An Excitatory GABAergic Plexus in Developing Neocortical Layer 1

R. S. DAMMERMAN, A. C. FLINT, S. NOCTOR, AND A. R. KRIEGSTEIN
Department of Neurology and the Center for Neurobiology and Behavior, Columbia University College of Physicians and Surgeons, New York, New York 10032

Received 8 February 2000; accepted in final form 30 March 2000

Dammerman, R. S., A. C. Flint, S. Noctor, and A. R. Kriegstein. An excitatory GABAergic plexus in developing neocortical layer 1. J Neurophysiol 84: 428–434, 2000. Layer 1 of the developing rodent somatosensory cortex contains a dense, transient GABAergic fiber plexus. Axons arising from the zona incerta (ZI) of the ventral thalamus contribute to this plexus, as do axons of intrinsic GABAergic cells of layer 1. The function of this early-appearing fiber plexus is not known, but these fibers are positioned to contact the apical dendrites of most postmigratory neurons. Here we show that electrical stimulation of layer 1 results in a GABA_A−mediated postsynaptic current (PSC) in pyramidal neurons. Gramicidin perforated patch recording demonstrates that the GABAergic layer 1 synapse is excitatory and can trigger action potentials in cortical neurons. In contrast to electrical stimulation, activation of intrinsic layer 1 neurons with a glutamate agonist fails to produce PSCs in pyramidal cells. In addition, responses can be evoked by stimulation of layer 1 at relatively large distances from the recording site. These findings are consistent with a contribution of the widely projecting incertocortical pathway, the only described GABAergic projection to neonatal cortex. Recording of identified neonatal incertocortical neurons reveals a population of active cells that exhibit high frequencies of spontaneous action potentials and are capable of robustly activating neonatal cortical neurons. Because the fiber plexus is confined to layer 1, this pathway provides a spatially restricted excitatory GABAergic innervation of the distal apical dendrites of pyramidal neurons during the peak period of cortical synaptogenesis.

INTRODUCTION

A prominent feature of the early postnatal rodent somatosensory cortex is a transient, dense GABAergic fiber plexus confined to layer 1 (Del Rio et al. 1992; Lauder et al. 1986). Neurites of intrinsic GABAergic cortical neurons, including a subset of Cajal-Retzius (CR) cells and neurogliaform cells potentially contribute to this plexus (Cobas et al. 1991; Hestrin and Armstrong 1996; Imamoto et al. 1994; Marin-Padilla 1998). The only demonstrated extrinsic source of GABA, however, is a lamina-specific projection from the zona incerta (ZI) (Castro-Alamancos and Connors 1997; Lin et al. 1990). Incertocortical fibers ramify within the upper half of layer 1 where they make contact with the apical dendrites of pyramidal neurons (Lin et al. 1997). This incertocortical pathway develops at the same time as the major thalamocortical pathway from the dorsal thalamus (Catalano et al. 1991; Nicoletis et al. 1995) and both pathways are driven by sensory input from the trigeminal system (Carstens et al. 1990). The layer 1 GABAergic plexus is positioned to provide GABAergic activation during early stages of synaptogenesis. Whether, or to what degree, these fibers make functional synapses with developing cortical neurons is unknown. Additionally, while the incertocortical pathway contributes fibers to this plexus, the physiological properties of the cells providing this projection have not been examined.

Unlike the action of GABA in more mature cortex, GABA released from fibers in layer 1 is likely to have an excitatory effect on developing cortical neurons. Gramicidin perforated patch recordings have demonstrated that GABA_A− receptor activation produces depolarization of neonatal neurons (Owens et al. 1996). This is due to a high intracellular chloride concentration in immature neurons maintained by an active chloride transport mechanism (Clayton et al. 1998; Rivera et al. 1999). Because of the relatively high intracellular chloride concentration in immature neurons, GABAergic activation in layer 1 would be predicted to depolarize the apical dendrites of pyramidal neurons. Studies of the developing neocortex have indicated a role for membrane depolarization as a necessary step in associative processes including paradigms involving synaptic plasticity and the conversion of “silent” synapses to functional synapses (Feldman et al. 1998; Isaac et al. 1997). Given the presumed paucity of AMPA mediated excitatory synaptic plasticity in neonatal cortex (Isaac et al. 1997; Kim et al. 1995), the synaptic source for depolarization at this age is unclear. A widespread excitatory GABAeric projection within layer 1 could provide the membrane depolarization necessary to induce long-term changes in synaptic efficacy.

Here we characterize the postsynaptic current resulting from selective stimulation of layer 1. We find, in all neonatal pyramidal cells examined, a GABA_A−-mediated depolarizing response that can often initiate action potential firing. This GABA_A−-mediated postsynaptic current (PSC) could be elicited by electrical but not chemical stimulation of layer 1 and could be elicited by electrical stimulation at relatively large distances from the recorded cells. These observations suggest that the presynaptic source furnishing the excitatory GABAergic response is provided by fibers of passage rather than intrinsic layer 1 cells. The only described extrinsic GABAergic innervation of developing rodent somatosensory cortex is provided by the ZI. We therefore identified incertocortical projection neurons which could contribute to the presynaptic source of the evoked response, and found them to be physiologically mature at neonatal stages and to exhibit high rates of spontaneous synaptic plasticity.

The costs of publication of this article were defrayed in part by the payment of page charges. The article must therefore be hereby marked “advertisement” in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.
activity. The GABAergic plexus in layer 1 therefore provides robust, spatially restricted excitation of pyramidal cells at early stages of development.

METHODS

Whole cell recording

Sprague-Dawley rats were used for all electrophysiological experiments, and the day of birth was defined as postnatal day 0 (P0). Standard methods were used for preparation of vibratome slices as previously described (Blanton et al. 1989). A sapphire blade (Delaware Diamond Knives, Wilmington, DE) was used to maintain the fragile layer 1 in early postnatal slices. The extracellular artificial cerebrospinal fluid (ACSF) solution contained (in mM): 124 NaCl, 2.5 KCl, 1.25 NaH₂PO₄, 1 MgSO₄, 2 CaCl₂, 25 NaHCO₃, and 20 glucose, pH 7.4 at 25°C, and was bubbled with 95% O₂-5% CO₂. Borosilicate IR-DIC: cells were recorded as assessed by the presence of the following under junction potentials were determined and corrected for using the computer running Pulse v. 8.0 software (Heka Electronic). Liquid (Heka Electronic, Lambrecht, Germany) controlled by a Macintosh interference contrast videomicroscopy (IR-DIC) using an Olympus Patch clamp recordings were obtained under infrared differential [Cl].

Molecular Probes (Eugene, OR) was injected (1 μL) for 10–30 min prior to recording. Perforated patch recordings were obtained as previously described (Flint et al. 1999). Cells were loaded with the calcium indicator dye fluo-3-AM (Molecular Probes,15 μM) and imaged under epifluorescent illumination with appropriate filters. Excitation light exposure was minimized using neutral density filters and a shutter in the light path. Images were acquired with a cooled CCD camera (Dage) using NIH Image software on a Macintosh computer. Changes in fluorescence were converted to ΔF/F format according to standard calculations (Owens et al. 1996).

RESULTS

Whole-cell patch clamp recordings were made from pyramidal neurons in coronal cortical slices obtained from neonatal rats. Bipolar stimulation of layer 1 (Fig. 1A) induced fast monophasic PSCs in recordings from pyramidal cells in layers 2–6 (n = 70 cells, 14 cells in each lamina). The layer 1-evoked PSC had a latency to onset of 3.9 ± 0.2 ms (mean ± SE), a latency to peak of 9.0 ± 2 ms, and decay constant τ = 56.7 ± 4.2 ms. The latency of this response is similar to that of the well-characterized, monosynaptic thalamocortical synapse at this early age recorded at room temperature (Kidd and Isaac 1999). During the first postnatal week (P0–7), the layer 1-evoked PSC was completely and reversibly abolished (>95% reduction in peak amplitude) by the GABA_A receptor antagonist bicuculline (50 μM, n = 35, Fig. 1A), demonstrating that, in developing layer 1, GABAergic fibers provide the major input to neocortical neurons. We found that the GABAergic layer 1-evoked PSC could follow repetitive stimulation in the 0.1–5 Hz range without signs of fatigue including increased failure frequency or diminished peak amplitude (Fig. 1A).

In contrast to our findings in neonates, stimulation of layer 1 in the adult cortex has been reported to produce a predominantly glutamate-mediated response in cortical pyramidal neurons (Cauller and Connors 1994). We therefore examined developmental changes in the response to layer 1 stimulation. There was no glutamatergic contribution to the layer 1-evoked PSC from P0–7, as the PSC was entirely abolished by bicuculline at these ages (n = 35, Fig. 1A) and was unaffected by the glutamatergic antagonists DNQX (10 μM) and d-AP5 (50 μM) (n = 10). By contrast, in recordings made after P11 (P11–35), we found a consistent contribution to the layer 1-evoked PSC by ionotropic glutamate receptors (n = 15) as previously described (Cauller and Connors 1994). Detection of a persistent bicuculline-sensitive GABAergic component, however, was possible at these later ages in the presence of the nonselective glutamate receptor antagonist, kynurenic acid (1

Calcium imaging

Measurements of relative changes in [Ca²⁺], were made using epifluorescence microscopy of neocortical slices as previously described (Flint et al. 1999). Cells were loaded with the calcium indicator dye fluo-3-AM (Molecular Probes,15 μM) and imaged under epifluorescent illumination with appropriate filters. Excitation light exposure was minimized using neutral density filters and a shutter in the light path. Images were acquired with a cooled CCD camera (Dage) using NIH Image software on a Macintosh computer. Changes in fluorescence were converted to ΔF/F format according to standard calculations (Owens et al. 1996).

Perforated patch recording

Perforated patch recordings were obtained as previously described (Owens et al. 1996). Briefly, patch pipettes were filled with a KCl-based filling solution (as above), to which gramicidin (Calbiochem, San Diego, CA) was added on the day of the experiment (solution kept on ice). The tip of the electrode was filled with 40 mg/ml gramicidin and the remainder of the electrode was filled with 10 mg/ml gramicidin. Following tight seal formation, access was allowed to develop for 10–30 min prior to recording.

Retrograde fluorescent microsphere labeling

A solution of rhodamine-conjugated microspheres (FluoSpheres, Molecular Probes, Eugene, OR) was injected (1 μL) onto the surface of the cortex beneath the meninges of P2/3 postnatal rat pups under hypothermic anesthesia. The surgical wound was closed using New Skin (Medtech, Jackson, WY), and the pups were killed at P6/P7 to prepare coronal diencephalic slices for patch clamp recording. The zona incerta was identified at low power (4 × objective), and cells containing fluorescent microspheres were identified under high power (60 × objective) prior to patch clamp recording with a filling solution containing 1–2% Lucifer yellow (Sigma, St. Louis, MO). Lucifer yellow and FluoSphere (rhodamine) fluorescence were recorded with a cooled CCD camera (Dage, Stamford, CT) using epifluorescence and appropriate filters during recording.
mM) or the combination of DNQX and D-AP5 (n = 5 P11 pyramidal cells, n = 5 P35 pyramidal cells). To determine the relative contribution of ionotropic glutamate and GABA<sub>A</sub> receptor activation to the layer 1 evoked response, the sodium channel antagonist QX-314 (2-[Diethylamino]-N-[2,6-dimethylphenyl]acetamide) was added to the intracellular solution to prevent escape spikes, which were otherwise often observed even at low stimulus intensities. At P11 (n = 5 pyramidal cells in 3 slices) 82 ± 4% (SD) of the evoked response was reversibly blocked by a combination of DNQX and D-AP5. The response that persisted in the presence of these glutamate antagonists was reversibly blocked (>95% reduction in peak amplitude) by bicuculline (Fig. 1A). Thus, during the second postnatal week, the GABAergic component of the layer 1 response represented approximately 18% of the total response.

Electrical stimulation of cortical lamina other than layer 1 produces large polysynaptic currents predominantly mediated by glutamate (Burgard and Hablitz 1993; Flint et al. 1997; Kim et al. 1995). This was confirmed in recordings with the stimulation electrode placed within layer 3/4 (Fig. 2A). Evoked GABA receptor-mediated synaptic responses have generally not been described during the first postnatal week (Agmon and O'Dowd 1992; Burgard and Hablitz 1993; Kim et al. 1995; Luhmann and Prince 1991). Low intensity stimulation in the subcortical white matter during the first postnatal week produces a purely glutamatergic synaptic response (Agmon and O'Dowd 1992; Burgard and Hablitz 1993; Kim et al. 1995); however, more intense white matter stimulation can recruit a GABA<sub>A</sub> receptor-mediated polysynaptic response which fatigues rapidly on repeated stimulation (Agmon et al. 1996).

Unlike the inhibitory effect of GABA<sub>A</sub> receptor activation in adult cortical neurons, activation of GABA<sub>A</sub> receptors depolarizes developing postnatal pyramidal cells because immature neurons have relatively high [Cl<sup>-</sup>] (Owens et al. 1996). The reversal potential of the layer 1-evoked PSC depends on the concentration of chloride in the whole-cell recording pipette (see Fig. 2A), as expected for a response mediated by GABA<sub>A</sub> receptors. To test whether the GABAergic layer 1-evoked PSC is excitatory, we used the gramicidin perforated patch recording method that leaves [Cl<sup>-</sup>] undisturbed (Kyrozis and Reichling 1995) (Fig. 2B). Focal stimulation of layer 1 in the first postnatal week produced a depolarizing excitatory postsynaptic potential in all cells recorded (n = 6/6), and in several cells (4/6) stimulation of this pathway led to action potential discharge (Fig. 2C). Therefore the GABAergic fiber plexus in layer 1 provides an early excitatory input to pyramidal cells in the developing neocortex.

To confirm that we were exclusively stimulating fibers in layer 1, we employed a slice preparation in which layer 1 was surgically isolated (Cauller and Connors 1994) (Fig. 3A). In this slice preparation, where only a band of layer 1 connected the stimulus site to the region of cortex containing the recorded cell, GABAergic PSCs persisted (n = 5/5, Fig. 3A). Stimulation of layer 1 at increasing distances from the recording site (up to 5 mm) in this preparation and in intact slices yielded GABAergic PSCs that were indistinguishable from those ob-
This indicates that GABAergic fibers project over relatively long distances in layer 1. Intrinsic GABAergic layer 1 cells, including Cajal-Retzius and neurogliaform cells, have axons that ramify locally and extend up to approximately 700 μm within layer 1 (Hestrin and Armstrong 1996; Zhou and Hablitz 1996a). Thus intrinsic cortical neurons are unlikely to contribute significantly to the layer 1 response evoked by stimulation at distances of several millimeters.

In a further attempt to examine the contribution of intrinsic layer 1 neurons to the evoked layer 1 response, we used focal applications of glutamate to stimulate intrinsic cells of layer 1 but not incertocortical axons. We have previously demonstrated that focal application of glutamate in cortical layers other than layer 1 evokes a barrage of GABAergic PSCs in developing cortical pyramidal neurons (Owens et al. 1999). As expected, glutamate applied within 250–500 μm of recorded P5–6 layer 5 pyramidal cells (n = 7/7 from 4 slices) resulted in a barrage of PSCs (Fig. 3). In these same cells, glutamate application at multiple sites in layer 1, however, failed to elicit
an increase in PSC frequency above the baseline frequency of 0.2 PSC/sec, while bipolar electrical stimulation of these same sites in layer 1 resulted in the characteristic monophasic PSC (Fig. 3C). This observation, in conjunction with the ability to evoke a response by distant electrical stimulation, suggests that the presynaptic source eliciting this PSC is provided largely by GABAergic fibers of passage in layer 1 rather than by intrinsic layer 1 cells (Chai et al. 1988; Sandkühler et al. 1988).

The GABAergic fiber plexus in developing layer 1 contains axon projections from GABAergic neurons of the ZI (Imamoto et al. 1994; Lin et al. 1990). While the physiological properties of neonatal layer 1 neurons have been examined (Hestrin and Armstrong 1996; Kim et al. 1995; Zhou and Hablitz 1996b), the properties of neonatal ZI cells, including incertocortical projection cells, have not been studied. Cytochrome oxidase staining of the ZI suggests that this nucleus has high metabolic activity at early developmental stages (Nicoletis et al. 1995). We therefore assessed the physiological maturity of incertocortical projection neurons by patch clamp recording. Incertocortical projection neurons in the neonatal (P6/7) ZI were identified by two complementary approaches: 1) observation of their characteristic spindle-shaped morphology (Nicoletis et al. 1995) by IR-DIC microscopy and dye-filling with Lucifer yellow, and 2) retrograde labeling of individual incertocortical projection neurons with fluorescent microspheres (Figure 4A) (Katz et al. 1984). Whole cell recordings from spindle-shaped and fluorescent microsphere-labeled incertocortical projection neurons (Fig. 4B) revealed high rates of spontaneous synaptic events, spontaneous action potential discharge, and occasional burst firing (n = 22, Fig. 4C). ZI neurons had the following mean physiologic membrane properties: $V_m = -48.4 \pm 1.9$ mV (mean ± SE), $R_{input} = 595 \pm 60$ MΩ, $\tau_{mem} = 73.3 \pm 8.9$ ms, and $dV/dT$ of the rising phase of the action potential = 32.4 ± 3.9 mV/ms. Therefore projection neurons of the ZI, like those of the dorsal thalamus (Perez Velazquez and Carlen 1996), are spontaneously active and functionally well-developed by the first postnatal week. We used epifluorescence calcium imaging methods in neonatal brain slices to sample simultaneous activity in large numbers of incertal cells. Consistent with the high levels of local activity inferred from our patch clamp recordings, calcium imaging of fluo-3-loaded slices of the ZI showed high rates of spontaneous calcium transients (Fig. 4D, n = 200 cells showing multiple calcium

![Image](https://via.placeholder.com/150)

**FIG. 4.** Incertocortical projection neurons were identified for patch clamp recording by retrograde labeling with fluorescent microspheres. A: rhodamine-conjugated microspheres were surgically injected onto the surface of cortex and allowed to be transported retrogradely for 3–4 days in vivo. Acute slice preparations of the diencephalon had high-density accumulations of rhodamine in the dorsal thalamus (primarily in the ventrolateral (VL) and ventrobasal (VB) nuclei) and in the zona incerta (ZI). DIC was used to visualize the diencephalic slice. B: high power micrographs of an incertocortical projection neuron before (left) and during (middle) recording with a Lucifer yellow (LY) containing patch pipette. The superimposed Rhodamine fluorescence and LY images (right) demonstrate co-localization of retrogradely transported rhodamine-conjugated microspheres and LY introduced via the patch pipette. Scale bar indicates 8 μm. C: physiology of first week postnatal incertocortical projection neurons. Incertocortical neurons displayed relatively mature membrane properties and a high degree of spontaneous activity. Left: several firing patterns were observed among ZI neurons, including regular spiking cells with variable spike widths (top 2 traces) and single-spiking cells with narrow spike widths (bottom trace). Right: ZI neurons were highly active at rest, displaying burst discharges and regular trains of action potentials (top 2 traces). A high degree of spontaneous synaptic activity was observed, as shown here with the cell voltage clamped at −60 mV (bottom trace). D: calcium imaging also revealed a high degree of network activity present in the ZI. Upper left: a representative example of an acute slice of the ZI loaded with the calcium indicator dye fluo-3. Right: patterns of spontaneous change in epifluorescence (ΔF/F) are shown for the 3 cells circled on the left. Lower left: calcium transients were reversibly antagonized during bath application of extracellular solution containing 0 mM Ca$^{2+}$, 3 μM TTX, 1 mM La$^{3+}$ and 3 mM EGTA as depicted for a single incertal neuron.)
transients in 4 slices) and confirmed that the ZI is metabolically active in the perinatal period. These transients were completely and reversibly abolished \((n = 100\) cells in 2 slices) on bath application of the sodium channel antagonist tetrodotoxin and a combination of agents to prevent extracellular calcium entry \((0 \text{Ca}^{2+}, 1 \text{mM La}^{3+},\) and \(3 \text{mM EGTA})\) suggesting that these calcium transients are mediated by extracellular mechanisms rather than by release from intracellular stores.

**DISCUSSION**

In the present study, we have shown that the GABAergic plexus in rodent developing layer 1 forms functional synaptic contacts with pyramidal neurons from birth. GABAergic synapses in layer 1 provide localized excitatory GABAergic drive onto the distal apical dendrites of immature cortical neurons. The layer 1 GABAergic plexus is composed of axons from intrinsic layer 1 cells and projecting fibers from the ZI. Because GABAergic layer 1 neurons and GABAergic incertocortical neurons exhibit features of physiological maturity by this stage of development, this excitatory drive may help to sculpt early synaptic connections.

The relative contribution of incertal neurons and cortical layer 1 interneurons to the GABAergic synapse in layer 1 is difficult to assess. A slice preparation akin to the thalamocortical slice preparation (Agmon and Connors 1991), in which the intact incertocortical projection was reserved, would simplify this analysis. Unlike the thalamocortical projection, however, the more tortuous incertocortical projection cannot be preserved in a \(500 \mu\text{m}\) acute slice (R.C.S. Lin, personal communication). Nevertheless, the ability to evoke the layer 1 GABAergic PSC at distances of several millimeters argues against a contribution of intrinsic layer 1 interneurons whose axons have not been reported to extend that far (Hestrin and Armstrong 1996; Zhou and Hablitz 1996a). Responses evoked at these distances appear to support an extrinsic source, in particular the incertocortical pathway, which courses over relatively long distances within layer 1 and is the only described extrinsic GABAergic projection to the neonatal cortex (Lin et al. 1990; Nicolelis et al. 1995). Similarly, failure of glutamatergic stimulation of layer 1 to evoke a PSC in pyramidal cells suggests that the PSC evoked by bipolar stimulation arises from activation of fibers of passage rather than of intrinsic layer 1 cells (Chai et al. 1988; Sandkühler et al. 1988).

Several lines of evidence suggest that the layer 1 GABAergic synapse may play a role in early stages of cortical development. This plexus provides an example of synthetically released GABA exciting neocortical neurons. The excitatory actions of GABA are generally confined to immature cortical neurons which have a high intracellular chloride concentration (Owens et al. 1996). A chloride extruding activity develops postnatally, rendering intracellular chloride low and GABA inhibitory (Rivela et al. 1999). GABAergic synapses in layer 1 develop during corticogenesis, as revealed by GABA immunohistochemistry, with a peak density during neonatal stages and subsequent decrease into adulthood (Lauder et al. 1986; Nicolelis et al. 1995). These observations suggest a possible functional role of GABA in layer 1 to provide a depolarizing influence limited to early postnatal stages of cortical development.

Neuroanatomical studies show that incertocortical axons express multiple varicosities, presumed presynaptic specializations, as they course within layer 1 (Lin et al. 1997). Individual incertocortical fibers can thus contact the apical dendrites of multiple pyramidal neurons. This is in marked contrast with the intracortical ramification of specific dorsal thalamic afferents. These predominantly glutamatergic fibers originate in nuclei of the dorsal thalamus, course below the cortical plate, arborize within a restricted radial domain in layers 3 and 4 (Castro-Alamancos and Connors 1997), and synapse predominantly onto pyramidal neurons (Elston et al. 1997). These two pathways develop prior to birth (Catalano et al. 1991; Nicolelis et al. 1995), and their nuclei of origin both receive somatosensory information from the trigeminal nucleus (Carstens et al. 1990). Thus activity of specific dorsal thalamocortical afferents will influence local cortical regions, while activity of incertocortical afferents could influence multiple pyramidal cells dispersed throughout the developing cortex.

In the somatosensory cortex, the first postnatal week is a critical period for sensory plasticity in the formation of layer 4 “barrels,” the cortical representations of individual whiskers (Woolsey and Wann 1976). Sensory plasticity during the critical period has been shown to be dependent on the NMDA subtype of glutamate receptor (Schlaggar et al. 1993), which can act as a detector of coincident neuronal activity. Protocols shown to induce thalamocortical synaptic plasticity in neonates involve pairing of thalamocortical stimulation with direct experimental depolarization, presumably to relieve the magnesium blockade of NMDA receptors (Caird and Malenka 1995; Feldman et al. 1998; Isaac et al. 1997). However, the native source of depolarization is unknown, as many thalamocortical synapses at this age lack synaptic non-NMDA-type glutamate receptors and are therefore functionally silent synapses at negative membrane potentials (Isaac et al. 1997). In other brain regions it has been suggested that synaptic release of GABA in early development provides the necessary depolarization to induce long-term potentiation at silent glutamatergic synapses (Ben-Ari et al. 1997). During the first postnatal week GABAergic synapses in layer 1 are capable of depolarizing pyramidal cells. Further, the layer 1 evoked GABAergic excitatory postsynaptic current can sustain stimulation at rates exceeding those commonly used to induce synaptic plasticity at this age (Caird and Malenka 1995; Feldman et al. 1998). The GABAergic depolarization in layer 1 may, therefore, act in concert with thalamocortical activation during the critical period for somatosensory plasticity to produce an activity-dependent refinement of thalamocortical circuitry.

We thank D. Owens for helpful comments.

This work was supported by National Institute of Neurological Disorders and Stroke Grants NS-21223 and NS-35710 and a grant from the Robert Leet and Clara Guthrie Patterson Trust.

**REFERENCES**


WOOLSEY TA and WANN JR. Areal changes in mouse cortical barrels follow-