Adjunctive sarcosine plus benzoate improved cognitive function in chronic schizophrenia patients with constant clinical symptoms: A randomised, double-blind, placebo-controlled trial

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Abstract

Objectives: Hypofunction of NMDA receptor is implicated in the pathophysiology, particularly cognitive impairment, of schizophrenia. Sarcosine, a glycine transporter I (GlyT-1) inhibitor, and sodium benzoate, a D-amino acid oxidase (DAAO) inhibitor, can both enhance NMDA receptor-mediated neurotransmission. We proposed simultaneously inhibiting DAAO and GlyT-1 may be more effective than inhibition of either in improving the cognitive and global functioning of schizophrenia patients.

Methods: This study compared add-on sarcosine (2 g/day) plus benzoate (1 g/day) vs. sarcosine (2 g/day) for the clinical symptoms, as well as the cognitive and global functioning, of chronic schizophrenia patients in a 12-week, double-blind, randomised, placebo-controlled trial. Participants were measured with the Positive and Negative Syndrome Scale and the Global Assessment of Functioning Scale every 3 weeks. Seven cognitive domains, recommended by the Measurement and Treatment Research to Improve Cognition in Schizophrenia Committee, were measured at weeks 0 and 12.

Results: Adjunctive sarcosine plus benzoate, but not sarcosine alone, improved the cognitive and global functioning of patients with schizophrenia, even when their clinical symptoms had not improved.

Conclusions: This finding suggests N-methyl-D-aspartate receptor-enhancement therapy can improve the cognitive function of patients with schizophrenia, further indicating this pro-cognitive effect can be primary without improvement in clinical symptoms.

Introduction

Schizophrenia is a severe mental disorder, influencing 1% of the global population (Schultz and Andreasen 1999). Its phenotypes include positive symptoms, negative symptoms and cognitive impairments. Cognitive dysfunction is thought to be the core manifestation of schizophrenia due to its detrimental impact throughout the lifelong illness (Kasper 2006; Carrión et al. 2011; Green et al. 2011). There is controversy regarding the effect of current antipsychotic medications on cognitive dysfunction (Sergi et al. 2007; Meltzer 2013). Evidence of the role of N-methyl-D-aspartate receptor (NMDA) receptors in the cognition and pathophysiology of schizophrenia (Krystal et al. 1994; Sawa 2009; Javitt et al. 2012; Errico et al. 2013) suggests that enhancement of the NMDA receptor function may improve the cognitive impairment of schizophrenia (Krystal et al. 1994; Javitt et al. 2012; Hashimoto et al. 2013; Paoletti et al. 2013). Previous research showed that adjuvant NMDA-enhancing agents which directly or indirectly enhance the NMDA function via the NMDA-glycine site, including D-serine (Tsai et al. 1998; Heresco-Levy et al. 2005, 2015), glycine (Javitt et al. 2001; Heresco-Levy et al. 2004), glycine transporter I (GlyT-1) inhibitor (Tsai et al. 2004), and D-amino acid oxidase (DAAO)
inhibitor (Lane et al. 2013), reveal beneficial efficacy on clinical symptoms. For example, sarcosine, increasing glycine supply to the synapse by blocking the reuptake of glycine through inhibiting GlyT-1 (McBain et al. 1989), could decrease positive and negative symptoms of some patients (Tsai et al. 2004; Lane et al. 2005). Whether sarcosine can improve cognitive function is not yet known. Roche also recently announced two Phase III studies of another GlyT-1 inhibitor, bitopertin, in adults with persistent, predominant negative symptoms, but failed to meet their primary goals (FirstWord Pharma 2011) and healthy subjects (Levin et al. 2015). However, Weiser et al. (2012) reported add-on D-serine showed no improvement in negative or cognitive symptoms of schizophrenia and the lower D-serine doses could be a possible factor. Although higher-dose D-serine may provide additional opportunities to test its pro-cognitive effects in schizophrenia, the nephrotoxicity is a concern. Another potential approach to simulate NMDA function is to prevent D-serine degradation. DAAO, a flavoenzyme of peroxisomes that exists in the central nervous system, is responsible for degrading D-serine and D-alanine (Verrall et al. 2010; Burnet et al. 2011). One of the potential agents is sodium benzoate (benzoate), an inhibitor of DAAO, which can slow the metabolism and elevate the synaptic concentration of D-amino acids such as D-serine and therefore enhance NMDA receptor-mediated neurotransmission (Van den Berghe-Snorek and Stankovich 1985). In an animal study, a single oral dose of sodium benzoate did not change D-serine levels in plasma or in the brain; however, sodium benzoate induced antipsychotic effects in the phencyclidine model of schizophrenia (Matsuura et al. 2015). Small-scale pilot trials showed that benzoate improves the cognitive function of patients with chronic schizophrenia (Lane et al. 2013) and patients with early-phase Alzheimer’s disease (Lin et al. 2014a), supporting the safety and pro-cognitive potential of this DAAO inhibitor.

While D-serine and glycine are endogenous coagonists of NMDA receptors, their availability are different (Papouin et al. 2012). The preferential affinity of D-serine is for synaptic NMDA receptors and that of glycine is for extrasynaptic NMDA receptors. Moreover, long-term potentiation relies on synaptic NMDA receptors, and long-term depression requires both the activations of synaptic and extrasynaptic receptors (Papouin et al. 2012). Whether modulating D-serine and glycine simultaneously can activate NMDA-related functions, such as cognitive function, more efficiently than one approach alone remains unclear. The feasibility of combining two NMDA-enhancement approaches by inhibiting both GlyT-1 and DAAO deserves further studies.

To date, few studies have applied comprehensive cognitive measures, such as the domains recommended by the Measurement and Treatment Research to Improve Cognition in Schizophrenia (MATRICS) Committee (Green et al. 2011), to assess the pro-cognitive effects of NMDA enhancing agents (Kantrowitz et al. 2010; D’Souza et al. 2013; Lane et al. 2013). This study compared the cognitive and clinical efficacy as well as safety of add-on sarcosine plus benzoate vs. sarcosine in patients with chronically stable schizophrenia in a 12-week, placebo-controlled trial.

**Methods**

**Participants**

Ethical approval was obtained from the Institutional Review Board. Patients with chronic schizophrenia were recruited from the inpatient units of the Department of Psychiatry, Changhua Hospital, Taiwan. All patients provided written informed consent following a complete description of the study. Patients were enrolled in this study if they: (1) were aged from 18 to 60 years, (2) were physically healthy and had laboratory assessments (including urine/blood routine, biochemical tests and electrocardiograph) within normal limits, (3) fulfilled the diagnosis of schizophrenia according to the criteria of the Diagnostic and Statistical Manual of Mental Disorders-Fourth Edition (DSM-IV) (American Psychiatric Association 1994), (4) remained symptomatic but without clinically significant fluctuation and the antipsychotic doses were unchanged for at least 2 months prior to this study, and (5) had a minimum baseline total score of 60 on the Positive and Negative Syndrome Scale (PANSS) (Kay et al. 1987). Exclusion criteria included DSM-IV (American Psychiatric Association 1994) diagnosis of mental retardation, substance/alcohol abuse or dependence, history of epilepsy, head trauma or central nervous system (CNS) diseases, pregnancy or breast-feeding, and an inability to follow the protocol.

**Study design**

All subjects had been receiving a balanced hospital diet and unchanged institutionalisation before and during
the trial. After achieving optimal clinical treatment response, patients’ antipsychotic doses remained constant for at least 2 months prior to enrolment into the study and during the study period. All patients were randomly assigned to receive the 12-week trial of stable antipsychotic regimens concomitant with placebo, sarcosine (2 g/day) or sarcosine (2 g/day) plus benzoate (1 g/day) in a 1:1:1 ratio (Figure 1). These doses were safely used in previous trials (Lane et al. 2005; Lane et al. 2013; Tsai et al. 2004). Study medications were given twice daily and were provided in coded containers with a supply of identical in appearance capsules of placebo or either of the active compounds. Patients, caregivers, and investigators (except for the investigational pharmacist) were all masked to the assignment. Patient’s adherence and safety were closely monitored by the research psychiatrists and the nursing staff.

**Measurement of clinical symptoms**

Clinical assessments were measured with the PANSS and the Global Assessment of Functioning (GAF) scale.

Figure 1. Flow chart of the subjects throughout the study period. Seventy-five schizophrenia patients were screened. Sixty-three were eligible and randomised: 21 received placebo; 21, sarcosine; and 21, sarcosine plus benzoate. Participants were measured with PANSS and GAF every 3 weeks. Cognitive domains recommended by the MATRICS Committee, were measured at weeks 0 and 12. Side-effect assessments including the Simpson-Angus Rating Scale, the AIMS, the Barnes Akathisia Scale and UKU Side-effects Rating Scale, were conducted at weeks 0, 3, 6, 9 and 12. Routine laboratory tests were arranged at the baseline and endpoint. In total, 49 of the participants completed the trial: 16 received placebo; 16 received sarcosine; and 17 received sarcosine plus benzoate.
(American Psychiatric Association 1994), and the Clinical Global Impression-Severity scale (CGI-S; Guy 2000) at weeks 0, 3, 6, 9 and 12, and cognitive function was measured at the baseline and at the endpoint. The ratings were performed by the research psychiatrists who were experienced in the rating scales. Inter-rater reliability was analysed with the analysis of variance (ANOVA) test. Only raters reaching the intra-class correlation coefficients of $>0.90$ during pre-study training were allowed to rate the study patients.

Primary outcome measures were PANSS total, global composite and neurocognitive composite of seven cognitive domains (see later); secondary outcome measures were the three PANSS subscales, CGI, GAF and seven cognitive domains (see later).

**Measurement of cognitive function**

This study was started before a commercial Chinese version of Measurement and Treatment Research to Improve Cognition in Schizophrenia (MATRICS) Consensus Cognitive Battery (MCCB) was available. Cognitive function in the current study was assessed using a battery of tests, which were the same tests or the analogues of tests from MCCB (Green et al. 2004), due to the lack of Chinese versions of some tests. The tests utilised in this study were reviewed by one of the developers of the MCCB, Dr Green, in our previous study (Lane et al. 2013). There were seven domains: (1) speed of processing (consisting of three tests: Category Fluency, Trail Marking A and WAIS-III Digit Symbol-Coding); (2) sustained attention (Continuous Performance Test) (Sternberg 1966; Chen et al. 1998); (3) working memory, verbal (backward digit span, Silver 2003) and nonverbal (WMS-III, Spatial Span); (4) verbal learning and memory (WMS-III, word listing); (5) visual learning and memory (WMS-III, visual reproduction); (6) reasoning and problem solving (WISC-III, Maze); and (7) social cognition (the Mayer–Salovey–Caruso Emotional Intelligence Test [MSCEIT] Version 2) (Mayer et al. 2003; Green et al. 2005). The Chinese version of the MSCEIT tasks was translated and back translated from English to Mandarin Chinese with satisfactory reliability, validity (Ma et al. 2012), and applicability (Lin et al. 2013). The first six domains were defined as neurocognition, the processes of linking and appraising information, while the seventh domain, social cognition, was defined as the mental operations underlying social interactions such as the perception, interpretation, and generation of responses to the intentions and behaviours of others (Mayer et al. 2003).

Cognitive performance data of 78 healthy comparison (HC) participants, matched with patients in age ($P = 0.11$) and gender ($P = 0.71$), were collected. Inclusion criteria for HC participants included no Axis I or Axis II psychiatric disorder, no neurological illness, no substance dependence or abuse, no family history of major psychiatric disorder, good general physical health, and age between 18 and 60 years. Participants provided written informed consent after the test procedures were fully explained. All cognitive raw scores of schizophrenia patients were standardised to $t$ scores based on the data of the 78 HC participants. For the cognitive domain that included more than one test, the summary score for the domain was calculated by summing the $t$ scores of the tests included in that domain and then standardising the sum to a $t$ score (Kern et al. 2008). A global composite score (for all seven domains) and a neurocognitive composite score (for the six neurocognitive domains without social cognition) were also calculated.

**Measurement of safety**

Side-effect assessments included the Simpson-Angus Rating Scale (Simpson and Angus 1970) for extrapyramidal side effects, the Abnormal Involuntary Movement Scale (AIMS) for dyskinesia (Guy 1976), the Barnes Akathisia Scale (Barnes 1989) and Udvalg for Kliniske Undersøgelser (UKU) Side-effects Rating Scale (Lingjærde et al. 1987) for systemic side effects. These assessments were conducted at weeks 0, 3, 6, 9 and 12. Routine laboratory tests, including complete blood count, biochemistry, urine routine and electrocardiogram were arranged at the baseline and endpoint.

**Statistical analysis**

At baseline, the demographic and clinical characteristics, including age, gender, years of education, duration of illness, classification of current antipsychotics, number of previous hospitalisations, the number of poor responses to antipsychotics, daily antipsychotic dose, severity of clinical symptoms and cognitive function were compared by Kruskal–Wallis tests for continuous variables and Pearson’s $\chi^2$-tests or Fisher’s exact tests for categorical variables.

The effects of the study drugs on the changes in PANSS, GAF, CGI-S and MATRICS T scores from the baseline to endpoint were assessed according to the linear mixed effects’ model with treatment, visit and treatment–visit interaction as fixed effects; baseline value as the covariate (to adjust the effect of baseline severity); the intercept is the only random effect (to adjust the individual’s effect). The autoregressive of order 1, named AR(1), was used as the covariance type to specify the within-patients’ dependence due to repeated
measurements from the same patient. In addition to the placebo group, the sarcosine group was also set as a reference group to compare the differences in clinical and cognitive change between the sarcosine group and the sarcosine plus benzoate group. All data were analysed by the SPSS 18.0 statistical package. All P values were based on two-tailed tests with a significance level of 0.05.

Results

Patients’ characteristics

To enhance drug adherence and minimise environmental factors, we enrolled inpatients in this study. A total of 75 patients were screened. Among them, 63 were eligible and randomised: 21 received placebo; 21 received sarcosine; and 21 received sarcosine plus benzoate (Figure 1). The mean age of the 63 patients was 38.4 ± 9.3 (SD) years, the duration of illness was 13.7 ± 6.7 years, the education duration was 11.1 ± 2.7 years, the number of previous hospitalisations was 3.6 ± 5.6, the number of poor response to antipsychotics, defined as records of failure to achieve clinical improvement after >8 weeks treatment of an antipsychotic agent, was 4.1 ± 1.1, and antipsychotic dose was 13.7 ± 5.6 mg/day (olanzapine-equivalent) (Gardner et al. 2010). Patients’ characteristics at the baseline were similar among the three treatment groups (Table 1).

In total, 49 of them completed the trial: 16 received placebo; 16 received sarcosine; and 17 received sarcosine plus benzoate (Figure 1). There was no significant baseline difference between the completers and the patients who dropped out (Supplementary Table 1 available online). Moreover, the characteristics of the three groups of completers were also similar (Supplementary Table 1 available online).

PANSS, GAF and CGI-S

Of all the 63 patients, their PANSS, GAF and CGI-S at the baseline were similar among the three treatment groups (Table 2). The sarcosine plus benzoate group was better than the placebo group in improving the GAF (mean difference in score changing rate [±SE] = 0.16 ± 0.06, P = 0.005) after 12 weeks of treatment; however, there was no significant group difference in improvement in the PANSS and CGI-S scores (Table 2).

Of the 49 completers, their PANSS, GAF and CGI-S at the baseline were also similar among the three treatment groups (Supplementary Table S2 available online). Likewise, the sarcosine plus benzoate group was also better than the placebo group in improving GAF after 12 weeks of treatment; however, there was no significant group difference in improvement in the PANSS and CGI-S scores (Supplementary Table S2 available online).

Cognitive battery

Table 3 presents the cognitive functions among the three treatment groups over the 12-week treatment. The cognitive function of the patients who did not complete the 12-week study was not measured at the endpoint. Therefore, the improvement in cognitive function was compared among the 49 completers. In line with previous research (Corbera et al. 2013; McCleery et al. 2014), schizophrenia patients had lower baseline cognitive performance than HC participants (not shown). Similarly to previous reports on the psychometric

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Table 1. Baseline characteristics of the 63 patients randomly assigned to three treatment groups.

|                      | Placebo N=21 | Sarcosine N=21 | Sarcosine + benzoate N=21 | P value*     |
|----------------------|--------------|----------------|---------------------------|--------------|
| Age (years)          | 39.1 ± 9.5   | 38.2 ± 9.3     | 37.8 ± 9.6                | 0.91         |
| Gender (N %)         |              |                |                           |              |
| Male                 | 13 (61.9%)   | 15 (71.4%)     | 11 (52.4%)                | 0.45         |
| Female               | 8 (38.1%)    | 6 (28.6%)      | 10 (47.6%)                |              |
| Education level (years) | 11.6 ± 2.4  | 11.7 ± 3.3     | 11.1 ± 3.2                | 0.91         |
| Age at illness onset(years) | 24.1 ± 6.0  | 23.7 ± 9.5     | 25.4 ± 8.7                | 0.65         |
| Illness duration (years) | 13.8 ± 8.5  | 14.7 ± 6.6     | 12.6 ± 7.3                | 0.65         |
| No. of previous hospitalisations | 3.4 ± 5.4 | 2.8 ± 2.3      | 4.5 ± 7.9                 | 0.65         |
| No. of poor response to antipsychotics | 4.1 ± 1.1 | 4.0 ± 1.1      | 4.2 ± 1.1                 | 0.95         |
| Body mass index (kg/m²) | 21.9 ± 3.7  | 21.5 ± 3.9     | 21.9 ± 3.8                | 0.75         |
| Olanzapine equivalent dose (mg/day) | 13.2 ± 5.5  | 14.5 ± 5.9     | 13.5 ± 5.6                | 0.84         |
| No. of patients using antipsychotics |          |                |                           | 0.70         |
| Amisulpride          | 3            | 2              | 0                         |              |
| Aripiprazole         | 3            | 2              | 4                         |              |
| Olanzapine           | 2            | 4              | 4                         |              |
| Paliperidone         | 1            | 4              | 2                         |              |
| Quetiapine           | 1            | 1              | 0                         |              |
| Risperidone          | 6            | 4              | 8                         |              |
| Zotepine             | 5            | 4              | 3                         |              |

*Kruskal–Wallis tests for continuous variables, Pearson’s χ²-tests for gender variable and Fisher’s exact test for number of patients using antipsychotics.
Table 2. Clinical measures over the 12-week treatment with the comparisons of changing rates among the three treatment groups of for all the 63 patients.

| Scale                     | Treatment group | Baseline* Mean ± SD | Week 3 Mean ± SD | Week 6 Mean ± SD | Week 9 Mean ± SD | Week 12 Mean ± SD | Difference in score changing rate (±SE) vs. placebo** t-Value P-Value | Difference in score changing rate (±SE) vs. Sarcosine** t-Value P-Value |
|---------------------------|-----------------|---------------------|------------------|------------------|------------------|-------------------|---------------------------------------------------------------------|------------------------------------------------------------------------|
| PANSS total               | Placebo         | 86.1 ± 12.3 (N=21)  | 84.5 ± 12.5 (N=19) | 82.4 ± 11.4 (N=16) | 80.9 ± 10.9 (N=16) | 80.1 ± 11.1 (N=16) | -0.04 ± 0.08 -0.53 0.60                                               | -0.11 ± 0.07 -1.53 0.13                                                |
|                           | Sarcosine       | 87.1 ± 10.9 (N=21)  | 84.7 ± 10.8 (N=19) | 82.0 ± 8.9 (N=18)  | 80.5 ± 10.2 (N=16) | 80.5 ± 10.1 (N=16) | 0.04 ± 0.08 0.53 0.54                                               |                                                                         |
|                           | S + B           | 84.9 ± 8.0 (N=21)   | 82.7 ± 8.4 (N=19)  | 81.6 ± 8.8 (N=19)  | 79.3 ± 9.6 (N=17)  | 78.6 ± 9.7 (N=17)  | -0.07 ± 0.07 -1.00 0.32                                               | -0.11 ± 0.07 -1.53 0.13                                                |
| PANSS subscale            |                 |                     |                  |                  |                  |                   |                                                                      |                                                                       |
| Positive                  | Placebo         | 19.6 ± 3.9          | 19.3 ± 3.6       | 18.6 ± 3.6       | 18.3 ± 3.4       | 18.1 ± 3.5       | -0.01 ± 0.03 -0.26 0.80                                               |                                                                         |
|                           | Sarcosine       | 20.3 ± 3.3          | 19.9 ± 2.9       | 19.0 ± 2.5       | 18.8 ± 2.7       | 18.9 ± 2.6       | 0.01 ± 0.03 0.26 0.80                                               |                                                                         |
|                           | S + B           | 18.9 ± 3.1          | 18.3 ± 3.2       | 17.8 ± 3.0       | 17.7 ± 2.4       | 17.5 ± 2.3       | -0.02 ± 0.03 -0.68 0.50                                               | -0.02 ± 0.03 -0.94 0.35                                                |
| Negative                  | Placebo         | 22.6 ± 3.9          | 22.0 ± 4.0       | 21.7 ± 3.5       | 21.4 ± 3.5       | 21.1 ± 3.6       | -0.03 ± 0.02 -1.17 0.24                                               |                                                                         |
|                           | Sarcosine       | 22.8 ± 3.8          | 21.8 ± 3.3       | 21.2 ± 2.8       | 20.7 ± 3.1       | 20.6 ± 3.1       | 0.03 ± 0.02 1.17 0.24                                               |                                                                         |
|                           | S + B           | 22.3 ± 4.2          | 21.7 ± 4.7       | 21.6 ± 4.7       | 20.8 ± 4.7       | 20.7 ± 4.6       | 0.02 ± 0.02 1.02 0.31                                               | -0.00 ± 0.02 -0.17 0.86                                                |
| General psychopathology   | Placebo         | 44.0 ± 7.2          | 43.7 ± 7.4       | 42.2 ± 6.8       | 41.3 ± 6.6       | 41.0 ± 6.8       | -0.01 ± 0.05 -0.15 0.88                                               |                                                                         |
|                           | Sarcosine       | 44.0 ± 6.1          | 43.0 ± 6.0       | 41.8 ± 5.3       | 41.0 ± 6.1       | 41.1 ± 6.2       | 0.01 ± 0.05 0.15 0.88                                               |                                                                         |
|                           | S + B           | 43.7 ± 3.6          | 42.6 ± 3.8       | 42.1 ± 4.1       | 40.9 ± 4.5       | 40.4 ± 4.6       | -0.07 ± 0.05 -1.54 0.13                                               | -0.08 ± 0.05 -1.70 0.09                                                |
| GAF                       | Placebo         | 48.9 ± 7.6          | 49.8 ± 7.1       | 51.8 ± 5.3       | 52.0 ± 5.2       | 52.4 ± 5.2       | -0.06 ± 0.06 -1.00 0.32                                               |                                                                         |
|                           | Sarcosine       | 47.0 ± 7.2          | 47.2 ± 6.6       | 48.6 ± 5.9       | 48.7 ± 6.8       | 48.8 ± 7.0       | 0.06 ± 0.06 1.00 0.32                                               |                                                                         |
|                           | S + B           | 47.0 ± 7.1          | 48.5 ± 6.7       | 49.1 ± 6.7       | 51.1 ± 6.3       | 51.4 ± 6.5       | 0.16 ± 0.06 2.85 0.005***                                               | 0.10 ± 0.06 1.85 0.066                                                |
| CGI                       | Placebo         | 4.0 ± 0.5           | 3.9 ± 0.6        | 4.0 ± 0.5        | 3.9 ± 0.6        | 3.9 ± 0.6        | -0.01 ± 0.01 -0.91 0.36                                               | 0.01 ± 0.01 0.91 0.36                                                |
|                           | Sarcosine       | 4.1 ± 0.5           | 4.0 ± 0.5        | 3.9 ± 0.5        | 4.0 ± 0.5        | 3.9 ± 0.5        | -0.01 ± 0.01 -0.91 0.36                                               |                                                                         |
|                           | S + B           | 3.8 ± 0.5           | 3.8 ± 0.5        | 3.8 ± 0.5        | 3.7 ± 0.6        | 3.7 ± 0.6        | 0.00 ± 0.01 0.70 0.48                                               | 0.10 ± 0.01 1.63 0.11                                                |

*Clinical severity at baseline was similar among the three treatment groups by Kruskal–Wallis tests (PANSS-Total, P=0.75; PANSS-Positive, P=0.54; PANSS-Negative, P=0.77; PANSS-General psychopathology, P=0.87; GAF, P=0.41; CGI, P=0.09).

**Mixed-model repeated measure (MMRM) methods analysis with treatment, visit and treatment–visit interaction as fixed effects and intercept as random effect; baseline value as the covariance. An autoregressive AR(1) covariance matrix was fit to the within-patient repeated measures.

*** P<0.05. P values were based on two-tailed tests.

PANSS, Positive and Negative Syndrome Scale; GAF, Global Assessment Function; CGI, The Clinical Global Impression – Severity scale; S + B, Sarcosine + Benzoate.
properties of multiple cognitive domains like the MATRICS Consensus Cognitive Battery (Kern et al. 2011), standardised scores from domains measured by more than one test tended to be lower than domains measured by a single test (Table 3). Moreover, the patients in this study exhibited severe levels of cognitive impairment in the neurocognitive composite and global composite scores (Table 3). These results were consistent with those reported by McCleery et al. (2014). After 12 weeks, the sarcosine plus benzoate group was superior to the placebo group in improving global composite cognition (mean difference in score changing rate [±SE] = 8.41 ± 3.07, P = 0.009). The sarcosine plus benzoate group was modestly superior to the sarcosine group in improving global composite cognition (mean difference in score changing rate [±SE] = 6.37 ± 2.86, P = 0.03), neurocognitive composite score (mean difference in score changing rate [±SE] = 7.02 ± 2.42, P = 0.03), and verbal learning and memory (mean difference in score changing rate [±SE] = 5.67 ± 2.72, P = 0.04). The sarcosine group was modestly superior to the placebo group in improving reasoning and problem solving (mean difference in score changing rate [±SE] = 8.12 ± 3.86, P = 0.03)** (Table 3).

### Table 3. Cognitive function measured with the seven domains recommended by the Measurement and Treatment Research to Improve Cognition in Schizophrenia (MATRICS) Committee over the 12-week treatment among three treatment groups of the 49 completers, who had both baseline and endpoint cognitive assessments.

| Cognitive domain | Treatment group | Baseline* Mean ± SD | Endpoint Mean ± SD | Difference in score changing rate [±SE]** t value | P value | Difference in score changing rate [±SE]** t value | P value |
|------------------|-----------------|----------------------|---------------------|---------------------------------------------|--------|---------------------------------------------|--------|
| Speed of processing | Placebo | 21.9 ± 8.4 (N=16) | 24.2 ± 9.1 (N=16) | 0.93 ± 2.19 | 0.43 | 0.67 | 0.04 | 0.68 |
|                   | Sarcosine | 22.1 ± 8.6 (N=16) | 25.8 ± 10.3 (N=16) | 3.02 ± 2.17 | 1.39 | 0.17 | 2.15 | 0.26 |
|                   | S + B | 25.1 ± 5.7 (N=17) | 30.7 ± 6.7 (N=17) | 7.77 ± 2.74 | 2.23 | 0.03*** | 6.37 ± 2.86 | 2.09 | 0.04*** |
| Attention/Vigilance | Placebo | 40.0 ± 11.4 (N=16) | 40.7 ± 15.0 (N=16) | 3.62 ± 4.54 | 0.80 | 0.43 | 3.62 ± 4.54 | 0.80 | 0.43 |
|                   | Sarcosine | 35.3 ± 15.2 (N=16) | 35.5 ± 8.9 (N=16) | 2.72 ± 2.61 | 1.04 | 0.30 | 5.67 ± 2.72 | 2.09 | 0.04*** |
|                   | S + B | 35.2 ± 6.5 (N=16) | 38.4 ± 6.8 (N=16) | 0.72 ± 3.51 | 0.21 | 0.84 | 4.32 ± 3.61 | 1.20 | 0.24 |
| Visual learning and memory | Placebo | 36.5 ± 10.4 (N=16) | 40.2 ± 9.1 (N=16) | 2.72 ± 2.61 | 1.04 | 0.30 | 5.67 ± 2.72 | 2.09 | 0.04*** |
| Social cognition | Sarcosine | 38.5 ± 9.5 (N=16) | 45.8 ± 12.1 (N=16) | 0.72 ± 3.51 | 0.21 | 0.84 | 4.32 ± 3.61 | 1.20 | 0.24 |
| Neurocognitive composite | Placebo | 37.2 ± 17.4 (N=16) | 42.1 ± 17.7 (N=16) | 8.12 ± 3.86 | 2.11 | 0.05*** | 8.08 ± 3.86 | 2.09 | 0.04*** |
| Verbal learning and memory | Sarcosine | 33.9 ± 17.1 (N=16) | 41.1 ± 13.5 (N=16) | 7.71 ± 3.83 | 2.01 | 0.05 | 5.67 ± 2.72 | 2.09 | 0.04*** |
|                   | S + B | 35.2 ± 6.5 (N=16) | 38.4 ± 6.8 (N=16) | 0.72 ± 3.51 | 0.21 | 0.84 | 4.32 ± 3.61 | 1.20 | 0.24 |
| Working memory | Sarcosine | 35.0 ± 7.6 (N=16) | 38.6 ± 11.7 (N=16) | 1.90 ± 2.50 | 0.76 | 0.45 | 2.10 ± 2.58 | 0.81 | 0.42 |
| Social cognition | S + B | 33.7 ± 9.7 (N=16) | 39.6 ± 12.3 (N=16) | 3.91 ± 2.48 | 1.57 | 0.12 | 2.10 ± 2.58 | 0.81 | 0.42 |
| Neurocognitive composite | Placebo | 33.7 ± 9.7 (N=16) | 39.6 ± 12.3 (N=16) | 3.91 ± 2.48 | 1.57 | 0.12 | 2.10 ± 2.58 | 0.81 | 0.42 |
| Global composite score | S + B | 33.9 ± 16.8 (N=16) | 39.4 ± 10.8 (N=16) | 3.46 ± 4.53 | 0.76 | 0.45 | 2.10 ± 2.58 | 0.81 | 0.42 |

All cognitive raw scores of patients were standardised to t scores based the data of 78 healthy comparison participants. For cognitive domains that included more than one measure, the summary score for the domain was calculated by summing the t scores of the tests included in that domain and then standardising the sum to a t score (Kern et al. 2008).

**For assessing neurocognitive function, a composite t score including all six neurocognitive domains, excluding social cognition, was calculated by standardising the sum of the t scores.

**For assessing the global cognitive function, an overall composite t score including all seven domains was calculated by standardising the sum of the t scores.**

**Cognitive performance at baseline was similar among the three treatment groups by Kruskal–Wallis tests.**
the placebo group, 0.2 ± 0.7 in the sarcosine group and 1.0 ± 1.0 in the sarcosine plus benzoate group. The baseline AIMS was 0.2 ± 1.1 in the placebo group, 0.2 ± 0.7 in the sarcosine group and 0.1 ± 0.4 in the sarcosine plus benzoate group. The baseline Barnes Akathesia Scale was 0.5 ± 1.5 in the placebo group, 0.2 ± 0.6 in the sarcosine group and 0.1 ± 0.4 in the sarcosine plus benzoate group. There were no significant differences among the three groups in the Simpson-Angus Rating Scale (P = 0.11), AIMS (P = 0.79) and Barnes Akathesia score (P = 0.56).

At the endpoint, the severity of extrapyramidal syndrome remained minimal and did not reveal significant differences among the three groups. The mean of the Simpson-Angus Rating Scale at the endpoint was 0.4 ± 1.3 in the placebo group, 0 ± 0 in the sarcosine group, and 0.4 ± 0.7 in the sarcosine plus benzoate group (P = 0.14). The endpoint AIMS was 0 ± 0 in the placebo group, 0.1 ± 0.5 in the sarcosine group, and 0 ± 0 in the sarcosine plus benzoate group (P = 0.36). The endpoint Barnes Akathesia score was 0.6 ± 1.8 in the placebo group, 0.1 ± 0.5 in the sarcosine group and 0 ± 0 in the sarcosine plus benzoate group (P = 0.32).

Treatment-emergent adverse events other than extrapyramidal syndrome in the placebo group included increased dream activity (N = 2), micturition disturbance (N = 1), increased tendency to sweating (N = 1), pruritus (N = 1), weight gain (N = 2), weight loss (N = 1) and increased sexual desire (N = 1); in the sarcosine group, increased tendency to sweating (N = 1), increased dream activity (N = 1), weight gain (N = 1) and weight loss (N = 2); in the sarcosine plus benzoate group, increased sleepiness (N = 1), increased duration of sleep (N = 2), weight gain (N = 1) and weight loss (N = 1). These systemic side effects were all mild and brief, not warranting medical treatment. They were likely coincidental.

The routine blood cell count, biochemistry, urine routine, and electrocardiogram after treatment remained unchanged (data not shown). No dropout occurred due to adverse effects.

Discussion

Cognitive impairment, a core feature throughout the course of schizophrenia, is related to functional outcome. However, it is poorly managed with currently available antipsychotics (Green et al. 2000; Mohamed et al. 2008; Allott et al. 2011). Moreover, there have been few positive responses from putative pro-cognitive drugs in humans (Millan et al. 2012; Preskorn et al. 2014). The present study suggests that adjunctive sarcosine plus benzoate, but not sarcosine alone, can improve the cognitive and global functioning of patients with chronic schizophrenia, even when their clinical symptoms cannot be improved. This finding lends support to the previous notion that NMDA-enhancement therapy can improve the neurocognition of patients with schizophrenia (Kantrowitz et al. 2010; Lane et al. 2013) and further indicates that this pro-cognitive effect can be primary without improvement of the clinical outcome. Although it may not achieve a striking clinical difference, the effect of sarcosine plus benzoate on GAF was statistically significant after 12 weeks. It has been suggested that the global functioning of patients with schizophrenia may be related to cognitive function rather than to clinical symptoms (Carrión et al. 2011; Green et al. 2011). This result indicates that pro-cognitive effects may reflect in the improvement of global functioning.

This clinical trial is the first to examine the combination effect of a DAAO inhibitor and a GlyT-1 inhibitor on symptomatic or cognitive domains of schizophrenia. The finding on its safety is akin to those of previous studies (Tsai et al. 2004; Lane et al. 2005, 2006, 2010, 2013), indicating add-on sarcosine and sarcosine plus benzoate are well tolerated. The mild side effects were likely coincidental observations because there was no significant group difference. Use of direct NMDA receptor agonists has been limited by the high doses that must be given or the relatively poor penetration of the CNS (Javitt 2008). Therefore, combining two kinds of NMDA receptor-enhancing agents is of a great interest in research (Labrie and Roder 2010). For example, d-serine in combination with a DAAO antagonist produced greater ameliorative effects than either compound applied alone in animals (Hashimoto et al. 2009). Previous pre-clinical and clinical studies supported compounds providing selective modulation of the NMDA receptor d-serine/glycine site such as diminishing d-serine catabolism by inhibition of DAAO or stimulating the glycine modulatory sites by blocking GlyT-1 could demonstrate beneficial effects on schizophrenia (Tsai and Lin 2009). Our study supports that coadministration of GlyT-1 and DAAO inhibitors could provide a potential therapeutic approach. However, there has not yet been a preclinical study examining the combination effect of a DAAO inhibitor, such as benzoate, and a GlyT-1 inhibitor, such as sarcosine in the NMDA receptor models of cognitive impairment associated with schizophrenia. Further studies are warranted.

Although there was no significant improvement in global composite score from the sarcosine add-on treatment, the sarcosine group also displayed improved reasoning and problem solving (P value of 0.04) when compared to the placebo group. Therefore, it remains possible whether a dose higher than the dose in the
current study, to reach a higher level of NMDA activation, would generate a better response (Lane et al. 2008; Kantrowitz et al. 2010).

The current result shows that adjunctive sarcosine plus benzoate improved the cognitive function of patients but not their negative symptoms. The relationships between positive, negative and cognitive symptoms have been equivocal in both cross-sectional and longitudinal studies. While some studies suggest that cognitive deficits are modestly associated with negative symptoms (Berman et al. 1997; Savilla et al. 2008), no significant associations between changes in negative symptoms and cognitive impairment have been raised (Bell and Mishara 2006; Umbricht et al. 2014). Our current result supports the notion that negative symptoms and cognition should be viewed as independent targets for intervention (Nasrallah et al. 2014). As in benzoate alone treatment (Lane et al. 2013), the study drugs in the current study did not affect social cognition. This may reflect the multi-determined nature of the social cognitive domain (Couture et al. 2006) and may be related to the inadequate study duration. The current results encourage further study of longer treatment duration and NMDA-enhancement therapy plus social rehabilitation in patients with chronic schizophrenia.

Different from most sarcosine or benzoate trials, the current study showed that all three groups have modest improvement in clinical manifestation; however, the improvements by sarcosine or sarcosine plus benzoate treatment were similar to that of the placebo group. In comparison with two previous sarcosine trials which showed effects on clinical symptoms, including positive and negative symptoms, in chronic schizophrenia, the current subjects tended to be older (38.4 (mean value) vs. 31.8 (Tsai et al. 2004) and 30.9 years old (Lane et al. 2010)), have longer illness durations (14.7 vs. 9.6 (Tsai et al. 2004) and 9.5 years (Lane et al. 2010)), and have more hospitalisations ([3.6 vs. 2.8 (Lane et al. 2010)]. The age and illness duration of the current study subjects were closer to those in the trial of sarcosine added to clozapine, which failed to reduce positive and negative symptoms of schizophrenia [38.4 vs. 36.1 years old and 14.7 vs. 14.9 years (Lane et al. 2006)], suggesting that adjunctive 2 g/day sarcosine reveals limited efficacy in older patients and those with longer illness duration.

A recent study (Lane et al. 2013) demonstrated that benzoate was beneficial for patients with schizophrenia. The age, illness duration, and previous number of hospitalisations (38.4 years, 13.7 years and 3.2, respectively) of the current study subjects were similar to those (37.3 years, 14.7 years and 3.6, respectively) in the previous trial of benzoate (Lane et al. 2013). However, the current study showed that sarcosine plus benzoate had limited effects in patients who had failed to achieve clinical improvement with multiple trials of antipsychotic agents. Without a group of only add-on benzoate, it remains unclear whether sarcosine plus benzoate may have generated overactive neurotransmission; thereby somewhat hampering the beneficial effect of benzoate per se. Studies using only add-on benzoate is necessary to clarify this issue.

Although the precise mechanisms are unknown, the involvement of neuroimmune dysregulation and the glutamatergic system in the pathophysiology of schizophrenia is intriguing (Steiner et al. 2012; Heresco-Levy et al. 2015). Agents with both anti-inflammatory and glutamatergic transmission modification properties are promising in the treatment of schizophrenia (Hashimoto 2014). It was reported that sodium benzoate can suppress the mevalonate pathway and reduce microglial and astroglial inflammatory responses (Brahmachari et al. 2009). Whether patients can benefit from benzoate treatment through both the anti-inflammatory and NMDA receptor-mediated pathways deserves further investigation.

This study has several limitations. First, there were only three study arms: (1) placebo, (2) sarcosine, and (3) sarcosine plus benzoate. Therefore, comparisons of benzoate vs. the current three arms remain unclear. For example, whether sarcosine plus benzoate is superior or equal to benzoate alone is unknown. That is, whether there is a synergistic or additive effect from sarcosine plus benzoate remains to be determined. Second, the sample size was small and the study subjects were chronic patients in a single psychiatric hospital; therefore the generalizability of the findings would be limited. Studies focussing on subjects who are younger, at an earlier stage of illness or have less severe psychotic symptoms are necessary. Third, the changes in blood levels of amino acids such as glycine and D-serine after treatment were not measured. Further studies should be conducted. Fourth, the treatment duration of 12 weeks may have been insufficient for assessment of the efficacy and safety for patients with chronic schizophrenia. Fifth, the sarcosine plus benzoate group tended to have more females and a shorter duration of illness, albeit insignificantly, which might have influenced the practise effect on cognitive testing. Sixth, we did not follow cognitive function at a time point post the trial to examine the sustained influence of NMDA-enhancement treatment after the subjects discontinued the study drugs. Finally, the study was started before a commercial Chinese version of MCCB was available; therefore the t scores in the current study could not be compared with other studies directly. Further trials which refer to the
normative data for Chinese version of MCCB should be encouraged.

In conclusion, a combination of NMDA-enhancing agents (sarcosine and benzoate), but not sarcosine alone, can improve cognitive function in patients with chronic schizophrenia, even when their clinical symptoms cannot be improved. These findings support the NMDA theory of cognitive impairment in schizophrenia (Krystal et al. 1994; Javitt et al. 2012; Hashimoto et al. 2013; Paoletti et al. 2013; Lin et al. 2014b). Future larger-sized studies in other racial populations are warranted.

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Statement of interest

Dr Tsai is a director and shareholder of SyneuRx International Corporation, which plans to develop glycine transporter-1 inhibitor and D-amino acid oxidase inhibitors, including sodium benzoate, for the treatment of central nervous system disorders. SyneuRx International Corporation was not involved in the funding or execution of the study. All other authors report no biomedical financial interests or potential conflicts of interest.

References

Allott K, Liu P, Proffitt T-M, Killackey E. 2011. Cognition at illness onset as a predictor of later functional outcome in early psychosis: systematic review and methodological critique. Schizophr Res. 125:221–235.

American Psychiatric Association. 1994. Diagnostic and Statistical Manual of Mental Disorders, 4th ed. Washington, DC: American Psychiatric Press.

Bado P, Madeira C, Vargas-Lopes C, Moulin T, Wasilewska-Sampaio A, Maretti L, et al. 2011. Effects of low-dose D-serine on recognition and working memory in mice. Psychopharmacology (Berl). 218:461–470.

Balu DT, Coyle JT. 2014. Chronic D-serine reverses arc expression and partially rescinds dendritic abnormalities in a mouse model of NMDA receptor hypofunction. Neurochem Int. 75:76–78.

Barnes TR. 1989. A rating scale for drug-induced akathisia. Br J Psychiatry. 154:672–676.

Bell MD, Mishara AL. 2006. Does negative symptom change relate to neurocognitive change in schizophrenia? Implications for targeted treatments. Schizophr Res. 81:17–27.

Berman I, Viegner B, Merson A, Allan E, Pappas D, Green Al. 1997. Differential relationships between positive and negative symptoms and neuropsychological deficits in schizophrenia. Schizophr Res. 25:1–10.

Brahmachari S, Jana A, Pahan K. 2009. Sodium benzoate, a metabolite of cinnamon and a food additive, reduces microglial and astroglial inflammatory responses. J Immunol. 183:5917–5927.

Buchanan RN, Javitt DC, Marder SR, Schooler NR, Gold JM, McMahen RP, et al. 2007. The Cognitive and Negative Symptoms in Schizophrenia Trial (CONSIST): the efficacy of glutamatergic agents for negative symptoms and cognitive impairments. Am J Psychiatry. 164:1593–1602.

Burnet BW, Anderson PN, Chen L, Nikiforova N, Harrison PJ, Wood MJ. 2011. D-amino acid oxidase knockdown in the mouse cerebellum reduces NR2A mRNA. Mol Cell Neurosci. 46:167–175.

Carrión RE, Goldberg TE, McLaughlin D, Auther AM, Correll CU, Cornblatt BA. 2011. Impact of neurocognition on social and role functioning in individuals at clinical high risk for psychosis. Am J Psychiatry. 168:806–813.

Chen WJ, Liu SK, Chang CJ, Lien YJ, Chang YH, Hwu HG. 1998. Sustained attention deficit and schizotypal personality features in nonpsychotic relatives of schizophrenic patients. Am J Psychiatry. 155:1214–1220.

Corbera S, Wexler BE, Ikezawa S, Bell MD. 2013. Factor Structure of Social Cognition in Schizophrenia: Is Empathy Preserved? Schizophr Res Treatment. 2013:409205

Couture SM, Penn DL, Roberts DL. 2006. The functional significance of social cognition in schizophrenia: a review Schizophr Bull. 32:445–463.

D’Souza DC, Radhakrishnan R, Perry E, Bhakta S, Singh NM, Yadav R, et al. 2013. Feasibility, safety, and efficacy of the combination of d-serine and computerized cognitive retraining in schizophrenia: an international collaborative pilot study. Neuropsychopharmacology. 38:492–503.

Errico F, Napolitano F, Squillace M, Vitucci D, Blasi G, de Bartolomeis A, et al. 2013. Decreased levels of D-aspartate and NMDA in the prefrontal cortex and striatum of patients with schizophrenia. J Psychiatr Res. 47:1432–1437.

FirstWord Pharma. 2014. Roche’s bitopertin fails to meet main goals of late-stage schizophrenia trials; January 21st, 2014: [Available from: http://www.firstwordpharma.com/node/1180713#axzz3En2j9x4U.

Gardner DM, Murphy AL, O’Donnell H, Centorrino F, Baldessarini RJ. 2010. International consensus study of antipsychotic dosing. Am J Psychiatry. 167:686–693.

Green MF, Kern RS, Braff DL, Mintz J. 2000. Neurocognitive deficits and functional outcome in schizophrenia: are we measuring the “right stuff”? Schizophr Bull. 26:119–136.

Green MF, Nuechterlein KH, Gold JM, Barch DM, Cohen J, Essock S, et al. 2004. Approaching a consensus cognitive battery for clinical trials in schizophrenia: the NIMH-MATRICS conference to select cognitive domains and test criteria. Biol Psychiatry. 56:301–307.

Green MF, Olivier B, Crawley JN, Penn DL, Silverstein S. 2005. Social cognition in schizophrenia: recommendations from the measurement and treatment research to improve cognition in schizophrenia new approaches conference. Schizophr Bull. 31:882–887.
Green MF, Schooler NR, Kern RS, Frese FJ, Granberry W, Harvey PD, et al. 2011. Evaluation of functionally meaningful measures for clinical trials of cognitive enhancement in schizophrenia. Am J Psychiatry. 168:400–407.

Guy W. 1976. ECDEU Assessment Manual for Psychopharmacology: U.S. Dept. of Health, Education, and Welfare, Public Health Service, Alcohol, Drug Abuse, and Mental Health Administration, National Institute of Mental Health, Psychopharmacology Research Branch, Division of Extramural Research Programs.

Guy W. 2000. Clinical Global Impressions (CGI) Scale. Modified From: Rush J, et al. Psychiatric Measures. Washington DC: APA.

Hashimoto K. 2014. Targeting of NMDA receptors in new treatments for schizophrenia. Expert Opin Ther Targets. 18:1049–1063.

Hashimoto K, Fujita Y, Horio M, Kunitachi S, Iyo M, Ferraris D, et al. 2009. Co-administration of a D-amino acid oxidase inhibitor potentiates the efficacy of D-serine in attenuating prepulse inhibition deficits after administration of dizocilpine. Biol Psychiatry. 65:1103–1106.

Hashimoto K, Malchow B, Falkai P, Schmitt A. 2013. Glutamate modulators as potential therapeutic drugs in schizophrenia and affective disorders. Eur Arch Psychiatry Clin Neurosci. 263:367–377.

Heresco-Levy U, Durrant AR, Ermilov M, Javitt DC, Miya K, Mori H. 2015. Clinical and electrophysiological effects of D-serine in a schizophrenia patient positive for anti-N-methyl-D-aspartate receptor antibodies. Biol Psychiatry. 77:e27–e29.

Heresco-Levy U, Ermilov M, Lichtenberg P, Bar G, Javitt DC. 2004. High-dose glycine added to olanzapine and risperidone for the treatment of schizophrenia. Biol Psychiatry. 55:165–171.

Heresco-Levy U, Javitt DC, Ebstein R, Vass A, Lichtenberg P, Bar G, et al. 2005. D-serine efficacy as add-on pharmacotherapy to risperidone and olanzapine for treatment-refractory schizophrenia. Biol Psychiatry. 57:577–585.

Javitt DC. 2008. Glycine transport inhibitors and the treatment of schizophrenia. Biol Psychiatry. 63:6–8.

Javitt DC, Silipo G, Cienfuegos A, Shelley AM, Bark N, Park M, et al. 2001. Adjunctive high-dose glycine in the treatment of schizophrenia. Int J Neuropsychopharmacol. 4:385–391.

Javitt DC, Zukin SR, Heresco-Levy U, Umbricht D. 2012. Has an angel shown the way? Etiological and therapeutic implications of the PCP/NMDA model of schizophrenia. Schizophr Bull. 38:958–966.

Kantrowitz JT, Malhotra AK, Cornblatt B. 2010. High dose D-serine in the treatment of schizophrenia. Schizophrenia Res. 121:125–130.

Kasper S. 2006. Optimisation of long-term treatment in schizophrenia: Treating the true spectrum of symptoms. Eur Neuropsychopharmacol. 16:s135–s141.

Kay SR, Fiszbein A, Opler LA. 1987. The positive and negative syndrome scale (PANSS) for schizophrenia. Schizophr Bull. 13:261–276.

Kern RS, Nuechterlein KH, Green MF, Baade LE, Fenton WS, Gold JM, et al. 2008. The MATRICS Consensus Cognitive Battery, Part 2: Co-Norming and Standardization. Am J Psychiatry. 165:214–220.

Kern RS, Gold JM, Dickinson D, Green MF, Nuechterlein KH, Baade LE, et al. 2011. The MCCB Impairment Profile for Schizophrenia outpatients: results from the MATRICS psychometric and standardization study. Schizophr Res. 126:124–131.

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. Arch Gen Psychiatry. 51:199–214.

Labrie V, Roder JC. 2010. The involvement of the NMDA receptor D-serine/glycine site in the pathophysiology and treatment of schizophrenia. Neurosci Biobehav Rev. 34:351–372.

Laney H, Chang YC, Liu YC, Chiu CC, Tsai GE. 2005. Sarcosine or D-serine add-on treatment for acute exacerbation of schizophrenia: a randomized, double-blind, placebo-controlled study. Arch Gen Psychiatry. 62:1196–1204.

Lanen YH, Huang CL, Wu PL, Liu YC, Chang YC, Lin PY, et al. 2006. Glycine transporter I inhibitor, N-methylglycine (sarcosine), added to clozapine for the treatment of schizophrenia. Biol Psychiatry. 60:645–649.

Lanen YH, Lin CH, Green MF, Hellermann G, Huang CC, Chen PW, et al. 2013. Add-on treatment of benzoate for schizophrenia: a randomized, double-blind, placebo-controlled trial of D-amino acid oxidase inhibitor. JAMA Psychiatry. 70:1267–1275.

Lanen YH, Lin CH, Huang YJ, Liao CH, Chang YC, Tsai GE. 2010. A randomized, double-blind, placebo-controlled comparison study of sarcosine (N-methylglycine) and d-serine add-on treatment for schizophrenia. Int J Neuropsychopharmacol. 13:451–460.

Lanen YH, Liu YC, Huang CL, Chang YC, Liau CH, Perng CH, et al. 2008. Sarcosine (N-methylglycine) treatment for acute schizophrenia: a randomized, double-blind study. Biol Psychiatry. 63:9–12.

Levin R, Dor-Abarbanel AE, Edelman S, Durrant AR, Hashimoto K, Javitt DC, et al. 2015. Behavioral and cognitive effects of the N-methyl-D-aspartate receptor co-agonist D-serine in healthy humans: Initial findings. J Psychiatr Res. 61:188–195.

Liu CH, Chen PK, Chang YC, Chuo LJ, Chen YS, Tsai GE, et al. 2014a. Benzoate, a D-amino acid oxidase inhibitor, for the treatment of early-phase Alzheimer disease: A randomized, double-blind, placebo-controlled trial. Biol Psychiatry. 75:678–685.

Lin CH, Tseng YL, Huang CL, Chang YC, Tsai GE, Lane HY. 2013. Synergistic effects of COMT and TPH2 on social cognition. Psychiatry. 76:273–294.

Lin CY, Tsai GE, Lane HY. 2014b. Assessing and treating cognitive impairment in schizophrenia: current and future. Curr Pharm Des. 20:5127–5138.

Lingjærde O, Ahlfors UG, Bech P, Dencker SJ, Elgen K. 1987. The Clinical Global Impression (CGI) Scale. Modified From: Rush J, et al. Psychiatric Measures. Washington DC: APA.
McBain CJ, Kleckner NW, Wyrick S, Dingledine R. 1989. Structural requirements for activation of the glycine coagonist site of N-methyl-D-aspartate receptors expressed in Xenopus oocytes. Mol Pharmacol. 36:556–565.

McCleery A, Ventura J, Kern RS, Subotnik KL, Gretchen-Doorly D, Green MF, et al. 2014. Cognitive functioning in first-episode schizophrenia: MATRICS Consensus Cognitive Battery (MCCB) Profile of Impairment. Schizophr Res. 157:33–39.

Meltzer HY. 2013. Update on typical and atypical antipsychotic drugs. Annu Rev Med. 64:393–406.

Millan MJ, Agid Y, Brüne M, Bullmore ET, Carter CS, Clayton NS, et al. 2012. Cognitive dysfunction in psychiatric disorders: characteristics, causes and the quest for improved therapy. Nat Rev Drug Discov. 11:141–168.

Mohamed S, Rosenheck R, Swartz M, Stroup S, Lieberman J, Keefe R. 2008. Relationship of cognition and psychopathology to functional impairment in schizophrenia. Am J Psychiatry. 165:978–987.

Nasrallah HA, Keefe RSE, Javitt DC. 2014. Cognitive deficits and poor functional outcomes in schizophrenia: clinical and neurobiological progress. Current Psychiatry. 13:1–s11.

Paoletti P, Bellone C, Zhou Q. 2013. NMDA receptor subunit diversity: impact on receptor properties, synaptic plasticity and disease. Nat Rev Neurosci. 14:383–400.

Papouin T, Ladépêche L, Ruel J, Sacchi S, Labasque M, Hanini M, et al. 2012. Synaptic and Extrasynaptic NMDA Receptors Are Gated by Different Endogenous Coagonists. Cell. 150:633–646.

Preskorn SH, Gawryl M, Dgetluck N, Palfreyman M, Bauer LO, Hilt D. 2014. Normalizing effects of EVP-6124, an α7 nicotinic partial agonist, on event-related potentials and cognition: a proof of concept, randomized trial in patients with schizophrenia. J Psychiatr Pract. 20:12–24.

Savilla K, Kettler L, Galletly C. 2008. Relationships Between Cognitive Deficits, Symptoms and Quality of Life in Schizophrenia. Aust N Z J Psychiatry. 42:496–504.

Sawa A. 2009. Cortical Development and Glutamatergic Dysregulation in Schizophrenia. Biol Psychiatry. 66:530–532.

Schultz SK, Andreasen NC. 1999. Schizophrenia. Lancet. 353:1425–1430.

Sergi MJ, Green MF, Widmark C, Reist C, Erhart S, Braff DL, et al. 2007. Social cognition [corrected] and neurocognition: effects of risperdone, olanzapine, and haloperidol. Am J Psychiatry. 164:1585–1592.

Simpson GM, Angus JW. 1970. A rating scale for extrapyramidal side effects. Acta Psychiatr Scand Suppl. 212:11–19.

Steiner J, Bogerts B, Sarnyai Z, Walter M, Gos T, Bernstein HG, et al. 2012. Bridging the gap between the immune and glutamate hypotheses of schizophrenia and major depression: Potential role of glial NMDA receptor modulators and impaired blood-brain barrier integrity. World J Biol Psychiatry. 13:482–492.

Sternberg S. 1966. High-speed scanning in human memory. Science. 153:652–654.

Tsai GE, Lane HY, Yang P, Chong MY, Lange N. 2004. Glycine transporter I inhibitor, N-methylglycine (sarcosine), added to antipsychotics for the treatment of schizophrenia. Biol Psychiatry. 55:452–456.

Tsai GE, Lin PY. 2009. Strategies to enhance N-methyl-D-aspartate receptor-mediated neurotransmission in schizophrenia, a critical review and meta-analysis. Curr Pharm Des. 16:522–537.

Tsai GE, Yang P, Chung LC, Lange N, Coyle JT. 1998. D-serine added to antipsychotics for the treatment of schizophrenia. Biol Psychiatry. 44:1081–1089.

Umbricht D, Alberati D, Martin-Facklam M, Borroni E, Youssef EA, Ostland M, et al. 2014. Effect of bitopertin, a glycine reuptake inhibitor, on negative symptoms of schizophrenia: A randomized, double-blind, proof-of-concept study. JAMA Psychiatry. 71:637–646.

Van den Berghe-Snorek S, Stankovich MT. 1985. Thermodynamic control of D-amino acid oxidase by benzoate binding. J Biol Chem. 260:3373–3379.

Verrall L, Burnet PW, Betts JF, Harrison PJ. 2010. The neurobiology of D-amino acid oxidase and its involvement in schizophrenia. Mol Psychiatry. 15:122–137.

Weiser M, Heresco-Levy U, Davidson M, Javitt DC, Werbeloff N, Gershon AA, et al. 2012. A multicenter, add-on randomized controlled trial of low-dose d-serine for negative and cognitive symptoms of schizophrenia. J Clin Psychiatry. 73:e728–e734.