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Presynaptic depression of inhibitory postsynaptic potentials by metabotropic glutamate receptors in rat hippocampal CA1 pyramidal cells

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Abstract

The effects of the metabotropic glutamate (mGlu) receptor agonists (±)-*trans*-1-aminocyclopentane-1,3-dicarboxylic acid (*trans*-ACPD) or 1*S*,3*R*-ACPD on γ-aminobutyric acid (GABA)-mediated inhibitory synaptic responses have been investigated in vitro in CA1 pyramidal cells of rat hippocampal slices. Bath application of both agonists depolarized the resting membrane potential and increased membrane resistance. Simultaneously, the afterhyperpolarization induced by a burst of spikes as well as spike accomodation were blocked. Stimulation of the stratum radiatum induced in CA1 pyramidal cells an early excitatory postsynaptic potential (EPSP) followed by a fast $GABA_A$ and a slow $GABA_B$-mediated inhibitory postsynaptic potentials (IPSPs). All synaptic responses were dose dependently depressed by mGlu receptor agonists. At low concentration, (±)-*trans*-ACPD (10–100 μM) and 1*S*,3*R*-ACPD (10 μM) consistently reduced the EPSP, slightly depressed the fast IPSP but greatly decreased the slow IPSP. Increasing the concentration of mGlu receptor agonists to 200 μM and 50 μM, respectively further depressed the EPSP and dramatically reduced the amplitude of both IPSPs. In the presence of the glutamate receptor antagonists 6-cyano-7-nitroquinoxaline-2,3-dione (10 μM) and D-(−)-2-amino-5-phosphonovaleric acid (30 μM), monosynaptically evoked IPSPs were still depressed by mGlu receptor agonists. In the same conditions, the discharge frequency of spontaneous IPSPs which reflect the activity of GABAergic interneurons was enhanced by low doses of mGlu receptor agonists but depressed with higher concentrations. On the other hand, the postsynaptic hyperpolarization and decrease in membrane resistance induced by the $GABA_B$ receptor agonist baclofen applied in the bath or by microiontophoresis were not affected by mGlu receptor agonists. These results indicate that the GABA-mediated IPSPs of CA1 hippocampal pyramidal cells are depressed by mGlu receptors located on presynaptic GABAergic terminals. Such heteroreceptors by inhibiting the release of GABA may account for the facilitatory effect of mGlu receptors in the mechanisms of neuronal plasticity.

Keywords: *trans*-ACPD ((±)-*trans*-1-aminocyclopentane-1,3-dicarboxylic acid); Metabotropic glutamate receptor; Inhibitory postsynaptic potential; Hippocampus; CA1 pyramidal cell

1. Introduction

Besides its role in mediating fast excitatory synaptic transmission through ionotropic receptors, glutamate has also a powerful neuromodulatory influence on neuronal activity through the activation of metabotropic receptors (mGlu receptors) (see Baskys, 1992; Schoepp and Conn, 1993 for review). The selective mGlu receptor agonist (±)-*trans*-1-aminocyclopenthyl-1,3-dicarboxylic acid ((±)-*trans*-ACPD) and its active isomer 1*S*,3*R*-ACPD were shown to alter neuronal excitability by inhibiting intrinsic K^+ conductances (Stratton et al., 1989; Baskys et al., 1990; Charpak et al., 1990). Besides, the depression of the excitatory synaptic transmission by (±)-*trans*-ACPD has been intensively investigated in several structures of the central nervous system and shown to involve a presynaptic autoreceptor inhibiting the release of glutamate (Forsythe and Clements, 1990; Baskys and Malenka, 1991; Crepel et al., 1991; Calabresi et al., 1992; Lovinger et al., 1993; Burke and Hablitz, 1994). In contrast, the control of GABAergic synaptic transmission by mGlu receptors is less documented. However, this is of particular importance since the activation of mGlu receptors was also shown to induce a slow-onset potentiation of extracel-

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lular field excitatory postsynaptic potential (EPSP) in the hippocampus (Bortolotto and Collingridge, 1992, 1993). In fact, one possibility is that the potentiation may reflect a reduction of the inhibitory tone on pyramidal cells. Depression by mGlu receptors of inhibitory postsynaptic potentials (IPSPs) induced in CA1 pyramidal cells by stimulation of the Schaffer collaterals was previously reported (Desai and Conn, 1991; Pacelli and Kelso, 1991; Liu et al., 1993). However, the magnitude, the time-course and the possible mechanisms of this depression remain to be determined. It may reflect either a direct effect of mGlu receptors on γ-aminobutyric acid (GABA)-mediated processes or alternatively an indirect effect by decreasing the excitatory drive of the Schaffer collaterals on the inhibitory interneurons.

In the present study, we studied the depression of GABA-mediated IPSPs by mGlu receptors in rat hippocampal slices and tried to determine the possible mechanisms of this decrease. The effects of the mGlu receptors on $GABA_A$-and $GABA_B$-mediated IPSPs, respectively, were carefully studied because these responses were recently suggested to be activated by distinct subtypes of GABAergic interneurons in the hippocampus (Lacaille and Schwartzkroin, 1988; Segal, 1990; Williams and Lacaille, 1992; Samulack and Lacaille, 1993, Samulack et al., 1993).

2. Materials and methods

Male Sprague-Dawley rats (200–350 g) were anesthetized with halothane and decapitated. The hippocampus was quickly removed and placed in a cold medium. Slices (400 μm thick) were cut on a tissue chopper and placed in a holding chamber for at least 1 h. A single slice was then transfered to the recording chamber. In the chamber, the slice was held between two nylon nets, submerged beneath a continuously superfusing medium that had been warmed to 30–33° C and pre-gassed with 95% O_2 and 5% CO_2. The composition of the medium was (mM): NaCl, 119; KCl, 3; $MgSO_4$, 1.3; $CaCl_2$, 2.5; $NaHCO_3$, 26.2; NaH_2PO_4, 1.0; glucose, 11.

Conventional intracellular recordings from CA1 pyramidal neurons were obtained using glass micropipettes filled with 3 M potassium acetate and having resistances of 60–100 MΩ. Intracellular voltage and current recordings were stored on a rectilinear strip chart recorder. Fast events (EPSP, IPSP or spikes) were stored on a digital oscilloscope and plotted on a digital plotter.

The synaptic responses elicited by electrical stimulation of the Schaffer collaterals/commissural fibers which reflect the release of endogenous neurotransmitters were compared before and after bath application of (±)-*trans*-ACPD (10–200 μM) or 1*S*,3*R*-ACPD (10–50 μM) purchased from Research Biomedical International (RBI). Stimuli (0.1 ms duration) were applied between the two poles of a bipolar electrode, one pole inserted into the slice and the other just above the slice.

In order to isolate monosynaptic IPSPs, the early EPSP was eliminated by bath application of D-(−)-2-amino-5-phosphonovaleric acid, 2-APV (30 μM, Cambridge Research Biomedical, CRB) and 6-cyano-7-nitroquinoxaline-2,3-dione, CNQX (10 μM, CRB). In addition, the fast IPSPs were isolated in some cases by bath application of the $GABA_B$ receptor antagonist 3-amino-propyl-diethoxymethylphosphinic acid (CGP 35348, 500 μM) kindly provided by Ciba-Geigy.

The IPSP amplitudes were determined at their maximal magnitude, e.g. 30 and 250 ms after stimulus onset for the fast IPSP and the slow IPSP, respectively. In fact, at 30 ms after the stimulus onset, the amplitude of the slow IPSP is extremely small due to the delayed protein G-dependent mechanism of this synaptic response. On the other hand, it was reported that the amplitude of the fast IPSP isolated in the presence of the $GABA_B$ receptor antagonist CGP 35348 was very small (and even absent) when measured 250 ms after the stimulus onset (Solis and Nicoll, 1992). These data indicate that even if some overlap occurred between the fast and the slow IPSP, this should not introduce a significant bias in our measurements.

The effects of mGlu receptor agonists on the postsynaptic responses to the $GABA_B$ receptor agonist baclofen was also investigated. In these experiments, baclofen (Ciba-Geigy) was applied either in the superfusion medium (20–40 μM for 1 min) or by microionophoresis (10 mM for 15 s). In addition to the membrane hyperpolarization, the extra conductance induced by the $GABA_B$ receptor agonist was determined. Using a constant hyperpolarizing current pulse of 0.2 nA intensity, a decrease in membrane resistance (increase in membrane conductance) occurred in the presence of baclofen which might be seen as a reduction of the magnitude of the pulse deflection. The total conductance (g_T) then measured represented the sum of the resting membrane conductance (g_R) and the specific conductance activated by the agonist (g_{Baclo}). In order to determine the latter conductance, g_R was then measured at the same level of membrane potential as achieved during baclofen application. It was then possible to calculate the extra conductance induced by baclofen as $g_{Baclo} = g_T - g_R$.

In some experiments, spontaneous IPSPs which reflect the synaptic activity of inhibitory interneurons (Alger and Nicoll, 1980; Pitler and Alger, 1992) were recorded. This was performed using 3 M KCl filled electrodes with a resistance of 30–50 MΩ. Under these conditions, diffusion of chloride ions from the elec-

Table 1
Percentage depression of the excitatory postsynaptic potential (EPSP) induced with 30 V stimulus intensity, of the fast and the slow inhibitory postsynaptic potential amplitudes as a function of metabotropic glutamate receptor agonist concentrations

	(±)-*trans*-ACPD		1*S*,3*R*-ACPD	
	50–100 μM	200 μM	10 μM	50 μM
EPSP	48 ± 4 (n = 12)	61 ± 8 (n = 6)	42 ± 8 (n = 4)	54 ± 16 (n = 6)
Fast IPSP	15 ± 3 (n = 17)	74 ± 8 (n = 8)	12 ± 2 (n = 3)	75 ± 14 (n = 6)
Slow IPSP	41 ± 4 (n = 17)	82 ± 7 (n = 8)	26 ± 4 (n = 3)	82 ± 11 (n = 6)

Results are expressed as means ± S.E.M. Numbers in parentheses indicate a number of tested cells.

trode leads to the inversion of the IPSPs. Bicuculline (10 μM, Sigma) was used in these experiments to demonstrate the GABAergic nature of these events. The spontaneous IPSPs were stored on line on a digital audiotape. The spontaneous IPSP frequency was then quantified off line by converting the discharge rate during 1 or 2 s to an analog voltage which was plotted on a chart recorder.

3. Results

The resting membrane potential of the neurons recorded in control conditions averaged 68.2 ± 6 mV (mean ± S.D., n = 48). In all cases, bath application of (±)-*trans*-ACPD (10–100 μM) or 1*S*,3*R*-ACPD (10–50 μM) for 10–30 min induced a membrane depolarization, an increase in input resistance, an inhibition of the slow afterhyperpolarization induced by a burst of spikes and a blockade of spike-frequency adaptation. All these modifications in membrane properties were used as an index of the correct wash-in of the mGlu receptor agonists.

3.1. Effects of mGlu receptor agonists on synaptic responses of CA1 pyramidal cells

In control medium, stimulation of the stratum radiatum elicited in CA1 pyramidal cells an early EPSP followed by a fast $GABA_A$-and a slow $GABA_B$-mediated IPSPs. When added to the bath for 10–30 min, mGlu receptor agonists dose dependently affected all these synaptic responses. Indeed, at low concentration, (±)-*trans*-ACPD (10–100 μM) or 1*S*,3*R*-ACPD (10 μM) consistently reduced the EPSP amplitude measured at −85 mV (Table 1). Simultaneously, the fast IPSP amplitude measured at the threshold for spiking was slightly decreased while the slow IPSP was much

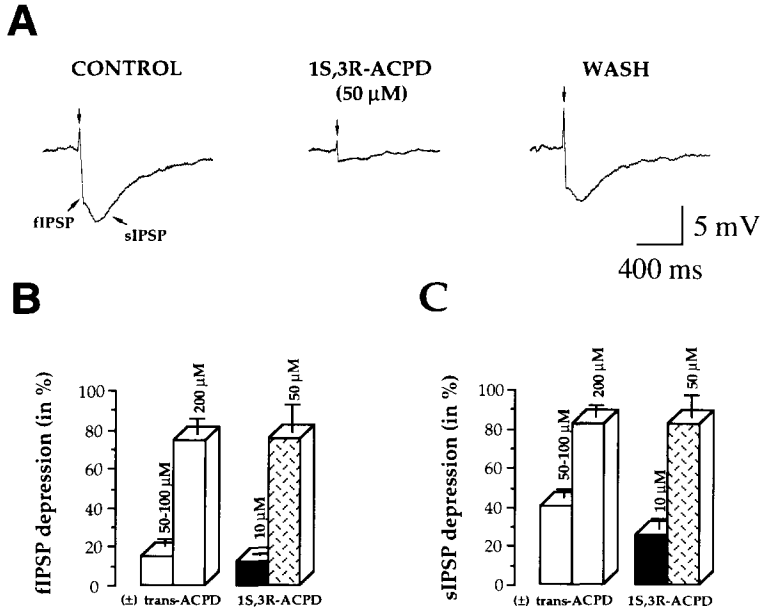

Fig. 1. Effects of mGlu receptor agonists on GABA-mediated IPSPs. (A) Examples of fast and slow IPSPs (fIPSP and sIPSP) induced by the stimulation of the stratum radiatum (arrows) in a CA1 pyramidal cell before, during and after wash-out of 1*S*,3*R*-ACPD (50 μM). Notice the strong depression of the amplitude of both IPSPs during the application of the mGlu receptor agonist and the recovery after wash-out. (B) Mean value of the fast IPSP (left) and the slow IPSP (right) depression induced by (±)-*trans*-ACPD and 1*S*,3*R*-ACPD at different concentrations.

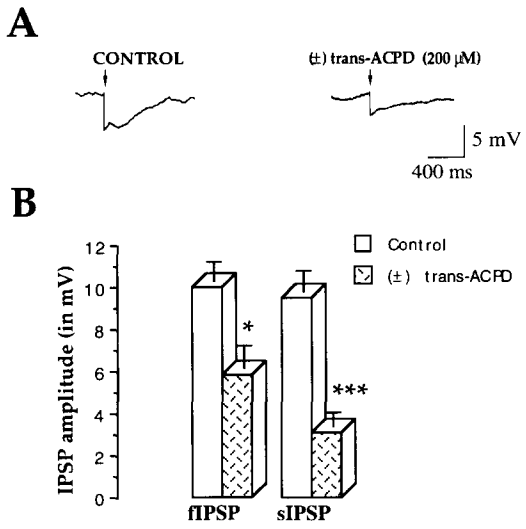

Fig. 2. Effects of mGlu receptor agonists on monosynaptically evoked GABA-mediated IPSPs. (A) Examples of monosynaptic IPSPs induced in a CA1 pyramidal cells by the stimulation of the stratum radiatum (arrows) in the presence of the excitatory amino acid antagonists APV (30 μM) and CNQX (10 μM). (A, left) Control medium. (A, right) In the presence of (±)-*trans*-ACPD (200 μM). (B) Mean value of the amplitude of the monosynaptically evoked fast IPSP and slow IPSP before (open columns) and during application of (±)-*trans*-ACPD (200 μM) (dashed columns). Notice that the amplitude of both monosynaptic IPSPs is significantly decreased by the mGlu receptor agonist (** $P < 0.01$ and *** $P < 0.001$ respectively, ANOVA).

more depressed (Table 1 and Fig. 1). Increasing the concentration of (±)-*trans*-ACPD to 200 μM and 1*S*,3*R*-ACPD to 50 μM further depressed the EPSP (see Table 1) and dramatically reduced the amplitude of both IPSPs (Fig. 1).

After wash-out of mGlu receptor agonists, both EPSP and IPSPs recovered (Fig. 1A.). However in a few neurons, EPSP amplitude was larger than in control conditions while IPSPs remained depressed.

3.2. Possible mechanisms of IPSP depression by mGlu receptor agonists

The results described above indicate that the inhibitory GABAergic responses of CA1 pyramidal cells are greatly depressed by mGlu receptors. However, since the EPSPs were also reduced by the agonists, the possibility remains that the depression of IPSP amplitude may reflect a decrease of the excitatory drive of the Schaffer collaterals converging on the inhibitory interneurons (see Desai and Conn, 1991). In order to answer this question, we studied the effects of mGlu receptor activation on monosynaptically elicited IPSPs by direct stimulation of the inhibitory interneurons in the presence of the glutamate receptor antagonists CNQX (10 μM) and 2-APV (30 μM).

Under these conditions, bath application of (±)-

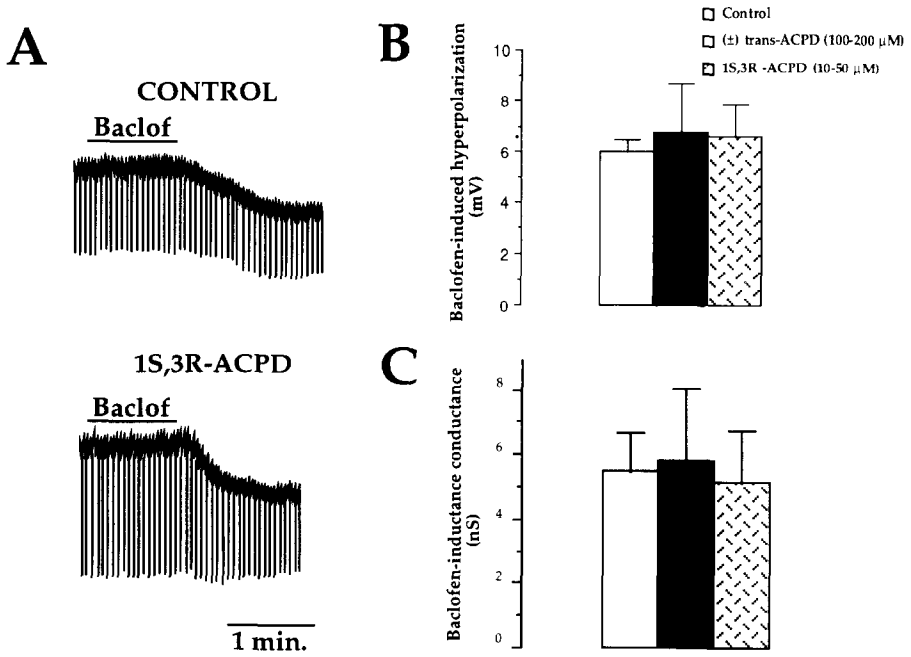

Fig. 3. Effects of mGlu receptor agonists on postsynaptic GABA receptor activation. (A) Examples of hyperpolarization and increase in membrane conductance induced in two different CA1 pyramidal cells by bath application of the $GABA_B$ receptor agonist baclofen (Baclof 30 μM) in control medium (top) and in the presence of 1*S*,3*R*-ACPD (50 μM) (bottom). (B) Mean value of the hyperpolarization and (C) mean value of the extra conductance induced by bath application of baclofen in control medium (open columns), in the presence of (±)-*trans*-ACPD (100–200 μM) (filled columns) and in the presence of 1*S*,3*R*-ACPD (10–50 μM) (dashed columns). Notice the absence of significant differences in the three conditions for both parameters.

trans-ACPD (200 μM) still significantly depressed the IPSPs (Fig. 2A). The fast IPSP was decreased by 41.1 ± 10.7% ($n = 8$) and the slow IPSP by 67.4 ± 6.4% ($n = 6$). A similar amplitude of depression by mGlu receptor agonists was obtained for isolated fast IPSPs after bath application of the $GABA_B$ receptor antagonist CGP 35348 to block the slow IPSP (500 μM, $n = 4$). These results indicate a significantly higher sensitivity of the $GABA_B$-mediated IPSPs to $mGlu_R$ agonists (ANOVA, $P < 0.01$ and $P < 0.001$ for the fast and the slow IPSP, respectively, Fig. 2B) as also observed in control medium (see Table 1). On the other hand, it must be pointed out that the depression obtained for both monosynaptically elicited IPSPs in the presence of CNQX and 2-APV is significantly weaker than the decrease recorded in control conditions.

These results indicate that mGlu receptors directly depress GABA-mediated IPSPs in the hippocampus. We then tried to determine the possible mechanism of this decrease. A first hypothesis was a dysfunction of the postsynaptic GABA receptors. Since the $GABA_B$-mediated IPSP appeared the most strikingly affected by mGlu receptor agonists, we focussed on the responses of CA1 pyramidal cells to the $GABA_B$ receptor agonist baclofen.

When applied in the bath at resting membrane potential, baclofen (20–40 μM for 1 min) induced a membrane hyperpolarization and a decrease in membrane resistance (i.e. an increase in membrane conductance) which reflect the activation of postsynaptic $GABA_B$ receptors (Fig. 3A, top). In control medium, a mean hyperpolarization of 6 ± 0.5 mV ($n = 6$) was recorded (Fig. 3B) while the extra conductance opened by the $GABA_B$ receptor agonist averaged 5.5 ± 1.2 nS (Fig. 3C). In the presence of (±)-*trans*-ACPD (100–200 μM, $n = 7$) or 1*S*,3*R*-ACPD (10–50 μM, $n = 5$), the activation of the postsynaptic $GABA_B$ receptors persisted (Fig. 4A, bottom): the mean hyperpolarization reached 6.7 ± 2 mV and 6.6 ± 1.3 mV, respectively (Fig. 3B), while the mean baclofen-induced conductance was 5.7 ± 2.2 nS and 5.2 ± 1.6 nS (Fig. 3C). These results indicate that the effects of the $GABA_B$ receptor agonist on CA1 pyramidal cells were not significantly affected in the presence of mGlu receptor agonists. This was also the case when baclofen was locally applied on the pyramidal cell dendrites by microionophoresis. In these conditions, (±)-*trans*-ACPD (100–200 μM, $n = 2$) did not affect the hyperpolarization and the increase in membrane conductance induced by brief application of the $GABA_B$ receptor

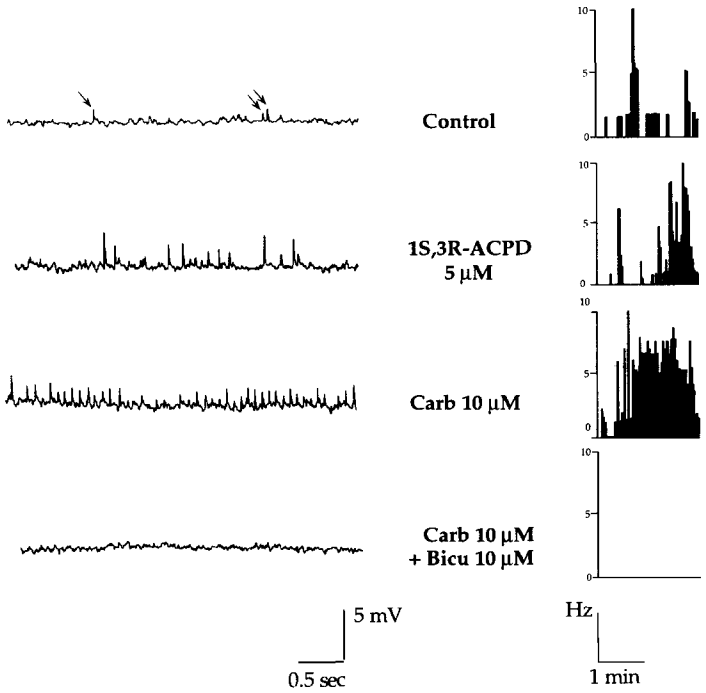

Fig. 4. Effects of low dose of mGlu receptor agonists on synaptic release of GABA. Examples (left) and frequency versus time plot (right) of spontaneous IPSPs (arrows) recorded from a pyramidal cell in control medium or during bath application of 1*S*,3*R*-ACPD (5 μM, 15 min), the cholinergic agonist carbachol (Carb, 10 μM, 2 min) and carbachol plus the $GABA_A$ antagonist bicuculline (Bicu, 10 μM, 2 min). All records are from the same neuron and each drug application was successively performed after 15 min wash-out. Each frequency bin is the rate (in Hz) averaged over a 2 s period. The spontaneous IPSPs reflect the firing of the GABAergic interneurons since they are abolished by the $GABA_A$ receptor antagonist (lower trace). Notice that the spontaneous IPSPs frequency which is low in basal condition is significantly enhanced by both mGlu receptor and cholinergic agonists.

agonist (10 mM for 15 s) while at the same time, the $GABA_B$-mediated IPSP was dramatically attenuated (not illustrated).

These results indicate that IPSP depression by mGlu receptors does not reflect a dysfunction of postsynaptic $GABA_B$ receptors. We then considered the possibility that mGlu receptors might presynaptically decrease the release of GABA. In order to test this hypothesis, we studied the effects of mGlu receptor agonists on spontaneous IPSPs reflecting the synaptic release of GABA caused by the activity of the inhibitory interneurons (Alger and Nicoll, 1980; Pitler and Alger, 1992; Miles and Poncer, 1993).

In the presence of the glutamate antagonists CNQX (10 μM) and 2APV (30 μM), 1*S*,3*R*-ACPD at low concentration (5–10 μM, $n = 4$) enhanced the frequency of spontaneous IPSPs by increasing the occurence of events of large amplitude which were easily blocked by the $GABA_A$ antagonist bicuculline (Fig. 4). However, increasing the concentration of 1*S*,3*R*-ACPD (40–50 μM, $n = 7$) induced a depression of the spontaneous IPSP frequency (Fig. 5). On the other hand, bath application of the cholinergic agonist carbachol (10 μM, $n = 5$) dramatically enhanced the frequency of these synaptic events (Fig. 5). When carbachol was applied during a prolonged application of 1*S*,3*R*-ACPD (40–50 μM), the increase induced by the cholinergic agonist was much less pronounced (Fig. 5). These results indicate that at a concentration which decreases evoked IPSPs, mGlu receptor activation also depressed

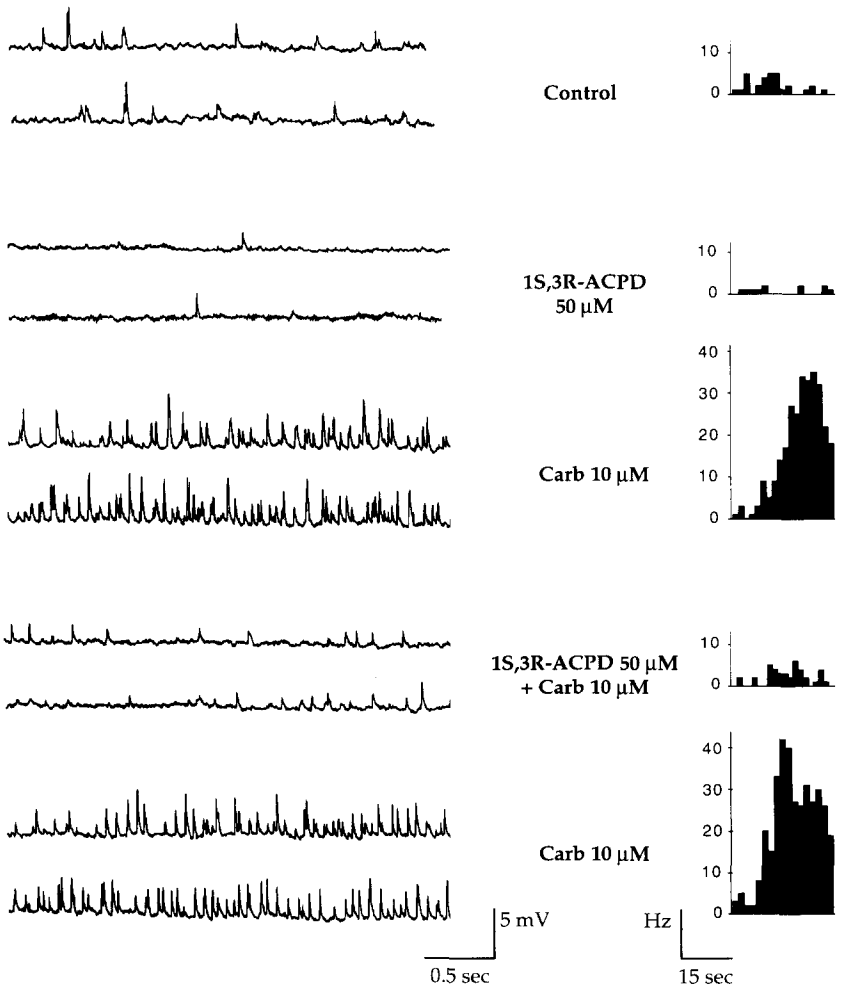

Fig. 5. Effects of high doses of mGluR agonists on synaptic release of GABA. Examples (left) and frequency versus time plot (right) of spontaneous IPSPs recorded from a pyramidal cell in control medium or during bath application of 1*S*,3*R*-ACPD (50 μM, 15 min), the cholinergic agonist carbachol (Carb, 10 μM, 1 min), carb in the presence of 1*S*,3*R*-ACPD (50 μM, 15 min), and finally in the presence of carbachol only. All records are from the same neuron and each drug application was successively performed after 15 min wash-out. Each frequency bin is the rate (in Hz) averaged over a 2 s period. Notice the clear inhibition of the spontaneous as well as the carbachol-induced increase of spontaneous IPSP frequency by the mGlu receptor agonist.

the spontaneous as well as the cholinergic-induced synaptic release of GABA from the presynaptic terminals of the inhibitory interneurons.

4. Discussion

The results of the present study show that the amplitude of the inhibitory responses of CA1 pyramidal cells mediated by the GABAergic interneurons are dose dependently decreased by mGlu receptor activation. A depression of GABA-mediated IPSPs was previously reported in the hippocampus (Desai and Conn, 1991; Pacelli and Kelso, 1991) as well as in other structures such as the nucleus tractus solitarii (Glaum and Miller, 1993), the striatum (Calabresi et al., 1992), the accessory olfactory bulb (Hayashi et al., 1993) and the neocortex (Burke and Hablitz, 1994). Our results further indicate that the $GABA_B$-mediated IPSPs were more sensitive to the effects of mGlu receptor agonists than the fast IPSPs induced by $GABA_A$ receptor activation. This might be due to the fact that a higher concentration of GABA is needed to activate $GABA_B$ receptors (Dutar and Nicoll, 1988). A second hypothesis could be a higher density of mGlu receptors on the interneurons mediating the $GABA_B$ response. Indeed, it was reported that mGlu receptors which belong to a family of G protein-coupled receptors consisting of at least eight distinct subtypes, show specific localization within the hippocampus (Tanabe et al., 1992). The $mGlu_1$ receptor α-subtype was found to be prefentially located on a population of GABAergic interneurons which also appeared to be specifically somatostatin-immunoreactive (Baude et al., 1993; Craig et al., 1993). This subset of interneurons localized in the stratum radiatum was recently proposed to selectively induce the slow $GABA_B$-mediated IPSPs in the hippocampus (Williams and Lacaille, 1992).

In an attempt to determine the possible mechanism of IPSP depression by mGlu receptors, we found that the depressive effect of the metabotropic agonists was also observed on monosynaptic IPSPs evoked by direct activation of the GABAergic interneurons (see also Liu et al., 1993). This result suggests that the mGlu receptor directly interacts with GABA-mediated processes in the hippocampus. Nevertheless, since the magnitude of the depression of the monosynaptic IPSPs was smaller than that observed in control medium, it is likely that part of the decrease in the latter condition might be due to a depression by mGlu receptors of the excitatory drive of the Schaffer collaterals impinging on the inhibitory interneurons (see Desai and Conn, 1991). Indeed, our results showed a clear depression of the excitatory transmission at the Schaffer collateral terminals by mGlu receptors, particularly at the junction with the CA1 pyramidal cells in accordance with previous reports (Forsythe and Clements, 1990; Baskys and Malenka, 1991; Pacelli and Kelso, 1991 but see Liu et al., 1993).

Considering that mGlu receptors directly affect GABAergic synaptic efficacy, at least two possible hypothesis remain: a dysfunction of the postsynaptic receptors and/or a decrease in GABA release. We showed here that despite a suppression of the $GABA_B$-mediated IPSP by mGlu receptors, the postsynaptic $GABA_B$ receptors remained functionnal. This result suggests that changes in the sensitivity of the postsynaptic sites did not account for the IPSP depression by mGlu receptors. However, we do not definitively exclude that part of the decrease may also possibly involve some postsynaptic mechanisms. In fact, Liu et al. (1993) have shown that the depression of the fast IPSP by mGlu receptors might be alleviated if postsynaptic G-proteins were previously activated by a non-hydrolyzable analogue of GTP.

Anyway, our findings suggest that IPSP depression by mGlu receptors may rather imply a presynaptic mechanism. In the last few years, evidence has accumulated which indicates that the control of glutamate release by mGlu receptors in the hippocampus might involve a presynaptic autoreceptor (Forsythe and Clements, 1990; Baskys and Malenka, 1991) but no evidence was yet available for the presence of mGlu receptors on inhibitory terminals. Spontaneous IPSPs reflect the synaptic release of GABA caused by the activity of the inhibitory interneurons (Alger and Nicoll, 1980; Pitler and Alger, 1992). We showed here that the frequency of spontaneous IPSPs was enhanced by *trans*-ACPD at low concentration. This reflects the primary excitatory effect of mGlu receptor agonists acting on the soma of the interneurons (see Miles and Poncer, 1993; McBain et al., 1994). In contrast, increasing the concentration to doses which reduced evoked IPSPs also decreased spontaneous IPSPs. This was not due to a depolarisation block of inhibitory cell firing since the cholinergic agonist carbachol could still increase the spontaneous IPSP frequency during mGlu receptor agonist application. This result rather suggests the presence of another type of mGlu receptors presynaptically located on inhibitory terminals which may reduce GABA release. Presynaptic depression of GABA-mediated responses was recently reported in the striatum (Calabresi et al., 1992), the accessory olfactory bulb (Hayashi et al., 1993) and the neocortex (Burke and Hablitz, 1994) as well as at the periphery, e.g. the lobster neuromuscular synapse (Miwa et al., 1993).

Our findings showing a delayed depression of the GABA-mediated IPSPs by mGlu receptor activation may account for the slow-onset potentiation of extracellularly field EPSP recorded in the hippocampus during application of mGlu receptor agonists (Borto-

lotto and Collingridge, 1992,1993). A similar increase of synaptic excitability by mGlu receptor agonists was recently reported in rat dorsolateral septal nucleus which was abolished in the presence of the mGlu receptor antagonist 2-amino-3-phosphonopropionic acid (Zheng and Gallagher, 1992). Besides, a wealth of evidence indicates a facilitatory effect of mGlu receptor agonists in the mechanisms of synaptic plasticity such as those involved in long term potentiation (see Schoepp and Conn, 1993 for review). Different hypotheses were proposed to account for the role of mGlu receptors in long term potentiation including an increase of NMDA receptor activation (Aniksztejn et al., 1992; Harvey and Collingridge, 1993; O'Connor et al., 1994) or the activation of some postsynaptic mGlu receptor-linked NMDA-independent calcium signal (Bortolotto and Collingridge, 1993). Our present results showing a clear depression of synaptic inhibition provide evidence for another mechanism of NMDA-independent long term potentiation induced by mGlu receptor agonists in CA1 pyramidal cells (see Desai and Conn, 1991). In fact, mGlu receptor activation was suggested to play a major role in the maintenance of long term potentiation (Behnisch and Reymann, 1993; Bortolotto and Collingridge, 1993) and a clear depression of GABAergic inhibition on CA1 pyramidal cells was recently reported during this phase of the potentiation (Stelzer et al., 1994). We may then hypothesize that the decrease of inhibitory synaptic activity in the maintenance of long term potentiation may be partly due to the activation of the metabotropic glutamate heteroreceptors present on the terminals of the inhibitory GABAergic interneurons. A definitive demonstration of the contribution of mGlu receptors in these mechanisms needs the development of new potent antagonists since results obtained with (*RS*)-α-methyl-4-carboxyphenyl-glycine are rather controversial (see Bashir et al., 1993; Chinestra et al., 1993; Manzoni et al., 1994).

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