Special Focus: Synapse-Glia Interactions

Polysialic acid and activity-dependent synapse remodeling

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Polysialic acid (PSA) is a large carbohydrate added post-translationally to the extracellular domain of the Neural Cell Adhesion Molecule (NCAM) that influences its adhesive and other functional properties. PSA-NCAM is widely distributed in the developing nervous system where it promotes dynamic cell interactions, like those responsible for axonal growth, terminal sprouting and target innervation. Its expression becomes restricted in the adult nervous system where it is thought to contribute to various forms of neuronal and glial plasticity. We here review evidence, obtained mainly from hypothalamic neuroendocrine centers and the olfactory system, that it intervenes in structural synaptic plasticity and accompanying neuronal-glial transformations, making possible the formation and elimination of synapses that occur under particular physiological conditions. While the mechanism of action of this complex sugar is unknown, it is now clear that it is a necessary molecular component of various cell transformations, including those responsible for activity-dependent synaptic remodeling.

Introduction

Although stable neurochemical and functional specificities are fundamental features of neuronal connectivity in the adult mammalian central nervous system (CNS), another important attribute is the ability to undergo function-related structural changes. Structural transformations can affect the overall morphology and therefore, interrelationships of CNS cells, both neurons and glia. They can also affect synapses, the fundamental elements of neuronal connectivity. While the concept of ‘synaptic plasticity’ is usually associated with functional modifications in preexisting synapses (like long-term potentiation or LTP), it connotes structural changes as well, including formation and elimination of synapses that can take place during normal conditions of changing neuronal activity (reviewed in refs. 1–3). Moreover, to the classic view of synapses composed of neuronal pre- and post-synaptic elements, we must now include a third player, glia (reviewed in ref. 4). CNS synapses are generally encapsulated by fine distal processes of astrocytes that actively contribute to the regulation of synaptic function.¹ This intimate neuronal-glial association is not stable but is modifiable in several brain regions, which adds further complexity to neurotransmission and neuronal communication (reviewed in ref. 5). Such morphological transformations are induced by physiological conditions of heightened neuronal activity and highlight the nervous system’s remarkable capacity for restructuring to meet particular functional conditions.

In the CNS, architectural specificity is attained during development and maintained in the adult through the action of a cohort of membrane-bound and extracellular matrix molecules, particularly adhesion-type glycoproteins with permissive and/or instructive functions. They include cell adhesion molecules like the cadherins, ephrins, neurexins and members of the immunoglobulin superfamily, such as the Neural Cell Adhesion Molecule (NCAM), as well as their receptors. Complex glycoproteins secreted from astrocytes into the extracellular space, like the chondroitin sulphate proteoglycans and tenascins, also contribute.⁶ These molecules play critical roles in the establishment, maintenance and functional modulation of neuronal circuitry. During synapse formation, they intervene in axonal growth, pathfinding and target recognition and in the differentiation of pre- and post-synaptic specializations; they play a role in the regulation of synaptic size, stability, strength and plasticity as well. It is not unexpected then that such molecules play significant roles in the capacity of synapses for restructuring. The topic has gained increasing attention and there are several excellent reviews summarising recent findings.⁷–¹⁰

We here focus on NCAM, and in particular, its highly sialylated isoforms, in activity-dependent changes of synaptic connections in the adult. As we shall discuss, we now have considerable evidence showing that polysialic acid (PSA), a large, highly complex sugar on the extracellular domain of NCAM, intervenes actively in the dynamic cell interactions responsible for synaptic remodeling. While mechanisms of action are still a matter of speculation, numerous observations from different neuronal systems make it obvious that PSA-NCAM is a necessary molecular participant in activity-dependent structural synaptic plasticity in the adult CNS.

PSA-NCAM, its Structure and Possible Mode of Action

Structure. Polysialic acid (PSA) is a linear homopolymer of α 2-8-N acetylenuraminic acid whose major carrier in vertebrates is NCAM (reviewed in refs. 11–14). NCAM is a member of the immunoglobulin superfamily of adhesion molecules coded by a single copy gene from which at least 20–30 distinct forms can be generated by alternative splicing and post-translational modifications.
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(reviewed in ref. 15). There are three major classes of polypeptides, differing in their size (NCAM 180, 140 and 120), cytoplasmic domains (NCAM 180 and 140) or in their means of attachment to the cell membrane (NCAM 120).

Polysialic acids are polymers of derivatives of nine carbon neuraminic acid which can be found in a wide range of biological components, from the coat of certain bacteria to the membranes of vertebrate neural cells. PSA is a relatively simple and large molecule, the number of monomers ranging from about 8 to over 100 (reviewed in refs. 11–13). Polysialylation can account for up to 30% of relative molecular mass. The carbohydrate is added posttranslationally through two Golgi-associated polysialyltransferases: ST8SialV (PST) and ST8SialII (STX) (reviewed in ref. 16).

Most of the interest linked to NCAM was prompted by the discovery that its functional properties are strongly influenced by polysialylation (reviewed in ref. 14). Indeed, PSA can be added to all the three forms of NCAM by post-translational modification, thus introducing new possibilities of modulating adhesion.

**Mode of action.** PSA serves as a potent negative regulator of cell interactions via its unusual biophysical properties (reviewed in refs. 12 and 14), although how it intervenes to permit restructuring in the CNS remains basically undetermined.

The classical view of PSA action refers to its ability to decrease NCAM-mediated membrane-membrane adhesion through steric properties resulting from a high density of negative charges which contribute to the hydrated volume of NCAM. The complex mechanism(s) allowing PSA to modulate NCAM-mediated cis, trans, homophilic and heterophilic interactions involve the charge and hydration of the polymer, its influence on the width of the extracellular space as well as its influence on receptors located on the same cell, other cells and/or the extracellular substrate. That there can be different degrees of polysialylation on different NCAM isoforms varying in location and time renders identification of specific mechanisms of action particularly difficult (reviewed in refs. 18–20).

Many effects involving the attenuation of cell-cell interactions are not directly mediated through the adhesion properties of NCAM, but rather, are linked to a global effect due to steric inhibition of membrane-membrane apposition based solely on PSA. The importance of such an effect resides in the simple fact that cell-cell interactions are more influenced by the rate of receptor encounter (e.g., IgCAM, L1, cadherins, integrins) than by the intrinsic affinities of the receptors themselves. In other words, PSA decreases the rate of binding among receptors on apposing cells, without affecting the intrinsic binding properties of those receptors. Alternatively, PSA can increase the magnitude of repulsive pressure between membranes sufficient to abrogate the attractive forces exerted by proteins like the cadherins, integrins and NCAM itself.

The expression of PSA on NCAM is regulated by a combination of developmental or physiological programs that control its synthesis, delivery and degradation. The amount of PSA on the cell surface appears to depend on two major regulatory mechanisms: (1) the synthesis of PSA-NCAM on the basis of transcription and/or activity of the sialyltransferases, and (2) the turnover of the molecule at the cell surface (reviewed in refs. 12, 14 and 20). In many systems, biosynthesis of PSA-NCAM is regulated by neuronal activation, including electrical activity in axons, which can also influence its expression in the target cell, or the loss of input from the periphery. Nevertheless, there is also strong evidence that in some cells, including glia and neuroendocrine neurons, PSA-NCAM arrives at the surface in a rapid, constitutive fashion that is not dependent on calcium entry or cell activation.

PSA-NCAM is generally viewed as a regulator of the extracellular space. The hydrated volume of NCAM together with PSA has been estimated to be about three times that of the polypeptide, thus directly increasing the width of the extracellular space and the close-ness of membrane-membrane contact. The extracellular space is comparatively wide in embryonic tissues which are highly hydrated, whereas it becomes progressively reduced with postnatal matura-tion. The fact that there are high amounts of PSA-NCAM in sites of persistent plasticity in the adult CNS with a specific time-course during postnatal brain maturation suggests that polysialylation may provide a suitable extracellular environment in the highly static, mature nervous tissue (reviewed in ref. 33).

**Regulation.** The fact that PSA can be rapidly expressed at the cell surface, along with the demonstration of relatively fast changes in the morphology and contacts of cells expressing high amounts of PSA-NCAM, raises the possibility that polysialylation may be involved in rapid modulation of cell surface interactions. Rapid conformational changes can affect neurons and glia, either when both cell types express PSA, like in the hypothalamic magnocellular nuclei, or when it is expressed only by the neuronal compartment, such as migrating neuroblasts in the process of adult neurogenesis.

Apart from specific molecular mechanisms, "the coupling of changes in PSA expression with the relevant biological process is the key to the creation of permissive conditions at the appropriate time and place." This concept is supported by observations from PSA-negative but NCAM-positive mice obtained by simultaneous deletion of the polysialyltransferases PST and STX, observations that highlight the interplay between NCAM and PSA in regulating NCAM interactions and neuronal differentiation. These animals display a severe phenotype characterized by postnatal growth and brain defects beyond features described in NCAM-null mice. Since these defects are rescued by additional deletion of NCAM, it is possible that they originate from a gain of NCAM functions due to the deficiency in PSA. Accordingly, differentiation can be induced by enabling heterophilic NCAM signals at cell-cell contacts that are otherwise prevented by polysialylation and dynamic changes in PSA can modulate the cell response by affecting the integration of homophilic and heterophilic NCAM interactions.

Through these different mechanisms, PSA-NCAM can play a prominent role in various forms of physiologically-occurring structural plasticity in the adult CNS, which include changes in the shape of neurons and associated glia and therefore in their interrelationships, in the length and arborization pattems of axons and dendrites, in the formation and elimination of synapses, in activity-induced plasticity, and in adult neurogenesis.

**PSA-NCAM in Synaptic Plasticity**

Synaptic plasticity and its modulation through adhesion molecules appeared very early in evolution and both processes are functionally present in simple nervous systems. For example, a group of cell surface proteins designated Aplysia cell adhesion molecules (apCAM’s) and fasciclin II, which are homologues of vertebrate
NCAM in the marine mollusk Aplysia and in Drosophila, respectively, are required for morphological rearrangement of synapses.\textsuperscript{31,42} Polysialylation, from the evolutionary point of view appears more important in vertebrates since invertebrate NCAM homologues are not polysialylated and the attenuation of cell interactions can occur through downregulation of adhesion receptor distribution on the cell surface\textsuperscript{43} or by expression of other anti-adhesive proteins.\textsuperscript{44} The appearance of PSA in complex nervous systems may thus represent a mechanism allowing more global effects on cell interactions with respect to an increase in complexity due to the regulation of many different genes (reviewed in ref. 18).

During maturation of neuronal systems throughout the postnatal period, the expression of PSA-NCAM becomes progressively confined to pre- and post-synaptic sites.\textsuperscript{45-47} Indeed, as shown by many observations from different regions, polysialylation is critical for establishing proper synaptic contacts. For instance, experimental PSA removal either by genetic manipulation or enzymatic digestion can induce ectopic synapse formation (as in the hippocampus\textsuperscript{48}) or may alter the very formation of synapses (as between chick oculomotor axons and ciliary neurons\textsuperscript{49}). More recently, it was seen that developmental and activity-dependent regulation of PSA plays a major role in the maturation of certain GABAergic innervations in the visual cortex since premature removal of PSA resulted in precocious innervation and enhanced inhibitory synaptic transmission.\textsuperscript{50} By the third postnatal week in rodents, most synapses lose PSA-NCAM, a change that is paralleled by important modifications in PSA expression related to other types of plasticity, such as persistent neurogenesis (reviewed in ref. 33 and Fig. 1).

There is now a considerable literature showing that polysialylation plays an essential role in functional synaptic plasticity in the adult. The most studied and perhaps most complex model of this kind of neuronal plasticity is the hippocampus, where it is clear that long-lasting changes at excitatory synapses like long-term potentiation (LTP) and long-term depression (LTD) are dependent on PSA expression.\textsuperscript{51-54} Selective PSA removal prevents induction of both LTP and LTD in cultured hippocampal neurons, and in slices prepared from NCAM knock-out mice.\textsuperscript{52} A more recent study, in which the role of PSA and NCAM at synapses formed by entorhinal cortex axons in the dentate gyrus were analysed in vivo in NCAM\textsuperscript{-}, STX- and PST-deficient mice,\textsuperscript{55} revealed further complexity, suggesting that PSA may be important for basal synaptic transmission in newly generated granule cells, whereas LTP in the dentate gyrus depends on NCAM glycoprotein alone.

The phenomenon of structural synaptic plasticity in the hippocampus appears more complex since there is an ongoing remodeling\textsuperscript{56-58} together with that linked to neurogenesis as a result of proliferative/differentiative activity of stem/progenitor cells.\textsuperscript{59,60} Since this issue has been widely addressed in several recent reviews (reviewed in refs. 14, 33 and 61) it shall not be discussed further here. Rather, in the following sections we will address the issue of PSA and synaptic remodeling in two other contexts in which they are highly correlated (1) in certain hypothalamic neuroendocrine centers, where PSA expression, synaptic rewiring and concomitant astrocytic changes are functionally linked to specific physiological states and (2) in the olfactory system and olfactory pathways, where polysialylation is associated with a wide range of structural plastic events, among which synaptic plasticity and indirectly, learning and memory.

**PSA-NCAM is Necessary for Axonal and Synaptic Remodeling in Neuroendocrine Systems**

The strongest evidence showing that PSA is essential for axon and synapse remodeling has been obtained from hypothalamic neuroendocrine structures, like the supraoptic (SON) and paraventricular (PVN) nuclei of the hypothalamo-neurohypophysial system (HNS), the arcuate nuclei and the median eminence of several species (reviewed in refs. 3, 5, 62 and 63). It is now clear that in these particular hypothalamic nuclei, certain circuits controlling the activity of the neuroendocrine neurons remain relatively plastic throughout life, a plasticity coincident with restructuring of adjacent astrocytes and dependent on neuronal activity. In neurohemal projection sites of these neurons, like the neurohypophysis and the external area of the median eminence, there are concomitant changes in the number and morphology of neurosecretory axons and terminals as well as remodeling of associated glia. All these brain areas express high levels of PSA.\textsuperscript{26,64-68}

The SON and magnocellular portions of the PVN are composed of magnocellular neurons secreting the vital neurohormones, oxytocin and vasopressin. In these nuclei, there is a striking synapse turnover in relation to specific physiological conditions leading to enhanced oxytocin secretion, like parturition, lactation and chronic dehydration (reviewed in refs. 3 and 62). An obvious manifestation is the appearance of boutons making synaptic contact onto more than one postsynaptic elements simultaneously (‘multiple’ synapses) but careful synaptic density analyses reveal that synapse formation affects boutons making single synaptic contacts as well. Analyses of ultrathin sections in which pre- and post-synaptic elements were immunoidentified allowed to show that the circuits undergoing plasticity are inhibitory (GABAergic) and excitatory (glutamatergic and noradrenergic) impinging on oxytocin neurons. The most important increases affect GABAergic inputs so that in the SON of lactating rats, about 50% of all axo-somatic and axo-dendritic synapses are GABAergic, compared to 35% in the SON of virgin rats under basal conditions. Once stimulation is over, synaptic densities revert to control values. These synaptic transformations are clearly associated with astrocytic changes since an enhanced number of synapses is invariably associated with a reduction in astrocytic coverage of oxytocinergic somata and dendrites; the return to control synaptic levels occurs together with a reestablishment of glial coverage. As seen in more recent work, synaptic remodeling in this system can be reproduced in vitro, in acute slices from adult hypothalamus that include the SON.\textsuperscript{34,69}

Electron microscopy and patch clamp electrophysiology of these preparations allowed to show unequivocally that synapse formation is very rapid, occurring within an hour, is reversible within hours as well and results in the formation of functional, inhibitory synapses. Another hypothalamic center in which there are synaptic changes clearly linked to varying conditions of physiological conditions is the arcuate nucleus. In adult females, there is a natural phasic synaptic turnover that occurs in relation to the ovarian cycle.\textsuperscript{63} Thus, the number of axo-somatic GABAergic synapses on arcuate neurons falls significantly between the morning and afternoon of proestrus, remains low throughout estrus, and rises to baseline levels on metestrus, to fall again at proestrus. Concomitantly, like in the magnocellular nuclei,\textsuperscript{3} there is increased and decreased astrocytic ensheathment of neuronal profiles.\textsuperscript{70} This negative correlation between the level of glial ensheathment and number of synaptic
As noted earlier (Section 2), neurons and glia in these structures constitutively express high levels of PSA-NCAM (Fig. 1), an expression that is not markedly affected by changing conditions of neurosecretion. This does not mean that PSA is of no consequence to their ability to undergo activity-dependent axonal and synaptic remodeling. On the contrary, it is a prerequisite for all dynamic phases of such plasticity. Thus, specific enzymatic removal of PSA from NCAM in the SON in situ inhibited glial and neuronal remodeling associated with lactation and chronic dehydration; there was no effect if PSA was removed once morphological changes had already taken place. Likewise, enzymatic removal of PSA from cell surfaces in the neurohypophysis prevented stimulation-related induction and reversal of axon and glial changes but had no effect once remodeling had occurred. In the arcuate nucleus, enzymatic removal of PSA inhibited the onset of synaptic changes associated with reproductive status. It is noteworthy that such morphofunctional changes, at least in the magnocellular nuclei, modify the activity of both polysialyltransferases responsible for polysialylation.

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Figure 1. Highly sialylated Neural Cell Adhesion Molecule (PSA-NCAM) in adult brain cell populations involved in different types of synaptic plasticity. Top: diagrammatic representation of sagittal (A) and coronal (B) views of the rat brain showing areas containing cells enriched in PSA-NCAM. In grey, neurogenic sites. H, hippocampus; OB, olfactory bulb; OM, olfactory mucosa; PC, piriform cortex; SON, supraoptic nucleus; SVZ, subventricular zone. Bottom: Immunoreactions for polysialic acid (PSA) reveal highly immunopositive neurons (C, D and F) in restricted areas known to undergo plasticity, like the olfactory bulb (C), hippocampus (D) and the piriform cortex (F). In neuroendocrine centers like the SON (E), a strong reaction fills the neuropile and is due to immunoreaction on neuronal and glial (astrocytic) processes. Note that newly generated (top, green nuclei) and non-newly generated cells (top, unstained nuclei) display a typical punctate reaction on their cell membrane (see also C and D). DCX, doublecortin.

inputs in relation to fluctuations in steroid levels has been detected in the arcuate nucleus of rodents and primates. In sheep, seasonal variations in glial ensheathment and synaptic inputs occur in the preoptic area. Thus, the number of synapses decreases while astrocytic coverage of neurons increases between the breeding and non-breeding seasons in conjunction with the diminished pulsatile GnRH secretion that leads to anestrous.

In the neurohemal structures containing the axons of these neurons, glial transformations give rise to a reduced glial ensheathment of neurosecretory terminals which, in perivascular areas, leads to a significantly enlarged neurovascular contact zone (reviewed in ref. 5). For example, in the rat neurohypophysis under basal conditions of neurosecretion, about 40% of the perivascular basal lamina is contacted by neurosecretory terminals and about 60% by processes of astrocytic-like pituicytes. During all conditions that stimulate neurosecretion, there is retraction of pituicyte processes from the perivascular zone as well as proliferation of neurosecretory terminals and these proportions are reversed. At the end of stimulation, glial coverage of perivascular areas returns to control levels. The axons of arcuate GnRH neurons project to the external layer of the median eminence where fluctuating steroid levels lead to axonal-glial changes similar to those characterising the neurohypophysis. They result in retraction of end feet of tanyctyes (modified ependymoglial cells) from the perivascular zone and exposure of GnRH terminals to fenestrated portal capillaries.

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A molecule like NCAM can be considered permissive for morphological plasticity in the neuroendocrine centers since its expression is not tightly linked to neuronal activity yet it appears obligatory for their morphological plasticity. As discussed above, this was clearly evident from enzyme perturbation experiments in which PSA was punctually removed by endosialidase application at particular stages of physiological function. Nonetheless, there can be compensa-
ory replacement by other molecules since remodeling is visible in lactating and salt loaded transgenic mice genetically deficient in NCAM. It is highly likely that these observations reveal a redundancy in the molecular systems permitting morphological plasticity. Similar phenomena have been described in other neuronal systems in which NCAM is highly expressed and which continue to undergo plasticity in NCAM−/− mutants. In a sense, the specific identity of molecules permissive for remodeling may not be important provided that they activate the same intracellular mechanism. The response of neurons and astrocytes implicated in remodeling would be invariant and independent of the identity of the factor, provided the proper stimulus intervenes.

**PSA-NCAM in the Olfactory System and Related Pathways: An Indirect Involvement in Synapse Remodeling**

Formation/elimination/remodeling of synapses are decisive for memory storage and for learning-induced changes in the wiring diagram of many neuronal circuits (reviewed in refs. 2, 57 and 76). It has been recognised that PSA-NCAM plays a prominent role in this context, a role which nonetheless remains poorly understood.

The olfactory pathway, in which immunocytochemical evidence for polysialylation was first obtained, is a striking illustration of several aspects of structural neuronal plasticity, including synaptic remodeling at different levels of the olfactory pathway, from olfactory receptors located in the nose to cortical circuits, axonal/dendritic outgrowth towards and within the olfactory bulb, and adult neurogenesis both at peripheral and central levels (reviewed in ref. 78). PSA-NCAM is associated with most anatomical/cellular elements of this system, including the olfactory sheet and olfactory nerve, the main and accessory olfactory bulb, the rostral migratory stream originating from the forebrain subventricular zone (SVZ), and cortical areas receiving and processing olfactory inputs. (Fig. 1).

**PSA-NCAM and plasticity in the olfactory bulb.** A multipotent stem cell generates new olfactory receptor neurons in the olfactory sheet throughout adult life. These sensory neurons are physiologically replaced with an average life-span of 30 days. It is highly likely that these observations reveal a redundancy in the molecular systems permitting morphological plasticity. Similar phenomena have been described in other neuronal systems in which NCAM is highly expressed and which continue to undergo plasticity in NCAM−/− mutants. In a sense, the specific identity of molecules permissive for remodeling may not be important provided that they activate the same intracellular mechanism. The response of neurons and astrocytes implicated in remodeling would be invariant and independent of the identity of the factor, provided the proper stimulus intervenes.

**Is PSA-NCAM important for learning/memory in the olfactory pathway?** Experimental manipulations affecting the expression or the function of PSA-NCAM alter the ability of animals to learn, conversely, learning alters the expression of the molecule (reviewed in refs. 95 and 96). By examining PSA-NCAM distribution (Fig. 1), it becomes obvious that polysialylation characterizes complex, widely interconnected olfactory neural circuits and pathways: (1) the olfactory bulb and the primary olfactory cortex, consisting of three-layered cortical structures (allocortical areas and transitional allo/iso-cortical areas) and (2) the medial temporal lobe (including the hippocampal formation, the perirhinal and parahippocampal—postrhinal—cortices), a bidirectional pathway between cortical regions and the hippocampal formation. Most of these neuronal circuits represent multiple levels of the same pathway (primary sensory, projection and associative) and are involved in memory storage and learning. Their distribution provides correlative evidence for an involvement of polysialylation in synaptic remodeling.

The hippocampus can be considered an extension of the olfactory pathway, both structurally, due to its connections with the primary olfactory cortex and functionally, since selective types of olfactory memory depend on the hippocampal formation (reviewed in ref. 78). Polysialylation is important at this level as well as in the olfactory system proper. For example, changes in NCAM and PSA-NCAM expression occur in the piriform cortex and hippocampus in relation to learning of olfactory discrimination tasks, and transient polysialylation in the dentate gyrus is linked to memory for odor-reward association.

Structures of the medial temporal lobe are involved in memory consolidation, a process in which memories are initially dependent on the medial temporal lobe but gradually become established as long-term memories in other brain areas. Also, spatial and contextual
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memory/learning depend on corticocerebellar circuitry. In most cases, there is PSA-mediated plasticity. In the hippocampus, transient modulations of polysialylation follow spatial learning and memory while enzymatic removal of PSA affects spatial learning. A transient increase in the frequency of polysialylated cells in the adult hippocampus, and entorhinal cortex is detectable during memory consolidation. These changes occur early (between 6 and 12 hours) and return to basal levels within 72 hours following periods of consolidation related to the selective retention and/or elimination of spines and polysialylation (reviewed in ref. 106). Also consolidation of passive avoidance learning is associated with transient increases of polysialylated neurons in layer II of the entorhinal/perirhinal cortex. Finally, recent studies provided evidence that PSA regulates synaptic plasticity at synapses formed by entorhinal cortex axons in the dentate gyrus in vivo, thereby extending previous in vitro results concerning the role of NCAM in LTP and presumably learning and memory.

In conclusion, there is now much evidence to support the involvement of PSA in several aspects of brain cognitive functions, exerting its effects at different levels of widely interconnected neuronal circuitries. Although suggestive of synaptic remodeling, the role of PSA-NCAM in isolated (non-newly generated) cortical neurons intercalated on the olfactory pathways remains a matter of debate. Hence, if at some levels of the olfactory/hippocampal systems PSA-NCAM is unequivocally related to neurogenesis, at other levels, it could be independent, as it is probably the case in most of the temporal lobe cortices.

PSA-NCAM on immature neurons in the mature brain: a potential reservoir for new synapses? It is generally thought that in areas of the hard-wired adult brain like the cerebral cortex, the morphology of most neuronal cells and their interconnections become fixed after specific time points or critical periods (reviewed in ref. 109). Nevertheless, an increasing number of observations indicate that there are cortical neurons that continue to express molecules involved in structural remodelling, like PSA-NCAM, and this could be illustrating their capacity for global morphological remodelling throughout life. In addition, they typically display a ‘punctate’ immunolabeling of their surface membranes and this could be illustrating their capacity for global morphological remodelling throughout life.

Examples such as the above provide a model in which polysialylation and dynamic processes of astrocytes may provide a space suitable for new synapse formation on a group of neurons within the static environment of the mature nervous tissue (reviewed in ref. 5). As we have described in this review, high levels of PSA are expressed throughout life by particular groups of neurons and their associated astrocytes (as in the hypothalamic magnocellular nuclei) as well as by isolated neuronal cells with some participation of glia (as in the olfactory bulb and cortex). Ongoing studies aim to unravel the function of the latter cells as well as the mechanisms that allow their persistent potential for plasticity.

Conclusions

As seen from the numerous observations described in this review, the presence of large amounts of PSA on the extracellular surface of neurons and astrocytes of the adult CNS appears critical for the morphological transformations linked to synapse formation and elimination occurring during diverse physiological conditions. The complex carbohydrate may provide particular extracellular space conditions favorable for such remodeling, for example, by greatly reducing adhesivity between adjacent cells and their synapses and/or the cells and their immediate extracellular matrix. In addition, PSA and/or its carrier NCAM may intervene more actively to allow interaction with other molecules on the cell surface of neurons and/or astrocytes, including other cell adhesion molecules, growth factors and receptors. These interactions may be critical for synapse remodeling since they will then trigger intracellular signaling mechanisms that ultimately result in gene activation and new protein synthesis underlying the morphological transformations. Analysis of mechanisms of PSA action as well as the identification of molecular factors with which it may intervene to permit structural synaptic plasticity is therefore of utmost relevance and remains a challenging task for future research.

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