CLINICAL STUDY PROTOCOL

A RANDOMIZED, PLACEBO-CONTROLLED, DOUBLE-BLIND STUDY TO ASSESS THE SAFETY AND EFFICACY OF INTRAVENOUS PCN-101 IN TREATMENT-RESISTANT DEPRESSION

Protocol Number: **PCN-101-21**

Study Phase: **2a**

EudraCT Number: **2020-005457-25**

Sponsor: **Perception Neuroscience, Inc.**
c/o atai Life Sciences, WeWork, 524 Broadway,
New York, NY 10012

Date of Original Protocol: **16 November 2020**

Date of Amendment 4: **19 May 2022**

NCT: NCT05414422

Confidentiality Statement:

SPONSOR SIGNATURE PAGE

Study Title:	A Randomized, Placebo-Controlled, Double-Blind Study to Assess Safety and Efficacy of Intravenous PCN-101 in Treatment Resistant Depression
Protocol Number:	PCN-101-21
Version/Date:	Original Protocol/16 November 2020 Amendment 1/27 April 2021 Amendment 2/ 10 February 2022 (US only) Amendment 3 / 14 April 2022 (US only) Amendment 4 / 19 May 2022
Sponsor Name and Address:	Perception Neuroscience, Inc. c/o atai Life Sciences, WeWork, 524 Broadway, New York, NY , 10012

Approval of protocol by Sponsor:

[redacted] **Date**

GCP Statement: This study will be conducted in compliance with this protocol, Good Clinical Practice, and applicable regulatory requirements.

The Investigator Agreement Page is provided in Appendix 7. The Investigator should retain the original in the study center study files and return a copy to the Sponsor or Contract Research Organization (CRO) for archiving.

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1.0 PROTOCOL SUMMARY

1.1 Synopsis

Name of Sponsor:	Perception Neuroscience, Inc.
Name of Investigational Product:	PCN-101 Solution for Injection
Name of Active Ingredient:	R-ketamine
Study Title:	A RANDOMIZED, PLACEBO-CONTROLLED, DOUBLE-BLIND STUDY TO ASSESS THE SAFETY AND EFFICACY OF INTRAVENOUS PCN-101 IN TREATMENT-RESISTANT DEPRESSION
Protocol Number:	PCN-101-21
Study Phase:	2a

Rationale:

Perception Neuroscience, Inc., is developing PCN-101 (R-ketamine) for at-home treatment of treatment-resistant depression (TRD). Ketamine is a parenterally administered anesthetic agent that was initially approved in the United States (US) in 1970. Since then, it has seen wide use in both adult and pediatric populations. Intranasally administered S-ketamine was approved in 2019 in the US and European Union (EU) for the treatment of treatment-resistant depression. Evidence from nonclinical depression model studies of subanesthetic doses in rodents suggests that R-ketamine has equal or potentially greater efficacy compared with S-ketamine, and both nonclinical and preliminary clinical studies suggest that R-ketamine may have a more favorable safety profile with a decreased incidence of adverse events (AEs) (eg, dissociative, cognitive impairment, and psychotomimetic effects) compared with S-ketamine. Based on nonclinical studies, R-ketamine may also have less abuse potential than S-ketamine. Overall, the available data support the concept of the use of PCN-101 as a potentially better tolerated, rapidly acting antidepressant compared with ketamine and S-ketamine, which may support the use of PCN-101 outside of a supervised, in-clinic environment.

Objectives and Estimands:

Objectives	Estimands
Primary	
To determine the efficacy of 2 doses (30 mg and 60 mg) of intravenous (IV) PCN-101 compared with placebo in improving depressive symptoms in subjects with TRD as assessed by change from baseline to 24 hours after the start of the infusion of PCN-101 in the Montgomery Åsberg Depression Rating Scale (MADRS) total score.	The primary estimand is defined as follows: • **Population**: Subjects with TRD in the full analysis set (FAS) and analyzed according to their randomized treatment. The analysis population includes all randomized subjects who receive any amount of study treatment and have at least 1 postbaseline assessment available. • **Variable**: Change in total MADRS score from baseline at 24 hours after the start of infusion. • **Intercurrent event (ICE)**: Incidents where a subject does not receive the full treatment infusion will be considered an intercurrent event. The intercurrent event will however be handled using a treatment policy where all observed values will be

Objectives	Estimands
	used regardless of occurrence of an intercurrent event. No imputation is performed in the primary efficacy analysis. • **Population-level summary**: Difference in mean change from baseline between each PCN-101 group versus placebo. Additional methodology will be specified in the Statistical Analysis Plan (SAP) for sensitivity and supplementary analyses for assessing the robustness of results. These sensitivity or supplementary analyses will explore different methods for handling intercurrent events and different assumptions for missing data.
Secondary	
• To assess the proportion of subjects with a response (defined as ≥ 50% improvement in MADRS total score from predose).	All secondary efficacy estimands are defined using the same population, ICE strategy, and population level summary (difference in means or proportions between each randomized treatment and placebo, as appropriate) as described above. The variables for the estimands are the following: • MADRS total score assessed at 2 and 4 hours, 7 and 14 days after the start of the infusion (Days 8 and 15, respectively). Note, for the 2-hour and 4-hour recall periods, the sleep and appetite items are not assessed. • Proportion of subjects with ≥ 50% improvement in MADRS total score at 24 hours, 7 days, and 14 days after start of infusion (Days 8 and 15, respectively).
• To assess the proportion of subjects with remission (defined as MADRS total score ≤ 10).	• Proportion of subjects with a MADRS total score ≤ 10 at 24 hours, 7 days, and 14 days after start of infusion (Days 8 and 15, respectively).
• To define changes in Hamilton Depression Rating Scale (HAM-D).	• Changes in HAM-D on Day 8 and Day 15 after start of infusion.
• Generalized Anxiety Disorder 7-Item (GAD-7).	• Change from baseline in GAD-7 by visit.
• Clinical Global Impression - Severity (CGI-S) and Clinical Global Impression - Improvement (CGI-I).	• Change from baseline in CGI-S by visit and CGI-I (calculated from predose CGI-S).
• Quick Inventory of Depressive Symptomatology - 16 Items (QIDS-SR-16).	• Change from baseline in QIDS-SR-16 by visit.
• European Quality - 5 Dimensions - 3 Levels (EQ-5D-3L).	• Change from baseline in EQ-5D-3L by visit.
• To determine the safety and tolerability of 2 doses of PCN-101 administered IV in subjects with TRD compared with placebo.	• Safety as assessed by: o Vital signs o 12-lead electrocardiogram (ECG) o Oxygen saturation (SpO_2) o Clinical laboratory parameters

Objectives	Estimands
	○ AEs ○ Modified Observer's Assessment of Alertness/Sedation (MOAA/S) ○ Clinician-Administered Dissociative States Scale (CADSS) ○ Brief Psychiatric Rating Scale - Modified 4 Components (BPRS+) ○ 5-Dimensional Altered States of Consciousness Rating Scale (5D-ASC) ○ Columbia Suicide Severity Rating Scale (C-SSRS).

Overall Design:

This is a double-blind, randomized, placebo-controlled, multicenter study comprised of 3 phases: screening (up to 2 weeks [Day -15 to Day -2]), In-Clinic Treatment (Day -1 to Day 2; including double-blind treatment [Day 1]), and post-treatment follow-up (7 and 14 days after infusion on Days 8 and 15, respectively). The study consists of 3 arms: placebo, PCN-101 Solution for Injection 30 mg, and PCN-101 Solution for Injection 60 mg. A total of 93 adult subjects with TRD will be randomly allocated in equal cohorts of 31 subjects/arm to the 3 arms of the study in a blinded manner. Randomization will be stratified by region (US, EU).

Subjects will be randomized within 14 days of screening (Visit 1). Subjects will be admitted to the clinic the evening prior to study treatment administration (Day -1, Visit 2) and undergo baseline testing to ensure continued study eligibility. Study treatments will be infused IV over 40 minutes the next morning (Day 1, Visit 2). Starting immediately prior to dosing, subjects will be monitored closely for safety. Additionally, the subjects' alertness, mood, and other psychological parameters will be assessed by clinician- and patient completed scales and questionnaires. Subjects will be discharged no earlier than 24 hours postinfusion and after the final in-clinic assessments have been completed (Day 2, Visit 2). Subjects will be asked to return to the clinic approximately 6 days (Day 8, Visit 3) and 13 days (Day 15, Visit 4) after discharge to assess the safety and tolerability of the study treatments and to determine the durability of the antidepressant effect.

During the coronavirus disease 2019 (COVID-19) pandemic, the specific guidance from local public health and other competent authorities regarding the protection of individuals' welfare must be applied. If subjects cannot attend follow up visits in person because of restrictions arising as a consequence of the COVID-19 pandemic, the Investigator must discuss with the Medical Monitor potential mitigation.

Number of Investigators and Study Centers:

Approximately 20 study centers in the EU and the US are expected to participate in this study.

Number of Subjects:

The enrollment of 93 subjects is planned. Subjects who do not receive a dose of study treatment on Day 1 (Visit 2) will be replaced.

Duration of Study:

The duration of study participation for each subject will be up to 29 days including a screening period, a 3-day in-clinic visit, and 2 follow-up visits.

Inclusion Criteria:

Subjects are eligible to be included in the study only if all of the following criteria apply:

1. Be capable of giving and give signed informed consent, which includes compliance with the requirements and restrictions listed in the Informed Consent Form (ICF) and in this protocol.
2. Be male or female 18 to 65 years of age inclusive at the time of signing the ICF.
3. Weigh ≥ 50 kg and have a body mass index (BMI) ≥ 18 and ≤ 35.
4. Have a diagnosis of recurrent major depressive disorder (MDD) without psychotic features per the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-V), confirmed by the Mini-International Neuropsychiatric Interview (MINI).
5. Have an HAM-D total score > 20 at screening and baseline (Day -1).
6. Have an inadequate response to at least 2 antidepressants in the current episode of depression that were each given for ≥ 6 weeks at an adequate dose as defined by the Massachusetts General Hospital Antidepressant Response Questionnaire (MGH-ATRQ).
7. Must be on stable oral antidepressant treatment without a dose change for at least 30 days before screening (a missed dose, or reasonable number of missed doses per Investigator's discretion, in that period does not exclude a subject).
8. A male subject must be medically confirmed sterile for at least 6 months prior to screening or agree to use highly effective contraception (see Appendix 5) during the treatment period and for at least 3 months after the last dose of study treatment and refrain from donating sperm during this period. If a male with a partner who is of childbearing potential (OCBP) is included, his partner also needs to use highly effective birth control measures.
9. A female subject is eligible to participate if she is not pregnant, not breastfeeding, and at least 1 of the following conditions applies:
    - Not of childbearing potential (see Appendix 5).
    - A subject OCBP who agrees to follow the highly effective contraceptive guidance (see Appendix 5) on highly effective birth control measures during the treatment period and for at least 3 months after the last dose of study treatment and refrain from donating eggs during this period.
10. Be medically stable on the basis of physical examination, medical history, vital signs, and 12-lead ECG performed at screening. If there are abnormalities, the subject may be included only if the Investigator judges the abnormalities not to be clinically important. This determination must be recorded in the subject's source documents and initialed and dated by the Investigator.

Exclusion Criteria:

Subjects are excluded from the study if any of the following criteria apply:

1. History of, or current signs and symptoms of, diseases or conditions that would make participation not be in the best interest (eg, compromise the well-being) of the subject or that could prevent, limit, or confound the protocol-specified assessments.
2. History of moderate or severe head trauma (for example, loss of consciousness for more than 15 minutes) or other neurological disorders (including a diagnosis of epilepsy or has had a seizure in the last 6 months), neurodegenerative disorder (Alzheimer's disease, Parkinson's disease, multiple sclerosis, Huntington's disease, etc.) or systemic medical diseases that are, in the opinion of the Investigator, likely to interfere with the conduct of the study or confound the study assessments. A history of febrile seizures in childhood is not exclusionary.

3. Has a primary DSM-V diagnosis of current (active) MDD with psychotic features, panic disorder, obsessive compulsive disorder, posttraumatic stress disorder, anorexia nervosa, or bulimia nervosa. Comorbid anxiety or panic disorder that does not dominate the clinical presentation is acceptable.
4. Has a current or prior DSM-V diagnosis of a primary psychotic disorder (eg, schizophrenia), bipolar or related disorders (confirmed by the MINI), intellectual or autism spectrum disorder, or borderline personality disorder.
5. Has any significant disease or disorder (eg, cardiovascular, pulmonary, gastrointestinal, hepatic, renal, neurological, musculoskeletal, endocrine, metabolic, malignant, psychiatric, major physical impairment) that, in the opinion of the Investigator, may either put the subject at risk because of participation in the study, influence the results of the study, or affect the subject's ability to participate in the study.
6. Has uncontrolled hypertension, despite medication, at Screening (systolic blood pressure [SBP] > 160 mm Hg or diastolic blood pressure [DBP] > 90 mm Hg) or any past history of hypertensive crisis. An abnormal blood pressure value at screening may be repeated once after 10-15 minutes of relaxation to determine the subject's eligibility.
7. Has an abnormal ECG of clinical relevance at screening or baseline (Day -1) including, but not limited to, the following:
    - QT interval corrected according to Fridericia's formula (QTcF) interval > 450 msec for male subjects and >470 msec for female subjects.
    - Evidence of 2nd and 3rd degree atrioventricular block, complete left bundle branch block (LBBB), or complete right bundle branch block (RBBB).
    - Features of new ischaemia.
    - Arrhythmia (except premature atrial contractions [PACs] and premature ventricular contractions [PVCs]).
    - Has a history of risk factors including hypokalemia or a family history of Long QT Syndrome.
8. Has known history of, or positive serology for, human immunodeficiency virus (HIV); has a positive hepatitis B surface antigen, and/or confirmed current hepatitis C infection (positive hepatitis C virus [HCV] antibody confirmed with reflex HCV ribonucleic acid [RNA] test). Subjects with a history of hepatitis B vaccination without a history of hepatitis B are allowed to enroll.
9. Has a history of malignancy within the 5 years prior to screening (exceptions are squamous and basal cell carcinomas of the skin and carcinoma in situ of the cervix, or a malignancy that in the opinion of the Investigator, with concurrence with the Sponsor's Medical Monitor, is considered to have minimal risk of recurrence).
10. Has homicidal ideation/intent per the Investigator's clinical judgment; or has suicidal ideation with some intent to act within 1 month prior to the start of screening per the Investigator's clinical judgment or based on the C-SSRS, corresponding to a response of "Yes" on Item 4 (active suicidal ideation with some intent to act, without specific plan) or Item 5 (active suicidal ideation with specific plan and intent); or a history of suicidal behavior within the past year prior to the start of the screening/prospective observational phase.
11. Has had major surgery (eg, requiring general or local anesthesia) within the 4 weeks before screening, or will not have fully recovered from surgery or planned surgery during the time the subject is expected to participate in the study.
12. Has moderately impaired hepatic function at screening, defined as serum alanine aminotransferase (ALT) or aspartate aminotransferase (AST) > 2 × upper limit of normal (ULN) or total bilirubin (TBL) > 2 × ULN.
13. Has received any disallowed therapies as follows (see Section 6.5 for further details):

a. Receipt of a known potent inhibitor of hepatic cytochrome P450 (CYP) 2B6, or CYP3A, activity within 1 week or within a period 5 times the drug's half-life, whichever is longer, before the first administration of study drug on Day 1.
b. Treatment with a disallowed antipsychotic within the past 30 days prior to screening, except subjects who are on stable doses of quetiapine, aripiprazole, brexpiprazole, or olanzapine prescribed as adjunct treatment for depression (without psychosis) may be included in the study.
c. Any changes in psychotropic medication type or dose within the past 30 days prior to screening.
d. Treatment with monoamine oxidase inhibitors (MAOIs) currently or within the past 30 days of screening.
e. Doses of oral contraception should not contain more than 30 micrograms of ethinyl estradiol per day.
f. Refer to Appendix 6 for a list of permitted and prohibited concomitant medications. The Medical Monitor should be contacted for any questions regarding concomitant medications.

14. Has initiated psychotherapy (eg, Cognitive Behavior Therapy, Interpersonal Psychotherapy, Psychodynamic Psychotherapy other than psycho-education, or acupuncture within the past 90 days of screening. Patients planning to initiate individual or group therapy during the study are also not eligible.
15. Has received electroconvulsive therapy, transcranial magnetic stimulation, vagal nerve stimulation, deep brain stimulation, or other brain stimulation treatment within the past 4 weeks or currently used as either an acute or maintenance treatment of depression.
16. Has received any investigational product (IP) within 30 days or 5 half-lives prior to dosing with PCN-101.
17. Has a history of substance abuse (drug or alcohol) or dependence (except nicotine or caffeine) within the previous 6 months prior to the screening visit.
    - Has a positive urine drug screen for ketamine, opiates, cocaine, barbiturates, and/or amphetamine/methamphetamine or positive alcohol screen at Screening or Day -1.
    - Subjects who have a positive test result at screening due to prescribed opiates or amphetamines may be permitted to continue the screening phase if the prohibited medication is discontinued at least 1 week or 5 half-lives, whichever is longer, before the first dose of study medication. Retesting is not permitted for positive test result(s) from nonprescription use of drugs of abuse.
18. Has a history of previous nonresponse to ketamine, R-ketamine or S-ketamine, or has received 8 or more doses of ketamine, R-ketamine or S-ketamine in their lifetime.
19. Has a previous history of intolerance to ketamine, R-ketamine, or S-ketamine.
20. History of abuse of ketamine, R-ketamine, S-ketamine, or phencyclidine.
21. Subjects should not consume grapefruit, grapefruit juice, or Seville orange -related products for 72 hours before IP administration and throughout the study.
22. Has the presence of clinically relevant long-term COVID-19 symptoms. Has current signs or symptoms of COVID-19.
23. COVID-19 vaccination is allowed as long as the doses are administered ≥ 30 days before study drug administration; vaccination is not allowed during the course of the study.

Treatment Groups and Duration:

- The IP (also referred to as Investigational Medicinal Product [IMP]) is PCN-101 Solution for Injection (R-ketamine hydrochloride in sterile water for injection). Subjects will receive PCN-101 Solution for Injection at either 30 mg or 60 mg or placebo.
- A placebo control will be used in the study.
- The active ingredient of the study treatment is R-ketamine.
- [redacted]
- PCN-101 or placebo will be administered on Day 1 as a single IV infusion via an electronic infusion pump over 40 minutes.
- IP/IMP dose modifications are not permitted in this study.
- The duration of study participation for each subject will be up to 29 days including a screening period, a 3-day in-clinic visit, and 2 follow-up visits.

Statistical Methods:

General Considerations

The comprehensive SAP with detailed description of all statistical analyses will be developed and finalized before database lock.

All statistical analyses, including summary tables and data listings will be performed using SAS® software (version 9.4 or higher). Continuous endpoints will be summarized using descriptive statistics (number of subjects [n], mean, standard deviation [SD], median, minimum and maximum). Categorical endpoints will be summarized using frequency counts and percentages. All individual subject data will be presented in listings.

Sample Size Calculation

The intent of the primary efficacy analysis is to demonstrate superiority of at least 1 therapeutic dose of PCN-101 Solution for Injection (30 mg or 60 mg) versus placebo based on the change in the MADRS total score from predose to 24 hours postdose.

For the primary analysis, a sample size of 93 randomized subjects (31:31:31) will provide 80% power to detect an 8-point difference between each PCN-101 Solution for Injection dose and placebo in the mean change from baseline MADRS total score at 24 hours postdose, using a t-test with $\alpha = 0.05$ (2-tailed) and assuming a common SD of 11.

The enrollment of 93 subjects is planned. Subjects who do not receive a dose of study treatment on Day 1 (Visit 2) will be replaced.

Populations for Analysis

Analysis Set	Description
Full Analysis Set (FAS)	The FAS will consist of all randomized subjects who receive at least 1 dose of study treatment and have at least 1 postbaseline assessment available. This population will serve as the basis for efficacy analysis. Subjects will be analyzed according to their randomized treatment.
Safety Analysis Set	The Safety Population will consist of all randomized subjects who receive any study treatment, even a partial dose. This population will be used for all summaries of subject disposition, demographic and baseline data, and safety information including AE incidence. Subjects will be analyzed according to the treatment they actually received.

Abbreviations: AE = adverse event; FAS = Full Analysis Set.

Full details of the efficacy analyses and any further supplementary analyses deemed appropriate, along with details on handling of missing data will be provided in the SAP.

Subject Disposition and Demographics

Subject disposition events (screened, randomized, completed the study, early discontinuation from the study along with the reason for discontinuation) as well as the number of subjects in each analysis population will be summarized using frequency counts and percentages by treatment groups.

Efficacy Analyses

Endpoint	Statistical Analysis Methods
Primary • Change from baseline in MADRS total score assessed at 24 hours after the start of the infusion.	The primary efficacy estimand will be evaluated in the FAS using a mixed effects model for repeated measure (MMRM) analysis with observed cases only. The MMRM model will include fixed effects for treatment, region, visit, study site and treatment by visit interaction, with subject as a random effect and baseline score as a covariate. From this analysis the least squares (LS) mean estimates for each treatment arm at each visit, along with the standard error and 95% confidence intervals (CIs) will be presented separately. The primary comparison for the MADRS is the estimate of the treatment difference at 24 hours after the start of the infusion.
Secondary • Change from baseline in MADRS total score at 2 hours, 4 hours, 7 days, and 14 days after the start of the infusion. • Proportion of subjects with ≥ 50% improvement in MADRS total score at 24 hours, 7 days, and 14 days after start of infusion. • Proportion of subjects with a MADRS total score ≤ 10 at 24 hours, 7 days, and 14 days after start of infusion. • Changes in HAM-D on Day 8 and Day 15 after start of infusion.	Observed and change from baseline values in continuous secondary endpoints will be summarized descriptively by treatment group. Continuous secondary efficacy endpoints assessed at more than 1 postbaseline visit will be analyzed using an MMRM analysis with observed cases only. The MMRM model will include factors for treatment, region, visit, study site, and treatment by visit as fixed effect; subjects as a random effect; and baseline score as a covariate. Categorical secondary endpoints will be summarized by frequency counts and percentages and analyzed by a chi-square test between each PCN-101 dose versus placebo.

Endpoint	Statistical Analysis Methods
• Change from baseline in QIDS-SR-16 by visit. • Change from baseline in GAD-7 by visit. • Change from baseline in EQ-5D-3L by visit. • Change from baseline in CGI S by visit and CGI I (calculated from predose CGI S).	
	Alternate sensitivity analyses will be detailed in the SAP.

Abbreviations: CI = confidence interval; MMRM = mixed effects model for repeated measure; SAP = Statistical Analysis Plan.

Safety Analysis

All safety analyses will be performed on the Safety Analysis Set.

Adverse Events

Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA; Version 23.0 or higher). Treatment-emergent adverse events (TEAEs) are AEs with onset or worsening after the start of study treatment. The AE summaries will be primarily based on TEAEs. The number and percentage of subjects with TEAEs will be summarized by treatment group, system organ class, and preferred term for all TEAEs, treatment-related TEAEs, serious adverse events (SAEs), serious TEAEs, severe TEAEs, TEAEs of special interest, and all TEAEs leading to study drug discontinuation. All TEAEs will be further summarized by maximum severity and causality. The TEAEs of special interest will be identified in the SAP.

All AEs will be presented in a by-subject listing. Serious AEs, severe AEs, and AEs leading to study discontinuation or death will be presented in separate listings.

Clinical Laboratory Tests

Laboratory data will be summarized using descriptive statistics for each parameter, including change from baseline at each assessment time by treatment group. Frequency tables or shift tables will be used to present the number and percentage of subjects with laboratory values within/outside reference ranges. All laboratory data will be present in the listings. A by-subject listing will also be provided for subjects with abnormal laboratory results.

Electrocardiogram

Descriptive statistics will be used to summarize the observed and change from baseline values of the following ECG parameters at each assessment time by treatment group: heart rate, PR interval, QRS interval, QT interval, and QTcF. All ECG data will be present in listings.

Vital Signs

Vital sign parameters including temperature, pulse/heart rate, respiratory rate, SpO2, and blood pressure will be summarized descriptively for both the observed and change from baseline values at each assessment time by treatment group. Frequency tabulations will be provided for abnormalities as applicable.

Other Safety Questionnaires and Assessments

All physical examination results will be listed by subject

Prior and concomitant therapies will be summarized separately and tabulated for each treatment group using frequencies and percentages. Percentages will be based on the number of subjects in each treatment group. Prior and concomitant medications will be listed by subject.

For the C-SSRS: suicidal ideation and behavior scores at each assessment time as well as the incidence rate of TEAEs will be summarized using frequency counts and percentages by treatment group.

For the CADSS, BPRS+, 5D-ASC, and MOAA/S: Descriptive statistics of scores and their changes from baseline will be summarized at each assessment time by treatment group.

Pregnancy test and urine drug and alcohol screen will be presented in by-subject listings.

Other Analyses

Exploratory analyses may be done if indicated.

Missing Data

In general, the missing data which is observed from this study will be assumed to be missing-at-random (MAR).

Since subjects are hospitalized at the time of the primary endpoint, it is anticipated that missing data will be infrequent. No imputation will be applied to primary analysis.

Further details on the handling of missing data will be provided in the SAP.

Interim Analyses

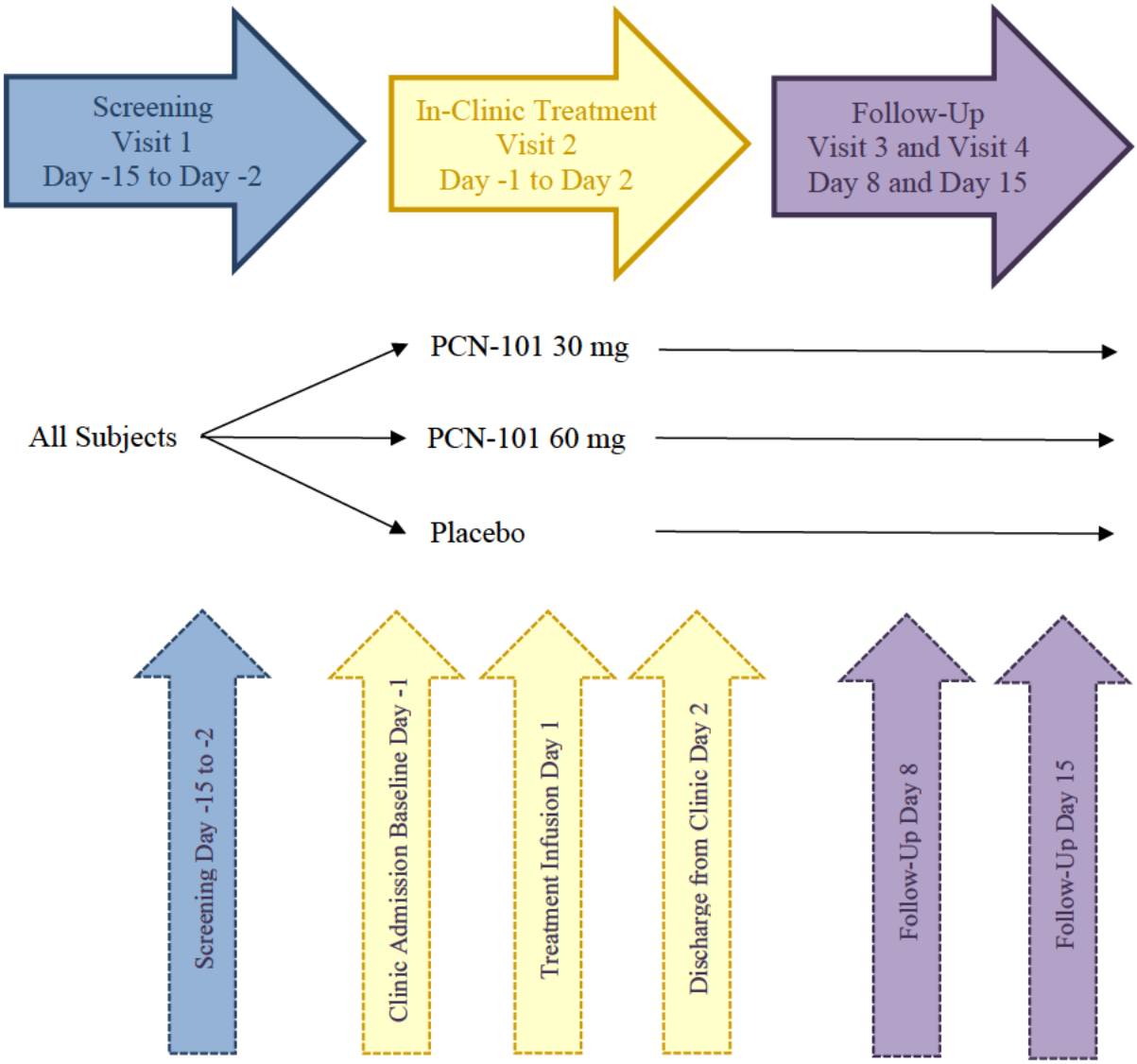
No interim analysis for efficacy is planned.

Data Monitoring Committee:

Because of the short timeframe of the study, there will not be a data monitoring committee.

1.2 Schema

Figure 1 Study Schema



1.3 Schedule of Activities

Procedure	**Visit 1 Screening Day -15 to -2**	**Visit 2 Baseline Day -1**	**Visit 2 In-clinic Dosing Day 1**							**Visit 2 Discharge Day 2**	**Visit 3 Follow-up Day 8 (±1 Day)**	**Visit 4 Follow-up Day 15 (±1 Day)[a]**
After Start of Infusion[b]			Predose[c]	15 min[c]	40 min (EoI) [b,c,d]	60 min[c]	2 hr[c]	4 hr[c]	6 hr[c]	24 hr[c]	7 days[c]	14 days[c]
Informed consent	X											
Inclusion/exclusion criteria	X[e]	X[e]										
Demography	X											
Physical examination	X	X								X		X
Height, weight, and BMI	X											
Medical history	X											
Prior/concomitant medications	X	X	X							X	X	X
Serum pregnancy test (subject OCBP)	X											
Urine pregnancy test (subject OCBP)		X									X	X
HIV, hepatitis B and C test	X											
Laboratory assessments[f]	X	X									X	X
12-Lead ECG[g]	X	X	X		X		X	X	X		X	X
Vital signs[h]	X	X	X		X		X	X	X	X	X	X
Pulse oximetry (SpO_2)			Continuous from predose to 4 hour postdose									
Urine drug/alcohol screen[i]	X	X										
Ketamine screen	X	X										
AE review	X	X	X							X	X	X

Procedure	**Visit 1 Screening Day -15 to -2**	**Visit 2 Baseline Day -1**	**Visit 2 In-clinic Dosing Day 1**							**Visit 2 Discharge Day 2**	**Visit 3 Follow-up Day 8 (±1 Day)**	**Visit 4 Follow-up Day 15 (±1 Day)[a]**
After Start of Infusion[b]			Predose[c]	15 min[c]	40 min (EoI) [b,c,d]	60 min[c]	2 hr[c]	4 hr[c]	6 hr[c]	24 hr[c]	7 days[c]	14 days[c]
Scales and Questionnaires												
MINI	X											
MGH-ATRQ	X											
HAM-D[j]	X	X	X							X	X	X
MADRS[j,k]	X		X				X	X		X	X	X
QIDS-SR-16			X							X[l]	X	X
CGI-S[j]			X[m]							X	X	X
CGI-I[j]										X	X	X
GAD-7[j]			X							X	X	X
EQ-5D-3L[j]			X							X	X	X
MOAA/S			X	Every 20 min for first 2 hr				X	X			
CADSS			X		X		X	X				
BPRS+			X		X		X	X				
5D-ASC									X			
C-SSRS[n]	X	X								X	X	X

Procedure	**Visit 1 Screening Day -15 to -2**	**Visit 2 Baseline Day -1**	**Visit 2 In-clinic Dosing Day 1**							**Visit 2 Discharge Day 2**	**Visit 3 Follow-up Day 8 (±1 Day)**	**Visit 4 Follow-up Day 15 (±1 Day)[a]**
After Start of Infusion[b]			Predose[c]	15 min[c]	40 min (EoI) [b,c,d]	60 min[c]	2 hr[c]	4 hr[c]	6 hr[c]	24 hr[c]	7 days[c]	14 days[c]
Study Administration												
Admission		X										
Randomization			X									
Treatment Administration				40 min								

Abbreviations: AE = adverse event; BMI = body mass index; BPRS+ = Brief Psychiatric Rating Scale - Modified 4 Components; CADSS = Clinician-Administered Dissociative States Scale; CGI-I = Clinical Global Impression - Improvement; CGI-S = Clinical Global Impression - Severity; C-SSRS = Columbia Suicide Severity Rating Scale; CRF = case report form; DBP = diastolic blood pressure; 5D-ASC = 5-Dimensional Altered States of Consciousness Rating Scale; ECG = electrocardiogram; EQ-5D-3L = European Quality - 5 Dimensions - 3 Levels; EoI = End of Infusion; GAD-7 = Generalized Anxiety Disorder 7-Item Scale; HAM-D = Hamilton Depression Rating Scale; HIV = human immunodeficiency virus; MADRS = Montgomery Åsberg Depression Rating Scale; MGH-ATRQ = Massachusetts General Hospital Antidepressant Response Questionnaire; MINI = Mini-International Neuropsychiatric Interview; MOAA/S = Modified Observer's Assessment of Alertness/Sedation; OCBP = of childbearing potential; PK = pharmacokinetic; QIDS-SR-14 = Quick Inventory of Depressive Symptomatology – 14 Items; QIDS-SR-16 = Quick Inventory of Depressive Symptomatology – 16 Items; SBP = systolic blood pressure.

a. Subjects who discontinue the study early will complete Visit 4 assessments and procedures as early termination procedures.

b. All post-dose timepoints are relative to the start of infusion except for the 40-minute (EoI) timepoint. EoI procedures should occur 40 minutes after the start of infusion or at the end of infusion if the infusion continues beyond 40 minutes.

c. Where activities at a given timepoint coincide, consideration should be given to ensure that the following order of activities is maintained: Efficacy rating scales (where more than one efficacy scale is assessed at a timepoint, the order of assessments should be MADRS, CGI-I and CGI-S, QIDS-SR-14/16, HAM-D, GAD-7 and EQ5D-3L); 12-lead ECGs; vital signs and pulse oximetry; safety rating scales (order: MOAA/S, CADSS and BPRS+); AE review, concomitant medication review, physical exam, and clinical laboratory assessment (including urine pregnancy test and drug/alcohol screen).

d. Assessments and procedures should be started as close as possible after the EoI.

e. Check eligibility.

f. Assess clinical hematology and chemistry tests (non-fasting blood samples will be requested, but both fasting and non-fasting samples are acceptable).

g. To be collected prior to clinical laboratory assessments. An ECG will be performed after 5 minutes in a supine position, if possible.

h. To be collected prior to clinical laboratory assessments. Vital signs will include SBP/DBP, heart rate, respiratory rate, and temperature (tympanic/temporal). Blood pressure and pulse measurements should be taken after the subject has been in a supine or semi-supine position in a rested and calm state for at least 5 minutes. Blood pressure must be recorded using the same arm and position (supine or semi-supine) throughout the study, where possible. Three consecutive blood pressure readings will be recorded at intervals of at least 1 minute and the average of the 3 blood pressure readings will be recorded in the CRF. An abnormal blood pressure value at screening may be repeated once after 10-15 minutes of relaxation to determine the subject's eligibility.

i. Urine drug screen will be conducted by a local laboratory and include alcohol, ketamine, opiates, cocaine, barbiturates, and/or amphetamine/methamphetamine. Benzodiazepines are allowed up to 6 hours before the start of the infusion, and if needed as rescue medication.

j. The HAM-D, MADRS, CGI-S, CGI-I, GAD-7, and EQ-5D-3L should be evaluated by a separate Investigator who is not involved with the assessment of safety to avoid functional unblinding.

k. There are 3 versions of MADRS: 7-day recall, 24-hour recall, and 2-hour recall. For the 2 hour and 4 hour periods, the sleep and appetite items will not be assessed; predose scores for these items obtained on the same day will be carried forward.

l. The QIDS-SR-14, with a 24 hour recall period, will be used on Visit 2 Day 2 assessment due to recall period.

m. Note: The predose CGI-S will be used as the basis for calculating the CGI-I at Visit 2.

n. Note: C-SSRS may be evaluated by either efficacy or safety rater.

2.0 INTRODUCTION

As a racemate, ketamine consists of the 2 stereoisomers of R-ketamine and S-ketamine in equal proportions. Ketamine is a parenterally administered anesthetic agent that was initially approved in the United States (US) in 1970. Since then, it has seen wide use in both adult and pediatric populations. S-ketamine was approved in the US and later approved in the European Union (EU) in 2019 as the isolated stereoisomer in an intranasal formulation (Spravato™) for the treatment of treatment-resistant depression (TRD) in adults. However, this product has shown to have a limited therapeutic index, particularly with respect to dissociative side effects including illusions, distortions of time and space, derealization, and depersonalization. Further, in a human abuse potential study, the scores for "Drug Liking at the Moment" and "Take Drug Again" were similar to racemic ketamine, a known drug of abuse, and greater than placebo at both the maximum indicated antidepressant dose and 1.3 times this dose (1). Based in part upon these findings, the administration of Spravato is limited to the clinic and under the observation of medical personnel.

Perception Neuroscience, Inc. (hereafter referred to as Perception Neuroscience or the Sponsor), is developing PCN-101 (R-ketamine) for at-home treatment of TRD. Nonclinical studies suggest that R-ketamine could cause less dissociative and psychotomimetic effects at therapeutic doses and have a lower potential for misuse compared with S-ketamine or racemic ketamine (2, 3, 4, 5, 6, 7, 8, 9, 10).

Both R-ketamine and S-ketamine (and by extension, racemic ketamine) share complex pharmacology with receptor-binding studies revealing significant affinity at several receptors and ion channels, eg, glutamatergic, cholinergic, sigma, opioid, and hyperpolarization-activated cyclic nucleotide-gated channels. R-ketamine and S-ketamine are primarily regarded as N-methyl-D-aspartate (NMDA) receptor non-competitive antagonists, with S-ketamine having approximately 4 times the binding affinity for the phencyclidine site of this receptor than R-ketamine. Such pharmacological activity is thought to be the primary driver of the adverse psychotomimetic properties seen with ketamine, and S-ketamine is suggested to mainly cause these effects. However, data suggest that such NMDA receptor activity is not the only driver of antidepressant efficacy. Indeed, nonclinical depression model studies of subanesthetic doses (10 mg/kg) in rodents suggest R-ketamine possesses longer acting and more potent effects than S-ketamine despite R-ketamine's lower affinity to the NMDA receptor (2, 3, 5, 7, 9, 11, 12, 13, 14, 15, 16, 17, 18). Moreover, at doses that had effects in depression models in rodents (20 mg/kg), R-ketamine did not cause conditioned place preference (2, 7), a nonclinical test that is thought to suggest a clinical risk of substance abuse. However, it should be noted that at a high dose (40 mg/kg), R-ketamine did cause conditioned place preference in rodents (19).

Overall, the available data support the use of PCN-101 as a potentially better tolerated, rapid-acting antidepressant compared with ketamine and S-ketamine, which may support the use of PCN-101 outside of a supervised, in-clinic environment.

2.1 Study Rationale

Perception Neuroscience is developing PCN-101 (R-ketamine) for at -home treatment of TRD. Evidence from nonclinical depression model studies suggests that R-ketamine may have equal or potentially greater efficacy compared with S-ketamine, and both nonclinical and clinical studies suggest that R-ketamine may have a more favorable safety profile with a decreased incidence of adverse events (AEs) (eg, dissociative, cognitive impairment, and psychotomimetic effects) compared with S-ketamine. Based on nonclinical studies, R-ketamine may also have less abuse potential than S-ketamine. Overall, the available data support the concept of the use of PCN-101 as a potentially better tolerated, rapidly acting antidepressant compared with ketamine and S-ketamine. Protocol PCN-101-21 is a proof-of-concept study to assess efficacy and safety to provide evidence that PCN-101 can be developed as a rapidly acting antidepressant for the potential treatment of TRD.

2.2 Background

Although racemic ketamine has been extensively used for almost 50 years as an approved parenteral anesthetic worldwide, it has not been approved for the indication of TRD because of concerns over potential AEs related to repeated dosing, abuse liability, and its intravenous (IV) route of administration. S-ketamine, the isolated (S)-enantiomer of racemic ketamine, was approved in the US and later approved in the EU in 2019 in an intranasal formulation for the treatment of TRD in adults, but the therapeutic index of this product is limited by dissociative side effects including illusions, distortions of time and space, derealization, and depersonalization limiting it use to the clinic setting. In addition, the potential abuse profile has been demonstrated to be similar to that of racemic ketamine. Thus, the search for novel ketamine-like agents has been driven by the goal to reduce the side effects seen with ketamine and S-ketamine while providing antidepressant efficacy and a convenient route of administration to permit the use outside of a supervised clinical setting.

Perception Neuroscience plans to develop PCN-101 for the treatment of psychiatric conditions. The doses of 30 mg and 60 mg to be used in this study were selected based on the results from a Phase 1 single-ascending-dose study in healthy volunteers. A detailed description of the chemistry, pharmacology, efficacy, and safety of PCN-101 is provided in the current Investigator’s Brochure.

2.3 Benefit/Risk Assessment

Due to its 4-fold lower affinity to the receptor, it is hypothesized that R-ketamine may have fewer N-methyl-D-aspartate receptor (NMDAR)-related adverse effects compared with racemic ketamine or S-ketamine, such as psychotomimetic effects and dissociation.

[redacted] It has been suggested that this action of R-ketamine may play a role in the hallucinogenic effects of racemic ketamine and that it may be responsible for the lowering of the seizure threshold seen with racemic ketamine.

Reported AEs from limited academic clinical studies in healthy volunteers evaluating R-ketamine subanesthetic doses of up to 1 mg/kg IV as bolus or short infusion or 1.8 mg/kg intramuscular included transient increases in blood pressure, emotional changes, illusion, sedation, proprioceptive and sensory disturbances, decline in concentration capacity and primary memory, blurred vision, altered hearing, and dizziness. Overall, less pronounced psychotomimetic and dissociative-like effects were reported with R-ketamine than with S-ketamine (20, 21, 22, 23, 24, 25). Results from an open-label pilot study of a single IV infusion of R-ketamine (0.5 mg/kg) administered to 7 subjects with TRD suggested that R-ketamine might produce fast-onset and sustained antidepressant effects with a favorable safety profile (35).

To date, there has been 1 clinical trial that evaluated the safety of PCN-101. The primary objective of study PCN-101-02 was to identify an acceptable tolerated dose of PCN-101 in healthy subjects and compare this dose to the safety profile of 15 mg S-ketamine, a dose which demonstrated a robust antidepressant effect in a published study of TRD patients (36). Dose ranging using single ascending doses of PCN-101-02 demonstrated that doses of PCN-101 ≤ 150 mg had acceptable safety profiles as single 40-minute IV infusions. A comparison of the safety of dose levels of PCN-101 and 15 mg S-ketamine identified 60 mg PCN-101 as the dose level most similar to 15 mg S-ketamine in terms of safety profile. Both 30 mg and 60 mg doses of PCN-101 resulted in fewer transient elevations in mean systemic blood pressure, less sedation, fewer psychological or psychotic and dissociative effects, and fewer episodes of altered consciousness than 100 mg or 150 mg doses of PCN-101.

More detailed information about the known and expected benefits and risks and reasonably expected AEs of PCN-101 may be found in the current Investigator's Brochure.

There are no identified serious adverse reactions that are considered expected for the purpose of expedited regulatory reporting.

2.3.1 Drug Abuse and Dependence

Ketamine has been reported being used as a drug of abuse. Ketamine dependence and tolerance are possible following prolonged administration. Although R-ketamine may have a potential for abuse, nonclinical data suggest a lower abuse potential for R-ketamine than that for racemic ketamine or S-ketamine due to its pharmacological properties, such as lesser affinity to NMDAR and a lack of effects on the dopamine pathway (4, 30, 31, 32). The abuse and dependence potential of PCN-101 in humans is unknown and has not yet been studied.

2.3.2 Drug-Drug Interactions

Multiple in vitro studies examining the in vitro drug metabolism and pharmacokinetics of PCN-101 have been conducted. For more information, refer to the Investigator's Brochure.

[redacted]

There is a potential for drug-drug interactions with CYP2B6 and CYP3A4 substrates. The clinical impact of these DDI's is thought to be minimal.

Clinical drug-drug interaction studies have not been performed with PCN-101 to date. Concomitant medications should be avoided except for those specifically allowed (see Section 6.5).

2.3.3 Overdose

There is no specific antidote for PCN-101 overdose.

3.0 OBJECTIVES AND ESTIMANDS

The study objectives and estimands are presented in Table 1.

Table 1 Study Objectives and Estimands

Objectives	Estimands
Primary	
To determine the efficacy of 2 doses (30 mg and 60 mg) of IV PCN-101 compared with placebo in improving depressive symptoms in subjects with TRD as assessed by change from baseline to 24 hours after the start of the infusion of PCN-101 in the Montgomery Åsberg Depression Rating Scale (MADRS) total score.	The primary estimand is defined as follows • **Population**: Subjects with TRD in the full analysis set (FAS) and analyzed according to their randomized treatment. The analysis population includes all randomized subjects who receive any amount of study treatment and have at least 1 postbaseline assessment available. • **Variable**: Change in total MADRS score from baseline at 24 hours after the start of infusion. • **Intercurrent event (ICE)**: Incidents where a subject does not receive the full treatment infusion will be considered an intercurrent event. The intercurrent event will however be handled using a treatment policy where all observed values will be used regardless of occurrence of an intercurrent event. No imputation is performed in the primary efficacy analysis. • **Population-level summary**: Difference in mean change from baseline between each PCN-101 group versus placebo Additional methodology will be specified in the Statistical Analysis Plan (SAP) for sensitivity and supplementary analyses for assessing the robustness of results. These sensitivity or supplementary analyses will explore different methods for handling intercurrent events and different assumptions for missing data.
Secondary	
• To assess the proportion of subjects with a response (defined as ≥ 50% improvement in MADRS total score from predose).	All secondary efficacy estimands are defined using the same population, ICE strategy, and population level summary (difference in means or proportions between each randomized treatment and placebo, as appropriate) as described above. The variables for the estimands are the following: • MADRS total score assessed at 2 and 4 hours, 7 and 14 days after the start of the infusion (Days 8 and 15, respectively). Note, for the 2-hour and 4-hour recall periods, the sleep and appetite items are not assessed. • Proportion of subjects with ≥ 50% improvement in MADRS total score at 24 hours, 7 days, and 14

Objectives	Estimands
• To assess the proportion of subjects with remission (defined as MADRS total score ≤ 10). • To define changes in Hamilton Depression Rating Scale (HAM-D). • Generalized Anxiety Disorder 7-Item (GAD-7). • Clinical Global Impression - Severity (CGI-S) and Clinical Global Impression - Improvement (CGI-I). • Quick Inventory of Depressive Symptomatology - 16 Items (QIDS-SR-16). • European Quality - 5 Dimensions - 3 Levels (EQ-5D-3L). • To determine the safety and tolerability of 2 doses of PCN-101 administered IV in subjects with TRD compared with placebo.	days after start of infusion (Days 8 and 15, respectively). • Proportion of subjects with a MADRS total score ≤ 10 at 24 hours, 7 days, and 14 days after start of infusion (Days 8 and 15, respectively). • Changes in HAM-D on Day 8 and Day 15 after start of infusion. • Change from baseline in GAD-7 by visit. • Change from baseline in CGI-S by visit and CGI-I (calculated from predose CGI-S). • Change from baseline in QIDS-SR-16 by visit. • Change from baseline in EQ-5D-3L by visit. • Safety as assessed by: o Vital signs o 12-lead electrocardiogram (ECG) o Oxygen saturation (SpO_2) o Clinical laboratory parameters o AEs o Modified Observer's Assessment of Alertness/Sedation (MOAA/S) o Clinician-Administered Dissociative States Scale (CADSS) o Brief Psychiatric Rating Scale - Modified 4 Components (BPRS+) o 5-Dimensional Altered States of Consciousness Rating Scale (5D-ASC) o Columbia Suicide Severity Rating Scale (C-SSRS).

Abbreviations: 5D-ASC = 5-Dimensional Altered States of Consciousness Rating Scale; AE = Adverse event; BPRS+ = Brief Psychiatric Rating Scale - Modified 4 Components; CADSS = Clinician-Administered Dissociative States Scale; CGI-I = Clinical Global Impression - Improvement; CGI-S = Clinical Global Impression - Severity; C-SSRS = Columbia Suicide Severity Rating Scale; ECG = electrocardiogram; EQ-5D-3L = European Quality - 5 Dimensions - 3 Levels; FAS = full analysis set; GAD-7 = Generalized Anxiety Disorder 7-Item; HAM-D = Hamilton Depression Rating Scale; ICE = intercurrent event; IV = intravenous; MADRS = Montgomery Åsberg Depression Rating Scale; MOAA/S = Modified Observer's Assessment of Alertness/Sedation; QIDS-SR-16 = Quick Inventory of Depressive Symptomatology - 16 Items; SAP = Statistical Analysis Plan; SpO_2 = oxygen saturation; TRD = treatment-resistant depression.

4.0 STUDY DESIGN

4.1 Overall Design

This is a double-blind, randomized, placebo-controlled, multicenter study comprised of 3 phases: screening (up to 2 weeks [Day -15 to Day -2]), In-Clinic Treatment (Day -1 to Day 2; including double-blind treatment [Day 1]), and post-treatment follow-up (7 and 14 days after infusion, on Days 8 and 15, respectively). The study consists of 3 arms: placebo, PCN-101 Solution for Injection 30 mg, and PCN-101 Solution for Injection 60 mg. A total of 93 adult subjects with TRD will be randomly allocated in equal cohorts of 31 subjects/arm to the 3 arms of the study in a blinded manner. Randomization will be stratified by region (US, EU).

Subjects will be randomized within 14 days of screening (Visit 1). Subjects will be admitted to the clinic the evening prior to study treatment administration (Day -1, Visit 2) and undergo baseline testing to ensure continued study eligibility. Study treatments will be infused IV over 40 minutes the next morning (Day 1, Visit 2). Starting immediately prior to dosing, subjects will be monitored closely for safety. Additionally, the subjects' alertness, mood, and other psychological parameters will be assessed by clinician- and patient-completed scales and questionnaires. Subjects will be discharged no earlier than 24 hours postinfusion and after the final in-clinic assessments have been completed (Day 2, Visit 2). Subjects will be asked to return to the clinic approximately 6 days (Day 8, Visit 3) and 13 days (Day 15, Visit 4) after discharge to assess the safety and tolerability of the study treatments and to determine the durability of the antidepressant effect.

During the coronavirus disease 2019 (COVID-19) pandemic, the specific guidance from local public health and other competent authorities regarding the protection of individuals' welfare must be applied. If subjects cannot attend follow up visits in person because of restrictions arising as a consequence of the COVID-19 pandemic, the Investigator must discuss with the Medical Monitor potential mitigation.

4.2 Scientific Rationale for Study Design

The study population will include adult men and women, ages 18 to 65 years inclusive, who meet the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-V) diagnostic criteria for major depressive disorder (MDD) without psychotic features confirmed by the Mini-International Neuropsychiatric Interview (MINI). Based on published data from racemic ketamine, R-ketamine may have the potential for adverse fetal effects if administered to pregnant women. Therefore, female subjects of childbearing potential should be adequately protected from becoming pregnant and pregnant women should not be enrolled in clinical studies with PCN-101.

Two doses (30 mg and 60 mg) of IV PCN-101 will be compared with placebo to determine the most effective dose in improving depressive symptoms in subjects with TRD.

Randomization will be used to minimize the bias in the assignment of subjects to treatment arms and to increase the likelihood that known and unknown subject characteristics are evenly balanced across treatment arms. This will improve the validity of the statistical comparisons among treatment arms. Blinded treatment will be used to reduce any potential bias during the collection of data and the analysis of clinical endpoints.

PCN-101 30 mg and 60 mg will be administered as an IV infusion over 40 minutes. These doses are within the range shown to be well-tolerated in a Phase 1 study in healthy volunteers.

The World Health Organization reports that depression is the leading cause of disability and loss of work worldwide, affecting more than 300 million people (26). In severe cases, it can lead to suicide, which has become the third leading cause of death in the US with its incidence having increased 30% over the past 10 years (27, 28). The prevalence of TRD, generally defined as treatment failure with adequate trials of at least 2 antidepressant regimens, remains very high. A multicenter European study found that 51% of depressed patients recruited from specialist referral centers met such criteria (29). In addition, much of the cost and disability associated with depression is accounted for by TRD (30). Finally, it should be noted that many patients with MDD continue to have residual symptoms even without meeting the formal diagnosis of TRD, thus adding to the unmet burden.

Primary and secondary endpoints for this study will be to determine the efficacy of 2 doses of IV PCN-101 compared with placebo in improving depressive symptoms in subjects with TRD as assessed by change from baseline in the MADRS total score. The 10-item clinician-administered MADRS was designed to be used in subjects with MDD to measure the overall severity of depressive symptoms. The MADRS scale is validated, reliable, and acceptable to regulatory health authorities as a primary scale to determine efficacy in major depression.

Other secondary endpoints for this study will include scales such as:

- The HAM-D is a multiple-item questionnaire designed to provide an indication of depression in adults and used as a guide to evaluate recovery. The questionnaire is designed to rate the severity of depression by probing mood, feelings of guilt, suicide ideation, insomnia, agitation or retardation, anxiety, weight loss, and somatic symptoms.
- The QIDS-SR-16, which is a 16-item, self-reported scale of depression that has very similar sensitivity to the Inventory of Depressive Symptomatology Self-Report - 30 Items and the Hamilton Rating Scale for Depression - 24 Items in detecting changes in symptoms of depression.

- The GAD-7, which is a self-administered 7-item scale that is used to diagnose anxiety and has sensitivity and specificity as a screener for panic, social anxiety, and posttraumatic stress disorder.
- The CGI-S and CGI-I scales, which will allow the assessment of minimal clinically important differences using an anchor-based approach calculated from the global impressions of the clinician and the subject.
- The EQ-5D-3L, which will be used by subjects as a widely accepted, self-completion instrument to assess health-related quality of life for the domains of mobility, capacity for self-care, conduct of usual activities, pain/discomfort, and anxiety/depression.

Safety evaluations will include:

- Standard assessments of vital signs, 12-lead ECG, SpO_2, clinical hematology and chemistry assays, treatment-emergent adverse events (TEAEs), prior and concomitant medications, physical examination, height, weight, body mass index (BMI), serum and urine pregnancy tests, and urine drug and alcohol screens.
- The MOAA/S, which will be used to determine if PCN-101 causes the side effect of sedation and to what degree.
- The CADSS, which will be used to assess the alertness or dissociative state of subjects administered PCN-101.
- The BPRS+, which is one of the oldest, most widely used rating scales to measure psychotic symptoms and will be used by the study staff to measure psychiatric symptoms such as depression, anxiety, hallucinations and unusual behavior.
- The 5D-ASC, which will be used to assess the quality of acute psychological effects of the PCN-101 infusion.
- The C-SSRS, which will be used to assess the emergence of suicidal ideation. This instrument has been used frequently in clinical studies, and it is a standard measure for suicidal ideation assessment; its use is in accordance with US Food and Drug Administration guidance.

4.3 Justification for Dose

Study PCN-101-02 was a randomized, placebo-controlled, double-blind, single ascending dose study of the safety, tolerability, and pharmacokinetics of PCN-101 that included a relative safety comparison of PCN-101 and S-ketamine. Healthy subjects were administered a single IV infusion of PCN-101 at total doses of 5, 15, 30, 60, 100, and 150 mg or placebo over a 40-minute period. The safety and tolerability of PCN-101 at different doses and of 15 mg S-ketamine was assessed using reported AEs, changes in hematology and serum chemistry, changes in vital signs

and physical examination findings, ECG and pulse oximetry findings, sedation (as measured by the MOAA/S), and the development of dissociative or perceptual changes (as measured by the CADSS, BPRS, and 5D-ASC).

The study evaluated 58 healthy subjects. Forty-eight (48) subjects were randomized to 6 dose levels of PCN-101 (N=36) or placebo (N=12) in the single-ascending dose portion of the study. Ten additional subjects were evaluated in a parallel and cross-over relative safety comparison of 150 mg PCN-101 (N=10) and 15 mg S-ketamine (N=10).

The primary objective was to identify an acceptable tolerated dose of PCN-101 and compare this dose to the safety profile of 15 mg S-ketamine. Dose ranging using single ascending doses of PCN-101-02 demonstrated that doses of PCN-101 ≤ 150 mg had acceptable safety profiles as single IV infusions. [redacted] Both 30 mg and 60 mg doses of PCN-101 resulted in fewer transient elevations in mean systemic blood pressure, less sedation, fewer psychological or psychotic and dissociative effects, and fewer episodes of altered consciousness than 100 mg or 150 mg doses of PCN-101.

Based on the results of study PCN-101-02, 30 mg and 60 mg of PCN-101 were selected to be evaluated in this proof-of-concept study.

4.4 End of Study Definition

The duration of the study will be up to 29 days per subject, which includes a screening period, a 3-day in-clinic visit, and 2 follow-up visits. A subject will be considered to have completed the study if he/she has completed all phases of the study including the last visit.

The end of the study is defined as the date of the last visit of the last subject in the study.

5.0 STUDY POPULATION

Prospective approval of protocol deviations to recruitment and enrollment criteria, also known as protocol waivers or exemptions, is not permitted.

5.1 Inclusion Criteria

Subjects are eligible to be included in the study only if all of the following criteria apply:

1. Be capable of giving and give signed informed consent, which includes compliance with the requirements and restrictions listed in the Informed Consent Form (ICF) and in this protocol.
2. Be male or female 18 to 65 years of age inclusive at the time of signing the ICF.
3. Weigh ≥ 50 kg and have a BMI ≥ 18 and ≤ 35.
4. Have a diagnosis of recurrent MDD without psychotic features per the DSM-V, confirmed by the MINI.
5. Have an HAM-D total score > 20 at screening and baseline (Day -1).
6. Have an inadequate response to at least 2 antidepressants in the current episode of depression that were each given for ≥ 6 weeks at an adequate dose as defined by the Massachusetts General Hospital Antidepressant Response Questionnaire (MGH-ATRQ).
7. Must be on stable oral antidepressant treatment without a dose change for at least 30 days before screening (a missed dose, or reasonable number of missed doses per Investigator's discretion, in that period does not exclude a subject).
8. A male subject must be medically confirmed sterile for at least 6 months prior to screening or agree to use highly effective contraception (see Appendix 5) during the treatment period and for at least 3 months after the last dose of study treatment and refrain from donating sperm during this period. If a male with a partner who is of childbearing potential (OCBP) is included, his partner also needs to use highly effective birth control measures.
9. A female subject is eligible to participate if she is not pregnant, not breastfeeding, and at least 1 of the following conditions applies:
    - Not of childbearing potential (see Appendix 5).
    - A subject OCBP who agrees to follow the highly effective contraceptive guidance (see Appendix 5) on highly effective birth control measures during the treatment period and for at least 3 months after the last dose of study treatment and refrain from donating eggs during this period.

10. Be medically stable on the basis of physical examination, medical history, vital signs, and 12-lead ECG performed at screening. If there are abnormalities, the subject may be included only if the Investigator judges the abnormalities not to be clinically important. This determination must be recorded in the subject's source documents and initialed and dated by the Investigator.

5.2 Exclusion Criteria

Subjects are excluded from the study if any of the following criteria apply:

1. History of, or current signs and symptoms of, diseases or conditions that would make participation not be in the best interest (eg, compromise the well-being) of the subject or that could prevent, limit, or confound the protocol-specified assessments.

2. History of moderate or severe head trauma (for example, loss of consciousness for more than 15 minutes) or other neurological disorders (including a diagnosis of epilepsy or has had a seizure in the last 6 months), neurodegenerative disorder (Alzheimer's disease, Parkinson's disease, multiple sclerosis, Huntington's disease, etc.) or systemic medical diseases that are, in the opinion of the Investigator, likely to interfere with the conduct of the study or confound the study assessments. A history of febrile seizures in childhood is not exclusionary.

3. Has a primary DSM-V diagnosis of current (active) MDD with psychotic features, panic disorder, obsessive compulsive disorder, posttraumatic stress disorder, anorexia nervosa, or bulimia nervosa. Comorbid anxiety or panic disorder that does not dominate the clinical presentation is acceptable.

4. Has a current or prior DSM-V diagnosis of a primary psychotic disorder (eg, schizophrenia), bipolar or related disorders (confirmed by the MINI), intellectual or autism spectrum disorder, or borderline personality disorder.

5. Has any significant disease or disorder (eg, cardiovascular, pulmonary, gastrointestinal, hepatic, renal, neurological, musculoskeletal, endocrine, metabolic, malignant, psychiatric, major physical impairment) that, in the opinion of the Investigator, may either put the subject at risk because of participation in the study, influence the results of the study, or affect the subject's ability to participate in the study.

6. Has uncontrolled hypertension, despite medication, at Screening (systolic blood pressure [SBP] > 160 mm Hg or diastolic blood pressure [DBP] > 90 mm Hg) or any past history of hypertensive crisis. An abnormal blood pressure value at screening may be repeated once after 10-15 minutes of relaxation to determine the subject's eligibility.

7. Has an abnormal ECG of clinical relevance at screening or baseline (Day -1) including, but not limited to, the following:

- QT interval corrected according to Fridericia's formula (QTcF) interval > 450 msec for male subjects and >470 msec for female subjects.
- Evidence of 2nd and 3rd degree atrioventricular block, complete left bundle branch block (LBBB), or complete right bundle branch block (RBBB).
- Features of new ischaemia.
- Arrhythmia (except premature atrial contractions [PACs] and premature ventricular contractions [PVCs]).
- Has a history of risk factors including hypokalemia or a family history of Long QT Syndrome.

8. Has known history of, or positive serology for, human immunodeficiency virus (HIV); has a positive hepatitis B surface antigen, and/or confirmed current hepatitis C infection (positive hepatitis C virus [HCV] antibody confirmed with reflex HCV ribonucleic acid [RNA] test). Subjects with a history of hepatitis B vaccination without a history of hepatitis B are allowed to enroll.
9. Has a history of malignancy within the 5 years prior to screening (exceptions are squamous and basal cell carcinomas of the skin and carcinoma in situ of the cervix, or a malignancy that in the opinion of the Investigator, with concurrence with the Sponsor's Medical Monitor, is considered to have minimal risk of recurrence).
10. Has homicidal ideation/intent per the Investigator's clinical judgment; or has suicidal ideation with some intent to act within 1 month prior to the start of screening per the Investigator's clinical judgment or based on the C-SSRS, corresponding to a response of "Yes" on Item 4 (active suicidal ideation with some intent to act, without specific plan) or Item 5 (active suicidal ideation with specific plan and intent); or a history of suicidal behavior within the past year prior to the start of the screening/prospective observational phase.
11. Has had major surgery (eg, requiring general or local anesthesia) within the 4 weeks before screening, or will not have fully recovered from surgery or planned surgery during the time the subject is expected to participate in the study.
12. Has moderately impaired hepatic function at screening, defined as serum alanine aminotransferase (ALT) or aspartate aminotransferase (AST) > 2 × upper limit of normal (ULN) or total bilirubin (TBL) > 2 × ULN.
13. Has received any disallowed therapies as follows (see Section 6.5 for further details):

a. Receipt of a known potent inhibitor of hepatic cytochrome P450 (CYP) 2B6, or CYP3A, activity within 1 week or within a period 5 times the drug's half-life, whichever is longer, before the first administration of study drug on Day 1.

b. Treatment with a disallowed antipsychotic within the past 30 days prior to screening, except subjects who are on stable doses of quetiapine, aripiprazole, brexpiprazole, or olanzapine prescribed as adjunct treatment for depression (without psychosis) may be included in the study.

c. Any changes in psychotropic medication type or dose within the past 30 days prior to screening.

d. Treatment with monoamine oxidase inhibitors (MAOIs) currently or within the past 30 days of screening.

e. Doses of oral contraception should not contain more than 30 micrograms of ethinyl estradiol per day.

f. Refer to Appendix 6 for a list of permitted and prohibited concomitant medications. The Medical Monitor should be contacted for any questions regarding concomitant medications.

14. Has initiated psychotherapy (eg, Cognitive Behavior Therapy, Interpersonal Psychotherapy, Psychodynamic Psychotherapy other than psycho-education, or acupuncture within the past 90 days of screening. Patients planning to initiate individual or group therapy during the study are also not eligible.

15. Has received electroconvulsive therapy, transcranial magnetic stimulation, vagal nerve stimulation, deep brain stimulation, or other brain stimulation treatment within the past 4 weeks or currently used as either an acute or maintenance treatment of depression.

16. Has received any IP within 30 days or 5 half-lives prior to dosing with PCN-101.

17. Has a history of substance abuse (drug or alcohol) or dependence (except nicotine or caffeine) within the previous 6 months prior to the screening visit.

- Has a positive urine drug screen for ketamine, opiates, cocaine, barbiturates, and/or amphetamine/methamphetamine or positive alcohol screen at Screening or Day -1.

- Subjects who have a positive test result at screening due to prescribed opiates or amphetamines may be permitted to continue the screening phase if the prohibited medication is discontinued at least 1 week or 5 half-lives, whichever is longer, before the first dose of study medication. Retesting is not permitted for positive test result(s) from nonprescription use of drugs of abuse.

18. Has a history of previous nonresponse to ketamine, R-ketamine or S-ketamine, or has received 8 or more doses of ketamine, R-ketamine or S-ketamine in their lifetime.
19. Has a previous history of intolerance to ketamine, R-ketamine, or S-ketamine.
20. History of abuse of ketamine, R-ketamine, S-ketamine, or phencyclidine.
21. Subjects should not consume grapefruit, grapefruit juice, or Seville orange related products for 72 hours before IP administration and throughout the study.
22. Has the presence of clinically relevant long-term COVID-19 symptoms. Has current signs or symptoms of COVID-19.
23. COVID-19 vaccination is allowed as long as the doses are administered ≥ 30 days before study drug administration; vaccination is not allowed during the course of the study.

5.3 Lifestyle Considerations

No restrictions are required for this study.

5.3.1 Meals and Dietary Restrictions

Subjects should not consume grapefruit, grapefruit juice, or Seville orange-related products for 72 hours before PCN-101 administration and throughout the study.

5.3.2 Caffeine, Alcohol, and Tobacco

Subjects will abstain from alcohol for 24 hours prior to the start of the infusion of PCN-101 until 24 hours after dosing has been completed on Day 1. It is recommended that subjects abstain from alcohol use for the period of the study.

Subjects will abstain from using tobacco and nicotine products (except nicotine replacement therapies) for 4 hours prior to the start of the infusion of PCN-101 until 4 hours after dosing has been completed on Day 1.

5.3.3 Activity

Subjects will abstain from strenuous exercise while in the clinic. Subjects may participate in light recreational activities during studies (eg, watching television, reading).

5.4 Screen Failures

Screen failures are defined as subjects who consent to participate in the clinical study but are not subsequently entered in the study. A minimal set of screen failure information is required to ensure transparent reporting of screen failure subjects to meet the Consolidated Standards of Reporting Trials (CONSORT) publishing requirements and to respond to queries from regulatory authorities. Minimal information includes demography, screen failure details, eligibility criteria,

and any serious adverse event (SAE). Screen failure and rescreening information will be entered into the study's Electronic Data Capture System (EDC).

Individuals who do not meet the criteria for participation in this study (screen failure) may be rescreened once per the Investigator's judgement. However, subjects may not be rescreened if they did not meet diagnostic criteria, HAM-D minimum score, or had a positive test result(s) from nonprescription drugs of abuse on the first screening. Rescreened subjects should not be assigned the same subject number as for the initial screening. Rescreened subjects must first be registered as screen failures and subsequently registered as rescreens. Once the subject is registered as rescreened, a new screening window will begin. For all rescreens, all screening procedures, including ICF, must be repeated. Sites should discuss planned rescreens with the Medical Monitor prior to rescreening.

6.0 STUDY TREATMENT

Study treatment is defined as any investigational treatment(s), marketed product(s), placebo, or medical device(s) intended to be administered to a subject according to the study protocol.

The IP (also referred to as investigational medicinal product [IMP]) is PCN-101 Solution for Injection (R-ketamine hydrochloride in sterile water for injection). Subjects will receive PCN-101 Solution for Injection at either 30 mg or 60 mg or placebo.

A placebo control will be used in the study.

The active ingredient of the study treatment is R-ketamine. The International Union of Pure and Applied Chemistry (IUPAC) name is (2R)-2-(2-chlorophenyl)-2-(methylamino) cyclohexan-1-one.

Investigational product will be provided in vials. The site pharmacy will prepare 100 mL infusion bags containing IP. PCN-101 or placebo will be administered on Day 1 as a single IV infusion via an electronic infusion pump over 40 minutes (Table 2).

IP/IMP dose modifications are not permitted in this study.

The duration of study participation for each subject will be up to 29 days including a screening period, a 3-day in-clinic visit, and 2 follow-up visits.

6.1 Study Treatment(s) Administered

Table 2 Study Treatment Details

Study Treatment Name:	PCN-101 30 mg IUPAC name: (2R)-2-(2-chlorophenyl)-2-(methylamino) cyclohexan-1-one	PCN-101 60 mg IUPAC name: (2R)-2-(2-chlorophenyl)-2-(methylamino) cyclohexan-1-one	Placebo
Dosage Formulation:	Solution for injection	Solution for injection	Solution for injection
Unit Dose Strength(s)/Dosage Level(s):	30 mg	60 mg	n/a
Route of Administration	IV	IV	IV
Dosing Instructions:	Dilution with saline for infusion over 40 minutes, administered by site staff in the clinic	Dilution with saline for infusion over 40 minutes, administered by site staff in the clinic	Dilution with saline for infusion over 40 minutes, administered by site staff in the clinic
Packaging and Labeling	1 mL extractable study treatment will be provided in 2 mL vials. Each vial will be labeled as required per country.	1 mL extractable study treatment will be provided in 2 mL vials. Each vial will be labeled as required per country.	1 mL extractable study treatment will be provided in 2 mL vials. Each vial will be labeled as required per country.

Abbreviations: IUPAC = International Union of Pure and Applied Chemistry; IV = intravenous.

6.2 Preparation/Handling/Storage/Accountability

1. The Site Qualification Visit will confirm the knowledge and ability of the site staff to handle the study treatment in compliance with all local and national regulations regarding controlled substances. Before a site begins any work with PCN-101, the study monitor will provide training to the staff regarding licensing and registration requirements, record-keeping, and procedures for storage and disposal of controlled substances and waste products.

2. The Investigator or designee must confirm adherence to local regulations for the importation of controlled substances. The Investigator or designee must also ensure that appropriate temperature conditions have been maintained during transit for all study treatment received, and any discrepancies are reported and resolved before use of the study treatment.

3. Only subjects enrolled in the study may receive study treatment, and only authorized study center staff may supply and administer study treatment. All study treatments must be stored in a secure, environmentally controlled, and monitored (manual or automated) area in accordance with the labeled storage conditions with access limited to the Investigator and authorized study center staff and in compliance with local regulations for controlled substances.

4. PCN-101 and placebo must not be injected intravenously without dilution. The study treatment will be diluted with sodium chloride 0.9% United States Pharmacopeia according to the Pharmacy Manual.
5. The Investigator, institution, or the head of the medical institution (where applicable) is responsible for study treatment accountability, reconciliation, and record maintenance (ie, receipt, reconciliation, and final disposition records).
6. Further guidance and information for the final disposition of used and unused study treatment are provided in the Pharmacy Manual.

The Investigator, a member of the study center staff, or a hospital pharmacist must maintain an adequate record of the receipt and distribution of all study medication using a Drug Accountability Form. These forms must be available for inspection at any time.

6.3 Measures to Minimize Bias: Randomization and Blinding

All subjects will be centrally assigned to randomized study treatment using an Interactive Web Response System (IWRS). Before the study is initiated, the log in information and directions for the IWRS will be provided to each study center.

This is a double-blind study with limited access to the randomization code. The investigational treatment and placebo will be identical in physical appearance. The treatment each subject will receive will not be disclosed to the Investigator, study center staff, subject, Sponsor, or study vendors. The treatment codes will be held by the unblinded statistician and will be programmed into the IWRS.

The IWRS will be programmed with blind-breaking instructions. In case of an emergency, the Investigator has the sole responsibility for determining if unblinding of a subject's treatment assignment is warranted. Subject safety must always be the first consideration in making such a determination. If the Investigator decides that unblinding is warranted, the Investigator should make every effort to contact the Sponsor prior to unblinding a subject's treatment assignment unless this could delay emergency treatment of the subject. If a subject's treatment assignment is unblinded, the Sponsor must be notified within 24 hours after breaking the blind. The date and reason that the blind was broken must be recorded in the source documentation and case report form (CRF), as applicable.

6.4 Study Treatment Compliance

The prescribed dosage, timing, and mode of administration may not be changed, except as defined in Section 6.6. Any departures from the intended regimen must be recorded in the appropriate CRFs.

Noncompliance is defined as receiving less than the full infusion or a deviation from the 40-minute infusion time.

6.5 Concomitant Therapy

Any medication or vaccine (including over-the-counter or prescription medicines, vitamins, and/or herbal supplements) that the subject is receiving at the time of enrollment (within 30 days before the screening visit) or receives during the study must be recorded on the CRF along with:

- Type of drug.
- Reason for use.
- Dates of administration including start and end dates.
- Dosage information including dose and frequency.
- Route of administration.

COVID-19 vaccination is allowed as long as the doses are administered ≥ 30 days before the administration of study drug. Vaccination is not allowed during the course of the study.

Concomitant therapies must be recorded throughout the study beginning with the signing of the ICF (ie, screening) until the second follow-up visit. Concomitant therapies should also be recorded beyond this time only in conjunction with new or worsening AEs or SAEs. For subjects who fail screening, concomitant therapies do not need to be recorded unless there is an AE.

Modification of an effective pre-existing therapy should not be made for the explicit purpose of entering a subject into the study. The following medications are prohibited for the duration of the study beginning from 1 week (or 5 half-lives, whichever is longer) prior to Baseline (Day -1). If applicable, the possible effects of these medications on the efficacy endpoints will be considered during the assessment of the evaluable period (see Table 6 in Appendix 6 for further details).

- Receipt of a known potent inhibitor of hepatic cytochrome P450 (CYP) 2B6, or CYP3A, activity within 1 week or within a period 5 times the drug's half-life, whichever is longer, before the first administration of study drug on Day 1.
- Treatment with a disallowed antipsychotic within the past 30 days prior to screening, except subjects who are on stable doses of quetiapine, aripiprazole, brexpiprazole, or olanzapine prescribed as adjunct treatment for depression (without psychosis) may be included in the study.
- Any changes in psychotropic medication type or dose within the past 30 days prior to screening.
- Treatment with monoamine oxidase inhibitors (MAOIs) currently or within the past 30 days of screening.

The following medications are restricted for the duration of the study beginning from the date of the screening visit until the completion of assessments at Visit 4 (Day 15) or the discontinuation visit:

- Daily doses of benzodiazepine receptor agonists will be limited to 2 mg/day or less of lorazepam or equivalents. Benzodiazepines will be prohibited within 6 hours prior to dosing.

Short acting benzodiazepines except clobazam may be used as rescue medication to treat intolerable hallucinations/dissociative effects or any other symptoms in the judgement of the Investigator.

Rescue medications may be used for other symptoms including anxiety/agitation, nausea/vomiting, dissociation, and blood pressure/hypertension according to the judgement of the Investigator.

Doses of oral contraception should not contain more than 30 micrograms of ethinyl estradiol per day.

Nonpharmacologic therapies must also be recorded throughout the study (eg, special diets, exercise regimens, occupational therapy, etc.) and must be recorded in the CRF. In addition, they must be stable 4 weeks prior study entry and remain stable throughout the study.

Patients undergoing regular psychotherapy (ie, at least 90 days duration at the time of Screening) are eligible to participate in the study.

The Medical Monitor should be contacted if there are any questions regarding concomitant or prior therapy.

6.6 Dose Modification

IMP/IP dose modification is not permitted during the study.

6.7 Treatment after the End of the Study

Subjects will return to their normal standard of care upon completion of the study.

7.0 DISCONTINUATION OF STUDY TREATMENT AND SUBJECT DISCONTINUATION/WITHDRAWAL

7.1 Discontinuation of Study Treatment

If an AE occurs during the administration of study treatment, discontinuation of the infusion may occur if the Investigator believes that it is in best interest of the subject. The subject will then be withdrawn from the study.

If a subject who does not meet the enrollment criteria is inadvertently administered study treatment, that subject may be discontinued from the study and the Sponsor or Sponsor designee must be contacted. An exception may be granted in rare circumstances for which there is a compelling reason to allow the subject to continue. In these rare cases, the Investigator must obtain documented approval from the Sponsor or Sponsor designee to allow the subject to continue in the study.

Subjects who discontinue study treatment will not be replaced.

See the Schedule of Activities (Section 1.3) for the data to be collected at the time of treatment discontinuation and follow-up and for any further evaluations that need to be completed.

7.2 Subject Discontinuation/Withdrawal from the Study

A subject may withdraw from the study at any time at his/her own request or may be withdrawn at any time at the discretion of the Investigator for safety, behavioral, compliance, or administrative reasons.

Any female subject who becomes pregnant while participating in the study will be withdrawn from the study.

If the subject withdraws consent for disclosure of future information, the Sponsor may retain and continue to use any data collected before such a withdrawal of consent.

If a subject withdraws from the study, he/she may request destruction of any samples taken and not tested, and the Investigator must document this in the study center study records.

See Section 1.3 for data to be collected at the time of study discontinuation and follow-up and for any further evaluations that need to be completed.

7.3 Lost to Follow-up

A subject will be considered lost to follow-up if he or she repeatedly fails to return for scheduled visits and is unable to be contacted by the study center.

The following actions must be taken if a subject fails to return to the clinic for a required study visit:

- The study center must attempt to contact the subject and reschedule the missed visit as soon as possible and counsel the subject on the importance of maintaining the assigned visit schedule and ascertain whether or not the subject wishes to and/or should continue in the study.
- Before a subject is deemed lost to follow-up, the Investigator or designee must make every effort to regain contact with the subject (where possible, 3 telephone calls and, if necessary, a certified letter to the subject's last known mailing address or local equivalent methods). These contact attempts should be documented in the subject's medical record.
- If the subject continues to be unreachable, he/she will be considered to have withdrawn from the study.

8.0 STUDY ASSESSMENTS AND PROCEDURES

Investigator or his/her qualified designee must have completed the sponsor specified training for each rating scale (where appropriate), before any rating scales are completed, and some additional training certification may be required prior to site activation. To maintain the study blind, the staff member that completes the efficacy scales should not complete any safety scales, except for C-SSRS, or assessment of adverse events for any given subject. Similarly, the staff member that completes the safety scales should not complete any efficacy scales for any given subject. To ensure consistency, the same staff member (where possible) should complete the questionnaires for each subject throughout the study.

All post-dose timepoints in the Schedule of Activities (Section 1.3) are relative to the start of the infusion, except for the 40-minute (end-of-infusion [EoI]) timepoint. EoI procedures should occur 40 minutes after the start of infusion or at the end of infusion if the infusion continues beyond 40 minutes.

EoI procedures include 12-lead ECG, vital signs, pulse oximetry, CADSS, and BPRS+. MOAA/S continues on 20 minute intervals from the start-of-infusion and AE review will be continuous from predose to 4 hours postdose.

For the EoI, a time window of ± 2 minutes after the 40-minute total time is allowed. Assessments and procedures, however, should be started as close as possible after EoI.

From predose on Day 1 to the end of the study, the highest priority procedures will be performed closest to the nominal time. Where activities at a given timepoint coincide, consideration should be given to ensure that the following order of activities is maintained. Deviations are not required when the actual order differs from the following.

- Efficacy rating scales. Where more than one efficacy scale is assessed at a timepoint, the order of assessments should be MADRS, CGI-I and CGI-S, QIDS-SR-14/16, HAM-D, GAD-7 and EQ5D-3L.
- 12-lead ECGs
- Vital signs and pulse oximetry
- Safety rating scales. Where more than one safety scale is assessed at a timepoint, the order of assessments should be MOAA/S, CADSS and BPRS+.
- AE and concomitant medication review.
- Physical examination
- Clinical laboratory assessments (including urine pregnancy test and drug/alcohol screen).

Study procedures and their timing are summarized in the Schedule of Activities in Section 1.3.

Protocol waivers or exemptions are not allowed.

Immediate safety concerns should be discussed with the Sponsor immediately upon occurrence or awareness to determine if the subject should continue or discontinue study treatment.

Adherence to the study design requirements, including those specified in the Schedule of Activities, is essential and required for study conduct.

All screening evaluations must be completed and reviewed to confirm that potential subjects meet all eligibility criteria. The Investigator will maintain a screening log to record details of all subjects screened and to confirm eligibility or record reasons for screening failure, as applicable.

Procedures conducted as part of the subject's routine clinical management (eg, blood count) and obtained before signing of the ICF may be utilized for screening or baseline purposes provided the procedures met the protocol-specified criteria and were performed within the time frame defined in the Schedule of Activities.

Screening and/or baseline procedures will include:

- Obtain informed consent.
- Confirm that the subject meets all eligibility criteria.
- Record the subject's demographic information.
- Perform a physical exam (include recording of the subject's height, weight, and BMI).
- Record the subject's medical history.
- Conduct screening tests for HIV, hepatitis B, and hepatitis C.
- Record the subject's prior and concomitant medications.
- For subjects OCBP, perform a serum pregnancy test at screening and a urine pregnancy test at baseline.
- Conduct laboratory assessments.
- Conduct a 12-lead ECG, vital signs, pulse oximetry.
- Conduct a urine alcohol and drug screen (at a minimum will include ketamine, opiates, cocaine, barbiturates, and/or amphetamine/methamphetamine).
- Review AEs.
- Perform a MINI version 7.0.2 to assess the 17 most common psychiatric disorders in DSM-III-R, DSM-IV, DSM-V, and ICD-10. The MINI is designed as a brief structured diagnostic interview to meet the need for a short but accurate structured psychiatric

interview for multicenter clinical trials and epidemiology studies and to be used as a first step in outcome tracking in non-research clinical settings. The MINI is a structured interview in which patients are asked to answer questions "Yes" or "No" (eg, "Were you ever depressed or down, or felt sad, empty or hopeless most of the day, nearly every day, for two weeks?"). The MINI is designed to map onto diagnoses defined by the DSM-5 (37).

- Perform an MGH-ATRQ, HAM-D, MADRS, and C-SSRS.

8.1 Efficacy Assessments

Efficacy assessments will be conducted at the time points listed in the Schedule of Activities in Section 1.3.

8.1.1 Montgomery Åsberg Depression Rating Scale

The 10-item MADRS will be administered by the study staff to measure the overall severity of depressive symptoms. The MADRS total score will be assessed at screening and baseline (Day 1 predose).

- The primary efficacy endpoint of improvement in the MADRS total score will be evaluated prior to discharge on Day 2 of Visit 2, 24 hours after the start of the infusion of study treatment.
- The MADRS total score will also be assessed at 2 hours, 4 hours, 7 days (on Day 8), and 14 days (on Day 15) after the start of the infusion of PCN-101 to evaluate the secondary efficacy endpoints of (1) response (defined as ≥ 50% improvement in the MADRS total score from predose) and (2) remission (defined as MADRS total score ≤ 10) in the improvement of depressive symptoms in the subjects.
- While the MADRS is typically used with a 7-day recall period, versions of the MADRS including 2-hour and 24-hour recall periods will be used in this study. (For the 2 hour and 4 hour periods, the sleep and appetite items will not be assessed; predose scores for these items obtained on the same day will be carried forward).

8.1.2 Hamilton Depression Rating Scale

The Hamilton Depression Rating Scale is a 17-item questionnaire designed to assess a subject's level of depression before, during, and after treatment. The survey is used as a guide to evaluate recovery and is based on the clinician's interview with the subject. The HAM-D rates the severity of depression by probing mood, feelings of guilt, suicide ideation, insomnia, agitation or retardation, anxiety, weight loss, and somatic symptoms. Scoring take about 15 minutes. The rater enters a number for each symptom that ranges from 0 (not present) to 4 (extreme symptoms). During this study, the questionnaire will be completed at screening, baseline, predose on Day 1, at Day 2 discharge, and both follow-up visits.

8.1.3 Quick Inventory of Depressive Symptomatology - Self Report

Subjects will answer the questions in the QIDS-SR-16, an instrument that is sensitive in detecting changes in the symptoms of depression. The 16 items correlate with the 9 DSM-V symptom criterion domains that include:

- Sleep disturbance - initial, middle, and late insomnia or hypersomnia (Questions 1 to 4)
- Sad mood (Question 5)
- Decrease/increase in appetite/weight (Questions 6 to 9)
- Concentration (Question 10)
- Self-criticism (Question 11)
- Suicidal ideation (Question 12)
- Interest (Question 13)
- Energy/fatigue (Question 14)
- Psychomotor agitation/retardation (Questions 15 and 16).

While the QIDS-SR-16 is typically used with a 7-day recall period, the QIDS-SR-14, with a 24-hour recall period will be used on Day 2. For the 24 hour recall periods, the weight and appetite items will not be assessed; predose scores for these items obtained on Day 1 predose will be carried forward. The QIDS-SR-16 item version will be used on Day 1 predose and Day 8 and Day 15 follow-up.

8.1.4 Generalized Anxiety Disorder 7-Item Scale

The GAD-7 is a self-reported questionnaire for the screening and measurement of the severity of generalized anxiety disorder. It has sensitivity and specificity as a screener for panic, social anxiety, and posttraumatic stress disorder. The GAD-7 is comprised of 7 items that measure the severity of various signs of GAD according to reported response categories with assigned points. The scores for all 7 items are added together to obtain the total score on which the assessment of anxiety is made.

8.1.5 Clinical Global Impression

The CGI rating scales measure symptom severity, treatment response, and the efficacy of treatments in studies of patients with mental disorders. The questionnaires require the study staff to compare the subjects to typical patients in the clinician’s experience.

- The CGI-S assesses minimal clinically important differences in treatments with respect to the severity of depression symptoms.

- The CGI-I assesses minimal clinically important differences in treatments with respect to the improvement in depression symptoms.

8.1.6 European Quality - 5 Dimensions - 3 Levels

EQ-5D-3L (a questionnaire to assess the health-related quality of life for the domains of mobility, capacity for self-care, conduct of usual activities, pain/discomfort, and anxiety/depression).

The EQ-5D-3L is a questionnaire that essentially consists of 2 pages - the EQ-5D descriptive system and a visual analog scale (VAS). The descriptive system is comprised of 5 dimensions: mobility, self-care, usual activities, pain/discomfort, and anxiety/depression. Each dimension has 3 levels: no problems, some problems, and extreme problems. The subject will be asked to indicate his/her health state by marking the box for the most appropriate statement in each of the 5 dimensions. The VAS records the subject's self-rated health on a vertical VAS where the endpoints are labeled "best imaginable health state" and "worst imaginable health state." This information will be used as a quantitative measure of health outcome.

8.2 Safety Assessments

The planned time points for all safety assessments are provided in the Schedule of Activities in Section 1.3.

8.2.1 Physical Examinations

A complete physical examination will include, at a minimum, assessments of the cardiovascular, respiratory, gastrointestinal, and neurological systems. Height and weight will also be measured and recorded and BMI will be calculated and recorded at screening.

8.2.2 Vital Signs

Tympanic or temporal temperature, heart rate, blood pressure (SBP/DBP), respiratory rate, and SpO_2 will be assessed.

Blood pressure and pulse measurements will be assessed with a completely automated device while the subject is supine or semi-supine. Manual techniques will be used only if an automated device is not available. Blood pressure and pulse measurements should be preceded by at least 5 minutes of rest for the subject in a quiet setting without distractions (eg, television, cell phones). An abnormal blood pressure value at screening may be repeated once after 5 minutes of relaxation to determine the subject's eligibility. Blood pressure must be recorded using the same arm and position (supine or semi-supine) throughout the study, where possible.

Vital signs (to be taken before blood collection for laboratory tests) will consist of 1 pulse and 3 blood pressure measurements (3 consecutive blood pressure readings will be recorded at

intervals of at least 1 minute). The average of the 3 blood pressure readings will be recorded in the CRF.

A pulse oximeter will be used to measure peripheral capillary oxygen saturation (SpO_2) continuously from predose until 4-hours postdose. A pulse oximeter will be clipped onto the finger or foot of the subject, and light will be sent through the finger or foot and measured on the other side.

8.2.3 Concomitant Medication Review

Concomitant medications will be recorded at the timepoints indicated in Section 1.3 and throughout the study until the second follow up visit. See Section 6.5 for further details.

8.2.4 Electrocardiograms

12-Lead ECGs will be obtained as outlined in the Schedule of Activities (see Section 1.3) using an ECG machine that automatically calculates the heart rate and measures PR, QRS, QT, and corrected QT (QTc) intervals. If abnormal, the ECG needs to be repeated. All ECGs will be centrally reviewed by an independent cardiologist blinded to treatment allocation; however, central review of the baseline (Day -1) ECG is not required prior to dosing on Day 1.

8.2.5 Clinical Safety Laboratory Assessments

See Appendix 3 for the list of clinical laboratory tests to be performed and to the Schedule of Activities (Section 1.3) for the timing and frequency.

The Investigator must review the laboratory report, document this review, and record any clinically relevant changes occurring during the study in the AE section of the CRF. The laboratory reports must be filed with the source documents. Clinically significant abnormal laboratory findings are those that are not associated with the underlying disease, unless judged by the Investigator to be more severe than expected for the subject's condition.

All laboratory tests with values considered clinically significantly abnormal during participation in the study or within 15 days after the last dose of study treatment should be repeated until the values return to normal or baseline or are no longer considered clinically significant by the Investigator or Medical Monitor.

- If such values do not return to normal/baseline within a period of time judged reasonable by the Investigator, the etiology should be identified, and the Sponsor should be notified.
- All protocol-required laboratory assessments, as defined in Appendix 3, must be conducted in accordance with the laboratory manual and the Schedule of Activities.
- If laboratory values from non-protocol specified laboratory assessments performed at the institution's local laboratory require a change in subject management or are considered

clinically significant by the Investigator (eg, SAE, AE, or dose modification), then the results must be recorded in the CRF.

8.2.6 Modified Observer's Assessment of Alertness/Sedation

The MOAA/S will be used to determine if PCN-101 causes the side effect of sedation and to what degree. Sedation will be evaluated by the study staff using the MOAA/S. The observer will record the subject's alertness predose, every 20 minutes for first 2 hours postdose, and at 4 and 6 hours after the start of the infusion.

8.2.7 Clinician-Administered Dissociative States Scale

The CADSS will be used to assess the alertness or dissociative state of subjects administered PCN-101. The study staff will evaluate the subject's degree of dissociation by administering the CADSS questionnaire predose; at 40 minutes postdose (at EoI); and at and 2 and4 hours postdose.

8.2.8 Brief Psychiatric Rating Scale - Modified 4 Components

The BPRS+ is one of the oldest, most widely used rating scales to measure psychotic symptoms and will be used by the study staff to measure psychiatric symptoms such as depression, anxiety, hallucinations, and unusual behavior. Only the four-item positive symptom subscale (consisting of: suspiciousness, hallucinations, unusual thought content, and conceptual disorganization) will be used in the study to assess treatment emergent psychotic symptoms. The questionnaire will be administered predose; at 40 minutes postdose (at EoI); and at 2 and 4hours postdose.

8.2.9 5-Dimensional Altered States of Consciousness Rating Scale

The 5D-ASC will be used to assess the quality of any acute psychological effects of the PCN-101 infusion. It is a retrospectively assessed questionnaire to measure subjective experiences of altered states of consciousness and contains 94 items that are formulated as a visual analog scale. The test will be performed 6 hours after the start of the infusion of PCN-101.

8.2.10 Suicidal Risk Monitoring

PCN-101 is being investigated as an antidepressant/central nervous system-active study treatment. Although there has been some concern that these types of treatments may be associated with an increased risk of suicidal ideation or behavior when given to some subjects with MDD, other studies have indicated that ketamine may be associated with a decrease in suicidal ideation or behavior. Therefore, the Sponsor considers it important to monitor suicidal ideation and behavior and treatment-emergent suicidal ideation and behavior during the study.

The definitions of behavioral suicidal events used in this scale are based on those used in the Columbia-Suicide History Form. Questions are asked on suicidal behavior, suicidal ideation, and intensity of ideation. At screening (Visit 1), questions will be in relation to lifetime experiences.

Questioning at all subsequent visits (see Section 1.3, Schedule of Activities) will be in relation to the last assessment (since last visit). The adult C-SSRS will be used for all subjects.

The C-SSRS will be administered to subjects at screening, at baseline, and prior to discharge from the clinic 24 hours after the infusion of study treatment. The C-SSRS will be administered to subjects at follow-up Visit 3 (Day 8) and Visit 4 (Day 15). Note: the C-SSRS may be evaluated by either efficacy or safety rater.

8.3 Adverse Events

The definitions of an AE and SAE can be found in Appendix 4.

AEs will be reported by the subject (or, when appropriate, by a caregiver, surrogate, or the subject's legally authorized representative).

The Investigator and any designees are responsible for detecting, documenting, and recording events that meet the definition of an AE or SAE and remain responsible for following up AEs that are serious, considered related to the study treatment or study procedures, or that caused the subject to discontinue from the study (see Section 7.0).

8.3.1 Time Period and Frequency for Collecting Adverse Event and Serious Adverse Event Information

All AEs will be collected from the signing of the ICF until the second follow-up visit (Day 15) at the time points specified in the Schedule of Activities (Section 1.3).

Medical occurrences that begin before the start of study treatment but after obtaining informed consent will be recorded on the Medical History/Current Medical Conditions section of the CRF, not the AE section.

All SAEs will be recorded and reported to the Sponsor or designee within 24 hours as indicated in Appendix 4. The Investigator will submit any updated SAE data to the Sponsor or designee within 24 hours of it being available.

Investigators are not obligated to actively seek AE or SAE information after the conclusion of study participation. However, if the Investigator learns of any SAE, including a death, at any time after a subject has been discharged from the study, and he/she considers the event to be reasonably related to the study treatment or study participation, the Investigator must promptly notify the Sponsor or designee.

The method of recording, evaluating, and assessing the causality of AEs and SAEs and the procedures for completing and transmitting SAE reports are provided in Appendix 4.

8.3.2 Method of Detecting AEs and SAEs

Care will be taken not to introduce bias when detecting AEs and/or SAEs. Open-ended and non-leading verbal questioning of the subject is the preferred method to inquire about AE occurrences.

8.3.3 Follow-up of Adverse Events and Serious Adverse Events

After the initial AE/SAE report, the Investigator is required to proactively follow each subject at subsequent visits/contacts. All SAEs will be followed until resolution, stabilization, the event is otherwise explained, or the subject is lost to follow-up (as defined in Section 7.3). Further information on follow-up procedures is given in Appendix 4.

8.3.4 Regulatory Reporting Requirements for Serious Adverse Events

Prompt notification by the Investigator to the Sponsor or designee of an SAE is essential so that legal obligations and ethical responsibilities toward the safety of subjects and the safety of a study treatment under clinical investigation are met.

The Sponsor has a legal responsibility to notify both the local regulatory authority and other regulatory agencies about the safety of a study treatment under clinical investigation. The Sponsor will comply with country-specific regulatory requirements relating to safety reporting to the regulatory authority, Independent Ethics Committees (IECs), Independent Review Boards (IRBs), and Investigators.

Investigator safety reports must be prepared for suspected unexpected serious adverse reactions according to local regulatory requirements and Sponsor or designee's policy and forwarded to Investigators as necessary.

An Investigator who receives an Investigator safety report describing an SAE or other specific safety information (eg, summary or listing of SAEs) from the Sponsor will review and then file it along with the Investigator's Brochure and will notify the IEC/IRB, if appropriate according to local requirements.

8.3.5 Pregnancy

Details of all pregnancies in female subjects and, if indicated, female partners of male subjects will be collected after the start of study treatment and until the second follow-up visit.

If a pregnancy is reported, the Investigator should inform the Sponsor within 24 hours of learning of the pregnancy and should follow the procedures outlined in Appendix 5. Information on the status of the mother and child will be forwarded to the Sponsor. Generally, the follow-up will be no longer than 6 to 8 weeks following the estimated delivery date. Any termination of the pregnancy will be reported regardless of fetal status (presence or absence of anomalies) or indication for the procedure.

Abnormal pregnancy outcomes (eg, spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy) are considered SAEs.

8.3.6 COVID-19

If a subject tests positive for COVID-19, this should be recorded as either an AE of "symptomatic COVID-19 disease" or "asymptomatic COVID-19 disease."

Symptoms, signs, and sequelae of COVID-19 should be reported as an AE per the Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting (Appendix 4). Medications for the prevention or treatment of COVID-19 should be entered as concomitant medications. (Please refer to the EDC Completion Guidelines for further details.)

8.4 Treatment of Overdose

An overdose is unlikely during this study since all treatment will be administered in a clinic setting using an infusion pump. However, if the study treatment is administered in less than 40 minutes due to a pump error, the Investigator should:

1. Contact the Medical Monitor immediately.
2. Closely monitor the subject for any AE/SAE and laboratory abnormalities for at least 24 hours.
3. Obtain a plasma sample for pharmacokinetic (PK) analysis as soon as possible and note the time from the start of the infusion of study treatment if requested by the Medical Monitor (determined on a case-by-case basis).
4. Document the quantity of the excess dose as well as the duration of the overdose in the CRF.

8.5 Pharmacokinetics

No assessment of the PK of PCN-101 is planned in this study.

8.6 Pharmacodynamics

Pharmacodynamic parameters will not be evaluated in this study.

8.7 Genetics

Genetics will not be evaluated in this study.

8.8 Biomarkers

Biomarkers will not be evaluated in this study.

8.9 Health Economics and/or Medical Resource Utilization and Health Economics

Health economic and/or medical resource utilization parameters will not be evaluated in this study.

9.0 STATISTICAL CONSIDERATIONS

9.1 General Considerations

The comprehensive SAP with detailed description of all statistical analyses will be developed and finalized before database lock.

All statistical analyses, including summary tables and data listings will be performed using SAS® software (version 9.4 or higher). Continuous endpoints will be summarized using descriptive statistics (number of subjects [n], mean, standard deviation [SD], median, minimum and maximum). Categorical endpoints will be summarized using frequency counts and percentages. All individual subject data will be presented in listings.

9.2 Sample Size Determination

The intent of the primary efficacy analysis is to demonstrate superiority of at least 1 therapeutic dose of PCN-101 Solution for Injection (30 mg or 60 mg) versus placebo based on the change in the MADRS total score from predose to 24 hours postdose.

For the primary analysis, a sample size of 93 randomized subjects (31:31:31) will provide 80% power to detect an 8-point difference between each PCN-101 Solution for Injection dose and placebo in the mean change from baseline MADRS total score at 24 hours postdose, using a t-test with $\alpha = 0.05$ (2-tailed) and assuming a common SD of 11.

Approximately 20 study centers in the EU and US are expected to participate in this study. The enrollment of 93 subjects is planned. Subjects who do not receive a dose of study treatment on Day 1 (Visit 2) will be replaced.

9.3 Populations for Analyses

For purposes of analysis, the analysis sets in Table 3 are defined.

Table 3 Analysis Sets

Analysis Set	Description
Full Analysis Set (FAS)	The FAS will consist of all randomized subjects who receive at least 1 dose of study treatment and have at least 1 postbaseline assessment available. This population will serve as the basis for efficacy analysis. Subjects will be analyzed according to their randomized treatment.
Safety Analysis Set	The Safety Population will consist of all randomized subjects who receive any study treatment, even a partial dose. This population will be used for all summaries of subject disposition,

	demographic and baseline data, and safety information including AE incidence. Subjects will be analyzed according to the treatment they actually received.

Abbreviations: AE = adverse event; FAS = Full Analysis Set.

9.4 Statistical Analyses

9.4.1 Subject Disposition and Demographics

Subject disposition events (screened, randomized, completed the study, early discontinuation from the study along with the reason for discontinuation) as well as the number of subjects in each analysis population will be summarized using frequency counts and percentages by treatment groups.

9.4.2 Efficacy Analyses

Primary Endpoint

The change from baseline in MADRS scores will be summarized separately by treatment arm for each visit. Associated baseline scores will be taken as the last corresponding measurement prior to the first dose of IMP in the treatment period (i.e., Visit 2, predose).

The primary endpoint of change from baseline MADRS total score at 24 hours post start of infusion will be evaluated using a mixed effects model for repeated measure (MMRM) analysis with observed cases only. The MMRM model will include fixed effects for treatment group, region, visit, study site and treatment group by visit interaction, with subject as a random effect and baseline score as a covariate. An unstructured covariance matrix will be used to estimate the variance-covariance structure within subjects across time points. If convergence is not obtained, then other covariance structures to be used will be specified in the SAP. Objective criteria for assessing normality assumptions and proposed alternative analyses will be specified in the SAP.

From this analysis the least squares (LS) mean estimates for each treatment arm at each visit, along with the standard error and 95% confidence intervals (CIs) will be presented separately. In addition, estimates of the treatment difference at each visit will be presented along with standard errors of the difference and 95% CIs. The primary comparison for the MADRS is the estimate of the treatment difference at 24 hours after the start of the infusion.

Secondary endpoints

The change from baseline to Day 14 in MADRS, HAM-D, GAD-7 and QIDS-SR scores will be analyzed using similar model approaches as for MADRS. Observed and change from baseline values in continuous secondary endpoints will be summarized descriptively by treatment group. Continuous secondary efficacy endpoints assessed at more than 1 postbaseline visit will be analyzed using an MMRM analysis. The MMRM model will include fixed effects for treatment group, region, visit, study site and treatment group by visit interaction, with subject as a random effect and baseline score as a covariate.

From this analysis the LS mean estimates for each treatment arm at each visit, along with the standard error and 95% CIs will be presented separately. In addition, estimates of the treatment

difference at each visit will be presented along with standard errors of the difference and 95% CIs.

The numeric values of CGI-S and CGI-I assessments will be analyzed separately using similar model approaches as for MADRS. For CGI-S, baseline score will be included as a covariate. Both values for the original categorical scale and the converted numerical scale at each visit (including change from baseline for the CGI-S numerical scale) will be summarized using standard summary statistics.

Binary exploratory endpoints will be analyzed separately and odds ratios, 95% CI and p-value will be presented.

Full details of the efficacy analyses and any further supplementary analyses deemed appropriate will be provided in the SAP.

9.4.3 Safety Analyses

All safety analyses will be performed on the Safety Analysis Set.

Adverse Events

Adverse events will be coded using the Medical Dictionary for Regulatory Activities (MedDRA; Version 23.0 or higher). Treatment-emergent adverse events (TEAEs) are AEs with onset or worsening after the start of study treatment. The AE summaries will be primarily based on TEAEs. The number and percentage of subjects with TEAEs will be summarized by treatment group, system organ class, and preferred term for all TEAEs, treatment-related TEAEs, SAEs, serious TEAEs, severe TEAEs, TEAEs of special interest, and all TEAEs leading to study drug discontinuation. All TEAEs will be further summarized by maximum severity and causality. The TEAEs of special interest will be identified in the SAP.

All AEs will be presented in a by-subject listing. Serious AEs, severe AEs, and AEs leading to study discontinuation or death will be presented in separate listings.

Clinical Laboratory Tests

Laboratory data will be summarized using descriptive statistics for each parameter, including change from baseline at each assessment time by treatment group. Frequency tables or shift tables will be used to present the number and percentage of subjects with laboratory values within/outside reference ranges. All laboratory data will be present in the listings. A by-subject listing will also be provided for subjects with abnormal laboratory results.

Electrocardiogram

Descriptive statistics will be used to summarize the observed and change from baseline values of the following ECG parameters at each assessment time by treatment group: heart rate, PR interval, QRS interval, QT interval, and QTcF. All ECG data will be present in listings.

Vital Signs

Vital sign parameters including temperature, pulse/heart rate, respiratory rate, SpO_2, and blood pressure will be summarized descriptively for both the observed and change from baseline values at each assessment time by treatment group. Frequency tabulations will be provided for abnormalities as applicable.

Other Safety Questionnaires and Assessments

All physical examination results will be listed by subject.

Prior and concomitant therapies will be summarized separately and tabulated for each treatment group using frequencies and percentages. Percentages will be based on the number of subjects in each treatment group. Prior and concomitant medications will be listed by subject.

For the C-SSRS: suicidal ideation and behavior scores at each assessment time as well as the incidence rate of TEAEs will be summarized using frequency counts and percentages by treatment group.

For the CADSS, BPRS+, 5D-ASC, and MOAA/S: Descriptive statistics of scores and their changes from baseline will be summarized at each assessment time by treatment group.

Pregnancy test and urine drug and alcohol screen will be presented in by-subject listings.

9.4.4 Other Analyses

Exploratory analyses may be done if indicated.

9.4.5 Missing Data

In general, the missing data which is observed from this study will be assumed to be missing-at-random (MAR).

Since subjects are hospitalized at the time of the primary endpoint, it is anticipated that missing data will be infrequent. No imputation will be applied to primary analysis.

Further details on the handling of missing data will be provided in the SAP.

9.5 Interim Analyses

No interim analysis for efficacy is planned.

9.6 Data Monitoring Committee

Because of the short timeframe of the study, there will not be a data monitoring committee.

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11.0 APPENDICES

Appendix 1 Abbreviations

Abbreviation	Definition
5D-ASC	5-Dimensional Altered States of Consciousness Rating Scale
AE	Adverse event
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
BMI	Body mass index
BPRS+	Brief Psychiatric Rating Scale - Modified 4 Components
CADSS	Clinician-Administered Dissociative States Scale
CFR	Code of Federal Regulations
CGI-I	Clinical Global Impression - Improvement
CGI-S	Clinical Global Impression - Severity
CI	Confidence interval
COVID-19	Coronavirus disease 2019
CRF	Case report form
CRO	Contract Research Organization
C-SSRS	Columbia Suicide Severity Rating Scale
CYP	Cytochrome P450
DBP	Diastolic blood pressure
DSM-V	Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition
ECG	Electrocardiogram
EDC	Electronic data capture
EoI	End-of-infusion
EQ-5D-3L	European Quality - 5 Dimensions - 3 Levels
EU	European Union
EudraCT	European Union Drug Regulating Authorities Clinical Trials (database)
FAS	Full Analysis Set
GAD-7	Generalized Anxiety Disorder 7-Item Scale
GCP	Good Clinical Practice
GGT	Gamma-glutamyl transferase
HAM-D	Hamilton Depression Rating Scale
HCV	Hepatitis C virus
HIV	Human immunodeficiency virus

Abbreviation	Definition
ICE	Intercurrent Event
ICF	Informed Consent Form
ICH	International Council for Harmonisation
IEC	Independent Ethics Committee
IMP	Investigational medicinal product
IRB	Independent Review Board
IUPAC	International Union of Pure and Applied Chemistry
IV	Intravenous
IWRS	Interactive Web Response System
LBBB	Left bundle branch block
LS	Least squares
LDH	Lactate dehydrogenase
MADRS	Montgomery Åsberg Depression Rating Scale
MAOI	Monoamine oxidase inhibitor
MAR	Missing-at-random
MDD	Major depressive disorder
MedDRA	Medical Dictionary for Regulatory Activities, Version 23.0 or higher
MGH-ATRQ	Massachusetts General Hospital Antidepressant Response Questionnaire
MINI	Mini-International Neuropsychiatric Interview
MMRM	Mixed effects model for repeated measure
MOAA/S	Modified Observer's Assessment of Alertness/Sedation
n	Number of subjects
NMDA	N-methyl-D-aspartate
NMDAR	N-methyl-D-aspartate receptor
OCBP	Of childbearing potential
PAC	Premature atrial contractions
PK	Pharmacokinetic(s)
PVC	Premature ventricular contractions
QIDS-SR-14	Quick Inventory of Depressive Symptomatology - 14 Items
QIDS-SR-16	Quick Inventory of Depressive Symptomatology - 16 Items
QTc	Corrected QT interval
QTcF	QT interval corrected according to Fridericia's formula
RBBB	Right bundle branch block

Abbreviation	Definition
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SBP	Systolic blood pressure
SD	Standard deviation
SpO_2	Oxygen saturation
TBL	Total bilirubin
TEAE	Treatment-emergent adverse event
TRD	Treatment-resistant depression
ULN	Upper limit of normal
US	United States
VAS	Visual analog scale

Appendix 2 Regulatory, Ethical, and Study Oversight Considerations

Regulatory and Ethical Considerations

This study will be conducted in accordance with the protocol and with the following:

- Consensus ethical principles derived from international guidelines including the Declaration of Helsinki and Council for International Organizations of Medical Sciences International Ethical Guidelines.
- Applicable International Council for Harmonisation (ICH) Good Clinical Practice (GCP) Guidelines.
- Applicable laws and regulations.

The protocol, protocol amendments, ICF, Investigator's Brochure, and other relevant documents (eg, advertisements) must be submitted to an IEC/IRB by the Investigator and reviewed and approved by the IEC/IRB before the study is initiated.

Any amendments to the protocol will require IEC/IRB and regulatory authority approval, when applicable, before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to subjects.

The Investigator will be responsible for the following:

- Providing written summaries of the status of the study to the IEC/IRB annually or more frequently in accordance with the requirements, policies, and procedures established by the IEC/IRB.
- Notifying the IEC/IRB of SAEs or other significant safety findings as required by IEC/IRB procedures.
- Providing oversight of the conduct of the study at the study center and adherence to requirements of 21 Code of Federal Regulations (CFR), ICH guidelines, the IEC/IRB, European regulation 536/2014 for clinical studies (if applicable), and all other applicable local regulations.

After reading the protocol, each Investigator will sign the protocol signature page and send a copy of the signed page to the Sponsor or representative (Appendix 7). The study will not start at any study center at which the Investigator has not signed the protocol.

Financial Disclosure

Investigators and sub-Investigators will provide the Sponsor with sufficient, accurate financial information as requested to allow the Sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are

responsible for providing information on financial interests during the course of the study and for 1 year after completion of the study.

Insurance

The Sponsor will provide insurance in accordance with local guidelines and requirements as a minimum for the subjects in this study. The terms of the insurance will be kept in the study files.

Informed Consent Process

- The Investigator or his/her representative will explain the nature of the study to the subject or his/her legally authorized representative and answer all questions regarding the study.
- Subjects must be informed that their participation is voluntary. Subjects or their legally authorized representatives will be required to sign a statement of informed consent that meets the requirements of 21 CFR 50, local regulations, ICH guidelines, Health Insurance Portability and Accountability Act requirements, where applicable, and the IEC/IRB or study center.
- The medical record must include a statement that written informed consent was obtained before the subject was entered in the study and the date the written consent was obtained. The authorized person obtaining the informed consent must also sign the ICF.
- Subjects must be re-consented on updated ICF(s) that may impact the subject's willingness to continue to participate during their participation in the study.
- A copy of the ICF(s) must be provided to the subject or the subject's legally authorized representative.
- Subjects who are rescreened are required to sign a new ICF.

Data Protection

- Subjects will be assigned a unique identifier by the Sponsor. Any subject records or datasets that are transferred to the Sponsor will contain the identifier only; subject names or any information that would make the subject identifiable will not be transferred.
- The subject must be informed that his/her personal study-related data will be used by the Sponsor in accordance with local data protection law. The level of disclosure must also be explained to the subject.
- The subject must be informed that his/her medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the Sponsor, by appropriate IEC/IRB members, and by inspectors from regulatory authorities.

Administrative Structure

This trial will be sponsored by Perception Neuroscience, Inc.

CROs will provide clinical operations management, data management, and medical monitoring.

Accredited local laboratories will be used for routine monitoring; local laboratory ranges will be collected.

Table 4 Study Administrative Structure

Function	Responsible Organization
Study Operations Management	CRO
Medical Monitoring	CRO
Trial Master File	CRO
Randomization Code	CRO
Data Management	CRO
Clinical Supply Management	Rave Medidata
Quality Assurance Auditing	CRO
Biostatistics	CRO
Medical Writing	CRO
Laboratory Assessments	Site
Electrocardiogram Collection, Review, and Analysis	Connected Devices

Sponsor Contact Details

Sponsor:	Perception Neuroscience, Inc. c/o atai Life Sciences, WeWork, 524 Broadway, New York, NY 10012
Safety Management and Pharmacovigilance Department — SAE Reporting:	Worldwide Toll: 1-919-313-7111 USA (Toll Free): 1-866-758-2798 Fax: +1-919-313-1412 Email: SafetyInbox.Biotech@IQVIA.com

Dissemination of Clinical Study Data

- Disclosure of clinical study reports, periodic safety reports, and clinical study summary reports after review by regulatory authorities. This includes access to clinical study reports from studies with negative outcomes and from terminated development programs.
- The posting of company-sponsored study information and tabular study results on the US National Institutes of Health's website www.ClinicalTrials.gov and other publicly accessible sites.
- Publication planning and other activities related to non-promotional, peer-reviewed publications, to ensure the scientific integrity and credibility of publication activities performed by or on behalf of the company. The granting of access to analyzable datasets from clinical studies through a secure system, following an independent assessment of the scientific merit of a rigorously defined research question from a third party.

A summary of the results of this trial will be made available on http://www.clinicaltrials.gov and http://www.clinicaltrialsregister.eu/ (as applicable), as required by US and EU Law.

Data Quality Assurance

- All subject data relating to the study will be recorded on printed CRFs unless transmitted to the Sponsor or designee electronically (eg, laboratory data). The Investigator is responsible for verifying that data entries are accurate and correct by physically or electronically signing the CRF.
- The Investigator must maintain accurate documentation (source data) that supports the information entered in the CRF.
- The Investigator must permit study-related monitoring, audits, IEC/IRB review, and regulatory agency inspections and provide direct access to source data documents.
- The Sponsor's designee is responsible for the data management of this study including quality checking of the data.
- Study monitors will perform ongoing source data verification to confirm that data entered into the CRF by authorized study center personnel are accurate, complete, and verifiable from source documents; that the safety and rights of subjects are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.
- Records and documents, including signed ICFs, pertaining to the conduct of this study must be retained by the Investigator for 15 years after study completion unless local regulations or institutional policies require a longer retention period. No records may be destroyed during the retention period without the written approval of the Sponsor. No

records may be transferred to another location or party without written notification to the Sponsor.

Source Documents

The Investigator/institution should maintain adequate and accurate source documents and study records that include all pertinent observations on each of the study center's subjects. Source data should be attributable, legible, contemporaneous, original, accurate, and complete. Changes to source data should be traceable, should not obscure the original entry, and should be explained if necessary (eg, via an audit trail).

- Source documents provide evidence for the existence of the subject and substantiate the integrity of the data collected. Source documents are filed at the Investigator's study center.
- Data entered in the CRF that are transcribed from source documents must be consistent with the source documents or the discrepancies must be explained. The Investigator may need to request previous medical records or transfer records, depending on the study. Also, current medical records must be available.
- Definition of what constitutes source data can be found in the CRF completion guidelines.

Study and Study Center Closure

The Sponsor designee reserves the right to close the study center or terminate the study at any time for any reason at the sole discretion of the Sponsor. Study centers will be closed upon study completion. A study center is considered closed when all required documents and study supplies have been collected and a study center closure visit has been performed.

The Investigator may initiate study center closure at any time, provided there is reasonable cause and sufficient notice is given in advance of the intended termination.

Reasons for the early closure of a study center by the Sponsor or Investigator may include but are not limited to:

- Failure of the Investigator to comply with the protocol, the requirements of the IEC/IRB or local health authorities, the Sponsor's procedures, or GCP guidelines.
- Inadequate recruitment of subjects by the Investigator.
- Discontinuation of further study treatment development.

Publication Policy

The data generated by this study are confidential information of the Sponsor. The Sponsor will make the results of the study publicly available.

- The results of this study may be published or presented at scientific meetings. If this is foreseen, the Investigator agrees to submit all manuscripts or abstracts to the Sponsor before submission. This allows the Sponsor to protect proprietary information and to provide comments.
- The Sponsor will comply with the requirements for the publication of study results. In accordance with standard editorial and ethical practice, the Sponsor will generally support publication of multicenter studies only in their entirety and not as individual study center data. In this case, a Coordinating Investigator will be designated by mutual agreement.
- Authorship will be determined by mutual agreement and in line with the International Committee of Medical Journal Editors authorship requirements.

Appendix 3 Clinical Laboratory Tests

Procedure Notes:

- Pregnancy test: Serum testing will be performed at screening and to confirm a positive urine test. Local urine pregnancy testing will be performed at both follow-up visits.
- Hepatitis B and hepatitis C screening: hepatitis B surface antigen and hepatitis C virus (HCV antibody) testing will be required.
- The tests detailed in Table 5 will be performed by a local laboratory. Urine drug screen may be performed on site.
- Protocol-specific requirements for inclusion or exclusion of subjects are detailed in Section 5.0 of the protocol.
- Additional tests may be performed at any time during the study as determined necessary by the Investigator or required by local regulations.

Table 5 Protocol-required Safety Laboratory Assessments

Laboratory Assessments	**Parameters**			
Hematology	Platelet count	RBC indices: Mean corpuscular volume (MCV) Mean corpuscular hemoglobin (MCH) % Reticulocytes		White blood cell count with differential: Neutrophils Lymphocytes Monocytes Eosinophils Basophils
	Red blood cell (RBC) count			
	Hemoglobin			
	Hematocrit			
Clinical chemistry	Blood urea nitrogen or urea Lactate dehydrogenase (LDH)	Potassium	Aspartate aminotransferase (AST)/serum glutamic-oxaloacetic transaminase (SGOT)	Total and direct bilirubin
	Creatinine	Sodium Chloride	Alanine aminotransferase (ALT)/serum glutamic-pyruvic transaminase (SGPT)	Total protein
	Glucose [non-fasting[a]]	Calcium	Alkaline phosphatase Gamma-glutamyl transferase (GGT) Albumin	

Table 5 Protocol-required Safety Laboratory Assessments

Laboratory Assessments	Parameters
Routine urinalysis	• Specific gravity • pH, glucose, protein, blood, ketones, bilirubin, urobilinogen, nitrite, leukocyte esterase by dipstick • Microscopic examination (if blood or protein is abnormal)
Other screening tests	• Follicle stimulating hormone and estradiol (as needed in women of non-childbearing potential only) • Urine alcohol and drug screen (to include at minimum: amphetamines/methamphetamines, ketamine, barbiturates, cocaine, and opiates) • Serum or urine human chorionic gonadotropin (hCG) pregnancy test (as needed for women of childbearing potential)[b] • Serology (HIV antibody, hepatitis B surface antigen, and HCV antibody) All study-required laboratory assessments will be performed by a local laboratory. Urine drug screen may be done on site. The results of each test must be entered into the CRF.

[a] Non-fasting blood samples will be requested, but both fasting and non-fasting samples are acceptable.
[b] Local urine testing will be standard for the protocol unless serum testing is required by local regulation or IEC/IRB.

Investigators must document their review of each laboratory safety report.

Laboratory/analyte results that could unblind the study will not be reported to study centers or other blinded personnel until the study has been unblinded.

Appendix 4 Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting

Definition of AE

AE Definition
• An AE is any untoward medical occurrence in a patient or subject, temporally associated with the use of study treatment, whether or not considered related to the study treatment. • NOTE: An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding), symptom, or disease (new or exacerbated) temporally associated with the use of study treatment.

Events <u>Meeting</u> the AE Definition
• Any abnormal laboratory test results (hematology, clinical chemistry, or urinalysis) or other safety assessments (eg, ECG, radiological scans, vital signs measurements), including those that worsen from baseline, considered clinically significant in the medical and scientific judgment of the Investigator (ie, not related to progression of underlying disease). • Exacerbation of a chronic or intermittent pre-existing condition including either an increase in frequency and/or intensity of the condition. • New conditions detected or diagnosed after study treatment administration even though it may have been present before the start of the study. • Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction. • Signs, symptoms, or the clinical sequelae of a suspected overdose of either study treatment or a concomitant medication. Overdose per se will not be reported as an AE/SAE unless it is an intentional overdose taken with possible suicidal/self-harming intent. Such overdoses should be reported regardless of sequelae. • "Lack of efficacy" or "failure of expected pharmacological action" per se will not be reported as an AE or SAE. Such instances will be captured in the efficacy assessments. However, the signs, symptoms, and/or clinical sequelae resulting from lack of efficacy will be reported as AEs or SAEs if they fulfill the definition of an AE or SAE.

Events NOT Meeting the AE Definition
• Any clinically significant abnormal laboratory findings or other abnormal safety assessments which are associated with the underlying disease, unless judged by the Investigator to be more severe than expected for the subject's condition. • The disease/disorder being studied or expected progression, signs, or symptoms of the disease/disorder being studied, unless more severe than expected for the subject's condition. • Medical or surgical procedure (eg, endoscopy, appendectomy): the condition that leads to the procedure is the AE. • Situations in which an untoward medical occurrence did not occur (social and/or convenience admission to a hospital, including planned hospitalization for this study). • Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

Definition of SAE

If an event is not an AE per the definition above, then it cannot be an SAE even if serious conditions are met (eg, hospitalization for signs/symptoms of the disease under study).

An SAE is defined as any untoward medical occurrence that, at any dose:
a) Results in death
b) Is life-threatening The term "life-threatening" in the definition of "serious" refers to an event in which the subject was at risk of death at the time of the event. It does not refer to an event, which hypothetically might have caused death, if it were more severe.
c) Requires inpatient hospitalization or prolongation of existing hospitalization In general, hospitalization signifies that the subject has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether "hospitalization" occurred or was necessary, the AE should be considered serious. Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE, nor is planned hospitalization for this study.
d) Results in persistent disability/incapacity • The term disability means a substantial disruption of a person's ability to conduct normal life functions. • This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, and

accidental trauma (eg, sprained ankle) which may interfere with or prevent everyday life functions but do not constitute a substantial disruption.
e) Is a congenital anomaly/birth defect
f) Other situations: • Medical or scientific judgment should be exercised in deciding whether SAE reporting is appropriate in other situations such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the subject or may require medical or surgical intervention to prevent 1 of the other outcomes listed in the above definition. These events should usually be considered serious. Examples of such events include invasive or malignant cancers, intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias, or convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

Recording and Follow-up of AE and/or SAE

AE and SAE Recording

- When an AE/SAE occurs, it is the responsibility of the Investigator to review all documentation (eg, hospital progress notes, laboratory reports, and diagnostics reports) related to the event.
- The Investigator will then record all relevant AE/SAE information in the CRF. Each event must be recorded separately.
- It is **not** acceptable for the Investigator to send photocopies of the subject's medical records to Safety Management in lieu of completion of the AE/SAE CRF page.
- There may be instances when copies of medical records for certain cases are requested by Safety Management. In this case, all subject identifiers, with the exception of the subject number, will be redacted on the copies of the medical records before submission to Safety Management.
- The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.

Assessment of Intensity

The Investigator will make an assessment of intensity for each AE and SAE reported during the study and assign it to 1 of the following categories:

- Mild: An event that is easily tolerated by the subject, causing minimal discomfort and not interfering with everyday activities.
- Moderate: An event that causes sufficient discomfort and interferes with normal everyday activities.
- Severe: An event that prevents normal everyday activities. An AE that is assessed as severe should not be confused with a SAE. Severe is a category utilized for rating the intensity of an event; and both AEs and SAEs can be assessed as severe.

  An event is defined as 'serious' when it meets at least 1 of the predefined outcomes as described in the definition of an SAE, NOT when it is rated as severe.

Assessment of Causality

- The Investigator is obligated to assess the relationship between study treatment and each occurrence of each AE/SAE. The AE must be characterized as unrelated, unlikely to be related, possibly related, probably related, or definitely related.
  - "Definitely related" suggests that the AE has a timely relationship to administration of study treatment, and there is no apparent potential alternate etiology.
  - "Probably related" conveys that there are facts, evidence, and/or arguments to suggest a causal relationship, rather than a relationship cannot be ruled out.
  - "Possibly related" suggests that the association of the AE with the study treatment is unknown; however, the AE is not reasonably supported by other conditions.
  - "Unlikely to be related" suggests that only a remote connection exists between the study treatment and the AE. Other conditions, including chronic illness, progression or

expression of the disease state or reaction to concomitant therapy, appear to explain the reported AE.
    - "Unrelated" is used if there is not a reasonable possibility that the study treatment caused the AE.
- The Investigator will use clinical judgment to determine the relationship.
- Alternative causes, such as underlying disease(s), concomitant therapy, and other risk factors, as well as the temporal relationship of the event to study treatment administration will be considered and investigated.
- The Investigator will also consult the IB and/or Product Information, for marketed products, in his/her assessment.
- For each AE/SAE, the Investigator must document in the medical notes that he/she has reviewed the AE/SAE and has provided an assessment of causality.
- There may be situations in which an SAE has occurred, and the Investigator has minimal information to include in the initial report to Safety Management. However, it is very important that the Investigator always makes an assessment of causality for every event before the initial transmission of the SAE data to Safety Management.
- The Investigator may change his/her opinion of causality in light of follow-up information and send an SAE follow-up report with the updated causality assessment.
- The causality assessment is one of the criteria used when determining regulatory reporting requirements.

Follow-up of AEs and SAEs

- The Investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated or as requested by Safety Management to elucidate the nature and/or causality of the AE or SAE as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.
- If a subject dies during participation in the study or during a recognized follow-up period, the Investigator will provide Safety Management with a copy of any postmortem findings including histopathology.
- New or updated information will be recorded in the originally completed CRF.
- The Investigator will submit any updated SAE data to the Sponsor or designee within 24 hours of receipt of the information.

Reporting of SAEs

SAE Reporting to Safety Management via Paper CRF

- E-mail transmission of the SAE paper CRF is the preferred method to transmit this information to Safety Management with facsimile transmission as the back-up method.

- In rare circumstances and in the absence of e-mail/facsimile equipment, notification by telephone is acceptable with a copy of the SAE data collection tool sent by overnight mail or courier service.
- Initial notification via telephone does not replace the need for the Investigator to complete and sign the SAE CRF pages within the designated reporting time frames.
- Contacts for SAE reporting can be found in the Investigator Study Binder and in the contacts listed in Appendix 2. All SAEs must also be reported to Safety Management within 24 hours of first awareness of the event. A paper SAE Report Form should be completed and submitted via e-mail to SafetyInbox.Biotech@IQVIA.com or faxed to +1-919-313-1412.

Appendix 5 Contraceptive Guidance and Collection of Pregnancy Information

Definitions:

Subject of Childbearing Potential (OCBP)

A woman is considered fertile following menarche and until becoming postmenopausal unless permanently sterile (see below).

Female Subjects in the following categories are not considered OCBP

1. Premenarchal
2. Premenopausal female with 1 of the following > 6 months prior to screening:
   a) Documented hysterectomy.
   b) Documented bilateral salpingectomy.
   c) Documented bilateral oophorectomy.

   Note: Documentation can come from the study center personnel's: review of the subject's medical records, medical examination, or medical history interview.
3. Postmenopausal female:
   a) A postmenopausal state is defined as no menses for 24 months without an alternative medical cause.

Contraception Guidance

Male subjects

- Male subjects with female partners of childbearing potential are eligible to participate if they agree to use a male condom plus partner use of a contraceptive method with a failure rate of < 1% per year when having penile-vaginal intercourse with a partner who is of childbearing potential who is not currently pregnant (during the protocol-defined time frame in Section 5.1):
- In addition, male subjects must refrain from donating sperm during the treatment period and for at least 3 months after the last dose of study treatment.

Female subjects

Female subjects of childbearing potential are eligible to participate if they agree to use a highly effective method of contraception consistently and correctly as described in the table below. Hormonal contraception may be susceptible to interaction with the study treatment, which may reduce the efficacy of the contraceptive method. For subjects using hormonal contraception as the primary form of birth control, a secondary (barrier) method should be utilized during the treatment period and for at least 30 days after the last dose of study treatment. Doses of oral contraception should not contain more than 30 micrograms of ethinyl estradiol per day.

Highly Effective Contraceptive Methods

Highly Effective Contraceptive Methods That Are User Dependent[a] Failure rate of < 1% per year when used consistently and correctly.
Combined (estrogen and progestogen containing) hormonal contraception associated with inhibition of ovulation in conjunction with a secondary method[b] • Oral. • Intravaginal. • Transdermal.
Progestogen only hormonal contraception associated with inhibition of ovulation in conjunction with a secondary method[b] • Oral. • Injectable.
Highly Effective Methods That Are User Independent[a]
• Implantable progestogen only hormonal contraception associated with inhibition of ovulation in conjunction with a secondary method[b] • Intrauterine device (IUD). • Intrauterine hormone-releasing system (IUS) in conjunction with a secondary method[b] • Bilateral tubal occlusion.
Vasectomized Partner • Vasectomized male partner (sterilization performed at least 90 days prior to the Screening visit, with verbal confirmation of surgical success, and the sole partner for the female subject)

[a] Typical use failure rates may differ from those when used consistently and correctly. Use should be consistent with local regulations regarding the use of contraceptive methods for subjects participating in clinical studies.

[b] Hormonal contraception may be susceptible to interaction with the study treatment, which may reduce the efficacy of the contraceptive method. In this case, 2 methods (1 primary and 1 secondary method) of birth control should be utilized during the treatment period and for at least 30 days after the last dose of study treatment.

Secondary (barrier) methods of contraception

Combined (estrogen and progestogen containing) hormonal contraception associated with inhibition of ovulation[b] • male condom with spermicide • female condom with spermicide • over-the-counter sponge with spermicide • cervical cap with spermicide (as prescribed) • diaphragm with spermicide (as prescribed).

Female subjects of childbearing potential should refrain from donation of ova from Check-in (Day -1) until 90 days after the Follow–up visit.

For subjects who are exclusively in same-sex relationships, contraceptive requirements do not apply. If a subject who is in a same-sex relationship at the time of signing the ICF becomes engaged in a heterosexual relationship, they must agree to use contraception as described previously.

Pregnancy Testing:

- Subjects OCBP should only be included after a negative serum pregnancy test.
- Additional pregnancy testing should be performed at the times specified in the Schedule of Activities.

Pregnancy testing will be performed whenever a menstrual cycle is missed or when pregnancy is otherwise suspected.

- Pregnancy testing will be performed and assayed in the local laboratory.

Collection of Pregnancy Information

Male subjects with partners who become pregnant

- The Investigator will attempt to collect pregnancy information on any male subject's female partner who becomes pregnant while the male subject is in this study. This applies only to male subjects who receive PCN-101.
- While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication or elective termination of a pregnancy will be reported as an AE or SAE. A spontaneous abortion is always considered to be an SAE and will be reported as such.
- Any post-study pregnancy-related- SAE considered reasonably related to the study treatment by the Investigator will be reported to Safety Management as described in Section 8.3.3. While the Investigator is not obligated to actively seek this information, he or she may learn of an SAE through spontaneous reporting.

Female subjects who become pregnant

- The Investigator will collect pregnancy information on any female subject who becomes pregnant while participating in this study. Information will be recorded on the appropriate form and submitted to Safety Management within 24 hours of learning of a subject's pregnancy. The subject will be followed to determine the outcome of the pregnancy. The Investigator will collect follow-up information on the subject and the neonate, and the information will be forwarded to the Sponsor. Generally, follow-up will not be required for longer than 6 to 8 weeks beyond the estimated delivery date. Any termination of pregnancy will be reported, regardless of fetal status (presence or absence of anomalies) or indication for the procedure.
- While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication or elective termination of a pregnancy will be reported as an AE or SAE. A spontaneous abortion is always considered to be an SAE and will be reported as such.
- Any post-study pregnancy-related SAE considered reasonably related to the study treatment by the Investigator will be reported to the Sponsor as described in Section 8.3.3. While the Investigator is not obligated to actively seek this information in former subjects, he or she may learn of an SAE through spontaneous reporting.
- Any female subject who becomes pregnant while participating in the study will be withdrawn from the study.

Appendix 6 Permitted and Prohibited Concomitant Medications

Table 6 provides a list of permitted and prohibited concomitant medications throughout the study.

Table 6 List of Permitted and Prohibited Concomitant Medications

Drug	Episodic Use	Continuous Use	Comments
Amantadine	N	N	
Analgesics (except opiates)	Y	Y	
Anti-anginal drugs	N	N	Angina is exclusion
Anti-arrhythymics	N	N	Arrhythmia is exclusionary
Anticholenergics	N	N	
Anticholinesterase inhibitors	N	N	
Anticoagulants	N	N	
Anticonvulsants	N	N	
Antidepressants	N	Y	MAOIs are always excluded
Antidiarrheal	Y	N	
Anti-emetics	Y	N	
Anti-inflammatory	Y	Y	
Antipsychotics	N	Y	Stable doses of quetiapine, aripiprazole, olanzapine, and brexpiprazole are permitted for continuous use
Antifungals	N	N	No ketoconazole, fluconazole due to CYP3A interaction
Artemisinin			
Aspirin	Y	Y	
Benzodiazepines	Y	Y	Except clobazam Daily doses of benzodiazepine receptor agonists will be limited to 2 mg/day or less of lorazepam or equivalents. Benzodiazepines will be prohibited within 6 hours prior to dosing

Table 6 List of Permitted and Prohibited Concomitant Medications

Drug	Episodic Use	Continuous Use	Comments
Clarithromycin	N	N	CYP3A interaction
Erythromycin	N	N	CYP3A interaction
Elagolix	N	N	CYP3A interaction
Fish oils	Y	Y	
HIV drugs	N	N	HIV excluded
Hormones	N	Y	Glucocorticoids excluded due to CYP3A interaction
Itraconazole	N	N	CYP3A interaction
Ketoconazole	N	N	CYP3A interaction
Lithium	N	N	
Methyldopa	N	N	
Modafinil	N	N	
Opiates	N	N	
Oritavancin	N	N	CYP3A interaction
Pioglitazone	N	N	CYP3A interaction
Rifabutin	N	N	CYP3A interaction
Rifampin	N	N	CYP interaction
St. john's wort	N	N	CYP3A interaction
Stimulants	N	N	
Steroids (oral)	N	N	
Steroids (inhaled, topical, ophthalmic)	Y	Y	
Telithromycin	N	N	CYP3A interaction
Telotristat	N	N	CYP3A interaction
Ticlopidine	N	N	CYP2B6 interaction

Table 6 List of Permitted and Prohibited Concomitant Medications

Drug	Episodic Use	Continuous Use	Comments
Troglitazone	N	N	CYP3A interaction
Troleandomycin	N	N	CYP3A interaction
Tryptophan	N	N	
Voriconazole	N	N	CYP3A interaction

Note: Permitted episodic or continuous use of a concomitant medication is indicated with a "Y" (yes).
Prohibited episodic or continuous use of a concomitant medication is indicated with an "N" (no).

Appendix 7 Signature of Investigator

PROTOCOL TITLE: A RANDOMIZED, PLACEBO-CONTROLLED, DOUBLE-BLIND STUDY OF INTRAVENOUS PCN-101 TO ASSESS THE SAFETY AND EFFICACY IN TREATMENT-RESISTANT DEPRESSION

PROTOCOL NO: PCN-101-21

VERSION: Protocol Amendment 4

This protocol is a confidential communication of Perception Neuroscience, Inc. I confirm that I have read this protocol, I understand it, and I will work according to this protocol. I will also work consistently with the ethical principles that have their origin in the Declaration of Helsinki and that are consistent with Good Clinical Practice and the applicable laws and regulations. Acceptance of this document constitutes my agreement that no unpublished information contained herein will be published or disclosed without prior written approval from the Sponsor.

Instructions to the Investigator: Please SIGN and DATE this signature page. PRINT your name, title, and the name of the study center in which the study will be conducted. Return the signed copy to the CRO.

I have read this protocol in its entirety and agree to conduct the study accordingly:

Signature of Investigator: _____ Date: _____

Printed Name: _____

Investigator Title: _____

Name/Address of Center: _____

_____

_____