within minutes, at sites of contact formation in nascent synapses [52].

Because NCAM binds to spectrin coated trans-Golgi derived organelles, it was proposed that NCAM could work as a trap to allow accumulation of postsynaptic proteins and components necessary for the formation of the synaptic contact [53]. Consistent with this idea, the same authors recently showed that disruption of NCAM-spectrin complexes results in a decrease in the size of postsynaptic densities and a reduced targeting of proteins such as spectrin, NMDA receptors and CaMKIIa to the synapse [54]. They propose that this mechanism could be important not only for the formation of a nascent synapse, but also for activity-dependent plasticity, since, in NCAM deficient neurons, recruitment of CaMKIIa to the synapse by NMDA receptor activation is prevented [54]. Together, this very elegant work clearly emphasizes the important role of NCAM as a functional building block of the synapse. How is it then that NCAM knockout mice do not appear to show deficiencies in synapse number or even in synapse morphology? One possibility clearly is that some degree of overlap or compensation is made possible between the different adhesion molecules expressed at synapses. If neurons from NCAM knockout mice are cultivated together with neurons expressing NCAM and PSA-NCAM, they do show a reduced number of synapses and reduced excitatory activity, indicating preferential formation of synapses with NCAM expressing cells [55]. Similarly, removal of PSA from NCAM also abolished preferential formation of synapses on NCAM expressing cells, pointing to a role of PSA in this process [56]. Another study also indicates that an NCAM mimetic peptide promotes synapse formation [57]. Further analyzes showed that this synaptogenetic effect of NCAM probably involved an interaction between PSA-NCAM and a heparan sulfate proteoglycan and signaling through the FGF receptor [56]. Interference with this mechanism prevented both NCAM-driven synaptogenesis and activity-dependent structural remodeling of activated synapses as indicated by the absence of formation of perforated synapses upon induction of LTP [56, 57]. Together, these results are consistent with the idea that PSA-NCAM, by interfering with NCAM homophilic binding, plays a permissive role that promotes growth and reorganization of the postsynaptic structure such as is required both for the formation of a new contact and for activity-dependent plasticity. A further argument supporting a role for NCAM and more specifically PSA-NCAM in synaptogenesis is the very recent finding that elimination of PSA from NCAM results in the developing visual cortex in an early maturation of perisomatic GABAergic innervation by basket interneurons. In this example, PSA-NCAM also modulates synaptogenesis, but probably through a presynaptic expression, which prevents interneuron axons to form synapses until visual activity sets the stage for ocular dominance plasticity [58]. It is not unlikely therefore that NCAM and PSA-NCAM may have different functional and regulatory role at different synapses.

Dynamic Aspect of Spine Turnover and Synapse Formation

An important new aspect of the mechanisms of synaptic plasticity was revealed by experiments of repetitive confocal imaging examining spine remodeling in living mice. Studies by several groups provided evidence that dendritic spines undergo some

sort of turnover and that there exist a process of continuous growth and elimination of spines [1–4, 59]. Although there has been some debate about the magnitude of this turnover and the technological approach used to image living neurons in the cortex [60], current evidence indicates that spine turnover varies greatly during development, affecting as many as 10%–15% of spines per 24 h in very young animals [2, 3]. Later on, spines become progressively more and more stable with probably only a few percent of spines undergoing replacement in adult tissue [4]. Furthermore, this process appears to vary in different cortical regions and even show some cell-type specificity [6]. Finally, more and more data suggest that this spine turnover process is also affected by sensory activity and may again become very prominent in regions submitted to a lesion [5, 6].

To be able to better analyze the mechanisms regulating spine turnover, we recently developed an in vitro approach based on the use of hippocampal slices cultures. Through repetitive imaging of the same cells over several days (Fig. 1), we found that dendritic spines showed a high level of turnover, affecting about 20% of all spines over a 24 h period in 15 days old cultures, but only about 10% after 25 days in vitro [21]. These values, which are quite close to those obtained in very young animals, suggest that spine turnover retains similar properties in vitro and in vivo and particularly its developmental dependency. Another interesting aspect was that this rate of spine turnover is actually underestimated by the use of long observation intervals. It turns out that in 15 DIV slice cultures, the basal rate of protrusion formation reaches values in the order of 2% of all spines per hour, which represents hundreds of new protrusions per day and per neuron. The reason why spine density remains nevertheless stable is that most of these new protrusions do in fact disappear fairly quickly and only a small proportion of them become stable spine synapses. Interestingly, while filopodia were proposed in previous studies to be precursors of spine synapses [18], we found that they only exceptionally lead to the formation of stable spine synapses (Fig. 1d), a result consistent with other in vivo data [3]. Protrusions are thus generated at a high rate, but only a fraction of them become finally stable spines (Fig. 1e). We also found that this required a process of maturation that lasted about 24 h. During this period, new spines usually grew in size (Fig. 1f) and started to express a PSD (Figs. 1g, h). It took however about 5 h in our in vitro experiments to start detecting PSDs on newly formed spines (Fig. 1i), a result that is consistent with results obtained through EM reconstruction of newly formed spines [22]. Interestingly, expression of this PSD was activity-dependent in hippocampal slice cultures, since blockade of AMPA and NMDA receptors prevented its expression and reduced the probability of the spine to be stabilized.

exceptionally lead to the formation of a stable spine (*black columns*). (**e**) Stability of newly formed spines (*black columns*) and of their transformation into stubby spines (*grey columns*). (**f**) Progressive enlargement of newly formed spines (bar: 0.5 µm). (**g**) Illustration of a newly formed spine (age < 5 h; *arrow head*, *middle panel*), which do not express PSD-95-DsRed2 (*arrow head*, *right panel*; bar: 0.5 µm). (**h**) Electron microscopic illustration of a spine devoid of PSD and presynaptic partner (bar: 0.5 µm). (**i**) Time course of Psd-95-DsRed2 expression in newly formed spines

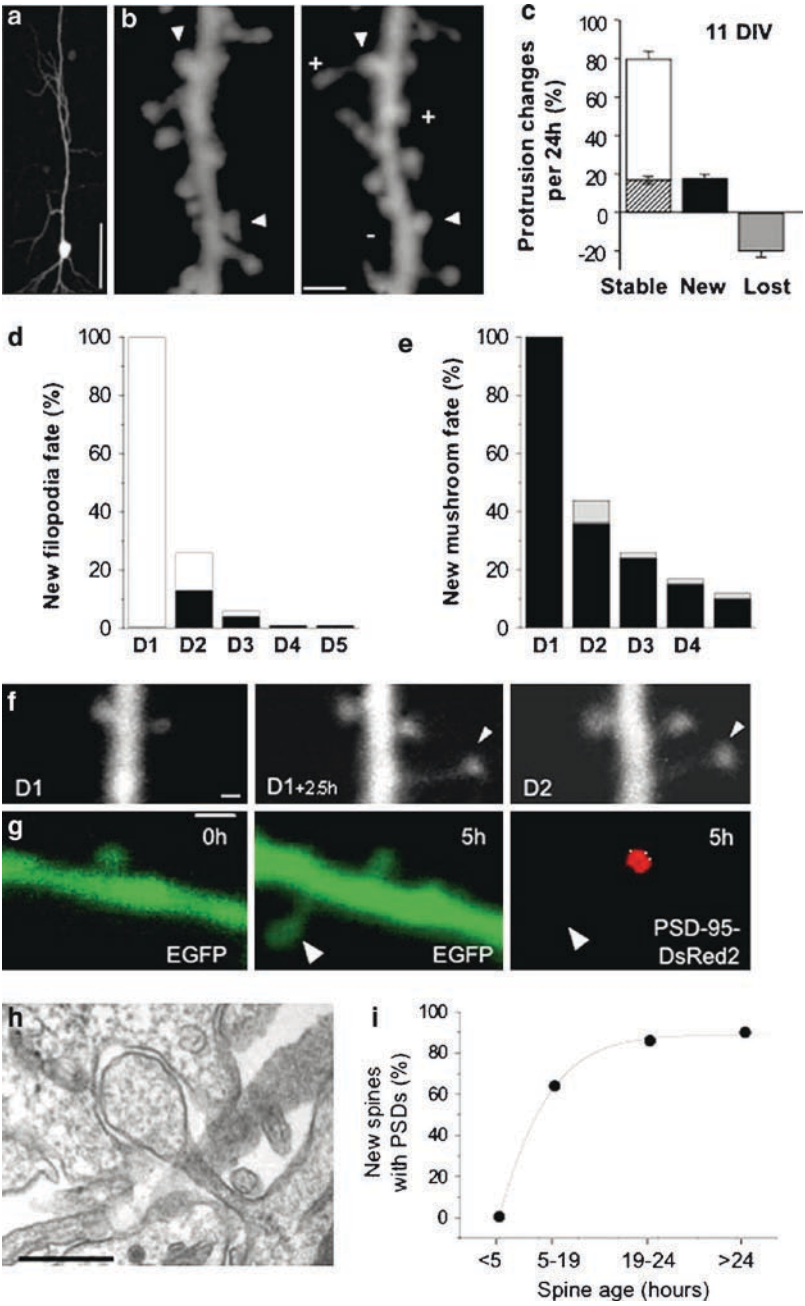

Fig. 1 Spine dynamics in hippocampal organotypic slice cultures. (**a**) EGFP transfected CA1 pyramidal neuron (bar: 100 μm). (**b**) Repetitive imaging of a dendritic segment at 24 h interval reveals the occurrence of new and lost protrusions (bar: 1 μm). (**c**) Summary of the proportion of stable spines (*open column*), which include spines exhibiting changes in morphology (*dashed column*), of newly formed (*black column*) and disappearing (*gray column*) spines. (**d**) Stability over 5 days of newly formed filopodia. Note that most of them disappear within 1–2 days and only

Taken together, these experiment suggest a model in which development of synaptic networks proceed through an extensive, non specific growth of dendritic protrusions, followed by the stabilization of a small number of spines. This stabilization process involves the expression of a PSD through mechanisms that appear to be driven by synaptic activity [21].

Regulation of Spine Stability and Function by PSA-NCAM/NCAM Ratio

The capacity to now examine on a longer time scale the behavior of specific synapses makes it possible to investigate new aspects of the role of adhesion molecules and in particular how they may regulate some of the initial or activity-dependent steps of synaptogenesis. In the case of NCAM and PSA-NCAM, preliminary experiment in this laboratory (Mendez et al., unpublished) suggest that upon removal of PSA from NCAM by the enzyme Endo-N, the rate of spine formation is reduced and more importantly the stability of spines is decreased. At the presynaptic level, other studies also suggest that the lack of NCAM function leads to a reduction of the stability of synaptic contacts [61]. These observations would actually be consistent with the different results reported above and in particular with the hypothesis that one important function of NCAM might be to help build the architecture of the postsynaptic density, probably by interacting with molecules such as spectrin and indirectly with constituents of the PSD such as PSD-95 or CaMKII [54]. One might also hypothesize that NCAM molecules through homophillic cis- or trans-interactions tend to form clusters that could give some rigidity to the synapse. It is interesting in this respect that electron microscopic analyzes of high pressure quickly frozen hippocampal synapses revealed the presence of a protein network within the synaptic cleft that shows the periodic arrangement expected from a zipper organization of NCAM or cadherin molecules [62]. However, in order to remodel or enlarge the PSD, as it happens during the initial formation of the PSD in a new synapse or following activity-induced plasticity, one might assume that less rigid interactions and additional signaling promoting turnover of PSD components would be important. These changes could be promoted by increasing the ratio of PSA-NCAM to NCAM, a change that can be mediated by neuronal and synaptic activity [29, 40]. The increase in PSA-NCAM could at the same time disrupt or affect the zipper organization of NCAM and, through interactions with heparan sulfate proteoglycans and FGF receptors or even other neurotrophin receptors, activate the signaling pathways regulating receptor and other PSD protein dynamics [43, 57]. An increased ratio of PSA-NCAM to NCAM could represent therefore an important regulator or trigger of plasticity. Accordingly, suppression of PSA from NCAM by the enzyme Endo-N would be expected to affect several important properties of excitatory synapses: this should interfere with the process of synapse formation by limiting the initial growth of the PSD in newly formed spines and thus reduce the probability for new protrusions to become stabilized, accounting therefore for the decreased

number of synapses detected on young NCAM knockout neurons [56]; the same mechanism would however also account for the role of PSA-NCAM in LTP, since the remodeling of the postsynaptic density and namely the expression of new receptors and other PSD constituents such as PSD-95 or CaMKII could also require to loosen the zipper organization of adhesion molecules at the synapse. Elimination of PSA from NCAM has indeed been shown to prevent the formation of perforated synapses upon application of high frequency stimulation [56]. Finally, the ratio of PSA-NCAM to NCAM could also have direct consequences on the long-term stability of excitatory synapses, if, as suggested by several results, stability correlates with size [63] and if size changes are mediated or associated to expression of properties of plasticity [24, 39, 64]. The interest of this general hypothesis about the role of PSA-NCAM and NCAM is that it makes predictions that should now become testable with the new developments carried out in confocal imaging. For example the dynamic of synaptic proteins at the PSD should depend upon the ratio of PSA-NCAM to NCAM or the stabilization of new spines by activity should also require expression of PSA-NCAM. Additionally, the development of new molecular tools such as peptides mimicking specific regions of these adhesion molecules could represent powerful new approaches to identify specific interactions involving adhesion molecules and their role in synapse formation and plasticity [65]. Evidence from recent results indeed indicates the great potential interest of these new tools with regard to therapeutic applications [57, 66].

Conclusion

The work reviewed here clearly points to an important function of NCAM and PSA-NCAM at excitatory synapses. While the evidence that these molecules regulate synapse formation mechanisms remains as yet scarce, solid data now firmly established that they play an important role in synaptic plasticity. As proposed here, these two aspects might actually reflect the same function of NCAM and PSA-NCAM at synapses, i.e., to regulate the capacity of the PSD to undergo remodeling and therefore set the stage for the formation and stabilization of the synapse. This hypothesis is already supported by interesting recent data, but it is very likely that some new developments in several methodological approaches will make it possible to test this idea more thoroughly.

References

1. Trachtenberg JT, Chen BE, Knott GW, Feng G, Sanes JR, Welker E, Svoboda K (2002) Long-term in vivo imaging of experience-dependent synaptic plasticity in adult cortex. Nature 420:788–794
2. Holtmaat AJ, Trachtenberg JT, Wilbrecht L, Shepherd GM, Zhang X, Knott GW, Svoboda K (2005) Transient and persistent dendritic spines in the neocortex in vivo. Neuron 45:279–291

3. Zuo Y, Lin A, Chang P, Gan WB (2005) Development of long-term dendritic spine stability in diverse regions of cerebral cortex. Neuron 46:181–189
4. Grutzendler J, Kasthuri N, Gan WB (2002) Long-term dendritic spine stability in the adult cortex. Nature 420:812–816
5. Brown CE, Li P, Boyd JD, Delaney KR, Murphy TH (2007) Extensive turnover of dendritic spines and vascular remodeling in cortical tissues recovering from stroke. J Neurosci 27:4101–4109
6. Holtmaat A, Wilbrecht L, Knott GW, Welker E, Svoboda K (2006) Experience-dependent and cell-type-specific spine growth in the neocortex. Nature 441:979–983
7. Laumonnier F, Cuthbert PC, Grant SG (2007) The role of neuronal complexes in human x-linked brain diseases. Am J Hum Genet 80:205–220
8. Dalva MB, McClelland AC, Kayser MS (2007) Cell adhesion molecules: signalling functions at the synapse. Nat Rev Neurosci 8:206–220
9. Yamada S, Nelson WJ (2007) Synapses: sites of cell recognition, adhesion, and functional specification. Annu Rev Biochem 76:267–294
10. Craig AM, Kang Y (2007) Neurexin-neuroligin signaling in synapse development. Curr Opin Neurobiol 17:43–52
11. Gerrow K, El-Husseini A (2006) Cell adhesion molecules at the synapse. Front Biosci 11:2400–2419
12. Akins MR, Biederer T (2006) Cell-cell interactions in synaptogenesis. Curr Opin Neurobiol 16:83–89
13. Garner CC, Waites CL, Ziv NE (2006) Synapse development: still looking for the forest, still lost in the trees. Cell Tissue Res 326:249–262
14. Prange O, Murphy TH (2001) Modular transport of postsynaptic density-95 clusters and association with stable spine precursors during early development of cortical neurons. J Neurosci 21:9325–9333
15. Gerrow K, Romorini S, Nabi SM, Colicos MA, Sala C, El-Husseini A (2006) A preformed complex of postsynaptic proteins is involved in excitatory synapse development. Neuron 49:547–562
16. Malinow R, Malenka RC (2002) AMPA receptor trafficking and synaptic plasticity. Annu Rev Neurosci 25:103–126
17. Gray NW, Weimer RM, Bureau I, Svoboda K (2006) Rapid redistribution of synaptic PSD-95 in the neocortex in vivo. PLoS Biol 4:e370
18. Ziv NE, Smith SJ (1996) Evidence for a role of dendritic filopodia in synaptogenesis and spine formation. Neuron 17:91–102
19. Engert F, Bonhoeffer T (1999) Dendritic spine changes associated with hippocampal long-term synaptic plasticity. Nature 399:66–70
20. Jourdain P, Fukunaga K, Muller D (2003) Calcium/calmodulin-dependent protein kinase II contributes to activity-dependent filopodia growth and spine formation. J Neurosci 23:10645–10649
21. De Roo M, Klauser P, Mendez P, Poglia L, Muller D (2008) Activity-dependent PSD formation and stabilization of newly formed spines in hippocampal slice cultures. Cereb Cortex 18:151–161
22. Knott GW, Holtmaat A, Wilbrecht L, Welker E, Svoboda K (2006) Spine growth precedes synapse formation in the adult neocortex in vivo. Nat Neurosci 9:1117–1124
23. McAllister AK (2007) Dynamic aspects of CNS synapse formation. Annu Rev Neurosci 30:425–450
24. Ehrlich I, Klein M, Rumpel S, Malinow R (2007) PSD-95 is required for activity-driven synapse stabilization. Proc Natl Acad Sci USA 104:4176–4181
25. Tang L, Hung CP, Schuman EM (1998) A role for the cadherin family of cell adhesion molecules in hippocampal long-term potentiation. Neuron 20:1165–1175
26. Okamura K, Tanaka H, Yagita Y, Saeki Y, Taguchi A, Hiraoka Y, Zeng LH, Colman DR, Miki N (2004) Cadherin activity is required for activity-induced spine remodeling. J Cell Biol 167:961–972

27. Cremer H, Lange R, Christoph A, Plomann M, Vopper G, Roes J, Brown R, Baldwin S, Kraemer P, Scheff S et al (1994) Inactivation of the N-CAM gene in mice results in size reduction of the olfactory bulb and deficits in spatial learning. Nature 367:455–459
28. Luthi A, Laurent JP, Figurov A, Muller D, Schachner M (1994) Hippocampal long-term potentiation and neural cell adhesion molecules L1 and NCAM. Nature 372:777–779
29. Muller D, Wang C, Skibo G, Toni N, Cremer H, Calaora V, Rougon G, Kiss JZ (1996) PSA-NCAM is required for activity-induced synaptic plasticity. Neuron 17:413–422
30. Becker CG, Artola A, Gerardy-Schahn R, Becker T, Welzl H, Schachner M (1996) The polysialic acid modification of the neural cell adhesion molecule is involved in spatial learning and hippocampal long-term potentiation. J Neurosci Res 45:143–152
31. Bukalo O, Fentrop N, Lee AY, Salmen B, Law JW, Wotjak CT, Schweizer M, Dityatev A, Schachner M (2004) Conditional ablation of the neural cell adhesion molecule reduces precision of spatial learning, long-term potentiation, and depression in the CA1 subfield of mouse hippocampus. J Neurosci 24:1565–1577
32. Eckhardt M, Bukalo O, Chazal G, Wang L, Goridis C, Schachner M, Gerardy-Schahn R, Cremer H, Dityatev A (2000) Mice deficient in the polysialyltransferase ST8SiaIV/PST-1 allow discrimination of the roles of neural cell adhesion molecule protein and polysialic acid in neural development and synaptic plasticity. J Neurosci 20:5234–5244
33. Markram K, Gerardy-Schahn R, Sandi C (2007) Selective learning and memory impairments in mice deficient for polysialylated NCAM in adulthood. Neuroscience 144:788–796
34. Stoenica L, Senkov O, Gerardy-Schahn R, Weinhold B, Schachner M, Dityatev A (2006) In vivo synaptic plasticity in the dentate gyrus of mice deficient in the neural cell adhesion molecule NCAM or its polysialic acid. Eur J Neurosci 23:2255–2264
35. Rutishauser U, Acheson A, Hall AK, Mann DM, Sunshine J (1988) The neural cell adhesion molecule (NCAM) as a regulator of cell-cell interactions. Science 240:53–57
36. Geinisman Y (1993) Perforated axospinous synapses with multiple, completely partitioned transmission zones: probable structural intermediates in synaptic plasticity. Hippocampus 3:417–433
37. Buchs PA, Muller D (1996) Induction of long-term potentiation is associated with major ultrastructural changes of activated synapses. Proc Natl Acad Sci USA 93:8040–8045
38. Toni N, Buchs PA, Nikonenko I, Povilaitite P, Parisi L, Muller D (2001) Remodeling of synaptic membranes after induction of long-term potentiation. J Neurosci 21:6245–6251
39. Matsuzaki M, Honkura N, Ellis-Davies GC, Kasai H (2004) Structural basis of long-term potentiation in single dendritic spines. Nature 429:761–766
40. Kiss JZ, Wang C, Olive S, Rougon G, Lang J, Baetens D, Harry D, Pralong WF (1994) Activity-dependent mobilization of the adhesion molecule polysialic NCAM to the cell surface of neurons and endocrine cells. EMBO J 13:5284–5292
41. Kasper C, Rasmussen H, Kastrup JS, Ikemizu S, Jones EY, Berezin V, Bock E, Larsen IK (2000) Structural basis of cell-cell adhesion by NCAM. Nat Struct Biol 7:389–393
42. Hinsby AM, Berezin V, Bock E (2004) Molecular mechanisms of NCAM function. Front Biosci 9:2227–2244
43. Ditlevsen DK, Povlsen GK, Berezin V, Bock E (2007) NCAM-induced intracellular signaling revisited. J Neurosci Res 86(4):727–743
44. Gascon E, Vutskits L, Kiss JZ (2007) Polysialic acid-neural cell adhesion molecule in brain plasticity: from synapses to integration of new neurons. Brain Res Rev 56:101–118
45. Rutishauser U (2008) Polysialic acid in the plasticity of the developing and adult vertebrate nervous system. Nat Rev Neurosci 9:26–35
46. Williams EJ, Walsh FS, Doherty P (2003) The FGF receptor uses the endocannabinoid signaling system to couple to an axonal growth response. J Cell Biol 160:481–486
47. Kiselyov VV, Soroka V, Berezin V, Bock E (2005) Structural biology of NCAM homophilic binding and activation of FGFR. J Neurochem 94:1169–1179
48. Kiryushko D, Korshunova I, Berezin V, Bock E (2006) Neural cell adhesion molecule induces intracellular signaling via multiple mechanisms of Ca2+ homeostasis. Mol Biol Cell 17:2278–2286

49. Muller D, Djebbara-Hannas Z, Jourdain P, Vutskits L, Durbec P, Rougon G, Kiss JZ (2000) Brain-derived neurotrophic factor restores long-term potentiation in polysialic acid-neural cell adhesion molecule-deficient hippocampus. Proc Natl Acad Sci USA 97:4315–4320
50. Gascon E, Vutskits L, Jenny B, Durbec P, Kiss JZ (2007) PSA-NCAM in postnatally generated immature neurons of the olfactory bulb: a crucial role in regulating p75 expression and cell survival. Development 134:1181–1190
51. Lu Y, Christian K, Lu B (2007) BDNF: a key regulator for protein synthesis-dependent LTP and long-term memory? Neurobiol Learn Mem 89(3):312–23
52. Sytnyk V, Leshchyns'ka I, Delling M, Dityateva G, Dityatev A, Schachner M (2002) Neural cell adhesion molecule promotes accumulation of TGN organelles at sites of neuron-to-neuron contacts. J Cell Biol 159:649–661
53. Sytnyk V, Leshchyns'ka I, Dityatev A, Schachner M (2004) Trans-Golgi network delivery of synaptic proteins in synaptogenesis. J Cell Sci 117:381–388
54. Sytnyk V, Leshchyns'ka I, Nikonenko AG, Schachner M (2006) NCAM promotes assembly and activity-dependent remodeling of the postsynaptic signaling complex. J Cell Biol 174:1071–1085
55. Dityatev A, Dityateva G, Schachner M (2000) Synaptic strength as a function of post- versus presynaptic expression of the neural cell adhesion molecule NCAM. Neuron 26:207–217
56. Dityatev A, Dityateva G, Sytnyk V, Delling M, Toni N, Nikonenko I, Muller D, Schachner M (2004) Polysialylated neural cell adhesion molecule promotes remodeling and formation of hippocampal synapses. J Neurosci 24:9372–9382
57. Cambon K, Hansen SM, Venero C, Herrero AI, Skibo G, Berezin V, Bock E, Sandi C (2004) A synthetic neural cell adhesion molecule mimetic peptide promotes synaptogenesis, enhances presynaptic function, and facilitates memory consolidation. J Neurosci 24:4197–4204
58. Di Cristo G, Chattopadhyaya B, Kuhlman SJ, Fu Y, Belanger MC, Wu CZ, Rutishauser U, Maffei L, Huang ZJ (2007) Activity-dependent PSA expression regulates inhibitory maturation and onset of critical period plasticity. Nat Neurosci 10:1569–1577
59. De Paola V, Holtmaat A, Knott G, Song S, Wilbrecht L, Caroni P, Svoboda K (2006) Cell type-specific structural plasticity of axonal branches and boutons in the adult neocortex. Neuron 49:861–875
60. Xu HT, Pan F, Yang G, Gan WB (2007) Choice of cranial window type for in vivo imaging affects dendritic spine turnover in the cortex. Nat Neurosci 10:549–551
61. De Paola V, Arber S, Caroni P (2003) AMPA receptors regulate dynamic equilibrium of presynaptic terminals in mature hippocampal networks. Nat Neurosci 6:491–500
62. Zuber B, Nikonenko I, Klauser P, Muller D, Dubochet J (2005) The mammalian central nervous synaptic cleft contains a high density of periodically organized complexes. Proc Natl Acad Sci USA 102:19192–19197
63. Kasai H, Matsuzaki M, Noguchi J, Yasumatsu N, Nakahara H (2003) Structure-stability-function relationships of dendritic spines. Trends Neurosci 26:360–368
64. Harvey CD, Svoboda K (2007) Locally dynamic synaptic learning rules in pyramidal neuron dendrites. Nature 450:1195–1200
65. Berezin V, Bock E (2004) NCAM mimetic peptides: pharmacological and therapeutic potential. J Mol Neurosci 22:33–39
66. Skibo GG, Lushnikova IV, Voronin KY, Dmitrieva O, Novikova T, Klementiev B, Vaudano E, Berezin VA, Bock E (2005) A synthetic NCAM-derived peptide, FGL, protects hippocampal neurons from ischemic insult both in vitro and in vivo. Eur J Neurosci 22:1589–1596