Psychopharmacology (2005) 179: 292–302
DOI 10.1007/s00213-004-2126-x

ORIGINAL INVESTIGATION

**Simona Spinelli · Theresa Ballard · Silvia Gatti-McArthur · Grayson J. Richards · Martin Kapps · Thomas Woltering · Jurgen Wichmann · Heinz Stadler · Joram Feldon · Christopher R. Pryce**

# Effects of the mGluR2/3 agonist LY354740 on computerized tasks of attention and working memory in marmoset monkeys

Received: 2 June 2004 / Accepted: 19 November 2004 / Published online: 28 January 2005


**Abstract** *Rationale:* LY354740 is a recently developed metabotropic glutamatergic receptor 2 and 3 (mGluR2/3) agonist. A high density of mGluR2 has been reported in terminal fields of the perforant path in rodents and humans, suggesting its involvement in cognitive functions mediated by the temporal lobe, including memory. A small number of in vivo studies in rodents have assessed the effects of LY354740 on memory tasks, reporting the induction of impaired memory for spatial orientation in a water maze task and for delayed match and non-match to position in an operant version of these tasks. *Objective:* In the present primate study, we used radioautography to describe the distribution and intensity of $^{3}$H-LY354740 binding in the hippocampal formation of the common marmoset monkey (*Callithrix jacchus*) relative to the rat. In the major, in vivo part of the study, the effects of systemic LY354740 on computerized tasks of attention and memory were investigated. *Methods:* Adult common marmosets were trained to perform a five-choice serial reaction time (5-CSRT) task and a concurrent delayed match-to-position (CDMP) task from the Cambridge Neuropsychological Automated test Battery (CANTAB). Filter tests of LY354740 effects on motor dexterity and motivation for reward revealed high inter-individual variation in sensitivity; therefore, on the 5-CSRT, subjects were tested at a dose range of 3–10 mg/kg, and on the CDMP, subjects were tested at 1–3 or 3–10 mg/kg. *Results:* Radioautography revealed a relatively low level of $^{3}$H-LY354740 binding in the marmoset hippocampal formation compared to the rat. Despite low binding, LY354740 reduced sustained-attention accuracy in the 5-CSRT, and reduced accuracy in two stages of the CDMP. *Conclusions:* The current study provides novel evidence for the importance of mGluR2/3 in the regulation of primate cognitive functioning.

**Keywords** Metabotropic glutamatergic receptor · LY354740 · Primate · Marmoset · CANTAB · Attention · Working memory

S. Spinelli · J. Feldon · C. R. Pryce (✉)
Behavioural Neurobiology Laboratory,
Swiss Federal Institute of Technology Zurich,
Schorenstrasse 16,
8603 Schwerzenbach, Switzerland
e-mail: pryce@behav.biol.ethz.ch
Tel.: +41-1-6557386
Fax: +41-1-6557203

T. Ballard · S. Gatti-McArthur · G. J. Richards · M. Kapps · T. Woltering · J. Wichmann · H. Stadler
F. Hoffmann-La Roche AG, Pharmaceuticals Division,
CNS-Preclinical Research,
Grenzacherstrasse,
4070 Basel, Switzerland

## Introduction

Glutamate receptors are classified into two different functional groups: ionotropic receptors and metabotropic receptors. At least eight metabotropic glutamate receptors (mGluRs; with splice variants) are described, subdivided into three groups according to their primary structure, second-messenger coupling, and pharmacology. Group I receptors include mGluR1 and 5, group II mGluR2 and 3, and group III mGluR 4, 6, 7 and 8 (Conn and Pin 1997). mGluRs are considered to have a modulatory function and therefore to constitute a pharmacological target for selective drugs with therapeutic potential (Conn and Pin 1997; Holden 2003).

mGluR2 has a distinct distribution in the rodent brain. Of particular interest is the high density of mGluR2 identified in the perforant path, the main input projection between entorhinal cortex and the hippocampal formation (Higgins et al. 2004; Neki et al. 1996; Ohishi et al. 1998; Shigemoto et al. 1997). The entorhinal cortex receives input from multiple neocortical association areas and represents an important convergent site for information into the hippocampus (Eichenbaum 2000; Squire and Knowlton 1995). However, the potential role of mGluR2 in memory regulation has received little attention to date.

Recently, pharmacological agents acting at specific mGluR subtypes have been developed, and these include

the group II-selective agonist LY354740 and antagonist LY341495 (Schoepp et al. 1999). Group II mGluR agonists have been reported to have neuroprotective, anxiolytic/antipanic and anti-Parkinsonism properties, as well as antipsychotic potential (Holden 2003; Pilc 2003; Schoepp et al. 2003). Since LY354740 binds to both mGluR2 and 3, it is likely that some of its effects are mediated more specifically by the activation of one or other of these two receptor subtypes. Such functional specificity is also suggested by the cell types expressing these two receptor subtypes, with mGluR2 expressed primarily in neurons and mGluR3 primarily in glial cells (Ohishi et al. 1993a,b, 1994; Tanabe et al. 1993) Although mGluR2/3 agonists have been considered as targets with respect to a broad range of psychiatric and neurodegenerative disorders, studies of the effects of mGluR2/3 activation on cognitive functions have been confined to a small number of rodent studies. LY354740 (1–10 mg/kg) induced a delay-dependent performance deficit in a T-maze task (Aultman and Moghaddam 2001), supporting a role of mGluR2/3 in working memory, and also induced working memory deficits in operant versions of both delayed match and delayed non-match to position (D(N)MP) tasks (Higgins et al. 2004). The latter study also reported a deficit in spatial learning in a water maze task with LY354740 (10 mg/kg, i.p.) and enhanced acquisition in the water maze with the group II antagonist LY341495 (1 mg/kg, i.p). These findings provide evidence that group II receptors can have an important role in learning and memory in rodents.

In a previous study (Spinelli et al. 2004), we demonstrated that a New World primate, the common marmoset monkey (*Callithrix jacchus*), can perform the five-choice serial reaction time (5-CSRT) task and a concurrent delayed match to position (CDMP) task, from the CAmbridge Neuropsychological Test Automated Battery (CANTAB; Cambridge Cognition Ltd., Cambridge, UK), a computerized battery of neuropsychological tasks presented as icons on a touch-sensitive computer screen. The 5-CSRT task has been used widely to assess visual attention in rodents (Carli et al. 1983; Robbins 2002) and, in the case of the CANTAB version, humans and rhesus macaques (Sahakian and Coull 1993; Weed et al. 1999). The CDMP task, comprising two interpolated delayed match to position tasks, exhibits similarities with the operant DMP task for rodents (Dunnett 1985, 1993), and also with CANTAB memory tasks used widely in humans, including the delayed match to sample, spatial recognition, and visuo-spatial paired associates learning memory tasks (Robbins et al. 1997). We recently demonstrated that the performance of marmoset monkeys in the 5-CSRT and CDMP tasks is sensitive to pharmacological manipulation of the cholinergic system, using nicotine and scopolamine (Spinelli 2004).

The aims of the present common marmoset study were to describe the distribution and abundance of mGluR2 and 3 in the temporal lobe of the adult brain using $^{3}$H-DCG-IV and $^{3}$H-LY354740-based radioautography, and to investigate the effects of LY354740 on attention and memory using the CANTAB 5-CSRT and CDMP tasks.

## Materials and methods

All experimental procedures were conducted under permit and in accordance with the Animal Protection Act (1978), Switzerland.

### [$^{3}$H]LY354740 and [$^{3}$H]DCG-IV binding to marmoset tissue sections and brain membranes

Common marmosets, two male and two female, were sedated using Saffan (alphaxalone/alphadolone, 10 mg/kg i.m.) and sacrificed with an overdose of sodium pentobarbital. The brain was rapidly removed, frozen in isopentane at −60°C and then stored at −80°C prior to sectioning. Cryostat sections of 15 μm in the coronal plane through the hippocampal formation were thaw-mounted onto gelatin-coated slides. In addition, parasagittal and coronal sections of Wistar rats were also prepared. The rats were sacrificed by decapitation under anesthesia with halothane, and then the brain was rapidly removed, frozen in dry ice and then stored at −80°C. Parasagittal cryostat sections (12 μm) were thaw-mounted on pre-cleaned slides and stored at −20°C until use. The brains of two adult female marmosets were used for the preparation of membranes. Blocked tissue from the prefrontal cortex, striatum, hippocampus, temporal cortex and cerebellum was stored at −80°C prior to preparation. Brain tissue from Wistar rats was used as reference for binding studies. Methodological differences in the processing of marmoset and rat brains reflect the histology protocols of the Swiss Federal Institute of Technology and Hoffmann La Roche laboratories, respectively.

Quantitative receptor radioautography with [$^{3}$H]LY354740 and [$^{3}$H]DCG-IV was performed as described in Higgins et al. (2004). Membrane binding studies were performed as described in Schaffhauser et al. (1998) and Mutel et al. (1998), with protein content measured using the bicinchoninic acid method.

### Western blot analysis of mGluR2/3 expression in the marmoset brain

Two polyclonal antibodies were used to detect mGluR2 and 3 gene products in the marmoset brain (human and rat mGluR2 are 97% identical at the amino acid level). Membranes were distributed in aliquots containing 20 μg protein in assay buffer with protease inhibitors. The aliquot was thawed and centrifuged at 13,000 rpm for 20 min at 4°C. The pellet was resuspended in 40 μl Laemmli buffer (with or without 20 mM DTT), heated for 5 min at 50°C and loaded on a 7.5% polyacrylamide gel. The transfer on to nitrocellulose membranes (Bio-Rad) was carried out in 25 mM TRIS, 192 mM glycine and 20% methanol using the Semidry Transfer in Trans blot SD transfer Cell (Bio-Rad) (45 min, 20 V). After rinsing in TBS Tween, blocking was carried out in 1% BSA/TBST for 1 h. The selective mGluR2 rabbit polyclonal antibody (Upstate Biotechnology, Lake Placid, NY, USA; 1:2,000, epitope corresponding to AA

829–845 of rat mGluR2), and a rabbit polyclonal mGluR2/3 antibody (Calbiochem, 1:1,000, raised against the carboxy-terminus peptide NGREVVDSTTSSL) were used as primary antibodies. After overnight incubation at 4°C, the filter was incubated (1 h; RT) with a secondary antibody, anti-POD (1:10,000 in 0.5% milk/TBST) or with Alexa Fluor 680 goat anti-rabbit antibody (1:20,000 in PBST for fluorescent detection; Molecular Probes, Eugene, OR, USA). The detection system was either ECL Plus (Amersham Biosciences) following manufacturer's instructions, or Odissey Scanner analysis for protein semi-quantification. In the case of fluorescent staining, actin was also stained in the same gel using a mouse anti-actin monoclonal antibody (1:10,000) and the IRDye 800 anti-goat IgG (Rockland, Gilbertville, PA).

## Marmoset subjects and care

The behavioural study was conducted with 17 adult common marmosets (ten females and seven males), aged 2–12 years. Details of caging, maintenance and home-cage behavioural training and testing are provided in Spinelli et al. (2004). Eight subjects were allocated to the non-cognitive tasks used to establish a dose range of LY354740 that did not impair motor dexterity or motivation. Two of these subjects and a further nine subjects were studied in either an attention task or a memory task. Training on these cognitive tasks required several months. Five and six subjects were trained to a level of stable accurate performance on the attention and memory tasks, respectively. We attempted to train several additional animals on these tasks but they did not attain a stable level of above-chance performance. Prior to the present study, the same marmosets had been included in a pharmacological validation of the cognitive tasks based on two reference cholinergic drugs, scopolamine and nicotine (Spinelli 2004). These validation studies were completed 2 months prior to the onset of the present study.

## ORD task performed with the WGTA

The object reaching with detour (ORD) task was used to establish the effects of LY354740 on motor capabilities (Taylor et al. 1990), and was presented using a Wisconsin General Test Apparatus (WGTA) [42 (*L*)×40 (*W*)×45 (*H*) cm]. The ORD task required the marmoset to retrieve a reward from within a transparent Plexiglas cube that measured 4×4×4 cm and was open on one side. Pieces of dry biscuit soaked briefly in banana-flavored milkshake were used as reward. The cube was presented with the open side on the left, right or front of the monkey. Two black diagonal lines were drawn on each side and the back of the cube; therefore, subjects could see the reward placed inside the cube and also had visual cues (Hauser 1999). During test sessions of a maximum of 24 trials, the reward was placed in the centre of the cube and the orientation of the open side was to the front, left, or right relative to the subject, according to a pseudo-random schedule, but with an equal number of each trial type. The following measures were calculated: number of trials per session; number of successful responses, i.e. reward retrieved at the first attempt; number of impulsive responses, i.e. at the first attempt the subject reaches to the front when the open side was on the left or right; number of perseverative responses, i.e. at the first attempt the subject reaches to a wrong side on the left or right, where this side was correct on the previous trial. After several test sessions, marmosets attained a stable level of performance of ca. 80% successful responses. Against this background, the effects of LY354740 were assessed, with the major aims of detecting doses that did or did not (1) impair dexterity, defined as the first response on a trial being to the open side of the cube but the first retrieval attempt unsuccessful, or (2) reduce motivation, defined as a reduced number of trials completed.

## CANTAB: apparatus, training and tasks

The Monkey CANTAB Testing Station (marmoset set-up, Cambridge Cognition Ltd., Cambridge, UK) and the training procedures used for home cage CANTAB testing are described extensively in Spinelli et al. (2004).

### *Progressive ratio schedule of reinforcement*

In four subjects, effects of LY354740 on feeding motivation were assessed using the progressive ratio (PR) schedule of reinforcement. The major measures of motivation were the total numbers of responses made and reinforcements obtained (Spinelli et al. 2004).

### *Five-choice serial reaction time task*

The 5-CSRT task was applied in five subjects in order to assess the effects of LY354740 on divided and sustained attention (Spinelli et al. 2004; Taffe et al. 2002). The duration for which the lever had to be depressed before the blue stimulus appeared was set to a range of 0.1–1.0 s. The dependent measures studied were: total number of trials performed; percent correct responses/total trials; percent accuracy, i.e. percent correct responses/total (correct+incorrect) responses; percent omissions (trials where a stimulus was presented but the subject did not touch the screen); percent release lever too soon (subject did not depress the lever until the blue target stimulus appeared on the screen); lever release latency (time taken to release the lever after the stimulus appeared on the screen, when response was correct); and movement time (time from lever release until screen touching, when response was correct).

*Concurrent delayed match to position task*

The CDMP task, studied here in terms of LY354730's effects in six subjects, was derived from the monkey CANTAB visuo-spatial paired associates learning (vsPAL) task (Spinelli et al. 2004; Taffe et al. 2002). The design of the task is given in Fig. 1.

LY354740 and behaviour

The compound (1*S*,2*S*,5*R*,6*S*)-2-amino-bicyclo[3.1.0]hexane-2,6-dicarboxylic acid (LY354740; synthesized at F. Hoffmann-La Roche, Basel, Switzerland), was dissolved in 0.9% NaCl solution and pH was adjusted to ≈7.0 with sodium hydroxide. Doses were injected i.p. in a volume of 0.6 ml/kg and with a pre-treatment time of 40 min. Drug doses of 1–5 mg/kg were counterbalanced in each experiment, whereas higher doses (7.5 and 10 mg/kg) were injected in ascending order and only in cases where subjects had not exhibited specific side effects at a lower dose. Animals were typically tested daily from Monday to Friday, with injections occurring twice per week, saline usually on Tuesday or Wednesday and LY354740 on Thursday or Friday; LY354740 testing was performed only if the saline session performance was similar to baseline performance in that week, otherwise the saline session was repeated.

*Experiment 1: ORD task and PR schedule*

The effects of LY354740 at 1.0–7.5 mg/kg i.p. on motor performance and motivation were assessed using the ORD task and PR schedule, and results indicated marked individual differences in LY354740 sensitivity. However, 3 mg/kg was well-tolerated by most of the eight subjects, and this dose was used for cognitive task pre-injections. One week prior to the beginning of cognitive testing (Experiments 2 and 3), subjects were pre-exposed for 2 consecutive days to 3 mg/kg LY354740; and observed for behavioural effects. According to reactions to pre-injections, "high sensitivity" and "low sensitivity" subjects were identified. The high-sensitivity group was tested at 1, 2 and 3 mg/kg and the low-sensitivity group at 3 and 5 mg/kg. In the latter group, if LY354740 did not affect task performance, doses of 7.5 and finally 10 mg/kg were administered. The maximum dose of 10 mg/kg was based on rat findings of reduced spontaneous locomotor activity and operant chain pulling behaviour at this dose (unpublished data).

*Experiment 2: 5-CSRT task*

All five subjects were in the LY354740 low-sensitivity group and tested with 3 and 5 mg/kg LY354740. When injected with 5 mg/kg, two animals did not complete all trials and showed considerable side effects. The other three monkeys were also tested at 7.5 and 10 mg/kg. In Experiment 2a, test conditions were a maximum session length of 20 min or a maximum number of 45 trials, with a random-balanced design of three stimulus display durations (0.1, 0.5 and 1.0 s) and 15 trials per duration. For each subject, the highest dose that did not induce a reduction in the number of trials completed was repeated in Experiment 2b. Since in Experiment 2a there was no effect of LY354740 on accuracy, the attentional load of the task was increased in Experiment 2b by using two SDs, 0.1 and 0.5 s, and increasing the number of trials to 60. Test conditions were a maximum session length of 25 min or 60 trials, with a random-balanced design of 30 trials per SD.

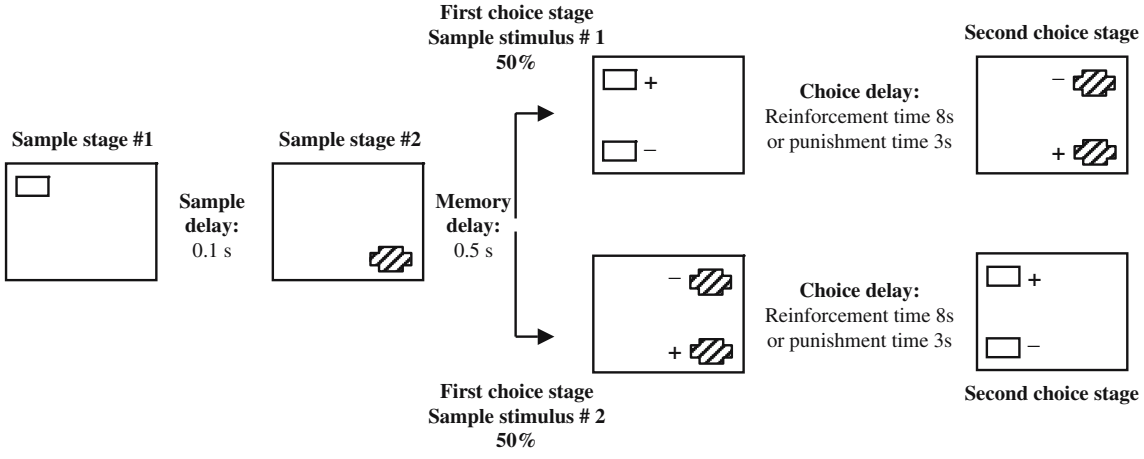


**Fig. 1** Schematic of the test sequence in the concurrent delayed match-to-position task. Each trial comprises either a sequence of stimulus presentation in which the first choice stage uses sample stimulus/position #1 such that the subject has to shift between stimuli and positions (*upper scheme*), or a sequence of stimulus presentation in which the first choice stage uses sample stimulus/position #2 and therefore does not require a shift (*lower scheme*). Each stimulus disappears immediately after responding. For both sequences, a correct choice at choice stage 1 results in 8-s reinforcement followed by a return to the screen for choice stage 2, whilst an incorrect choice at choice stage 1 results in choice stage 2 following a 3-s punishment time out. It should be noted that marmosets did not need to discriminate between stimuli #1 and #2 in order to make correct responses. This was due to the non-overlap between the sample and distracter positions used for stimulus #1 and stimulus #2 within each trial

*Experiment 3: CDMP task*

The maximum session length was 25 min or 35 trials. Two of six subjects performed consistently less than 35 trials and therefore their maximum number of trials was reduced to 30. Three of six subjects were of high sensitivity and tested with 1, 2 and 3 mg/kg (doses counterbalanced) and three were of low sensitivity and tested with 3 and 5 mg/kg, doses counterbalanced, and then at 7.5 and 10 mg/kg.

LY354740 pharmacokinetics using HPLC

After cognitive testing, in an attempt to gain insight into observed individual differences in LY354740 sensitivity, plasma concentrations were measured in low- and high-sensitivity subjects. In five marmosets, a blood sample (0.3 ml from the femoral vein) was withdrawn into an EDTA-primed syringe. LY354740 was administered at 2–3 mg/kg ($n$=3 high-sensitivity subjects) or 5–10 mg/kg ($n$=2 low-sensitivity subjects). Further blood samples were then withdrawn at 30 min, 60 min and 24 h post-treatment. Bloods were placed immediately on ice and then centrifuged, and plasma was stored at −80°C prior to analysis. Plasma samples were treated with 3 volumes of methanol also containing an internal standard. The samples were centrifuged (3,500 $g$, 20 min 10°C) and the supernatant injected directly onto a high pressure liquid chromatography (HPLC) system. HPLC analysis was conducted on a normal phase column (GROM Spherisorb-CN 125×2 mm) at 40°C using a polarity gradient (phase A methanol and phase B methanol/ formic acid 1% 20:80) with a flow rate of 300 μl/min. A PE-Sciex API-2000 MS/MS mass spectrometer was used for detection (ion source: turbo ion spray ionisation).

Data analysis

Scores on the ORD task and PR schedule were analysed using one-way analysis of variance (ANOVA), with LY354740 dose as a within-subject factor. Performance and latency scores on the 5-CSRT task were analysed using a two-way ANOVA with stimulus duration and LY354740 as within-subject factors. For Experiment 2b, data were divided into two trial bins (1–30, 31–60) and analysed using a nested three-way ANOVA with within-subject factors of SD, LY354740 and trial bins. Performance and latency scores on the CDMP task were also analysed using a two-way ANOVA with choice stage and LY354740 as within-subject factors. Statistical significance was set at $p<0.05$, and significant effects were analysed post-hoc using Fisher's protected least significant difference for pair-wise comparisons or $t$-test based on the error term derived from the appropriate overall ANOVA.

## Results

mGluR2/3 radioautography and Western blot

The binding density of both [$^3$H]LY354740 and [$^3$H]DCG-IV by marmoset tissue was considerably reduced relative to that of rat tissue. Table 1 presents a comparison of the binding densities of different brain regions in adult marmoset and adult rat, and the distribution and abundance of total binding sites (>85% specific) is illustrated in Fig. 2. In the rat, the highest level of binding was to be observed in the lacunosum moleculare/perforant path of the hippocampal formation (Fig. 2a). In the marmoset there was rarely the same high level of binding, and the lacunosum moleculare was barely distinguishable from the surrounding tissue, other than by histology (Fig. 2b). Marmoset membrane binding studies using homogenate from different brain areas showed the presence of specific binding for [$^3$H]LY354740 with a $K_d$ of about 10 nM in hippocampus and 30 nM in striatum, using one-site model curve fitting. Calculated $B_{max}$ values were always below 300 fmol/mg of protein with the lowest specific binding in cerebellum. The relatively low expression of mGluR2 and mGluR3 in marmoset versus rat cortical regions was also confirmed by the Western blot studies (data not shown).

Neuropsychological tasks

*Experiment 1: ORD task and PR schedule*

Table 2 presents the percent trials completed in the ORD task. One subject already exhibited a marked reduction in trials completed at 1 mg/kg, as well as gastrointestinal side effects at 3 mg/kg, and therefore was not injected with 5 mg/kg. Three animals completed at least 70% of trials at all doses; in these subjects, for percent successful responses, there was no significant effect of LY354740, with performance consistent at a mean of 75–80% at each dose. In the PR schedule, LY354740 at 1 or 3 mg/kg did not exert a significant effect on the number of responses performed or reinforcements obtained, e.g. total responses were: SAL

**Table 1** [$^3$H]LY354740 and [$^3$H]DCG-IV binding (fmol/mg protein) by marmoset and rat brain

| Region | [$^3$H]LY354740 | | [$^3$H]DCG-IV | |
|---|---|---|---|---|
| | Rat | Marmoset | Rat | Marmoset |
| Lacunosum moleculare | 7,507±1,181 | 1,382±227 | 4,757±408 | 464±93 |
| Dentate gyrus | 4,594±820 | 1,560±343 | 2,411±309 | 605±153 |
| CA1 | 1,062±236 | 1,144±377 | 432±24 | 128±49 |
| Caudate putamen | 3,249±502 | 1,537±251 | 2,530±527 | 524±53 |
| Cortex | 4,795±653 | 1,310±199 | 3,086±147 | 384±49 |

Values are mean±SEM based on $N$=3–4 subjects, and two to four sections per subject

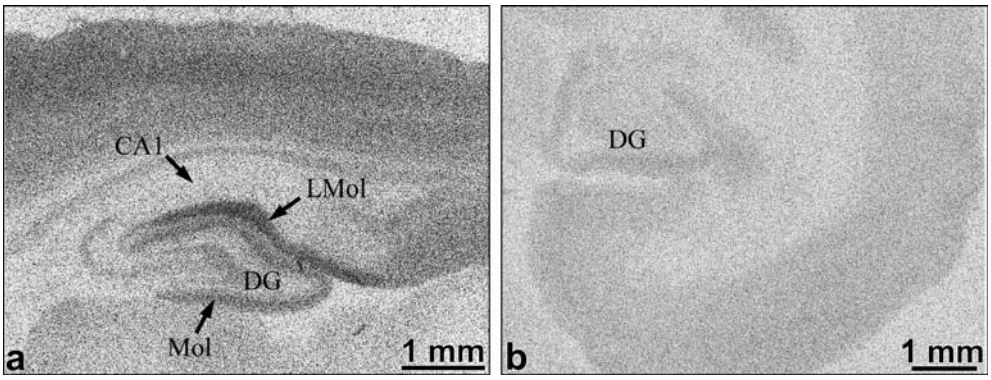


**Fig. 2** Representative autoradiograms of [$^3$H]LY354740 binding in the hippocampal formation in **a** a rat parasagittal section and **b** a marmoset coronal section

147±41; 1 mg/kg 136±24; 3 mg/kg 156±27. Two subjects that did not exhibit behavioural side effects at 3 mg/kg were treated with up to 7.5 mg/kg and at these doses also did not exhibit altered motivation, e.g. total responses at 7.5 mg/kg: 140±3.

Therefore, Experiment 1 identified high individual variability in LY354740 effects on motivation. The dose of 3 mg/kg was well tolerated by six of eight subjects tested and this was confirmed by qualitative behavioural observations conducted across 60 min post-injection. The side effects observed with LY354740 were increased frequency of some natural marmoset behaviours, e.g. head rubbing on branches, head and body shaking, and repeated scratching of body and tail; tail swirling, which is not in the marmoset's natural repertoire, was also observed. The dose of 3 mg/kg was selected for pre-treatment prior to cognitive testing, and behavioural reaction to two such pre-treatments provided the main indication of individual sensitivity. High-sensitive subjects demonstrated high frequencies of head rubbing on branches, shaking and scratching, while low-sensitive subjects did not.

*Experiment 2: 5-CSRT tasks*

Five low-sensitive subjects were treated with 3 and 5 mg/kg LY354740, and at 5 mg/kg two monkeys performed only 42% or 71% of trials and three performed all trials up to 10 mg/kg. With SDs of 0.1, 0.5 and 1.0 s and 45 trials/session (Experiment 2a), ANOVA was conducted with two marmosets at 3 mg/kg and three at 10 mg/kg. There was a significant main effect of SD on accuracy [$F_{(2,8)}$=16.12, $p$<0.002] in the absence of a significant main effect of LY354740. Fisher's post-hoc analysis indicated that percent accuracy was significantly reduced at SD 0.1 s (68±5) compared to SD 0.5 (94±2; $p$<0.02) and 1.0 s (96±2; $p$<0.01). A significant main effect of SD was also found on percent correct responses/total responses [$F_{(2,8)}$=7.92, $p$<0.02], with Fisher's post-hoc analysis indicating that percent correct responses was significantly reduced at SD 0.1 s (53±4) compared to SD 0.5 s (75±4; $p$<0.03) and 1.0 s (83±4; $p$<0.005). With this same measure there was no main effect of LY354740. There was a trend towards a significant decrease in lever release latency due to LY354740 [$F_{(1,4)}$=6.64, $p$<0.07]. None of the other measures of the task (omissions, release lever too soon, response latency) was affected by SD ($p$>0.1) or LY354740 ($p$>0.1).

**Table 2** Effects of LY354740 (mg/kg) on motivation to perform the ORD task

| Subject | Percent trials completed | | | |
|---|---|---|---|---|
| | Saline | LY 1 | LY 3 | LY 5 |
| Con | 100.0 | 100.0 | 100.0 | 12.5 |
| Ing | 100.0 | 41.7 | 0.0 | – |
| Lul | 100.0 | 100.0 | 100.0 | 0.0 |
| Dia | 100.0 | 100.0 | 100.0 | 83.3 |
| Dan | 100.0 | 83.3 | 100.0 | 100.0 |
| Ivo | 100.0 | 83.3 | 70.8 | 100.0 |

The maximum number of trials per session was 24

In Experiment 2b, the attentional load of the task was increased by presenting 60 trials and employing SDs of 0.1 and 0.5 s. Under these task conditions, four marmosets performed all trials across several sessions; one did not and was subsequently excluded. Three animals were tested at 10 mg/kg and one at 3 mg/kg (this latter subject performed only 71% of 45 trials in Experiment 2a at 5 mg/kg). For accuracy (Fig. 3a), there was a significant SD × LY354740 × trial-block interaction [$F_{(1,3)}$=10.73, $p$<0.05], and a significant SD × trials block interaction [$F_{(1,3)}$= 32.55, $p$<0.02], in addition to a main effect of LY354740 [$F_{(1,3)}$= 43.44, $p$<0.008] and a main effect of SD [$F_{(1,3)}$= 631.4, $p$<0.0001]. Post-hoc $t$-comparisons indicated that LY354740 significantly reduced accuracy at SD 0.1 s during the second block of trials (31–60) [$t_{(3)}$=4.65, $p$<0.02], in the absence of significant effects during the second block of trials (31–60) at SD 0.5 s or during the first block of trials (1–30) at either SD 0.1 or 0.5 s (Fig. 3a). For percent correct responses/total trials, there was a significant main effect of SD [$F_{(1,3)}$=533.63, $p$< 0.0002], a significant main effect of trial block [$F_{(1,3)}$= 13.96, $p$<0.04], and a trend to a significant main effect of LY354740 [$F_{(1,3)}$=9.12, $p$<0.06]. These findings reflected the decrease in correct responses at SD 0.1 s (55±5.1) versus 0.5 s (78±3.6), the decrease in correct responses at trials 31–60 (61±6.2) versus 1–30 (72±3.8), and the tendency for LY354740 to decrease correct responses at trials 31–60 (SAL 71±6.3; LY 51±9.9). For omissions, there was no significant effect involving LY354740, but a significant trial-block main effect [$F_{(1,3)}$=108.9,

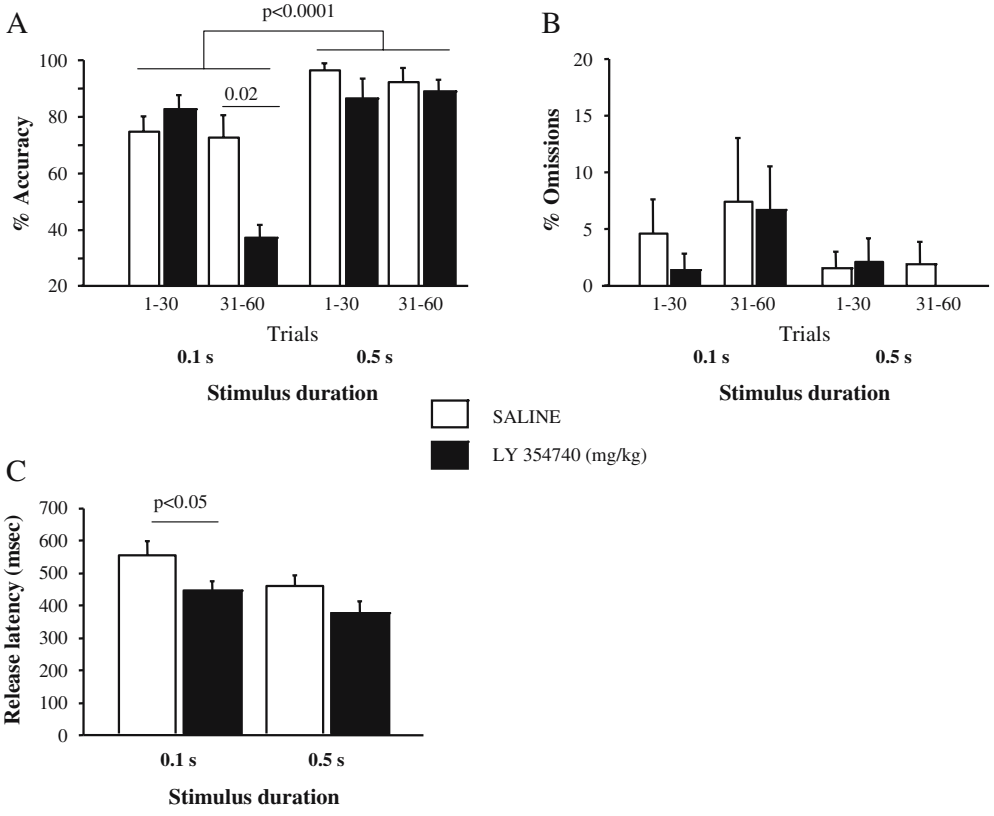


**Fig. 3** Effect of LY354740 (3 or 10 mg/kg) on performance (mean±SEM) of four marmosets in the five-choice serial reaction time task (stimulus durations = 0.1 and 0.5 s, trials = 60) as measured in terms of **a** percent accuracy, **b** percent omissions/ total trials, and **c** lever release latency

$p<0.002$], and a trend to a significant SD main effect [$F_{(1,3)}=9.78$, $p<0.06$] (Fig 3b). For lever release latency, there was a significant main effect of LY354740 [$F_{(1,3)}=21.24$, $p< 0.02$] and a trend to a significant main effect of SD [$F_{(1,3)}= 9.83$, $p<0.06$] (Fig. 3c). Post-hoc *t*-comparison showed that LY354740 induced a significant reduction in lever release latency at SD 0.1 s [$t_{(3)}=3.66$, $p<0.05$], and tended to do so at SD 0.5 s [$t_{(3)}=2.75$, $p<0.1$]. LY354740 did not significantly affect any other measure of the 5-CSRT task.

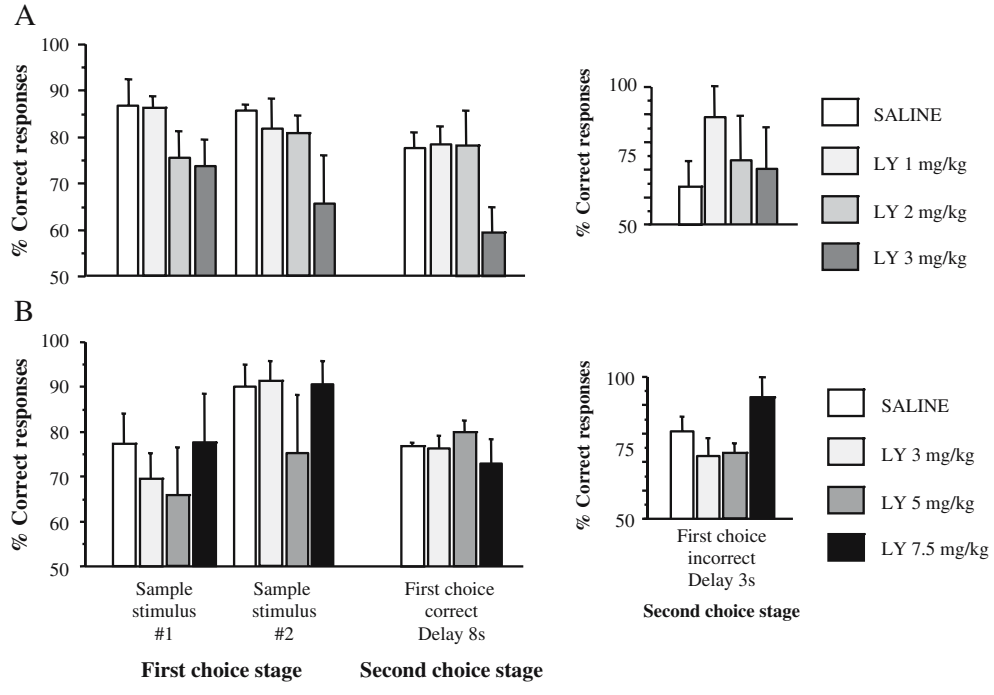


**Fig. 4** Effects of LY354740 on performance in the concurrent delayed match-to-position task in **a** high-sensitivity subjects that received 1–3 mg/kg and **b** low-sensitivity subjects that received 3–7.5 mg/kg. Percent correct responses (mean±SEM, $n$=3) across the four possible sample stimuli × choice stage conditions are given. In the *main figure*, the values at choice stage 2 are based on the average of both trial types, i.e. sample #1 presented at choice stage 1, sample #2 presented at choice stage 1, correct response at choice stage 1. The *inset figure* shows performance at choice stage 2 based on the average of both trial types, i.e. sample #1 presented at choice stage 1, sample #2 presented at choice stage 1, incorrect response at choice stage 1

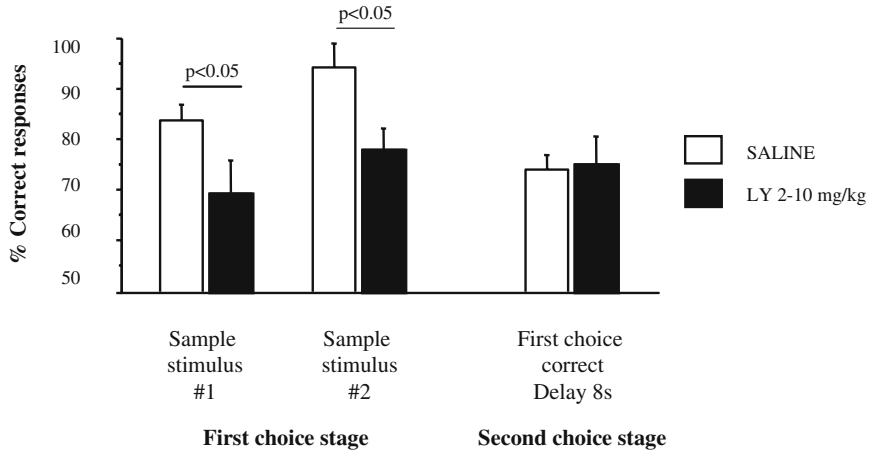


**Fig. 5** Effects of LY354740 (2–10 mg/kg) in six marmosets on the concurrent delayed match-to-position task. Mean ±SEM percent correct responses at choice stage 1 with sample stimuli #1 and #2, and at choice stage 2 when response at choice stage 1 was correct and subjects received 8 s reward. For each subject, the dose selected for this analysis was the highest at which it performed 80–100% of trials. The saline data were those obtained in the same study week as the selected LY354740 dose data

*Experiment 3: CDMP task*

Of the six marmosets trained in the CDMP task, three were high-sensitive and tested at 1, 2 and 3 mg/kg, and three low-sensitive and tested at 3, 5, 7.5 mg/kg and two also at 10 mg/kg. Figure 4a presents the CDMP performance of the high-sensitivity group: there was a significant main effect of LY354740 on percent correct responses [$F_{(3,6)}$= 24.9, $p$<0.0009], in the absence of an interaction with or a main effect of stage. Fisher's post-hoc analysis revealed a dose-dependent effect of LY354740, with performance at 3 mg/kg significantly reduced compared to saline ($p$<0.0002), 1 mg/kg ($p$<0.0004) and 2 mg/kg ($p$< 0.002). For response latency, there was a significant main effect of stage [$F_{(2,4)}$=139.21, $p$<0.0002] in the absence of a main effect of LY354740: first choice stage with sample stimulus #1: 1,006±67 ms, first choice stage with sample stimulus #2: 1,028±65 ms, second choice stage after 8 s delay: 3,334±225 ms. For the low-sensitivity group, there was a trend towards a significant main effect of LY354740 on percent correct responses [$F_{(3,6)}$=4.12, $p$<0.07] in the absence of a significant effect involving stage (Fig. 4b). Doses of 3 and 5 mg/kg tended to decrease performance at the first stage of the task, but this was not significant for any single stage type. As for the high-sensitivity group, there was a significant main effect of stage on response latency [$F_{(2,4)}$=21.44, $p$<0.008], in the absence of any significant LY354740 effect: first choice stage with sample stimulus #1: 1,095±153 ms, first choice stage sample with stimulus #2: 806±46 ms, second choice stage after 8 s delay: 3,077 ±259 ms. Although these three subjects performed sessions at each of these doses, at 7.5 mg/kg one subject performed only 25 of 35 trials.

An individual-specific dose analysis was performed that included all six subjects, with the dose selected for each subject being the highest at which it performed at least 80% of the trials; this yielded a dose range of 2–10 mg/kg LY354740. There was a significant main effect of LY354740 on percent correct responses [$F_{(1,5)}$=11.4, $p$< 0.02], and a tendency for a LY35740 × stage interaction [$F_{(2,10)}$=3.26, $p$<0.09], in the absence of any significant effect involving stage (Fig. 5). Post-hoc $t$-comparison revealed a significant LY354740-induced performance impairment at the first choice stage when sample stimulus was #1 [$t_{(10)}$=2.8, $p$<0.05] and when sample stimulus was #2 [$t_{(10)}$=3.14, $p$<0.05], and no effect of LY354740 at the second choice stage after 8 s delay. Response latency was significantly affected by stage [$F_{(2,10)}$=56.4, $p$<0.001], in the absence of a LY354740 effect: first choice stage with sample stimulus #1: SAL 1,101±250 ms, LY354740 934±80 ms; first choice stage sample with stimulus #2: SAL 852±58 ms, LY354740 952±62 ms; second choice stage after 8 s delay: SAL 3,208±457 ms, LY354740 3,168±227 ms.

## Pharmacokinetics

Given the marked individual differences in LY354740 sensitivity, we investigated the existence of differences in drug metabolism. Five subjects were administered LY354740 at the maximum dose at which they had been studied, and their plasma concentrations were monitored. As given in Table 3, the plasma titres of LY354740 were proportional to the dose injected at 30 min post-injection. Comparing titres at 60 and 30 min post-injection, the relative decrease in the two low-sensitivity marmosets was

**Table 3** LY354740 plasma titres (ng/ml) in marmosets with different behavioural sensitivity

| Subject | Task | Sensitivity | Dose (mg/kg) | Time relative to i.p. injection | | | |
|---|---|---|---|---|---|---|---|
| | | | | −1 min | 30 min | 60 min | 24 h |
| Cat | CDMP | Low | 5 | <50 | 15,500 | 7,100 | <50 |
| Hel | CDMP | Low | 10 | <50 | 36,500 | 19,350 | <50 |
| Con | CDMP | High | 2 | <50 | 9,400 | 6,980 | <50 |
| Cas | CDMP | High | 3 | <50 | 9,870 | 4,470 | <50 |
| Ils | 5-CSRT | High | 3 | <50 | 8,600 | 10,800 | <50 |

greater than that in two of the three high-sensitivity marmosets. This suggests that sensitivity differences could be related in part to differences in the efficacy of LY354740 metabolism.

## Discussion

To the best of our knowledge, this is the first study of the effects of the mGluR2/3 agonist LY354740 on the performance of cognitive tasks in a primate species, and also the first description of mGluR2/3 ligand binding in the hippocampal formation of a non-human primate species.

Using [$^3$H]LY354740 and [$^3$H]DCG-IV, mGlurR2/3 binding by the hippocampal formation, and in particular the lacunosum moleculare, was low in the marmoset compared to the rat; indeed, the marmoset appears to exhibit low levels of hippocampal mGluR2/3 relative to both rat and human (Blumcke et al. 1996; Crook et al. 2002; Lee et al. 2004). Western blot studies, using prefrontal cortex tissue, suggest that marmoset mGluR3 expression was greater than that of mGluR2; despite this, it is unlikely that low marmoset binding was due to exclusive mGluR3 binding. mGluR2 knock-out mice exhibit normal basal synaptic transmission and are without cognitive impairment relative to the wild-type in terms of water maze spatial navigation (Higgins et al. 2004; Yokoi et al. 1996). These results indicate that mGluR2 is non-essential in at least some learning and memory tasks known to require temporal lobe integrity (Eijkenboom et al. 2000; Gerlai et al. 2002; Skelton and McNamara 1992). However, in the present study and despite the low levels of mGluR2/3 binding to LY354740, this agonist exerted marked behavioural effects and impaired cognition at doses at and below those demonstrated to impair cognition in rodent species.

Establishing the appropriate dose range of LY354740 for application in cognitive tasks was achieved using the ORD task to assess motor dexterity (Taylor et al. 1990) and the PR schedule for appetitive motivation (Spinelli et al. 2004). ORD and PR schedule performance and, in addition, observations of general post-treatment behaviour, each indicated marked individual differences in sensitivity to LY354740. Since 3 mg/kg was generally well tolerated, cognitive-task subjects were pre-treated with this dose. Depending on their general behavioural reactions, the subjects were categorized as either low or high LY354740-sensitive. Pharmacokinetic analysis confirmed that LY354740 was circulating at the time of cognitive testing, and at levels proportional to the amount of drug injected.

For the 5-CSRT task, when subjects were tested using three SDs and 45 trials, LY354740 did not affect performance on any measure. When the attentional load of the task was increased by using SD of 0.1 and 0.5 s and increasing the number of trials to 60, LY354740 impaired accuracy, specifically at the shortest SD in the second block of trials. This suggests that LY354740 impairs sustained attention, with the effect becoming evident when a short SD is used. This deficit in sustained attention was obtained with three animals treated with 10 mg/kg and one with 3 mg/kg of LY354740; the drug effect was similarly severe in all subjects, suggesting that an appropriate dose was selected for each animal. Another potential mediating process other than impaired attention would be reduced motivation, but three lines of evidence argue against this. Firstly, animals performed all trials, and in a previous study (Spinelli et al. 2004), we demonstrated that reduced motivation is mainly manifested as a reduction in the number of trials performed. Second, response omissions, a measure considered to be a motivation index in the rodent version of the 5-CSRT task (Robbins 2002), were not increased. Finally, lever release latency was decreased by LY354740, whereas an increase would be expected in the case of reduced motivation.

In the CDMP task, in the high-sensitivity group LY354740 impaired performance dose-dependently, suggesting that this subgroup comprised marmosets with a similar sensitivity to this mGluR2/3 agonist. In contrast, in the low-sensitivity group impairments were individual-specific. Because of the limited number of subjects in each group, the most important findings were obtained with the overall individual-specific dose analysis, which indicated that LY354740 reduced CDMP performance. At the first choice stage when the sample stimulus was #2, subjects tended to show a LY354740-induced impairment. At this stage, the delay between the sample stimulus and the choice presentation is short (0.5 s memory delay+response time) and the animal does not need to move to another position to respond correctly. LY354740 impaired performance at the first choice stage when the sample stimulus was #1: the memory load of this stage of the task is also low (0.1 s sample delay+response time to stimulus 2+0.5 s memory delay+response time), but there is a high potential for within-trial retroactive interference due to presentation and responding to sample stimulus #2 being interpolated between presentation of and the position-choice stage for sample stimulus #1. LY354740 did not affect the performance at the second stage after 8 s of reinforcement, when the interval between sample presentation and choice stage is longest and the memory load of the task is highest. The lack of effect of LY354740 at this stage of the task suggests (1) that the mGluR2/3 agonist did not specifically affect memory, and (2) that the ability to process two spatial engrams concurrently was not impaired, given that this trial type always followed a correct response at the first choice stage. Furthermore, this finding also supports the interpretation that the impairments obtained at the other stages of the task were not due to reduced motivation or a general inability of the monkeys to apply the matching rule that underlies each stage of the task. Within-trial interference is likely to act at each choice stage of the CDMP, but the lack of LY354740 effect at the second choice stage/8 s delay, even when the sample stimulus was #1, rather suggests that it does not impair the on-line holding of positional information but increases interference of retrieval, and specifically at short delays between sample stimuli and choice. An alternative explanation is that LY354740 impairs inhibitory control,

expressed as a higher tendency to switch towards a "novel" position, such that inhibition deficits are likely to be higher in the first choice stage given that the second choice stage occurs immediately after the subject has consumed reward. Indeed, a marked natural predisposition to alternate position has been reported in marmoset monkeys using a T-maze spatial delayed non-match to sample task (Easton et al. 2003), and faster learning for the non-matching rule has been reported in various species including macaques (Mishkin and Delacour 1975). Inhibition measures are also provided by the ORD task (incorrect responses to the front side of the cube) and 5-CSRT task (premature lever release), but neither of these measures was affected by LY354740. However, the large numbers of sessions performed may have reduced the sensitivity of these measures to modulation by LY354740.

The current findings for the effects of LY354740 on the CDMP task differ from similar studies in rodents. In rats, a delay-dependent deficit induced by LY354740 has been demonstrated in a T-maze task (Aultman and Moghaddam 2001) and in the operant version of the D(N)MP task (Higgins et al. 2004), strongly suggesting that LY354740 induces a memory deficit. In mice, LY354740 has been demonstrated to impair spatial navigation memory using the water maze (Higgins et al. 2004). Our results in marmoset monkeys do no support a specific effect of LY354740 in terms of mnemonic impairment in the CDMP task. This discrepancy between species could clearly be related to the differences described here in terms of the density/affinity of mGluR2/3. In vitro and in vivo studies will need to be performed in additional primate species in order to clarify the rodent–primate comparison at neurobiological and behavioural levels.

In conclusion, in this study we have been able to extend previous works on mGluR2/3 regulation of rodent cognition by describing the effects of systemic LY354740 administration on the performance of marmoset monkeys in CANTAB cognitive tasks. In the 5-CSRT task, LY354740 impaired accuracy with respect to detecting brief visual stimuli across a relatively large number of trials. In the CDMP task, LY354740 impaired performance at two stages of the task with low memory retention load but high memory interference effect. Therefore, although the marmoset monkey may not be the species of choice given the low level of specific binding for LY354740 in hippocampus and cortex identified in this study, the overall findings indicate clearly that even under conditions of low central expression, mGluR2/3 are important regulators of primate cognitive functions. Future studies in other non-human primate species, such as macaque monkeys, may help to clarify the role of these receptors and their relevance as a pharmacological target for the treatment of cognitive dysfunction, including that associated with neurodegenerative diseases.

**Acknowledgements** We are extremely grateful to Silvana Ressegatti and Jeanne Michel for marmoset care, Guy Higgins for insightful discussions, and to Giancarlo Tomio for writing the PERL computer programmes for analysis of the results output generated by the CANTAB software. We are also extremely grateful to Dr. Huguenin, Jurg Messer, Jennifer Beck and Petra Paszkiewicz for technical support in neuroanatomical studies. Simona Spinelli was in receipt of a Studentship funded by F. Hoffmann La Roche, Basel, Switzerland, and additional support was provided by the Swiss Federal Institute of Technology Zurich.

## References

Aultman JM, Moghaddam B (2001) Distinct contributions of glutamate and dopamine receptors to temporal aspects of rodent working memory using a clinically relevant task. Psychopharmacology (Berl) 153:353–364

Blumcke I, Behle K, Malitschek B, Kuhn R, Knopfel T, Wolf HK, Wiestler OD (1996) Immunohistochemical distribution of metabotropic glutamate receptor subtypes mGluR1b, mGluR2/3, mGluR4a and mGluR5 in human hippocampus. Brain Res 736:217–226

Carli M, Robbins TW, Evenden JL, Everitt BJ (1983) Effects of lesions to ascending noradrenergic neurones on performance of a 5-choice serial reaction task in rats; implications for theories of dorsal noradrenergic bundle function based on selective attention and arousal. Behav Brain Res 9:361–380

Conn PJ, Pin JP (1997) Pharmacology and functions of metabotropic glutamate receptors. Annu Rev Pharmacol Toxicol 37: 205–237

Crook JM, Akil M, Law BC, Hyde TM, Kleinman JE (2002) Comparative analysis of group II metabotropic glutamate receptor immunoreactivity in Broadmann's area 46 of the dorsolateral prefrontal cortex from patients with schizophrenia and normal subjects. Mol Psychiatry 7:157–164

Dunnett SB (1985) Comparative effects of cholinergic drugs and lesions of nucleus basalis or fimbria–fornix on delayed matching in rats. Psychopharmacology (Berl) 87:357–363

Dunnett SB (1993) Operant delayed matching and non-matching to position in rats. In: Sahgal A (ed) Behavioural Neuroscience: a practical approach, Vol. 1 (The practical approach series). IRL Press at Oxford University Press, Oxford, pp 149–163

Easton A, Parker K, Derrington AM, Parker A (2003) Behaviour of marmoset monkeys in a T-maze: comparison with rats and macaque monkeys on a spatial delayed non-match to sample task. Exp Brain Res 150:114–116

Eichenbaum H (2000) A cortical–hippocampal system for declarative memory. Nat Rev Neurosci 1:41–50

Eijkenboom M, Blokland A, van der Staay FJ (2000) Modelling cognitive dysfunctions with bilateral injections of ibotenic acid into the rat entorhinal cortex. Neuroscience 101:27–39

Gerlai RT, McNamara A, Williams S, Phillips HS (2002) Hippocampal dysfunction and behavioral deficit in the water maze in mice: an unresolved issue? Brain Res Bull 57:3–9

Hauser MD (1999) Perseveration, inhibition and the prefrontal cortex: a new look. Curr Opin Neurobiol 9:214–222

Higgins GA, Ballard TM, Kew JN, Grayson Richards J, Kemp JA, Adam G, Woltering T, Nakanishi S, Mutel V (2004) Pharmacological manipulation of mGlu2 receptors influences cognitive performance in the rodent. Neuropharmacology 46:907–917

Holden C (2003) Psychiatric drugs. Excited by glutamate. Science 300: 1866–1868

Lee HG, Ogawa O, Zhu X, O'Neill MJ, Petersen RB, Castellani RJ, Ghanbari H, Perry G, Smith MA (2004) Aberrant expression of metabotropic glutamate receptor 2 in the vulnerable neurons of Alzheimer's disease. Acta Neuropathol (Berl) 107:365–371. Epub 2004 Feb 11

Mishkin M, Delacour J (1975) An analysis of short-term visual memory in the monkey. J Exp Psychol Anim Behav Process 1:326–334

Mutel V, Adam G, Chaboz S, Kemp JA, Klingelschmidt A, Messer J, Wichmann J, Woltering T, Richards JG (1998) Characterization of (2*S*,2′*R*,3′*R*)-2-(2′,3′-[$^3$H]-dicarboxycyclopropyl)glycine binding in rat brain. J Neurochem 71:2558–2564

Neki A, Ohishi H, Kaneko T, Shigemoto R, Nakanishi S, Mizuno N (1996) Pre- and postsynaptic localization 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 (1993a) Distribution of the messenger RNA for a metabotropic glutamate receptor, mGluR2, in the central nervous system of the rat. Neuroscience 53:1009–1018

Ohishi H, Shigemoto R, Nakanishi S, Mizuno N (1993b) Distribution of the mRNA for a metabotropic glutamate receptor (mGluR3) in the rat brain: an in situ hybridization study. J Comp Neurol 335:252–266

Ohishi H, Ogawa-Meguro R, Shigemoto R, Kaneko T, Nakanishi S, Mizuno N (1994) Immunohistochemical localization of metabotropic glutamate receptors, mGluR2 and mGluR3, in rat cerebellar cortex. Neuron 13:55–66

Ohishi H, Neki A, Mizuno N (1998) Distribution of a metabotropic glutamate receptor, mGluR2, in the central nervous system of the rat and mouse: an immunohistochemical study with a monoclonal antibody. Neurosci Res 30:65–82

Pilc A (2003) LY-354740 (Eli Lilly). IDrugs 6:66–71

Robbins TW (2002) The 5-choice serial reaction time task: behavioural pharmacology and functional neurochemistry. Psychopharmacology (Berl) 163:362–380

Robbins TW, Semple J, Kumar R, Truman MI, Shorter J, Ferraro A, Fox B, McKay G, Matthews K (1997) Effects of scopolamine on delayed-matching-to-sample and paired associates tests of visual memory and learning in human subjects: comparison with diazepam and implications for dementia. Psychopharmacology (Berl) 134:95–106

Sahakian BJ, Coull JT (1993) Tetrahydroaminoacridine (THA) in Alzheimer's disease: an assessment of attentional and mnemonic function using CANTAB. Acta Neurol Scand Suppl 149:29–35

Schaffhauser H, Richards JG, Cartmell J, Chaboz S, Kemp JA, Klingelschmidt A, Messer J, Stadler H, Woltering T, Mutel V (1998) In vitro binding characteristics of a new selective group II metabotropic glutamate receptor radioligand, [$^3$H]LY354740, in rat brain. Mol Pharmacol 53:228–233

Schoepp DD, Jane DE, Monn JA (1999) Pharmacological agents acting at subtypes of metabotropic glutamate receptors. Neuropharmacology 38:1431–1476

Schoepp DD, Wright RA, Levine LR, Gaydos B, Potter WZ (2003) LY354740, an mGlu2/3 receptor agonist as a novel approach to treat anxiety/stress. Stress 6:189–197

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

Skelton RW, McNamara RK (1992) Bilateral knife cuts to the perforant path disrupt spatial learning in the Morris water maze. Hippocampus 2:73–80

Spinelli S (2004) A neuropsychopharmacological study of attention and memory in the marmoset monkey. Unpublished Ph.D. thesis, Swiss Federal Institute of Technology Zurich, Switzerland

Spinelli S, Pennanen L, Dettling AC, Feldon J, Higgins GA, Pryce CR (2004) Performance of the marmoset monkey on computerized tasks of attention and working memory. Brain Res Cogn Brain Res 19:123–137

Squire LR, Knowlton B (1995) Memory, hippocampus, and brain systems. In: Gazzaniga MS (ed) The cognitive neurosciences. MIT Press, Cambridge, MA, pp 825–837

Taffe MA, Weed MR, Gutierrez T, Davis SA, Gold LH (2002) Differential muscarinic and NMDA contributions to visuospatial paired-associate learning in rhesus monkeys. Psychopharmacology (Berl) 160:253–262

Tanabe Y, Nomura A, Masu M, Shigemoto R, Mizuno N, Nakanishi S (1993) Signal transduction, pharmacological properties, and expression patterns of two rat metabotropic glutamate receptors, mGluR3 and mGluR4. J Neurosci 13:1372–1378

Taylor JR, Elsworth JD, Roth RH, Sladek JR Jr, Redmond DE Jr (1990) Cognitive and motor deficits in the acquisition of an object retrieval/detour task in MPTP-treated monkeys. Brain 113:617–637

Weed MR, Taffe MA, Polis I, Roberts AC, Robbins TW, Koob GF, Bloom FE, Gold LH (1999) Performance norms for a rhesus monkey neuropsychological testing battery: acquisition and long-term performance. Brain Res Cogn Brain Res 8:185–201

Yokoi M, Kobayashi K, Manabe T, Takahashi T, Sakaguchi I, Katsuura G, Shigemoto R, Ohishi H, Nomura S, Nakamura K, Nakao K, Katsuki M, Nakanishi S (1996) Impairment of hippocampal mossy fiber LTD in mice lacking mGluR2. Science 273:645–647