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Presynaptic inhibitory action of the group II metabotropic glutamate receptor agonists, LY354740 and DCG-IV

John Kilbride [a], LingQian Huang [a], Michael J. Rowan [b], Roger Anwyl [a,*]

[a] *Department of Physiology, Trinity College, Dublin 2, Ireland*
[b] *Department of Pharmacology and Experimental Therapeutics, Trinity College, Dublin 2, Ireland*

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Abstract

Electrophysiological studies were carried out on the presynaptic inhibitory action of the group II metabotropic glutamate (mGlu) receptor agonists (+)-2-aminobicyclo[3.1.0]hexane-2-6-dicarboxylic acid (LY354740) and (2*S*,1′*R*,2′*R*,3′*R*)-2-(2′,3′-dicarboxycyclopropyl)glycine (DCG-IV) in three paths of the rat hippocampus, the medial and lateral perforant path to the dentate gyrus, and the Schaffer collateral/commissural path to CA1. LY354740 caused a dose-dependent reversible inhibition of the field excitatory postsynaptic potential (EPSP) in the medial and lateral perforant paths, with an EC_{50} of 115 ± 16 nM and 230 ± 58 nM, respectively. Maximal inhibition by LY354740 was much greater in the medial path (about 80%) than in the lateral path (about 50%). No inhibition was observed in CA1. A presynaptic inhibition was confirmed by LY354740 inducing dose-dependent changes in paired-pulse depression/facilitation. DCG-IV had a similar action to LY354740, but with a lower potency.

Keywords: Glutamate receptor, metabotropic; Perforant path; Paired-pulse

1. Introduction

The mediation of presynaptic inhibition by metabotropic glutamate (mGlu) receptors was first established in studies in which 1*S*,3*R*-aminocyclopentane-1,3-dicarboxylic acid (*trans*-ACPD) was shown to reversibly depress excitatory synaptic transmission in hippocampal CA1 (Baskys and Malenka, 1991; Desai and Conn, 1991; McGuinness et al., 1991; Pacelli and Kelso, 1991). A large number of studies have subsequently confirmed the presence of presynaptic mGlu receptors in many other areas of the brain, such as the hippocampal dentate gyrus (Macek et al., 1996; Bushell et al., 1996), neocortex (Sladeczek et al., 1993; Burke and Hablitz, 1994) and the striatum (Lovinger, 1991, Calabresi et al., 1992). Strong evidence that the inhibition of excitatory synaptic transmission was mediated presynaptically was shown by several lines of evidence. Firstly, inhibition by mGlu receptor agonists occurred without postsynaptic changes (Lovinger, 1991; Baskys and Malenka, 1991; Calabresi et al., 1992; Glaum et al., 1992; Lovinger et al., 1993; Burke and Hablitz, 1994); secondly, mGlu receptor activation reduced the AMPA- and NMDA-receptor mediated excitatory synaptic transmission with a similar potency (Baskys and Malenka, 1991; Lovinger, 1991; Pacelli and Kelso, 1991). Thirdly a change in paired-pulse facilitation or depression (indicative of a presynaptic modulation of transmitter release), was evoked by mGlu receptor agonists. Thus paired-pulse facilitation in CA1 was enhanced by mGlu receptor agonists (Baskys and Malenka, 1991; Burke and Hablitz, 1994; Gereau and Conn, 1995; Manzoni et al., 1997) and paired-pulse depression in the medial perforant path of the dentate gyrus being reduced by mGlu receptor agonists (Kahle and Cotman, 1993; Brown and Reymann, 1995).

Recent studies using a number of agonists selective for mGlu receptor group subtypes have shown the widespread presence of presynaptic group II mGlu receptors. In the hippocampus, a number of group II mGlu receptor selective agonists have been found to inhibit excitatory synaptic transmission, including (1*S*,3*S*)-1-aminocyclopentane-1,3-dicarboxylic acid {(1*S*,3*S*)-ACPD} (Vignes et al., 1995) and (2*S*,1′*R*,2′*R*,3′*R*)-2-(2′,3′-dicarboxycyclopropyl)-glycine (DCG-IV) (Yokoi et al., 1996) in young CA1; (2*S*,1′*S*,2′*S*)-2-carboxycyclopropyglycine (LCCG-1)

* Corresponding author. Tel.: +353-1-608-1624; Fax: +353-1-679-3545.

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(Ugolini and Bordi, 1995) and DCG-IV (Macek et al., 1996; Huang et al., 1997) in the medial perforant path of adult dentate gyrus, and DCG-IV (Macek et al., 1996; Bushell et al., 1996) and (1*S*,3*S*)-ACPD (Bushell et al., 1996) in the lateral perforant path of neonatal (Bushell et al., 1996) and adult (Macek et al., 1996) dentate gyrus.

(+)-2-Aminobicyclo[3.1.0]hexane-2-6-dicarboxylic acid (LY354740) is a recently synthesised high affinity efficacious and selective group II mGlu receptor agonist. (Bond et al., 1997; Monn et al., 1997; Schoepp et al., 1997a,b). LY354740 suppressed forskolin-stimulated cyclic 3′,5′-adenosine monophosphate (cAMP) formation at group II mGlu receptor with nanomolar potency, but had little or no agonist or antagonist action at group I mGlu receptor or group III mGlu receptor. The agent has potentially important clinical uses—it was found to prevent anxiety in the elevated plus maze and also prevent ACPD-induced limbic seizures. Moreover, it is orally active.

In the present study, we have investigated the presynaptic inhibitory action of LY354740 in the medial and lateral

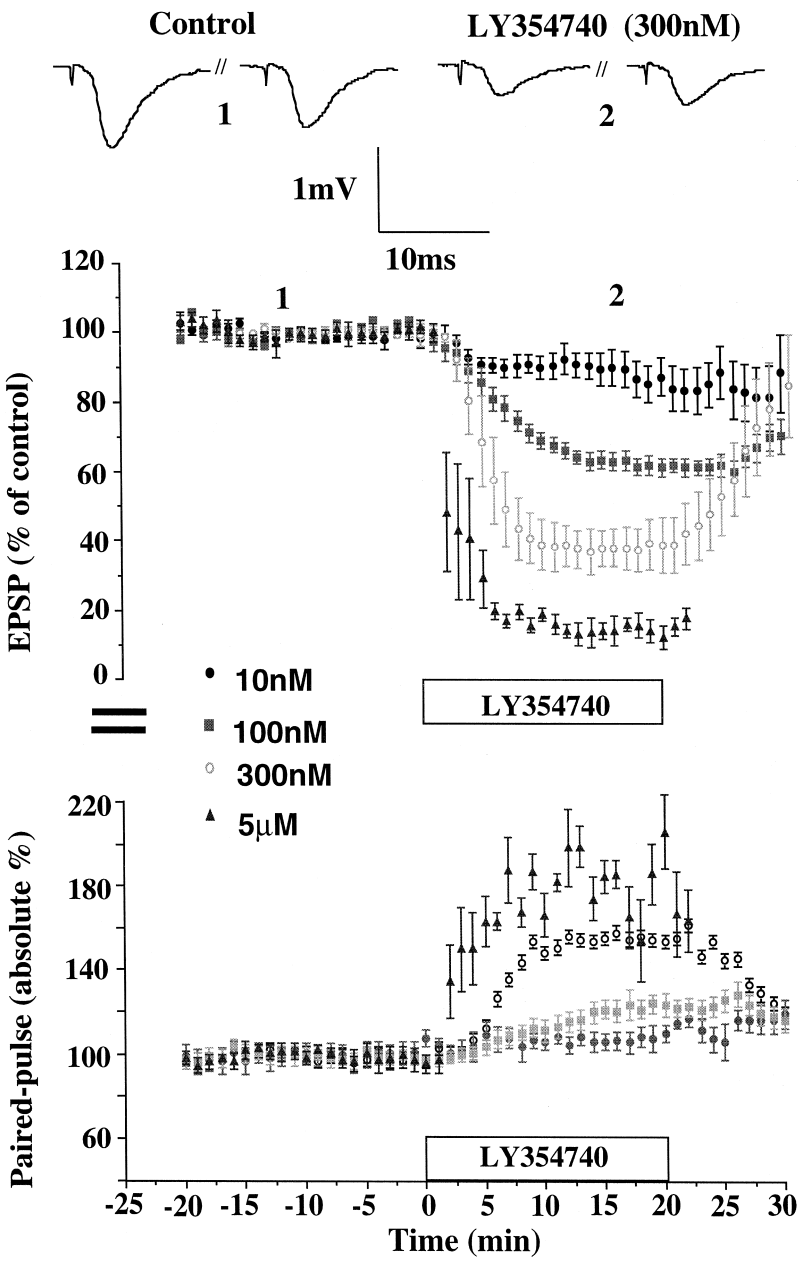


Fig. 1. LY354740 evokes a dose-dependent inhibition of excitatory synaptic transmission in the medial perforant path of the dentate gyrus in vitro. Upper graph, following a stable baseline for 20 min, perfusion of LY354740 at doses of 10 nM, 100 nM, 300 nM and 5 μM resulted in an increasing inhibition of the field EPSPs. Lower graph, 10 nM, 100 nM, 300 nM and 5 μM LY354740 caused an increasing reduction in paired-pulse depression accompanying the inhibition of the EPSP. The original traces show pairs of EPSPs in control and following application of 300 nM LY354740.

perforant path of the hippocampal dentate gyrus, and also CA1 hippocampus, comparing its action with the well established group II mGlu receptor agonist, DCG-IV.

2. Materials and methods

All experiments were carried out on hippocampal slices obtained from Wistar rats (50–70 g) (BioResources Unit, Trinity College, Dublin, Ireland). Slices were obtained as described previously (Huang et al., 1997). Briefly, the brain was rapidly removed after decapitation and placed in cold (5°C) oxygenated (95% O_2, 5% CO_2) artificial cerebro-spinal fluid (ACSF) containing in mM: NaCl, 120; $NaHCO_3$, 26; NaH_2PO_4, 1.25; KCl, 2.5; Mg_2SO_4, 2; $CaCl_2$, 2; glucose, 10). Hippocampal slices (350 μM) were cut using a Campden vibroslice (Campden Group Instruments, London, UK) and transferred immediately to an incubation chamber, maintained at room temperature, for a period of at least 60 min. Single slices were then transferred to a submersion type recording chamber at 30–31°C.

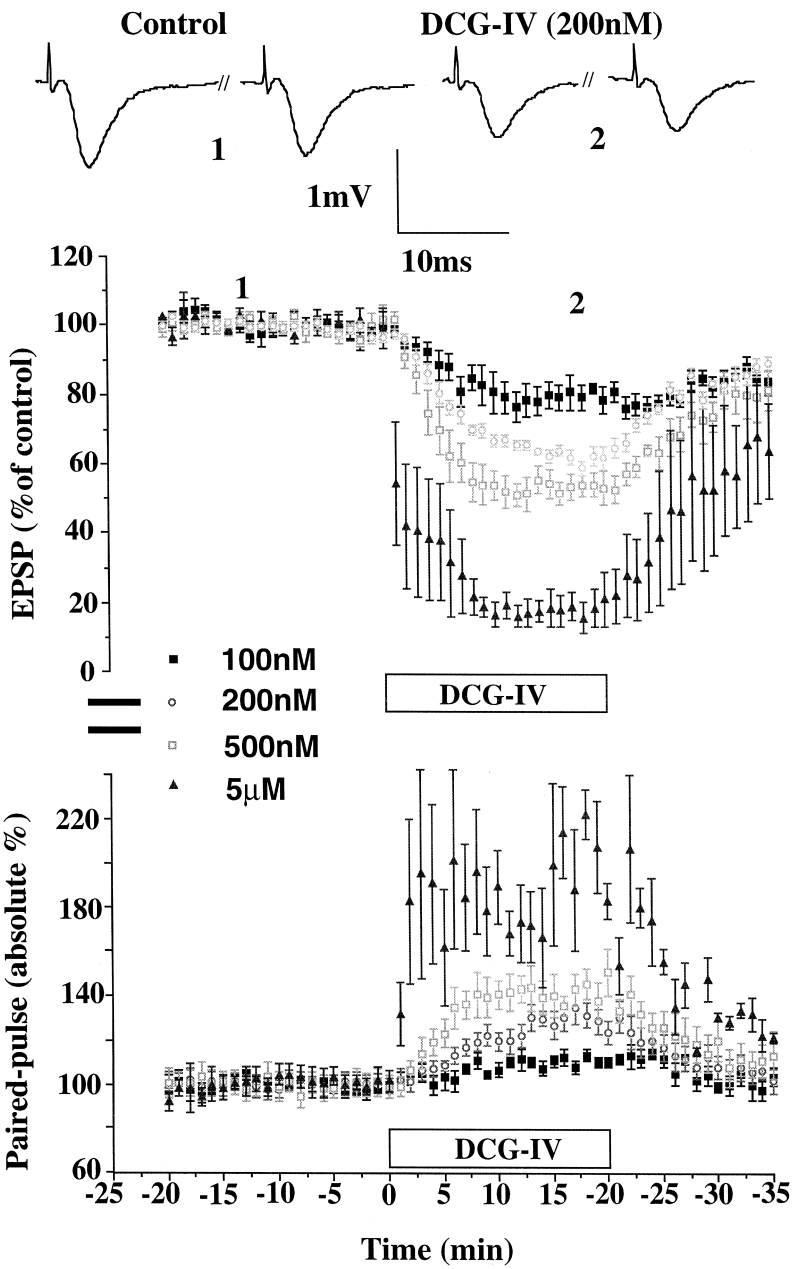


Fig. 2. DCG-IV evokes a dose-dependent inhibition of excitatory synaptic transmission in the medial perforant path of the dentate gyrus in vitro. Upper graph, following a stable baseline for 20 min, perfusion of DCG-IV at doses of 100 nM, 200 nM, 500 nM and 5 μM resulted in an increasing inhibition of the field EPSPs. Lower graph, 100 nM, 200 nM, 500 nM and 5 μM DCG-IV caused an increasing reduction in paired-pulse depression accompanying the inhibition of the EPSP. The original traces show pairs of EPSPs in control and following application of 200 nM DCG-IV.

Field excitatory postsynaptic potentials (EPSP) were recorded using standard glass electrodes filled with ACSF. Both recording and stimulating electrodes were placed in either the middle or outer third of the molecular layer of the dentate gyrus in order to stimulate and record from either the medial or lateral perforant path, respectively, and in the Schaffer collateral/commissural path in the stratum radiatum of CA1. Test EPSPs were evoked using a Grass S48 stimulator (0.0166 Hz, pulse width 0.1 ms) via a bipolar insulated tungsten wire electrode, adjusted to give about 30% of the maximal response (~1 mV). EPSP amplitude was measured using Maclab Scope, version 3.4. Paired-pulse stimulation (interstimulus interval of 40 ms) was applied in all experiments. For each pair, the amplitude of the second EPSP was divided by the first and multiplied by 100 to give the paired-pulse 'percentage'. A paired-pulse percentage of less than 100 was indicative of paired-pulse depression; a percentage greater than 100 was indicative of paired-pulse facilitation. The effect of LY354740 in the lateral perforant path and CA1 are presented in this way, in the text and in Figs. 3 and 5. However, the effects of LY354740 in the medial perforant path and also DCG-IV in the medial and lateral perforant path are represented in the text and Figs. 1, 2 and 4 as the 'absolute' percentage change from the normalised baseline control period, in order that the dose-dependent change in

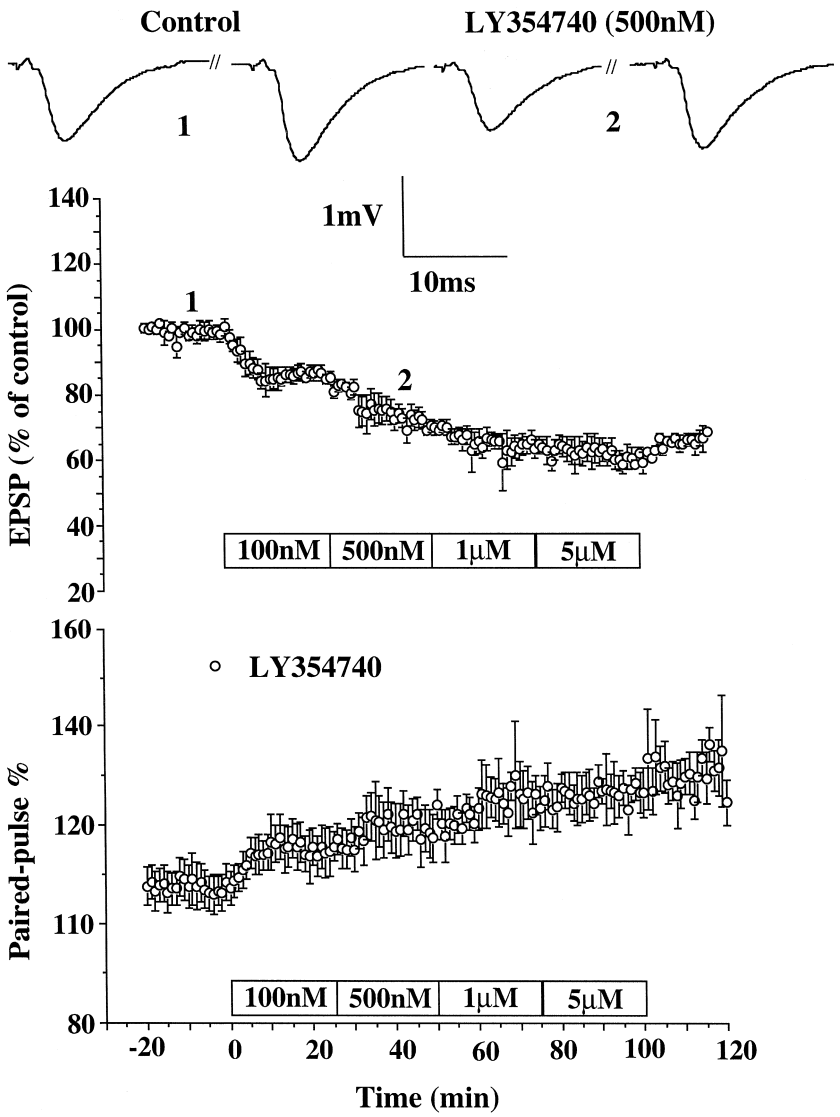


Fig. 3. LY354740 causes a dose-dependent inhibition of excitatory synaptic transmission in the lateral perforant path of the dentate gyrus in vitro. Upper graph, subsequent to a 20 min baseline period, perfusion of LY354740 at doses of 100 nM, 500 nM, 1 and 5 μM caused an increasing inhibition in the amplitude of the field EPSPs. Lower graph, paired-pulse facilitation in the lateral perforant path undergoes a concomitant increase in response to increasing doses of LY354740. The original traces show pairs of EPSPs elicited with a 40 ms inter-stimulus interval. Note the paired-pulse facilitation under control conditions, indicative of the lateral perforant pathway, and the subsequent increase in paired-pulse facilitation induced by LY354740 (500 nM).

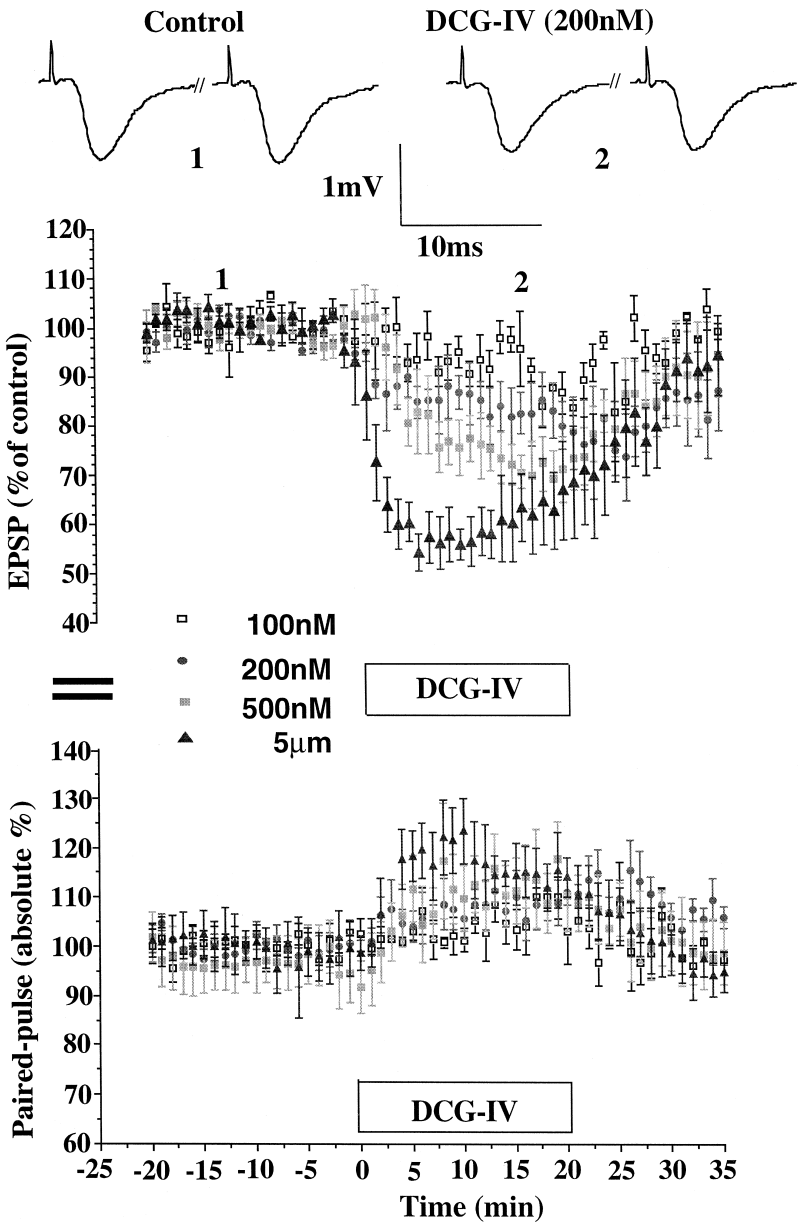


Fig. 4. DCG-IV causes a dose-dependent inhibition of excitatory synaptic transmission in the lateral perforant pathway of the dentate gyrus in vitro. Upper graph, following a stable baseline for 20 min, perfusion of DCG-IV at doses of 100, 200 and 500 nM and 5 μM caused a dose-dependent reduction in the EPSP amplitude. Lower graph, The absolute percentage changes in the level of paired-pulse facilitation induced by the application of stated doses of DCG-IV. The traces show the increase in paired-pulse facilitation in response to the application of 200 nM.

paired-pulse could be easily visualised in the figures with four different doses displayed (compare with Figs. 3 and 5).

All experiments in the dentate gyrus were carried out in the presence of 100 μM picrotoxin to block $GABA_A$ receptor-mediated inhibition. Drugs were added directly to the perfusate after establishing a steady baseline. Each slice was exposed to only one concentration of either LY354740 or DCG-IV except in some of the experiments carried out on LY354740 in the lateral perforant pathway in which cumulative doses were tested. LY354740 was a generous gift from Eli Lilly, USA. DCG-IV was obtained from Tocris Cookson.

Summarised results are expressed as normalised EPSP mean amplitude ± S.E.M. Data was analysed using Student's paired *t*-test, and repeated measures analysis of variance. Dose–response curves were constructed using Graphpad (Prism) software.

3. Results

3.1. LY354740 and DCG-IV inhibit the EPSP in the medial perforant path

The placement of the recording and stimulating electrodes in the medial perforant path was verified by the

presence of paired-pulse depression of EPSPs in response to paired-pulse stimulation.

After establishing a stable amplitude test EPSP for at least 20 min, bath application of LY354740 (20–25 min) caused a dose-dependent and reversible inhibition of the EPSP amplitude in the medial perforant path. The approximate threshold concentration of LY354740 was 10 nM, which induced a small inhibition of the EPSP amplitude of $11 \pm 4\%$ ($P < 0.05$, $n = 5$). LY354740 at higher concentrations of 100 nM and 300 nM reduced the EPSP amplitude by $36\% \pm 2\%$ ($P < 0.05$, $n = 7$) and $62 \pm 6\%$ ($P < 0.05$, $n = 4$), respectively (Fig. 1). Maximal inhibition of $85 \pm 3\%$ ($P < 0.05$, $n = 4$) was evoked with 5 μM LY354740. The dose–response curve for LY354740 (Fig. 6) was best fitted with a one-site binding hyperbola. The EC_{50} was estimated to be 115 ± 16 nM from this dose–response curve.

Evidence that the LY354740-evoked inhibition of the EPSPs was presynaptic was shown by an accompanying reduction in paired-pulse depression. In control media, EPSPs evoked in pairs, at an interval of 40 ms, resulted in paired-pulse depression of about 16%. Paired-pulse depression was reduced by LY354740 in a dose-dependent manner. Thus 10 nM, 100 nM, 300 nM and 5 μM caused absolute percentage changes in paired-pulse depression of $8 \pm 4\%$ ($P < 0.05$, $n = 5$), $21 \pm 12\%$ ($P < 0.05$, $n = 4$), $56 \pm 9\%$ ($P < 0.05$, $n = 4$) and $78 \pm 11.7\%$ ($P < 0.05$, $n = 5$), respectively (Fig. 1).

Perfusion of DCG-IV also resulted in dose-dependent and reversible inhibition of the EPSP amplitude, although less potently than LY354740. In the presence of 100 nM, 200 nM, 500 nM and 5μM DCG-IV, the EPSP amplitude was reduced by $21 \pm 4\%$ ($P < 0.05$, $n = 6$), $36 \pm 2\%$ ($P < 0.05$, $n = 8$), $47 \pm 3\%$ ($P < 0.05$, $n = 7$) and $82 \pm 4\%$ ($P < 0.05$, $n = 4$) of control values, respectively (Fig. 2, also see Fig. 4). The EC_{50} value was estimated to be 317 ± 54 nM from the dose–response curve of Fig. 6 fitted with a one-site binding hyperbola. Similar to that observed with LY354740, the inhibition of the test EPSP by DCG-IV was accompanied by a reduction in paired-pulse depression. For example, in 100 nM, 200 nM, 500 nM and 5 μM, the absolute percentage changes in paired-pulse were

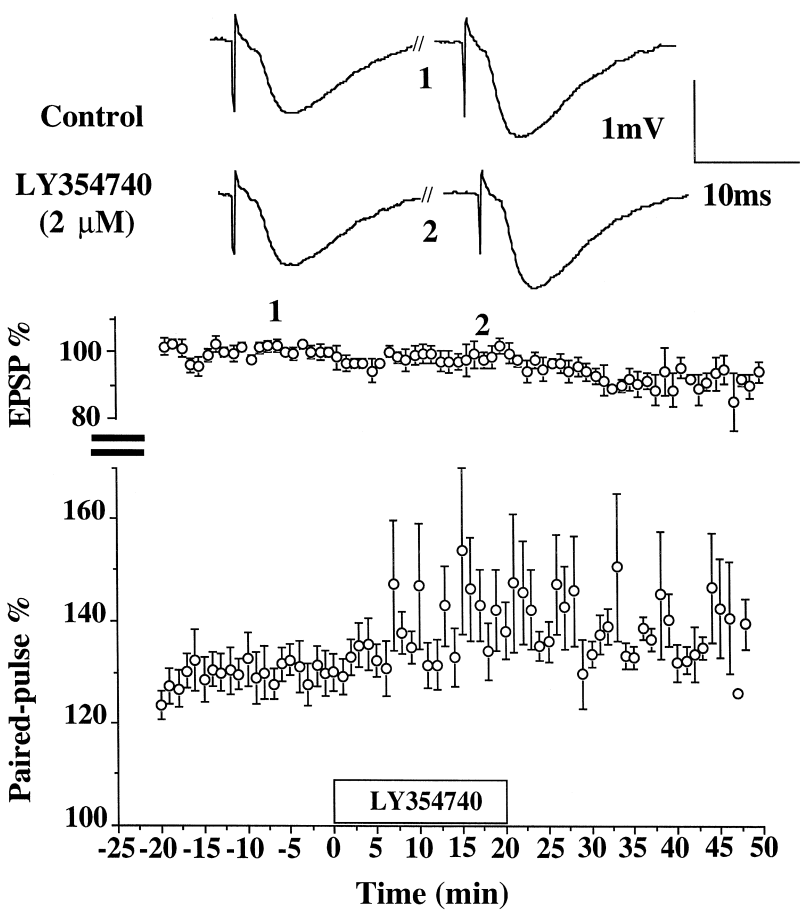


Fig. 5. Dose–response curves for the action of LY354740 and DCG-IV in the medial and lateral perforant paths. The dose–response curves were best fitted with a one-site binding hyperbola (Prism software) $Y = B_{max} \cdot X/(K_d + X)$ where B_{max} is the maximal binding, and K_d is the concentration of ligand required to reach half-maximal binding. The EC_{50} for LY354740 was estimated to be 115 ± 16 nM and 230 ± 58 nM in the medial and lateral perforant paths, respectively. The EC_{50} for DCG-IV was estimated to be 317 ± 54 nM and 334 ± 69 nM in the medial and lateral perforant paths, respectively. Each point plots the mean ± S.E.M. values for four to eight slices.

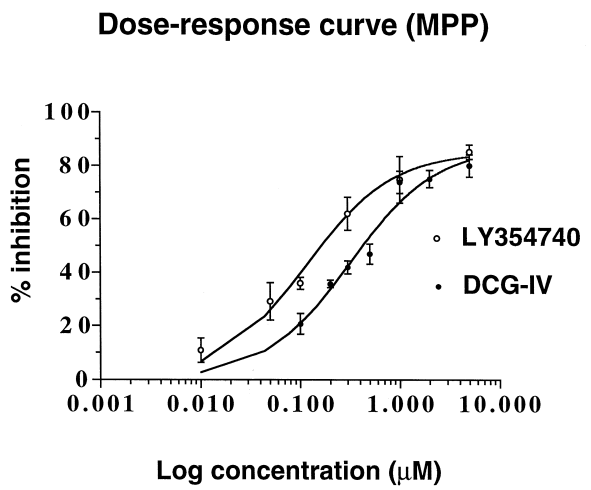
11 ± 3% ($P < 0.05$, $n = 6$), 29 ± 6% ($P < 0.05$, $n = 7$), 42 ± 8% ($P < 0.05$, $n = 7$) and 89 ± 19% ($P < 0.05$, $n = 4$; Fig. 2), respectively.

3.2. LY354740 and DCG-IV inhibit the EPSP in the lateral perforant path

The placement of the recording and stimulating electrodes in the lateral perforant path was verified by the presence of paired-pulse facilitation of EPSPs in response to paired-pulse stimulation.

Both LY354740 and DCG-IV inhibited the EPSP in the lateral perforant path, but with a lower potency and lower maximal inhibition than in the medial perforant path. The EPSP amplitude was significantly reduced by 15 ± 2%, 28 ± 3%, 46 ± 4% and 48 ± 4% in 100 nM, 500 nM, 1 μM and 5 μM LY354740, respectively (Fig. 3, also see Fig. 6). All values were significant ($P < 0.01$, $n = 5$) when tested using a repeated measures analysis of variance. The EC_{50} value was estimated to be 230 ± 58 nM from the dose–response curve (Fig. 6). A dose-dependent significant increase in paired-pulse facilitation accompanied the LY354740-induced depression of the EPSPs, in the lateral perforant path. In control conditions the paired-pulse facilitation was 7 ± 4%. This increased to 15 ± 4%, 20 ± 4%, 25 ± 3% and 25 ± 4% in 100 nM, 500 nM, 1 μM and 5 μM, respectively. All values were significant when tested using the repeated measures analysis of variance ($P < 0.01$, $n = 5$) (Fig. 3).

DCG-IV also inhibited the EPSP in the lateral perforant path, but like the medial perforant path, with a lower potency than LY354740. Perfusion of DCG-IV reduced the EPSP amplitude by 7 ± 3%, 16 ± 5%, 17 ± 5% and 39 ± 7% in 100 nM, 200 nM, 500 nM and 5 μM, respectively. All values were significant when tested using the Student's paired *t*-test ($P < 0.01$, $n = 5$) (Fig. 4). The estimated EC_{50} value was 334 ± 69 nM. Like that with LY354740, the increase in paired-pulse facilitation was relatively small and was dose-dependent. DCG-IV caused an absolute percentage increase in paired-pulse facilitation of 7 ± 4%, 9 ± 4%, 13 ± 7% and 14 ± 6% in 100 nM, 200 nM, 500 nM and 5 μM, respectively. All values were significant when tested using the Student's paired *t*-test ($P < 0.01$, $n = 5$) (Fig. 4).



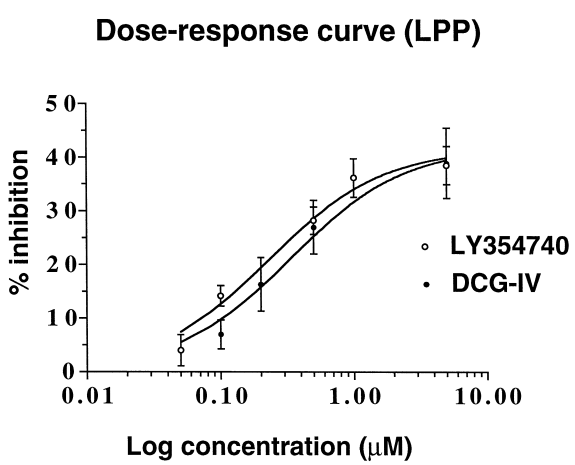


Fig. 6. LY354740 does not produce an inhibition of excitatory synaptic transmission in the CA1 hippocampus. Upper graph, application of 2 μM LY354740 does not produce an inhibition of the EPSP in CA1. Lower graph, LY354740 does not produce a significant change in paired-pulse facilitation in CA1.

3.3. LY354740 has no effect on EPSP amplitude in CA1

LY354740 was applied at a concentration which was sufficient to maximally depress synaptic transmission in both pathways of the dentate gyrus (2 μM). At this concentration LY354740 had no discernible effect, 98.9 ± 2.6% ($P > 0.05$, $n = 4$; Fig. 5).

4. Discussion

The present electrophysiological study has shown that LY354740 is a potent agonist at the group II mGlu receptor responsible for mediating inhibition of EPSPs at the medial perforant path in the dentate gyrus in vitro, with the threshold dose about 10 nM and the EC_{50} close to 100 nM. This is the most potent agonist action at group II mGlu receptors, demonstrated in electrophysiological studies, in this pathway. In comparison with other agonists, electrophysiological studies have shown that DCG-IV has an EC_{50} of close to 300 nM (present study), while LCCG-1 has an EC_{50} of 30 μM (Ugolini and Bordi, 1995). The particularly potent properties of LY354740 are in agreement with neurochemical studies in which LY354740 has been shown to be the most potent group II agonist synthesised, with an EC_{50} of 5 nM and 24 nM for the inhibition of forskolin-stimulated cAMP at expressed mGluR2 receptor and mGlu receptors, respectively (Schoepp et al., 1997b)

and a similar potency for group II mGlu receptor in the rat cerebral cortex and hippocampus group II mGlu receptor (Monn et al., 1997; Schoepp et al., 1997b).

Both LY354740 and DCG-IV were also found to cause a depression of the EPSP in the lateral perforant path, although this path was less sensitive to the agonists, the EC_{50} for LY354740 and DCG-IV being close to 200 nM and 300 nM, respectively, much higher than in the medial path. In addition, the maximal inhibition (40–50%) was much lower than in the medial path (80–90%). Such a reduced sensitivity in the lateral perforant path is likely to reflect a lower density of group II mGlu receptors in this path. A difference in the extent of inhibition by group II mGlu receptor agonists between the medial and lateral perforant path was shown previously by Macek et al. (1996), although this study also found a much lower sensitivity to DCG-IV in both the medial and lateral perforant path than the present study, with the EC_{50} for the action of DCG-IV being 1.6 μM and >3 μM, respectively. This lower sensitivity in the study of Macek et al. (1996) may be due to a developmental decrease in group II mGlu receptor sensitivity, similar to that occurring in CA1 (Vignes et al., 1995; Shigemoto et al., 1997). Thus Macek et al. (1996) used animals of 100–150 g weight, compared to 50–70 g weight in the present study. However, we have not found any developmental decrease in sensitivity to group II mGlu receptor agonists in the medial perforant path of the dentate gyrus (unpublished results). The complete lack of effect of LY354740 in the CA1 region of the hippocampus reflects the absence of group II mGlu receptors in this region of the adult rat, and confirms previous electrophysiological studies using lower potency agonists than LY354740, such as DCG-IV (Gereau and Conn, 1995; Breakwell et al., 1997)

The inhibition of the EPSP by LY354740 and DCG-IV was accompanied by a change in paired-pulse depression/facilitation. Such changes demonstrate that the group II mGlu receptor agonists caused a decrease in the presynaptic probability of transmitter release. The most likely receptor mediating the presynaptic inhibition demonstrated in the present study is mGluR2. Several immunohistochemical studies have shown that mGluR2 is located at a very high density on perforant path axons in the molecular layer of the dentate gyrus, especially in the medial perforant path (Ohishi et al., 1993; Neki et al., 1996; Petralia et al., 1996; Shigemoto et al., 1997). We cannot rule out inhibition via mGluR3 receptors, although this receptor is only present at a much lower density in the hippocampus and is mainly located on glial cells (Petralia et al., 1996). Activation of the group II mGlu receptors may inhibit Ca^{2+} influx, thereby reducing the probability of transmitter release. Alternatively, as the mGluR2 receptors are located preterminally at some distance from the transmitter release sites and corresponding Ca^{2+} channels, rather than presynaptically (Shigemoto et al., 1997), activation of mGluR2 receptors may be linked to opening of K^+ channels, a reduction in the amplitude/duration of the axonal action potential, and thereby reduction of Ca^{2+} influx and the probability of transmitter release.

Acknowledgements

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References

Baskys, A., Malenka, R.C., 1991. Agonists at metabotropic glutamate receptors presynaptically inhibit epsc's in neonatal rat hippocampus. J. Physiol. 444, 687–701.

Bond, A., Monn, J.A., Lodge, D., 1997. A novel orally active group 2 metabotropic glutamate receptor agonist LY354740. NeuroReport 8, 1463–1466.

Breakwell, N.A., Huang, L.-Q., Rowan, M.J., Anwyl, R., 1997. DCG-IV inhibits synaptic transmission by activation of NMDA receptors in area CA1 of rat hippocampus. Eur. J. Pharmacol. 322, 173–178.

Brown, R.E., Reymann, K.G., 1995. Metabotropic glutamate receptor agonists reduce paired pulse depression in the dentate gyrus of the rat in vitro. Neurosci. Lett. 196, 17–20.

Burke, J.P., Hablitz, J.J., 1994. Presynaptic depression of synaptic transmission mediated by activation of metabotropic glutamate receptors in rat neocortex. J. Neurosci. 14, 5120–5130.

Bushell, T.J., Jane, D.E., Tse, H.-W., Watkins, J.C., Garthwaite, J., Collingridge, G.L., 1996. Pharmacological antagonism of the actions of group II and group III mGluR antagonists in the lateral perforant path of rat hippocampal slices. Br. J. Pharmacol. 117, 1457–1462.

Calabresi, P., Mercuri, N.B., Bernadi, G., 1992. Activation of quisqualate glutamate receptors reduces glutamate and GABA-mediated synaptic potentials in the rat striatum. Neurosci. Lett. 139, 41–44.

Desai, M., Conn, P.J., 1991. Excitatory effects of ACPD receptor activation in the hippocampus are mediated by direct effects on pyramidal cells and blockade of synaptic inhibition. J. Neurophysiol. 66, 40–52.

Gereau, R.W., Conn, P.J., 1995. Multiple presynaptic metabotropic glutamate receptors modulate excitatory and inhibitory synaptic transmission in hippocampal area CA1. J. Neurosci. 15, 6879–6889.

Glaum, S.R., Slaber, N.T., Rossi, T.Y., 1992. Role of metabotropic glutamate receptors at the parallel fiber–Purkinje cell synapse. J. Neurophysiol. 60, 1453–1462.

Huang, L.-Q., Rowan, M.J., Anwyl, R., 1997. Group II mGluR agonist inhibition of LTP induction, and group II mGluR antagonist inhibition of LTD induction, in the dentate gyrus in vitro. NeuroReport 8, 687–693.

Kahle, J.S., Cotman, C.W., 1993. L-2-Amino-4-phosphonobutanoic acid and 1*S*,3*R*-1-aminocyclopentane-1,3-dicarboxylic acid reduce paired pulse depression recorded from medial perforant path in the dentate gyrus of hippocampal slices. J. Pharmacol. Exp. Ther. 266, 207–215.

Lovinger, D.M., 1991. *trans*-1-Aminocyclopentane-1-3-dicarboxylic acid (*t*-ACPD) decreases synaptic excitation in rat striatal slices through a presynaptic action. Neurosci. Lett. 129, 17–21.

Lovinger, D.M., Tyler, E., Fidler, S., Merritt, A., 1993. Properties of a presynaptic metabotropic glutamate receptor in neostriatal slices. J. Neurophysiol. 69, 1236–1244.

Macek, T.A., Winder, D.G., Gereau, R.W., Ladd, C.O., Conn, P.J., 1996. Differential involvement of group II and group III mGluRs as autoreceptors at lateral and medial perforant path synapses. J. Neurophysiol. 76, 3798–3806.

Manzoni, O., Michel, J.-M., Bockaert, J., 1997. Metabotropic glutamate receptors in the rat nucleus accumbens. Eur. J. Neurosci. 9, 1514–1523.

McGuinness, N., Anwyl, R., Rowan, M., 1991. The effects of *trans*-ACPD on long-term potentiation in the rat hippocampus. NeuroReport 2, 688–690.

Monn, J.A., Valli, M.J., Massey, S.M., Wright, R.A., Salhoff, C.R., Johnson, B.G., Howe, T., Alt, C.A., Rhodes, G.A., Robey, R.L., Griffey, K.R., Tizzano, J.P., Kallman, M.J., Helton, D.R., Schoepp, D.D., 1997. Design, synthesis and pharmacological characterisation of (+)-2-aminobicyclo[3.1.0]hexane-2-6-dicarboxylic acid (LY354740): a potent, selective and orally active group 2 metabotropic glutamate receptor agonist possessing anticonvulsant and anxiolytic properties. J. Med. Chem. 40, 528–537.

Neki, A., Ohishi, H., Kaneko, T., Shigemoto, R., Nakanishi, S., Mizuno, N., 1996. Pre- and postsynaptic localisation of a metabotropic glutamate receptor, mGluR2, in the rat brain: an immunohistochemical study with a monoclonal antibody. Neurosci. Lett. 202, 197–200.

Ohishi, H., Shigemoto, R., Nakanishi, S., Mizuno, N., 1993. Distribution of the messenger RNA for metabotropic glutamate receptor, mGluR2, in the central nervous system of the rat. Neuroscience 53, 1009–1018.

Pacelli, G.J., Kelso, S.R., 1991. *trans*-ACPD reduces multiple components of synaptic transmission in the rat hippocampus. Neurosci. Lett. 132, 267–269.

Petralia, R.S., Wang, Y.-X., Niedzielski, Wenthold, R.J., 1996. The metabotropic glutamate receptors, mGluR2 and mGluR3, show unique postsynaptic, presynaptic and glial localisations. Neuroscience 71, 949–976.

Schoepp, D.D., Johnson, B.G., Wright, R.A., Salhoff, C.R., Mayne, N.G., Wu, S., Cockerham, S.L., Burnett, J.P., Belegaje, R., Bleakman, D., Monn, J.A., 1997a. LY354740 is a potent and highly selective group II metabotropic glutamate receptor agonist in cells expressing human glutamate receptors. Neuropharmacology 36, 1–11.

Schoepp, D.D., Johnson, B.G., Wright, R.A., Salhoff, C.R., Monn, J.A., 1997b. Second messenger responses induced by the group II metabotropic glutamate receptor agonist LY354740 in the rat brain. Neuropharmacology 36, 1–11, Soc. Neurosci. Abstr. 23, 2024.

Shigemoto, R., Kinoshita, A., Wada, E., Nomura, S., Ohishi, H., Takada, M., Flor, P.J., Neki, A., Abe, T., Nakanishi, S., Mizuno, N., 1997. Differential presynaptic localisation of metabotropic glutamate receptor subtypes in the rat hippocampus. J. Neurosci. 17, 7503–7522.

Sladeczek, F., Momiyama, A., Takahashi, T., 1993. Presynaptic inhibitory action of a metabotropic glutamate receptor agonist on excitatory transmission in visual cortical neurons. Proc. R. S. London B 253, 297–303.

Ugolini, A., Bordi, F., 1995. Metabotropic glutamate group II receptors are responsible for the depression of synaptic transmission induced by ACPD in the dentate gyrus. Eur. J. Pharmacol. 294, 403–410.

Vignes, M., Clarke, V.R.J., Davies, C.H., Chambers, A., Jane, D.E., Watkins, J.C., Collingridge, G.L., 1995. Pharmacological evidence for an involvement of group II and group III mGluRs in the presynaptic regulation of excitatory synaptic responses in the CA1 region of rat hippocampal slices. Neuropharmacology 34, 973–982.

Yokoi, M., Kobayashi, K., Manabe, T., Takahashi, T., Sakaguchi, I., Katssura, G., Shigemoto, R., Ohishi, H., Nomura, S., Nakamura, K., Nakao, K., Katsuki, M., Nakanishi, S., 1996. Impairment of hippocampal mossy fibre LTD in mice lacking mGluR2. Science 273, 645–650.