

D-cycloserine Augmentation of Cognitive Behavioral Therapy for Delusions
Detailed Study Protocol Principal Investigator: Donald Goff, MD

Purpose of the Study

Specific Aims and Design:

Antipsychotic-resistant delusions in schizophrenia are common, are often distressing, and increase the risk for incarceration and self-injury. Converging evidence from our group and others suggests that the persistence of delusions represents an inability to “unlearn” or “extinguish” false beliefs and that this deficit may respond to D-cycloserine (DCS) augmentation of cognitive behavioral therapy (CBT). We have demonstrated a selective impairment of memory consolidation for fear extinction in schizophrenia subjects which is highly correlated with persistence of delusions. In animal models, fear extinction is facilitated by DCS; in anxiety disorders DCS has well-established efficacy as an augmenter of CBT. We have shown in two trials that DCS similarly enhances memory consolidation in schizophrenia subjects. We recently completed a two-session placebo-controlled crossover trial of DCS augmentation of a CBT exercise in schizophrenia subjects with persistent delusions and found marked improvement of delusions and delusional distress. While the evidence for this approach is compelling, we believe a parallel-group proof-of-concept trial employing a full course of CBT is needed prior to embarking on a larger, multi-center trial which, if successful, could fundamentally change our approach to the treatment of medication-resistant delusions.

We therefore propose a placebo-controlled 12 week trial of DCS augmentation of once-weekly CBT sessions in 60 schizophrenia subjects with antipsychotic-resistant delusions. In addition to testing efficacy, this trial will characterize DCS effects in terms of time course and persistence of response and will examine DCS effects on memory consolidation and cognitive flexibility as possible mediators of DCS enhancement of CBT for delusions.

Primary

1. Evaluate efficacy of DCS facilitation of a 12 week course of CBT for delusions.
 - a. Hypothesis: DCS will significantly reduce severity of delusions measured by the PSYRATS Delusions Subscale total score compared to placebo.

Secondary:

2. Evaluate the time course of DCS facilitation of CBT.
 - a. Onset Hypothesis: The rate of response of delusions with DCS defined as a 20% reduction in PSYRATS Delusions Subscale total score will be superior to placebo after 2 weeks of DCS treatment.
 - b. Persistence Hypothesis: DCS improvement of delusions compared to placebo will persist at 3 and 6 month follow-up as defined as maintenance of a 20% or greater reduction in the PSYRATS Delusions Subscale total score from baseline.

Tertiary

3. Identify mediators of delusional response to DCS.
 - a. Memory Consolidation Hypothesis: Change in 7-day thematic recall on the Logical Memory Test after the first dose of DCS will predict improvement on the PSYRATS Delusions Subscale total score at 12 weeks.
 - b. Cognitive Flexibility Hypothesis: Change in the number of alternative beliefs generated during the first compared to the second administration of the Alternative Beliefs exercise in the DCS group will predict improvement of the PSYRATS Delusions Subscale total score at 12 weeks.
4. Identify neural differences in patients compared to age-matched healthy control population.
 - a. Network connectivity hypothesis: Decreased connectivity between the hippocampus and prefrontal cortex in patients at rest and during encoding of stories

Study #:S12-02991

and images, DCS enhancement of connectivity.

b. Memory Hypothesis: Patient impairment of network connectivity will predict performance on immediate and 1-week delay memory relative to healthy controls

Background Significance:

Public Health Significance: Delusions remain a serious public health problem, with persistent antipsychotic-resistant delusions present in approximately 30% of schizophrenia patients. In addition, most patients experience intermittent psychotic relapse associated with major disruptions in rehabilitation and recovery. Persecutory delusions are associated with great psychological distress to patients and family members, whereas other forms of delusions commonly result in social and vocational disability, incarceration, and can result in harm to self or others.

Alternative Pharmacological Treatments: The current approach of dopamine D2 receptor blockade for the treatment of delusions, while effective in many patients, is only partially effective or is ineffective in approximately 30% of patients. Findings from PET imaging reveal that excessive release by stimulants of striatal dopamine in schizophrenia subjects predicts response of delusions to D2 antagonists, but a substantial proportion of individuals with schizophrenia overlap with healthy subjects on this measure and display minimal response to antipsychotic medication (1). Of the second generation antipsychotics, only olanzapine demonstrated greater effectiveness than perphenazine in the CATIE trial, although this effect was modest and at the cost of substantial medical morbidity (2). In contrast, clozapine is effective in up to 50% of refractory patients. While the mechanism for clozapine's efficacy remains unclear, it is noteworthy that clozapine (and not haloperidol) attenuates psychosis produced by NMDA receptor blockade (3), suggesting that dysregulation of NMDA receptors may play a role in delusions that are resistant to traditional antipsychotics but potentially responsive to clozapine. Unfortunately, the use of clozapine is constrained by clinician reluctance to prescribe, patient refusal, requirements for hematological monitoring, and metabolic side effects and sedation, so that less than 10% of patients with refractory delusions receive it. No other pharmacologic treatment has demonstrated consistent efficacy for antipsychotic-resistant delusions, although the most promising results have come from agents acting at glutamatergic systems, including modulators of the NMDA receptor complex (sarcosine, D-serine) (4) and of glutamate release (lamotrigine) (5).

Cognitive Behavioral Therapy (CBT): CBT was recommended as an evidenced-based treatment for refractory psychosis in the most recent PORT guidelines (6). However, efficacy for positive symptoms is quite modest; in a meta-analysis by Wykes and colleagues (7), the effect size was 0.22 for methodologically-rigorous trials that included rater blinding and an adequate control condition. Predictors of response included greater insight at baseline and recent inpatient admission; duration of treatment was not related to outcome. A clear relationship between CBT approach and effectiveness for psychosis has not emerged; treatments involving individual or group treatment and lasting from 6 weeks to 18 months have all produced positive results (8). Kingdon and Turkington (9) demonstrated that CBT for psychosis can be disseminated within community mental health systems. They trained psychiatric nurses from six community mental health centers to conduct a six-session CBT protocol and demonstrated significant benefit in a randomized controlled trial involving 443 schizophrenia patients. CBT aims to facilitate the patient's capacity to develop alternative explanations for delusions, but this approach is limited by cognitive deficits that prevent most delusional patients from recognizing and changing or "unlearning" their false beliefs. CBT also helps patients cope with distressing beliefs and to modify resulting dysfunctional behaviors.

Summary of Significance: Given the high prevalence and serious adverse consequences of treatment refractory delusions, a major unmet need exists for a new and effective treatment approach. Our model, based on animal and human studies of memory consolidation and fear

extinction, posits a deficit in the capacity to “unlearn” delusional beliefs which can be reversed by DCS combined with CBT. While the evidence supporting this model is compelling and the results from our preliminary treatment trial were highly promising, we believe a placebo-controlled, parallel-group trial is the necessary next step to validate this approach prior to embarking on a larger trial that, if successful, could lead to a fundamental change in our understanding and treatment of antipsychotic-resistant delusions. This potentially high-impact approach involves an inexpensive generic medication that is extremely safe and well-tolerated outcome demonstrated by Turkington, Kindgon and colleagues (9) of a six-session CBT protocol administered by psychiatric nurses.

Innovation:

Memory consolidation and thalamocortical dysfunction in schizophrenia: Prior to Dr. Goff's recent move to New York University Medical Center (NYUMC), his group at the Massachusetts General Hospital (MGH) and others identified a selective impairment of memory consolidation in schizophrenia associated with abnormal thalamocortical connections and with a failure to activate ventromedial prefrontal cortex. These deficits, in turn, are highly correlated with persistence of delusions in medicated and unmedicated subjects. Dara Manoach (MGH) demonstrated a failure to consolidate learning of a procedural memory task in schizophrenia subjects which is highly correlated with abnormalities of sleep spindles and with psychosis (10, 11). Sleep spindles are generated by the thalamus and are necessary for transfer of information to the prefrontal cortex for memory consolidation. Ferrarelli and colleagues (12) also reported a marked deficit in sleep spindles in schizophrenia that is unrelated to medication status and highly correlated with psychotic symptoms. More recently, this group demonstrated a selective slowing in prefrontal/thalamic oscillations that was highly correlated ($r=0.62$) with delusions (13). Daphne Holt (MGH) demonstrated that individuals with schizophrenia exhibit a selective failure to consolidate extinction memory (14) which highly correlates with a failure to activate ventrolateral prefrontal cortex (15). This deficit was unrelated to medication status and was highly correlated with delusions. Hence the failure to transfer learning to prefrontal cortex during memory consolidation and to access this information appropriately appears to play a role in the persistence of delusions. The improvement of delusions by enhancement of memory consolidation would represent important evidence that this association is causal.

DCS facilitates memory consolidation: In animal models a similar deficit in memory consolidation is produced by administration of an NMDA antagonist in the ventromedial prefrontal (infralimbic) cortex (16). Following injection of the NMDA antagonist, CPP, into the prefrontal cortex, mice successfully extinguish fear conditioning but fail to retain extinction learning when examined after 24 hours (16); this finding of preserved extinction and failure to consolidate extinction learning is identical to the pattern exhibited by schizophrenia subjects compared to healthy controls (14). DCS, which is a potent positive modulator of NMDA receptors containing the NR2C subunit, markedly enhances consolidation of extinction memory. DCS has twice the activity at the subpopulation of NR2C-containing NMDA receptors compared to D-serine and glycine and 50% activity at other NMDA receptors (17, 18). A recently-developed NR2C knockout mouse is behaviorally normal except for deficits in memory, including consolidation of fear extinction learning (19). Whereas NMDA receptor density (represented by the obligatory NR1 subunit) is decreased postmortem in schizophrenia prefrontal cortex, only the NR2C subunit is selectively reduced in schizophrenia compared to bipolar disorder and healthy controls (20).

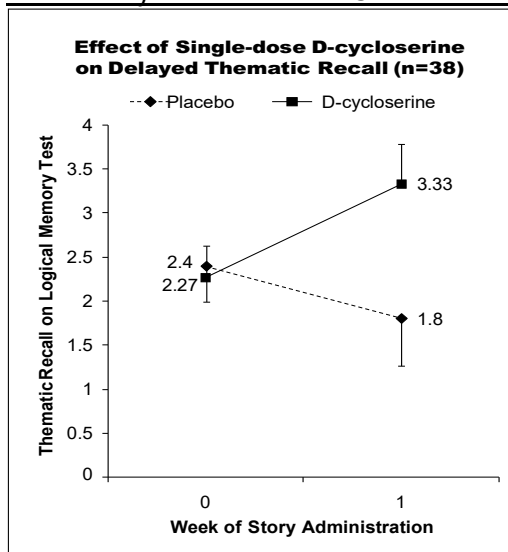
When administered within one hour after extinction training, DCS increases 24 hour retention by three-fold, but tolerance rapidly develops with daily dosing (21). When administered prior to training, DCS has no effect on performance during training—enhanced performance is only

observed when animals are tested 24 hours later. Importantly, DCS facilitates consolidation of novel learning only. For example, if an animal has previously received extinction training, DCS has no effect on subsequent extinction training (22). In addition, DCS does not just enhance new inhibitory learning; it appears to “erase” the previous fear conditioning learning as reflected by endocytosis of synaptic AMPA receptors which are expressed during fear conditioning (23). Based on the robust finding of enhancement of fear extinction training in animals, DCS has been used as an adjunct to CBT for anxiety disorders and has consistently demonstrated enhanced efficacy relative to placebo when administered within one hour of training (24, 25).

DCS effects on cognitive flexibility: Agonists at the glycine site of the NMDA receptor may also enhance cognitive flexibility. In an 8-week placebo-controlled daily-dosing trial of DCS 50 mg/d, we found that DCS significantly enhanced temporal lobe activation during a verbal fluency task (26). Tsai and colleagues found a reduction in perseverative errors on the Wisconsin Card Sort Task following daily dosing with D-serine (27). In animal models, D-serine significantly enhanced “reversal learning” in rats as measured by the time required to find the platform in the Morris Water Test after the location of the platform was moved (28).

Preliminary study of CBT for delusions: We conducted a 16 week randomized parallel group controlled feasibility trial of CBT for delusions in 30 schizophrenia patients who exhibited psychotic symptoms despite optimal pharmacological treatment and recorded a 93% study retention rate; 75% of subjects attended all 16 sessions (29). The manualized CBT protocol, developed by Dr. Gather (MGH) included cognitive restructuring, exposure, and coping skills. Compared with a psychoeducational control condition, the effect size of CBT for delusions was 0.18, which was not statistically significant. This study demonstrated the feasibility of retaining subjects with refractory psychosis in a CBT trial and sustained the relatively small effect of CBT for delusions in the absence of pharmacologic augmentation.

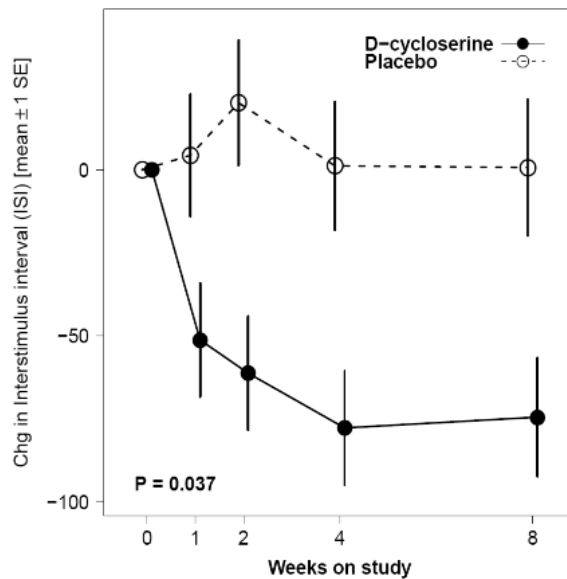
Preliminary studies of DCS for memory consolidation: In two studies we demonstrated enhancement of memory consolidation in schizophrenia subjects with DCS. In one study, a single dose of DCS 50 mg significantly improved 7-day thematic recall on the Logical Memory Test compared to placebo (figure, left) (30). We selected this measure because consolidation of narrative themes represents a cognitive function particularly relevant to CBT for delusions and because memory consolidation is known to solidify the “gist” of new learning (31). As shown in the figure, DCS may have enhanced cognitive flexibility since, unlike the DSC group, the placebo group displayed worsening in 7-day recall of story B compared to baseline. Interference in learning story B may have resulted from the administration of story A one week earlier at baseline.



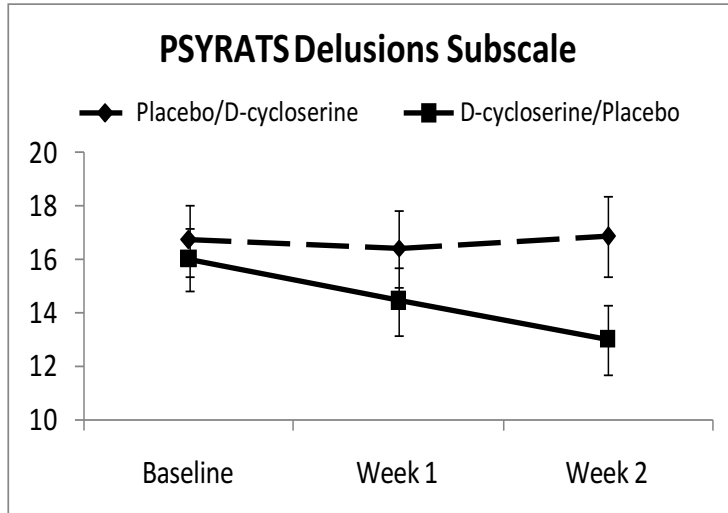
Recently we combined once-weekly DCS 50 mg with the Brain Fitness cognitive remediation program which emphasizes auditory discrimination training, in 38 schizophrenia subjects. Compared to placebo, DCS significantly improved learning of the auditory discrimination task (figure 2, below). However, enhancement of learning with DCS did not generalize to cognitive domains that were not practiced; performance on the MATRICS battery did not improve from baseline ($p>0.1$). Of note, with weekly dosing of DCS, learning on the auditory discrimination task did not plateau until week 4 and no loss of efficacy was observed over the 8 week trial. No adverse effects were reported. This study replicates the

Study #:S12-02991

finding that DCS enhances learning in schizophrenia subjects and extends the finding to an 8 weekly, once-weekly dosing schedule.



Preliminary study of DCS facilitation of CBT for delusions: We also conducted a random-order, counter-balanced, two session placebo-controlled trial of DCS facilitation of CBT for delusions (32). Twenty-one schizophrenia subjects with medication-resistant delusions despite adequate treatment with an antipsychotic other than clozapine received DCS 50 mg or placebo one hour prior to each of two sessions of CBT. The CBT exercise consisted of an alternative beliefs task developed by Jennifer Gottlieb (MGH) which trained subjects to formulate alternative explanations for a series of benign social vignettes leading up to the formulation of alternative explanations for their delusional belief. Twenty subjects (95%) completed the two CBT sessions. Subjects who received DCS first exhibited a large reduction (effect size 0.8) in PSYRATS measures of delusional severity (figure 3, below) compared to subjects who received placebo first ($p < 0.05$). Consistent with the finding in animal studies that effects of DCS are limited to memory consolidation and are only observed after a delay of 24 hours, DCS did not affect the number of responses generated during the first alternative beliefs exercise. This study establishes the feasibility of a DCS augmentation of CBT and the results are consistent with animal studies that demonstrate that DCS only enhances learning during the first exposure to a new form of training. However, this result requires confirmation from a parallel group design as we are now proposing.



Summary of Innovation: This study builds upon recent findings in translational neuroscience supporting a role for deficits in thalamo-cortical connections and memory consolidation in the persistence of delusions and follows from recent data demonstrating that, at low concentrations, DCS is a highly potent and selective agonist at a subpopulation of NMDA receptors involved in memory consolidation. This subgroup of NMDA receptors appears to be specifically down-regulated in schizophrenia. Although DCS has been well-established as an effective augmentor of CBT for anxiety disorders, this study (and our pilot trial) represents the first application of this approach for delusions. We are also the first group (MGH) to examine memory consolidation and fear extinction in schizophrenia and the first to examine measures of memory consolidation and cognitive flexibility as potential mediators of DCS effects when combined with CBT. If positive, this study could lead to an entirely novel approach for the treatment of delusions.

Characteristics of the Research Population:

Number of Subjects: Total planned enrollment of randomized subjects for the study is 64 subjects since this will ensure sufficient statistical power to detect any effects. In order to achieve adequate enrollment, we will consent and screen up to 140 individuals which will allow for a 40% screen failure rate. 77 participants will be enrolled for a total of 44 randomized subjects at NYU, and 33 participants will be consented at Sheppard Pratt for a total of 20 randomized participants. Additionally, 30 age and gender matched healthy subjects will be consented for the MRI portion of the study in order to enroll 15 healthy subjects.

The recruitment of 15 healthy subjects will be used as a comparison group against schizophrenia patients when analyzing fMRI scans. Due to the innovative nature of the tasks patients are doing while in the scanner, the addition of healthy participants will allow us to better identify any neural differences in these two groups, specifically to see if D-cycloserine enhances connectivity in specific neural regions.

Age of Subjects: Individuals between the ages of 18-68 will be included. Children below age 18 will not be included in this study. Schizophrenia is primarily an illness of adulthood and we would first like to explore the effectiveness of this intervention on adults. Healthy controls will have no history of mental illness.

Gender: Patients enrolled in this study will reflect the population of schizophrenia patients treated at each New York University Langone Medical Center and Sheppard Pratt Hospital. The estimated rate of females among chronic schizophrenia patients is 35% at both sites.

Racial and Ethnic Origin: Minorities will be included in this project in proportion to their representation in the New York City segment of the population served by Bellevue Hospital and the Baltimore segment of the population served by Sheppard Pratt Hospital. The targeted demographic profile for this study is 70% White or Caucasian, 26% Black or African American and 4% Asian or multi-racial with at least 17% identifying as Hispanic. Dr. Goff and the DSMB will monitor inclusion rates of women and minorities. We will target a female enrollment rate of

Study #:S12-02991

at least 35%. If the inclusion rates are not representative, Dr. Goff will implement corrective action including the establishment of additional referral sources as needed.

Inclusion and Exclusion Criteria: Subjects will be adult outpatients with schizophrenia, delusional disorder or schizoaffective disorder, ages 18-68, treated with any antipsychotic agent except clozapine at an adequate, stable dose for at least 8 weeks, or antipsychotic medication naïve within the last six months. Since a significant population of psychotic patients are often unable or unwilling to take antipsychotic medications, and these patients would also benefit from D-cycloserine augmented CBT, we will enroll antipsychotic medication-free patients only if they are unable or unwilling to take antipsychotic medication. To be eligible, subjects must have persistent delusions as defined by a score of at least 3 (moderate) on the Scale for Assessment of Psychotic Symptoms (SAPS) global delusion score at two assessments, four weeks apart.

However in some study participants, the SAPS may not capture all relevant symptoms related to the participant's delusions which are a requirement of enrollment. If this occurs, the study PI and study psychologist will confirm the subject's diagnosis and appropriateness for the study by completing the Structured Clinical Interview for DSM-IV Axis I Disorders (SCID). With the subject's consent, the study team will also confirm the symptoms of the subject with his/her current treatment provider to further ascertain appropriateness for the study.

Additionally, subjects must be free of serious or unstable medical illness, including renal impairment (creatinine greater than 1.4), seizures, dementia, anemia, and cannot be pregnant or nursing or have abused substances in the previous 6 weeks. Subjects with renal insufficiency are excluded because DCS is excreted unchanged in the urine; anemia is excluded because it is a rare side effect of DCS. A urine toxic screen and pregnancy test (in females) must be negative at screening. Subjects treated with clozapine will be ineligible because we and others have found that DCS worsens negative symptoms when added to clozapine (33).. Subjects treated with any SSRI will be ineligible because the PI has reviewed literature which posits that SSRI's prevent the effects of D-cycloserine on learning.

Healthy subjects will be adults between ages 18-67 with no history or current status of mental illness. In addition, healthy subjects will have normal intellectual functioning and not have any MRI contraindications.

Targeted/Planned Enrollment Table:

Ethnic Category	Sex/Gender		
	Females	Males	Total
Hispanic or Latino	2	4	6
Not Hispanic or Latino	19	35	54
Ethnic Category: Total of All Subjects *	21	39	60
Racial Category			
American Indian/Alaska Native	0	0	0
Asian	1	1	2
Native Hawaiian or Other Pacific Islander	0	0	0
Black or African American	5	11	16
White	12	30	42

Study #:S12-02991

Racial Categories: Total of All Subjects*	18	42	60
---	----	----	----

* The "Ethnic Category: Total of All Subjects" must be equal to the "Racial Categories: Total of All Subjects."

Methods and Procedures:**Approach:**

Study #:S12-02991

Location: The study will be conducted at two sites. The primary site will be Dr. Goff's lab within the Adult Psychiatry Clinical Trials Research space on the 8th floor of One Park Avenue and at the Bellevue Hospital Outpatient Psychiatry Department on the 2nd fl of the C & D building. Study drug will be dispensed by the NYU Research Pharmacy. Subject recruitment will occur at Bellevue Hospital. If additional subjects are required, the study will also be conducted at the Nathan Kline Institute for Psychiatric Research (NKI) which recruits subjects from the 520-bed Rockland Psychiatric Center on the NKI campus and from a network of affiliated mental health clinics. Dr. Goff is Vice Chair for Research in the Department of Psychiatry at NYU and Director of the Schizophrenia Program at NYU/Bellevue as well as Director of the NKI.

The second site will be the Sheppard Pratt Hospital in Baltimore, Maryland. The study will be carried out by the Stanley Research Program within Sheppard Pratt Hospital. Dr. Faith Dickerson, PhD, will be the site PI. Healthy subjects will be recruited from the NYU campus using web based advertisements and flyers posted in NYU buildings, including in waiting rooms and bulletin boards at the medical center.

Screening: An initial diagnostic assessment will be performed by a research psychiatrist or psychologist using the Structured Clinical Interview for DSM-IV (SCID), all medical records, and interviews with family and clinicians. The diagnosis will be confirmed at a weekly consensus diagnostic conference chaired by Dr. Goff which will also serve to verify adequacy of antipsychotic treatment. A medical review, physical examination and routine laboratory tests including electrolytes, creatinine, BUN, glucose, liver enzymes, calcium, phosphate, magnesium, albumin, CBC and urinalysis will be completed to identify unstable medical illness. A urine toxicology screen and, in females, a pregnancy test, will also be obtained. Additionally, DNA will be taken at this visit in order to analyze BDNF Val66Met polymorphism.

Healthy subjects will be screened using the SCID, Peters Delusion Index to rule out history or current mental illness or delusional thinking. Additionally, a WAIS-IV will be performed to exclude participants with below standard IQ.

CBT/DCS Protocol: Individual manualized CBT sessions will last 50 minutes and will be offered weekly for 12 weeks. The first two sessions will focus on engagement, characterization of delusions, and formulation of a treatment plan. Sessions 3 & 4 will consist of an alternative beliefs exercise as employed in our pilot trial in which subjects are trained to produce as many explanations as possible for a series of benign social vignettes (e.g., a taxi driver is staring at you), culminating with their own delusional belief (32). The exercise is structured with a fixed set of vignettes which allows us to record the number of alternative explanations generated by the subject during the exercise as a measure of cognitive flexibility. The remaining 8 sessions will include exploration of antecedents of delusional beliefs, development of a normalizing rationale, cognitive restructuring, testing of explanatory models, and review and consolidation. Study drug (DCS 50 mg or matching placebo) will be administered orally, 1 hour prior to sessions 3-12. Placebo will be administered prior to sessions 1 & 2. DCS and matching placebo capsules will be formulated by the NYUMC research pharmacy.

If a subject misses three consecutive study visits and corresponding CBT sessions, he/she will be withdrawn from the study. These missed visits and sessions may compromise the integrity of the study data and adversely affect the therapeutic alliance which is important in the CBT treatment.

CBT will be provided by Dr. Iruma Bello, a PhD psychologist who trained under Dr. Cather at MGH using manualized CBT protocols. A second PhD level therapist may be added and will have at least one year of intensive CBT training and experience working with psychosis. To ensure that there will be no effect due to CBT therapist, balance will be maintained such that each therapist will be assigned to the same number of subjects assigned to DCS as to placebo. This will be accomplished by a statistician independent of the study. Before initiation of the study, training will be provided by Dr. Cather who will also provide weekly supervision by video-

Study #:S12-02991

conference and will monitor treatment fidelity by evaluating audiotapes from 20% of sessions using the Cognitive Therapy for Psychosis Adherence Scale (CTPAS) (34). Dr. Cather recently participated in the development of the CBT protocol and conducted trainings for the NIMH-funded RAISE Study. Competency will be defined as a score of at least 10 on the CTPAS. Competency must be achieved by each study therapist with a pilot case prior to participation in the study and must be maintained throughout the study. Additional training will be provided if CTPAS scores drop below 10.

Rationale for DCS/CBT protocol design: The design of our protocol for combining DCS with CBT follows from several findings: 1.) DCS effects may decrease over time with repeated dosing (21) although we believe that once-weekly dosing reduces the risk of tachyphylaxis, 2.) DCS is effective for novel learning only (22) and 3.) we previously found large effects when DCS was combined with the first of two sessions of an alternative beliefs exercise which was designed to exploit DCS effects on memory consolidation and cognitive flexibility (32). Therefore, we will begin with two introductory sessions unaccompanied by study drug; study drug will first be administered with an alternative beliefs exercise at session #3. We will then continue weekly study drug administration one hour prior to CBT sessions and will follow session #3 with a second session of the alternative beliefs exercise in order to replicate the intervention provided in our pilot trial (two consecutive sessions of an alternative beliefs exercise) since delusions continued to improve following the second exposure to the exercise. The remaining 8 sessions will consist of standard CBT approaches to delusions to consolidate therapeutic effects.

We selected a 12 session CBT protocol to ensure an adequate CBT intervention in the placebo group consistent with clinical practice. Because our treatment is focused on delusions and their behavioral consequences, 12 sessions are adequate whereas longer treatment, such as the 16 session protocol that we developed for our CBT monotherapy trial, may be necessary for treatments that address the full range of schizophrenia symptoms. In support of a brief treatment, Turkington, Kingdon and colleagues (9) developed a 6-session CBT protocol which demonstrated efficacy compared to treatment as usual in a sample of 442 schizophrenia subjects (9). In studies of DCS augmentation of CBT for anxiety disorders, DCS efficacy compared to placebo was greatest in trials of short duration (2-5 weeks) compared to longer trials, possibly reflecting tachyphylaxis or a ceiling effect due to the efficacy of CBT administered alone for anxiety disorders (24). Because of the relatively small effect of CBT for delusions we do not expect to encounter a ceiling effect. Subjects will receive 10 weekly doses of DCS in our proposed trial and we will track the response of delusions over this time frame. In our 8 week study of DCS facilitation of cognitive remediation, enhancement of learning plateaued between weeks 4-8; we anticipate a similar pattern with DCS facilitation of CBT. Our previous manualized CBT protocols were developed by Dr. Corinne Cather based on manuals and casebooks produced by Tarrington and Kingdon (35). Dr. Iruma Bello and Dr. Cather will develop a manualized CBT protocol based on these resources with consultation from Dr. Kingdon. Other texts such as those produced by Fowler and colleagues (36) Chadwick and colleagues (37) and Nelson (38) will also be used.

Rationale for DCS Dosing: DCS was developed as an antibiotic and used at doses of 500-2000 mg/d for tuberculosis but is now rarely used due to psychiatric and neurological side effects (activation, confusion and seizures) at these doses. It has a serum half-life of 4 hours and maximum CSF concentrations equal to 80% of serum concentrations are achieved 2 hours after oral dosing (39). It has been well-tolerated and effective at single weekly doses of 50 to 500 mg/d in subjects with anxiety disorders (40). A recent review found that DCS doses of 50-100 mg were most effective in anxiety disorder subjects provided they were administered within one hour of CBT sessions (25). In a placebo-controlled dose finding trial, we found that daily dosing with DCS 50 mg for two weeks improved negative symptoms and working memory, whereas doses of 15 mg and 250 mg daily were ineffective (41). In three trials in which DCS 50 mg was administered as a single dose or once-weekly, we found enhancement of memory and enhancement of CBT efficacy for delusions without adverse effects (32, 42). At doses above 100 mg/d DCS has been associated with worsening of psychosis in two studies (43, 44) and in

Study #:S12-02991

a study of DCS 50 mg daily we observed worsening of delusions in one subject who achieved unusually high serum concentrations (45). Recent studies by Dravid and colleagues (17)

Study #:S12-02991

suggest that at low concentrations DCS acts as a high-potency selective agonist at NR2C-containing NMDA receptors which mediate memory consolidation; at higher concentrations DCS binds as a partial agonist at NR2A and NR2B subunits which may contribute to worsening of psychosis in some patients. Taken together, it appears that the optimal dose of DCS is approximately 50 mg; unlike subjects with anxiety disorders, schizophrenia subjects appear at risk for worsening of psychosis at higher doses. Given the extensive prior experience and the risk that higher doses will worsen psychosis, we do not believe a dose finding trial is necessary and do not believe exposing schizophrenia subjects to a higher dose is justified. In addition, DCS serum concentrations will not be informative in this once-weekly dosing trial since steady state will not be achieved and given the variability in T_{max}.

IND Exemption:

D-cycloserine is approved and has been used for over 50 years at oral daily doses of 500 mg to 1000 mg for the treatment of tuberculosis although it is now a second-line therapy. Dr. Goff has been studying D-cycloserine 50 mg/d in schizophrenia for over 20 years [51], including 6 published randomized placebo-controlled trials [52-57]; no serious adverse events have been observed except for worsening of negative symptoms when added to clozapine [58, 59], which is excluded in the current trial. The IRB at the Massachusetts General Hospital, where these studies were performed, agreed with Dr. Goff's judgment that an IND was not necessary. Since then, D-cycloserine 50 mg- 100 mg has been widely used in psychiatry for facilitation of cognitive behavioral therapy for anxiety disorders and to reduce drug craving—more than 15 studies have been published without any serious adverse effects identified [60]. In the current study oral D-cycloserine 50 mg or placebo is administered once-weekly for 9 weeks, prior to cognitive behavioral therapy sessions. Given the safety record of D-cycloserine in individuals with schizophrenia and other psychiatric disorders and the fact that single doses of 50 mg are only 10% of the lower range of doses used for treatment of tuberculosis, we believe this study does not increase the risk of the drug or the acceptability of risks of the drug. In addition, results will not be used to support a new indication, a change in labelling, or a change in advertising of the drug. On January 23, 2013, Steve Hardemann of the FDA responded to a letter from Dr. Goff as follows: "...Thus, the determination of whether an investigation is exempt is yours to make, and on its face your study does appear to be exempt." Dr. Goff did not proceed to formally request an exemption by completing an IND application but will if requested by the IRB.

DNA Collection, Extraction, Genotyping:

Blood will be collected from all subjects at the screening visit for DNA analysis. DNA extraction, genotyping, and storage will occur at the NYU CORE lab. DNA samples will be labeled with a numeric ID only and delivered same day to the lab. Subjects have the option during consent to allow the biological materials (DNA) collected from them to be stored and possibly made available to qualified researchers in the scientific community. Other researchers who may conduct research on the DNA collected from this research project will not have access to any confidential information, and any data shared will contain no PHI, but subject codes. The samples will have a random code and other researchers will have no access to the link. Only this research group will have the link in a password protected database.

Subjects may request at a later date, through the study PI, that their genetic material not be used for this study. If the subject decides to withdraw permission to use the DNA for this study,

Study #:S12-02991

the NYU CORE lab will be notified of the request to remove the subject's genetic material by using the assigned code number.

DNA extraction will be performed immediately after samples are obtained, and DNA will be frozen and stored at -80 C until genotyping is conducted. BDNF Val66Met genotype (rs6265) will be determined with the Sequenom MassArray Platform (Sequenom Inc, San Diego, CA) using primers that have been previously described [55].

Given the low frequency of Met/Met genotype (3.3% in HapMap Caucasian subjects), Met/Met and Met/Val subjects will be combined into a Met carrier group for statistical analysis, which will contrast Val/Val and Met carrier subjects on response to clinical measures and gray matter volumes. To control for the possibility of population stratification artifact, race will be entered as a co-variate in genetic analyses.

MRI Imaging, Data Acquisition, Processing and Analysis Methods:

Theories of systems-level memory consolidation propose that memories are initially encoded in the hippocampus and then are distributed across cortical regions over time (51,52). In accordance with this theory, successful long term memory in healthy adults has been linked to increased connectivity between the hippocampus and prefrontal cortex (53). Thus, the impairment in memory consolidation in schizophrenia patients might be explained by a corresponding decrement in hippocampal-prefrontal connectivity.

MRI images will be performed at the NYU site only. Two MRIs without contrast will be performed as part of the study, before and after the first administration of D-Cycloserine. Specifically, each of these will consist of the acquisition of a high-resolution MPRAGE scan that will be motion corrected (Siemens 3 T Skyra, resolution 1 mm³, slices = 192, TR = 23000 ms, TE = 2.96 ms, FA = 9°, FOV = 256mm) to create a single image volume with high contrast-to-noise, a T2-weighted anatomical image (1mm³, slices = 192, TR = 3200 ms, TE = 422 ms, FOV = 256mm). In addition, several scans will be acquired that will allow us to determine whether D-cycloserine-induced changes in cortical thickness and hippocampal volume in the patients are accompanied by related changes in connectivity of the frontal cortex and hippocampus (measured using diffusion tensor imaging (DTI), and resting-state, functional connectivity MRI (FcMRI)). Thus, one 6 minute, 18 second long fcMRI scan (TR = 2500 msec; TE = 30 msec; flip angle = 90; slices = 46, 3 mm³ voxel size), and one approximately 9 minute DTI scan (TR/TE=7000/82 ms, b=700 s mm⁻², 256x256 mm FOV, 128x128 matrix, 2 mm slice thickness, 10 T2 + 60 DWI) will be acquired. We will also collect fMRI data to examine whether d-cycloserine has effects on the function of hippocampal subfields. Subjects will look at stimuli presented via projector on a screen placed at the back of the bore. They will be instructed to indicate, using a button response box, how the stimuli relate to categories and/or one another. During fMRI, subjects will complete 5 blocks of 90 trials, during which time we will collect Blood Oxygen Dependent Level (BOLD) data (5 x 184 x 19 slices, TR = 2000 ms, TE = 30 ms, Flip Angle = 70°). This task will take approximately 30 min. During anatomical scans, subjects will be asked to merely keep their eyes open and relax. During fMRI and FcMRI subjects will be asked to view images and make choices (former) and rest with their eyes open and focused loosely on the screen. The entire scan will take approximately 1 ½ to 2 hours.

Assessments:

The SAPS will be performed at screening and four weeks later at baseline to identify subjects with persistence of sufficient delusional severity to enter the study. The clinical assessment battery will be comprised of the Psychotic Symptom Rating Scale (PSYRATS), Brief Psychiatric Rating Scale (BPRS), Scale for Assessment of Negative Symptoms (SANS), Calgary Depression Scale for Schizophrenia (CDSS), Clinical Global Impression (CGI) and Heinrich's Quality of Life Scale (QLS). We are using this broad battery of tests to compare treatment groups at baseline and to allow exploratory analyses to help us understand the effects of DCS on our primary outcomes and to guide selection of outcome measures for future studies. The clinical battery will be administered at baseline and weeks 4, 8, 12, 24, and, in most subjects,

Study #:S12-02991

36. In addition, the Delusion Subscale of the PSYRATS will be administered at weeks 2, 3, 6 & 10 to assess the time course of DCS effects on delusions. Ratings will be performed prior to the DCS/CBT session each week. Additionally, the Drug and Alcohol Questionnaire and the Pittsburgh Sleep Quality Index will be performed at baseline. Assessment of cognitive functioning will be measured by the MATRICS battery at baseline only since we have found no effect of daily or weekly DCS administration on this and similar cognitive batteries in four randomized controlled trials. The SAFTEE will be administered at weeks 3, 4, 8, and 12. The SAFTEE is a self-report measure on which the participant rates the severity of 55 side effects on a 4 point Likert Type scale. The scale also asks the participant to furnish an overall rating on the severity of all side effects. Participants will be followed regardless of whether they remain on study drug or comply with CBT sessions to permit an unbiased causal estimate of D-cycloserine treatment efficacy under the intention-to-treat principle. Additionally, a per protocol analysis will be performed using data only from subjects who have substantially complied with the protocol.

The Psychotic Symptom Rating Scale (PSYRATS) (46): The PSYRATS is a well-validated 17 item, 5 point likert scale which contains 6 items that characterize delusions according to 1.) Amount and duration of preoccupation, 2.) Conviction, 3.) Amount and intensity of distress, and 4.) Disruption to life. We selected the PSYRATS because it frequently has been used in CBT studies, because we previously demonstrated that it captured response to DCS augmentation of CBT, and, unlike the Positive and Negative Syndrome Scale (PANSS), Scale for Assessment of Psychotic Symptoms (SAPS) and the Brief Psychiatric Rating Scale (BPRS), it rates subjective distress and the degree to which delusions disrupt life, both of which are important targets of CBT. Factor analysis has revealed three dimensions (47): preoccupation, conviction and distress/life disruption, with modest correlation between preoccupation and conviction ($r=0.28$) (46). We will use the Delusion Subscale total score as our primary outcome measure and, as an exploratory analysis, we will look at the three domains to further characterize DCS effects.

Assessment of memory consolidation: The Logical Memory Test of the Wechsler Memory Scale (WMS-III) will be used to measure memory consolidation, as in our pilot trial (30). Subjects will be administered Story A one week prior to baseline and will be tested on immediate item and thematic recall. One week later, at baseline they will be tested on 7-day delayed thematic recall. At week 3, one hour after administration of study drug, (immediately before the CBT session), participants will be administered story B, which will be tested for delayed thematic recall immediately prior to administration of study drug the following week. This will allow us to assess the effect of the first dose of DCS on memory consolidation following the same protocol that we used successfully in a previous study (30). The 2-week gap between administration of story A and story B will minimize potential interference between tests.

Assessment of cognitive flexibility: As our primary measure of cognitive flexibility we will record the number of alternative explanations generated by the subject during the Alternative Beliefs Exercises at weeks 3, 4 & 12. This measure of cognitive flexibility is most relevant to our primary outcome, improvement of delusions. We will not administer this measure at baseline because the exercise is an essential element of the treatment; it will be first administered at week 4 following the first dose of DCS (or placebo) to maximize the treatment effect of the first dose of DCS. We have previously shown that performance on the Alternative Beliefs Exercise is not affected when measured immediately after DCS administration (32), so the week 2 administration will serve as a baseline value. This is consistent with the large body of evidence from animal and human studies that DCS does not affect performance during training—effects are only detectable after a 24 hour delay. Given that other measures of cognitive flexibility (and negative symptoms) improved in previous studies after 8 weeks of daily dosing with DCS (26) or

Study #:S12-02991

D-serine (27), we will repeat the Alternative Beliefs Exercise at week 12 to assess a possible delayed improvement of cognitive flexibility. We will also examine perseverative errors on the Wisconsin Card Sort Test and performance on the Verbal Fluency Test at baseline and week 12 as measures of cognitive flexibility unrelated to training.

Rater Training: Training on rating scales will be provided by Drs. Cather and Bello and will require agreement ($r = 0.85$) with gold-standard measures on each of 10 videotaped ratings of the SAPS, PSYRATS, SANS, BPRS, CDSS and QLS as well as observed rating interviews conducted by the trainee. Inter-rater reliability and fidelity with gold –standard measures will be maintained with quarterly training which will consist of review of ratings on taped interviews conducted as part of the study. Raters will be required to maintain a criterion of $r = .8$ correlation with the gold-standard ratings. Raters who demonstrate drift from these standards will receive additional training and must demonstrate improvement before recommencing study ratings. Training and certification for administration of the MATRICS cognitive battery will be provided by Dr. Lila Davachi.

Study Visits for Healthy Subjects

Screening (Visit 1)

Healthy subjects will be consented and screened for eligibility. Then, participants will perform a logical memory test, immediate recall test, and 25 minute recall. Participants will also perform a verbal fluency test. KVL-T-R and BVMT-R and Rey Complex Figure test (RCFT).

Visit 2

Healthy subjects will perform the 1 week retrieval for the logical memory task, be given instructions for the scan test, and undergo the first set of MRI and fMRI scans.

Visit 3

Healthy subjects will complete the second set of MRI and fMRI scans.

Visit 4

Subjects will perform a memory recall task from the fMRI scan on visit 3 and a story segmentation task based on the fMRI scan on visit 2..

Data Analysis and Data Monitoring

Analysis Plan:

Primary: Between treatment differences in PSYRATS Delusions Subscale

Mean effects in week 12. The mean effect of DCS + CBT compared to Placebo + CBT on delusions will be tested using a linear mixed model repeated measures (MMRM) analysis of modified PSYRATS with fixed terms for study visit, DCS/Placebo treatment group and visit by treatment interaction. The model will include random subject effects. Baseline PSYRATS will be included in the model together with a baseline by treatment interaction term. The MMRM will assume a means model, an unstructured covariance matrix and parameter estimation will be based on restricted maximum likelihood. Because of the potential for failure of model convergence due to the requirement of a large number of interaction terms, only a few other covariates will be considered. These may include baseline SANS total score and demographic variables including age and gender, which may be predictive of outcome. To test treatment difference in change from baseline in PSYRATS, we will use a pairwise contrast on the terms for DCS treatment by visit interaction. To examine differential treatment effects at each CBT week if there is significant interaction, we will perform tests of simple main effects. A significantly larger 12-week decline in mean PSYRATS total score for subjects receiving DSC supports the

Study #:S12-02991

effectiveness of DCS. The probability of a type one error, the size of the test, will be controlled at 0.05, and all tests will be two-tailed. Overall alpha will be maintained at 0.05 for analyses of primary parameters. No adjustments will be made for multiplicity among secondary parameters analyzed. *Missing data*: The analysis will utilize the intention-to-treat principle. All randomized subjects, both completers and non-completers, will be included in the analysis without imputation. Estimates will be unbiased if follow-up data that are missing are “missing at random” conditional on the observed data and model assumptions.

Secondary: Characterizing onset and duration of effectiveness of DCS

a and b. Testing equality of the distributions of onset and duration. Onset is defined as the first week at which the criterion for response is met that lasts at least to the final week of the treatment period. Kaplan-Meier estimates of the survival distributions of time to onset of response will be obtained and the survival distributions will be compared using the log-rank test. The survival distribution, $H(t)$, is the probability that onset will occur after time t . Duration is defined as the number of weeks from onset to offset. Offset for a responder is the first week at which the criterion for response is no longer met. The treatment specific survival distributions of duration will similarly be compared using the log-rank test. The null hypothesis for the second primary outcome measure is that there is no difference between the two treatment groups in the proportion of subjects who are responders (defined as a 20% or greater reduction in PSYRATS Delusions Subscale total score) beginning in week two to the end of the treatment phase. The statistical analysis will be based on a repeated measures logistic regression model. The independent terms in the model will include the same variables and covariates used in the MMRM analysis. By the definition of responder, there cannot be any missing data.

c. Characterizing the population of responders. An alternative approach to characterizing onset when there is a substantial fraction of the population who will never respond is to use a “cure model” (48, 49). The approach was recommended by consensus, in a specially organized conference to develop an approach to measuring onset in GAD (49). The recommended model assumes that $H(t) = (1-p) + pS(t)$ where $H(t)$ is the survival distribution for the full population, $S(t)$ is the conditional survival probability of onset, given that onset will occur and p is the probability that onset will occur e.g., that the subject is a responder. In the parametric approach, the probability of response, p , will be modeled with a logistic and $S(t)$ will be modeled with a Weibull. The equality of the values of p for placebo and DSC will be compared using a likelihood ratio test. There are many tests of equal conditional survival distribution summarized in Laska (49). In the statistical models for p and for $S(t)$, baseline variables can be included as covariates. Those that are statistically significant and that have high influence will be used to identify the population of patients most likely to be responders in subsequent trials. Nonparametric estimates of the probability of response at the end of the treatment period and of the conditional survival distribution of onset for those achieving onset can also be obtained to confirm the results of the parametric model. The Laska-Meisner test will be used to test equal probability of achieving onset (50). (Remove the current 50 and change reference 51 to 50)

Tertiary: Identify mediators of delusional response to DCS

a. Memory Consolidation Hypothesis. The primary MMRM model described above will be used to test the predictive value of 7-day delayed thematic recall on the Logical Memory Test after the first dose of DCS. The baseline covariates in MMRM analysis of modified PSYRATS, will be augmented by the recall value. If statistically significant, a test of the hypothesis that the coefficient of the recall term in the model is not zero will demonstrate the predictive value of the measure. The magnitude of the coefficient is a measure of the degree to which the variable effects the PSYRATS score at week 12. The data for this analysis is restricted to those subjects randomized to DCS.

b. Cognitive Flexibility Hypothesis. Just as for the memory consolidation hypothesis, the primary MMRM model described above will be used to test the predictive value of change in the number of alternative beliefs generated during the first and second administrations of the Alternative Beliefs Exercise in the DCS group on the week 12 PSYRATS score. The baseline covariates in MMRM analysis of modified PSYRATS, will be augmented by the change score.

c. Synergy Hypothesis. The primary MMRM model described above will be used to test whether

Study #:S12-02991

positive outcomes on both the 7-day delayed thematic recall measure and the change in the number of alternative beliefs measure produces better outcomes on the week 12 PSYRATS score than would be expected by additivity. The analysis will also investigate whether outcomes needs to be positive in both variables for the DCS treatment to be effective. In this analysis, the model will contain both covariates and an interaction term. If the interaction term is statistically significant, the sign of the estimated coefficient will indicate synergy/antagonism and the magnitude of the coefficient together with the magnitude of the coefficients of the recall and change measure quantifies the joint effect on the PSYRATS week 12 score.

Data Storage and Confidentiality:

Data Management and Quality Assurance: The Principal Investigator has extensive experience in conducting clinical research studies. Based upon this experience, a comprehensive data management methodology combined with strong planning, control, coordination and quality assurance functions will be implemented. The primary objectives of the data management methodology are to insure the completeness, accuracy and overall integrity of the study data.

Easy to complete case report forms (CRFs) will be developed to standardized data collection across all subjects. Most CRFs will be forms used in previous studies that have a track record of successful data collection. The database will be maintained by the project coordinator, Iruma Bello, PhD and will be stored in her and the research assistant's office. To protect confidentiality, all information will be identified by code only. DNA samples will be encoded with a numeric study ID and the name of the patient will then be stored in a password protected database that only the research staff will have access to. DNA samples will be stored indefinitely at the NYU CORE lab. Subjects have the option during consent to allow DNA samples to be stored and used for future research related to schizophrenia. Genetic tests will not be shared with the participants or their clinicians, nor will the results be placed in the patient's medical records. Additionally, results from the study will not be released to participants. Data that are identifiable (consent forms, laboratory results, study roster and key) will be stored separately in a locked drawer in these offices. All databases will be password-protected. Coded information will be entered and stored in two secure, password-protected, databases. Subject name or any other personally identifying information will not be stored in either of the databases. Although subject date of birth will be entered into our database for administrative purposes, this information will not be viewable and will not be shared with anyone.

Once data collection has started, staff will review all data, with a special focus on the primary outcome measures and safety data. Missing fields and forms are reviewed to ensure that research staff are collecting data as instructed by the protocol, and entering data into the study database within the recommended time frame. All data are reviewed for accuracy, completeness and logical consistency. Inconsistencies are documented as data management queries. Trends in the queries are monitored and intervention strategies generated to resolve any data issue. Final data clean up will be completed shortly after the last subject visit and the study database will be locked and provided to the study statistician for analyses.

Study recruitment, enrollment, and retention will be monitored to insure that recruitment goals are attained. Study screening information is reviewed by comparing the characteristics of subjects enrolled versus screened out, assisting with potential strategies to improve study enrollment. The ratio of screened to randomized subjects is also reviewed to reflect the effort required to successfully enroll subjects in the study. Research study visit attendance will also be monitored on an ongoing basis.

Estimation of statistical power: We will enroll up to 100 subjects until 60 are evaluable, defined as completing the week 3 clinical assessment which is the first clinical assessment following study drug administration. Subjects will be randomly assigned to DCS or Placebo in a 1:1 ratio. This sample size provides greater than 85% power to detect an effect size of 0.8 in the two-group mean comparisons of 12-week PSYRATS Delusions Subscale total score, which we consider to be clinically-significant. We found an effect size of 0.8 for the difference between treatment groups in our two-session pilot trial and expect a substantially larger effect with 10 DCS-facilitated CBT sessions in this trial. Further, the sample size provides greater than 95%

Study #:S12-02991

power to detect a difference in the response rate of DCS vs placebo assuming the response rate for placebo is 10% and the response rate for DCS is 50%.

Risk/Benefit Assessment

Potential Risks: Loss of confidentiality regarding psychiatric or medical information is a possible risk for which precautions will be taken. Phlebotomy may cause soreness, bruising, bleeding and rarely, infection. A single dose of D-cycloserine 50 mg combined with CBT was well-tolerated in one cross-over study of 20 subjects exposed to drug and weekly dosing of D-cycloserine; 50 mg was well-tolerated in two studies of schizophrenia patients (n=38 subjects exposed to DCS) and in eight studies of anxiety disorder patients. However, it is possible that D-cycloserine could worsen symptoms of schizophrenia. At high doses (500-1,000 mg/d) used for the treatment of tuberculosis, D-cycloserine has been associated with confusion, lethargy, and seizures. High dose, long-term daily administration of D-cycloserine has also been associated with B12 and/or folate deficiency anemia (macrocytic anemia). It is also possible, although unlikely, that CBT will cause anxiety or agitation, insofar as it will entail participants examining their delusional beliefs.

Protection against Risks: All patients are carefully screened to avoid enrollment of anyone for whom study procedures or study drug would be potentially harmful, including subjects with unstable medical or psychiatric illness, recent substance abuse, anemia or insufficient renal function. Subjects will be evaluated by a physician during screening and after the last dose of D-Cycloserine. A complete blood count (CBC) will additionally be performed at screening and after the last dose of D-Cycloserine. If the hemoglobin drops below 13.5 mg/100 ml for male or 12 mg/100 ml for female subjects with a mean corpuscular volume (MCV) greater than 100 (macrocytic anemia consistent with B 12 or folate deficiency), the anemia will be evaluated and treated as clinically indicated. The Side Effect Checklist for assessment of drug side effects will be administered at every visit and neurological side effect scales will be completed monthly. All information will be identified by code only. Data that are identifiable (consent forms, laboratory results, study roster and key) will be stored separately. All databases will be password-protected. Study drug will be suspended if significant clinical worsening is detected by formal assessment or clinical observation. The CBT protocol will be designed to avoid the triggering of excessive anxiety or stress. In addition, a DSMB will review the protocol prior to initiation and will monitor safety during the trial.

Justification of Potential Risks: The risks associated with this trial are minimal and the potential scientific and clinical benefits are considerable. Appropriate safeguards will be in place to ensure that the risk does not outweigh the scientific benefit.

Potential Benefits to the Subjects: All subjects will receive a comprehensive psychiatric and medical evaluation and optimal clinical treatment as part of the NYU/Bellevue Outpatient Psychiatry Department. They will also receive a CBT intervention which is not routinely available. Subjects will be closely monitored for safety. It is possible that subjects randomized to d-cycloserine may benefit with an improved clinical course, although this potential benefit has not been established by prior research.

Subject Identification, Recruitment, and Consent/Assent

Method of Subject Identification and Recruitment: The research team's psychiatrists and psychologist are credentialed treating clinicians at the Bellevue Hospital outpatient clinic where these patients will be referred for or are currently receiving standard psychiatric care, and are part of the treating team for clinical care purposes. In order to further insure protection of privacy for this patient population, patients will not be contacted by any member of the research team unless they have agreed to hear about the research process beforehand from the initial treating clinician. Clinicians in the Outpatient Psychiatry Clinic, Inpatient Psychiatry Units and the Comprehensive Psychiatry Emergency Program at Bellevue Hospital will be provided with flyers

Study #:S12-02991

as well as a document listing the inclusion/exclusion criteria in order to facilitate recruitment. This will allow the initial treating clinicians to help identify potential subjects as well as provide interested individuals with basic study-related information. Potential subjects identified by the initial treating clinicians will be asked whether they would like to speak to a researcher. If the patient agrees, the initial treating clinician will then contact the study psychologist and they will determine whether the study clinician (psychologist or psychiatrist) will go and meet with the patient at that time or whether they will meet with the patient during the next scheduled clinic appointment. During this meeting, the study will be explained to the potential participant and in the event that the individual is interested in participating, s/he will then be given the consent form to read and review. In addition to the study consent form, participants will be given a second consent form asking them to participate in providing anonymous quotes to be used in the research group's website for advertisement purposes. All recruitment and screening will be conducted only during the face to face consenting process.

Process of Consent: Patients who are interested in participating in the study will meet with a doctoral level member of the study staff who will review the consent form and assess the patient's capacity for participation in research in accordance with our standardized research procedures. After signing consent, participants will be assessed for eligibility. Subjects will be remunerated for each study visit in amounts commensurate with the time commitment. Reimbursement will also be provided for participant travel expenses to and from the study site.

Subject Capacity and Subject/Representative Comprehension: Subjects capacity for participation in research will be reviewed by a doctoral level member of the study staff during the consenting process as well as by a psychiatrist on staff. The clinician obtaining the informed consent will take steps to ensure that the participant is capable of consenting and participating in the study. The clinician will ensure that the individual understands the content and procedures of the study, their rights as a participant, and their right to discontinue participation at any time. Individuals who are not able to demonstrate this level of comprehension will be excluded from participation. In addition, the potential subject must also achieve a perfect score on a ten-item true/false quiz that asks questions about the study procedure and potential risks. After the consent form has been signed, the subject will be provided with a letter detailing study-related information and contact information for key study staff.

Debriefing Procedures: The only information withheld from subjects is whether the type of medication they are on is active drug or placebo. Subjects will understand that they are blinded.

Consent Forms: Consent forms will be stored in a personal file with the patients initials. This file will be separate from any of the subject's study data in order to separate their data from any identifying information.

Costs to the Subject: There are no costs to the subject associated with the study. Transportation will be reimbursed. Study procedures are paid for via an NIMH grant.

Payment for Participation: Total compensation for the study will be \$400.00. Compensation breakdown is as follows:

- | | |
|---|-------|
| ▪ \$10 for visits 2, 4, 8, 9, 10, 12, 13, and 14 | \$80 |
| ▪ \$30 for the screening visit, visits 3, 7, 11, 15, 16, and 17 | \$210 |
| ▪ \$50 for the first MRI | \$50 |
| ▪ \$60 for visit 6 | \$60 |

TOTAL: \$400.00

Study #:S12-02991

Total compensation for healthy controls will be \$160.00. Compensation breakdown is as follows:

- \$30 for visit 1 and visit 4 \$60
- \$50 for visit 2 and visit 3 \$100

TOTAL: \$160.00

References

1. Laruelle M, Abi-Dargham A. Dopamine as the wind of the psychotic fire: new evidence from brain imaging studies. *J Psychopharmacol*. 1999;13(4):358-71.
2. Lieberman JA, Stroup TS, McEvoy JP, Swartz MS, Rosenheck RA, Perkins DO, et al. Effectiveness of antipsychotic drugs in patients with chronic schizophrenia. *N Engl J Med*. 2005;353(12):1209-23.
3. Malhotra A, Adler C, Kennison S, Elman I, Pickar D, Breier A. Clozapine blunts N-methyl-D-aspartate antagonist-induced psychosis: A study with ketamine. *Biological psychiatry*. 1997;42:664-8.
4. Tsai GE, Lin PY. Strategies to enhance N-methyl-D-aspartate receptor-mediated neurotransmission in schizophrenia, a critical review and meta-analysis. *Current pharmaceutical design*. 2010;16(5):522-37
5. Tiihonen J, Wahlbeck K, Kiviniemi V. The efficacy of lamotrigine in clozapine-resistant schizophrenia: a systematic review and meta-analysis. *Schizophrenia research*. 2009;109(1-3):10-4.
6. Dixon LB, Dickerson F, Bellack AS, Bennett M, Dickinson D, Goldberg RW, et al. The 2009 schizophrenia PORT psychosocial treatment recommendations and summary statements. *Schizophrenia bulletin*. 2010;36(1):48-70.
7. Wykes T, Steel C, Everitt B, Tarrier N. Cognitive behavior therapy for schizophrenia: effect sizes, clinical models, and methodological rigor. *Schizophrenia bulletin*. 2008;34(3):523-37.
8. Turkington D, Kingdon D, Weiden PJ. Cognitive behavior therapy for schizophrenia. *Am J Psychiatry*. 2006;163(3):365-73.
9. Turkington D, Kingdon D, Turner T. Effectiveness of a brief cognitive-behavioural therapy intervention in the treatment of schizophrenia. *The British journal of psychiatry : the journal of mental science*. 2002;180:523-7.
10. Manoach DS, Cain MS, Vangel MG, Khurana A, Goff DC, Stickgold R. A failure of sleep-dependent procedural learning in chronic, medicated schizophrenia. *Biological psychiatry*. 2004;56(12):951-6.
11. Manoach DS, Thakkar KN, Stroynowski E, Ely A, McKinley SK, Wamsley E, et al. Reduced overnight consolidation of procedural learning in chronic medicated schizophrenia is related to specific sleep stages. *J Psychiatr Res*. 2010;44(2):112-20.
12. Ferrarelli F, Huber R, Peterson MJ, Massimini M, Murphy M, Riedner BA, et al. Reduced sleep spindle activity in schizophrenia patients. *Am J Psychiatry*. 2007;164(3):483-92.
13. Ferrarelli F, Sarasso S, Guller Y, Riedner BA, Peterson MJ, Bellesi M, et al. Reduced Natural Oscillatory Frequency of Frontal Thalamocortical Circuits in Schizophrenia. *Archives of general psychiatry*. 2012.
14. Holt DJ, Lebron-Milad K, Milad MR, Rauch SL, Pitman RK, Orr SP, et al. Extinction memory is impaired in schizophrenia. *Biological psychiatry*. 2009;65(6):455-63.
15. Holt DJ, Coombs G, Zeidan MA, Goff DC, Milad MM. Failure of neural responses to safety in schizophrenia. *Archives of general psychiatry*. (in press).

16. Burgos-Robles A, Vidal-Gonzalez I, Santini E, Quirk GJ. Consolidation of fear extinction requires NMDA receptor-dependent bursting in the ventromedial prefrontal cortex. *Neuron*. 2007;53(6):871-80.
17. Dravid SM, Burger PB, Prakash A, Geballe MT, Yadav R, Le P, et al. Structural determinants of D-cycloserine efficacy at the NR1/NR2C NMDA receptors. *The Journal of neuroscience : the official journal of the Society for Neuroscience*. 2010;30(7):2741-54.
18. Sheinin A, Shavit S, Benveniste M. Subunit specificity and mechanism of action of NMDA partial agonist D-cycloserine. *Neuropharmacology*. 2001;41(2):151-8.
19. Hillman BG, Gupta SC, Stairs DJ, Buonanno A, Dravid SM. Behavioral analysis of NR2C knockout mouse reveals deficit in acquisition of conditioned fear and working memory. *Neurobiol Learn Mem*. 2011.
20. Beneyto M, Meador-Woodruff JH. Lamina-specific abnormalities of NMDA receptor-associated postsynaptic protein transcripts in the prefrontal cortex in schizophrenia and bipolar disorder. *Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology*. 2008;33(9):2175-86.
21. Parnas AS, Weber M, Richardson R. Effects of multiple exposures to D-cycloserine on extinction of conditioned fear in rats. *Neurobiol Learn Mem*. 2005;83(3):224-31.
22. Langton JM, Richardson R. D-cycloserine facilitates extinction the first time but not the second time: an examination of the role of NMDA across the course of repeated extinction sessions. *Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology*. 2008;33(13):3096-102.
23. Mao SC, Hsiao YH, Gean PW. Extinction training in conjunction with a partial agonist of the glycine site on the NMDA receptor erases memory trace. *The Journal of neuroscience : the official journal of the Society for Neuroscience*. 2006;26(35):8892-9.
24. Norberg MM, Krystal JH, Tolin DF. A meta-analysis of D-cycloserine and the facilitation of fear extinction and exposure therapy. *Biological psychiatry*. 2008;63(12):1118-26.
25. Rothbaum BO. Critical parameters for D-cycloserine enhancement of cognitive-behavioral therapy for obsessive-compulsive disorder. *Am J Psychiatry*. 2008;165(3):293-6.
26. Yurgelun-Todd DA, Coyle JT, Gruber SA, Renshaw PF, Silveri MM, Amico E, et al. Functional magnetic resonance imaging studies of schizophrenic patients during word production: effects of D-cycloserine. *Psychiatry research*. 2005;138(1):23-31.
27. Tsai G, Yang P, Chung L-C, Lange N, Coyle J. D-Serine added to antipsychotics for the treatment of schizophrenia. *Biological psychiatry*. 1998;44:1081-9.
28. Labrie V, Duffy S, Wang W, Barger SW, Baker GB, Roder JC. Genetic inactivation of D-amino acid oxidase enhances extinction and reversal learning in mice. *Learn Mem*. 2009;16(1):28-37.
29. Cather C, Penn D, Otto MW, Yovel I, Mueser KT, Goff DC. A pilot study of functional Cognitive Behavioral Therapy (fCBT) for schizophrenia. *Schizophrenia research*. 2005;74(2-3):201-9.
30. Goff DC, Cather C, Gottlieb JD, Evins AE, Walsh J, Raeke L, et al. Once-weekly d-cycloserine effects on negative symptoms and cognition in schizophrenia: An exploratory study. *Schizophrenia research*. 2008;106(2-3):320-7.
31. Budson AE, Todman RW, Schacter DL. Gist memory in Alzheimer's disease: evidence from categorized pictures. *Neuropsychology*. 2006;20(1):113-22.
32. Gottlieb JD, Cather C, Shanahan M, Creedon T, Macklin EA, Goff DC. D-cycloserine facilitation of cognitive behavioral therapy for delusions in schizophrenia. *Schiz Res*. 2011;131:69-74.
33. Goff D, Henderson D, Evins A, Amico E. A placebo-controlled crossover trial of D-cycloserine added to clozapine in patients with schizophrenia. *Biological psychiatry*. 1999;45:512-4.

34. Startup M, Jackson M, Pearce E. Assessing therapist adherence to cognitive-behavioural therapy for psychosis. *Behavioral and Cognitive Psychotherapy*. 2002;30:10-8.
35. Kingdon D, Turkington D. *Cognitive Therapy of Schizophrenia* New York, NY Guilford Press 2005.
36. Fowler DG, Garety P, Kuipers E. Cognitive behaviour therapy for psychosis: Preliminary studies. In: Werbart A, Cullberg J, editors. *The Psychotherapy of Schizophrenia: Facilitating and Obstructive Factors*. Oslo, Norway: Scandinavian University Press; 1992.
37. Chadwick P, Birchwood M. The omnipotence of voices: a cognitive approach to auditory hallucinations. *The British journal of psychiatry : the journal of mental science*. 1994;164:190-210.
38. Nelson H. *Cognitive Behavioral Therapy With Schizophrenia A Practice Manual*. Cheltenham, England Stanley Thornes Publishers, Ltd; 1997.
39. Nair KGS, Epstein IG, Baron H, Mulinos HG. Absorption, distribution and excretion of cycloserine in man. *Antibiotic Annual*. 1956:136-40.
40. Ressler KJ, Rothbaum BO, Tannenbaum L, Anderson P, Graap K, Zimand E, et al. Cognitive enhancers as adjuncts to psychotherapy: use of D-cycloserine in phobic individuals to facilitate extinction of fear. *Archives of general psychiatry*. 2004;61(11):1136-44.
41. Goff DC, Tsai G, Manoach DS, Coyle JT. Dose-finding trial of D-cycloserine added to neuroleptics for negative symptoms in schizophrenia. *Am J Psychiatry*. 1995;152:1213-5.
42. Goff DC, Cather C, Gottlieb DJ, Walsh JP, Raeke L, Otto MW, et al. Once weekly D-cycloserine effects on negative symptoms and cognition. *Schizophrenia research*. in press.
43. van Berckel BNM, Evenblij CN, van Loon BJAM, Maas MF, van der Geld MAM, Wynne HJ, et al. D-cycloserine increases positive symptoms in chronic schizophrenic patients when administered in addition to antipsychotics: A double-blind, parallel, placebo-controlled study. *Neuropsychopharmacology : official publication of the American College of Neuropsychopharmacology*. 1999;21:203-10.
44. Cascella NG, Macciardi F, Cavallini C, Smeraldi E. d-Cycloserine adjuvant therapy to conventional neuroleptic treatment in schizophrenia: An open-label study. *J Neural Transm [Gen Sect]*. 1994;95:105-11.
45. Goff D, Tsai G, Levitt J, Amico E, Manoach D, Schoenfeld D, et al. A placebo-controlled trial of D-cycloserine added to conventional neuroleptics in patients with schizophrenia. *Arch Gen Psych*. 1999;56:21-7.
46. Steel C, Garety PA, Freeman D, Craig E, Kuipers E, Bebbington P, et al. The multidimensional measurement of the positive symptoms of psychosis. *International journal of methods in psychiatric research*. 2007;16(2):88-96.
47. Peters ER, Joseph SA, Garety PA. Measurement of delusional ideation in the normal population: introducing the PDI (Peters et al. Delusions Inventory). *Schizophrenia bulletin*. 1999;25(3):553-76.
48. Laska EM, Mallinckrodt CH, Mundt JC, Leber P, Vaccarino AL, Kalali AH, et al. Assessing onset of treatment benefit in depression and anxiety: conceptual considerations. *The Journal of clinical psychiatry*. 2009;70(8):1138-45.
49. Laska EM, Siegel C. Assessing the onset of relief of a treatment for migraine. *Cephalalgia : an international journal of headache*. 2000;20(8):724-31.
50. Laska EM, Meisner MJ. Nonparametric estimation and testing in a cure model. *Biometrics*. 1992;48(4):1223-34.
51. Goff, D.C., D-cycloserine: an evolving role in learning and neuroplasticity in schizophrenia. *Schizophr Bull*, 2012. 38(5): p. 936-41.
52. Cain, C.K., et al., d-Cycloserine augmentation of cognitive remediation in schizophrenia. *Schizophr Res*, 2014. 153(1-3): p. 177-83.
53. Gottlieb, J.D., et al., D-cycloserine facilitation of cognitive behavioral therapy for delusions in schizophrenia. *Schiz Res*, 2011. 131: p. 69-74.

54. Goff, D., et al., A placebo-controlled trial of D-cycloserine added to conventional neuroleptics in patients with schizophrenia. *Arch Gen Psych*, 1999. 56: p. 21-27.
55. Goff, D.C., et al., Once-weekly d-cycloserine effects on negative symptoms and cognition in schizophrenia: An exploratory study. *Schizophr Res*, 2008. 106(2-3): p. 320-7.
56. Goff, D.C., et al., A six-month, placebo-controlled trial of D: -cycloserine co-administered with conventional antipsychotics in schizophrenia patients. *Psychopharmacology (Berl)*, 2005. 179(1): p. 144-50.
57. Goff, D.C., et al., A placebo-controlled trial of D-cycloserine added to conventional neuroleptics in patients with schizophrenia. *Arch Gen Psychiatry*, 1999. 56(1): p. 21-7.
58. Goff, D., et al., A placebo-controlled crossover trial of D-cycloserine added to clozapine in patients with schizophrenia. *Biol Psychiatry*, 1999. 45: p. 512-514.
59. Goff, D.C., et al., D-cycloserine added to clozapine for patients with schizophrenia. *Am J Psychiatry*, 1996. 153: p. 1628-1630.
60. Norberg, M.M., J.H. Krystal, and D.F. Tolin, A meta-analysis of D-cycloserine and the facilitation of fear extinction and exposure therapy. *Biol Psychiatry*, 2008. 63(12): p. 1118-26.
61. Squire, L. R.; Alvarez, P. (1995). "Retrograde amnesia and memory consolidation: A neurobiological perspective". *Current Opinion in Neurobiology* 5 (2): 169–177. doi:10.1016/0959-4388(95)80023-9
62. Nadel, L.; Moscovitch, M. (1997). "Memory consolidation, retrograde amnesia and the hippocampal complex". *Current Opinion in Neurobiology* 7 (2): 217–227. doi:10.1016/S0959-4388(97)80010-4
63. Gais, Steffen, Geneviève Albouy, Mélanie Boly, Thien Thanh Dang-Vu, Annabelle Darsaud, Martin Desseilles, Géraldine Rauchs, et al. "Sleep Transforms the Cerebral Trace of Declarative Memories." *Proceedings of the National Academy of Sciences* (104): 18778–83. doi:10.1073/pnas.0705454104.