



# Group II metabotropic glutamate receptor agonist prodrugs LY2979165 and LY2140023 attenuate the functional imaging response to ketamine in healthy subjects

Mitul A. Mehta<sup>1</sup> · Anne Schmechtig<sup>1</sup> · Vasileia Kotoula<sup>1</sup> · Juliet McColm<sup>2</sup> · Kimberley Jackson<sup>2</sup> · Claire Brittain<sup>2</sup> · Sitra Tauscher-Wisniewski<sup>3</sup> · Bruce J. Kinon<sup>4</sup> · Paul D. Morrison<sup>5</sup> · Thomas Pollak<sup>5</sup> · Timothy Mant<sup>6</sup> · Steven C. R. Williams<sup>1</sup> · Adam J. Schwarz<sup>3</sup>

Received: 7 September 2017 / Accepted: 8 March 2018 / Published online: 21 March 2018  
© Springer-Verlag GmbH Germany, part of Springer Nature 2018

## Abstract

**Background** Aberrant glutamate neurotransmission, and in particular dysfunction of the *N*-methyl-D-aspartate receptor (NMDAR), has been implicated in psychiatric disorders and represents a novel therapeutic target. Low-dose administration of the NMDA antagonist ketamine in healthy volunteers elicits a strong blood oxygenation level dependent (BOLD) imaging signal that can be attenuated by pretreatment with single, therapeutically effective doses of marketed medicines interacting with the glutamate system.

**Objective** To test the attenuation of the ketamine-induced BOLD signal by pretreatment with either a metabotropic glutamate receptor (mGluR) 2/3 or a mGluR2 agonist in healthy volunteers

**Methods** We used a ketamine challenge pharmacological magnetic resonance imaging (phMRI) paradigm to assess the modulatory effects of single acute doses of LY2140023 (pomaglumedad methionil), the methionine prodrug of the mGluR2/3 agonist LY404039 (10, 40, and 160 mg; *N* = 16 subjects) and of LY2979165, and the alanine prodrug of the selective orthosteric mGluR2 agonist 2812223 (20 and 60 mg; *N* = 16 subjects).

**Results** A reduction in the ketamine-evoked BOLD phMRI signal relative to placebo was observed at the highest doses tested of both LY2140023 and LY2979165. A relationship was observed between reduction of the BOLD signal and increasing plasma levels of 2812223 in the LY2979165 cohort.

**Conclusions** These results identify pharmacologically active doses of the group II mGluR agonist prodrugs LY2140023 and LY2979165 in humans. They also extend the classes of compounds that have been experimentally shown to reverse the ketamine-evoked phMRI signal in humans, further supporting the use of this method as a neuroimaging biomarker for assessing functional effects.

**Keywords** phMRI · fMRI · Ketamine · mGlu · Glutamate · Pharmacological

---

Juliet McColm, Sitra Tauscher-Wisniewski, and Bruce J. Kinon were employed at Eli Lilly and Company when the study was conducted.

**Electronic supplementary material** The online version of this article (<https://doi.org/10.1007/s00213-018-4877-9>) contains supplementary material, which is available to authorized users.

✉ Mitul A. Mehta  
Mitul.Mehta@kcl.ac.uk

✉ Adam J. Schwarz  
a.schwarz@lilly.com

<sup>1</sup> Department of Neuroimaging, Institute of Psychiatry, Psychology & Neuroscience, King's College London, De Crespigny Park, London SE5 8AF, UK

<sup>2</sup> Eli Lilly and Company, Sunninghill Road, Windlesham, Surrey, UK

<sup>3</sup> Eli Lilly and Company, Indianapolis, IN 46285, USA

<sup>4</sup> Lundbeck LLC, Deerfield, IL, USA

<sup>5</sup> Psychosis Studies Department, Institute of Psychiatry, Psychology & Neuroscience, King's College London, London, UK

<sup>6</sup> QuintilesIMS, Reading, UK

## Introduction

Dopamine-based therapies remain first-line medications for schizophrenia; blockade of dopamine D<sub>2</sub> receptors is thought to underlie—at least in part—the effectiveness of most current antipsychotics (Ginovart and Kapur 2012; Kapur et al. 2000). However, a significant unmet need remains, with approximately 20–40% of patients unresponsive to existing therapies (Ginovart and Kapur 2012). In addition to dopaminergic abnormalities, glutamate dysfunction has long been associated with neural impairments in schizophrenia (Egerton et al. 2012a, b; Howes et al. 2015; Merritt et al. 2016; Olney and Farber 1995).

Several lines of evidence suggest that modulation of glutamatergic activity could provide a novel therapeutic mechanism of action in the treatment of psychosis (Egerton and Stone 2012; Javitt et al. 2011; Moghaddam and Javitt 2012). First, dysfunction of the glutamatergic *N*-methyl-D-aspartate receptor (NMDAR) has been linked to schizophrenia and related syndromes (Egerton and Stone 2012). Subanaesthetic doses of the NMDAR antagonist, ketamine, induce behavior reminiscent of positive and negative symptoms (Krystal et al. 1994). This, along with a body of preclinical data linking NMDAR antagonists and aberrant glutamate neurotransmission, led to the glutamate hypothesis of schizophrenia (Moghaddam and Javitt 2012). Second, genes that encode proteins involved in glutamate neurotransmission are associated with schizophrenia (Schizophrenia Working Group of the Psychiatric Genetics Consortium 2014). Third, imaging studies have revealed glutamatergic abnormalities associated with psychosis (Egerton and Stone 2012), which may have implications for treatment decisions. These studies suggest that schizophrenic patients who do not respond to first-line dopamine-based antipsychotics have normal striatal dopamine function (Demjaha et al. 2012), but may have elevated glutamate levels in the anterior cingulate cortex (Egerton et al. 2012a, b), whereas patients who do respond show the reverse (Egerton et al. 2014). These studies suggest that glutamatergic dysfunction is present in schizophrenia and may relate to poor treatment response.

Metabotropic glutamate receptors (mGluRs) are plasma membrane-associated G-protein-coupled proteins functioning via presynaptic, postsynaptic, and glial mechanisms to modulate neuronal excitability (Monn et al. 2015; Sanacora et al. 2008). Activation of group II (mGluR2, mGluR3) subtypes by glutamate or exogenous agonists leads to the inhibition of synaptic glutamate release, dampening downstream postsynaptic excitation (Javitt et al. 2011; Sanacora et al. 2008). Novel treatments for psychiatric disorders based upon glutamatergic mechanisms of action are currently of interest (Dunayevich et al. 2008; Hashimoto 2014; Howes et al. 2015; Kendell et al. 2005; Maksymetz et al. 2017). However, recent late-stage failures of several such candidate therapeutics (Iwata

et al. 2015; Zink and Correll 2015) underscore how much remains to be understood about how such compounds interact with the human brain (Chaves et al. 2009). Both glutamatergic abnormalities associated with schizophrenia (Poels et al. 2014) and treatment effects of glutamatergic compounds in patients (Chaves et al. 2015) have been detected using imaging markers. Experimental medicine imaging studies could also play an important role to better understand these mechanisms.

NMDAR antagonists are widely used as pharmacological models of glutamate dysfunction relevant for schizophrenia (Large 2007; Northoff et al. 2005). This is in part due to their ability to induce positive, negative, and cognitive schizophrenia-like symptoms in healthy volunteers and exacerbate symptoms in patients (Lahti et al. 1995; Malhotra et al. 1997). Additionally, in healthy volunteers, glutamate levels in the prefrontal cortex correlate with positive symptom scores during acute ketamine infusion (J. M. Stone et al. 2012). Acute administration of NMDAR antagonists also elicits a robust functional and pharmacologically reversible imaging signal, in both animals (Duncan et al. 1999; Gozzi et al. 2008) and humans (Deakin et al. 2008), suggesting that this paradigm could serve as a translational imaging biomarker (Bifone and Gozzi 2012; Javitt et al. 2017). We recently established a ketamine challenge pharmacological magnetic resonance imaging (phMRI) model in healthy humans and showed that a reliable pattern of ketamine-evoked blood oxygenation level dependent (BOLD) signal change could be reversed by single doses of lamotrigine or risperidone (De Simoni et al. 2013; Doyle et al. 2013). The imaging signal was more sensitive to these modulatory effects than concomitantly obtained subjective scales. However, ketamine-evoked functional imaging changes in specific brain regions have been shown to correlate with self-rated perceptual distortions, delusional thoughts, anhedonia and cognitive disorganization, linking NMDAR antagonist infusion, localized brain activity, and schizophrenia-like symptoms (Anticevic et al. 2012; Pollak et al. 2015; J. Stone et al. 2015).

There is convergent evidence from behavioral, neurochemical, and imaging studies that the effects of NMDAR antagonists in both rodents and humans can also be reversed by pretreatment with group II mGluR agonists (Gozzi et al. 2008; Krystal et al. 2005; Lorrain et al. 2003; Moghaddam 1998). LY2140023 (pomaglumetad methionil) is the methionine prodrug of the mGluR2/3 agonist LY404039 and has been tested in clinical trials for schizophrenia (Adams et al. 2013; Adams et al. 2014; Kinon and Gomez 2013; Patil et al. 2007; Stauffer et al. 2013). The initial positive trial in schizophrenia (Patil et al. 2007) was not replicated, although a post hoc analysis of the data suggested a positive response in patients within the first 3 years of onset or those previously treated only with drugs with predominantly dopamine D<sub>2</sub> receptor antagonist activity (Kinon et al. 2015). LY2979165 is

the alanine prodrug of 2812223, a selective orthosteric mGluR2 agonist (McColm et al. 2017; Monn et al. 2015). Both compounds have been developed with the aim of normalizing aberrant glutamatergic activity in the human brain, and both dose-dependently reversed ketamine-evoked increases in glucose metabolism in the rat brain as assayed by 2-deoxyglucose (2-DG) autoradiography. LY404039 was also shown to attenuate both ketamine- and memantine-induced 2-DG uptake in the mouse brain (Dedeurwaerdere et al. 2011).

In the present study, we applied the ketamine challenge pHMRI paradigm to assess functional central nervous system effects of single, acute doses of LY2140023 and LY2979165 in healthy volunteers. LY2140023 or LY2979165 was administered to two separate cohorts of healthy volunteers prior to ketamine administration during MRI scanning. We hypothesized that both compounds would attenuate the ketamine-evoked BOLD signal in a dose- and exposure-dependent fashion.

## Materials and methods

### Study design

The present study, I4S-EW-HHCC (<http://clinicaltrials.gov/NCT01524237>), comprised two parts and was approved by the Plymouth Ethics Committee (11/IE/0166) and the Medicines and Healthcare Products Regulatory Agency (EUDRACT number 2011-000058-50). An evaluation of the safety and tolerability of each compound in combination with the ketamine infusion was undertaken (see [Supplementary Material](#)) before proceeding to the imaging part of the protocol. After demonstrating safety and tolerability in the first part, the imaging cohorts were tested and are reported in this paper. No subject participated in both the safety and imaging cohorts. Inclusion criteria specified healthy, right-handed, Caucasian males, aged 21–50 years, and a body mass index 18–30 kg/m<sup>2</sup>. Subjects were recruited via a third party clinical research organization, Quintiles Drug Research Unit at Guy's Hospital, which also provided safety oversight and clinical monitoring. Before each study visit, participants were confined to the unit where suitability for study entry was confirmed. An alcohol test and a drug abuse urine test were performed at screening and on each admission (see [Supplementary Material](#) for details on the latter). A target sample size of  $N = 16$  completers for each cohort was chosen based on a power analysis using the results of the previous study (Doyle et al. 2013) (see [Supplementary Material](#)).

LY2140023 was tested at 10, 40, and 160 mg in comparison to placebo. The 40-mg dose demonstrated (at twice-daily dosing) efficacy in an initial phase 2 schizophrenia study (Patil et al. 2007). The 10-mg dose was also tested in Phase III schizophrenia studies (Stauffer et al. 2013), whereas 160 mg

was well tolerated in clinical pharmacology studies (Zhang et al. 2015) and equivalent to the highest total daily dose evaluated in phase 2 trials (80 mg, twice daily) (Kinon and Gomez 2013).

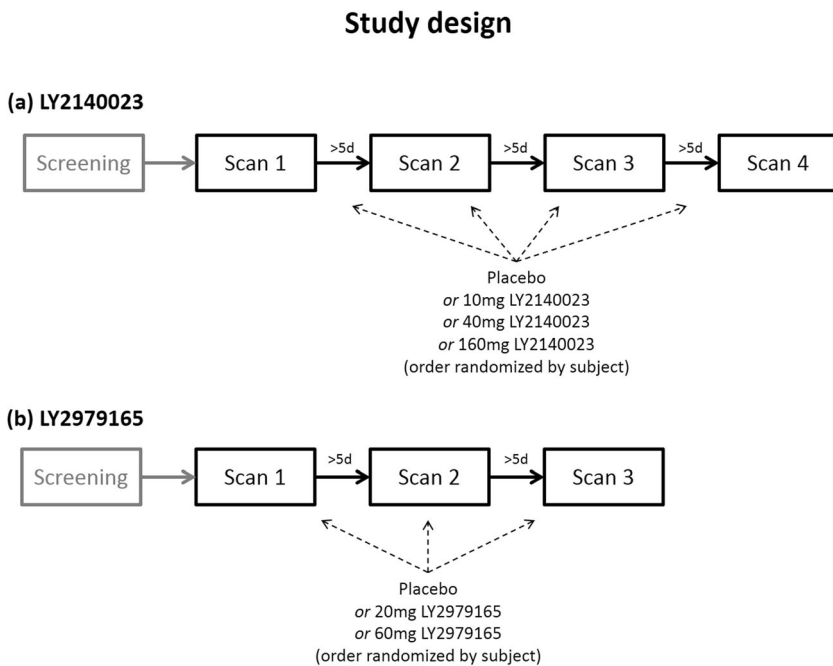
LY2979165 was tested at 20 and 60 mg in comparison to placebo. The 60-mg dose was considered the upper tolerable single dose based on emesis in the single ascending dose phase I study (McColm et al. 2017). The mean plasma exposure of the active moiety 2812223 in that study (area under the curve 11,200 nM·hr fed) (McColm et al. 2017) was within targeted efficacious range predictions from preclinical data, and the mean maximal cerebrospinal fluid exposure exceeded the EC<sub>50</sub> values of an in vitro mGluR2 assay approximately 4-fold. At 20 mg, plasma exposure was expected to be close to or below the lower boundary of this targeted exposure range.

Both cohorts were assessed in a randomized, placebo-controlled, investigator- and subject-blind crossover design in 16 healthy male volunteers. LY2140023 was tested within a four-period cross-over design, and LY2979165 was tested within a three-period cross-over design (Fig. 1). Replacement subjects could be recruited to ensure 16 completers for each cohort. There was a washout period of at least 5 days between scanning visits. The primary objective was to evaluate the effect of single doses of LY2140023 and LY2979165 in attenuating the ketamine-evoked BOLD pHMRI signal in healthy male volunteers. Secondary objectives included an evaluation of the relationship between the exposure of the active moieties and the ketamine-evoked BOLD signal. Exploratory objectives included an evaluation of effects on subjective rating scales (see below).

### Study procedures

Study procedures are illustrated in Fig. 2. Subjects were admitted to the Quintiles Drug Research Unit the day before each study day and fasted overnight for at least 8 h. On the morning of the scanning day, subjects were assessed and transferred to the scanning facility where dosing and scanning took place. Subjects were administered a single oral dose of LY2140023, LY2979165, or placebo with approximately 200 mL of water, followed by a light meal. The ketamine infusion was started near the expected time of maximum concentration observed for each compound, which after taking into account both plasma and cerebrospinal fluid data (see [Supplementary Material](#)) was 4.0 h after dosing for LY2140023 and 6.5 h after dosing for LY2979165. Ketamine infusions were staggered to accommodate infusion and scanning schedules and to enable one subject from each cohort to undergo MRI scanning on the same day. MRI scanning commenced approximately 30 min before the ketamine infusion. The duration of the ketamine infusion ranged from 38 to 53 min across both cohorts.

**Fig. 1** Schematic illustration of the study design for the LY2140023 (a) and LY2979165 (b) cohorts



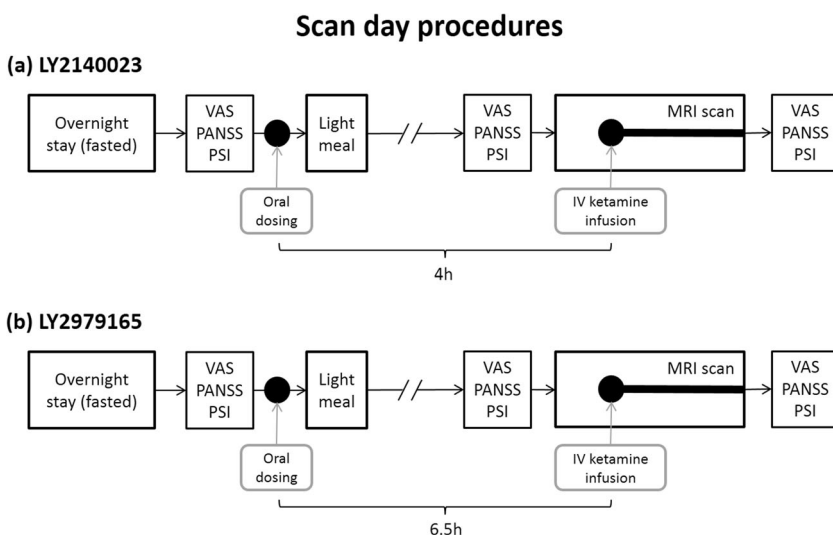
Racemic ketamine (Ketalar; Pfizer, New York, NY) was infused via the cubital vein with a temporal concentration profile calculated from the Clements 250 model (Absalom et al. 2007) and a target plasma level of 75 ng/mL, as used previously (De Simoni et al. 2013; Doyle et al. 2013). The infusion model was implemented in Stanpump ([www.opentci.org](http://www.opentci.org)) using a Graseby 3400 pump (Smiths Medical, Kent, UK) and took into account the subject’s height and weight. The resulting ketamine dose (mean ± SD) delivered was  $0.123 \pm 0.003$  mg/kg during the first minute followed by a pseudo-continuous infusion of an average of  $0.317 \pm 0.034$  mg/kg/h for LY2140023 and  $0.125 \pm 0.021$  mg/kg during the first minute followed by a pseudo-continuous infusion of an average of  $0.331 \pm 0.022$  mg/kg/h for LY2979165. Infusion rates did not

differ between drug conditions as would be expected in this crossover study design.

Imaging data were acquired using a 3T General Electric Signa HDx MRI system (Milwaukee, WI). A 15-min resting state, eyes open, BOLD phMRI time series was acquired, with the ketamine infusion initiated after the first 5 min had elapsed, as performed previously (De Simoni et al. 2013; Doyle et al. 2013). This scan used a gradient-echo echo-planar imaging sequence with 38 slices (3 mm thickness plus 0.3 mm interslice gap), repetition time (TR)=2 s, echo time (TE)=30 ms, flip angle = 75°, in-plane resolution = 3.3 mm, and matrix = 64 × 64.

Blood samples were taken at specified intervals following the oral drug administration in order to determine the plasma

**Fig. 2** Schematic illustration of the scan day procedures for the LY2140023 (a) and LY2979165 (b) cohorts. IV intravenous, MRI magnetic resonance imaging, PANSS cognitive and positive subscales, PSI psychometric states inventory, VAS visual analog scale



pharmacokinetic (PK) of LY2140023 or LY2979165 and the active moieties. Samples were also taken at approximately 14 and 42 min following commencement of the ketamine infusion to determine plasma concentrations of ketamine and nor-ketamine. The Bond-Lader Visual Analog Scale (VAS) (Bond and Lader 1974), the Positive and Negative Syndrome Scale (PANSS), and the Psychometric States Inventory (PSI) (Mason et al. 2008) were each administered prior to oral dosing and immediately before and approximately 2 h after ketamine infusion (following completion of the scanning session). The factor summary scores for each of these scales, to which we have previously demonstrated sensitivity to the effects of ketamine (De Simoni et al. 2013; J. M. Stone et al. 2012), were analyzed (see Supplementary Table 5).

## Data analysis

The BOLD pHMRI time series were processed using Statistical Parametric Mapping (SPM) software, version 8 (SPM8; Wellcome Department of Cognitive Neurology, London, UK; <http://www.fil.ion.ucl.ac.uk/spm/>) using identical operations and parameters as used in the previous study (Doyle et al. 2013). Briefly, standard spatial preprocessing steps were applied, comprising slice-timing correction, motion correction, nonlinear spatial normalization to the standard Montreal Neurological Institute (MNI) space template and spatial smoothing using an isotropic 8-mm full-width at half-maximum Gaussian kernel (De Simoni et al. 2013; Doyle et al. 2013). The amplitude of the BOLD response to the onset of ketamine infusion was estimated on a voxelwise basis using a general linear model with a design matrix comprising a gamma variate signal model of the predicted drug effect along with additional regressors to capture linear drift and head motion confounds (De Simoni et al. 2013).

Primary statistical analyses were performed on the average amplitude of the BOLD signal from voxels within each predefined region of interest (ROI). The set of ROIs was the same as that used in the previous study (Doyle et al. 2013) and was defined as binary masks based on regions in the Harvard-Oxford brain atlas, thresholded at 25% (De Simoni et al. 2013) (Table 1). Each cohort was analyzed separately, using a predefined mixed-effect model over all ROIs with fixed terms of ROI, treatment, and period. To allow covariance among the regions, ROIs were fitted as a repeated effect within subject and period. Subject was fitted as a random effect, and no imputation of missing data was performed. The posterior probability that the BOLD response in the active treatment conditions was different from placebo was calculated as the appropriate tail-area in the *t*-distribution, under the assumption of vague priors, with the threshold for significance at a given dose level versus placebo predefined as a posterior probability of 90%. Approximate effect sizes for active treatment versus placebo conditions were estimated based on least-squares

**Table 1** Predefined regions of interest (ROIs) based on atlas structures

ROI abbreviation	Description
ACC	Anterior cingulate cortex
PCC	Posterior cingulate cortex
SupGPaCC <sup>a</sup>	Supragenual paracingulate gyrus
SMA	Supplementary motor area
aIns-L <sup>b</sup>	Anterior insula (L)
aIns-R <sup>b</sup>	Anterior insula (R)
frOper-L	Frontal operculum (L)
frOper-R	Frontal operculum (R)
preCun	Precuneus
Thal	Thalamus
medOcc	Medial occipital lobes

<sup>a</sup> Atlas paracingulate cortex region was truncated to  $Z > 0$  mm (MNI coordinates) to retain only the supragenual portion and avoid the trend to negative BOLD responses in the ventral portion approaching the subcallosal cortex (De Simoni et al. 2013)

<sup>b</sup> Atlas insula cortex regions were split at  $Y > 0$  mm (MNI coordinates) to retain only the anterior portion, where a focal response was observed in previous studies (De Simoni et al. 2013; Doyle et al. 2013)

mean and 95% confidence intervals, yielding observed standard deviations of 1.86 (LY2140023 cohort) and 1.34 (LY2979165 cohort). Planned secondary analyses were also performed using similar mixed-effect models for individual ROIs. In addition, an ROI centered on the subgenual cingulate, previously shown to exhibit a negative BOLD response to ketamine infusion (De Simoni et al. 2013; Deakin et al. 2008), was examined. This ROI was defined as a sphere of 10 mm diameter, centered on MNI coordinate (2, 4, 31 mm). Modeling of pharmacokinetic/pharmacodynamic (PK/PD) relationships and analysis of subjective rating scales was performed as described in the [Supplementary Material](#). Subjects with evaluable PK data (concentrations that were above the limit of quantification of the assay) were included in the PK analyses. Results are summarized in the [Supplementary Material](#).

Additional exploratory analyses of the imaging data were performed on a voxelwise level within the SPM framework. First-level model contrasts of the pHMRI regressor were entered into second-level ANOVA models including factors of subject and treatment condition. Only subjects completing all scheduled scanning sessions were used for these SPM analyses.

## Results

### Subject sample

Participants in the LY2140023 cohort were  $28.3 \pm 6.4$  years of age (range 22–44) with weight  $74.7 \pm 7.2$  kg (range 63.4–

87.6) and BMI  $23.5 \pm 2.4$  kg (range 19.4–28.6). Participants in the LY2979165 cohort were  $29.0 \pm 7.4$  years of age (range 21–49) with weight  $75.9 \pm 10.1$  kg (range 56.6–94.4) and BMI  $23.6 \pm 2.5$  kg (range 19.6–29.1). Of the 38 subjects who entered the study and were randomized, all received at least one dose of study drug (LY2979165, LY2140023, or placebo), and 32 (16 in each cohort) completed the study. Four subjects were discontinued due to adverse events that occurred during period 1, and two subjects were withdrawn due to a positive drug abuse test at admission to period 2 (see [Supplementary Material](#)). All subjects were included in the safety and PK analyses.

### Ketamine and nor-ketamine plasma concentrations

Plasma levels of both ketamine and nor-ketamine were similar across treatment groups in both cohorts (see [Supplementary Material](#)).

### Imaging results for the LY2140023 cohort

The least-squares mean results of the BOLD phMRI response across all ROIs for the four treatment periods, corresponding to the primary analysis model, are shown in Fig. 3a and Table 2, along with the 95% confidence intervals. The main finding was a significant mean decrease of 54% in the signal at the 160-mg dose of LY2140023 compared to placebo, while the 10- and 40-mg doses showed no separation from placebo. The corresponding effect size at the 160-mg dose was  $-0.45$ . One subject was identified as a statistical outlier at the 160-mg dose; however, when either the 160-mg data point or all data from that subject were removed, the statistical separation from placebo at the 160-mg dose level was maintained. Secondary analyses for the individual ROIs revealed signal decreases consistent with those found in the mixed-effects model, with numerical decreases in BOLD signal at the 160-mg dose compared to placebo in all regions (see [Supplementary Material](#)). These differences were nominally significant for all regions except the supplementary motor area, right anterior insula, and right frontal operculum and were consistent with a generalized reduction in the ketamine-evoked BOLD signal at the 160-mg dose of LY2140023. No significant modulation of the ketamine-evoked response in the subgenual cingulate ROI was observed.

The PK profile of the active moiety, LY404039, was as expected and consistent with previous studies (see [Supplementary Material](#)). No significant relationship between reduction in the ketamine-evoked BOLD response and plasma levels of LY404039 was found (Fig. 3b). Contrast maps illustrating the anatomical distribution of the main effect of ketamine and the comparison between the 160-mg dose and placebo on the ketamine response are shown in Figs. 3c and 4d, respectively.

### Imaging results for the LY2979165 cohort

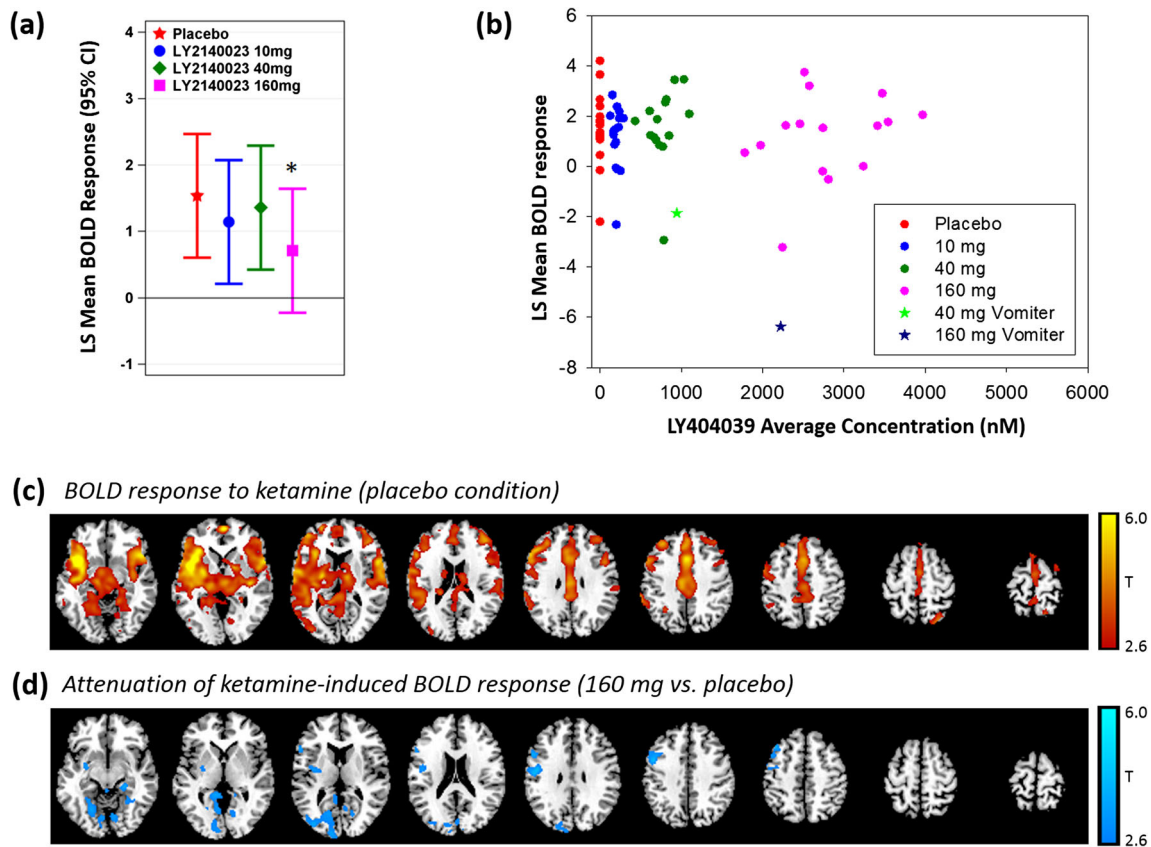
The least-squares mean results of the BOLD phMRI response across all ROIs for the three treatment periods, corresponding to the primary analysis model, are shown in Fig. 4a and Table 3, along with the 95% confidence intervals. The main finding was a significant mean decrease of 45% in the ketamine-evoked signal at the 60-mg dose of LY2979165 compared to placebo, while the 20-mg dose showed an intermediate level of effect but no statistical separation from placebo. The corresponding effect size at the 60-mg dose was  $-0.50$ . Secondary analyses for the individual ROIs revealed dose-dependent signal decreases consistent with those found in the mixed-effects model, with numerical decreases in BOLD signal at the 60-mg dose compared to placebo in all regions (see [Supplementary Material](#)). These differences were nominally significant for all regions except the anterior cingulate cortex, right frontal operculum, and precuneus and medial occipital lobes and were consistent with a generalized reduction in the ketamine-evoked BOLD signal at the 60-mg dose of LY2979165. No significant modulation of the ketamine-evoked response in the subgenual cingulate ROI was observed.

The PK profile of the active moiety 2812223 was as expected and consistent with previous studies (see [Supplementary Material](#)). A significant relationship was observed between the reduction in ketamine-evoked BOLD response and the increasing plasma levels of LY2812223 at the time of scanning (Fig. 4b). Contrast maps illustrating the anatomical distribution of the main effect of ketamine and the comparison between the 60-mg dose and placebo on the ketamine response are shown in Fig. 4c, d, respectively.

### Subjective rating scales

There was a clear main effect of ketamine on subjective ratings, defined as the response at the first postinfusion time point (Supplementary Table 5). In both cohorts, ketamine elicited increases in drowsiness (VAS); total 30, cognitive and positive subscales (PANSS); and the total score, delusory thinking, perceptual distortion, and cognitive disorganization items of the PSI. Additionally, for the LY2140023 cohort, ketamine elicited increases in the negative and general psychopathology subscale (PANSS) and mania and paranoia (PSI), and in the LY2979165 cohort, ketamine elicited increases in the excited subscale (PANSS).

Both mGluR2(3) compounds attenuated the ketamine-induced increases in delusory thinking (PSI). For LY2140023, it was significant at the 10- and 40-mg doses ( $P = 0.014$  and  $P = 0.037$ , respectively); for LY2979165, this was monotonically dose-dependent and significant at both 20- and 60-mg doses ( $P = 0.003$  and  $P \leq 0.001$ , respectively). Additionally, pretreatment with LY2979165 dose-



**Fig. 3** Results for the LY2140023 (pomaglumetad methionil) cohort. **a** Dose-response analysis: least-squares (LS) mean and 95% confidence intervals (CIs) for the blood oxygenation level dependent (BOLD) response to ketamine challenge for each experimental condition (oral placebo, 10 mg LY2140023, 40 mg LY2140023, or 160 mg LY2140023 pretreatment) and the mixed model across all atlas regions of interest (ROIs). **b** Exposure-response analysis: relationship between BOLD response to ketamine (mean across all atlas ROIs) and measured plasma levels of the active moiety LY404039. The pharmacokinetic

endpoint was the average of the two samples immediately before and after the scanning session. For the purposes of plotting the data, the placebo response data have been assigned a value of 0 nM for  $C_p$ , avg. A significant linear relationship was not found. **c** Voxelwise group map of the BOLD response to ketamine ( $P < 0.01$ ,  $P_{cluster} < 0.05$ ; color scale  $2.6 < T < 6$ ). **d** Voxelwise group map for the contrast of placebo versus 160 mg condition ( $P < 0.01$ ,  $P_{cluster} < 0.05$ ; color scale  $2.6 < T < 6$ ). These maps are shown to illustrate the anatomical pattern of effects and were not used for inference

independently augmented the ketamine-induced increases in the cognitive and positive subscales (PANSS), statistically significant ( $P = 0.008$  and  $P = 0.005$ , respectively) at the 60-mg dose compared to placebo). There were no significant correlations between the overall change in pHMRI attenuation of the ketamine-evoked BOLD response and the change in subjective ratings.

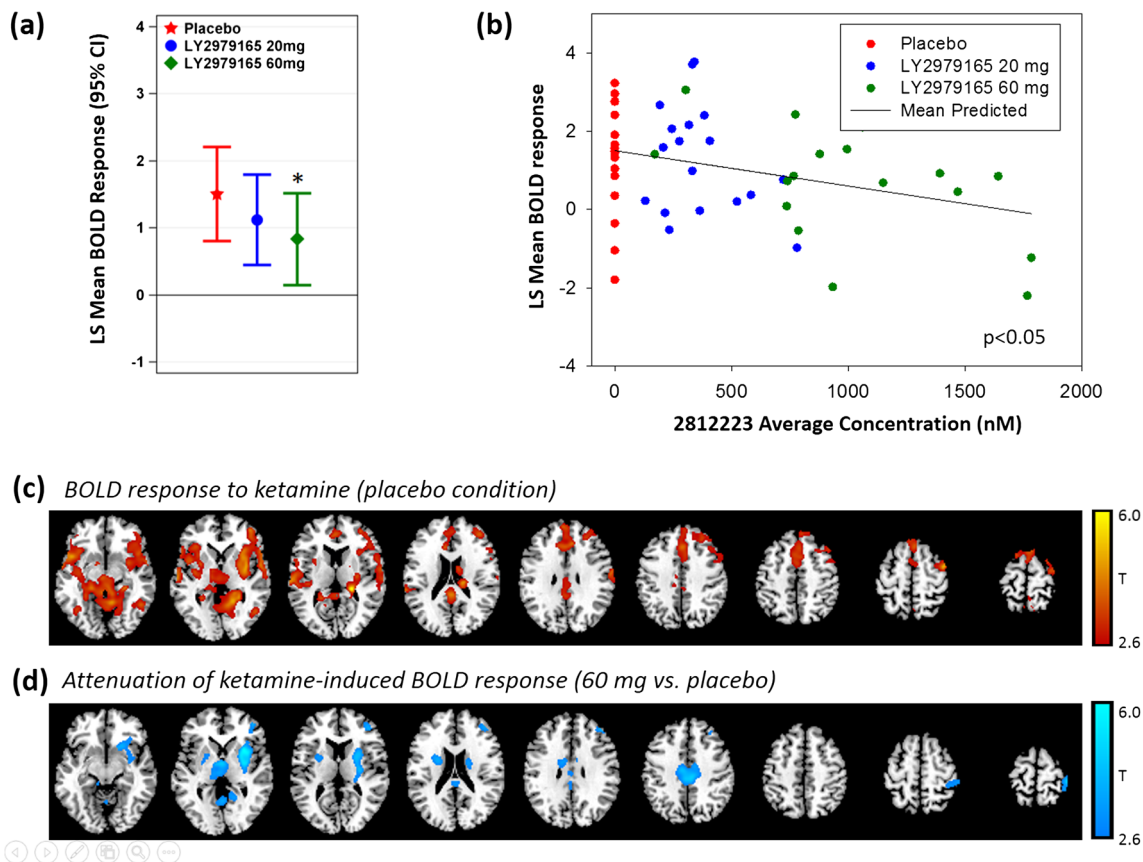
### Discussion

Single doses of two different group II mGluR agonist prodrugs, LY2140023 and LY2979165, attenuated the ketamine-evoked BOLD pHMRI signal in healthy volunteers at the highest doses tested (160 and 60 mg, respectively). Moreover, a significant relationship between reduction of the

**Table 2** Primary statistical analysis of BOLD response to ketamine infusion across predefined atlas regions of interest (ROIs) for the LY2140023 (pomaglumetad methionil) cohort

Treatment	Number	LS mean (95% CI)	Difference from placebo (95% CI)	Posterior probability
Placebo	16	1.54 (0.60, 2.47)		
10 mg LY2140023	16	1.14 (0.21, 2.07)	-0.39 (-1.14, 0.35)	0.855
40 mg LY2140023	16	1.36 (0.43, 2.29)	-0.18 (-0.92, 0.57)	0.683
160 mg LY2140023	16	0.71 (-0.22, 1.64)	-0.83 (-1.57, -0.08)	0.985

CI confidence interval, LS least squares, N sample size



**Fig. 4** Results for the LY2979165 cohort. **a** Dose-response analysis: least-squares (LS) mean and 95% confidence intervals (CIs) for the BOLD response to ketamine challenge for each experimental condition (oral placebo, 20 mg LY2979165, or 60 mg LY2979165 pretreatment) and the mixed model across all atlas regions of interest (ROIs). **b** Exposure-response analysis: relationship between BOLD response to ketamine (mean across all atlas ROIs) and measured plasma levels of the active moiety 2812223. The pharmacokinetic endpoint was the average of the two samples immediately before and after the scanning

session. For the purposes of plotting the data, the placebo response data have been assigned a value of 0 nM for Cp, avg. The straight line indicates linear regression (slope =  $-0.000905$  nM, 95% confidence interval =  $[-0.0015, -0.000299]$ ). **c** Voxelwise group map of the BOLD response to ketamine ( $P < 0.01$ ,  $P_c < 0.05$ ; color scale  $2.6 < T < 6$ ). **d** Voxelwise group map for the contrast of placebo versus 60 mg conditions ( $P < 0.01$ ,  $P_c < 0.05$ ; color scale  $2.6 < T < 6$ ). These maps are shown to illustrate the anatomical pattern of effects and were not used for inference

BOLD signal and increasing plasma levels of 2812223 (the active moiety) was observed in the LY2979165 cohort. Therefore, the hypothesis that group II mGluR agonists would attenuate the BOLD response following ketamine was confirmed using two different compounds in two cohorts of healthy volunteers.

These findings are consistent with a body of preclinical experimental data. Imaging studies in rodents have shown that the earlier generation mGluR2/3 agonist, LY354740, blocks the pHMRI response to the NMDAR antagonist phencyclidine

in the rat brain (Gozzi et al. 2008), and LY404039 blocks the 2-DG signal evoked by the NMDAR antagonists ketamine and memantine in the mouse (Dedeurwaerdere et al. 2011). One of the primary effects of ketamine, thought to arise via disinhibition of gamma-aminobutyric acid (GABA)-ergic inhibitory neurons downstream from blockade of NMDARs, is an increase in glutamate release. This has been demonstrated in the frontal cortex of rats following phencyclidine (Moghaddam 1998) and ketamine (Lorrain et al. 2003) and in humans using magnetic resonance spectroscopy (Stone

**Table 3** Primary statistical analysis of BOLD response to ketamine infusion across predefined atlas regions of interest (ROIs) for the LY2979165 cohort

Treatment	Number	LS Mean (95% CI)	Difference from placebo (95% CI)	Posterior probability
Placebo	16	1.50 (0.80, 2.21)		
20 mg LY2979165	18	1.12 (0.44, 1.79)	-0.38 (-1.10, 0.33)	0.862
60 mg LY2979165	17	0.83 (0.15, 1.52)	-0.67 (-1.39, 0.05)	0.967

CI confidence interval, LS least squares, N sample size



et al. 2012). Moreover, the mGluR2/3 agonist LY354740 was shown to block glutamate, but not dopamine, release elicited by phencyclidine in the rat (Moghaddam 1998). Taken together, these findings suggest that the blockade of the ketamine-induced BOLD signal observed in the present study is related to the compounds' effects in attenuating glutamate efflux and is consistent with their expected mechanism of action and underscores the translational nature of this approach.

For LY2140023, the signal attenuation at 10 mg was numerically greater than that at 40 mg, although the mean responses at both doses showed less attenuation on average than at the 160-mg dose, and neither the 10-mg nor 40-mg doses were significantly different from placebo in the ROI analysis. For LY2979165, the dose-response was monotonic, with the mean ketamine-induced BOLD signal at the 20-mg dose numerically intermediate between that observed for the placebo and 60-mg conditions. This, coupled with the significant association observed between the plasma levels of the active moiety 2812223, is consistent with monotonic dose- and exposure-response relationships over the 0- to 60-mg dose range. Modulatory effects on individual ROIs were, overall, consistent with a global reduction in the ketamine-induced BOLD signal change. Voxel-based analyses—not used for statistical inference but to visualize the anatomical distribution of the pharmacological effects—support the a priori ROI analysis, with regions of attenuation generally corresponding to regions that responded most strongly to the ketamine challenge. Apparent region-specific effects of both compounds should be interpreted with caution, as they may be due to variability in the data in this small experimental medicine study. While both active metabolites are classed as group II mGluR agonists, LY404039 (the active metabolite of LY2140023) has comparable affinity at both mGluR2 and mGluR3 receptors, whereas 2812223 (the active metabolite of LY2979169) preferentially targets the mGluR2 receptor. The precise role of each of these receptors and their relative utility as drug targets for psychiatric disorders remains incompletely understood. Indeed, the recent availability of mGluR2-selective ligands will enable this role to be better understood. In the present study, the largely overlapping effects of each compound on reversing the ketamine-evoked response, and the parallel cohort study design, preclude identification of any impact of the additional mGluR3 activity of LY2140023 in this model.

The doses at which a significant reversal of the ketamine signal was observed in the present study reduced the mean ketamine response by 54% (160 mg LY2140023) and 45% (60 mg LY2979165). This represents a similar relative degree of attenuation as previously observed with clinically relevant doses of risperidone (57%) and lamotrigine (43%) (Doyle et al. 2013). Those results suggest that this degree of effect in this imaging model may indicate a clinically meaningful

dose, although that hypothesis remains to be confirmed across a wider range of compounds and mechanisms. Although the ketamine model is often considered in the context of schizophrenia, this imaging signal is likely not specific to that clinical indication given that lamotrigine also attenuates the ketamine response in this model (Deakin et al. 2008; Doyle et al. 2013) and is indicated for bipolar disorder and epilepsy, but has not robustly shown benefit in schizophrenia trials (Reid et al. 2013). Rather, it reflects a functionally effective action of the compound in modulating glutamate-related brain activity (Doyle et al. 2013). In this context, and given the negative clinical findings with pomaglumetad methionil (LY2140023) at the 40-mg dose, the present imaging results suggest that a higher dose of this compound might be required to achieve more robust clinical efficacy. On the other hand, the nonmonotonic dose-response relationship suggests that the relationship between the ketamine imaging findings, clinical efficacy, and clinical population may be significant for the mGluR2(3) agonist mechanism. Additional imaging and clinical exploration with both compounds tested in the current study will help elucidate the relationship between the dose-dependent effects observed in the ketamine imaging model and those required for efficacy in clinical populations.

In the present study, the ketamine BOLD response in the placebo condition was on average weaker and more variable across subjects than observed in the previous study (Doyle et al. 2013). This was despite the use of identical inclusion/exclusion criteria, MRI scanner, staff, and acquisition and analysis procedures and may reflect the different recruitment mechanism used, resulting in a population more typical of phase I safety studies, in contrast to the more homogeneous study population obtained via the academic recruiting network used previously. This emphasizes the importance of the recruitment mechanism and a clean study sample when using functional imaging in biomarker studies.

The low dose of ketamine used in this study was previously established as eliciting a robust BOLD pHMRI response, but relatively mild subjective effects (De Simoni et al. 2013). Nevertheless, significant ketamine-induced increases in many items and subscales of the Bond-Lader VAS, PANSS, and PSI were recorded in both cohorts, consistent with findings from previous studies (De Simoni et al. 2013; Doyle et al. 2013). While pretreatment reversed few of these ratings overall, it is noteworthy that both compounds reversed the increase in PSI delusory thinking, suggesting that this psychotomimetic subscale might be particularly sensitive to the mGluR2(3) agonist mechanism, which potentially could be tested in patient populations using symptom-level outcomes. Since psychometric factors and delusory thinking are sensitive to the dose of ketamine (Pollak et al. 2015; Stone et al. 2015), replication with a higher dose of ketamine will provide data more amenable to test the neural basis of the mGluR2(3)-based mechanism of this effect. Surprisingly, LY2979165 additionally augmented

the ketamine-induced increases in the PANSS cognitive and positive subscales, despite having no effect per se on these subscales immediately prior to the scan and the ketamine infusion. Given the exploratory nature of the subjective rating scales in the present study, and that these findings were nominally significant but not formally corrected for multiple testing, further studies are needed to confirm and elucidate these subjective effects.

## Conclusion

Two novel mGluR2(3) agonist prodrugs were shown to attenuate the ketamine-evoked BOLD pHMRI signal and subjective effects of ketamine, relative to placebo, in healthy volunteers. Only the highest doses of each compound tested produced significant attenuation across the ROIs, and a significant relationship between reduction of the BOLD signal and increasing plasma levels of 2812223 was observed in the LY2979165 cohort. These results indicate functionally active doses of these compounds, extend the classes of compounds that have been experimentally shown to reverse the ketamine-evoked pHMRI signal in humans, and further support the use of this method as an imaging biomarker for assessing functional effects within the central nervous system.

**Acknowledgements** Authors SCRW and MAM acknowledge the ongoing support of the National Institute for Health Research (NIHR) Biomedical Research Centre at South London and Maudsley NHS Foundation Trust and King's College London. The authors acknowledge Darren Wilbraham for overseeing the clinical assessments and monitoring for this study at the Quintiles Clinical Research Unit. The views expressed are those of the authors and not necessarily those of the NIHR, NHS, or the Department of Health.

**Funding** The study was funded by Eli Lilly and Company.

## Compliance with ethical standards

This study was conducted in accordance with the Declaration of Helsinki and Council for International Organizations of Medical Sciences (CIOMS) International Ethical Guidelines. The study protocol was approved by the ethics review board (ERB). Informed consent was obtained from all participants.

**Conflict of interest** MAM has acted as a consultant for Taisho Pharmaceutical and Cambridge Cognition and has sat on the advisory board for FORUM Pharmaceuticals and Autifony (unpaid). He currently holds research grants from Johnson & Johnson. AJS, JM, KJ, and CB are all employees and stockholders of Eli Lilly and Company. ST-W was an Eli Lilly and Company employee when this study was carried out; she is currently employed by Astellas. BJK was an Eli Lilly and Company employee when this study was carried out; he is currently employed by Lundbeck LLC and a stockholder of Eli Lilly and Company. PDM is supported by the National Institute for Health Research (NIHR) Biomedical Research Centre at South London and Maudsley NHS Foundation Trust and King's College London. TP is an employee of the Psychosis Studies Dept., Institute of Psychiatry, King's College, London,

UK and receives funding in the form of a clinical research training fellowship from the Wellcome Trust (grant number 105758/Z/14/Z). TM is an employee of QuintilesIMS and is supported by the NIHR Biomedical Research Centre at Guy's and St. Thomas' Hospitals and King's College London. SCRW has received project funding from Pfizer, Takeda, GSK, Eli Lilly and Company, Johnson & Johnson, Roche, Evotec, Bionomics, GE Healthcare, and P1Vital. He has also previously acted as a consultant for GE Healthcare, GSK, Novartis, and Takeda. VK and AS declare that they have no conflict of interest.

## References

- Absalom AR, Lee M, Menon DK, Sharar SR, De Smet T, Halliday J et al (2007) Predictive performance of the Domino, Hijazi, and Clements models during low-dose target-controlled ketamine infusions in healthy volunteers. *Br J Anaesth* 98(5):615–623. <https://doi.org/10.1093/bja/aem063>
- Adams DH, Kinon BJ, Baygani S, Millen BA, Velona I, Kollack-Walker S, Walling DP (2013) A long-term, phase 2, multicenter, randomized, open-label, comparative safety study of pomaglumetad methionil (LY2140023 monohydrate) versus atypical antipsychotic standard of care in patients with schizophrenia. *BMC Psychiatry* 13(1):143. <https://doi.org/10.1186/1471-244X-13-143>
- Adams DH, Zhang L, Millen BA, Kinon BJ, Gomez JC (2014) Pomaglumetad Methionil (LY2140023 monohydrate) and aripiprazole in patients with schizophrenia: a phase 3, multicenter, double-blind comparison. *Schizophr Res Treat* 2014:758212–758211. <https://doi.org/10.1155/2014/758212>
- Anticevic A, Gancsos M, Murray JD, Repovs G, Driesen NR, Ennis DJ et al (2012) NMDA receptor function in large-scale anticorrelated neural systems with implications for cognition and schizophrenia. *Proc Natl Acad Sci U S A* 109(41):16720–16725
- Bifone A, Gozzi A (2012) Neuromapping techniques in drug discovery: pharmacological MRI for the assessment of novel antipsychotics. *Expert Opin Drug Discov* 7(11):1071–1082
- Bond A, Lader M (1974) The use of analogue scales in rating subjective feelings. *Br J Med Psychol* 47:211–218
- Chaves C, Marque CR, Trzesniak C, Machado de Sousa JP, Zuardi AW, Crippa JAS et al (2009) Glutamate-N-methyl-D-aspartate receptor modulation and minocycline for the treatment of patients with schizophrenia: an update. *Braz J Med Biol Res* 42(11):1002–1014
- Chaves C, Marque CR, Maia-de-Oliveira JP, Wichert-Ana L, Ferrari TB, Santos AC et al (2015) Effects of minocycline add-on treatment on brain morphometry and cerebral perfusion in recent-onset schizophrenia. *Schizophr Res* 161(2–3):439–445. <https://doi.org/10.1016/j.schres.2014.11.031>
- De Simoni S, Schwarz AJ, O'Daly OG, Marquand AF, Brittain C, Gonzales C et al (2013) Test-retest reliability of the BOLD pharmacological MRI response to ketamine in healthy volunteers. *NeuroImage* 64:75–90. <https://doi.org/10.1016/j.neuroimage.2012.09.037>
- Deakin JFW, Lees J, McKie S, Hallak JEC, Williams SR, Dursun SM (2008) Glutamate and the neural basis of the subjective effects of ketamine. *Arch Gen Psychiatry* 65(2):154–164
- Dedeurwaerdere S, Wintmolders C, Straetmans R, Pemberton D, Langlois X (2011) Memantine-induced brain activation as a model for the rapid screening of potential novel antipsychotic compounds: exemplified by activity of an mGlu2/3 receptor agonist. *Psychopharmacology* 214(2):505–514. <https://doi.org/10.1007/s00213-010-2052-z>
- Demjaha A, Murray RM, McGuire PK, Kapur S, Howes OD (2012) Dopamine synthesis capacity in patients with treatment-resistant

- schizophrenia. *Am J Psychiatry* 169(11):1203–1210. <https://doi.org/10.1176/appi.ajp.2012.12010144>
- Doyle OM, De Simoni S, Schwarz AJ, Brittain C, O'Daly OG, Williams SC, Mehta MA (2013) Quantifying the attenuation of the ketamine pharmacological magnetic resonance imaging response in humans: a validation using antipsychotic and glutamatergic agents. *J Pharmacol Exp Ther* 345(1):151–160. <https://doi.org/10.1124/jpet.112.201665>
- Dunayevich E, Erickson J, Levine L, Landbloom R, Schoepp DD, Tollefson GD (2008) Efficacy and tolerability of an mGlu2/3 agonist in the treatment of generalized anxiety disorder. *Neuropsychopharmacology* 33(7):1603–1610. <https://doi.org/10.1038/sj.npp.1301531>
- Duncan GE, Miyamoto S, Leipzig JN, Lieberman JA (1999) Comparison of brain metabolic activity patterns induced by ketamine, MK-801 and amphetamine in rats: support for NMDA receptor involvement in responses to subanesthetic dose of ketamine. *Brain Res* 843(1–2): 171–183
- Egerton A, Stone J (2012) The glutamate hypothesis of schizophrenia: neuroimaging and drug development. *Curr Pharm Biotechnol* 13(8): 1500–1512
- Egerton A, Brugger S, Raffin M, Barker GJ, Lythgoe DJ, McGuire PK, Stone JM (2012a) Anterior cingulate glutamate levels related to clinical status following treatment in first-episode schizophrenia. *Neuropsychopharmacology* 37(11):2515–2521. <https://doi.org/10.1038/npp.2012.113>
- Egerton A, Fusar-Poli P, Stone J (2012b) Glutamate and psychosis risk. *Curr Pharm Des* 18(4):466–478
- Egerton A, Stone JM, Chaddock CA, Barker GJ, Bonoldi I, Howard RM et al (2014) Relationship between brain glutamate levels and clinical outcome in individuals at ultra high risk of psychosis. *Neuropsychopharmacology* 39(12):2891–2899. <https://doi.org/10.1038/npp.2014.143>
- Genovart N, Kapur S (2012) Role of dopamine D(2) receptors for antipsychotic activity. *Handb Exp Pharmacol* (212):27–52. doi:[https://doi.org/10.1007/978-3-642-25761-2\\_2](https://doi.org/10.1007/978-3-642-25761-2_2)
- Gozzi A, Large CH, Schwarz A, Bertani S, Crestan V, Bifone A (2008) Differential effects of antipsychotic and glutamatergic agents on the pHMRI response to phencyclidine. *Neuropsychopharmacology* 33(7):1690–1703. <https://doi.org/10.1038/sj.npp.1301547>
- Hashimoto K (2014) Targeting of NMDA receptors in new treatments for schizophrenia. *Expert Opin Ther Targets* 18(9):1049–1063. <https://doi.org/10.1517/14728222.2014.934225>
- Howes O, McCutcheon R, Stone J (2015) Glutamate and dopamine in schizophrenia: an update for the 21st century. *J Psychopharmacol* 29(2):97–115. <https://doi.org/10.1177/0269881114563634>
- Iwata Y, Nakajima S, Suzuki T, Keefe RS, Plitman E, Chung JK et al (2015) Effects of glutamate positive modulators on cognitive deficits in schizophrenia: a systematic review and meta-analysis of double-blind randomized controlled trials. *Mol Psychiatry* 20(10): 1151–1160. <https://doi.org/10.1038/mp.2015.68>
- Javitt DC, Schoepp D, Kalivas PW, Volkow ND, Zarate C, Merchant K et al (2011) Translating glutamate: from pathophysiology to treatment. *Sci Transl Med* 3(102):102mr102
- Javitt DC, Carter CS, Krystal JH, Kantrowitz JT, Girgis RR, Kegeles LS et al (2017) Utility of imaging-based biomarkers for glutamate-targeted drug development in psychotic disorders: a randomized clinical trial. *JAMA Psychiatry*. <https://doi.org/10.1001/jamapsychiatry.2017.3572>
- Kapur S, Zipursky R, Jones C, Remington G, Houle S (2000) Relationship between dopamine D(2) occupancy, clinical response, and side effects: a double-blind PET study of first-episode schizophrenia. *Am J Psychiatry* 157(4):514–520
- Kendell SF, Krystal JH, Sanacora G (2005) GABA and glutamate systems as therapeutic targets in depression and mood disorders. *Expert Opin Ther Targets* 9(1):153–168. <https://doi.org/10.1517/14728222.9.1.153>
- Kinon BJ, Gomez JC (2013) Clinical development of pomaglumetad methionil: a non-dopaminergic treatment for schizophrenia. *Neuropharmacology* 66:82–86. <https://doi.org/10.1016/j.neuropharm.2012.06.002>
- Kinon BJ, Millen BA, Zhang L, McKinzie DL (2015) Exploratory analysis for a targeted patient population responsive to the metabotropic glutamate 2/3 receptor agonist pomaglumetad methionil in schizophrenia. *Biol Psychiatry* 78:754–762. <https://doi.org/10.1016/j.biopsych.2015.03.016>
- Krystal JH, Karper LP, Seibyl JP, Freeman GK, Delaney R, Bremner JD et al (1994) Subanesthetic effects of the noncompetitive NMDA antagonist, ketamine, in humans. Psychotomimetic, perceptual, cognitive, and neuroendocrine responses. *Arch Gen Psychiatry* 51(3): 199–214
- Krystal JH, Abi-Saab W, Perry E, D'Souza DC, Liu N, Gueorguieva R et al (2005) Preliminary evidence of attenuation of the disruptive effects of the NMDA glutamate receptor antagonist, ketamine, on working memory by pretreatment with the group II metabotropic glutamate receptor agonist, LY354740, in healthy human subjects. *Psychopharmacology* 179(1):303–309. <https://doi.org/10.1007/s00213-004-1982-8>
- Lahti AC, Koffel B, LaPorte D, Tamminga CA (1995) Subanesthetic doses of ketamine stimulate psychosis in schizophrenia. *Neuropsychopharmacology* 13:9–19
- Large CH (2007) Do NMDA receptor antagonist models of schizophrenia predict the clinical efficacy of antipsychotic drugs? *J Psychopharmacol* 21(3):283–301. <https://doi.org/10.1177/0269881107077712>
- Lorrain DS, Bacceti CS, Bristow LJ, Anderson JJ, Varney MA (2003) Effects of ketamine and n-methyl-d-aspartate on glutamate and dopamine release in the rat prefrontal cortex: modulation by a group II selective metabotropic glutamate receptor agonist LY379268. *Neuroscience* 117(3):697–706. [https://doi.org/10.1016/s0306-4522\(02\)00652-8](https://doi.org/10.1016/s0306-4522(02)00652-8)
- Maksymetz J, Moran SP, Conn PJ (2017) Targeting metabotropic glutamate receptors for novel treatments of schizophrenia. *Mol Brain* 10(1):15. <https://doi.org/10.1186/s13041-017-0293-z>
- Malhotra AK, Pinals DA, Adler CM, Elman I, Clifton A, Pickar D, Breier A (1997) Ketamine-induced exacerbation of psychotic symptoms and cognitive impairment in neuroleptic-free schizophrenics. *Neuropsychopharmacology* 17:141–150
- Mason OJ, Morgan CJ, Stefanovic A, Curran HV (2008) The psychotomimetic states inventory (PSI): measuring psychotic-type experiences from ketamine and cannabis. *Schizophr Res* 103(1–3):138–142. <https://doi.org/10.1016/j.schres.2008.02.020>
- McColm J, Brittain C, Suriyapperuma S, Swanson S, Tauscher-Wisniewski S, Foster J, Soon D, Jackson K (2017) Evaluation of single and multiple doses of a novel mGlu2 agonist, a potential antipsychotic therapy, in healthy subjects. *Br J Clin Pharmacol* 83: 1654–1667. <https://doi.org/10.1111/bcp.13252>
- Merritt K, Egerton A, Kempton MJ, Taylor MJ, McGuire PK (2016) Nature of glutamate alterations in schizophrenia: a meta-analysis of proton magnetic resonance spectroscopy studies. *JAMA Psychiatry* 73(7):665–674. <https://doi.org/10.1001/jamapsychiatry.2016.0442>
- Moghaddam B (1998) Reversal of phencyclidine effects by a group II metabotropic glutamate receptor agonist in rats. *Science* 281(5381): 1349–1352. <https://doi.org/10.1126/science.281.5381.1349>
- Moghaddam B, Javitt D (2012) From revolution to evolution: the glutamate hypothesis of schizophrenia and its implication for treatment. *Neuropsychopharmacology* 37(1):4–15. <https://doi.org/10.1038/npp.2011.181>
- Monn JA, Prieto L, Taboada L, Hao J, Reinhard MR, Henry SS et al (2015) Synthesis and pharmacological characterization of C4-

- (thiotriazolyl)-substituted-2-aminobicyclo[3.1.0]hexane-2,6-dicarboxylates. Identification of (1R,2S,4R,5R,6R)-2-amino-4-(1H-1,2,4-triazol-3-ylsulfanyl)bicyclo[3.1.0]hexane-2, 6-dicarboxylic acid (LY2812223), a highly potent, functionally selective mGlu2 receptor agonist. *J Med Chem*. <https://doi.org/10.1021/acs.jmedchem.5b01124>
- Northoff G, Richter A, Bempohl F, Grimm S, Martin E, Marcar VL, Wahl C, Hell D, Boeker H (2005) NMDA hypofunction in the posterior cingulate as a model for schizophrenia: an exploratory ketamine administration study in fMRI. *Schizophr Res* 72(2–3): 235–248. <https://doi.org/10.1016/j.schres.2004.04.009>
- Olney JW, Farber NB (1995) Glutamate receptor dysfunction and schizophrenia. *Arch Gen Psychiatry* 52:998–1007
- Patil ST, Zhang L, Martenyi F, Lowe SL, Jackson KA, Andreev BV et al (2007) Activation of mGlu2/3 receptors as a new approach to treat schizophrenia: a randomized phase 2 clinical trial. *Nat Med* 13(9): 1102–1107. <https://doi.org/10.1038/nm1632>
- Poels EM, Kegeles LS, Kantrowitz JT, Slifstein M, Javitt DC, Lieberman JA et al (2014) Imaging glutamate in schizophrenia: review of findings and implications for drug discovery. *Mol Psychiatry* 19(1):20–29. <https://doi.org/10.1038/mp.2013.136>
- Pollak TA, De Simoni S, Barimani B, Zelaya FO, Stone JM, Mehta MA (2015) Phenomenologically distinct psychotomimetic effects of ketamine are associated with cerebral blood flow changes in functionally relevant cerebral foci: a continuous arterial spin labelling study. *Psychopharmacology* 232(24):4515–4524. <https://doi.org/10.1007/s00213-015-4078-8>
- Reid JG, Gitlin MJ, Altshuler LL (2013) Lamotrigine in psychiatric disorders. *J Clin Psychiatry* 74(7):675–684. <https://doi.org/10.4088/JCP.12r08046>
- Sanacora G, Zarate CA, Krystal JH, Manji HK (2008) Targeting the glutamatergic system to develop novel, improved therapeutics for mood disorders. *Nat Rev Drug Discov* 7(5):426–437. <https://doi.org/10.1038/nrd2462>
- Schizophrenia Working Group of the Psychiatric Genetics Consortium, T (2014) Biological insights from 108 schizophrenia-associated genetic loci. *Nature* 511(7510):421–427. <https://doi.org/10.1038/nature13595>
- Stauffer VL, Millen BA, Andersen S, Kinon BJ, Lagrandeur L, Lindenmayer JP, Gomez JC (2013) Pomaglutetad methionil: no significant difference as an adjunctive treatment for patients with prominent negative symptoms of schizophrenia compared to placebo. *Schizophr Res* 150(2–3):434–441. <https://doi.org/10.1016/j.schres.2013.08.020>
- Stone JM, Dietrich C, Edden R, Mehta MA, De Simoni S, Reed LJ et al (2012) Ketamine effects on brain GABA and glutamate levels with 1H-MRS: relationship to ketamine-induced psychopathology. *Mol Psychiatry* 17(7):664–665. <https://doi.org/10.1038/mp.2011.171>
- Stone J, Kotoula V, Dietrich C, De Simoni S, Krystal JH, Mehta MA (2015) Perceptual distortions and delusional thinking following ketamine administration are related to increased pharmacological MRI signal changes in the parietal lobe. *J Psychopharmacol* 29:1025–1028. <https://doi.org/10.1177/0269881115592337>
- Zhang W, Mitchell MI, Knadler MP, Long A, Witcher J, Walling D, Annes W, Ayan-Oshodi M (2015) Effect of pomaglutetad methionil on the QT interval in subjects with schizophrenia. *Int J Clin Pharmacol Ther* 53(6):462–470
- Zink M, Correll CU (2015) Glutamatergic agents for schizophrenia: current evidence and perspectives. *Expert Rev Clin Pharmacol* 8(3): 335–352