A Dendritic $GABA_A$-Mediated IPSP Regulates Facilitation of NMDA-Mediated Responses to Burst Stimulation of Afferent Fibers in Piriform Cortex

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Studies in a number of cortical systems have shown that the NMDA component of the EPSP is strongly regulated by $GABA_A$-mediated inhibition. The present study explored the possibility that specificity in inhibitory circuitry could allow such regulation to occur during normal function without increasing the propensity for epileptiform bursting, which occurs with indiscriminate $GABA_A$ blockade. Specifically, the hypothesis was tested that a dendritic $GABA_A$-mediated IPSP is present which strongly modulates the NMDA component and can be activated independently of the somatic IPSP. The experiments were performed on slices of piriform cortex in which the NMDA component of the EPSP was pharmacologically isolated by bath-applied 6,7-dinitroquinoxaline-2,3-dione. A facilitation of NMDA responses to burst stimulation of afferent fibers is described, which required $GABA_A$ blockade and served as an assay for the presence of a functionally significant $GABA_A$ input. When bicuculline was applied focally in the somatic region, the feedback IPSP was blocked with little or no increase in the NMDA component of the response to burst stimulation of afferent fibers. In contrast, when bicuculline was applied focally in the dendritic region, the NMDA-mediated response to burst stimulation was facilitated with minimal effect on the somatic IPSP, confirming the hypothesis.

Key words: long-term potentiation; NMDA; $GABA_A$; inhibition; IPSP; pyramidal cells; cerebral cortex; piriform cortex; olfactory cortex

Studies in the hippocampus (Dingledine et al., 1986; Staley and Mody, 1992) and neocortex (Luhmann and Prince, 1990) have demonstrated that the NMDA component of EPSPs is accentuated by blockade of $GABA_A$-mediated inhibition. Such regulation of the NMDA component has implications for the control of long-term potentiation (LTP) and expression of epileptic activity. An important question is whether this modulation of the NMDA component by GABAergic processes is part of normal function or whether it can be induced only under experimental conditions. Uncertainty about the participation of this process in normal function stems from the fact that even partial indiscriminate antagonism of the fast GABAergic IPSP evokes epileptiform activity in most cortical systems (Scholfield, 1980; Gutnick et al., 1982; Traub et al., 1987). However, spatially segregated inputs to dendritic and somatic regions of pyramidal cells from different GABAergic circuits, as demonstrated in the hippocampal formation (Halasy and Somogyi, 1993; Han et al., 1993; Buhl et al., 1994), could allow such control. The rationale for this suggestion is that suppression of a dendritic IPSP could facilitate the NMDA component with less effect on the propensity for epileptic bursting than would suppression of a somatic IPSP. Somatic IPSPs, especially when mediated by feedback circuitry, can strongly suppress epileptiform bursting (Traub et al., 1987; Vu and Krasne, 1992), whereas dendritic inhibition would be expected to strongly modulate the NMDA component because excitatory synapses on pyramidal cells are confined to dendrites (Douglas and Martin, 1990). Selective suppression of dendritic inhibition without attenuation of somatic feedback inhibition therefore could provide a mechanism for regulation of NMDA-dependent processes.

To test this hypothesis, experiments were performed on piriform (olfactory) cortex. Previous study in piriform cortex (Kanter and Haberly, 1993) has shown that, as in the dentate gyrus (Tomasulo et al., 1993; Zhang and Levy, 1993), associative LTP or long-term depression (LTD) induced by coincidental activation of afferent and association fiber systems can be evoked only after $GABA_A$ blockade. Indirect evidence for piriform cortex also suggests that the NMDA component is enhanced by the GABAergic block that enables associative LTP (Kanter and Haberly, 1993). The present results reveal a $GABA_A$-mediated IPSP in the dendritic region that strongly modulates the NMDA component and that can be evoked independently of the $GABA_A$-mediated IPSP in the somatic region.

This work has been presented, in part, in abstract form (Kapur et al., 1992, 1993).

MATERIALS AND METHODS

Rats weighing ~200 gm were decapitated under ether anesthesia. A block of brain containing piriform cortex was removed and slices 500 μm thick were cut perpendicular to the cortical surface with a Vibratome (Lancer) in oxygen-saturated medium at 0–4°C. Slices were allowed to recover for 2 hr at room temperature before recording. The medium contained (in mM): 124 NaCl, 5.0 KCl, 2.4 $CaCl_2$, 1.3 $MgSO_4$, 26 $NaHCO_3$, 1.2 KH_2PO_4, and 10 mM D-glucose, equilibrated with 95% O_2/5% CO_2.

Recordings were taken from submerged slices continuously perfused with oxygenated medium at 30°C in the chamber described by Tseng and Haberly (1988). Dark-field illumination through a transparent base allowed visualization of all cortical layers for accurate placement of electrodes. Intracellular recordings were made from layer II pyramidal cells using glass micropipettes (60–90 MΩ) filled with 4 M potassium acetate.

Received April 21, 1995; revised Aug. 28, 1995; accepted Sept. 22, 1995.

This work was supported by Grant NS19865 from the NINDS (L.B.H.) and National Institutes of Health training Grant GMO7507 (E.D.K.). We thank Robert Pearce for helpful discussions and a critical reading of the manuscript. We thank Drs. L. Maître and H. Kaufmann of Ciba-Geigy Pharmaceuticals for their generous gift of CGP 35348.

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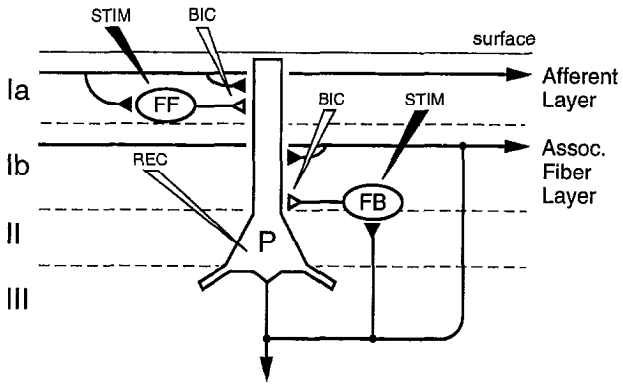

Figure 1. Lamination of piriform cortex, relevant inputs to pyramidal cells, postulated inhibitory circuitry, and placement of electrodes. Cell bodies of superficial pyramidal cells (*P*) are in layer II; their apical dendrites extend into layer I. Afferent fibers from the olfactory bulb are confined to layer Ia; association fibers (recurrent collaterals of pyramidal cell axons) are confined to layer Ib. Afferents synapse on distal segments of pyramidal cell apical dendrites and on GABAergic feedforward interneurons (*FF*). Association fibers synapse on proximal segments of pyramidal cell apical dendrites and on GABAergic feedback (*FB*) interneurons. It is postulated that output from feedforward interneurons is concentrated on apical dendrites of pyramidal cells, whereas output from feedback interneurons is concentrated in the vicinity of cell bodies. Responses were recorded intracellularly from pyramidal cell somata in layer II. Tungsten microelectrodes were placed in layer Ia for stimulation of afferent fibers and feedforward inhibitory circuitry, and in the deep part of layer Ib for stimulation of association fibers and feedback inhibitory circuitry. *BIC*, bicuculline-containing pipette; *REC*, intracellular recording electrode; *STIM*, stimulating microelectrode.

In some experiments the recording electrode also contained 50 mM QX-314, which blocks both sodium spikes and the $GABA_B$-mediated slow IPSP intracellularly (Connors and Prince, 1982; Nathan et al., 1990). In experiments that required a depolarized membrane potential, this was achieved by injection of current through the micropipette with a bridge circuit (Axoclamp 2A). Stimuli were 100 μsec shocks, delivered through bipolar tungsten microelectrodes placed under direct vision at the lateral border of the lateral olfactory tract (LOT) and layer Ia for stimulation of afferent fibers, and in the deep part of layer Ib for stimulation of association fibers (Fig. 1). Amplified responses were digitized and analyzed by computer.

Bicuculline methiodide (Sigma) was bath-applied or focally applied from a second micropipette by pressure injection (Pneumatic Picopump, WPI). D-2-Amino-5-phosphonovaleric acid (D-APV), D,L-2-amino-5-phosphonovaleric acid (D,L-APV), 2-hydroxysaclofen, 3-amino-propyl(diethoxymethyl)phosphinic acid (CGP 35348), and 6,7-dinitroquinoxaline-2,3-dione (DNQX) were bath-applied. D-APV was from Cambridge Research Biochemicals; DNQX and 2-hydroxysaclofen were from Research Biochemicals; D,L-APV was from Sigma; QX-314 was from Astra; and CGP 35348 was a gift from Ciba-Geigy.

RESULTS

The experiments were performed on neurons in layer II of piriform cortex. Based on results of previous studies with the same preparation in which dye injection was carried out (Tseng and Haberly, 1988), it can be assumed that most or all impalements were of superficial (layer II) pyramidal cells (Fig. 1). The experimental design took advantage of the fact that afferent fibers terminate exclusively on the distal-most segments of the apical dendrites of these cells (Fig. 1). The spatial separation of this input from cell bodies allowed the effects of dendritic and somatic disinhibition on the NMDA component of the afferent fiber-mediated responses to be compared. As in the hippocampus (Nicoll et al., 1990), responses to afferent fiber stimulation in the piriform cortex consist of fast and slow EPSPs mediated by AMPA and NMDA receptors, and fast and slow IPSPs mediated by $GABA_A$ and $GABA_B$ receptors (Tseng and Haberly, 1988; Haberly, 1990; Jung et al., 1990). In all experiments the NMDA component of the EPSP was isolated by blockade of the fast EPSP component with the AMPA receptor antagonist DNQX (Drejer and Honoré, 1988).

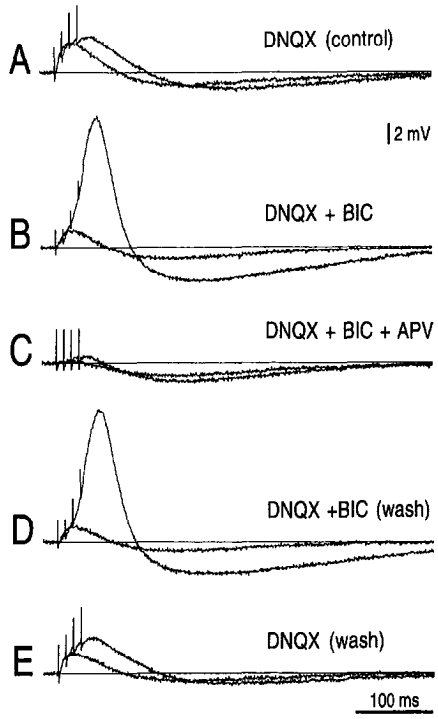

Figure 2. $GABA_A$ blockade facilitates responses to burst stimulation of afferent fibers. *A*, Superimposed responses to single pulse and burst of 4 pulses delivered to afferent fibers in DNQX at resting membrane potential (−68 mV). *Traces* are averages of three or more successive responses. *B*, Same as *A* but after addition of 10 μM bicuculline (*BIC*) to the bathing medium. Note that blockade of the fast IPSP by BIC induced a large increase in the response to the 4-pulse burst. *C*, Same as *B*, but after additional application of 100 μM D,L-APV. The response was largely blocked by APV, indicating that an NMDA-mediated EPSP underlies the large facilitated depolarizing component. *D*, After washout of APV. *E*, After washout of both APV and BIC. Time and amplitude calibration bars apply to all traces.

Facilitation of the NMDA component after $GABA_A$ blockade

Initial experiments performed with bath-applied bicuculline confirmed that the fast IPSP regulates the NMDA component in piriform cortex as in the hippocampus and neocortex. Four-pulse, 100 Hz bursts identical to those used to induce LTP in piriform cortex (Kanter and Haberly, 1990, 1993) were applied to afferent fibers in layer Ia to accentuate the NMDA component. Peak amplitudes of responses to these stimulus bursts in DNQX alone were typically larger than responses to single pulses by a factor of 3 or less (2.07 ± 0.17, mean ± SEM, n = 11). After bath application of 10 μM bicuculline, responses to 4-pulse bursts were greatly facilitated relative to single-pulse responses (7.32 ± 0.82, n = 8), as well as to 4-pulse responses in DNQX alone. A typical result is illustrated in Figure 2. In this experiment, a 4-pulse burst evoked a response that was 1.2× larger than the response to a single pulse when $GABA_A$ inhibition was intact (see superimposed responses to single pulse and burst in *A*). After bath application of 10 μM bicuculline, the response to the 4-pulse burst

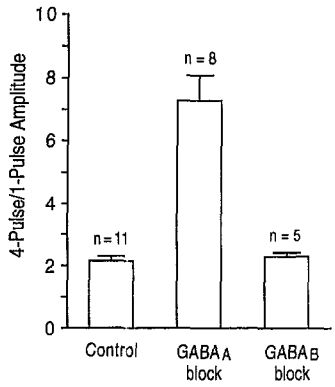

Figure 3. $GABA_A$, but not $GABA_B$ block induces facilitation of the response to burst stimulation of afferent fibers. Ratio of peak depolarization in response to 4-pulse burst to peak depolarization in response to single pulse under control condition (DNQX alone), after block of $GABA_A$-mediated inhibition alone by addition of 10 μM bicuculline, and after block of $GABA_B$-mediated inhibition alone by internal application of 50 mM QX-314 through the micropipette. The extent of facilitation by $GABA_A$ block is underestimated because suprathreshold responses were excluded. Error bars are SEMs. The increase after $GABA_A$ blockade is significant at $p < 0.001$.

was much larger than the response to the single pulse (approximately sevenfold). This action was reversible when the bicuculline was washed out (Fig. 2*E*). Responses to bursts were blocked by 90 ± 5% (mean ± SD, $n = 5$) in APV (15 mM D-APV or 30–100 mM D,L-APV), indicating that the depolarizing response in DNQX and bicuculline was dependent on the activation of NMDA receptors (Fig. 2*C*). Results from all experiments are summarized in Figure 3.

In contrast to the strong effect of $GABA_A$ blockade, blockade of $GABA_B$-mediated inhibition alone by inclusion of QX-314 in the recording pipette ($n = 5$) had no apparent effect on facilitation of the NMDA component (Fig. 3). A similar result was obtained with bath application of the $GABA_B$ antagonists, 2-OH saclofen ($n = 2$), or CGP 35348 ($n = 1$).

Polysynaptic activity is not required for facilitation of the NMDA component

In normal bathing medium, bicuculline induces a large facilitation of stimulus-evoked EPSPs in the piriform cortex by the induction of epileptiform responses (Scholfield, 1980). In the hippocampus, similar responses have been shown to be mediated by synaptic reverberation in the pyramidal cell population (Johnston and Brown, 1981; Traub, 1982). These bicuculline-induced epileptiform potentials can be recognized by their all-or-none character. Careful adjustment of stimulus strength in the present experiments failed to reveal all-or-none PSP components in facilitated burst responses in DNQX and bicuculline. This suggests that circuit-mediated regenerative positive feedback does not contribute to the facilitation of burst responses under present conditions in which the fast EPSP is absent. However, additional experiments were performed to confirm that polysynaptic activity does not underlie the facilitation. For these experiments, the divalent cation concentration was increased to a level greater than that at which epileptiform activity induced by bath-applied bicuculline is blocked (Kanter and Haberly, 1993). Highly facilitated burst responses were observed with a time course similar to that in normal bathing medium with Mg^{2+} elevated to 11 mM ($n = 4$) or Ca^{2+} to 8 mM ($n = 1$). A representative experiment with elevated Mg^{2+} is illustrated in Figure 4. In this experiment, intrinsic association

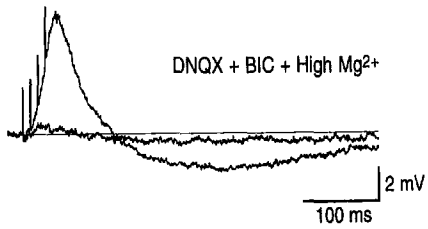

Figure 4. Facilitation of burst response by disinhibition is not blocked by elevation of threshold. Superimposed responses to single and 4-pulse stimuli applied to association fibers in bathing medium containing DNQX (20 μM) and bicuculline (10 μM), with Mg^{2+} elevated to 11 mM. Facilitation of the response to burst stimulation with threshold elevated by a high divalent cation concentration rules out a mechanism involving synaptic reverberation.

axons were stimulated in layer Ib to maximize the possibility for evoking polysynaptic activity.

Demonstration of a dendritic $GABA_A$ input that modulates NMDA facilitation

To test the hypothesis that there is an inhibitory process concentrated in the dendritic region that can modulate the expression of the NMDA component, bicuculline (1 mM) was delivered focally to the distal dendritic region or to the cell body region by pressure ejection from a micropipette. Fast EPSPs were blocked by bath-applied DNQX (20 μM), and slow IPSPs and action potentials were blocked by QX-314 in the recording electrode. The effects on responses to single-pulse stimuli and 4-pulse, 100 Hz bursts delivered to the afferent fiber layer were observed at resting membrane potential. The effects of local applications of bicuculline on the feedback IPSP evoked by stimulation in the association fiber layer (deep part of layer Ib) also were assessed at a depolarized membrane potential where it is of substantial amplitude. Figures 5 and 6 illustrate the results of this experiment in a representative cell.

After dendritic application of bicuculline, the response to a burst increased substantially (Figs. 5*A2*, 6*A*, *open circles*). During this increase in the burst response, the response to the single shock decreased (Fig. 5*A1*), consistent with block of the fast IPSP, which is depolarizing in sign at resting membrane potential in piriform cortex (Scholfield, 1978). Bath application of APV blocked the increase in the burst response (not illustrated), indicating that the NMDA component of the EPSP was facilitated as when bicuculline was bath-applied.

Facilitation of the burst response could occur with minimal effect on the feedback IPSP evoked from the association fiber layer (Figs. 5*A3*, 6*B*, *open circles*), indicating that the bicuculline did not spread appreciably to the vicinity of cell bodies (see below). After somatic application of a sufficient amount of bicuculline to block the IPSP evoked from the association layer (Figs. 5*B3*, 6*B*, *closed circles*), there was little change in single-pulse or burst responses to afferent layer stimulation (Fig. 5*B1,2*, Fig. 6*A*, *closed circles*).

Pooled results for six experiments are presented in Figure 7. Data from one experiment in which no facilitation could be obtained from injection of bicuculline at any depth were excluded from Figure 7. Differences between the effects of dendritically and somatically applied bicuculline on facilitation (Fig. 7*A*) and block of the feedback IPSP (Fig. 7*B*) were significant at $p < 0.001$.

The differential effect of bicuculline applied in dendritic and somatic regions on IPSPs evoked by stimulation in afferent and

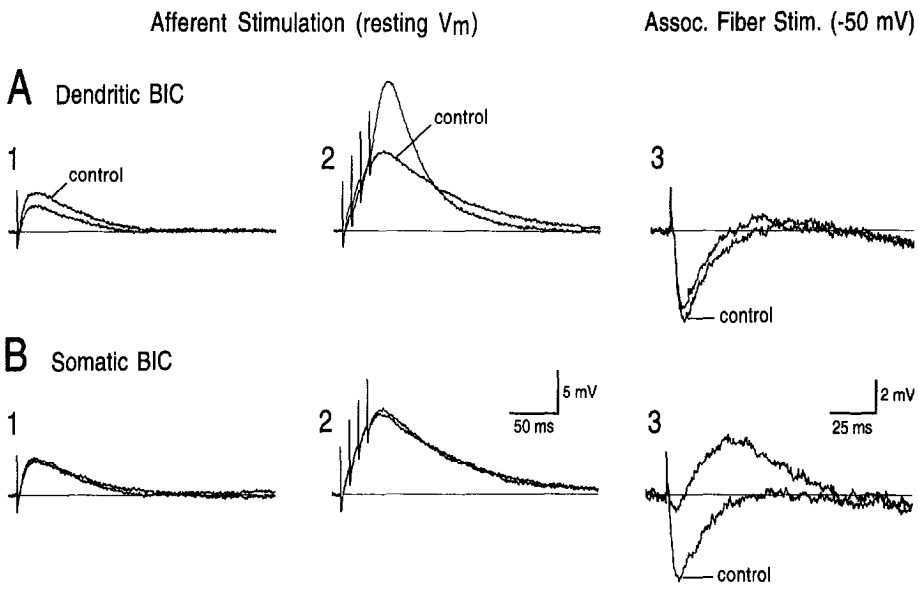

Figure 5. Blockade of dendritic but not somatic $GABA_A$-mediated inhibition enables NMDA burst facilitation. Bicuculline (*BIC*) (1 mM) was delivered focally at distal apical dendrites (layer Ia) or at the soma by pressure injection through a micropipette (300 msec pulses, 40 psi, 3 μm tip OD). Fast EPSPs were blocked throughout by bath-applied DNQX (20 μM). The $GABA_B$-mediated IPSP and Na^+ spikes were blocked by QX-314 (50 mM) in the recording pipette. Responses to afferent fiber stimulation (*A1,2; B1,2*) were recorded at resting membrane potential (−67 mV). Responses to association fiber stimulation were recorded at a depolarized membrane potential (−50 mV) to convert the fast IPSP to a large hyperpolarizing potential for easy visualization (*A3, B3*). All *traces* are superimposed averages of four consecutive responses (10 sec intertrial interval) before (*control*) and after bicuculline injection. *A*, $GABA_A$ blockade at the dendritic level. *A1*, Response to single afferent pulse was decreased by bicuculline. *A2*, Response to burst stimulation of afferent fibers (4 pulses, 100 Hz) was increased. *A3*, Fast IPSP from stimulation in the association fiber layer was only slightly affected. *B*, $GABA_A$ blockade at somatic level. *B1*, Single-pulse response to afferent stimulation was unchanged. *B2*, Response to burst stimulation of afferent fibers also was unchanged. *B3*, Fast IPSP from association fiber layer was largely blocked. Calibration bars in *B2* also apply to *B1* and *A1,2*; bars in *B3* also apply to *A3*.

association fiber layers indicates the presence of spatially segregated inhibitory circuitry (see Fig. 1). Focal application of bicuculline in the distal dendritic layer that had only a small, delayed effect on feedback IPSPs evoked from the association fiber layer (Fig. 5*A3*; Fig. 6*B, open circles*; Fig. 7*B, left bar*) could block the response from single pulse afferent layer stimulation by up to 30% (Fig. 5*A1*)—an underestimate of the degree of blockade of the IPSP as this response includes an NMDA component in addition to the depolarizing IPSP. Focal applications of bicuculline in the somatic region that had little effect on IPSPs evoked from the afferent layer (Fig. 5*B1*) largely blocked IPSPs evoked from the association fiber layer in all experiments ($n = 7$) (Figs. 5*B3*, 7*B*).

DISCUSSION

The results have confirmed that an NMDA component is present in afferent fiber-mediated EPSPs and that the amplitude of this component, when evoked by the same burst stimuli used to evoke LTP, is facilitated by blockade of the fast, but not the slow, GABAergic IPSP. Blockade of the fast IPSP at the distal apical dendritic level was found to be much more effective in facilitating the NMDA component of the response to afferent stimulation than blockade in the somatic region.

A central issue regarding the validity of the results is the extent to which the injections of bicuculline were spatially restricted to dendritic or near-somatic regions. Verification of spatial restriction took advantage of the segregation of the afferent input to distal dendritic segments and the feedback IPSP evoked from layer Ib to the vicinity of cell bodies. Restriction of dendritic injections was confirmed by the demonstration of minimal effect on the feedback IPSP when the afferent-evoked NMDA component was facilitated; restriction of the somatic injections was confirmed by their failure to result in significant facilitation of the NMDA component when the feedback IPSP was largely blocked.

In piriform cortex, as in other cortical areas, block of the fast IPSP can induce polysynaptic epileptiform activity. To confirm that this did not occur under the present recording conditions with the fast EPSP blocked by DNQX, control experiments were performed with an elevated divalent cation concentration (Fig. 4) that blocks polysynaptic activity in piriform cortex (Kanter and Haberly, 1993). The occurrence of facilitation under these conditions ruled out a contribution from reverberative activity in intrinsic excitatory fiber systems.

Because the postsynaptic potential evoked from afferent layer stimulation in the presence of AMPA and $GABA_B$ blockers consists of a fast IPSP in addition to the NMDA-mediated EPSP, it could also be argued that the observed facilitation resulted from an alteration in the IPSP rather than the NMDA component. As in the hippocampus (Deisz and Prince, 1989; Davies et al., 1990), the fast IPSP in piriform cortex is partially blocked via presynaptic $GABA_B$ receptors during repetitive stimulation (A. Kapur and L. B. Haberly, unpublished observations). Therefore, it is conceivable that a decrease in amplitude of this IPSP over the course of the burst stimulus could mask an increase in the NMDA component. Block of the IPSP by bicuculline could uncover the increase in the NMDA component rather than inducing its occurrence. Two arguments indicate that any contribution from this mechanism is minimal. First, the IPSP evoked by afferent layer stimulation is too small to account for the large facilitation of the burst response after disinhibition (even if the entire postsynaptic potential evoked from the afferent fiber layer were an IPSP that decremented completely over the course of the stimulus burst, it would be insufficient to account for the observed increase in the burst response). Second, bath application of 2-hydroxysaclofen or

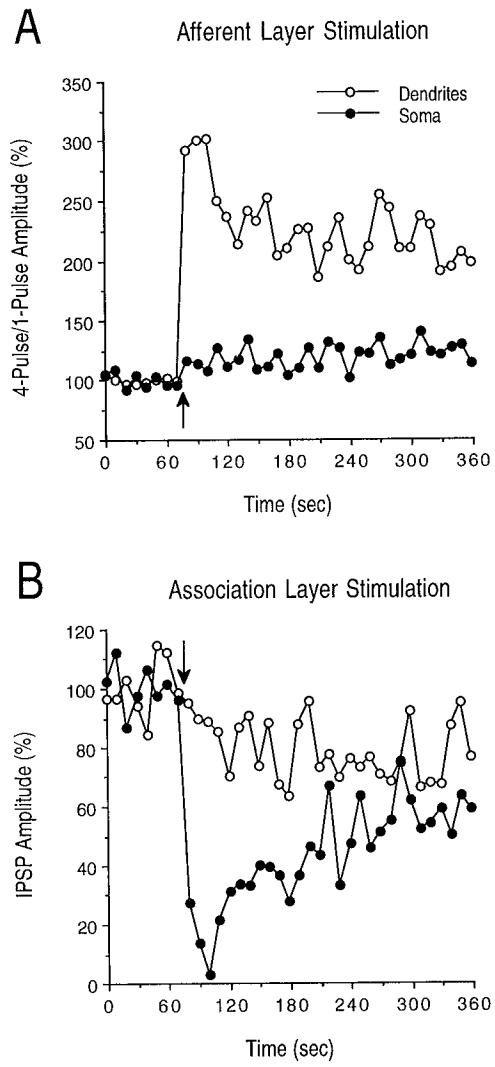

Figure 6. Time course of the effect of local bicuculline injection at dendritic and somatic levels. Same cell as in Figure 5. Baseline values before bicuculline pulse are normalized to 100%. *Arrows* indicate delivery of bicuculline pulse. *A*, Afferent layer (Ia) stimulation: ratio of peak amplitude of burst response to single-pulse response versus time at resting membrane potential. *Open circles* track the effect of bicuculline injection into layer Ia; *filled circles*, the effect of injection into the cell body layer. *B*, Amplitude of feedback IPSP evoked from layer Ib as a function of time after bicuculline injection into distal dendritic or somatic regions. Cell was depolarized to −50 mV by current injection. Symbols are as in *A*. Note the fast onset of IPSP block from somatic application and slow onset of limited block from dendritic application.

CGP 35348, which decreases the frequency reduction of the IPSP in piriform cortex as in hippocampus (Davies et al., 1990), did not alter the bicuculline-induced facilitation (Kapur and Haberly, unpublished observations).

Blockade of the facilitation of the burst-evoked EPSP by APV indicates that this process is dependent on an underlying NMDA component. It can be postulated that positive feedback between depolarization and increased channel current is responsible for the facilitation (Mayer and Westbrook, 1987): successive EPSPs would increase in amplitude because depolarization from preceding EPSPs would decrease the Mg^{2+} block. Studies with computer simulation modeling (Kapur et al., 1994) have confirmed the feasibility of this mechanism for bicuculline-induced facilitation in piriform cortex.

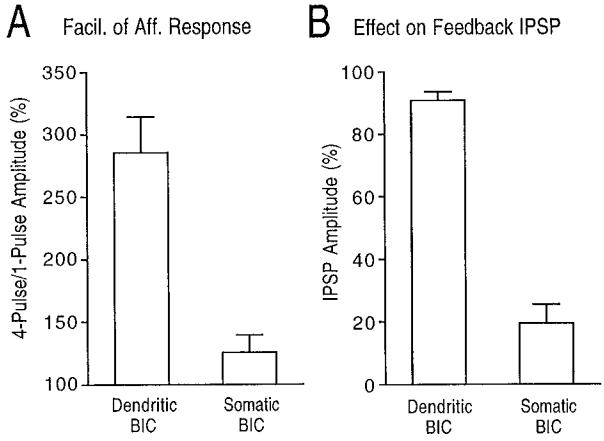

Figure 7. Differential effects of dendritic and somatic application of bicuculline: pooled results for six experiments. *A*, Facilitation of the response to burst (4-pulse) stimulation in the afferent fiber layer after local injection of bicuculline (*BIC*) at the level of distal apical dendrites (*left bar*) and in the vicinity of cell bodies (*right bar*). *Bars* show percent change in the ratio of the peak amplitude of the burst response to the peak amplitude of the single-pulse response as in Figure 6. Values were obtained at the times of maximum facilitation. *B*, Mean amplitude of intracellularly recorded feedback IPSPs, expressed as percent of control, measured at the same time that values for facilitation in *A* were obtained. Feedback IPSPs were evoked by stimulation in layer Ib. Error bars are SEMs; differences between the effects of dendritic and somatic application of bicuculline in *A* and *B* are significant at $p < 0.001$.

The amplitude of the NMDA-mediated EPSP is also presumably modulated by other factors, including postsynaptic voltage-dependent channels and presynaptic facilitation of transmitter release. It should be noted, however, that these other factors would not be directly altered by bicuculline, and although the contribution of voltage-dependent channels could increase after bicuculline-induced facilitation, only an increase in the NMDA-mediated EPSP could underlie the facilitation itself.

Implications concerning inhibitory processes

Previous experiments in piriform cortex using Cd^{2+} injection to locally block synaptic transmission had suggested that the fast, Cl^--mediated IPSP is concentrated in the vicinity of the soma, and that the slow, K^+-mediated IPSP is concentrated in the dendrites (Tseng and Haberly, 1988) as originally reported for the hippocampus (Newberry and Nicoll, 1984). The failure of these earlier studies to reveal the presence of a fast dendritic IPSP can be attributed to its small amplitude at somatic recording sites compared with the somatic-region fast IPSP. Facilitation of the NMDA component served as a powerful assay for the presence of this IPSP in the present experiments. The existence of fast dendritic inhibition in piriform cortex has been confirmed in studies of pharmacologically isolated IPSCs with whole-cell patch pipettes (Kapur et al., 1993). Previous studies in the hippocampus (Lambert et al., 1991; Solís and Nicoll, 1992; Pearce, 1993) have demonstrated fast dendritic inhibition in pyramidal cells.

GABAergic modulation of the association fiber-mediated NMDA response

EPSPs evoked by stimulation of association fibers have an NMDA component as observed with afferent fiber stimulation (Hoffman and Haberly, 1989). This component also is facilitated by application of bicuculline through the bath or by local application in the proximal dendritic region (E. D. Kanter, A. Kapur, and L. B.

Haberly, unpublished observations). However, it was not possible to determine whether facilitation of the association fiber-evoked NMDA component was a consequence of blockade of $GABA_A$ receptors in the dendritic region or a result of diffusion to the adjacent cell body region because the feedback IPSP could not be independently evoked to assess the extent of spread as in the studies of the afferent fiber response.

Relationship to LTP

Studies of the hippocampal formation (Tomasulo et al., 1993; Zhang and Levy, 1993) and piriform cortex (Kanter and Haberly, 1993) have demonstrated that associative LTP between fiber systems terminating on nonoverlapping dendritic segments can be induced only after blockade of $GABA_A$-mediated inhibition. It has been postulated that it is $GABA_A$ input at the level of dendrites that is responsible for this regulation (Tomasulo et al., 1993; Zhang and Levy, 1993). The present results, obtained with the same 4-pulse bursts used to induce associative LTP in piriform cortex, confirm a $GABA_A$-mediated input to dendrites that strongly regulates the NMDA component on which this LTP depends. Such an increase in the NMDA component could facilitate associative LTP by increasing the level of depolarization that results from a given excitatory input, allowing it to spread over a greater distance. Disinhibition also would increase the NMDA component evoked by a weak input at a given level of depolarization resulting from electrotonic spread from distant stronger inputs.

Of equal importance for understanding the regulation of LTP is the finding that somatic inhibition can be blocked without significantly altering the NMDA component. This finding means that the regulation of LTP by GABAergic tone could be largely independent of the regulation of cell excitability at the somatic level. If a set of interneurons is present that can selectively mediate the dendritic IPSP, then control of the activity of this population could regulate associative LTP by modulation of the NMDA component. Regulation of the postulated interneuron population could be effected by centrifugal projections from cholinergic and monoaminergic cell groups that have been demonstrated in the piriform cortex (Kanter and Haberly, 1993; Haberly et al., 1994).

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