Cortical maps: a role for astrocytes?
Mónica López-Hidalgo and James Schummers

Abstract
Astrocytes are a multifunctional cell type in the nervous system that can influence neurons and synapses in numerous ways. Astrocytes have been suggested to play important roles in synapse formation during development, as well as in multiple forms of synaptic plasticity in the developing and adult brain. Astrocytes respond to nearby neural activity with elevations in cytosolic calcium concentration, and in sensory cortex these calcium responses have been shown to be topographically aligned to neuronal sensory maps. Here, we review recent evidence for astrocyte interactions with neural circuits, with particular emphasis on how these interactions may shape the development, arrangement and plasticity of cortical sensory maps.

Addresses
Max Planck Florida Institute for Neuroscience, 1 Max Planck Way, Jupiter, FL 33458, United States

Corresponding author: Schummers, James
(James.Schummers@mpfi.org)

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Introduction
Maps refer to the systematic spatial clustering of neural circuits with similar functional roles in information representation. In circuits within sensory pathways, these maps represent similar aspects of the sensory world, in many cases a distorted representation of the sensory organ itself, including retinotopic, cocheleotopic and somatotopic maps. In some cases these maps are elaborated substantially to the point that there are specializations of the underlying structure to mimic the sensory apparatus, as in the case of the barrel cortex. During early development, coarse maps are delineated, in the absence of appropriate sensory-driven activity, by various molecular cues which guide axons to their appropriate target locations in combination with spontaneous activity patterns [1–3]. The onset of sensory input is typically followed by a period when the maps are particularly malleable, and the size, form and distribution of feature representations can be altered by plasticity rules that act upon the nature of the sensory inputs. The net effect of plasticity during this so-called ‘critical period’ is to enhance the representation of features that are more commonly present in the sensory world of the individual animal. As development progresses, the extent and ease by which maps can be altered by the nature and quantity of sensory inputs tends to decline. In the adult cortex, strong or prolonged changes in activity patterns, or additional factors such as behavioral attention are typically required to open the gate of plasticity.

While some of the synaptic and circuit mechanisms that underlie these processes are well delineated, many other remain unclear. Here we will examine the possible roles for astrocytes in the organization, development and plasticity of cortical sensory maps. The notion that astrocytes could be involved in activity-dependent map plasticity was proposed over twenty years ago [4], and supported by the demonstration that grafting immature astrocytes into adult cortex could enhance such plasticity [5]. Over the last two decades, the capacity of astrocytes to bi-directionally interact with synaptic activity, both by ‘listening in’ on, and modifying the activity of, synapses via morphological changes and/or the release of neuroactive compounds, has been increasingly appreciated. While the metabolic and support roles for astrocytes in neural function remain largely unchallenged [6,7], (though see [8,9]), there are now numerous lines of evidence that astrocytes are well equipped and well positioned to play important roles in synaptic plasticity. We propose that astrocytes are potentially important partners in sensory map organization and plasticity, and we emphasize that studies of cortical circuits should consider the possibility of their contributions.

Interactions between neurons and astrocytes
Neuron to astrocyte signaling
Astrocytes exhibit little in the way of electrical activity, but are quite active in terms of chemical signaling pathways, most notably large modulations of cytosolic calcium concentration, which have been visualized by the use of fluorescent calcium indicators. Other chemical signals are likely to play important roles in astrocyte signaling, but are not as readily observed without appropriate sensor technologies (e.g. [10]). Calcium signaling in astrocytes has long been known to be sensitive to nearby neural activity [11], though the specific mechanisms for translating neural activity to astrocyte signaling remain to be fully worked out, and are likely to be numerous [12]. Proto-plasmic astrocyte morphology is characterized by a small soma which gives rise to a small number of primary processes, each of which branches extensively in to either microscopic (nm scale) peripheral astrocyte processes
(PAPs) or end-feet which ensheathe blood vessels. At an ultrastructural level, astrocyte PAPs are positioned in close proximity to the synaptic cleft at nearly two-thirds of synapses [13]. Astrocytes have been shown to express a plethora of both ionotropic and metabotropic type receptors, including glutamatergic, cholinergic, noradrenergic, purinergic and GABAergic [14–16]. The metabotropic receptors include those of the Gq type, linked to the IP3 second-messenger pathway, and have been shown to trigger calcium elevations upon activation. During the last decade, a great deal of emphasis has been placed on the activation of astrocytic mGlURs by synaptically released glutamate as the primary means of triggering calcium signaling in the hippocampus and cortex [16,17]. The importance of this particular pathway in the adult cortex has been cast into doubt by the recent demonstration that mature astrocytes lack Gq type glutamate receptors [18*]. This report highlights the need for more careful characterization of many basic aspects of astrocyte cell biology and the risks of extrapolating from in vitro preparations (typically taken from immature brain tissue) to the in vivo situation (typically mature adults). Regardless of the specific mechanism, it remains clear that astrocytes are able to ‘listen in’ on synaptic transmission, which leads to calcium elevations even in adulthood [19–22].

Strong, synchronous or long lasting neural activity can elicit calcium elevations throughout astrocyte processes and somata. The physiological relevance of such stimulation protocols have been called into question [23], and the activity patterns of astrocytes under more natural ongoing activity levels remain largely unknown. Two recent papers have characterized localized calcium signals (<10 µm) in astrocyte processed in response to spontaneous or action potential evoked (minimal-stimulation-induced) synaptic release [24**,20**]. Notably, these events are substantially faster, both in rise time (~200 ms) and duration (~1 s) than those typically observed in the somatic compartment. These results suggest that astrocytes are capable of sensing local ongoing synaptic activity within the domain of their processes. Importantly, these events are detected in adult as well as juvenile hippocampal slices [20**]. These results highlight the need for a better understanding of the integration of synaptic activity by astrocytes: do they integrate inputs in a manner analogous to neurons, with sufficient activation of the processes leading to a somatic signal? If so, are there distinct consequences of somatic calcium signals?

Astrocyte to neuron communication

There is now a considerable literature suggesting that astrocytes are also capable of communicating with neurons by a number of means, including the physical interactions [15], metabolic supply [25,26], buffering of extracellular ions [27–29] and release of trophic factors [30] and neurotransmitters (referred to as gliotransmitters when released by astrocytes). The study of gliotransmission has grown substantially over the last decade, and has been largely responsible for a resurgence of interest in astrocytes. It is worth noting that the technical difficulty of isolating the source of released substances under physiological conditions in intact tissue has made the definitive demonstration of the phenomenon elusive, and the specifics and relevance remain controversial [23,31–33]. With that said, there is sufficient evidence in favor of the impact of gliotransmission on neural activity that it warrants consideration as a mechanism of neural circuit function and modulation.

The three best characterized gliotransmitters are adenosine triphosphate (ATP), glutamate and D-serine. There is now substantial evidence for the ability of astrocytes to release these substances [34–38] and the physiological responses in neurons which correlate with artificial or endogenous astrocyte calcium activity [24**,39]. ATP can be released from astrocytes in response to neuronal and glial activity and can activate neighboring astrocytes producing a propagating calcium wave [40,41]. The majority of astrocytes respond to ATP with calcium increases due to their expression of ionotropic (P2X) and metabotropic (P2Y) purinoceptors [41–43,44**,45]. ATP thought to be rapidly hydrolyzed to adenosine in the extracellular space, and is able to modulate individual neuronal activity as well as to coordinate neuronal and astrocytic networks [43–45]. Glutamate is released by astrocytes spontaneously [46,47] or in responses to neuronal activity [48,49]. In this context, it is able to modulate synaptic transmission through the activation of presynaptic mGlURs, thus regulating the amount of neuronal glutamate released [39,50] or by activating synaptic or extrasynaptic NMDARs [51,52,53*] where it can generate slow inward currents (SIC) [46,48,53*,54]. D-Serine is a co-agonist of NMDARs [55,56] targeting preferentially synaptic receptors [57], which is congruent with their parallel expression during development and in adulthood [58,59]. The main cellular origin of D-serine is currently under debate; while some work points to astrocytes [60–63], others suggest a neuronal source [64–66]. It was recently proposed that L-glutamate and D-serine are stored in the same vesicles [62] which could ensure the activation of NMDAR by both the agonist and co-agonist. This is an important factor, especially during development, when subunits with low-Mg2+ sensitivity are expressed presynaptically (NR2C and NR2D) [67,68].

Spatial arrangement of astrocytes

One of the inherently important aspects of maps is the spatial arrangement of information-content that converges onto neurons. In this context, it is noteworthy that sensory maps typically have spatial precision at a scale that is beyond the spatial extent of neuronal integration as judged by dendritic field size. For instance, in
the barrel cortex, the dendrites of superficial layer pyramidal neurons extend well beyond the boundaries of their home barrel [69], suggesting that they are likely to receive and integrate input from multiple whiskers (Figure 1a). This is evident in most sensory maps but has been most clearly demonstrated in orientation preference maps in the visual cortex of carnivores. Here, orientation preference maps are arranged radially around pinwheel centers with an impressive degree of precision [70,71]. However, these functional boundaries are sharper than the inter-neuronal distance, with a dramatic change in the orientation preference of neurons just 50 µm apart [72,73**]. The dendritic arbors of neurons can extend for hundreds of microns past the pinwheel center, into domains of the map representing widely varying orientation preference [74]. The evidence collected so far suggests that there is little specificity of connectivity near pinwheel centers, suggesting that neurons in these locations receive synaptic inputs carrying information representing a broad range of stimulus orientations [75,76]. Thus, it remains unclear what mechanisms are in place during the development and maintenance of map structure to ensure appropriately precise functional boundaries.

In contrast to the situation with neurons, the organization of cortical astrocytes is prima facie better suited to maintain the separation of information in a cortical map. Each astrocyte establishes a domain (~120,000 µm²; ~40 µm on a side) with little overlap with neighbors (3-5% in rodents and 12% in humans) where it has been estimated to contact ~140,000 synapses and at least one blood vessel [77,78]. Although there has been relatively little study of this organization across species or brain regions, the minimally overlapping domains, and the roughly even spacing of astrocyte somata has led to the notion that astrocytes effectively ‘tile’ neural tissue [77,79] such that any given volume of neuropil interacts with only a single astrocyte, a notion that has been referred to as ‘synaptic islands’ [80]. This arrangement would enable modular regulation of activity levels and synaptic processing (see below) on a fine spatial scale commensurate with the precision of sensory map structure (Figure 1b).

Since Ramon y Cajal, it has been clear that the morphology of astrocytes varies according to brain region and circuit location [81]. Although in some specialized astrocytes, such as Muller glia of the retina or Bergman glia in the cerebellum, their shape is related with their presumed functional role (for review see [82,83]), little is known about the diversity of cortical astrocyte morphology and its correlation with function. Interestingly, when comparing the morphology of cortical astrocytes across several species (mouse, squirrel monkey, chimpanzee and human) it has been observed that human astrocytes are ~2.5-fold larger than their rodent counterpart, which has been suggested to represent a phylogenetic change related with cognitive processing [84].

Information regarding the morphology of individual astrocytes in relation to maps remains sparse. In the olfactory bulb, astrocytes located within the olfactory glomeruli (OG) have an asymmetrical shape, prioritizing the extension of their process into the core of the same glomerulus, with minimal extension to the extraglomerular space [85,86], suggesting a compartmentalization of the inputs that arrives to the astrocytes — in these particular case, glomerular astrocytes would receive sensory information...
of a unique odor. However, in the barrel cortex, mature astrocytes do not show biased morphology within a barrel column, compared to those outside the barrels (in the septa) [87]. Given the precedent for a correspondence between astrocyte morphology and neural activity, the relationship of astrocyte organization to functional organization warrants further study.

Astrocyte networks

Overlaid on this modular organization is the connectivity between neighboring astrocytes by means of gap junctions. Astrocytes express high levels of connexins (Cx’s), especially the astrocyte specific Cx43 and Cx30 [88,89]. These connexins form gap junctions which enable the transmission of small molecules including calcium, IP₃ and other ions among neighbors. Under certain conditions, long-range communication through this gap junction network can be observed in vivo, including calcium waves, [90–92]. This connectivity pattern has led to the notion of astrocyte networks, which might have the capability to work in conjunction to regulate synaptic function over larger spatial scales [93,94]. However, the permeability of gap junctions is subject to regulation by a large number of factors [95], and the degree to which the gap junction network is functionally relevant under physiological conditions remains unclear. Recent evidence suggests that individual astrocytes can act independently of their neighbors in vivo, at least under anesthesia [73**,96]. There are reports of both sparse [47] and network-level [91] activity patterns in vivo, and future work will need to determine which conditions give rise to these apparently different modes of activity (which may additionally be brain-region, or even cortical layer, specific [92]). Thus, an important outstanding question is the degree to which, and the conditions under which, astrocytes function as a gap-junction coupled network, versus individual islands.

With that in mind, it is now clear that astrocyte calcium activity can accurately reflect the extent and boundaries of cortical sensory maps. The first demonstration of somatosensory-evoked calcium activity in astrocytes suggested that several aspects of the astrocyte activity matched the receptive field properties of surrounding neurons [22]. Ghosh et al. used fluorescent camera imaging of calcium-indicator loaded astrocytes to measure responses over large expanses in the somatosensory cortex [97]. They found that astrocyte calcium activity could effectively be used to map the boundaries of hindpaw and forepaw representations. The spatial correspondence of neural maps and astrocyte responses has been evaluated at higher resolution in the visual cortex of the ferret. Schummers et al. [73**] found that, at single-cell resolution, astrocyte activity in the visual cortex is matched to the activity of nearby neurons in the orientation preference map. Importantly, this relationship holds true at locations near the pinwheel center of the map, suggesting that the scale of neuron-astrocyte interactions is on the order of a few tens of microns.

Although the functional importance of gap-junction-coupled networks remains to be clarified, dye-coupling experiments in astrocytes located in the glomeruli and barrels reveals preferential connection with astrocytes located within the same processing unit and little coupling with neighboring units (OG or barrel) (Figure 1c; [85,87]). This raises the possibility that astrocyte modulation of neural activity (see below) is spatially constrained by neuronal maps.

Role of astrocyte and neural population dynamics

While most studies of astrocyte-to-neuron communication have focused on the effects on single neurons, several lines of study have proposed a role for astrocytes in the synchronization of neural population activity, ranging from local clusters of neurons to large-scale events, recognizable with EEG recordings. The cerebral cortex typically undergoes large-scale transitions between so-called UP-states and DOWN-states. The alternation between these two quasi-binary states has been observed in cortical brain slices, anesthetized and awake preparations [98]. At a single-cell level UP-states are characterized by depolarized (10–20 mV) membrane potential, compared to DOWN-states. At the network-level, the transition between states is synchronized among populations of neurons over large expanses (mm-scale) of cortical tissue, and can be readily recognized in field potential/EEG recordings, corresponding to low-frequency, large amplitude characteristics. The prevalence of DOWN states in the alert cortex is unclear, as are the physiological mechanisms.

The first suggestion that astrocytes regulate neural synchronization was the demonstration that astrocyte calcium signaling led to depolarization (SICs) in small clusters of hippocampal neurons that were synchronized on the time-scale of ~100 ms [48]. It was later reported that mice with deficient glutotransmission display abnormal slow-wave oscillations [99]. It was recently shown that astrocytes may specifically play a role in the initiation of UP states. Poskanzer and Yuste [100**] found that buffering calcium in the astrocyte network (by injecting calcium chelators into a single astrocyte, and presumably letting the buffer pass through the astrocyte network) reduced the number of UP-states. Further, stimulating individual astrocytes via depolarization or local glutamate uncaging increased the frequency of UP states. With pharmacological experiments, they concluded that both glutamatergic and purinergic signaling are required for the astrocyte-triggered increase in UP-state frequency. They further demonstrated that astrocyte calcium events are required for the initiation of this cascade. Thus, astrocytes may play pivotal roles in the neural synchronization phenomena that have long been recognized as hallmarks of different brain states.

To summarize, astrocytes are organized with domains that tile cortical tissue and are connected via gap junctions, thus enabling the potential to interact bi-directionally with neural activity at scales ranging from single synapses to large networks. Their activity is triggered by nearby synaptic activity, and they release gliotransmitters capable of modifying synaptic transmission on short and long time scales. In the cases where it has been investigated, astrocyte morphology and/or gap-junction coupling is biased in accordance with sensory map structure. But do they have any specific impact on the organization, development or plasticity of maps? Below we review some recent evidence.

**Role of astrocytes in activity-dependent developmental plasticity**

Multiple forms of synaptic plasticity have been suggested to play roles in refining the synaptic weights and connections of circuits in sensory cortex during early sensory experience. Though differences exist, similar mechanisms have been shown to be involved in the changes in response to manipulations of sensory input levels in both somatosensory and visual cortex [1,101]. Deprivation of specific sensory inputs, such as with whisker trimming or eyelid suture, gives rise to a dynamic set of synaptic and cellular processes, with the net result of a shift in the network representation away from the deprived inputs towards the spared inputs. In broad terms, these consist of a relatively rapid initial reduction in responses to the deprived inputs, followed later by increased responses to the intact inputs, as well as the deprived inputs. These processes have been mechanistically linked to hebbian LTP and LTD, STDP and homeostatic scaling [1,101]. There is evidence for a role for astrocytes in each of these phenomena.

**Astrocytes are involved in hebbian plasticity**

Hebbian plasticity modifies the strength of synapses that show coincident presynaptic and postsynaptic activity. In broad terms, it is based on a positive-feedback modulation where the synapses that are more active are strengthened while the less active are weakened. Because of their biophysical properties, NMDARs are a good coincident activity detector and they are involved in many forms of LTP, LTD and STDP [102]. Astrocytes are well equipped to modulate NMDAR activity which can be accomplished by regulating the availability of glutamate in the synaptic cleft through glutamate transporters (GLT-1 and GLAST [103]); releasing glutamate or D-serine [20**,60,63,104].

One of the pioneering works that analyzed the role of astrocytes in mediating NMDAR-dependent plasticity was done by Yang et al. a decade ago [105]. In this work, LTP was induced in cultured hippocampal neurons only if they were in direct contact with astrocytes. When neurons were fed with astrocytic conditioned media, without direct contact with astrocytes, LTP was not observed unless exogenous D-serine was administrated, suggesting a pivotal role of D-serine in the NMDAR-dependent LTP. The role of astrocyte-released D-serine in LTP induction in situ has been most clearly demonstrated in hippocampal slices. Henneberger et al. [36**] found that LTP of CA1 fEPSPs was blocked by either, first, clamping the calcium concentration, second, blocking D-serine synthesis or third, blocking vesicular release with TTC light chain, in a single nearby astrocyte, and rescued by the exogenous application of D-serine. Furthermore, the spatial extent of the effects of astrocyte on LTP of fEPSPs was commensurate with the size of astrocyte domains, and depended on whether the manipulation was expected to extend through gap junctions (calcium-clamp) or not (TTC light-chain). The role of astrocytic D-serine in NMDAR-dependent LTP has also been reported in hypothalamus, cerebellum and cortex [21**,36**,60,106], in juvenile animals [21**,36**,60], in adults [21**,106], and in aged rats [107,108].

Changes in synaptic strength are known to parallel morphological changes in neurons [109]. In this sense D-serine plays a role in promoting synaptogenesis [110,111] because mice lacking the enzyme that produces D-serine (serine racemase) showed a decrease in neuronal dendritic complexity and spine density in the prefrontal [112] and in the barrel cortex [110]. The mechanism by which D-serine promotes the formation of new synapses could be explained through the regulation of the two synaptogenic neurotrophic factors, TGF-β1 and BDNF, because blocking the synthesis of (or enzymatically degrading) D-serine led to changes in the production of these factors [110,111].

Long-term depression, the weakening of less active synapses, is also strongly involved in activity-dependent cortical plasticity. LTD has been proposed to be responsible for the rapid reduction of responses following reduction in sensory input in both barrel and visual cortices [101,113–115]. In rodent barrel cortex, the typical mature receptive fields are characterized by a spiking response primarily to the principle whisker (PW; the anatomically corresponding whisker), with minimal response to adjacent whiskers. Early in development, the receptive fields are poorly defined, and typically include responses to both the PW and one or more adjacent whiskers. In a use-dependent manner, these receptive fields are refined. Experimental manipulations of the activity levels of different whiskers can modify the receptive fields in a manner consistent with the notion of competition between neighboring whiskers for representation in cortical receptive field maps. In later stages of development, after the initial refinement phase, maps remain plastic, and a deprivation of sensory input leads to a rapid reduction in sensory-evoked response magnitude in layer 2/3. Both the initial refinement phase, and the...
deprivation-induced plasticity, have been proposed to depend on spike-timing-dependent long-term depression (st-LTD), based on the similarity of developmental time-course, pharmacological profile, and laminar specificity [116,117]. In the visual cortex of mice, a similar picture has emerged in the case of ocular dominance (OD) plasticity. In a monocular-deprivation protocol, the depression of layer 2/3 responses to the deprived eye has also been proposed to be attributable to LTD [113–115,118].

In both visual and somatosensory cortex, LTD, and visually driven response depression, depend on the activation of cannabinoid CB1-type receptors [119,120]. Until recently, the cellular localization of the CB1 receptors responsible for these phenomena was not clear. It has now been shown that these receptors are located on astrocytes, which serve as an intermediate step in a complex interaction at the synapse [121]. The first suggestion came from the finding that spatial working memory and hippocampal LTD are absent in mice lacking CB1 receptors in astrocytes, but not in neurons [122]. Additionally, Min and Nevinian [123–125] showed that activation of layer4-to-layer2/3 synapses in barrel cortex leads to increased astrocytic calcium events, specifically with stimulation protocols that elicit st-LTD. This increase in calcium events is dependent on CB1R activation. Furthermore, blocking calcium signaling in astrocytes prevents st-LTD in neighboring neurons, whereas pairing artificial astrocyte calcium activity with pre-synaptic activity can induce LTD. One interesting conclusion from this study is that astrocytes may act as a ‘memory-buffer’, using slow chemical signaling pathways to keep track of the recent activity of neighboring synapses over the time-scale of minutes.

Astrocytes are involved in homeostatic plasticity
The increase in responsiveness that constitutes the second phase of the sensory plasticity is thought to result in part from a homeostatic control mechanism [124], perhaps related to the phenomenon referred to as synaptic scaling. Such scaling can be induced in vitro by reducing activity levels pharmacologically for periods of hours to days [125,126]. Interestingly, the ability to express this form of plasticity depends on the presence of tumor necrosis factor alpha (TNFalpha) [127]. In these slice-culture preparations, the relevant source of TNFalpha has been shown to be astrocytes, suggesting that astrocytes serve as gate-keepers to homeostatic scaling [128]. Lending support to the prominent position of astrocytes in this signaling role of TNFalpha is the demonstration that TNFalpha is necessary for the normal kinetics of glutamate release from astrocyte small vesicles. In the absence of TNFalpha, astrocytes respond to GPCR stimulation with normal calcium signaling, but vesicle-mediated release of glutamate is less synchronous, and therefore exerts a moderated impact on nearby neurons due to a shift of the balance towards glutamate clearance, away from effects on glutamate receptors on neighboring neurons [129].

During OD plasticity, scaling up of visually responses following monocular deprivation exhibits many of the same features as synaptic scaling in vitro, suggesting that the same mechanisms may underlie the phenomenon. Of particular note, the scaling up of visual responses depends on the presence of TNFalpha. TNFalpha−/− mice exhibit normal early depression of responses to the deprived eye, but fail to exhibit the second scaling phase [130]. This demonstration lends strong support to the notion that cortical astrocytes play a critical enabling role in experience-dependent sensory map modifications during development.

Astrocytes regulate synaptic plasticity by other means
Although much emphasis has been placed on the role of astrocytic calcium signaling in the modulation of synaptic activity, there are other mechanisms by which astrocytes are capable of influencing synaptic circuits. Notably, the close apposition of astrocyte PAPs to synapses serves as a physical constraint on synaptic elements. The mobility of synapses, thought to be an important aspect of synaptic and developmental plasticity, is strongly regulated by astrocyte morphology [131,132]. Astrocytes have been shown to play an important role in the dramatic structural and synaptic remodelling in the hypothalamus that occurs during parturition and lactation [133,134].

On top of these structural interactions, the proximity of astrocytes to synapses is important in the spatial regulation of neurotransmitter in and around the synaptic cleft. The dynamics of the diffusion of neurotransmitter is a complex process involving the interaction of physical barriers to diffusion, and binding affinities of various synaptic and extra-synaptic receptors and transporters [103,135–138]. Astrocytes are key members in this process both because of their role in physically ensheathing synapses, and their dominant role in the uptake of glutamate by transporters (GLT-1 and GLAST). Astrocytic glutamate transporters are highly expressed during normal barrel development and their expression is restricted within the hollows where synaptic contacts are established [139]. Mice deficient in GLT-1 or GLAST have normal patterning of the barrels [139,140] and normal critical period [141]. However, the lack of these transporters decreases the glucose consumption induced by whisker stimulation [139,140] and impairs the lesion-induced refinement and remodeling of the barrel maps [141], suggesting that GLT-1 and GLAST are required for anatomical whisker map plasticity. The role of astrocytic glutamate transporters in barrel plasticity extends into adulthood, where artificially increased activity within a single whisker barrel leads to both an increase in expression of glutamate

transporters, and increased coverage of synapses by astrocyte PAPs [142]. This result suggests that astrocytes may additionally regulate synaptic efficacy via negative-feedback to modify the amount and/or duration of available glutamate subsequent to release. The finding that larger (and therefore stronger and more stable [109]) spines have a greater degree of astrocyte coverage [131] suggests the possibility that that astrocytes may regulate individual spines during the course of normal ongoing activity.

**Astrocytes and plasticity: CSPGs and PNN**

An essential factor that allows plasticity during development and limits it in the adult stage, is the amount and content of the extracellular matrix (ECM), and astrocytes are important determinant of this [143–145]. In visual cortex, chondroitin sulfate proteoglycans (CSPGs) reach maximum levels around the end of the critical period [146], when they condense around GABAAergic parvalbumin positive neurons in the form of perineuronal nets (PNNs) [146–148]. CSPGs restrain morphological changes necessary for plasticity, like the lateral movement of AMPA receptors [149]. Its degradation in the visual cortex of adult rats increases the motility of cortical spines and permits the induction of in vivo LTP [150]. Although there is some contradictory evidence in cats [151], degradation of CSPGs in visual cortex of the rat reactivates OD plasticity after monocular deprivation [152] which suggest CSPGs and PNN are involved in limiting the critical period and hence the plasticity in adults.

**Astrocytes and plasticity: TNC**

The astrocyte-derived glycoprotein Tenascin-c (TNC) is considered as a permissive factor for the morphological plasticity in developing brains as well as in adults [153,154]. In the barrel cortex of newborn mice, this protein is uniformly present in all cortical layers [155] and delineates the barrels field once the cortical map is established (P6). At the end of the critical period, TNC is no longer detectable in the barrel cortex [155,156] raising the possibility of a role in plasticity during this period. Despite its pattern and spatial distribution, TNC-deficient mice did not show differences in the histological structure of the barrel field [153,157]. However, a decrease in the numerical density of GABAAergic parvalbumin positive cells [158], higher whisker-evoked responses [158] and a reduction in barrel plasticity in adults was observed in these mice [153] confirming its role in modulating neuronal functions.

**Subcortical modulation**

During development, thalamocortical (TC) neurons located in the barreloids of the ventrobasal thalamic nucleus (VB) exhibit spontaneous oscillations that shape cortical barrels [159]. Small clusters of astrocytes in the VB also show coincident spontaneous calcium oscillations [46] and respond to either sensory fibers or corticothalamic stimulation with an increase in their calcium concentration [52]. Spontaneous and prolonged afferent stimulation induces SICs in TC neurons due to an increase of calcium-dependent release of astrocytic glutamate [53*], the magnitude of such currents (100 pA) would be capable of eliciting action potentials in TC neurons [160]. Interestingly, TC SICs are observed for up to an hour after cessation of the stimulus and do not depend on neuronal activity. Although the effect of this long-lasting SIC on synaptic function or the organization of the barrels remains to be clarified, this result suggests that neuronal activity can induce functional changes in astrocytes allowing them to ’store’ the history of neuronal activity. This raises the possibility that astrocytes could be involved in the experience-dependent plasticity of the thalamic topographic organization; in concordance with this, the long-term enhancement of astrocytic-neuron signaling is five times bigger during the critical period, compared with the first postnatal week [161].

**Astrocyte involvement in adult plasticity**

After the close of the classical critical period and through adulthood, cortical maps are more stable, and activity-driven changes in maps follow a more restrictive set of requirements. One way in which adult map plasticity can be induced is by substantially extended periods of modified sensory inputs [113,162–164]. Additionally, sensory inputs have been shown to drive plasticity of cortical receptive fields, and maps, when paired with activation of neuromodulatory systems, most notably acetylcholine [165,166]. Such changes can be quite robust, and have led to the notion that cortical plasticity post-critical period is gated. The similarities in the cortical changes induced by cholinergic activation and those induced by behavioral learning paradigms have led to the proposal that cholinergic systems may serve to highlight behaviorally relevant sensory stimuli and enhance their cortical representation [167].

The specific circuit mechanisms by which neuromodulatory systems enable adult plasticity have not been clearly defined. In the case of acetylcholine, the major cortical projections, arising from the nucleus basalis (NB) of the brainstem, are diffuse and likely to activate ionotropic nicotinic, and metabotropic muscarinic receptors non-specifically. These receptors are distributed throughout a range of subcellular compartments of a number of cell types [168–172]. Among them are astrocytes, which express high levels of both classes of receptors [170–175].

Several recent studies have focused attention on the possible role of AChRs expressed on astrocytes in mediating some of the roles of ACh in plasticity and learning. Lopez-Hidalgo et al. found that nicotinic enhancement of NMDA-dependent hippocampal synaptic transmission, as well as hippocampal-dependent memory, depend
critically on astrocyte-mediated release of d-serine [176]. They suggest that astrocytes play a necessary role in the influence of cholinergic systems on NMDA-mediated synaptic processing. In a complimentary study, Navarrete et al. [49] found that NMDA-independent enhancement of hippocampal synaptic transmission induced by stimulation of muscarinic AChRs also depends on astrocytes to be expressed. They observed that astrocyte calcium events were triggered by cholinergic stimulation in vivo, and that mAChR-induced plasticity was absent in a mouse line which lacks IP$_3$R2, and is therefore unable to translate mGluR activation to calcium elevations [177]. Interestingly, they found that this pathway also involves mGluRs which is likely to have a presynaptic role in this form of plasticity. Taken together, these studies have brought to light that astrocytes are likely to have roles in cholinergic modulation of multiple forms of hippocampal synaptic plasticity.

Two other recent studies have demonstrated more direct involvement in cholinergic effects in cortical plasticity. Takata et al. [214] paired electrical stimulation of the NB with sensory stimulation of the whiskers to investigate the role of astrocytes in ACh-mediated sensory plasticity. They found that sensory evoked potentials are enhanced by pairing with NB stimulation, a form of in vivo long-term synaptic potentiation. This phenomenon was absent in mice lacking IP$_3$R2-mediated calcium signaling, suggesting that it is also mediated in part by astrocyte activity. Furthermore, they suggested a mechanistic role for d-serine release by demonstrating that extracellular levels of d-serine are elevated following NB stimulation in wild-type mice but not in IP$_3$R2-deficient mice. These findings suggest the gating of cortical plasticity by cholinergic systems depends on astrocyte-mediated release of d-serine. In a more detailed study of ACh-induced plasticity of sensory responses in cortical circuits, Chen et al. [19] provided additional support for the importance of astrocytes, and importantly demonstrated stimulus-specificity of astrocyte actions. They found that pairing NB stimulation with visual stimulation led to a long-lasting enhancement of visual responses, which was dependent on mAChR activity, NMDAR-dependent, and absent in mice lacking normal calcium signaling mechanisms (IP$_3$R2-cKO mice). Notably, they found that visually evoked astrocyte calcium responses were enhanced by NB stimulation, consistent with a recent report that sensory-evoked responses in somatosensory cortex are enhanced in awake, compared to anesthetized states [141]. Most importantly, they demonstrated that the enhancement of visual responses was specific for the particular stimulus orientation that was paired with NB stimulation. This is the first demonstration that astrocytes are capable of mediating synapse-specific modifications within cortical circuits in vivo.

Together, this exciting emerging topic has placed astrocytes in a central position in the circuit mechanisms responsible for cholinergic regulation of cortical (and hippocampal) response plasticity (Figure 2), the most well documented example of adult activity-dependent cortical map reorganization [166,178,179]. It is worth noting that this may be only one of several such neuromodulatory systems that depend on astrocytes for its effects in cortex. Noradrenergic projections, which have also been demonstrated to enable adult cortical plasticity [180], have also been shown to directly activate astrocyte calcium signaling in vivo [181]. More generally, neuromodulatory systems have been implicated in a variety of roles in cortical information.
processing, related to arousal and attention (e.g. [182]);
the possible role of astrocytes in these phenomena warrants further investigation. These results certainly
do not argue against direct effects of ACh on cortical neurons. The ability to tease apart the circuit components (cell types) involved in ACh role on response plasticity has been enabled in no small part by using recently developed genetic tools to refine the picture provided by pharmacological experiments [177,183]. Additional studies will be required to dissect the inter-
action between these astrocyte-mediated effects, and direct neuromodulatory effects on different neuronal
subtypes, which also express AChRs.

Conclusion
In summary, their organization makes astrocytes well
suited to play a role in the spatial segregation of inform-
ation, as in the topographic organization of sensory
cortical maps. Cortical astrocytes have been shown to
respond to sensory stimulation with a high degree of
spatial specificity, commensurate with the resolution of
cortical maps. Astrocytes interact bi-directionally with
neuronal components of synapses, and they have been
shown to be important for synapse maturation and stabil-
ization, activity-dependent developmental plasticity, and
the gating of adult sensory cortical plasticity phenomena.
As such, astrocytes should not be ignored in future studies
of the circuit mechanisms responsible for cortical map
development, maintenance and plasticity. With that said,
the study of astrocytes at the level of brain circuits is only
recently gaining momentum, and many questions remain
to be addressed.

References and recommended reading
Papers of particular interest, published within the period of review,
have been highlighted as:
● of special interest
** of outstanding interest

1. Espinosa JS, Stryker MP: Development and plasticity of the
2. Huberman AD, Feller MB, Chapman B: Mechanisms underlying
development of visual maps and receptive fields. *Annu Rev
4. Muller CM: A role for glial-cells in activity-dependent central
   nervous plasticity – review and hypothesis. *Int Rev Neurobiol*
5. Muller CM, Best J: Ocular dominance plasticity in adult cat
   visual cortex after transplantation of cultured astrocytes.
6. Atwell D, Iadecola C: The neural basis of functional brain
   metabolic relationships: for better and for worse.* *Trends
   Neurosci* 2011, 34:76-87.
8. Nizam K, Uhliriova H, Tian P, Saiain PA, Cheng Q, Reznichenko L,
   Weidt KL, Steed TC, Siddhar VB, MacDonald CL, Cui J et al.: In
   vivo stimulus-induced vasodilation occurs without IP3
   receptor activation and may precede astrocytic calcium
   Cerebral blood flow modulation by basal forebrain or whisker
   stimulation can occur independently of large cytosolic Ca2+
10. Lamy CM, Chatton JY: Optical probing of sodium dynamics in
11. Cornellibe AL, Finkbeiner SM, Cooper MS, Smith SJ: Glutamate
    induces calcium waves in cultured astrocytes — long-range
12. Parpura V, Heneka MT, Montana V, Oliet SHR, Schousboe A,
    Haydon PG, Stout RF, Spray DC, Reichenbach A, Pannicke T,
    Pekny M et al.: Glial cells in (patho)physiology. *J Neurochem*
    2012, 121:4-27.
13. Ventura R, Harris KM: Three-dimensional relationships
    between hippocampal synapses and astrocytes. *J Neurosci*
14. Min R, Santello M, Nevan T: The computational power of
    astrocyte mediated synaptic plasticity. *Front Comput Neurosci*
    2012, 6:93.
15. Nedergaard M, Verhratsky A: Artifact versus reality-how
    astrocytes contribute to synaptic events. *Glia* 2012, 60:1013-
    1023.
    process and control synaptic information.* *Trends Neurosci*
17. Agulhon C, Sun MY, Murphy T, Myers T, Lauderdale K, Fiacco TA:
    Calcium signaling and gliotransmission in normal vs reactive
    astrocytes. *Front Pharmacol* 2012, 3:139.
18. Sun W, McConnell E, Pare JF, Xu Q, Chen M, Peng W, Lovatt D,
    Han X, Smith Y, Nedergaard M: Glutamate-dependent
    neuroglial calcium signaling differs between young and adult
   This paper demonstrates that mature astrocytes do not express mGluRS,
   and do now exhibit calcium elevations in response to neural activity. This
   paper highlights the risks in extrapolating from slice experiments to in vivo
   conditions.
    *Nucleus basalis-enabled stimulus-specific plasticity in the
    visual cortex is mediated by astrocytes.* *Proc Natl Acad Sci U S
20. Di Castro MA, Chuquet J, Liaudet N, Bhaukaularly K, Santello M,
    Bouvier D, Trett P, Volterra A: *Local Ca2+ detection and
    modulation of synaptic release by astrocytes.* *Nat Neurosci*
    2011, 14:1276-1284.
   This paper demonstrates that calcium transients are triggered in adult
   dentate gyrus astrocyte processes by spontaneous and evoked single
   synapse activation. These events are highly localized, exhibit rapid
   dynamics and are dependent on IP3 signaling. Blocking these pathways
   in astrocytes leads to a reduction in the reliability of adjacent synaptic
   release.
21. Takata N, Mishima T, Hisatsune C, Nagai T, Ebisu E, Mikoshita K,
    Hirase H: *Astrocyte calcium signaling transforms cholinergic
    modulation to cortical plasticity in vivo.* *J Neurosci* 2011,
    31:18155-18165.
   This paper provides the first evidence for the involvement of astrocytes in
   the cholinergic-induced plasticity in sensory systems.
    Nedergaard M: *Astrocytic Ca2+ signaling evoked by synaptic
23. Fiacco TA, Agulhon C, McCarthy KD: *Sorting out astrocyte
    physiology from pharmacology.* *Annu Rev Pharmacol Toxicol*
24. Panatier A, Vallee J, Haber M, Murai K, Lacaille JC, Robitaille R:
    *Astrocytes are endogenous regulators of basal transmission at
   This paper demonstrates that hippocampal CA1 astrocytes contain
   functional compartments that respond to single synapse activity. The
   failure probability of these events is only slightly less than that measured
electrophysiologically in adjacent neurons, suggesting that this is a very sensitive mechanism. They further show that basal synaptic activity in nearby synapses is modulated by a process that is dependent on astrocyte calcium fluctuations, and mediated by adenosine A2A receptors.


Using a well-designed method to buffer the increases in the intracellular calcium concentration in individual astrocytes, the authors block HFS-evoked LTP in a δ-serine-dependent manner. Interestingly, the block affects only nearby synapses which reinforce the astrocytic domain organization.


Although acetylcholine-induced plasticity in the visual cortex has been widely studied, this paper pointed to astrocytes as the main circuit component responsible for the neuronal plasticity of orientation-selective responses.


53. Pirttimaki TM, Hall SD, Parri HR: Sustained neuronal activity generated by glial plasticity. J Neurosci 2011, 31:7637-7647. This paper demonstrates that in the developing somatosensory thalamus, transient activation of afferent input pathways generated SICS in thalamocortical neurons that persisted for periods of hours. This form of plasticity was dependent on activation of astrocyte calcium pathways, and was mediated by activation of extrasynaptic NMDARs.


73. Schummers J, Yu H, Sur M: Tuned responses of astrocytes and their influence on hemodynamic signals in the visual cortex. Science 2008, 320:1638-1643. Using intrinsic optical imaging and two-photon laser scanning microscopy, the authors showed that astrocytes in the visual cortex of ferrets are highly tuned to visual stimuli with similar spatial distribution to neurons around pinwheel centers, but with sharper tuning curves.


In this paper, the authors reveal gap junction-mediated networks of astrocytes that depend on neuronal activity which arrangement is following the basic unit of processing in the olfactory bulb, the glomerulus.


Although several works had analyzed the role of astrocytes synchronizing networks, this elegant work combining calcium imaging and patch clamp recordings showed that single astrocyte manipulation is able to modulate UP network states. Moreover, this work reveals that astrocytes are responsible for the UP state generation in the cortex through the release of glutamate and ATP.


The authors show for the first time that postsynaptic release of endocannabinoids induced astrocytic release of glutamate that it is necessary for the induction of spike-timing-dependent depression in the developing sensory neocortex.


This paper reports that a mouse line deficient in TNF alpha, ocular dominance plasticity is abnormal. Specifically, the early reduction in response amplitudes is intact, but the subsequent increased responses to both eyes is absent, commensurate with the role of TNF alpha in synaptic scaling up in response to activity reduction.


