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Developmental decline in modulation of glutamatergic synapses in layer IV of the barrel cortex by group II metabotropic glutamate receptors

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Abstract

Previously, we demonstrated that group II mGluRs reduce glutamate release from thalamocortical synapses during early postnatal development (P7–11). To further examine the role of group II mGluRs in the modulation of somatosensory circuitry, we determined whether group II mGluRs continue to modulate thalamocortical synapses until adulthood and whether these receptors also modulate intra-cortical synapses in the barrel cortex. To address these issues, we examined the effect of the group II mGluR agonists on thalamocortical EPSCs and intra-barrel EPSCs in slices from animals of different ages (P7–53). We found that the depression of thalamocortical EPSCs by stimulation group II mGluRs rapidly declined after the second postnatal week. In contrast, adenosine continued to depress thalamocortical EPSCs via a presynaptic mechanism in young adult mice (P30–50). Activation group II mGluRs also reduced intra-barrel EPSCs through a postsynaptic mechanism in young mice (P7–11). Similar to the thalamocortical synapses, the group II mGluR modulation of intra-barrel excitatory synapses declined with development. In young adult animals (P30–50), group II mGluR stimulation had little effect on intra-barrel EPSCs but did hyperpolarize the neurons. Together our results demonstrate that group II mGluRs modulate barrel cortex circuitry by presynaptic and postsynaptic mechanisms depending on the source of the synapse and that this modulation declines with development.

Keywords

barrel cortex; metabotropic glutamate receptors; development

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INTRODUCTION

Group II metabotropic glutamate receptors (mGluRs) have been identified as new targets for the development of treatments for schizophrenia (Moreno et al., 2009; Fribourg et al., 2011; Matrisciano et al., 2012; Nasca et al., 2013), attentional deficits (Counotte et al., 2011; Pozzi et al., 2011), and depression (Nasca et al., 2013). However, many questions remain concerning the effects of group II mGluR activity on neuronal circuitry particularly in adult animals.

Immunohistochemical detection of group II mGluRs in the barrel hollows of adult rats (Ohishi et al., 1998) suggests that group II mGluRs could modulate synaptic excitation of the barrel cortex in adult rats. Furthermore, application of a nonselective group I and group II mGluR agonist (Cahusac, 1994; Taylor and Cahusac, 1994) or the selective group II mGluR agonist APDC (Cahusac and Wan, 2007) to the barrel cortex in vivo depresses the activation of cortical neurons by whisker stimulation indicating that group II mGluRs modulate thalamocortical excitation in the adult brain. However, several mechanisms could account for this depression. This depression could be due to group II mGluR modulation of thalamocortical synapses. In support of this possibility, we demonstrated that the activity-dependent stimulation of group II mGluRs decreases thalamocortical excitatory postsynaptic currents (EPSCs) in the barrel cortex during early postnatal development (Mateo and Porter, 2007). However, the modulation of thalamocortical synapses can change with development. For example, 5-HT_{1B} (Leslie et al., 1992; Bennett-Clarke et al., 1993; Laurent et al., 2002) and kainate (Kidd et al., 2002) receptors depress thalamocortical synapses only in the first two postnatal weeks. Group II mGluRs could follow a similar developmental pattern, since mGluRs can be lost during development (Baskys and Malenka, 1991; Ross et al., 2000; Doherty et al., 2004) or maintained into adulthood (Macek et al., 1996; Flavin et al., 2000).

The in vivo depression of barrel cortex activity by group II mGluRs could also be mediated by group II mGluR modulation of intra-barrel excitatory synapses or postsynaptic group II mGluR mediated hyperpolarization. In slices from P10–16 rodents, group II mGluR agonists decrease intra-cortical EPSCs by a postsynaptic mechanism (Egger et al., 1999) and hyperpolarize layer 4 neurons by activating postsynaptic K⁺ channels (Lee and Sherman, 2009). Either of these mechanisms could also mediate the group II mGluR depression of barrel excitation in adulthood.

Therefore, to better understand the role of group II mGluRs in the modulation of barrel cortex circuitry at later stages of development, we examined group II mGluR-mediated inhibition of thalamocortical afferents and intra-barrel excitatory synapses from the postnatal period to young adulthood. We demonstrate that both the presynaptic group II mGluR-mediated depression of thalamocortical synapses and the postsynaptic group II mGluR-mediated depression of intra-barrel excitatory synapses declines with development, whereas the postsynaptic hyperpolarization persists into adulthood.

METHODS

Thalamocortical slice preparation

Mice from postnatal days 7 to 55 were obtained from our breeding facility at the Ponce School of Medicine. In compliance with the National Institutes of Health (NIH) guidelines for the care and use of laboratory animals (Publication DHHS NIH 86–23), the Institutional Animal Care and Use Committee of the Ponce School of Medicine approved all procedures involving animals. Mice younger than P20 were anesthetized with halothane, whereas animals from P20–P55 were anesthetized with an intraperitoneal injection of 0.5 ml of pentobarbital (65 mg/ml). Anesthetized mice were decapitated, and the brains were removed and placed in ice-cold oxygenated artificial cerebrospinal fluid (ACSF) containing 126 mM NaCl₂, 3 mM KCl, 1.25 mM NaH₂PO₄, 2 mM CaCl₂, 1 mM MgSO₄, 26 mM NaHCO₃, and 20 mM glucose, bubbled with carbogen (95% O₂ and 5% CO₂). Brains slices (300 µm in thickness) with intact thalamocortical connectivity were prepared with a Vibratome 1000 Plus (Vibratome, St. Louis, MO) as previously described (Agmon and Connors, 1991; Porter et al., 2001) and incubated for one hour at room temperature in ACSF. To enhance neuronal survival (Schurr et al., 1995), brain slices from mice more than 20 days old were incubated with the NMDA receptor blocker, MK801 [(+)-5-methyl-10,11-dihydro-5H-dibenzo[a,d]cyclohepten-5,10-imine maleate] (10 µM).

Electrophysiology

Slices were incubated for one hour before being transferred to a submersion recording chamber mounted on a E600 upright microscope (Nikon Instruments, Melville, NY) and perfused with room temperature, carbogenated ACSF at a rate of 2–3 ml/min. Thalamocortical axons were stimulated with a bipolar tungsten microelectrode placed in the ventrobasal nucleus or internal capsule. EPSCs with latencies that varied by less than one millisecond were considered monosynaptic thalamocortical EPSCs and were included in the analysis (Agmon et al., 1996; Beierlein and Connors, 2002; Porter and Nieves, 2004). To avoid corticothalamic EPSCs which exhibit paired pulse facilitation (Beierlein and Connors, 2002), only EPSCs evoked by thalamic stimulation that displayed paired pulse depression were accepted as thalamocortical EPSCs (Fontanez and Porter, 2006; Mateo and Porter, 2007). To stimulate intra-cortical synapses, a microelectrode was placed in a single layer IV barrel close to the layer V border. EPSCs were then recorded from neurons within the same barrel.

EPSCs were recorded with a patch-clamp amplifier (MultiClamp 700A, Axon Instruments, Union City, CA) using glass micropipettes pulled on a Flaming/Brown micropipette puller (Sutter Instruments, CA). The positions of the stimulating electrode and micropipettes were adjusted to maximize EPSC amplitude.

Single neurons were visually identified using a video camera (Dage MTI, Michigan City, IN) connected to the Nikon E600 microscope. Whole-cell current-clamp and voltage-clamp recordings were done with patch pipettes filled with an internal solution containing 12 mM KCl, 140 mM KGluconate, 0.2 mM EGTA, 10 mM HEPES, 0.3 mM GTP, 0.4 mM ATP and biocytin (pH 7.3, 285 mOsm). In some experiments, 1 mM GDPβS was added to the

patch pipette solution to block any postsynaptic G protein-mediated effects of the agonists. Resting membrane potential was measured after achieving whole-cell configuration and cells with a membrane potential more depolarized than -50 mV were discarded. Recordings were not compensated for series resistance, but changes in series resistance were continuously monitored and recordings were eliminated from analysis if the series resistance changed by more than 15%. To examine the effect of mGluR and adenosine receptor activation on AMPA receptor-mediated EPSCs, we diluted the drugs in ACSF containing 100 μ M DL-2-amino-5-phosphonopentanoic acid (AP5) and 10 μ M bicuculline to block NMDA and GABA_A currents, respectively.

DCG IV (2S,2'R,3'R)-2-(2',3'-dicarboxycyclopropyl) glycine and (2R, 4R)-4-aminopyrrolidine-2,4-dicarboxylic acid (APDC), were purchased from Tocris (Ellisville, MO). Adenosine, GDP β S, bicuculline and AP5 were purchased from Sigma (St. Louis, MO).

Data analysis and statistics

Experiments were accepted for analysis if the effect of the agonist on the EPSCs reversed upon removal of the drug and there was no confounding di- or polysynaptic activity in the EPSCs. Data were analyzed using Clampfit (Axon Instruments, Union City, CA). Average EPSCs were taken from 10 consecutive traces including failures. The paired pulse ratio (PPR) was calculated from EPSCs evoked by paired stimuli as the mean amplitude of the EPSC evoked by the second stimulus (EPSC2) divided by the mean amplitude of the EPSC evoked by the first stimulus (EPSC1). The coefficient of variation of the EPSCs was calculated as the mean of 10 consecutive EPSCs divided by the standard deviation of the same 10 EPSCs. Data were analyzed using Student's *t*-tests or one-way ANOVA (Statistica, Statsoft, Tulsa, OK). After main effect, post hoc comparisons were done using Tukey HSD test. Statistical significance was set at $P < 0.05$. Values are reported as the mean $\pm$ the standard error of the mean (S.E.M.).

RESULTS

Group II mGluR inhibition of thalamocortical synapses declines after the second postnatal week

First, we determined whether the inhibition of thalamocortical synapses by group II mGluRs declines during development. Monosynaptic thalamocortical EPSCs were evoked by stimulating either the ventrobasal nucleus of the thalamus or the internal capsule in the presence of bicuculline and AP5 to block GABA_A and NMDA currents, respectively. We measured the sensitivity of the evoked thalamocortical EPSCs to the selective group II mGluR agonists APDC and DCG IV in slices from mice of ages P7 to P50 (Figure 1). All recordings were restricted to excitatory neurons that were identified by their characteristic discharge patterns in response to depolarizing current pulses as previously described (Mateo and Porter, 2007). Since we previously found that 10 μ M APDC and 10 μ M DCG IV inhibit thalamocortical EPSCs to the same degree (Mateo and Porter, 2007), we combined the data for both agonists (Figure 1A–C). Consistent with our initial findings (Mateo and Porter, 2007), bath application of 10 μ M APDC or 10 μ M DCG IV inhibited thalamocortical EPSCs

by half during the early postnatal period (P7–12, $45\% \pm 8$ inhibition, $n = 13$). As shown in Figure 1B–C, at later ages there was a dramatic decline in the group II mGluR-mediated inhibition (P20–30, $12\% \pm 5$, $n = 5$; P30–50, $2\% \pm 3$, $n = 5$). One-way ANOVA indicated a main effect of age ($F(2, 20) = 10.22$, $P = 0.0008$) and post-hoc comparisons indicated that the inhibition was greater at P7–12 than either P20–30 or P30–50 ($p < 0.014$). These experiments demonstrate that group II mGluRs depress thalamocortical EPSCs primarily during the early postnatal period.

Although APDC did not depress thalamocortical EPSCs in slices from adult mice (P30 to P50), application of APDC did change the holding current in these neurons. As shown in Figure 1D and 1E, bath application of $10 \mu\text{M}$ APDC reduced the amount of negative current needed to maintain the neurons at the holding potential of -60 mV ($17 \pm 7 \text{ pA}$ change from baseline, $n = 11$, $t = 2.78$, $p = 0.019$). Concurrent with the change in holding current, APDC also reduced the input resistance (Figure 1F, $76 \pm 9\%$ of baseline $n = 11$, $t = 2.81$, $p = 0.018$). The positive shift in the holding current and decrease in input resistance are consistent with hyperpolarization. Thus similar to the results recently found in the visual cortex (De Pasquale and Sherman, 2013), group II mGluRs continue to hyperpolarize barrel neurons in adult mice.

Adenosine continues to inhibit thalamocortical synapses in adult mice

To control for the possibility of a global reduction of presynaptic receptors on thalamocortical synapses during development, we examined the effect of adenosine receptors in young adult mice. Previously, we found that A1 adenosine receptors depress thalamocortical synapses in young (P13–21) mice (Fontanez and Porter, 2006). Therefore, we determined if the adenosine receptor-mediated depression of thalamocortical synapses also disappears with development. Figure 2 illustrates that bath perfusion of $100 \mu\text{M}$ adenosine inhibited thalamocortical EPSCs in slices from P30–50 mice by more than half ($54\% \pm 5$, $n = 4$, $t = 11.345$, $p = 0.0015$). In P13–21 mice, adenosine receptors depressed thalamocortical EPSCs by a presynaptic mechanism (Fontanez and Porter, 2006). To verify that the adenosine receptors in adult mice continued to modulate thalamocortical synapses presynaptically, we measured the effect of adenosine on the paired pulse ratio (PPR), the coefficient of variation of the EPSCs (CV), and the input resistance. Bath perfusion of adenosine showed a trend of increasing the PPR ($171\% \pm 25$, $n = 4$, $t = 2.81$, $p = 0.067$), increased the CV ($233\% \pm 22$, $n = 4$, $t = 6.15$, $p = 0.0087$), and decreased the input resistance ($74\% \pm 4$, $n = 4$, $t = 6.09$, $p = 0.009$), suggesting that adenosine produced both postsynaptic and presynaptic effects (Figure 2A–B). To determine which effects mediated the reduction in thalamocortical EPSCs, we measured the effect of adenosine on thalamocortical EPSCs while blocking postsynaptic adenosine receptor activity with intracellular $\text{GDP}\beta\text{S}$. In the presence of 1 mM of $\text{GDP}\beta\text{S}$ in the patch pipette (Figure 2C–D), adenosine still depressed thalamocortical EPSCs ($59\% \pm 0.6$ inhibition, $n = 3$, $t = -95.18$, $p = 0.001$) even though the input resistance did not change ($100\% \pm 3$ of baseline, $n = 3$, $t = 0.08$, $p = 0.94$). These results indicate that presynaptic adenosine receptors depress thalamocortical synapses in young adult mice and that not all the receptors located at thalamocortical terminals are lost with development.

Group II mGluR reduction of intra-barrel EPSCs also declines with development

To examine the developmental profile of group II mGluR-mediated modulation of intracortical synapses, we examined the effect of group II mGluR agonists on evoked intra-barrel EPSCs in cortical neurons from the early postnatal period to early adulthood. Intra-barrel excitatory fibers were stimulated with an electrode placed within a single barrel near the layer IV–V border (Figure 3A). EPSCs were recorded in the presence of bicuculline and AP5 to block NMDA and GABA_A currents, respectively. As shown in Figure 3B, C, 10 μ M APDC reduced by half the intra-barrel EPSCs ($56\% \pm 8$ inhibition, $n = 5$) in P7–12 mice. APDC continued to reduce the intra-barrel EPSCs until the fourth postnatal week (P20–30, $42\% \pm 13$ inhibition, $n = 6$). Similar to the thalamocortical synapses, the group II mGluR modulation of intra-barrel excitatory synapses also declined with development. In young adult mice (P40–47), APDC only slightly reduced the intra-barrel EPSCs ($11\% \pm 3$ inhibition, $n = 5$; Figure 3B–C). One-way ANOVA indicated a main effect of age ($F(2, 13) = 5.53$, $P = 0.018$) and post-hoc comparisons indicated that the inhibition was greater at P7–12 and P20–30 than at P30–50 ($p = 0.016$).

Group II mGluRs reduce intra-barrel EPSCs by a postsynaptic mechanism

Previously, we demonstrated that group II mGluRs are located both presynaptically on thalamocortical synapses and postsynaptic on the barrel neurons in early postnatal mice (Mateo and Porter, 2007). Therefore, to determine whether APDC reduced intra-barrel EPSCs by stimulating group II mGluRs located on the presynaptic axon terminals or on the postsynaptic membranes, we analyzed the effects of APDC on the PPR, coefficient of variation of the EPSCs, and input resistance (Figure 4A–C). Application of 10 μ M APDC reduced the input resistance ($82\% \pm 5$ of baseline, $n = 5$, $t = -3.77$, $p = 0.019$) and did not affect the PPR ($95\% \pm 13$ of baseline, $n = 5$, $t = 0.396$, $p = 0.71$) or the coefficient of variation of the EPSCs ($95\% \pm 13$ of baseline, $n = 5$, $t = 0.396$, $p = 0.71$) consistent with a postsynaptic mechanism. To confirm that APDC reduced the intra-barrel EPSCs via a postsynaptic mechanism, we blocked postsynaptic G protein signaling by adding 1 mM GDP β S to the solution in the recording electrode (Figure 4D–E). In barrel neurons dialyzed with GDP β S, APDC failed to reduce the intra-barrel EPSCs ($104\% \pm 7$ of baseline, $n = 4$, $t = 0.55$, $p = 0.62$) or the input resistance ($93\% \pm 5$ of baseline, $n = 4$, $t = -1.26$, $p = 0.30$). Taken together these results suggest that postsynaptic group II mGluRs modulate intra-barrel excitatory synapses during the first postnatal month.

DISCUSSION

In this study, we directly examined whether group II mGluRs continue to depress thalamocortical and intra-barrel glutamatergic synapses through to adulthood. Our results demonstrate that group II mGluR-mediated depression of thalamocortical synapses declines rapidly after the end of the early postnatal period. Similarly, early in development group II mGluRs depressed intra-barrel excitatory synapses via a postsynaptic mechanism. In adults, group II mGluRs had little or no effect on thalamocortical or intra-barrel excitatory synapses but did hyperpolarize the barrel neurons.

Developmental decline in group II mGluR-mediated inhibition

Application of a nonselective group I and group II mGluR agonist (Cahusac, 1994; Taylor and Cahusac, 1994) or the selective group II mGluR agonist APDC (Cahusac and Wan, 2007) to the somatosensory cortex in vivo depresses the activation of cortical neurons by whisker stimulation in adult rats indicating that group II mGluRs do modulate thalamocortical excitation in the adult brain. Our results show that the presynaptic inhibition provided by group II mGluRs at thalamocortical synapses declines rapidly after the first two postnatal weeks and; therefore, could not account for modulation seen in the adult barrel cortex in vivo (Cahusac and Wan, 2007). A similar developmental decline in presynaptic group II mGluR modulation of thalamocortical synapses was found in the primary visual cortex (De Pasquale and Sherman, 2013). Stimulation of group II mGluRs also depressed intra-cortical glutamatergic synapses via a postsynaptic mechanism. Although the mechanism is unknown, one possibility is that postsynaptic group II mGluRs depress the intra-barrel EPSCs by inhibiting adenylate cyclase, since the inhibition of PKA leads to internalization of AMPA receptors (Man et al., 2007). Our findings are consistent with previous studies demonstrating that the activation of postsynaptic group II mGluRs decreases in intra-cortical EPSCs in P12–14 (Egger et al., 1999) and adult rats (De Pasquale and Sherman, 2013). However, this mechanism is unlikely to account for the depression seen by Cahusac and colleagues in vivo since there was very little depression of intra-barrel EPSCs in the adult mice.

In addition to reducing intra-barrel EPSCs, group II mGluRs inhibit layer 4 neurons in the somatosensory, visual, and auditory cortices by a postsynaptic mechanism involving enhanced K^+ conductance through GIRK channels in P10–16 mice (Lee and Sherman, 2009). Consistent with recent findings in the primary visual cortex of adult mice (De Pasquale and Sherman, 2013), our results show that stimulating group II mGluRs also hyperpolarizes primary somatosensory neurons in slices from young adults. Therefore, the depression of whisker-evoked activity seen in adult rats (Cahusac and Wan, 2007) appears to be due primarily to hyperpolarization of the barrel neurons by postsynaptic group II mGluRs.

Developmental changes in the modulation of thalamocortical synapses

Early in development several receptors modulate glutamate release from thalamocortical terminals. In addition to group II mGluRs (Mateo and Porter, 2007), presynaptic kainate (Kidd et al., 2002) and 5-HT_{1B} (Laurent et al., 2002) receptors also decrease glutamate release from thalamocortical afferents during the first postnatal week. The presynaptic inhibition provided by these receptors is developmentally down-regulated after the first postnatal week (Kidd et al., 2002; Laurent et al., 2002). However, not all receptors, expressed on thalamocortical terminals, decline after the early postnatal period. Presynaptic GABA_B (Porter and Nieves, 2004), muscarinic (Gil et al., 1997), nicotinic (Gil et al., 1997), and A₁ adenosine (Fontanez and Porter, 2006) receptors continue to depress thalamocortical synapses after the early postnatal period. Although few studies have examined the presynaptic regulation of thalamocortical synapses in adults, our results indicate that at least A₁ adenosine receptors continue to depress thalamocortical synapses in young adults via a presynaptic mechanism. Therefore, the decline of presynaptic inhibition at thalamocortical

inputs represents a receptor-specific change rather than a global decline. This suggests that presynaptic group II mGluR, 5-HT_{1B}, and kainate receptors play an important role in barrel development, while GABA_B, muscarinic, nicotinic, and A₁ adenosine receptors play a more general role in maintaining glutamate release from thalamocortical terminals within normal limits. In the adult auditory cortex, presynaptic adenosine receptors restrict the induction of synaptic plasticity in thalamocortical inputs (Blundon et al., 2011). Whether adenosine receptors perform a similar function in the somatosensory cortex remains to be determined.

Potential roles of group II mGluR-mediated depression of thalamocortical synapses

Presynaptic group II mGluR may be important for thalamocortical synapse formation or refinement. At birth, the cortex of rodents is relatively immature and most of the thalamocortical axons have not yet arrived to their final cortical position (Agmon et al., 1993). Therefore chemotactic cues are very likely to participate in the proper development and elaboration of thalamocortical axons toward the final targets and to impose the periphery-related patterns in the barrel cortex. Previous studies implicate ionotropic (Schlaggar et al., 1993; Fox, 1996) and metabotropic (Hannan et al., 2001; She et al., 2009) glutamate receptors in the early development of the barrel formation. Although the exact role of group II mGluRs in barrel development remains to be determined, one can speculate that group II mGluRs may modulate growth cone movement given that cAMP facilitates growth cone extension (Rydel and Greene, 1988; Yamada et al., 2005), intracellular cAMP gradients promote growth cone turning (Lohof et al., 1992), and group II mGluRs inhibit cAMP formation (Tanabe et al., 1993; Schoepp et al., 1995; Wright and Schoepp, 1996). Consistent with this idea, barrel formation is disrupted (Salichon et al., 2001; Rebsam et al., 2002) and whisker-dependent tasks are impaired (Lee, 2009) by the excessive stimulation of presynaptic 5-HT_{1B} receptors on thalamocortical terminals, which also inhibit adenylate cyclase and thalamocortical synapses like group II mGluRs (Laurent et al., 2002). Furthermore, the blockade of group II mGluRs induces an abnormal outgrowth of hippocampal mossy fibers (Koyama et al., 2002).

In addition, group II mGluRs may regulate the plasticity of thalamocortical synapses, since the activation of these receptors induces LTD at hippocampal and cortical synapses (Yokoi et al., 1996; Li et al., 2002; Renger et al., 2002). Consistent with this possibility, thalamocortical afferents are most susceptible to LTD during the same postnatal two weeks that group II mGluRs modulate thalamocortical synapses (Feldman et al., 1998). Therefore group II mGluRs may modulate thalamocortical refinement during the early postnatal period by modulating growth cone movement or synaptic plasticity.

Conclusion

In conclusion, our results demonstrate that during postnatal development presynaptic group II mGluRs modulate thalamocortical excitation and postsynaptic group II mGluRs modulate intra-barrel glutamatergic synapses. After several weeks of development, this modulation is greatly reduced, but postsynaptic group II mGluRs continue to modulate barrel activity by hyperpolarizing barrel neurons. Therefore, both presynaptic and postsynaptic group II

mGluRs modulate glutamatergic transmission in the barrel cortex in a synapse-specific and age-dependent manner.

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Highlights

We examined the effects of group II mGluRs on cortical glutamatergic synapses in juveniles and adults.

Presynaptic group II mGluRs depressed thalamocortical synapses in the early postnatal period.

Early in development group II mGluRs depressed intra-barrel excitatory synapses via a postsynaptic mechanism.

In adults, group II mGluRs had little or no effect on thalamocortical or intra-barrel excitatory synapses.

Presynaptic adenosine receptors depressed thalamocortical synapses in adults.

Figure 1.

Group II mGluR depression of thalamocortical synapses declines after the second postnatal week. A) Time course showing that APDC or DCG IV reversibly reduced thalamocortical EPSCs in the animals aged P7–12. B) Average of 10 consecutive traces demonstrating that group II mGluR stimulation with APDC or DCG IV reduces thalamocortical EPSCs in slices from a P7 mouse more than in slices from a P15 or a P50 mouse. C) Percent inhibition of thalamocortical EPSCs by group II mGluR agonists, DCG IV and APDC, during development. D) Averages of ten consecutive traces before, during, and after 10 μ M APDC application during thalamocortical stimulation in slice from a P37 mouse indicating a positive shift in holding current. E–F) Time courses showing the average effect of APDC on holding current and input resistance in 11 neurons from young adult animals P34–50.

Figure 2.

Adenosine reduces thalamocortical EPSCs presynaptically in young adult mice. A) Time course showing that adenosine reversibly reduced thalamocortical EPSCs in the adult animals aged P30–50. B) Averages of ten consecutive thalamocortical EPSCs before and during 100 μ M adenosine application in a barrel neuron from a P37 mouse. C) Average effect of adenosine on thalamocortical EPSCs, PPR, CV, and input resistance in 5 neurons from P30–50 mice. D) Averages of ten consecutive thalamocortical EPSCs before and during 100 μ M adenosine in a barrel neuron from a P33 mouse recorded with an internal solution containing GDP β S. E) Average effect of adenosine on thalamocortical EPSCs and input resistance recorded in three neurons dialyzed with GDP β S from young adult animals. * $p < 0.05$

Figure 3.

Depression of intra-barrel EPSCs by group II mGluRs declines with development. A) Illustration of the placement of stimulating and recording electrode within the barrel cortex. B) Time course showing that APDC reversibly reduced intra-barrel EPSCs in the animals aged P7–P12. C) Averages of 10 consecutive traces showing intra-barrel EPSCs before and during 10 μ M of APDC in slices from a P12, P20 and P42 mouse. D) Average effect of APDC on intra-barrel EPSCs in slices from mice of different ages. * $p < 0.05$

Figure 4.

Group II mGluRs depress intra-barrel EPSCs by a postsynaptic mechanism. A) Averages of 10 consecutive traces showing paired intra-barrel EPSCs before and during 10 μ M of APDC. B) Averages of ten consecutive traces showing current evoked by 50 ms hyperpolarizing pulse in same neuron during baseline and application of APDC. Capacitive transients have been truncated for clarity. C) Average effect of 10 μ M APDC on intra-barrel EPSCs, PPR, CV, and input resistance in 5 barrel neurons from P20–30 mice. D) Averages of ten consecutive traces before and during 10 μ M APDC in a barrel neuron recorded with internal solution containing GDP β S. E) Average effect on intra-barrel EPSCs and input resistance by 10 μ M APDC in 4 barrel neurons dialyzed with GDP β S. * $p < 0.05$