CLINICAL STUDY PROTOCOL: SP-624-201 **Study Title:**	A Multicenter, Double-Blind, Randomized, Placebo-Controlled Study of the Safety and Efficacy of SP-624 in the Treatment of Adults with Major Depressive Disorder
Sponsor:	Sirtsei Pharmaceuticals, Inc.
Phase of Development:	Phase 2
NCT Number	NCT04479852
Protocol Number:	SP-624-201
Protocol Version:	Amendment 3
Protocol Version Number:	4.0
Protocol Version Date:	26 May 2021

CLINICAL STUDY PROTOCOL: SP-624-201

Study Title:	A Multicenter, Double-Blind, Randomized, Placebo-Controlled Study of the Safety and Efficacy of SP-624 in the Treatment of Adults with Major Depressive Disorder
Study Drug:	SP-624
IND Number:	[redacted]
Sponsor:	Sirtsei Pharmaceuticals, Inc. [redacted]
Phase of Development:	Phase 2
Protocol Number:	SP-624-201
Protocol Version:	Amendment 3
Protocol Version Number:	4.0
Protocol Version Date:	26 May 2021

Version History

Version Number	Version Date	Comment
1.0	09 July 2020	Original
2.0	17 September 2020	Amendment 1
3.0	27 January 2021	Amendment 2
4.0	26 May 2021	Amendment 3

INVESTIGATOR'S PROTOCOL AGREEMENT PAGE

Note: A signed copy of this page will be collected and filed in both the Sponsor and Investigator trial master files prior to the commencement of this study.

- I have read the SP-624-201 protocol and agree to conduct the study as outlined.
- I have received and read the Investigator's Brochure for SP-624.
- I agree to maintain the confidentiality of all information received or developed in connection with this protocol.
- I will personally oversee the conduct of this study as outlined herein and will make all reasonable efforts to complete the study within the time designated.
- I will provide all key study personnel under my supervision with copies of the protocol and access to all information provided by Sirtsei Pharmaceuticals, Inc.. I will discuss this material with them to ensure that they are fully informed about SP-624, the safety parameters, and the conduct of the study in general.
- I am aware that, before commencement of this study, the institutional review board responsible for such matters must approve this protocol in the clinical facility where it will be conducted.
- I agree to make all reasonable efforts to adhere to the attached protocol.
- I, or my designee, agree to be present at the site initiation visit. In addition, I will ensure the presence of all key study personnel under my supervision at this meeting.
- I agree to provide all subjects with informed consent forms, as required by government and International Council for Harmonisation regulations.
- I further agree to report to Sirtsei Pharmaceuticals, Inc. any adverse experiences in accordance with the terms of this protocol and Food and Drug Administration (FDA) regulation 21 Code of Federal Regulations (CFR) 312.64.

_____

Printed Name of Investigator

_____

Signature of Investigator

_____

Date

SPONSOR EMERGENCY CONTACT INFORMATION

Reporting of Serious Adverse Events (SAEs)

Any SAE occurring in a study subject must be reported to the Sponsor within 24 hours.

See Section 8.1.2 (Serious Adverse Events) for definition and reporting procedure for SAEs.

All relevant information must be captured, preferably on an SAE Report Form, and submitted via email within 24 hours to each of the individuals listed in Table 1.

Request email confirmation of receipt, and if not confirmed, follow-up by phone to confirm receipt.

Table 1: Emergency Contact Information

Role in Study	Name	Email Address	Telephone Number
Sponsor Clinical Study Leader	[redacted]	[redacted]	[redacted]
Sponsor Medical Monitor	[redacted]	[redacted]	[redacted]

PROTOCOL SYNOPSIS

Name of Sponsor/Company:

Sirtsei Pharmaceuticals, Inc.

Name of Investigational Product:

SP-624 (oral capsules)

Name of Active Ingredient:

Code Name: SP-624

[redacted]

Title of Study:

A Multicenter, Double-Blind, Randomized, Placebo-Controlled Study of the Safety and Efficacy of SP-624 in the Treatment of Adults with Major Depressive Disorder

Phase of development:

Phase 2

Study center(s):

Approxmiately 40 sites in the United States

Planned Study Duration:

The planned study duration encompasses the estimated time from the Screening of the first subject to the estimated time the last subject completes the Week 6 Follow-Up Visit.

- Estimated date first subject Screened: September 2020
- Estimated date of last subject first visit: October 2021
- Estimated date of last subject last visit: November 2021

Objectives:

Primary:

- Evaluate the efficacy of SP-624 administered once-daily for up to 4 weeks compared to placebo in the treatment of adults with major depressive disorder (MDD)

Secondary:

- Evaluate the safety and tolerability of SP-624 administered once-daily for up to 4 weeks compared to placebo in the treatment of adults with MDD
- Characterize the population pharmacokinetics (PK) of SP-624 in plasma when administered orally to subjects with MDD

Study Design:

This is a Phase 2, multicenter, double-blind, randomized, placebo-controlled study of the safety and efficacy of SP-624 in the treatment of adult subjects with MDD as defined by the Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition (DSM-5). Following the successful completion of a Screening Phase, subjects will be randomized to one of two treatment groups in a 1:1 ratio and will receive SP-624 or placebo over a treatment period of four weeks (Treatment Group #1: SP-624 20 mg/day; Treatment Group #2: Placebo).

Approximately 300 subjects (150 per treatment group) will be enrolled into the study. Each subject will complete Screening, Baseline, Treatment, and Follow-up Visits.

Prior to initiating the 4-week treatment period, subjects will complete a Screening/Baseline period of up to 28 days, during which time all Screening assessments will be performed, and any current depression medications discontinued. All Screening assessments should be completed before discontinuing any current depression medications. Subjects will return for a Baseline Visit to complete efficacy and safety assessments. During the treatment period, subjects will return to the investigative site to complete efficacy and safety assessments at the end of Weeks 1, 2, 3, and 4. After the final dose of study drug, subjects will complete a follow-up period of two weeks and will return at the end of Weeks 5 and 6. Subjects may be treated with anti-depressant medications according to physician recommendations after completion of the Week 5 Visit.

Unless there are any outstanding safety issues that require follow-up, subjects will be discharged from the study at the Week 6 Visit.

Number of subjects (planned):

Approximately 300 adult subjects with MDD as defined by DSM-5; 150 subjects per treatment group at approximately 40 centers.

Duration of Subject Participation:

Subjects will receive double-blind treatment for up to 4 weeks, following a Screening/Baseline period of up to 28 days. After the treatment period, subjects will complete a 2-week follow-up period. The planned study duration for each subject will be approximately 70 days.

Investigational Product, Dosage, and Mode of Administration:

Each dose of SP-624 will be supplied as two capsules, each containing 10 mg of active pharmaceutical ingredient (API). Matching placebo capsules identical in shape and color to the active capsules will also be used.

- Treatment Group #1: Two 10 mg capsules SP-624 each morning for 4 weeks
- Treatment Group #2: Two capsules placebo each morning for 4 weeks

If the two capsule dose is not well tolerated, the dose can be decreased to one capsule per dose after Week 1.

Study drug will be packaged in weekly blister cards (Weeks 1-4 of dosing), with each blister card containing 20 capsules of study drug (two capsules per dose for seven days of dosing, plus three extra days of dosing as a provision for study visit scheduling). Weekly blister cards will be assembled in subject kits containing four blister cards for use during the treatment period. Each kit will be labeled with a 3-part label that specifies the randomization number, cautionary statement, and storage conditions. Each blister card will have a one-part label that specifies the randomization number, week, number of capsules, cautionary statement, and storage conditions.

Each study drug dose should be taken orally, once daily at approximately the same time each morning. Study drug doses should be taken on an empty stomach before breakfast, and no food should be consumed for at least 1 hour after dosing. High fat foods at breakfast (e.g., bacon, cheese, eggs, etc.) should be avoided. Morning liquids such as coffee or tea are allowed without any additives (eg, cream, sugar, etc).

If dosing on an empty stomach is not practical or not well tolerated, then a low fat, low calorie breakfast 2 hours before dosing, or 30-60 minutes after dosing, is acceptable. If dosing remains not well tolerated, then subjects should dose 30 minutes after a low fat, low calorie meal.

Inclusion Criteria:

To be eligible for this study, a subject must meet all of the following inclusion criteria:

1. Willing and able to provide written informed consent to participate in the study.
2. Males and females, aged 18 to 65 years, inclusive.
3. In generally good physical health, in the opinion of the Investigator.
4. Body mass index (BMI) must be ≥ 18 and ≤ 40 kg/m^2
5. Females of reproductive potential and males with partners of reproductive potential must agree to remain abstinent or use adequate and reliable contraception throughout the study and for at least 30 days after the last dose of study drug.
    a. Adequate contraception is defined as continuous use of double-barrier method (e.g., condom and spermicide or diaphragm and spermicide), oral contraceptive, implant, dermal contraception, long-term injectable contraceptive, or intrauterine device.
    b. Non-reproductive potential is defined as surgically sterile, undergone tubal ligation or vasectomy, or postmenopausal (defined as at least 12 months of spontaneous amenorrhea).
    c. Subjects of reproductive potential but whose sole partner is of non-reproductive potential, or whose sole partner is of the same sex, are not required to maintain abstinence or contraception.

d. Subjects must agree to refrain from sperm and oocyte donation for at least 90 days after the last dose of study drug.

6. Meets DSM-5 criteria for moderate to severe MDD, as confirmed by the Mini International Neuropsychiatric Interview (MINI). Subjects with a diagnosis of co-morbid generalized anxiety disorder (GAD) may be included if MDD is the primary diagnosis.
7. Montgomery-Asberg Depression Rating Scale (MADRS) total score ≥ 27 at both Screening and Baseline.
8. History of inadequate or non-response to at least one adequate treatment, but no more than three of the same class of antidepressant, (i.e., an adequate dose for at least six weeks) for the current episode, in the opinion of the Investigator. NOTE: If immediately prior to this episode starting, treatment was working to the point of remission, but then the depression worsened, subject should not be enrolled. For example, subject should not have been on an antidepressant and in remission prior to the start of the current episode.
9. Willing and able to comply with the study design schedule and other requirements.

Exclusion Criteria:

A subject who meets any of the following exclusion criteria will not be enrolled in the study:

1. Female who is pregnant, breastfeeding, or less than six months postpartum at Screening.
2. History or presence of major bowel resection, total gastrectomy, or diabetes, and/or history or presence of any clinically significant medical condition, disease, or surgical history that could, in the opinion of the Investigator, jeopardize the safety of the subject or validity of the study data, or interfere with the absorption, distribution, metabolism, or excretion of the study drug.
3. A current DSM-5 diagnosis of depression with peripartum onset, panic disorder, obsessive compulsive disorder, post-traumatic stress disorder, anorexia nervosa, or bulimia nervosa.
4. A history of or current DSM-5 diagnosis of MDD with psychotic features, any schizophrenia spectrum and other psychotic disorders, bipolar disorder, or personality disorder.
5. History of treatment with electroconvulsive therapy (ECT), vagus nerve stimulation, deep brain stimulation, or transcranial magnetic stimulation within six months of Screening.
6. Current episode of depression exceeds 2 years or is less than six weeks from Screening.
7. History of inadequate or non-response, in the opinion of the Investigator, to an adequate treatment regimen of two or more classes of antidepressants during the current episode.
8. Subjects treated with an antipsychotic during the current episode should be excluded.
9. Subjects who meet DSM-5 criteria for moderate or severe substance use disorder for any substance except nicotine or caffeine, within 12 months of Screening. Subjects with a urine drug screen positive for suspected substance of abuse will be excluded. Urine drug screen positive for cannabinoids is not exclusionary if the subject does not meet criteria for moderate or severe substance use disorder.
10. Will require initiation of psychotherapy during the study period, in the Investigator's opinion. If a subject has been in ongoing psychotherapy for a minimum of three months, the therapy may continue if it remains unchanged. Cognitive behavioral therapy is excluded.

11. Poses a current or future suicidal risk, in the Investigator's opinion, or has a history of suicidal behavior within the past 12 months, and/or subject answers "yes" to "Suicidal Ideation" Item 4 (active suicidal ideation with some intent to act, without specific plan) or Item 5 (active suicidal ideation with specific plan and intent) on the Columbia-Suicide Severity Rating Scale (C-SSRS) assessment during the six months prior to the Screening period and/or at Baseline.
12. Failure to discontinue all psychoactive medications or psychoactive supplements including antidepressants and mood stabilizers, within a time period prior to Baseline corresponding to at least five half-lives of the medication in question.
13. Presence of a clinically significant abnormality on physical examination or electrocardiogram (ECG), including a corrected QT interval using Fridericia's formula (QTcF) >450 msec for males and >470 msec for females. Note: ECG may be repeated once at both Screening and Baseline for confirmatory purposes.
14. Presence or history of any known clinically significant cardiovascular disorders including, but not limited to: coronary artery disease, heart failure, valvular heart disease, cardiomyopathies, myocardial infarction, chamber enlargement or hypertrophy, or orthostatic hypotension.
15. Presence of uncontrolled hypertension, defined as consistent systolic blood pressure (SBP) >160 mmHg or consistent diastolic blood pressure (DBP) >95 mmHg despite present therapy. Note: Blood pressure measurements may be repeated twice at both Screening and Baseline for confirmatory purposes.
16. Screening laboratory value(s) outside the laboratory reference range that are considered to be clinically significant by the Investigator (clinical chemistry, hematology, coagulation, and urinalysis). Note: Screening labs may be repeated once for confirmatory purposes.
17. Screening liver function tests (ALT, AST, Alkaline phosphatase) > 2x the upper limit of normal.
18. Known history of current or chronic Hepatitis B or Hepatitis C infection, or a known history of human immunodeficiency virus (HIV) infection. Subjects with a history of treated Hepatitis C infection where liver function tests are within normal limits may participate.
19. Subjects with a history of seizures or seizure disorder.
    Note: Subjects with a history of only febrile seizures, limited to up to two episodes, are permitted.
20. Clinically significant acute infection, other than mild viral or bacterial infections from which the subject has fully recovered (e.g., common cold, localized skin infection, etc.), within the two weeks prior to Baseline.
21. Major trauma or surgery in the 12 weeks prior to Screening or at any time between Screening and randomization.
22. Presence or history of malignancy within the last five years. Note: History of adequately treated basal cell or squamous cell carcinoma of the skin or in situ cervical cancer is allowed.
23. History of an anaphylactic allergic reaction to previous medication.
24. Blood or plasma donation within 30 days of randomization, or planned donations or elective surgery during the study or during the 60 days following the study.
25. Subjects who, in the opinion of the Investigator, are not suitable candidates for the study.

26. Individuals who are currently enrolled in or have participated in a clinical trial (i.e., received study drug) involving an investigational product within 60 days or within five half-lives of the investigational product (whichever is longer) prior to Baseline.

Study Endpoints:

Primary Efficacy Endpoint:

- Change from Baseline to Week 4 in MADRS total score

Secondary Efficacy Endpoints:

- Change from Baseline to Weeks 1, 2, and 3 in MADRS total score
- Change from Baseline to Weeks 1, 2, 3, and 4 in Clinical Global Impression – Severity (CGI-S) total score
- Change from Baseline to Week 5 and change from Week 4 to Week 5 in MADRS total score and CGI-S total score
- Change from Baseline to Week 2 and Week 4 in the 17-item Hamilton Depression Rating Scale (HAM-D-17) total score
- Change from Baseline to Week 2 and Week 4 in the Sheehan Disability Scale (SDS)
- Change from Baseline to Week 2 and Week 4 in the Quick Inventory of Depressive Symptomology – Self Report (QIDS-SR)
- Change from Baseline to Week 2 and Week 4 in the Quality of Life Enjoyment and Satisfaction Questionnaire – Short Form (Q-LES-Q-SF)

Safety Endpoints:

- Adverse Events (AEs)
- Clinical safety laboratory tests (serum chemistry [including thyroid function], hematology, coagulation, and urinalysis tests)
- Vital signs (blood pressure, pulse rate, and oral body temperature)
- Body weight
- 12-lead ECGs
- Physical examinations
- C-SSRS

PK Endpoints:

A total of four blood samples will be collected from each subject for the measurement of plasma concentrations of SP-624 at the following visits:

- Visit 2 (Day -1)
- Visit 3 (Week 1): Pre-Dose
- Visit 4 (Week 2): Pre-Dose or approximately 2 to 10 hours post-dose (whichever is more convenient for the subject)

- Visit 6 (Week 4): approximately 2 to 10 hours post-dose

If a subject missed the dose the day before a scheduled "pre-dose" sample collection, study personnel should instead collect a post-dose sample at that visit. Should a subject inadvertently dose prior to a scheduled "pre-dose" sample collection at Week 1 or Week 2, study personnel should collect a post-dose sample.

In the event that samples cannot be conveniently collected for a given subject according to the scheduled sampling times specified above, study personnel should attempt to collect at least one pre-dose sample and at least one post-dose sample during the course of the study.

Date and time of most recent dose prior to PK sample collection, date and time of PK sample collection, and meal information relative to most recent dose, will be recorded on days of PK sample collection.

Plasma samples will be analyzed for SP-624 concentration using a validated bioanalytical method.

An aliquot of each subject's post-dose samples may be used by the bioanalytical laboratory for metabolite profiling. Potential metabolites will be identified using mass spectrometry techniques; and if conducted, these data will be summarized in a separate bioanalytical report.

[redacted]

Statistical Methods and Sample Size Justification:

Analysis Populations:

Within each population, analyses will be performed comparing subjects and by-treatment assignment (SP-624 vs placebo).

- All Subjects Population – all subjects who sign informed consent and are assigned a unique subject identification number. This population will be used to summarize disposition.
- Modified Intent-to-Treat (mITT) Population – all subjects who are randomized to receive treatment, receive at least one dose of study drug, and have a Baseline and at least one post-dose efficacy assessment. This population will be the primary population used to summarize demographics, baseline characteristics, medical history, treatment history, and all efficacy outcomes. Subjects will be analyzed according to randomized treatment group.
- Per Protocol (PP) Population – All subjects in the mITT population who received assigned study drug as randomized, completed the Week 4 assessments without any major protocol violations, and have 75-125% compliance. The PP population will be used as an additional population for summaries of efficacy outcomes.

- Safety Population — all subjects in the mITT population who receive at least one dose of study drug. Safety subjects will be summarized according to the treatment they actually received. This population will be used to summarize all safety assessments.
- PK Population — all subjects who receive active study drug and have evaluable PK data based on actual treatment received, protocol compliance, adequate numbers of samples, and successful sample assays.

Sample Size Justification:

Recent studies of MDD using change from Baseline in MADRS total scores suggest that placebo will yield an approximately nine unit decrease over four weeks with a standard deviation (SD) of around 9.2. SP-624 is expected to yield at least a 12 unit decrease in MADRS total score over four weeks with a similar SD of around 9.2. A minimum of 150 subjects per treatment arm (300 total) would be needed to detect a treatment difference of three units with 80% statistical power. The sample size was estimated using NQuery 8 (V8.5.2.0) two group test of equal means (MTT0-1: www.statsols.com).

Efficacy:

The primary efficacy variable is change from Baseline in MADRS total score at Week 4, for testing superiority of SP-624 vs. placebo. This variable will be analyzed using a Mixed Model for Repeated Measures (MMRM), with change from Baseline in MADRS total score at each visit as the response variable. The MMRM model will include age, weight, and baseline MADRS total score as covariates, with factors for treatment, visit (Weeks 1, 2, 3, and 4; as a categorical variable), treatment by visit interaction term, gender, and site as a fixed effect. An unstructured covariance matrix will be used for the within-subject correlation. Kenward-Rogers approximation will be used to calculate the denominator degrees of freedom. The primary efficacy endpoint will be calculated using an estimate statement. Subjects from low enrolling sites may be pooled in order to help with model convergence.

In the event of a model which fails to converge, analysis strategies will be detailed in the statistical analysis plan (SAP) which will be used to complete the analysis of the primary endpoint. This will include different model assumptions (e.g., restricted maximum likelihood vs maximum likelihood, covariance structure, and removing variables from the model [especially if they are associated with a high degree of missing values]).

Safety:

Laboratory parameters, vital sign data, and ECG results will be summarized by treatment group and presented in tabular and graphic formats where appropriate. Appropriate change from Baseline calculations will be provided to detect any significant changes in laboratory parameters, vital sign values, ECG results, and C-SSRS results, but no formal statistical tests are planned.

Pharmacokinetic Assessments:

Plasma concentration data for SP-624 will be listed and summarized using descriptive statistics.

If data allow, results will be used for other analyses (e.g., PopPK) which will be presented in a separate report.

The population PK analysis of SP-624 will be performed using a nonlinear mixed effects model (NONMEM). This analysis will provide estimates of population PK parameters such as CL/F, Vz/F, etc. and a measure of their variability. Pharmacokinetic parameters of individual subjects (e.g., AUC[0-tau], Cavg, Cmax) may be estimated and their correlation with relevant covariates (e.g., body weight, gender, concomitant medications) will be explored. The population PK analysis plan will be prepared separately.

Plasma concentration data will remain blinded until the unblinding of the clinical database at the end of the study.

SCHEDULE OF EVENTS

Table 2: Schedule of Events

Study Period	**Screening / Washout**	**Baseline**	**Treatment Period**				**Follow-up Period**	
Study Visit	**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**
Study Day / Week	**Day -28 to -2**	**Day -1**	**Week 1** (Day 7 ± 3 days)	**Week 2** (Day 14 ± 3 days)	**Week 3** (Day 21 ± 3 days)	**Week 4 / Prema-ture D/C** (Day 28 ± 3 days)	**Week 5** (7 days ±3 days post Week 4 Visit)	**Week 6** (14 days ±3 days post Week 4 Visit)
Event/Assessment								
Clinic Visit[1]	X	X	X	X	X	X	X	X
Informed Consent	X							
Inclusion/Exclusion	X	X						
Diagnosis Confirmation and Treatment History	X							
Demographics, Medical History	X	X						
Height and Weight[2]	X	X				X		
Physical Exam[3]	X	X	X	X	X	X	X	
C-SSRS[4]	X	X	X	X	X	X	X	X
Pregnancy Test[5]	X	X				X		
Urine Drug Screen	X							
Clinical Safety Labs[6]	X	X		X		X	X	
Vital Signs[7]	X	X	X	X	X	X	X	X
12-Lead ECG[8]	X	X	X	X	X	X	X	
MADRS	X	X	X	X	X	X	X	
CGI – S	X	X	X	X	X	X	X	
HAM-D, SDS, QIDS-SR; Q-LES-Q-SF		X		X		X		
Randomization		X						
Dispensation / Collection / Accountability of Study Drug		X	X	X	X	X		
Plasma PK Blood Sampling		X	X	X		X		
[redacted]		[redacted]		[redacted]		[redacted]	[redacted]	
Prior and Con Med Record	X	X	X	X	X	X	X	X
AE Monitoring	X	X	X	X	X	X	X	X

Abbreviations: AE=adverse event; con med=concomitant medication; CGI-S=clinical global impression-severity; C-SSRS=Columbia-Suicide Severity Rating Scale; ECG=electrocardiogram; MADRS=Montgomery-Asberg Depression Rating Scale; PK=pharmacokinetics; Premature D/C=Premature Discontinuation from treatment; QIDS-SR=Quick Inventory of Depressive Symptomology – Self Report; Q-LES-Q-SF=Quality of Life Enjoyment and Satisfaction Questionnaire – Short Form; SDS=Sheehan Disability Scale

[1] If a clinic visit is not possible (e.g., due to illness), a remote visit may be conducted to complete all assessments that can be done remotely and assess for adverse events and concomitant medications. The Screening, Baseline, and Week 4/Premature Discontinuation Visits cannot be done remotely. The sponsor should be contacted prior to conducting any remote visits whenever possible.

[2] Height and body weight at Visit 1. Body weight only at Visit 2 and Visit 6/Premature Discontinuation.

[3] Physical examinations will include a review of the following body systems: general appearance, skin and extremities, ear/eyes/nose/throat, head/neck, heart/chest, abdomen, musculoskeletal, and neurological.

[4] Baseline/Screening Version at Visit 1; Since Last Visit Version at Visits 2-8.

[5] All females; Serum pregnancy test at Visit 1 and Visit 6 (or Premature Discontinuation); Urine pregnancy test at Visit 2 – if urine pregnancy test at Visit 2 is positive, a confirmatory serum pregnancy test will be conducted.

[6] Clinical safety labs include clinical chemistry, hematology, coagulation, and urinalysis. Labs should be collected in a fasting state. Urinalysis not required at Visit 4 (Week 2 visit).

[7] Vital signs include blood pressure, pulse rate, and oral body temperature. Vital signs to be assessed in a sitting position after three mins or more of quiet sitting.

[8] Two ECGs to be collected at the Baseline Visit separated by approximately 10 minutes. One ECG collected at all other visits.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

1. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

The following abbreviations and terms are used in this study protocol.

Table 3: Abbreviations

Abbreviation	Explanation
AE	Adverse event
ALT	Alanine aminotransferase
ANCOVA	Analysis of covariance
API	Active pharmaceutical ingredient
AST	Aspartate aminotransferase
AUC	Area under the plasma concentration-time curve
AUC[0-tau]	Area under the plasma concentration-time curve during the 24-hour dosing interval
BMI	Body mass index
BOCF	Baseline observation carried forward
bpm	Beats per minute
BUN	Blood urea nitrogen
C	Celsius
Cavg	Average plasma concentration
CBC	Complete blood count
CFR	Code of Federal Regulations
CGI-S	Clinical Global Impression – Severity
CL/F	Apparent systemic clearance
CLss/F	Apparent systemic clearance after oral dosing at steady state
cm	Centimeter(s)
Cmax	Observed maximum serum concentration
CRF	Case report form
C-SSRS	Columbia-Suicide Severity Rating Scale
DBP	Diastolic blood pressure
DMP	Data management plan
DNA	Deoxyribonucleic acid
DSM-5	Diagnostic and Statistical Manual of Mental Disorders, Fifth Edition
ECG	Electrocardiogram

Abbreviation	Explanation
ECT	Electroconvulsive therapy
eGFR	Estimated glomerular filtration rate
FCS	Fully conditional specification
FDA	Food and Drug Administration
g	Gram(s)
GAD	Generalized anxiety disorder
GCP	Good Clinical Practice
GGT	Gamma-glutamyltransferase
GLP	Good Laboratory Practice
GMP	Good Manufacturing Practice
h	Hour(s)
HAM-D-17	17-item-Hamilton Depression Rating Scale
hCG	Human chorionic gonadotropin
HED	Human equivalent dose
HDL	High-density lipoprotein
HIV	Human immunodeficiency virus
ICF	Informed consent form
ICH	International Council for Harmonisation
IND	Investigational New Drug
INR	International normalized ratio
IRB	Institutional Review Board
LDH	Lactate dehydrogenase
LDL	Low-density lipoprotein
LOCF	Last observation carried forward
LTFU	Lost to follow up
MADRS	Montgomery-Asberg Depression Rating Scale
MAR	Missing at random
MCH	Mean corpuscular hemoglobin
MCHC	Mean corpuscular hemoglobin concentration
MCV	Mean cell volume
MDD	Major depressive disorder
MedDRA	Medical Dictionary for Regulatory Activities

Abbreviation	Explanation
mg	Milligram(s)
mL	Milliliter(s)
mm	Millimeter(s)
min	Minutes
MINI	Mini International Neuropsychiatric Interview
mITT	Modified intent-to-treat
MMRM	Mixed model for repeated measures
MNAR	Missing not at random
MPV	Mean platelet volume
MVN	Multivariate normal distribution
NAD+	Nicotinamide-adenine dinucleotide coenzyme
ng	Nanogram(s)
NONMEM	Nonlinear mixed effects model
PI	Principal Investigator
PK	Pharmacokinetic(s)
PP	Per protocol
PT	Prothrombin time
PTT	Partial thromboplastin time
QA	Quality assurance
QIDS-SR	Quick Inventory of Depressive Symptomology – Self-Report
Q-LES-Q-SF	Quality of Life Enjoyment and Satisfaction Questionnaire – Short Form
QTcF	Corrected QT interval using Fridericia's formula
RBC	Red blood cells
RDW	Red cell distribution width
REML	Restricted maximum likelihood
RNA	Ribonucleic acid
SAE	Serious adverse event
SAP	Statistical analysis plan
SBP	Systolic blood pressure
SD	Standard deviation
SDS	Sheehan Disability Scale

Abbreviation	Explanation
SIGH-D	HAM-D-17 – Structured Interview Guide Version
SIGMA	MADRS – Structured Interview Guide Version
[redacted]	[redacted]
SOE	Schedule of Events
SOPs	Standard operating procedures
SUSAR	Suspected and unexpected serious adverse reaction
t½	Terminal phase half-life
TEAE	Treatment-emergent adverse event
tmax	Time to reach maximum concentration
TSH	Thyroid stimulating hormone
ULN	Upper limit of normal
Vz/F	Apparent volume of distribution during the terminal phase
WBC	White blood cell
WHO	World Health Organization

2. INTRODUCTION

2.1. Major Depressive Disorder

Major depressive disorder, also known as unipolar or major depression, is characterized by a persistent feeling of sadness or a lack of interest in outside stimuli. The unipolar connotes a difference between MDD and bipolar depression, which refers to an oscillating state between depression and mania. Major depressive disorder is solely focused on the "lows," or the negative emotions and symptoms that are experienced (adapted from Lieber 2020). Depression is among the top public health concerns worldwide, causing significant disability and disease burden. In 2012, it was estimated that 16 million people were living with depression in the United States. The aim of antidepressant therapy is symptom remission or the reinstatement of euthymia—often defined as a score ≤7 on the total Hamilton Depression Rating Scale. Response, traditionally defined as a ≥50% decrease in the score from Baseline on a depression rating scale (commonly, the Hamilton Depression Rating Scale), has proven to be an inadequate goal, as many patients meeting the criteria for response will continue to have residual symptoms and functional impairments. Moreover, unremitted patients report poorer quality of life and are at a higher risk for relapse and recurrence of depression. Specifically, in the Sequenced Treatment Alternatives to Relieve Depression (STAR*D) trial—the largest randomized depression study to date—68% of responders relapsed within the first year following treatment, compared with 47% of remitters who relapsed. In addition, when more treatment steps are needed, lower acute remission rates and higher relapse rates are to be expected. Indeed, one third of the STAR*D cohort never remitted, even after four consecutive treatment trials. The remission rate was a mere 27.5% in Phase 1 treatment with citalopram, based on HAM-D scores, and 32.9% based on the Quick Inventory of Depressive Symptomology – Self-Report (QIDS-SR) scores (adapted from Ionescu et al., 2015). New antidepressant treatments with new mechanisms of action are needed to treat MDD.

2.2. Investigational Product

[redacted]

2.2.1. [redacted]

[redacted]

2.2.2. Non-clinical Studies

[redacted]

2.2.2.1. [redacted]

[redacted]

ere

[redacted]

2.2.2.2. [redacted]

[redacted]

2.2.3. Clinical Studies

2.2.3.1. SP-624-101

SP-624 was assessed in a single, oral dose study, SP-624-101. The doses assessed following an overnight fast were 3 mg, 10 mg, and 30 mg. Additionally, 10 mg was assessed in a food effect cohort following an overnight fast and 30 minutes after a high fat meal. Each cohort in the first part of the study (non-food effect) comprised of eight subjects each, of whom six subjects per

cohort received single oral doses of SP-624 and two subjects received placebo. In the food effect part of the study, a separate cohort of eight subjects received SP-624 once under fasting conditions and once under fed conditions.

Orally administered SP-624 was well-tolerated after each dose.

There were no severe AEs, SAEs, deaths, or AEs leading to withdrawal in this study.

No placebo subjects reported an AE, and three SP-624 subjects reported at least one AE. One subject in the 3 mg dose group reported headache and insomnia, one subject in the 10 mg food effect fasted dose group reported headache, and one subject in the 10 mg food effect fed dose group reported vessel puncture site pain. All AEs were assessed as mild in intensity and not related to study drug.

Shifts in laboratory values appeared to be transient, and no patterns of clinical significance were observed. There were no laboratory values in the study that were deemed clinically significant by the Principal Investigator (PI).

There were no apparent patterns of mean change from Baseline in vital signs to suggest any effect from SP-624. There were no clinically relevant changes from Baseline in ECG parameters.

SP-624 displayed linear, dose-proportional PK when dosed following an overnight fast. At all dose levels SP-624 appeared to have a distinct absorption profile with some absorption occurring over the first two hours followed by a spike to the Cmax. Geometric mean plasma half-lives were similar across dose levels at 7.1, 6.8 and 6.8 hours for the 3, 10 and 30 mg cohorts, respectively. Volumes of distribution (Vz/F) were similar between cohorts (175, 230, and 209 liters) and displayed low inter-subject variability. Apparent systemic clearance (CL/F) values were consistent between cohorts (17.1, 23.6, and 21.4 L/h) with low variability.

When administered 30 minutes after a high fat meal (10 mg oral dose), plasma exposure to SP-624 was unchanged as assessed by area under the curve (AUC), but mean Cmax was decreased by approximately 30% and mean tmax was delayed from 2.25 hours to 5.13 hours.

2.2.3.2. SP-624-102

SP-624 was assessed in a multiple dose study, SP-624-102. The doses assessed were 3 mg, 10 mg, and 20 mg. The 3 and 10 mg doses were orally administered, once daily, for five days. The 20 mg dose was orally administered, once daily, for 10 days. Cohorts 1 and 2 (3 mg and 10 mg) comprised eight subjects each, of whom six received SP-624 and two received placebo. Cohort 3 comprised seven subjects, of whom five received SP-624 and two received placebo.

Orally administered SP-624 was well-tolerated after each dose. No safety concerns were identified for electrocardiography, vital signs, safety laboratories or adverse events for any cohort of subjects.

There were no severe AEs, SAEs, deaths, or AEs leading to withdrawal in this study.

No placebo subjects reported an AE, and five SP-624 subjects reported a total of 12 AEs. All AEs were reported by single subjects. All AEs were assessed as mild in intensity with the exception of arthralgia that was reported twice for one subject in the 3 mg SP-624 dose group;

once at mild intensity and once at moderate intensity. No AEs were assessed as probably related to study drug; eight AEs were assessed as possibly related: pallor (3 mg QD), procedural dizziness (3 mg QD), defecation urgency (20 mg QD), diarrhea (20 mg QD), melanocytic nevus (20 mg QD), oropharyngeal discomfort (20 mg QD), tongue blistering (20 mg QD), and tongue discomfort (20 mg QD).

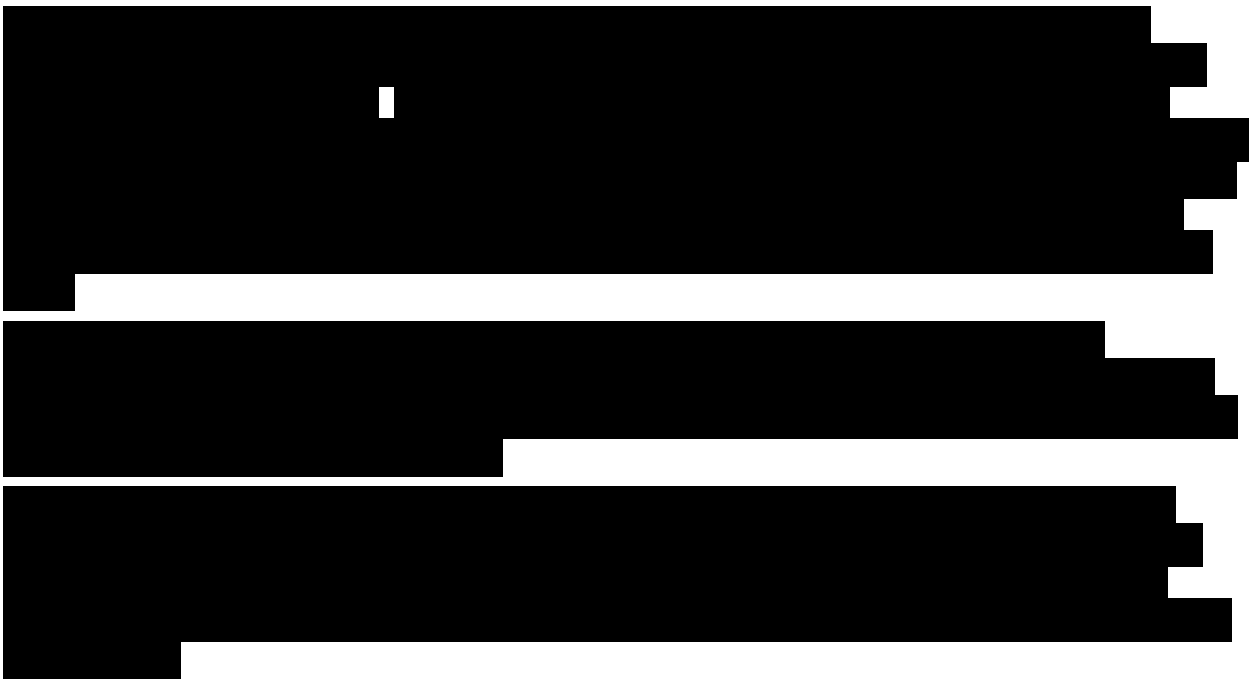
AUC and Cmax values increased in proportion to dose during multiple dosing of SP-624 at dose levels of 3, 10, and 20 mg per day. Cmax exceeded predicted efficacious plasma concentrations of 3.28 ng/mL at all doses. t½ and CLss/F remained constant as the dose was increased. There was little or no accumulation during 5 days of dosing at 3, 10, and 20 mg SP-624 or 10 days of dosing at 20 mg SP-624.

2.3. SP-624 Development Plan

Sirtsei Pharmaceuticals, Inc. plans to investigate the safety and efficacy of SP-624 in this pilot, double-blind, placebo controlled, multicenter study in adults with moderate to severe MDD. The study will evaluate the safety and efficacy of a 4-week, multidose regimen of SP-624 versus placebo in approximately 300 subjects.

Pending the efficacy and safety profile observed from this study, additional nonclinical and clinical studies will be conducted.

2.3.1. Rationale for Dose



2.3.2. Benefit and Risk Assessment

SP-624 was well tolerated at a dose of 20 mg per day for 10 days in study SP-624-102. There were no SAEs, AEs leading to discontinuation, or severe AEs. There were no changes of

concern in clinical laboratory findings, physical exams, vital signs, ECGs, or mental status. Steady state Cmax values were well in excess of predicted efficacious concentrations.

It is concluded that the dose of SP-624 planned in this study, together with the safety monitoring procedures, has a low foreseeable risk of inflicting harm or injury to study participants. This is the first study of SP-624 in subjects with MDD and the study may provide initial evidence of efficacy. The Sponsor believes that the potential benefit of SP-624 for MDD and good tolerability profile supports conduct of this clinical study.

3. STUDY OBJECTIVES AND ENDPOINTS

3.1. Objectives

3.1.1. Primary Objective

- Evaluate the efficacy of SP-624 administered once-daily for up to 4 weeks compared to placebo in the treatment of adults with MDD.

3.1.2. Secondary Objectives

- Evaluate the safety and tolerability of SP-624 administered once-daily for up to 4 weeks compared to placebo in the treatment of adults with MDD; and
- Characterize the population PK of SP-624 in plasma when administered orally to subjects with MDD.

3.2. Endpoints

3.2.1. Efficacy Endpoints

3.2.1.1. Primary Endpoint

- Change from Baseline to Week 4 in MADRS total score

3.2.1.2. Secondary Endpoints

- Change from Baseline to Weeks 1, 2, and 3 in MADRS total score
- Change from Baseline to Weeks 1, 2, 3, and 4 in CGI – S total score
- Change from Baseline to Week 5 and change from Week 4 to Week 5 in MADRS total score and CGI-S total score
- Change from Baseline to Week 2 and Week 4 in the HAM-D-17 total score
- Change from Baseline to Week 2 and Week 4 in the SDS
- Change from Baseline to Week 2 and Week 4 in the QIDS-SR
- Change from Baseline to Week 2 and Week 4 in the Q-LES-Q-SF

3.2.2. Safety Endpoints

- AEs
- Clinical safety laboratory tests (serum chemistry [including thyroid function], hematology, coagulation, and urinalysis tests)
- Vital signs (blood pressure, pulse rate, and oral body temperature)
- Body weight
- 12-lead ECGs

- Physical examinations
- C-SSRS

3.2.3. Pharmacokinetic Endpoints

A total of four blood samples will be collected from each subject for the measurement of plasma concentrations of SP-624 at the following visits:

- Visit 2 (Day -1)
- Visit 3 (Week 1): Pre-Dose
- Visit 4 (Week 2): Pre-Dose or approximately 2 to 10 hours post-dose (whichever is more convenient for the subject)
- Visit 6 (Week 4): approximately 2 to 10 hours post-dose

3.2.4. [redacted]

[redacted]

4. INVESTIGATIONAL PLAN

4.1. Overall Study Design

This is a Phase 2, multicenter, double-blind, randomized, placebo-controlled study of the safety and efficacy of SP-624 in the treatment of adult subjects with MDD as defined by the DSM-5. Following the successful completion of a Screening Phase, subjects will be randomized to one of two treatment groups in a 1:1 ratio and will receive SP-624 or placebo over a treatment period of four weeks (Treatment Group #1: SP-624 20 mg/day; Treatment Group #2: Placebo).

Approximately 300 subjects (150 per treatment group) will be enrolled into the study. Each subject will complete Screening, Baseline, Treatment, and Follow-up Visits.

The Schedule of Events (SOE) to be completed for subjects in the study is provided in Table 2.

An overview of the study schematics is displayed in Figure 1. Study periods are detailed below.

4.1.1. Screening Period and Baseline Visit

A Screening period of up to 28 days (Day -28 to Day -2) and a Baseline Visit (Day -1) will precede initiation of the Treatment Period. During the Screening Period, all Screening assessments will be completed and any current depression medications will be discontinued. All Screening assessments should be completed before discontinuing any current depression medications. An adequate number of subjects will be screened to ensure the planned number of subjects is enrolled (approximately 300 subjects).

The Screening Period may be extended for extenuating circumstances, such as when additional time is needed for medication taper/washout or to repeat laboratory testing, with the approval of the medical monitor.

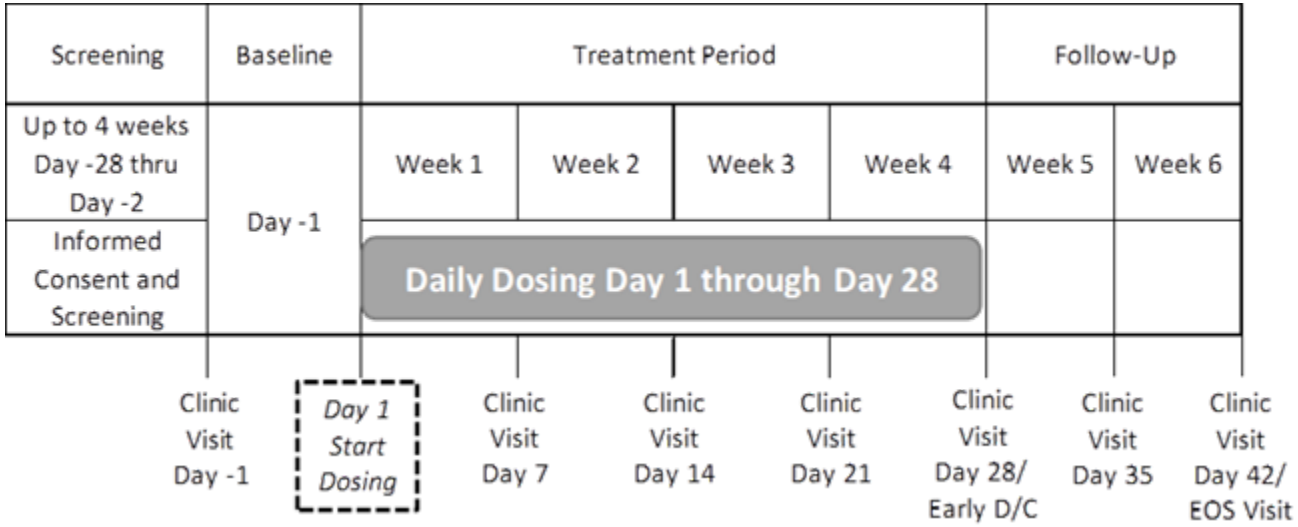
Subjects who continue to satisfy all entry criteria at the Baseline Visit will be randomized in a 1:1 ratio to one of two treatment groups (Treatment Group #1: SP-624 20 mg/day; Treatment Group #2: placebo). At the completion of the Baseline Visit, randomized subjects will be dispensed study drug and will be instructed to begin dosing the next morning (Day 1).

4.1.2. Treatment and Follow-up Periods

Subjects will be scheduled to return to the clinic for safety and efficacy assessments at the end of Weeks 1, 2, 3, 4, and 5 as specified in the SOE (Table 2). After the final dose of study drug, subjects will complete a Follow-up Period of two weeks and will return at the end of Weeks 5 and 6. Subjects may be treated with anti-depressant medications according to physician recommendations after completion of the Week 5 Visit unless a clinically significant lab value is identified at the Week 4 Visit. If a clinically significant lab value is identified at the Week 4 Visit, consult with the Medical Monitor to determine if the lab value needs to be followed prior to initiating new therapy.

Unless there are any outstanding safety issues that require follow-up, subjects will be discharged from the study at the Week 6 Visit.

Figure 1: Study Schematic



4.2. Randomization and Blinding

4.2.1. Subject Identification and Randomization to Treatment

A unique subject identification number (sometimes referred to as the Screening number and different from the randomization number) should be assigned to each subject who signs an informed consent form (ICF). The subject identification number should be used on all case report forms (CRFs) and in all correspondence between sites, the Sponsor, Institutional Review Boards (IRBs), and the Food and Drug Administration (FDA).

Sirtsei Pharmaceuticals or designee will generate a randomization scheme with appropriate blocking. Randomization numbers will be 3 digits starting with 101. When the decision is made to randomize a subject to treatment, the Investigator should pick the next available randomization number from the study drug kits assigned to the site. The subject randomization number will be recorded in the CRF, linking the subject's unique subject identification number to the subject's assigned randomization number.

4.2.2. Blinding

The study will be conducted under double-blind conditions, meaning the Investigator, study center personnel, study subjects, and Sponsor will be unaware of the treatment assignment for the subjects.

4.2.3. Emergency Unblinding

If an emergency situation occurs that, in the Investigator's opinion, cannot be adequately treated without knowing the identity of the study medication, the study treatment assignment can be unblinded via the tamper-evident covered disclosure panel attached to the study drug kit.

Every effort will be made to contact the Sponsor Medical Monitor prior to breaking the blind. If this is not possible and the situation is an emergency, the Investigator may break the blind for only the subject(s) involved in the emergency situation and must contact the Sponsor Medical Monitor as soon as possible (see Table 1 for Medical Monitor contact information).

In any case of emergency unblinding, pertinent information including the date and reasons for breaking the blind must be documented in the subject CRF and in the site and Sponsor study files.

4.3. Study Centers and Personnel

The study will be conducted at approximately 40 investigative sites located in the United States.

The PI at each site will be a licensed physician or clinical psychologist experienced in conducting studies in MDD. For those sites where a clinical psychologist is the PI, one of the sub-Investigators must be a licensed physician. The PI will be responsible for delegating study responsibilities to only those key site personnel who have the appropriate level of education, training and experience, including medical expertise and good clinical practice (GCP) training, as applicable for their assigned role in the study.

The clinical research facility at each investigative site will have adequate facilities and equipment to conduct the study procedures, and to allow for close supervision of study subjects,

including immediate access to equipment and appropriately qualified staff to respond in the case of an acute medical emergency.

4.4. Number of Subjects

Approximately 300 adult subjects with MDD as defined by DSM-5 will be enrolled with approximately 150 subjects per treatment group.

4.5. Study Duration

Subjects will receive double-blind treatment for up to 4 weeks, following a Screening/washout period of up to 28 days. After the treatment period, subjects will complete a 2-week follow-up period. The planned study duration for each subject will be approximately 70 days.

5. SELECTION AND WITHDRAWAL OF SUBJECTS

5.1. Subject Recruitment, Enrollment, and Identification

Subject recruitment and enrollment will be organized and overseen by the Investigator at the site following approval of the study protocol, ICF, and any study specific advertisements/recruitment materials by the respective IRB.

Subjects will be recruited via advertisements, physician referral, and from databases kept by the research center.

A sufficient number of subjects will be screened in order to ensure that a total of approximately 300 subjects who satisfy the entry criteria are randomized to treatment.

Each subject's signed and dated ICF to participate in the study must be obtained before conducting any study-related procedures. All subjects who sign an ICF will be given a unique Screening identification number and their study status will be tracked on a study Screening and Enrollment log maintained in the Investigator study files.

5.2. Subject Inclusion Criteria

To be eligible for this study, subjects must meet all of the following inclusion criteria.

1. Willing and able to provide written informed consent to participate in the study.
2. Males and females, aged 18 to 65 years, inclusive.
3. In generally good physical health, in the opinion of the Investigator.
4. Body mass index must be ≥ 18 and ≤ 40 kg/m^2
5. Females of reproductive potential and males with partners of reproductive potential must agree to remain abstinent or use adequate and reliable contraception throughout the study and for at least 30 days after the last dose of study drug.
    a. Adequate contraception is defined as continuous use of double-barrier method (e.g., condom and spermicide or diaphragm and spermicide), oral contraceptive, implant, dermal contraception, long-term injectable contraceptive, or intrauterine device.
    b. Non-reproductive potential is defined as surgically sterile, undergone tubal ligation or vasectomy, or postmenopausal (defined as at least 12 months of spontaneous amenorrhea).
    c. Subjects of reproductive potential but whose sole partner is of non-reproductive potential, or whose sole partner is of the same sex, are not required to maintain abstinence or contraception.
    d. Subjects must agree to refrain from sperm and oocyte donation for at least 90 days after the last dose of study drug.
6. Meets DSM-5 criteria for moderate to severe MDD, as confirmed by the MINI. Subjects with a diagnosis of co-morbid GAD may be included if MDD is the primary diagnosis.
7. Montgomery-Asberg Depression Rating Scale total score ≥ 27 at both Screening and Baseline.

8. History of inadequate or non-response to at least one adequate treatment, but no more than three of the same class of antidepressant, (i.e., an adequate dose for at least six weeks) for the current episode, in the opinion of the Investigator. NOTE: If immediately prior to this episode starting, treatment was working to the point of remission, but then the depression worsened, subject should not be enrolled. For example, subject should not have been on an antidepressant and in remission prior to the start of the current episode.
9. Willing and able to comply with the study design schedule and other requirements.

5.3. Subject Exclusion Criteria

A subject who meets any of the following exclusion criteria will not be enrolled in the study:

1. Female who is pregnant, breastfeeding, or less than six months postpartum at Screening.
2. History or presence of major bowel resection, total gastrectomy, or diabetes, and/or history or presence of any clinically significant medical condition, disease, or surgical history that could, in the opinion of the Investigator, jeopardize the safety of the subject or validity of the study data, or interfere with the absorption, distribution, metabolism, or excretion of the study drug.
3. A current DSM-5 diagnosis of depression with peripartum onset, panic disorder, obsessive compulsive disorder, post-traumatic stress disorder, anorexia nervosa, or bulimia nervosa.
4. A history of or current DSM-5 diagnosis of MDD with psychotic features, any schizophrenia spectrum and other psychotic disorders, bipolar disorder, or personality disorder.
5. History of treatment with ECT, vagus nerve stimulation, deep brain stimulation, or transcranial magnetic stimulation within six months of Screening.
6. Current episode of depression exceeds 2 years or is less than six weeks from Screening.
7. History of inadequate or non-response, in the opinion of the Investigator, to an adequate treatment regimen of two or more classes of antidepressants during the current episode.
8. Subjects treated with an antipsychotic during the current episode should be excluded.
9. Subjects who meet DSM-5 criteria for moderate or severe substance use disorder for any substance except nicotine or caffeine, within 12 months of Screening. Subjects with a urine drug screen positive for suspected substance of abuse will be excluded. Urine drug screen positive for cannabinoids is not exclusionary if the subject does not meet criteria for moderate or severe substance use disorder.
10. Will require initiation of psychotherapy during the study period, in the Investigator's opinion. If a subject has been in ongoing psychotherapy for a minimum of three months, the therapy may continue if it remains unchanged. Cognitive behavioral therapy is excluded.
11. Poses a current or future suicidal risk, in the Investigator's opinion, or has a history of suicidal behavior within the past 12 months, and/or subject answers "yes" to "Suicidal Ideation" Item 4 (active suicidal ideation with some intent to act, without specific plan)

or Item 5 (active suicidal ideation with specific plan and intent) on the C-SSRS assessment during the six months prior to the Screening period and/or at Baseline.

12. Failure to discontinue all psychoactive medications or psychoactive supplements including antidepressants and mood stabilizers, within a time period prior to Baseline corresponding to at least five half-lives of the medication in question.
13. Presence of a clinically significant abnormality on physical examination or ECG, including a QTcF >450 msec for males and >470 msec for females. Note: ECG may be repeated once at both Screening and Baseline for confirmatory purposes.
14. Presence or history of any known clinically significant cardiovascular disorders including, but not limited to: coronary artery disease, heart failure, valvular heart disease, cardiomyopathies, myocardial infarction, chamber enlargement or hypertrophy, or orthostatic hypotension.
15. Presence of uncontrolled hypertension, defined as consistent SBP >160 mmHg or consistent DBP >95 mmHg despite present therapy. Note: Blood pressure measurements may be repeated twice at both Screening and Baseline for confirmatory purposes.
16. Screening laboratory value(s) outside the laboratory reference range that are considered to be clinically significant by the Investigator (clinical chemistry, hematology, coagulation, and urinalysis). Note: Screening labs may be repeated once for confirmatory purposes.
17. Screening liver function tests (ALT, AST, Alkaline phosphatase) > 2x the upper limit of normal.
18. Known history of current or chronic Hepatitis B or Hepatitis C infection, or a known history of HIV infection. Subjects with a history of treated Hepatitis C infection where liver function tests are within normal limits may participate.
19. Subjects with a history of seizures or seizure disorder.
    Note: Subjects with a history of only febrile seizures, limited to up to two episodes, are permitted.
20. Clinically significant acute infection, other than mild viral or bacterial infections from which the subject has fully recovered (e.g., common cold, localized skin infection, etc.), within the two weeks prior to Baseline.
21. Major trauma or surgery in the 12 weeks prior to Screening or at any time between Screening and randomization.
22. Presence or history of malignancy within the last five years. Note: History of adequately treated basal cell or squamous cell carcinoma of the skin or in situ cervical cancer is allowed.
23. History of an anaphylactic allergic reaction to previous medication.
24. Blood or plasma donation within 30 days of randomization, or planned donations or elective surgery during the study or during the 60 days following the study.
25. Subjects who, in the opinion of the Investigator, are not suitable candidates for the study.

26. Individuals who are currently enrolled in or have participated in a clinical trial (i.e., received study drug) involving an investigational product within 60 days or within five half-lives of the investigational product (whichever is longer) prior to Baseline.

5.4. Subject Withdrawal from the Study

A subject may be withdrawn from the study at any time at the discretion of the Investigator.

Justifiable reasons to discontinue a subject from the study may include, but are not limited to, safety, ability or willingness to comply with conditions of the protocol, withdrawal of consent, lost to follow-up, Investigator discretion, Sponsor decision, or premature termination of the study.

The reason(s) for premature discontinuation will be documented on the subject's CRF.

Any AEs or SAEs that lead to study discontinuation should be followed as appropriate until the AE has resolved or stabilized (in the opinion of the Investigator and Sponsor Medical Monitor).

Subjects who prematurely discontinue after receiving a dose of study drug, should continue to be followed for scheduled safety assessments according to the SOE (Table 2), if possible. At a minimum, every attempt should be made to complete the Week 4 / Premature Discontinuation procedures; however, subjects are free to withdraw consent from participation in the study at any time, without prejudice to their medical care.

If a subject is lost to follow-up (i.e., fails to return for study visits), all reasonable efforts should be made to contact the subject and complete the Week 4 / Premature Discontinuation procedures.

Should it be discovered after a subject has begun study treatment that the subject did not meet all inclusion and exclusion criteria, or if during the study a subject ceases to meet all inclusion and exclusion criteria, the subject's continued participation in the study will be discussed with the Medical Monitor.

5.5. Premature Termination of the Study

If the study is prematurely terminated, written notification, documenting the reason for study termination, will be promptly provided by the Sponsor to the regulatory authorities. The Investigator will be responsible for informing the IRB.

The Sponsor reserves the right to terminate the study for any reason at any time.

6. STUDY TREATMENTS, MATERIALS, AND MANAGEMENT

6.1. Description of Study Drug

The study drug code name is SP-624. SP-624 is [redacted] intended for the treatment of MDD.

The clinical trial material for this study will consist of capsules, each containing 10 mg of SP-624 API. Matching placebo capsules identical in shape and color to the active capsules will also be used.

6.2. Acquisition of Study Drug Supplies

After the research site has submitted all approved regulatory and study start-up documents, the Sponsor will initiate a requisition for an initial shipment of study kits to be delivered to the site via the Sponsor's distribution facility.

All manufacturing, packaging, and labeling operations for the supplied study drug materials will be performed according to Good Manufacturing Practice (GMP) and GCP guidelines, as well as local regulations.

Storage and retest information will be provided for all supplied materials.

Upon receipt of the materials, the Investigator or designee will examine the package container, labeling, and appearance of the packaged materials to confirm the packaged materials were received in good condition.

All study materials will be stored at the study site in a restricted-access, securely locked area, under controlled temperature and humidity conditions in accordance with the manufacturer's instructions (see Section 6.7).

6.3. Subject Treatment Assignment

When the decision is made to randomize a subject, the Investigator should pick the next randomization number from the study drug kits assigned to the site. The subject randomization number will be recorded in the CRF.

6.4. Study Drug Administration

Each dose of SP-624 will be supplied as two capsules, each containing 10 mg of API. Matching placebo capsules identical in shape and color to the active capsules will also be used. Following successful completion of the Baseline Visit, subjects will be randomized in a 1:1 ratio to one of two treatment groups:

- Treatment Group #1: Two 10 mg capsules SP-624 each morning for 4 weeks
- Treatment Group #2: Two capsules placebo each morning for 4 weeks

Each study drug dose should be taken orally, once daily at approximately the same time each morning. Study drug doses should be taken on an empty stomach before breakfast, and no food should be consumed for at least 1 hour after dosing. High fat foods at breakfast (e.g., bacon,

cheese, eggs, etc.) should be avoided. Morning liquids such as coffee or tea are allowed without any additives (eg, cream, sugar, etc).

If dosing on an empty stomach is not practical or not well tolerated, then a low fat, low calorie breakfast 2 hours before dosing, or 30-60 minutes after dosing, is acceptable. If dosing remains not well tolerated, then subjects should dose 30 minutes after a low fat, low calorie meal.

Study drug will be packaged in subject kits and each kit will contain four weekly blister cards with each card containing 20 capsules of study drug (two capsules per dose for seven days of dosing, plus three extra days of dosing as a provision for study visit scheduling). Each subject kit will be labeled with a 3-part label that specifies the randomization number, cautionary statement, and storage conditions. Each blister card will have a one-part label that specifies the randomization number, week, number of capsules, cautionary statement, and storage conditions.

Study drug will be dispensed as follows:

- Baseline Visit – one blister card for Week 1
- Week 1 Visit – one blister card for Week 2
- Week 2 Visit – one blister card for Week 3
- Week 3 Visit – one blister card for Week 4

Subjects will be instructed to return study drug at the Week 1, Week, 2, Week 3, and Week 4 (or Premature Discontinuation) Visits.

If the two capsule dose, is not well tolerated, the dose can be decreased to one capsule per dose after Week 1.

6.5. Treatment Compliance

Study drug accountability will be recorded by study site staff, and any deviations from the administration instructions or dosing schedule will be entered into the subject's CRF.

6.6. Study Drug Accountability

The Investigator (or designee) is responsible for the accountability, reconciliation, and record maintenance of all study drug receipt, use, and disposition. In accordance with all applicable regulatory requirements, the Investigator or designated study personnel must maintain study drug accountability and storage records throughout the study. Unused drug will be returned to the investigative site and to the Sponsor or designee at the end of the study for destruction. No study drug usage outside the protocol will be allowed.

When a kit is assigned to a subject, the Investigator will tear off a 2 part segment of the label that contains a tamper-evident covered disclosure panel to be used for unblinding in case of emergency and affix the 2-part segment to the drug dispensing record. The drug dispensing record will be collected for the sponsor’s files at the end of the study. This will serve to connect the unique randomization number to the unique subject identification number assigned at Screening.

6.7. Study Drug Storage and Stability

All study materials will be stored at the study site in a restricted-access, securely locked area, under controlled temperature and humidity conditions in accordance with the manufacturer's instructions.

The storage conditions for the study drug will be indicated on the label.

All supplied study drug materials will be stored at the study site in a restricted-access, securely locked area at the long-term refrigerated storage condition, 2-8ºC. After a study drug kit has been assigned to a subject, the study drug kit and weekly blister cards dispensed to study subjects may be stored under room temperature conditions, 25°C (77°F), with excursions permitted between 15°C and 30°C (59°F-86°F).

In the solid state, the SP-624 drug substance is stable for at least 18 months when stored at 5ºC and for at least 6 months when stored at 25ºC/60% relative humidity.

SP-624 drug product stability is expected to directly correlate with that of the drug substance, since the drug product is neat drug substance filled into capsules. A 24-month stability program will be initiated for these capsules once manufacturing is completed, with storage conditions at 2-8ºC and 25ºC/60% relative humidity. Retest date information for the study drug will be provided.

At the Week 1, Week 2, Week 3, and Week 4 Visits, subjects will return the previously used blister cards, including any unused study drug. The used study drug kits will be maintained at the site under room temperature storage conditions until returned to the Sponsor or designee.

6.8. Prior and Concomitant Medications

Medications taken within 28 days before Day 1 will be documented as prior medications. Medications taken any time on or after Day 1 and until completion of the Week 4 / Premature Discontinuation Visit will be documented as concomitant medications. Medications that started after the date of the last dose of study drug will not be considered concomitant but will be considered post-treatment.

The use of all prior, concomitant, and post-treatment medications will be recorded in the subject's CRF, including the drug name, dose, route of administration, dates of administration, and indication.

Once all Screening assessments have been completed, any current psychoactive medications and supplements must be discontinued. Subjects should consult with the Investigator before taking any medications, including over-the-counter products.

Subjects may be treated with anti-depressant medications according to physician recommendations after completion of the Week 5 Visit unless a clinically significant lab value is identified at the Week 4 Visit. If a clinically significant lab value is identified at the Week 4 Visit, consult with the Medical Monitor to determine if the lab value needs to be followed prior to initiating new therapy.

Previous medications for medical, non-psychiatric illnesses, as well as oral contraceptives and contraceptive devices, are permitted.

Medications such as non-steroidal anti-inflammatories, antacids, and non-psychoactive allergy medications, may be given at the discretion of the Investigator.

A list of restricted and prohibited medications is included in Table 4:

Table 4: Restricted and Prohibited Medications

Medication Class	**Examples**[1]	**Comment**
Antidepressants	Trazodone TCAs SSRIs SNRIs MAOIs	All prohibited Any anti-depressants being taken at the time of Screen must be discontinued within a time period prior to Baseline corresponding to at least five half-lives of the medication in question.
Antipsychotics	Aripiprazole Brexpiprazole Haloperidol Olanzapine Risperidone	All prohibited If an antipsychotic was used to treat the current episode of depression, the subject should be excluded.
Anxiolytics	Benzodiazepines	Restricted Subjects on a stable dose of lorazepam (2 mg/day), or equivalent dose of another benzodiazepine (e.g., diazepam 15 mg), for anxiety at the time of Screening may continue with the current treatment regimen. The dose should remain consistent throughout the study. Should a benzodiazepine be required for increased anxiety during the study in a subject who had not been taking a consistent dose prior to Screening, lorazepam 2 mg/day, or an equivalent dose of another benzodiazepine (e.g., diazepam 15 mg), is limited to two days or less per week. Use the night before or the day of a study visit (prior to the visit) is prohibited. Any need to exceed this amount must be discussed with the Medical Monitor.
Hypnotics	Zolpidem Temazepam Sedating antihistamines	Restricted Zolpidem being taken daily for sleep at the time of the Screening visit is permitted for severe insomnia. The dose should remain consistent throughout the study. If zolpidem is being taken as needed at the time of Screening, use is limited to three nights or less per week and use the night before a study visit is prohibited. Prohibited All other hypnotics

Medication Class	**Examples**[1]	**Comment**
Mood stabilizers and anticonvulsants	Lithium Valproate Lamotrigine Gabapentin	<u>All prohibited</u> Any mood stabilizers or anticonvulsants being taken at the time of Screening must be discontinued within a time period prior to Baseline corresponding to at least five half-lives of the medication in question.
Muscle relaxers	Carisoprodol Cyclobenzaprine Metaxalone Baclofen	<u>Restricted</u> Use of muscle relaxants during the study is limited to episodic use for an acute situation (e.g., back injury during the study) and must be discussed with the Medical Monitor.
Opioid analgesics	Oxycodone Morphine Tramadol Codeine	<u>Restricted</u> Use of opioids during the study is limited to episodic use for an acute situation (e.g., back injury during the study) and must be discussed with the Medical Monitor.
Stimulants	Amphetamine Dextroamphetamine Pemoline Methylphenidate	<u>All prohibited</u> Any stimulants being taken at the time of Screening must be discontinued within a time period prior to Baseline corresponding to at least five half-lives of the medication in question.
Herbal and nutritional supplements	St. John's Wort Ginkgo Biloba S-Adenosyl methionine (SAM-e) Omega-3 supplements Cannabidiol products Melatonin	<u>Restricted</u> Only multivitamins that do not contain any of the example ingredients are allowed during the study.

[1] Medications including, but not limited to, examples provided.

7. STUDY PROCEDURES AND ASSESSMENTS

7.1. Schedule of Events

The SOE for participants in the study is provided in Table 2.

The actual dates of study drug administration and all study visits and assessments will be recorded in the subject's CRF. If a study visit or assessment occurs outside the specified time point and allowed time window (if applicable), then the reason for the deviation will be noted. Every attempt should be made to ensure that subsequent visits and assessments are performed on-time.

The Screening Period may be extended for extenuating circumstances, such as when additional time is needed for medication taper/washout or to repeat laboratory testing, with the approval of the medical monitor.

Study visits during the Treatment Period can be scheduled within a ± 3 day window relative to the Baseline Visit (i.e., the Week 1 Visit can be scheduled 7 ± 3 days relative to the completion of the Baseline Visit, the Week 2 Visit can be scheduled 14 ± 3 days relative to the completion of the Baseline Visit, the Week 3 Visit can be scheduled 21 ± 3 days relative to the completion of the Baseline Visit, etc.).

The Week 5 Visit can be scheduled 7 ± 3 days relative to the Week 4 / Premature Discontinuation Visit and the Week 6 Visit can be scheduled 14 ± 3 days relative to the Week 4 / Premature Discontinuation Visit.

Whenever ECGs, vital signs, and a blood draw for PK and/or safety labs are scheduled on a given day, the ECG and vital signs should be completed in the sequence of procedures either prior to the blood draw (preferred) or after the blood draw, such that the blood draw does not interfere with the ECG and vital sign recordings.

In addition to the study visits and assessments described in the SOE, unscheduled visits/assessments may be performed or repeated at the discretion of the Investigator if required to further evaluate laboratory parameters or the subject's clinical safety.

7.2. Study-Specific Procedures

7.2.1. Informed Consent

The Investigator must inform each prospective subject of the nature of the study, explain the potential risks, and obtain written informed consent from the subject prior to performing any study-related Screening procedures. Once written informed consent is obtained, prospective subjects will be screened for eligibility into the study based on the inclusion and exclusion criteria detailed in Section 5.

7.2.2. Demographic Information, Medical History/Current Medical Conditions, and Treatment History

Demographic information (e.g., date of birth, sex, race, ethnicity) will be collected and a detailed medical history, including review of past and ongoing medical conditions or procedures will be

obtained during the Screening Period as specified in the SOE (Table 2). Additionally, treatment history for the current depressive episode will also be collected.

7.2.3. Mini International Neuropsychiatric Interview

Subjects must meet DSM-5 criteria for moderate to severe MDD, as confirmed by the MINI (Sheehan et. al, 1998). Subjects with a diagnosis of co-morbid GAD may be included if MDD is the primary diagnosis. The MINI should only be completed by Sponsor approved raters. Raters will be approved by the Sponsor based on education and experience conducting the MINI in subjects with MDD.

Current DSM-5 diagnoses of depression with peripartum onset, panic disorder, obsessive compulsive disorder, post-traumatic stress disorder, anorexia nervosa, or bulimia nervosa are exclusionary.

A history of or current DSM-5 diagnosis of MDD with psychotic features, any schizophrenia spectrum and other psychotic disorders, bipolar disorder, or personality disorder are exclusionary.

7.2.4. Assessment of Efficacy

Efficacy assessments will be conducted by individuals with sufficient experience conducting the below rating scales who have received centralized rater training and have been approved by the PI and Sponsor. Whenever possible, the same rater should assess the same subject throughout the study.

The schedule of these safety assessments is provided in the SOE (Table 2), and the study-specific procedures for completion of these assessments are detailed below.

Assessments should not be performed on subjects who have missed two or more consecutive study drug doses immediately before the Week 1, Week 2, Week 3, or Week 4 / Premature Discontinuation visits.

7.2.4.1. Montgomery-Asberg Depression Rating Scale

Efficacy will be assessed using the MADRS – Structured Interview Guide Version (SIGMA) (Appendix A). The MADRS is a 10-item depression rating scale. The MADRS will be completed at study visits as specified in the SOE (Table 2). In order to qualify for the study, subjects must have a MADRS total score ≥ 27 at both Screening and Baseline.

7.2.4.2. Clinical Global Impression – Severity

The CGI-S is completed by the Investigator and measures the severity of illness (Appendix B). The CGI-S is used to rate subjects on a 7-point scale ranging from “Normal” to “Among the most extremely ill patients.”.

7.2.4.3. Hamilton-Depression Rating Scale (17-item)

The Hamilton-Depression Rating Scale is a 17-item depression rating scale (Appendix C). The HAM-D-17 – Structured Interview Guide Version (SIGH-D) will be completed at study visits as specified in the SOE (Table 2).

7.2.4.4. Sheehan Disability Scale

The SDS is a brief, subject-completed questionnaire that assesses functional impairment in work/school, social life, and family life/home responsibilities (Appendix D).

7.2.4.5. Quick Inventory of Depressive Symptomology – Self Report

The QIDS-SR is a subject-completed 16-item depression rating scale (Appendix E).

7.2.4.6. Quality of Life Enjoyment and Satisfaction Questionnaire – Short Form

The Q-LES-Q-SF is a subject-completed 16-item questionnaire that assesses life satisfaction over the past week (Appendix F). Each question is rated on a 5-point scale from 1 (Very Poor) to 5 (Very Good).

7.2.5. Assessment of Safety

Each subject will be closely monitored for safety throughout their study participation. Study-specific safety assessments will include the following:

- Physical examinations
- Vital signs (blood pressure, pulse rate, and oral body temperature)
- Body weight
- 12-lead ECGs
- Clinical safety laboratory tests (serum chemistry [including thyroid function], hematology, coagulation, and urinalysis tests)
- C-SSRS
- AEs

The schedule of these safety assessments is provided in the SOE (Table 2), and the study-specific procedures for completion of these assessments are detailed below.

7.2.5.1. Physical Examination

Physical examinations will be performed as specified in the SOE (Table 2).

Physical examinations will include a review of the following body systems: general appearance, skin and extremities, ears/eyes/nose/throat, head/neck, heart/chest, abdomen, musculoskeletal, and neurological.

Clinically significant findings on the Screening and Baseline physical examination will be evaluated to confirm the subject remains eligible for study participation. New clinically significant abnormalities or exacerbations that occur after the Screening visit will be captured as AEs.

7.2.5.2. Vital Signs, Height, and Weight Measurements

Vital sign measurements will be taken at the time points specified in the SOE (Table 2). Height will be measured (in centimeters) at Screen only; weight (in kilograms) will be measured at Screen and all other scheduled time points indicated in the SOE (Table 2).

Vital sign measurements will include systolic and diastolic blood pressure, pulse rate, and oral body temperature.

At each timepoint, vital signs will be assessed with the subject in the sitting position after approximately three minutes or more of quiet sitting.

Abnormal vital signs may be repeated twice for confirmatory purposes. Confirmed abnormal vital signs will be further categorized as clinically significant or not clinically significant by the Investigator based on his/her clinical judgement.

Subjects with confirmed clinically significant abnormal vital signs at the Screening or Baseline Visits should be excluded from the study. New clinically significant abnormalities or exacerbations that occur after the Screening Visit will be captured as AEs.

7.2.5.3. 12-lead Electrocardiograms

A 12-lead ECG will be obtained at the study site during the time points specified in the SOE (Table 2) using an ECG machine provided by the central ECG monitoring vendor. The central ECG vendor will supply the ECG quantitative and qualitative data. The ECG will be reviewed by medically qualified site personnel. The Investigator will review the ECG report for abnormalities and/or clinically significant changes from previous readings.

ECGs will be performed only after the subject is positioned supine, resting, and quiet for a minimum of three minutes.

Subjects with clinically significant abnormal ECG findings at the Screening or Baseline Visits should be excluded from the study as specified in the study inclusion/exclusion criteria (Section 5.2 and Section 5.3). Criteria for reporting abnormal ECG findings as AEs are provided in Section 8.1.1.

ECGs may be repeated once at both Screening and Baseline for confirmatory purposes.

The following quantitative ECG data will be recorded: heart rate (bpm), PR interval (msec), QRS duration (msec), QT interval (msec), and QTcF interval (msec).

The following qualitative ECG data will be documented: Overall interpretation (normal or abnormal) and if abnormal, comment describing the abnormality. Abnormal ECG results will be further categorized as clinically significant or not clinically significant by the Investigator.

7.2.5.4. Pregnancy Test

A serum pregnancy test for all females will be performed as indicated in the SOE (Table 2). A serum pregnancy test is required at the Screening and Week 4 / Premature Discontinuation Visit. A urine pregnancy test is to be performed at Day -1 as indicated in the SOE. If the urine pregnancy test at Day -1 is positive, a confirmatory serum pregnancy test will be conducted. If pregnancy is confirmed, the subject will be excluded from the study. If the urine pregnancy test is positive, randomization should be postponed until serum pregnancy test results can be

obtained and pregnancy definitively ruled-out. Any occurrence of pregnancy exposure (i.e., pregnancy in a female subject or the female partner of a male subject who has received study drug) must be reported to the Sponsor as summarized in Section 8.1.6.

7.2.5.5. Clinical Safety Laboratory Assessments

Blood samples for clinical safety laboratory tests will be collected as indicated in the SOE (Table 2) and analyzed at an accredited central laboratory approved by the Sponsor. The Clinical Laboratory Manual should be referenced for specific procedures with regards to sample collection, handling, labeling, storage, and shipping.

Clinical safety laboratory tests will include the following laboratory test panels: serum chemistry (including thyroid function), hematology, coagulation, and urinalysis.

A summary of the analytes included in each test panel is provided in Table 5.

Any value outside of the current reference ranges for the laboratory performing the test will be flagged on the laboratory results report.

Laboratory tests showing abnormal or exclusionary values at Screening may be repeated no more than once. After dose administration, abnormal laboratory test results may be repeated as often as deemed clinically necessary by the Investigator until the test values return to clinically acceptable limits or until an explanation other than drug effect is given.

Laboratory test results will be assessed in accordance with the reference ranges provided by the clinical laboratory. Abnormal laboratory results will be further categorized as clinically significant or not clinically significant by the Investigator. Subjects with clinically significant abnormal lab results at the Screening Visit should be excluded from the study. If subjects present with clinically significant lab results collected at the Baseline Visit, continued subject participation should be discussed with the Medical Monitor. New clinically significant abnormalities or exacerbations that occur after the Screening Visit will be captured as AEs.

An estimation of the total blood collection volume per subject is provided in Section 7.2.5.6 and Table 6.

7.2.5.6. Blood Volume Collection Summary

An estimation of the total blood collection volume per subject required for the Screening, Baseline, Treatment Period, and Follow-up Period, is provided in Table 6.

- A total of approximately 139.0 mL (about ½ of a cup) of blood is estimated to be drawn from subjects in the study over a period of about 70 days (depending on the length of the Screening period).

Table 5: Clinical Safety Laboratory Analytes

Thyroid Function	Fasting Serum Chemistry[1]		Hematology	Urinalysis[2]	Urine Drug Test
TSH T3 – Screen only T4 – Screen only	Albumin Alkaline Phosphatase ALT AST BUN Calcium Chloride CO_2 Creatinine eGFR[3] GGT Glucose LDH Magnesium Phosphorus	Potassium Sodium Total Bilirubin; if elevated obtain Direct Bilirubin Total Protein Uric Acid Total Cholesterol HDL Cholesterol LDL Cholesterol Triglycerides Creatine Phosphokinase Serum βhCG (all females)[4]	RBC (Erythrocyte) Count Hemoglobin Hematocrit Platelet Count WBC Count with differential (Absolute and %: Basophils, Eosinophils, Lymphocytes, Monocytes, and Neutrophils) MCV MCH MCHC RDW MPV Reticulocyte Count (Absolute and %)	pH Blood/ RBCs Protein Glucose Ketones Bilirubin Color Leukocytes or WBCs Nitrite Specific Gravity Urobilinogen Microscopic Exam Urine βhCG (all females)[5]	Amphetamines Barbiturates Cannabinoids Benzodiazepines Methadone Opiates Cocaine Phencyclidine
Coagulation					
PT PTT INR					

[1] All protocol scheduled Serum Chemistry samples should be collected with the subject in a fasting state (i.e., no food for a minimum of eight hours prior to sample collection).
[2] If urine is positive for blood, leukocyte esterase, nitrite, or protein, then will default to reflex microtesting.
[3] Cockcroft-Gault Method of calculation – subject weight is required for calculation
[4] Serum pregnancy test at Screen and Week 4/Premature D/C
[5] Urine pregnancy test at the Baseline Visit

ALT=alanine aminotransferase; AST=aspartate aminotransferase; BUN=blood urea nitrogen; CBC=complete blood count; CO2=carbon dioxide or bicarbonate; eGFR=estimated glomerular filtration rate; GGT=gamma glutamyl transferase; hCG= human chorionic gonadotropin; HDL=high density lipoprotein; LDH=lactate dehydrogenase; LDL=low density lipoprotein; MCH=mean corpuscular hemoglobin; MCHC=MCH Concentration; MCV=mean cell volume; MPV=mean platelet volume; Premature D/C=Premature Discontinuation from treatment; PT=prothrombin time; PTT=partial thromboplastin time; RBC=red blood cell; RDW=RBC distribution width; TSH=thyroid stimulating hormone; WBC=white blood cell

Table 6: Estimation of Blood Volumes Required from Screening through the End of the Follow-up Period

Tests	Estimated Blood Draw Volume/ Sample (mL)	Study Day								Estimated Total Blood Draw Volume (mL)
		Screen (Day -28 to -2)	Day -1 (Pre-Dose)	Week 1	Week 2	Week 3	Week 4/ Premature D/C	Week 5	Week 6	
Serum Chemistry Serum Pregnancy (females only)	7.5	7.5	7.5		7.5		7.5	7.5		**37.5**
TSH/T3/T4	5.0	5.0	5.0		5.0		5.0	5.0		**25.0**
Hematology	3.0	3.0	3.0		3.0		3.0	3.0		**15.0**
Coagulation	2.7	2.7	2.7		2.7		2.7	2.7		**13.5**
PK	6.0		6.0	6.0	6.0		6.0			**24.0**
[redacted]	[redacted]		[redacted]		[redacted]		[redacted]	[redacted]		[redacted]
Estimated Total mL of Blood Drawn:		**18.2**	**30.2**	**6.0**	**30.2**	**0.0**	**30.2**	**24.2**	**0.0**	**139.0 mL**

Abbreviations: PK=Pharmacokinetics; Premature D/C=Premature Discontinuation from treatment

7.2.5.7. Columbia-Suicide Severity Rating Scale

The C-SSRS (Appendix G and Appendix H) will be administered by a trained site staff member during the Screening Period (Baseline/Screening Version) and the Treatment and Follow-up Periods (Since Last Visit Version) as specified in the SOE (Table 2). Subjects who pose a current or future suicidal risk in the Investigator's opinion, or have a history of suicidal behavior within the past 12 months, and/or answers "yes" to "Suicidal Ideation" Item 4 (active suicidal ideation with some intent to act, without specific plan) or Item 5 (active suicidal ideation with specific plan and intent) on the C-SSRS assessment during the six months prior to the Screening period and/or at Baseline will not be eligible for study enrollment.

Subjects who at any time during this study spontaneously report AEs of suicidal ideation or suicidal behavior, either as an outpatient or during visit interviews, must be assessed by the PI or designee and referred for further mental health evaluation as clinically indicated.

7.2.6. Assessment of Pharmacokinetics [redacted]

7.2.6.1. Pharmacokinetics

The population PK of SP-624 will be assessed.

A total of four blood samples (6 mL) will be collected from each subject for the measurement of plasma concentrations of SP-624 at the following visits:

- Visit 2 (Day -1)
- Visit 3 (Week 1): Pre-Dose
- Visit 4 (Week 2): Pre-Dose or approximately 2 to 10 hours post-dose (whichever is more convenient for the subject)
- Visit 6 (Week 4): approximately 2 to 10 hours post-dose

If a subject missed the dose the day before a scheduled "pre-dose" sample collection, study personnel should instead collect a post-dose sample at that visit. Should a subject inadvertently dose prior to a scheduled "pre-dose" sample collection at Week 1 or Week 2, study personnel should collect a post-dose sample.

In the event that samples cannot be conveniently collected for a given subject according to the scheduled sampling times specified above, study personnel should attempt to collect at least one pre-dose sample and at least one post-dose sample during the course of the study.

Plasma samples will be analyzed for SP-624 concentration using a validated bioanalytical method.

An aliquot of each subject's post-dose samples may be pooled by the bioanalytical laboratory for metabolite profiling. Potential metabolites will be identified using mass spectrometry techniques; and if conducted, these data will be summarized in a separate bioanalytical report.

Labels on blood sample collection tubes and vials will include the protocol code number, subject identification code number, and visit number.

Samples will be stored at the study center until shipment under appropriate conditions to the Sponsor-designated bioanalytical laboratory for analysis. The bioanalytical lab will analyze the samples for plasma concentrations of SP-624 using a validated LC-MS method.

Detailed instructions for all blood sampling, processing, storage, and shipment procedures will be provided in a separate study guidance document.

An estimation of the total blood collection volume per subject is provided in Section 7.2.5.6 and Table 6.

7.2.6.2. [redacted]

[redacted]

7.2.7. [redacted]

[redacted]

8. ADVERSE EVENTS, SERIOUS ADVERSE EVENTS, AND OTHER REPORTABLE EVENTS

8.1. Adverse Events

An AE is any untoward medical occurrence in a clinical study subject, whether or not the event is considered causally related to the study drug. An AE can be a new occurrence or an existing process that increases significantly in intensity or frequency.

All AEs must be recorded in the appropriate section of the CRF.

Adverse event monitoring for each subject begins upon Screening and continues until study completion. Abnormalities present at Screening are considered AEs only if they reoccur after resolution or worsen in intensity or frequency during the study.

Each AE is to be classified as serious or non-serious, and categorized with regard to relationship to study drug, and intensity.

8.1.1. Clinical Safety Assessment Abnormalities

Abnormal findings on clinical safety assessments, including clinical safety laboratory test results, vital sign measurements, ECGs, and physical examinations, will be assessed by the Investigator as clinically significant or not clinically significant. Clinically significant abnormalities that are new or exacerbate after the Screening Visit will be reported as AEs. Classification of seriousness, relationship to study drug, and intensity should use the guidelines provided in Section 8.1.2, Section 8.1.4.3 and Section 8.1.4.4, respectively.

8.1.2. Serious Adverse Event (SAE)

An AE is serious, regardless of whether or not it is considered related to the study drug, if the event meets one or more of the following criteria:

- Results in death.
- Is life threatening; per the Investigator, the subject was at immediate risk of death from the event at the time that the event occurred. It does not include an event that hypothetically might have caused death if it had occurred in a more severe form.
- Requires in-patient hospitalization; initial or prolonged, meaning that a hospital admission and/or prolongation of a hospital stay was required for the treatment of the AE, or occurred as a consequence of the event. It does not include a pre-planned elective hospital admission for treatment or diagnostic procedures.
- Results in a persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions.
- Congenital anomaly or birth defect that occurs in the offspring of a subject exposed to the study drug.
- Important medical event that, although does not result in one of the other serious outcomes, requires medical or surgical intervention in order to prevent one of the other serious outcomes listed above. Examples of such medical events include

allergic bronchospasm requiring intensive treatment in the emergency room or at home, blood dyscrasias, or convulsions that do not result in inpatient hospitalization, or the development of drug dependency or drug abuse.

All SAEs that occur after any subject has been enrolled, before treatment, during treatment, or within 28 days following the Week 4 Visit, whether or not they are related to the study, must be recorded on forms provided by Sirtsei Pharmaceuticals.

8.1.3. Suspected and Unexpected Serious Adverse Reactions

A suspected and unexpected serious adverse reaction (SUSAR) is defined as an SAE that meets both of the following criteria with respect to study drug:

- **Suspected:** is assessed as probably or possibly related to study drug (see Section 8.1.4.3)
- **Unexpected:** in comparison to drug-related AEs described in the Investigator's Brochure, the event meets any of the following criteria:
  - The event was not previously described; or is now characterized more specifically (e.g., an event of "interstitial nephritis" in a subject receiving an agent previously described as associated with "acute renal failure"); and/or
  - The event is now characterized as more severe (see Section 8.1.4.4).

8.1.4. Evaluation of Adverse Events

Adverse events spontaneously reported by the subject and/or in response to an open question from the study personnel (i.e., how are you feeling?) or revealed by observation will be recorded during the study at the investigational site.

All AEs/SAEs will be recorded in the CRF and will include a concise AE term/diagnosis, date and time of event onset and resolution, relationship to study drug, intensity, action taken, and outcome as described in Section 8.1.4.1 through Section 8.1.4.6.

Abnormalities in vital signs, laboratory results, and other safety assessments will be recorded as an AE if they meet the definition of an AE (Section 8.1.1).

Events that are considered symptoms of MDD should not be recorded as medical history or AEs.

Lack of efficacy should not be considered an AE.

8.1.4.1. Adverse Event Term/Diagnosis

The diagnosis or description of the AE will be as specific and complete as possible (i.e., "lower extremity edema," rather than just "edema").

Whenever possible, signs, symptoms, and laboratory abnormalities due to a common etiology will be reported as an integrated diagnosis; for example, cough, runny nose, sneezing, sore throat, and head congestion would be reported as "upper respiratory infection." The AE term should be reported in standard medical terminology when possible.

A medical or surgical procedure is not an AE; rather the condition leading to the procedure should be recorded as the AE. If the condition is not known, the procedure may be reported as

an AE instead. Similarly, death is not an AE, but is rather the outcome of the AE(s) that resulted in death. If the AE(s) leading to death are not known, then death will be reported as an AE.

8.1.4.2. Date and Time of Adverse Event Onset and Resolution

The time, if available, and date at which the AE was first apparent will be recorded as the event onset. The date and time of onset of symptoms may be appreciably earlier than the date and time the Investigator becomes aware of the event. Some events may be apparent to the subject and Investigator independently, and information from each may contribute to the final report. The date and time of resolution of the event will be recorded when they are available.

8.1.4.3. Relationship to Study Drug

A causality assessment (i.e., assessment of relationship to study drug) must be provided for all AEs. This assessment must be made by a physician Investigator and recorded on the CRF. The definitions for the causality assessment are provided below.

- **Probably Related:** an event that (1) follows a reasonable temporal sequence from administration of the drug, (2) follows a known or expected response pattern to the drug, (3) is improved/resolved by stopping or reducing the dosage of the drug, and/or (4) could not be reasonably explained by the known characteristics of the subject's clinical state.
- **Possibly Related:** an event that follows a reasonable temporal sequence from administration of the drug and where a causal relationship between drug administration and event is a reasonable possibility, even if the event could readily have been produced by a number of other factors. The expression "reasonable possibility" is meant to convey in general that there are facts (evidence) or arguments to suggest a causal relationship.
- **Not Related:** an event that is unlikely to have any relationship to the study drug even if such relationship cannot be definitely ruled out.

If the relationship between the AE/SAE and the study drug is determined to be "possible" or "probable" the event will be classified as "related" to the study drug for the purposes of expedited regulatory reporting.

8.1.4.4. Intensity Categorization

The intensity of all AEs will be assessed by the Investigator using the following criteria:

- **Mild:** An AE that is transient, requires minimal or no treatment, and does not interfere with the subject's daily activities.
- **Moderate:** An AE that is usually alleviated with specific therapeutic intervention. The event causes some interference with usual activities of daily living, causing discomfort, but poses no significant or permanent risk of harm to the research participant.
- **Severe:** An AE that is incapacitating, prevents usual activities of daily living, or significantly affects clinical status, or may require intensive therapeutic intervention.

It is important to distinguish between serious and severe AEs. Intensity is a measure of severity whereas seriousness is defined by the criteria under Section 8.1.2. An AE of severe intensity may not be considered serious (e.g., a headache can be severe but not serious – e.g., is not life threatening, or does not result in hospitalization).

8.1.4.5. Action Taken for Adverse Event

Adverse events will be followed and managed by the Investigator, including obtaining any supplemental tests or assessments needed to define the nature and/or cause of the event (e.g., laboratory tests, diagnostic procedures, consultation with other health care professionals, etc.).

For each AE, the Investigator will categorize one or more actions taken to manage the AE as follows:

- **Discontinuation:** Scheduled study procedures and/or study drug dosing (if applicable) were stopped due to the AE.
- **Study drug dose reduction:** The study drug dose is decreased from two capsules per day to one capsule per day.
- **Concomitant medication:** One or more medications (prescription or over the counter) were started or increased in dosage.
- **Study drug dose taken with food**: The study drug dose is taken ≤ 2 hours following a meal or a meal is eaten < 1 hour after taking a dose.
- **Other action:** Non-medication action(s) were ordered as management of the AE (e.g., bed placed in Trendelenburg position, warm compresses applied to intravenous access site, etc.).
- **No action:** No actions were ordered for management of the AE.

8.1.4.6. Outcome of Adverse Event

If possible, AEs will be followed until resolved (i.e., recovered, recuperated, or ended) either with or without sequelae, or the Investigator and Sponsor Medical Monitor assess them as chronic or stable, and no longer requiring follow-up.

The outcome of each event (including time if available, and date) will be described using the following categories:

- **Recovered:** The event resolved, and subject returned to Baseline.
- **Recovered with Sequelae:** The event resolved but the subject is left with residual problems (e.g., functional deficits, pain).
- **Not Recovered:** At the last observation, the event was ongoing.
- **Death (Fatal):** To be used for the one AE which, in the judgment of the Investigator, was the primary cause of death.
- **Unknown:** There were no observations after the onset (initial observation or report) of the event, or subject is lost-to-follow-up and outcome of AE is unknown.

A subject who is discontinued from study drug because of an AE must be followed by the Investigator until clinical recovery is complete and laboratory results have returned to normal, or

until progression has been stabilized. Follow-up may need to continue after the subject has discontinued from the study, and additional investigations may be requested by the Investigator.

8.1.5. Reporting of Serious Adverse Events

The Investigator will review each SAE and evaluate the severity and the causal relationship of the event to study drug. Serious AEs that occur after the last study visit and come to the attention of the Investigator must be reported only if there is (in the opinion of the Investigator) reasonable causal relationship with the study drug. Serious AE reporting contact information is provided in Table 1.

The Investigator is responsible for providing notification to the Sponsor (Table 1) of any SAE, within 24 hours of when he or she became aware of the event, whether deemed study drug related or not.

An SAE report form template will be provided to the Investigator by the Sponsor (or designee) prior to study initiation, with the preferred format for reporting SAEs.

As a minimum requirement, the initial notification should include the following information:

- Study number
- Subject number
- Name of Investigator
- AE term
- Criterion for classification as 'serious'
- Study drug name
- Causality assessment (if sufficient information is available to make this classification)
- Identifiable reporter of event

If clarification is requested by the Sponsor for omitted or discrepant information in the initial notification, the Investigator or an authorized delegate is responsible for sending the requested information within 24 hours.

Initial reports of SAEs must be followed later with detailed descriptions, including clear copies of other documents as necessary (e.g., hospital reports, consultant reports, autopsy reports), with the study subject's personal identifiers removed. Additional information on the SAE may be requested, which the Investigator or an authorized delegate must send to the SAE reporting contact (Table 1).

The Sponsor is responsible for expedited reporting of qualifying events to relevant regulatory authorities. It is the Investigator's responsibility to notify the IRB of all SAEs that occur at his or her site. Investigators will also be notified by the Sponsor of all unexpected, serious, drug-related events [Investigational New Drug (IND) safety reports; also known as SUSARs] that occur during the clinical trial from any other studies or study sites. The Investigator is responsible for notifying its IRB of these events.

8.1.6. Other Reportable Events

Certain events that occur in the absence of an AE should be reported to the Sponsor. These include the following events:

- Pregnancy Exposure (subject or subject's partner becomes pregnant at any time during study participation and through 30 days post-dose).
  - Should a female subject or partner of a male subject become pregnant during the study, the subject will inform the Investigator.
  - The subject will be asked to follow up with the study site to report the eventual outcome of the pregnancy. The information will be tracked by the Sponsor.
  - The outcome of all pregnancies (spontaneous miscarriage, elective termination, normal birth or congenital abnormality) must be followed up and documented even if the subject was discontinued from the study.
  - Pregnancy in itself is not regarded as an AE.
  - All reports of congenital abnormalities/birth defects are SAEs. Spontaneous miscarriages should also be reported and handled as SAEs. Elective abortions without complications should not be handled as AEs.
- Lactation Exposure (subject was taking study drug while nursing an infant).
- Accidental Exposure (someone other than the study subject was exposed to study drug).
- Overdose of Study Drug (defined as dosing > 4 capsules in a 24-hour period) as a result of a dosing amount error should be reported. Any adverse outcomes of the overdose should be recorded as AEs/SAEs according to the procedures described in Section 8.1 through Section 8.1.5 . Intentional overdose of study drug should be evaluated as a potential AE/SAE and reported accordingly.
- Other medication errors that potentially place subjects at greater risk of harm than was previously known or recognized (i.e., study drug was administered via an incorrect route).

9. STATISTICS

Following are proposed statistical analyses. A separate Statistical Analysis Plan (SAP) will be generated and finalized prior to analysis. The SAP will specify the details of all statistical analyses, including all efficacy and safety analyses to be performed.

9.1. Determination of Sample Size

Recent studies of MDD using change from Baseline in MADRS total scores suggest that placebo will yield an approximately nine unit decrease over four weeks with a SD of around 9.2. SP-624 is expected to yield at least a 12 unit decrease in MADRS total score over four weeks with a similar SD of around 9.2. A minimum of 150 subjects per treatment arm (300 total) would be needed to detect a treatment difference of three units with 80% statistical power. The sample size was estimated using NQuery 8 (V8.5.2.0) two group test of equal means (MTT0-1: www.statsols.com).

9.2. Analysis Populations

Within each population, analyses will be performed comparing subjects and by-treatment assignment (SP-624 vs placebo).

- All Subjects Population – all subjects who sign informed consent and are assigned a unique subject identification number. This population will be used to summarize disposition.
- Modified Intent-to-Treat (mITT) Population – all subjects who are randomized to receive treatment, receive at least one dose of study drug, and have a Baseline and at least one post-dose efficacy assessment. This population will be the primary population used to summarize demographics, baseline characteristics, medical history, treatment history, and all efficacy outcomes. Subjects will be analyzed according to randomized treatment group.
- Per Protocol (PP) Population – All subjects in the mITT population who received assigned study drug as randomized, completed the Week 4 assessments without any major protocol violations, and have 75-125% compliance. The PP population will be used as an additional population for summaries of efficacy outcomes.
- Safety Population — all subjects in the mITT population who receive at least one dose of study drug. Safety subjects will be summarized according to the treatment they actually received, in case they received a different treatment than assigned at randomization. This population will be used to summarize all safety assessments.
- PK Population — all subjects who receive active study drug and have evaluable PK data based on actual treatment received, protocol compliance, adequate numbers of samples, and successful sample assays.

9.3. Description of Statistical Methods

9.3.1. General Approach

All data will be provided in listing files. Study results and information will be displayed in descriptive summary tables. Inferential tests are identified below. For measurements that have multiple intermediate visits, results for observed data will be summarized for each time.

When comparisons between groups are performed, two-sided tests at a 0.05 level of significance (α=0.05) will be used. The null hypothesis for all analyses is that there is no difference between the SP-624 and placebo treatment groups. There is only one primary outcome for analysis, and there will be no adjustment for multiplicity for secondary endpoints.

All summaries, statistical analyses, and individual subject data listings described below will be completed using Version 9.4 or later of the SAS Statistical Analysis System (SAS Institute, Inc. Cary, NC).

Subjects who are randomized but not treated and those who are treated but have no post-treatment follow-up assessments or data will be listed, but not analyzed.

9.3.2. Interim Analysis

No formal interim analyses are planned for this study.

9.3.3. Summary of Demographics, Baseline Characteristics, Medical History, and Treatment History

Listings and summaries of demographics, baseline characteristics, medical history, and treatment history will be provided for the mITT and safety populations. Categorical variables will be assessed for homogeneity across the treatment arms using a Fisher's exact test and numeric variables will be compared using a Wilcoxon rank-sum test. If there are any ordered categorical variables, the homogeneity of the treatment arms will be evaluated using a CMH row-mean score test calculated with modified ridit ranks. This summary will be provided for the mITT population.

9.3.4. Summary of Efficacy

Efficacy analyses will be conducted using the mITT population.

9.3.4.1. Primary Efficacy Endpoint Analysis

The MADRS is a 10-item scale whose total score can range from 0 to 60. The primary efficacy variable is change from Baseline in MADRS total score at Week 4, for testing superiority of SP-624 vs. placebo in adults with major depressive disorder. The analysis will be completed using observed data without imputation. Scores will be taken at 4 weeks regardless of subject discontinuation from treatment. Subjects will not be treated with any other antidepressant medication until at least the completion of the Week 5 visit.

The change from Baseline in MADRS total score at Week 4 will be analyzed from the difference in least square mean change using MMRM, with change from Baseline in MADRS total score at each visit as the response variable. The MMRM model will include age, weight, and baseline

MADRS total score as covariates, with factors for treatment, visit (Weeks 1, 2, 3, and 4; as a categorical variable), treatment by visit interaction term, gender, and site as a fixed effect. An unstructured covariance matrix will be used for the within-subject correlation. Kenward-Rogers approximation will be used to calculate the denominator degrees of freedom. The primary efficacy endpoint will be calculated using an estimate statement. Subjects from low enrolling sites may be pooled in order to help with model convergence.

In the event of a model which fails to converge, additional strategies to address convergence may be required such as alternative covariance structures, using a maximum likelihood versus restricted maximum likelihood (REML) estimation, or dropping variables.

The protocol may include approximately 40 sites and sites with less than 10 subjects will be pooled to obtain at least 10 subjects. Sites would be pooled in order of the site number and, if necessary, the last un-pooled site will be pooled with the last set of sites pooled.

Additional variables of interest that may be evaluated as covariates will be identified in the SAP, such as age of onset of depression and number of lifetime episodes.

Additionally, results from the primary outcome will be summarized for the PP population.

9.3.4.2. Secondary Efficacy Endpoint Analysis

Seven secondary efficacy endpoints will be analyzed using both the mITT and PP populations.

These include:

- Change from Baseline to Weeks 1, 2, and 3 in MADRS total score for SP-624 vs. placebo. The outcome will be analyzed using the same methods as the primary outcome.
- CGI-S is measured on a 7-point scale where 1 represents “normal” and 7 represents “most extremely ill”. A summary of percentage of subjects with each score at Baseline and at Weeks 1, 2, 3, and 4 will be presented. The change from Baseline in CGI-S can take on interval values between -6 (improvement from 7 to 1) and 6 (worsening from 1 to 7). Treatment comparison of change from Baseline to Weeks 1, 2, 3, and 4 in CGI-S scores will be performed using an MMRM model similar to that of the primary efficacy variable.
- Change from Baseline to Week 5 and change from Week 4 to Week 5 will be assessed using descriptive statistics for the MADRS and CGI-S.
- The HAM-D-17 is a 17-item scale whose total score can range from 0 to 52. Change from Baseline to Week 2 and Week 4 in the 17-item Hamilton Depression Rating Scale (HAM-D-17) total score will be assessed for treatment differences using an MMRM model similar to that of the primary efficacy variable.
- The Sheehan Disability Scale (SDS) is a 3-part scale that measures the degree of disruption on work, social and family life using an 11-point scale where 0 represents “no disruption” and 10 represents “extreme disruption”. In addition to the 11-point scale, subjects are asked to indicate the number of days in the past week that were “lost” and numbers of days that were “unproductive”. The results of these questions have a range from 0 to 7. A total global functioning impairment score can be utilized by summing the scores from work, social and family life scales for a value range from 0 to 30. Treatment

difference in change from Baseline to Week 2 and Week 4 for both global functioning impairment and days lost/unproductive will be assessed using an MMRM model similar to that of the primary efficacy variable. Change in days lost will be analyzed separately from change in unproductive days.

- The Quick Inventory of Depressive Symptomology – Self Report (QIDS-SR), is a 16-item self-reported scale where each item has a 4-point scale where 0 represents least impact scores while 3 represents greatest impact scores. Scale 6 and 7 are linked such that only one of these will be scored. Likewise, scales 8 and 9 are linked such that only one of them will be scored. Nine domains are scored based on the highest score of items within that domain. The total score from all domains can take a value ranging from 0 to 27. Treatment difference in change from Baseline to Week 2 and Week 4 in the QIDS-SR will be assessed using an MMRM model similar to that of the primary efficacy variable.
- The Quality of Life Enjoyment and Satisfaction Questionnaire – Short Form (Q-LES-Q-SF) is a 16-item satisfaction scale where each item has a 5-point scale. A score of 1 represents "very poor satisfaction", while a score of 5 represents "very good satisfaction". Only the first 14 items are summed for a total score that ranges from 14 to 70 and is expressed as a percentage based on the maximum total score of the items completed (0–100). Treatment difference in the change from Baseline to Week 2 and Week 4 in total scores will be assessed using an MMRM model similar to that of the primary efficacy variable. The last two items pertaining to satisfaction with medications and overall contentment will be summarized separately by treatment group.

9.3.5. Sensitivity Analyses

A dropout rate of 20-30% is typically seen in MDD studies. The mechanisms that cause missing data may be missing at random (MAR) or may be missing not at random (MNAR). The MMRM model (especially as implemented within the PROC MIXED procedure in SAS) used in the primary analysis of the primary efficacy endpoint relies on the assumption of MAR. Hence, a sensitivity analysis for the primary endpoint and secondary endpoints will be performed using a pattern-mixture modeling approach (Kenward, et al 2003).

Additionally, subjects will be grouped by the visit at which they had their last MADRS total score measured. This will result in five categories of discontinuation: Week 1 dropouts, Week 2 dropouts, Week 3 dropouts, Week 4 dropouts, and completers. Mean change from Baseline in MADRS total score will be plotted by the dropout category and by reason for discontinuation, in order to assess whether this efficacy measure appears to be correlated with study dropout.

If missing data are greater than 25% or the pattern-mixture model provides a different interpretation of the treatment comparison for the primary endpoint, a multiple imputation analysis may also be performed using a fully conditional specification (FCS) approach using variables identified in the SAP.

9.3.6. Summary of Pharmacokinetics

Plasma concentration data for SP-624 will be listed and summarized using descriptive statistics.

If data allow, results will be used for other analyses (e.g., PopPK) which will be presented in a separate report.

The population PK analysis of SP-624 will be performed using a nonlinear mixed effects model (NONMEM). This analysis will provide estimates of population PK parameters such as CL/F, Vz/F, etc. and a measure of their variability. Pharmacokinetic parameters of individual subjects (e.g., AUC[0-tau], Cavg, Cmax) may be estimated and their correlation with relevant covariates (e.g., body weight, gender, concomitant medications) will be explored. The population PK analysis plan will be prepared separately.

Plasma concentration data will remain blinded until the unblinding of the clinical database at the end of the study.

9.3.7. [redacted]

[redacted]

9.3.8. Concomitant Medications

Concomitant medications will be coded using the most current version of the World Health Organization (WHO) drug dictionary available.

Any medications taken during the course of the study with a start date on or after the date of the first dose of study drug and on or before the date of the last dose of study drug, or with a start date prior to, and an end date on or after, the date of the first dose of study drug, or marked as ongoing, will be considered concomitant medications. Medications that ended prior to the date of the first dose of study drug will be considered prior medications. Medications that started after the date of the last dose of study drug will not be considered concomitant but will be considered post-treatment. Prior and concomitant medications will be summarized for the number and percentage of subjects using each medication by treatment and by the drug class and preferred name for the safety population.

9.3.9. Summary of Safety and Tolerability

All summaries of safety and tolerability will be performed on the safety population.

Laboratory parameters, vital sign data, and ECG results will be summarized by treatment group and presented in tabular and graphic formats where appropriate. Appropriate change from Baseline calculations will be provided to detect any significant changes in laboratory parameters, vital sign values, ECG results, and C-SSRS results, but no formal statistical tests are planned.

9.3.9.1. Adverse events

Summaries of the number and percentage of subjects in the safety population with at least one treatment-emergent AE (TEAE), classified according to preferred term and/or body system using the MedDRA dictionary, will be provided overall and by treatment group for:

- Overall summary
- All TEAEs
- TEAEs by relationship (not related, possibly related, probably related)

- TEAEs by severity (mild, moderate, severe)
- SAEs
- TEAEs leading to withdrawal from the study
- TEAEs leading to study drug dose reduction
- TEAEs leading to study drug dose taken with food
- Analyses of AEs with time of onset beginning 24 hours after the final dose through up to 28 days after the final dose.

No formal statistical tests are planned for AE data.

9.3.9.2. Vital Signs and ECGs

Vital signs and ECG results will be summarized by treatment group and presented in tabular and graphic formats where appropriate. Appropriate change from Baseline calculations will be provided to detect any significant changes in vital sign values or ECG results, but no formal statistical tests are planned. Any abnormal finding or clinically significant changes will be captured as AEs.

9.3.9.3. Clinical Laboratory Assessments

Clinical laboratory test results for chemistry (including thyroid function), hematology, coagulation, and urinalysis will be summarized by treatment group and presented in tabular and graphic formats where appropriate. Appropriate change from Baseline calculations will be provided to detect any significant changes in laboratory parameters, but no formal statistical tests are planned. Any abnormal finding or clinically significant changes will be captured as AEs.

9.3.9.4. Physical Exams

Physical examination findings will be listed for each subject at each assessment period. Clinically significant new findings and adverse changes in physical examination findings will be captured as AEs, therefore eliminating the need to summarize over time.

9.3.9.5. C-SSRS

Frequency and severity of suicidal ideation and suicidal behavior as measured by the C-SSRS will be summarized by treatment for each visit.

10. SPONSOR'S AND INVESTIGATOR'S RESPONSIBILITIES

10.1. Sponsor's Responsibilities

- The Sponsor and its representatives will undertake the management of the study in compliance with all applicable national and local regulations, as well as the global standard International Council for Harmonisation (ICH) GCP E6 Guideline.
- The Sponsor will ensure that all local regulatory authority requirements of the Sponsor are met before the start of the study including receipt of regulatory authority approvals and/or compliance with the appropriate wait-time required prior to release of the study drug for shipment to the site
- Sponsor ensures that suitable clinical study insurance coverage is in place prior to the start of the study. An insurance certificate will be supplied as needed.
- When required by national law, Sponsor will post appropriate study information on government websites which may include Investigator name and contact information for participating sites.
- Sponsor representatives will make periodic visits to participating sites to review subjects' medical records/source documents, study drug accountability records, and site regulatory documents.

10.2. Investigator's Responsibilities

- The Investigator will conduct the study in compliance with all applicable national and local regulations, as well as the global standard ICH GCP E6 Guideline.
- The Investigator agrees to personally oversee the study, delegate study-related tasks to qualified personnel, and meet with Sponsor representatives during on-site visits.
- The Investigator must permit representatives of Sponsor, the IRB and national, local, and foreign regulatory authorities to have direct access to original source documents/medical records, regardless of media, relevant to the study.
- The Investigator is required to maintain all study records according to the ICH GCP E6 Guideline and may not destroy any records until the mandated record retention period is over. Note: Investigators are encouraged to consult with the Sponsor before record destruction to ensure that all retention requirements have been satisfied.

11. ETHICS

11.1. Ethical Conduct of the Study

The study will be performed in accordance with ethical principles that have their origin in the Declaration of Helsinki and are consistent with ICH GCP and applicable regulatory requirements.

11.2. Ethics Review

The final study protocol, including the final version of the ICF, must be approved in writing by an IRB as appropriate. The Investigator must submit written approval to the Sponsor before he or she can enroll any subject into the study.

The Investigator is responsible for informing the IRB of any amendment to the protocol in accordance with local requirements. In addition, the IRB must approve all advertising used to recruit subjects for the study. The protocol must be re-approved by the IRB upon receipt of amendments and annually, as local regulations require.

The Investigator is also responsible for providing the IRB with reports of any reportable serious adverse drug reactions from any other study conducted with the study drug. The Sponsor will provide this information to the Investigator.

Progress reports and notifications of serious adverse drug reactions will be provided to the IRB according to local regulations and guidelines.

11.3. Written Informed Consent

The Investigator will ensure that the subject is given full and adequate oral and written information about the nature, purpose, possible risk, and benefit of the study. The subject must also be notified that they are free to discontinue from the study at any time. The subject should be given the opportunity to ask questions and allowed time to consider the information provided.

The subject's signed and dated ICF must be obtained before conducting any study procedures.

The Investigator must maintain the original, signed ICF. A copy of the signed ICF must be given to the subject.

12. DATA HANDLING AND RECORD KEEPING

12.1. Data Collection and Recording

The Investigator or personnel designated by the Investigator will perform primary data collection based on original hospital or clinic records or other source documentation. All required study information must be reported on the appropriate CRF. The study monitor will review CRFs for accuracy and completeness to ensure maximum data integrity. In addition, as the person ultimately responsible for the accuracy of all CRF data, the Investigator will provide endorsement that the data on the CRFs are accurate and complete.

Data processed outside the study site at a central laboratory (e.g., safety lab, ECG, bioanalytical data, etc.) may be transferred electronically.

12.2. Retention of Records

The Investigator must maintain all documentation relating to the study for a period of two years after the last marketing application approval, or if not approved, two years following the discontinuance of the study drug for investigation. The Investigator agrees that any study documentation will be promptly disclosed to the Sponsor or the regulatory authority upon request for review.

12.3. Protocol Deviations

A protocol deviation is any non-compliance with the clinical study protocol, including compliance requirements and agreements. Examples include an Investigator's decision to deviate from the protocol, a subject's lack of adherence to the protocol, or external/environmental factors (e.g., severe weather or holidays) that change the conduct of a protocol from that which was planned.

The Sponsor will review the protocol deviations that occur during the study. A determination will be made as to which deviations are major – e.g., deviations that impact the study results, data integrity, subject safety, subject consent, or admission criteria.

Further details about the handling of protocol deviations will be included in the Sponsor Study Monitoring and Data Management Plans. The Investigator is responsible for complying with IRB protocol deviation/violation reporting requirements, as applicable.

12.4. Publication Policy

The publication policy is provided in a separate agreement between the Sponsor and those parties contracted to perform services for this study. In summary, all information received and developed in connection with this protocol is considered confidential by the Sponsor. Such information may only be disclosed as deemed appropriate and necessary by the Sponsor. The publication or presentation of data from this study is not allowed without prior agreement with the Sponsor.

12.5. Data Quality Assurance

Standard operating procedures (SOPs) are available for all activities performed by the investigational site relevant to the quality of this study. Designated personnel of the investigational site will be responsible for implementing and maintaining quality assurance (QA) and quality control systems to ensure that the study is conducted, and that data are generated, documented and reported in compliance with the study protocol, GCP and Good Laboratory Practice (GLP) requirements, as applicable, as well as applicable regulatory requirements and local laws, rules and regulations relating to the conduct of the clinical study.

CRFs will be 100% source data verified by a Sponsor study monitor against the source notes for identification and clarification of any discrepancies.

The Investigator must make study data accessible to the study monitor, to other authorized representatives of the Sponsor, and to the appropriate regulatory authority inspectors. Audits for QA of the database may be performed according to relevant SOPs from the contract research organization (if applicable) or at the request of the Sponsor.

12.6. Data Management

Following are proposed data management activities. A separate Data Management Plan (DMP) will be generated and finalized prior to initiating data management activities. The DMP will specify the details of all data management activities.

Sirtsei Pharmaceuticals, or their designee, will generate the CRFs. An electronic CRF will be used. Sirtsei Pharmaceuticals clinical and safety monitors will review entered data and ensure that they correspond to data kept by the site in source documents. Data management activities such as query management and coding of AE terms will be performed by Sirtsei Pharmaceuticals or their designee.

Each CRF will be reviewed and signed by the PI at each site, as applicable. A detailed DMP will be prepared by the assigned data manager and approved by the Sponsor.

All clinical data will undergo a quality control check prior to clinical database lock. Edit checks are performed for appropriate databases as a validation routine using SAS® or comparable program to check for missing data, data inconsistencies, data ranges, etc. Corrections will be made prior to database lock.

13. LIST OF REFERENCES

13.1. Publications Referenced

American Psychiatric Association. Depressive Disorders. In: Diagnostic and Statistical Manual of Mental Disorders. 5th ed. Washington, DC: American Psychiatric Association; 2013. https://dsm-psychiatryonline-org.ahecproxy.ncahec.net/doi/full/10.1176/appi.books.9780890425596.dsm04

Ionescu DF, Rosenbaum JF, Alpert JE. Pharmacological Approaches to the Challenge of Treatment-Resistant Depression. Dialogues Clin Neurosci. 2015 Jun;17(2):111-26.

Kenward MG, Molenberghs G, Thijs H. Pattern-Mixture Models with Proper Time Dependence. Biometrika, 2003;90(1):53-71.

Lieber A. Major Depression. https://www.psycom.net/depression.central.major.html. Updated January 7, 2020. Accessed July 7, 2020.

Sheehan DV, Lecrubier Y, Harnett-Sheehan K, Amorim P, Janavs J, Weiller E, Hergueta T, Baker R, Dunbar G. The Mini International Neuropsychiatric Interview (M.I.N.I.): The Development and Validation of a Structured Diagnostic Psychiatric Interview. J. Clin Psychiatry, 1998;59(suppl 20): 22-33. http://www.psychiatrist.com/JCP/article/Pages/1998/v59s20/v59s2005.aspx

[redacted]

13.2. Sponsor Documents and Study Reports Referenced

13.2.1. Investigator's Brochure

[SP-624 Investigator's Brochure 2020] Investigator's Brochure for SP-624. 2nd ed. Sirtsei Pharmaceuticals, Inc.; Release date 11 June 2020.

13.2.2. [redacted]

[redacted]

14. APPENDICES

APPENDIX A. MONTGOMERY-ASBERG DEPRESSION RATING SCALE

SIGMA 2011, v. 1.2

STRUCTURED INTERVIEW GUIDE FOR THE MONTGOMERY AND ASBERG DEPRESSION RATING SCALE (SIGMA)

Janet B.W. Williams, Ph.D. and Kenneth A. Kobak, Ph.D.

INTERVIEWING GUIDELINES: The questions in bold for each item should be asked exactly as written unless the information has been previously obtained, in which case it is appropriate to restate the information for confirmation. Follow-up questions are provided for use when further exploration or additional clarification of symptoms is necessary. The specified questions should be asked until you have enough information to rate the item confidently. In some cases, you may also have to add your own follow-up questions to obtain necessary information. Note that questions in parentheses are optional, for use, for example, if information is unknown. Statements in ALL CAPITALS are interviewer instructions and should not be read to the subject.

RATING GUIDELINES: Ratings should be based on the subject's condition as observed in the past week (past 7 days). As specified in the item descriptions, three of the items, Reduced Sleep, Reduced Appetite, and Inability to Feel, are rated as present only when they reflect a change from before the depression began (EUTHYMIC BASELINE). The interviewer should attempt to identify the most recent 2-month period of non-depressed functioning and use this as a reference point. In some cases, such as when the subject has dysthymia, the referent should be to the last time the subject felt alright (i.e., not depressed or high) for at least a few weeks. When a clear euthymic baseline cannot be established because of chronic depressive symptoms, these three items should be rated as observed over the past 7 days instead of comparing to a previous time point.

This interview guide is based on the Montgomery-Asberg Depression Rating Scale (MADRS) (Montgomery SA, Asberg M: A new depression scale designed to be sensitive to change. *Br J Psychiatry*; 1979 134: 382-9). The scale itself has been retained in its original form, except for reversing the order of the first two items. This guide adds interview questions to aid in the assessment and application of the MADRS. Previous versions of this guide appeared in 1988, 1992, 1996, 2005 and 2008.

SIGMA 2011, v. 1.2

STRUCTURED INTERVIEW GUIDE FOR THE MONTGOMERY AND ASBERG DEPRESSION RATING SCALE (SIGMA)

PT'S INITIALS: _____ **PT'S ID:** _____ **TIME BEGAN SIGMA:** _____ **AM / PM**

INTERVIEWER: _____ **DATE:** _____

OVERVIEW: **I'd like to ask you some questions about the past week. How have you been feeling since last (DAY OF WEEK)?** IF OUTPATIENT: Have you been working? (What kind of work do you do? Have you been able to work your normal hours?) IF NOT WORKING OR WORKING LESS, CLARIFY WHY.	
In the past week, have you been feeling sad or unhappy? (Depressed at all?) IF YES: Can you describe what this has been like for you? (IF UNKNOWN: How bad has that been?) IF DEPRESSED: Does the feeling lift at all if something good happens? How much does your mood lift? Does the feeling ever go away completely? (How often have you had life in your mood this week? What things have made you feel better?) How often did you feel (depressed/OWN EQUIVALENT) this past week? (IF UNKNOWN: How many days this week did you feel that way? How much of each day?) **In the past week, how have you been feeling about the future?** (Have you been discouraged or pessimistic?) What have your thoughts been? How (discouraged or pessimistic) have you been? How often have you felt that way? Do you think things will ever get better for you? **ESTABLISH EUTHYMIC BASELINE: When was the last time you were well, not depressed at all, for at least 2 months?**	**1. REPORTED SADNESS.** Representing reports of depressed mood, regardless of whether it is reflected in appearance or not. Includes low spirits, despondency or the feeling of being beyond help and without hope. Rate according to intensity, duration, and the extent to which the mood is reported to be influenced by events. 0 - Occasional sadness in keeping with the circumstances. 1 – 2 - Sad or low but brightens up without difficulty. 3 – 4 - Pervasive feelings of sadness or gloominess. The mood is still influenced by external circumstances. 5 – 6 – Continuous or unvarying sadness, misery, or despondency.

SIGMA 2011, v. 1.2

RATING BASED ON OBSERVATION DURING INTERVIEW AND THE FOLLOWING QUESTIONS. **In the past week, do you think you have looked sad or depressed to other people?** Did anyone say you looked sad or down? **How about when you've looked in the mirror; did you look gloomy or depressed?** IF YES: How sad or depressed do you think you have looked? How much of the time over the past week do you think you have looked depressed or down? **Has it been hard for you to laugh or smile in the past week?**	**2. APPARENT SADNESS.** Representing despondency, gloom and despair. (More than just ordinary transient low spirits) reflected in speech, facial expressions, and posture. Rate by depth and inability to brighten up. 0 – No sadness 1 – 2 - Looks dispirited but does brighten up without difficulty. 3 - 4 - Appears sad and unhappy most of the time. 5 – 6 – Looks miserable all the time. Extremely despondent.
Have you felt tense or edgy in the last week? Have you felt anxious or nervous? IF YES: Can you describe what that has been like for you? How bad has it been? **What about feeling fearful that something bad is about to happen?** How much of the time have you felt (anxious/tense/OWN EQUIVALENT) over the past week? **Have you felt panicky in the past week?** IF YES: Can you describe this feeling? How often have you felt this way? IF YES TO ANY TENSION SYMPTOM: How hard has it been to control these feelings? (What has it taken to help you feel calmer? Has anything worked to calm you down?)	**3. INNER TENSION.** Representing feelings of ill-defined discomfort, edginess, inner turmoil, mental tension mounting to either panic, dread, or anguish. Rate according to intensity, frequency, duration and the extent of reassurance called for. 0 - Placid. Only fleeting inner tension. 1 - 2 - Occasional feelings of edginess and ill-defined discomfort. 3 – 4 – Continuous feelings of inner tension or intermittent panic which the patient can only master with some difficulty. 5 – 6 – Unrelenting dread or anguish. Overwhelming panic.
How has your sleeping been in the last week? (How many hours have you been sleeping, compared to usual?) **Have you had trouble falling asleep?** (How long has it been taking you to fall asleep this past week? How many nights?) **Have you been able to stay asleep through the night?** (Have you been waking up at all in the middle of the night? How long does it take you to fall back to sleep? How many nights?) **Have there been any mornings this past week when you have awakened earlier than (EUTHYMIC BASELINE)?** IF UNKNOWN: Has your sleeping been restless or disturbed?	**4. REDUCED SLEEP.** Representing the experience of reduced duration or depth of sleep compared to the subject's own normal pattern when well. 0 - Sleeps as usual. 1 - 2 - Slight difficulty dropping off to sleep or slightly reduced, light, or fitful sleep. 3 – 4 – Sleep reduced or broken by at least 2 hours. 5 – 6 – Less than 2 or 3 hours sleep.

SIGMA 2011, v. 1.2

How has your appetite been this past week? (What about compared to your usual appetite?) IF NOT REDUCED: Have you been less interested in food? (How much less?) **Does food taste as good as usual?** IF LESS: How much less? Does it have any taste at all? (Have you had to push yourself to eat or have other people had to urge you to eat?)	**5. REDUCED APPETITE.** Representing the feeling of a loss of appetite compared with when well. Rate by loss of desire for food or the need to force oneself to eat. 0 - Normal or increased appetite. 1 – 2 - Slightly reduced appetite. 3 – 4 – No appetite. Food is tasteless. 5 – 6 – Needs persuasion to eat at all.
Have you had trouble concentrating or collecting your thoughts in the past week? (How about at home or at work?) IF YES: Can you give me some examples? (Have you been able to concentrate on reading a book or on the computer? Do you need to read things over and over again? Are you able to follow movies or television?) How often has that happened in the past week? Has this caused any problems for you? **Have you had any trouble following a conversation?** (IF YES: How bad has that been? How often has that happened this past week?) NOTE: ALSO CONSIDER BEHAVIOR DURING INTERVIEW.	**6. CONCENTRATION DIFFICULTIES.** Representing difficulties in collecting one's thoughts mounting to incapacitating lack of concentration. Rate according to intensity, frequency, and degree of incapacity produced. 0 - No difficulties in concentration. 1 - 2 - Occasional difficulties in collecting one's thoughts. 3 – 4 – Difficulties in concentrating and sustaining thought which reduces ability to read or hold a conversation. 5 – 6 – Unable to read or converse without great difficulty.
Have you had any trouble getting started at things in the past week? IF YES: What things? How bad has that been? Have you had difficulty getting started at simple routine everyday things (like getting dressed, brushing your teeth, showering)? Are you OK once you get started at things or is it still more of an effort to get something done? Has there been anything that you needed to do that you were unable to do? Have you needed help to do things? IF YES: What things? How often? Have you done everyday things more slowly than usual? IF YES: Like what, for example? How bad has that been?	**7. LASSITUDE.** Representing a difficulty getting started, or slowness initiating and performing everyday activities. 0 - Hardly any difficulty in getting started. No sluggishness. 1 - 2 - Difficulties in starting activities. 3 – 4 – Difficulties in simple routine activities, which are carried out with effort. 5 – 6 – Complete lassitude. Unable to do anything without help.

SIGMA 2011, v. 1.2

Have you been less interested in things around you, or in activities you used to enjoy? IF YES: What things? How much less interested in (those things) are you now compared to (EUTHYMIC BASELINE)? **What things have you enjoyed this week?** How much did you enjoy them? **Has there been any change in your ability to feel emotions in the past week?** (Do you feel things less intensely than you used to, things like anger, grief, pleasure?) IF YES: Can you tell me more about that? (IF UNKNOWN: Are you able to feel any emotions at all?) **Have your feelings towards family and friends changed at all since (EUTHYMIC BASELINE)?** IF YES: Do you feel less towards them than you used to?	**8. INABILITY TO FEEL.** Representing the subjective experience of reduced interest in the surroundings, or activities that normally give pleasure. The ability to react with adequate emotion to circumstances or people is reduced. 0 - Normal interest in the surroundings and in other people. 1 - 2 - Reduced ability to enjoy usual interests. 3 – 4 – Loss of interest in the surroundings. Loss of feelings for friends and acquaintances. 5 – 6 – The experience of being emotionally paralyzed, inability to feel anger, grief or pleasure, and a complete or even painful failure to feel for close relatives and friends.
Have you been putting yourself down, or feeling that you're a failure in some way, over the past week? (Have you been blaming yourself for things that you've done, or not done?) IF YES: What have your thoughts been? How often have you felt that way? **In the past week have you been feeling guilty about anything? What about feeling as if you have done something bad or sinful?** IF YES: What have your thoughts been? How often have you felt that way? ALSO CONSIDER RESPONSES TO QUESTIONS ABOUT PESSIMISM FROM ITEM 1.	**9. PESSIMISTIC THOUGHTS.** Representing thoughts of guilt, inferiority, self-reproach, sinfulness, remorse, and ruin. 0 - No pessimistic thoughts. 1 - 2 - Fluctuating ideas of failure, self-reproach, or self-depreciation. 3 – 4 – Persistent self-accusations, or definite but still rational ideas of guilt or sin. Increasingly pessimistic about the future. 5 – 6 – Delusions of ruin, remorse, or unredeemable sin. Self-accusations which are absurd and unshakeable.

SIGMA 2011, v. 1.2

This past week, have you felt like life isn't worth living? (IF NO: What about feeling as if you're tired of living?) IF YES: Tell me about that. How often have you felt that way? **This week, have you thought that you would be better off dead?** IF YES: Tell me about that. How often have you felt that way? **Have you had thoughts of hurting or even killing yourself this past week?** IF YES: What have you thought about? How often have you had these thoughts? How long have they lasted? Have you actually made plans? IF YES: What are these plans? Have you made any preparations to carry out these plans? (Have you told anyone about it?)	**10. SUICIDAL THOUGHTS.** Representing the feeling that life is not worth living, that a natural death would be welcome, suicidal thoughts, and preparation for suicide. Suicidal attempts should not in themselves influence this rating. 0 - Enjoys life or takes it as it comes. 1 – 2 - Weary of life. Only fleeting suicidal thoughts. 3 - 4 - Probably better off dead. Suicidal thoughts are common, and suicide is considered as a possible solution, but without specific plans or intention. 5 – 6 – Explicit plans for suicide when there is an opportunity. Active preparations for suicide.

TIME ENDED SIGMA:	_____ **AM / PM**
TOTAL MADRS SCALE SCORE:	_____ _____

APPENDIX B. CLINICAL GLOBAL IMPRESSION – SEVERITY

Clinical Global Impression - Severity (CGI-S)

Considering your total clinical experience with this particular population, how mentally ill is the patient at this time?

0 = Not assessed
1 = Normal, not at all ill
2 = Borderline mentally ill
3 = Mildly ill
4 = Moderately ill
5 = Markedly ill
6 = Severely ill
7 = Among the most extremely ill patients

APPENDIX C. 17-ITEM HAMILTON DEPRESSION RATING SCALE

SIGH-D-17 Feb. 2013

STRUCTURED INTERVIEW GUIDE FOR THE HAMILTON DEPRESSION RATING SCALE – 17 ITEM VERSION (SIGH-D-17)

Janet B.W. Williams, PhD

INTERVIEWER

The first question for each item should be asked exactly as written. Often this question will elicit enough information about the severity and frequency of a symptom for you to rate the item with confidence. Follow-up questions are provided for use when further exploration or additional clarification of symptoms is necessary. The specified questions should be asked until you have enough information to rate the item confidently. In some cases, you may also have to add your own follow-up questions to obtain necessary information. You should ask for examples for any symptoms acknowledged as present (e.g., "*Can you give me an example of that?*"). For some of the HAM-D items, you may find you have already asked about some of the symptoms (for a previous item). You do not need to repeat questions about these symptoms unless you need additional information to rate their severity.

Time period. The interview questions indicate that the ratings should be based on the subject's condition in the past week.

Administration method. This version includes interview questions to help the clinician rate psychomotor agitation and psychomotor retardation, when the interview is administered by telephone. Several research studies have demonstrated that depression scale scores are equivalent whether the scale is administered face-to-face, by telephone, or by video (Williams JBW and Kobak KA: Development and Reliability of a Structured Interview Guide for the Montgomery-Asberg Depression Rating Scale. Br J Psychiatry 192, 52-58, 2008; Kobak KA: A comparison of face-to-face and remote administration of the Hamilton Depression Rating Scale via videoconferencing. J Telemed Telecare 10, 231-235, 2004).

Referent of "usual" or "normal" condition. In the HAM-D, most items are rated positive only if they represent a change from usual functioning. For this reason, several of the interview questions in the HAM-D refer to the subject's usual or normal functioning. The referent should be to the last time they felt okay (i.e., not depressed or high and normal interest in things) for at least two months. When no clear euthymic baseline can be established, one should rate symptomatic behavior as one sees it, even if it is not a change from the subject's usual dysphoric self.

This instrument provides an interview guide for the Hamilton Depression Scale (Hamilton, Max: A rating scale for depression. J Neurol Neurosurg Psychiat 23:56-61, 1960). The anchor point descriptions for all items except Helplessness, Hopelessness, and Worthlessness, with very minor modifications, have been taken from the ECDEU Assessment Manual (Guy, William, ECDEU Assessment Manual for Psychopharmacology. Revised 1976, DHEW Publication No. (ADM) 76-338). The loss of weight item has been simplified to eliminate the section for ratings by ward staff. A reliability study of the SIGH-D (interview guide for the HAM-D alone) was published in the Archives of General Psychiatry (1988;45:742-747). Additional designators were added in parentheses to the anchor points by Kobak, Lipsitz and Williams to further standardize ratings.

For further information and permission to use or translate the SIGH-D, contact Mapi Research Trust (internet: https://eprovide.mapi-trust.org).

SIGH-D-17 Feb. 2013

STRUCTURED INTERVIEW GUIDE FOR THE HAMILTON DEPRESSION RATING SCALE – 17 ITEM VERSION (SIGH-D-17)

SUBJECT'S INITIALS: _____ TIME BEGAN SIGH-D: _____

INTERVIEWER: _____ DATE: ____/____/____

OVERVIEW: **I'd like to ask you some questions about the past week. How have you been feeling since last (DAY OF WEEK)? IF OUTPATIENT: Have you been working? IF NOT: Why not?**

What's your mood been like this past week (compared to when you feel okay)?

Have you been feeling down or depressed?

IF YES: Can you describe what this feeling has been like for you? How bad is the feeling?

Does the feeling lift at all if something good happens?

How are you feeling about the future?

IF UNKNOWN: Have you been feeling discouraged or pessimistic?

IF YES: What have your thoughts been?

In the last week, how often have you felt (OWN EQUIVALENT FOR DEPRESSED MOOD)? On how many days? For how long each day?

Have you been crying at all? How often?

IF SCORED 1-4 ABOVE, ASK: How long have you been feeling this way (OWN EQUIVALENT FOR DEPRESSED MOOD)?

1. DEPRESSED MOOD (sadness, hopeless, helpless, worthless):

0	Absent
1	Indicated only on questioning *(occasional, mild depression)*
2	Spontaneously reported verbally *(persistent, mild to moderate depression)*
3	Communicated non-verbally, i.e., facial expression, posture, voice, tendency to weep *(persistent, moderate to severe depression)*
4	VIRTUALLY ONLY those feeling states reported in spontaneous verbal and non-verbal communication *(persistent, very severe depression, with extreme hopelessness or tearfulness)*

Version 2013.1
© Janet B.W. Williams, 1988. All Rights Reserved
SIGH-D – United States/English – Original version
SIGH-D_AU2.0_eng-USori

SIGH-D-17 Feb. 2013

How have you been spending your time this past week (when not at work)?	2. WORK AND ACTIVITIES:
Have you felt interested in doing (THOSE THINGS), or do you feel you have to push yourself to do them? How much less interested in these things have you been this past week compared to when you're not depressed? How hard to do you have to push yourself to do them? Have you stopped doing anything you used to do? (What about hobbies?) IF YES: Why? About how many hours a day do you spend doing things that interest you? Is there anything you look forward to? IF WORKING (IN OR OUT OF THE HOME): Have you been able to get as much (work) done as you usually do? How much less productive or efficient are you compared to before you were depressed?	0 No difficulty 1 Thoughts and feelings of incapacity, fatigue or weakness related to activities, work or hobbies *(mild reduction in interest or pleasure; no clear impairment in functioning)* 2 Loss of interest in activity, hobbies or work - by direct report of the patient or indirect in listlessness, indecision and vacillation (feels he has to push self to work or activities) *(clear reduction in interest, pleasure or functioning)* 3 Decrease in actual time spent in activities or decrease in productivity. In hosp., pt. spends less than 3 hrs./day in activities *(hospital job or hobbies)* exclusive of ward chores *(profound reduction in interest, pleasure, or functioning)* 4 Stopped working because of present illness. In hospital, no activities except ward chores, or fails to perform ward chores unassisted *(unable to work or fulfill primary role because of illness, and total loss of interest)*

Now let's talk about your sleep. What were your usual hours of going to sleep and waking up, before this began? When have you been falling asleep and waking up over the past week?	
Have you had any trouble falling asleep at the beginning of the night? (Right after you go to bed, how long has it been taking you to fall asleep?) How many nights this week have you had trouble falling asleep? Have you changed the time at which you try to get to sleep since you've been depressed?	**3. INSOMNIA EARLY (INITIAL INSOMNIA):** 0 No difficulty falling asleep 1 Complains of occasional difficulty falling asleep *(i.e., 30 minutes or more, 2-3 nights)* 2 Complains of nightly difficulty falling asleep *(i.e., 30 minutes or more, 4 or more nights)*

SIGH-D-17 Feb. 2013

During the past week, have you been waking up in the middle of the night? IF YES: Do you get out of bed? What do you do? (Only go to the bathroom?) When you get back in bed, are you able to fall right back asleep? How long does it take you to fall back asleep? Do you wake up more than once during the night? (IF YES: How long does it take for you to fall back to sleep each time?) Have you felt your sleeping has been restless or disturbed some nights? How many nights this week have you had that kind of trouble?	**4. INSOMNIA MIDDLE:** 0 No difficulty 1 Complains of being restless and disturbed during the night *(or occasional difficulty, i.e., 2-3 nights, 30 minutes or more)* 2 Waking during the night; any getting out of bed (except to void) *(often, i.e., 4 or more nights of difficulty, 30 minutes or more)*
What time have you been waking up in the morning for the last time, this past week? IF EARLY: Is that with an alarm clock, or do you just wake up yourself? What time do you usually wake up (that is, when you feel well)? How many mornings this past week have you awakened early?	**5. INSOMNIA LATE (TERMINAL INSOMNIA):** 0 No difficulty 1 Waking in early hours of morning but goes back to sleep *(occasional, i.e., 2-3 nights difficulty)* 2 Unable to fall asleep again if gets out of bed *(often, i.e., 4 or more nights difficulty)*

Sometimes, along with depression or anxiety, people might lose interest in sex. This week, how has your interest in sex been? (I'm not asking about actual sexual activity, but about your interest in sex.) Has there been any change in your interest in sex (from when you were feeling OK)? IF YES: How much less interest do you have compared to when you're not depressed? (Is it a little less or a lot less?)	**6. GENITAL SYMPTOMS** (such as loss of libido, menstrual disturbances): 0 Absent 1 Mild (*somewhat less interest than usual*) 2 Severe (*a lot less interest than usual*)
How has your appetite been this past week? (What about compared to your usual appetite?) IF LESS: How much less than usual? Have you had to force yourself to eat? Have other people had to urge you to eat? (Have you skipped meals?)	**7. SOMATIC SYMPTOMS GASTROINTESTINAL:** 0 None 1 Loss of appetite but eating without encouragement (*appetite somewhat less than usual*) 2 Difficulty eating without urging (*appetite significantly less than usual*)

SIGH-D-17 Feb. 2013

Have you lost any weight since this (DEPRESSION) began? IF YES: Did you lose any weight this last week? (Was it because of feeling depressed or down?) How much did you lose? IF NOT SURE: Do you think your clothes are any looser on you? FOLLOW-UP: Have you gained any of the weight back? IF YES: How much?	**8. LOSS OF WEIGHT** Rate by history: 0 No weight loss 1 Probable weight loss due to current depression 2 Definite *(according to patient)* weight loss due to depression
How has your energy been this past week? IF LOW ENERGY: Have you felt tired? (How much of the time? How bad has it been?) This week, have you had any aches or pains? (What about backaches or muscle aches?) (How much of the time? How bad has it been?) Have you felt any heaviness in your limbs, back, or head?	**9. SOMATIC SYMPTOMS GENERAL:** 0 None 1 Heaviness in limbs, back, or head. Backaches, muscle aches. Loss of energy and fatigability *(somewhat less energy than usual; mild, intermittent loss of energy or muscle aches/heaviness)* 2 Any clear-cut symptoms *(persistent, significant loss of energy or muscle aches/heaviness)*
Have you been putting yourself down this past week, feeling you've done things wrong, or let others down? IF YES: What have your thoughts been? Have you been feeling guilty about anything that you've done or not done? IF YES: What have your thoughts been? What about things that happened a long time ago? Have you thought that you've brought (THIS DEPRESSION) on yourself in some way? (Have you been hearing voices or seeing visions in the last week? IF YES: Tell me about them.)	**10. FEELINGS OF GUILT:** 0 Absent 1 Self-reproach; feels he has let people down 2 Ideas of guilt or rumination over past errors or sinful deeds *(feelings of guilt, remorse or shame)* 3 Present illness is a punishment; delusions of guilt *(severe, pervasive feelings of guilt)* 4 Hears accusatory or denunciatory voices and/or experiences threatening visual hallucinations

Page 5 of 8

SIGH-D-17 Feb. 2013

This past week, have you had thoughts that life is not worth living? **What about thinking you'd be better off dead?** **Have you had thoughts of hurting or killing yourself?** IF YES: What have you thought about? Have you actually done anything to hurt yourself?	**11. SUICIDE:** 0 Absent 1 Feels life is not worth living 2 Wishes he were dead or any thoughts of possible death to self 3 Suicidal ideas or gesture 4 Attempts at suicide
Have you been feeling anxious or tense this past week? IF YES: Is this more than is normal for you? **Have you been feeling irritable this past week? (IF YES): Can you give me some example? How bad has it been?** Have you been worrying a lot about little things, things you don't ordinarily worry about? IF YES: Like what, for example? How about worrying about big problems more than you need to? How much of the time has that happened this week? Has this caused you any problems or difficulties? IF YES: Like what, for example?	**12. ANXIETY PSYCHIC:** 0 No difficulty 1 Subjective tension and irritability *(mild, occasional)* 2 Worrying about minor matters *(moderate, causes some distress)* 3 Apprehensive attitude apparent in face or speech *(severe; significant impairment in functioning due to anxiety)* 4 Fears expressed without questioning *(symptoms incapacitating)*

Page 6 of 8

SIGH-D-17 Feb. 2013

Tell me if you've had any of the following physical symptoms in the past week. (READ LIST) FOR EACH SX ACKNOWLEDGED AS PRESENT: How much has (THE SX) been bothering you this past week? [How bad has it gotten? How much of the time, or how often, have you had it? Did (the symptom) interfere at all with your functioning or your usual activities?] NOTE: DO NOT RATE SXS THAT ARE CLEARLY RELATED TO A DOCUMENTED PHYSICAL CONDITION.	**13. ANXIETY SOMATIC** (physiologic concomitants of anxiety, such as GI - dry mouth, gas, indigestion, diarrhea, stomach cramps, belching CV - heart palpitations, headaches Resp - hyperventilating, sighing Urinary frequency Sweating): 0 Not present 1 Mild *(symptom(s) present only infrequently, no impairment, minimal distress)* 2 Moderate *(symptom(s) more persistent, or some interference with usual activities, moderate distress)* 3 Severe *(significant impairment in functioning)* 4 Incapacitating
In the last week, how much have your thoughts been focused on your physical health or how your body is working (compared to your normal thinking)? (Have you worried a lot about being or becoming physically ill? Have you really been preoccupied with this?) Have you worried a lot that you have a specific medical illness? Do you complain much about how you feel physically? Have you seen a doctor about these problems? IF YES: What did the doctor say?	**14. HYPOCHONDRIASIS:** 0 Not present 1 Self-absorption *(bodily)* (*some inappropriate worry about his/her health OR slightly concerned despite reassurance)* 2 Preoccupation with health *(often has excessive worries about his/her health OR definitely concerned has specific illness despite medical reassurance)* 3 Frequent complaints, requests for help, etc. *(is certain there is a physical problem which the doctors cannot confirm; exaggerated or unrealistic concerns about body and physical health)* 4 Hypochondriacal delusions
RATING BASED ON OBSERVATION DURING INTERVIEW	**15. INSIGHT:** 0 Acknowledges being depressed and ill OR not currently depressed 1 Acknowledges illness but attributes cause to bad food, overwork, virus, need for rest, etc. 2 Denies being ill at all

SIGH-D-17 Feb. 2013

RATING BASED ON OBSERVATION DURING INTERVIEW **IF INTERVIEWING BY PHONE:** During this interview have you been fidgeting a lot or moving around a lot in your chair? Have you had trouble sitting still? Have you been shaking your legs or playing with your hair or your hands?	**16. AGITATION:** 0 None 1 Fidgetiness *(slight agitation or mild restlessness)* 2 Playing with hands, hair, etc. *(moderate to marked restlessness or agitation)* 3 Moving about, can't sit still *(cannot remain seated)* 4 Hand-wringing, nail biting, hair-pulling, biting of lips *(interview cannot be conducted; severe agitation)*
RATING BASED ON OBSERVATION DURING INTERVIEW **IF INTERVIEWING BY PHONE:** During this interview have you been moving slowly, reacting slowly, or speaking more slowly than usual for you?	**17. RETARDATION** (slowness of thought and speech; impaired ability to concentrate; decreased motor activity): 0 Normal speech and thought 1 Slight retardation at interview *(mild psychomotor retardation)* 2 Obvious retardation at interview *(moderate; some difficulty with interview, noticeable pauses and slowness of thought)* 3 Interview difficult *(severe psychomotor retardation; very long pauses)* 4 Complete stupor *(extreme retardation; interview barely possible)*

TIME ENDED SIGH-D-17:	**_____ AM / PM ET / CT / PT**
TOTAL HAM-D-17 SCORE:	**_____**

APPENDIX D. SHEEHAN DISABILITY SCALE

SHEEHAN DISABILITY SCALE

A BRIEF, PATIENT RATED, MEASURE OF DISABILITY AND IMPAIRMENT

Please mark ONE circle for each scale.

In the past (timeframe):

WORK* / SCHOOL

The symptoms have disrupted your work / school work:

☐ I have not worked /studied at all during the past week **for reasons unrelated to the disorder.**
* Work includes paid, unpaid volunteer work or training. If your symptoms interfered with your ability to find or hold a job or contributed in any way to your currently not working, you must give a score on this scale.

SOCIAL LIFE

The symptoms have disrupted your social life / leisure activities:

FAMILY LIFE / HOME RESPONSIBILITIES

The symptoms have disrupted your family life / home responsibilities:

DAYS LOST

On how many days in the last week did your symptoms cause you to miss school or work or leave you unable to carry out your normal daily responsibilities? _____

DAYS UNDERPRODUCTIVE

On how many days in the last week did you feel so impaired by your symptoms, that even though you went to school or work or had other daily responsibilities, your productivity was reduced? ____

APPENDIX E. THE QUICK INVENTORY OF DEPRESSIVE SYMPTOMOLOGY – SELF REPORT

SELF-REPORT QUICK INVENTORY OF DEPRESSIVE SYMPTOMATOLOGY (QIDS-SR16)

Name _____ Date _____

Please circle the one response to each item that best describes you for the past seven days.

1. Falling Asleep:
   - 0 I never take longer than 30 minutes to fall asleep.
   - 1 I take at least 30 minutes to fall asleep, less than half the time.
   - 2 I take at least 30 minutes to fall asleep, more than half the time.
   - 3 I take more than 60 minutes to fall asleep, more than half the time.
2. Sleep During the Night:
   - 0 I do not wake up at night.
   - 1 I have a restless, light sleep with a few brief awakenings each night.
   - 2 I wake up at least once a night, but I go back to sleep easily.
   - 3 I awaken more than once a night and stay awake for 20 minutes or more, more than half the time.
3. Waking Up Too Early:
   - 0 Most of the time, I awaken no more than 30 minutes before I need to get up.
   - 1 More than half the time, I awaken more than 30 minutes before I need to get up.
   - 2 I almost always awaken at least one hour or so before I need to, but I go back to sleep eventually.
   - 3 I awaken at least one hour before I need to, and can't go back to sleep.
4. Sleeping Too Much:
   - 0 I sleep no longer than 7-8 hours/night, without napping during the day.
   - 1 I sleep no longer than 10 hours in a 24-hour period including naps.
   - 2 I sleep no longer than 12 hours in a 24-hour period including naps.
   - 3 I sleep longer than 12 hours in a 24-hour period including naps.
5. Feeling Sad:
   - 0 I do not feel sad
   - 1 I feel sad less than half the time.
   - 2 I feel sad more than half the time.
   - 3 I feel sad nearly all of the time.
6. Decreased Appetite:
   - 0 There is no change in my usual appetite.
   - 1 I eat somewhat less often or lesser amounts of food than usual.
   - 2 I eat much less than usual and only with personal effort.
   - 3 I rarely eat within a 24-hour period, and only with extreme personal effort or when others persuade me to eat.
7. Increased Appetite:
   - 0 There is no change from my usual appetite.
   - 1 I feel a need to eat more frequently than usual.
   - 2 I regularly eat more often and/or greater amounts of food than usual.
   - 3 I feel driven to overeat both at mealtime and between meals.
8. Decreased Weight (Within the Last Two Weeks):
   - 0 I have not had a change in my weight.
   - 1 I feel as if I've had a slight weight loss.
   - 2 I have lost 2 pounds or more.
   - 3 I have lost 5 pounds or more.
9. Increased Weight (Within the Last Two Weeks):
   - 0 I have not had a change in my weight.
   - 1 I feel as if I've had a slight weight gain.
   - 2 I have gained 2 pounds or more.
   - 3 I have gained 5 pounds or more.
10. Concentration/Decision Making:
    - 0 There is no change in my usual capacity to concentrate or make decisions.
    - 1 I occasionally feel indecisive or find that my attention wanders.
    - 2 Most of the time, I struggle to focus my attention or to make decisions.
    - 3 I cannot concentrate well enough to read or cannot make even minor decisions.

11. View of Myself:
    - 0 I see myself as equally worthwhile and deserving as other people.
    - 1 I am more self-blaming than usual.
    - 2 I largely believe that I cause problems for others.
    - 3 I think almost constantly about major and minor defects in myself.
12. Thoughts of Death or Suicide:
    - 0 I do not think of suicide or death.
    - 1 I feel that life is empty or wonder if it's worth living.
    - 2 I think of suicide or death several times a week for several minutes.
    - 3 I think of suicide or death several times a day in some detail, or I have made specific plans for suicide or have actually tried to take my life.
13. General Interest:
    - 0 There is no change from usual in how interested I am in other people or activities.
    - 1 I notice that I am less interested in people or activities.
    - 2 I find I have interest in only one or two of my formerly pursued activities.
    - 3 I have virtually no interest in formerly pursued activities.

To Score:

1. Enter the highest score on any 1 of the 4 sleep items (1-4) _____
2. Item 5 _____
3. Enter the highest score on any 1 appetite/ weight item (6-9) _____
4. Item 10 _____
5. Item 11 _____
6. Item 12 _____
7. Item 13 _____
8. Item 14 _____
9. Enter the highest score on either of the 2 psychomotor items (15 and 16) _____

TOTAL SCORE (Range 0-27) _____

14. Energy Level:
    - 0 There is no change in my usual level of energy.
    - 1 I get tired more easily than usual.
    - 2 I have to make a big effort to start or finish my usual daily activities (for example, shopping, homework, cooking or going to work).
    - 3 I really cannot carry out most of my usual daily activities because I just don't have the energy.
15. Feeling slowed down:
    - 0 I think, speak, and move at my usual rate of speed.
    - 1 I find that my thinking is slowed down or my voice sounds dull or flat.
    - 2 It takes me several seconds to respond to most questions and I'm sure my thinking is slowed.
    - 3 I am often unable to respond to questions without extreme effort.
16. Feeling restless:
    - 0 I do not feel restless.
    - 1 I'm often fidgety, wringing my hands, or need to shift how I am sitting.
    - 2 I have impulses to move about and am quite restless.
    - 3 At times, I am unable to stay seated and need to pace around.

QIDS-SR16 - United States/English – Original version

APPENDIX F. QUALITY OF LIFE ENJOYMENT AND SATISFACTION QUESTIONNAIRE – SHORT FORM

Name:_____ Date:_____

Quality of Life Enjoyment and Satisfaction Questionnaire – Short Form (Q-LES-Q-SF)

Taking everything into consideration, during the past week how satisfied have you been with your.....

	Very Poor	Poor	Fair	Good	Very Good
.....physical health?	1	2	3	4	5
.....mood?	1	2	3	4	5
.....work?	1	2	3	4	5
.....household activities?	1	2	3	4	5
.....social relationships?	1	2	3	4	5
.....family relationships?	1	2	3	4	5
.....leisure time activities?	1	2	3	4	5
.....ability to function in daily life?	1	2	3	4	5
.....sexual drive, interest and/or performance?*	1	2	3	4	5
.....economic status?	1	2	3	4	5
.....living/housing situation?*	1	2	3	4	5
.....ability to get around physically without feeling dizzy or unsteady or falling?*	1	2	3	4	5
.....your vision in terms of ability to do work or hobbies?*	1	2	3	4	5
.....overall sense of well being?	1	2	3	4	5
.....medication? (If not taking any, check here_____and leave item blank.)	1	2	3	4	5
.....How would you rate your overall life satisfaction and contentment during the past week?	1	2	3	4	5

*If satisfaction is very poor, poor or fair on these items, please UNDERLINE the factor(s) associated with a lack of satisfaction.

APPENDIX G. COLUMBIA – SUICIDE SEVERITY RATING SCALE – BASELINE/SCREENING VERSION

COLUMBIA-SUICIDE SEVERITY RATING SCALE (C-SSRS)

Baseline/Screening Version

Version 1/14/09

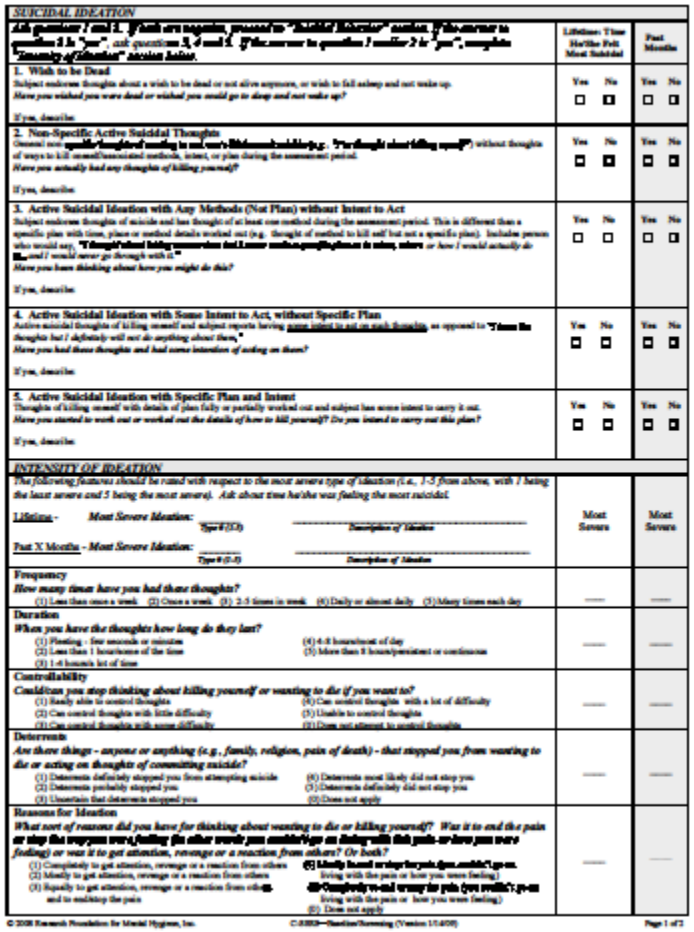
Posner, K.; Brent, D.; Lucas, C.; Gould, M.; Stanley, B.; Brown, G.; Fisher, P.; Zelazny, J.; Burke, A.; Oquendo, M.; Mann, J.

Disclaimer:

This scale is intended to be used by individuals who have received training in its administration. The questions contained in the Columbia-Suicide Severity Rating Scale are suggested probes. Ultimately, the determination of the presence of suicidal ideation or behavior depends on the judgment of the individual administering the scale.

Definitions of behavioral suicidal events in this scale are based on those used in ***The Columbia Suicide History Form****, developed by John Mann, MD and Maria Oquendo, MD, Conte Center for the Neuroscience of Mental Disorders (CCNMD), New York State Psychiatric Institute, 1051 Riverside Drive, New York, NY, 10032. (Oquendo M. A., Halberstam B. & Mann J. J., Risk factors for suicidal behavior: utility and limitations of research instruments. In M.B. First [Ed.] Standardized Evaluation in Clinical Practice, pp. 103 -130, 2003.)*

For reprints of the C-SSRS contact Kelly Posner, Ph.D., New York State Psychiatric Institute, 1051 Riverside Drive, New York, New York, 10032; inquiries and training requirements contact posnerk@nyspi.columbia.edu



SUICIDAL IDEATION

Ask questions 1 and 2. If both are negative, proceed to "Suicidal Behavior" section. If the answer to question 2 is "yes", ask questions 3, 4 and 5. If the answer to question 1 and/or 2 is "yes", complete "Intensity of Ideation" section below.

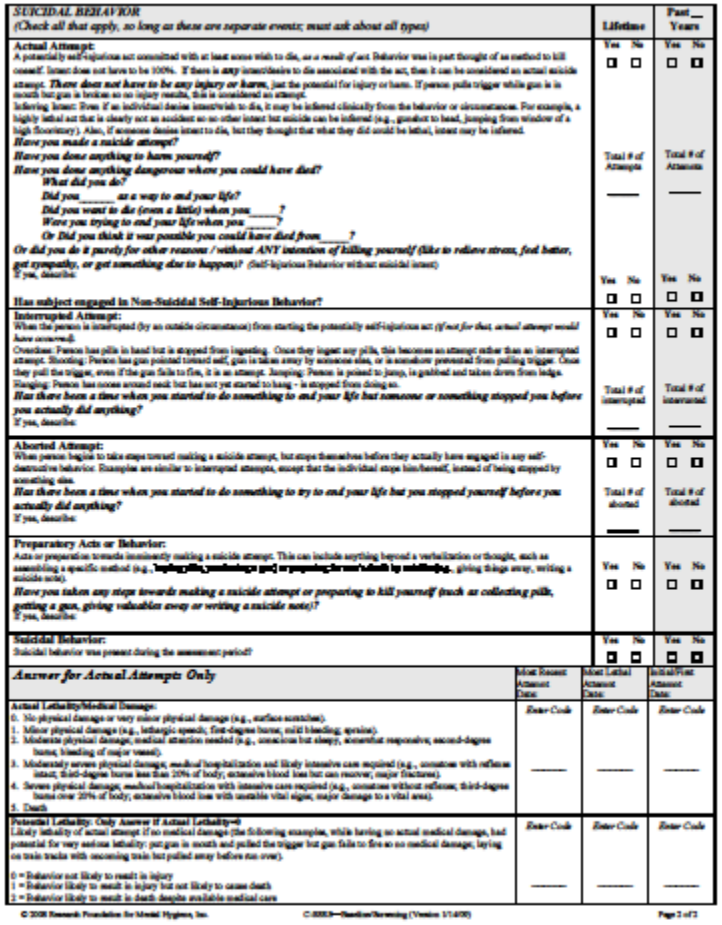
	Lifetime: Time He/She Felt Most Suicidal	Past Months
1. Wish to be Dead Subject endorses thoughts about a wish to be dead or not alive anymore, or wish to fall asleep and not wake up. *Have you wished you were dead or wished you could go to sleep and not wake up?* If yes, describe:	Yes ☐ No ☐	Yes ☐ No ☐
2. Non-Specific Active Suicidal Thoughts General non-specific thoughts of wanting to end one's life/commit suicide (e.g., *"I've thought about killing myself"*) without thoughts of ways to kill oneself/associated methods, intent, or plan during the assessment period. *Have you actually had any thoughts of killing yourself?* If yes, describe:	Yes ☐ No ☐	Yes ☐ No ☐
3. Active Suicidal Ideation with Any Methods (Not Plan) without Intent to Act Subject endorses thoughts of suicide and has thought of at least one method during the assessment period. This is different than a specific plan with time, place or method details worked out (e.g. thought of method to kill self but not a specific plan). Includes person who would say, *"I thought about killing myself but I never would or could, specifically how to, where or how I would actually do it... and I would never go through with it."* *Have you been thinking about how you might do this?* If yes, describe:	Yes ☐ No ☐	Yes ☐ No ☐
4. Active Suicidal Ideation with Some Intent to Act, without Specific Plan Active suicidal thoughts of killing oneself and subject reports having some intent to act on such thoughts, as opposed to *"I have the thoughts but I definitely will not do anything about them."* *Have you had these thoughts and had some intention of acting on them?* If yes, describe:	Yes ☐ No ☐	Yes ☐ No ☐
5. Active Suicidal Ideation with Specific Plan and Intent Thoughts of killing oneself with details of plan fully or partially worked out and subject has some intent to carry it out. *Have you started to work out or worked out the details of how to kill yourself? Do you intend to carry out this plan?* If yes, describe:	Yes ☐ No ☐	Yes ☐ No ☐

INTENSITY OF IDEATION

The following features should be rated with respect to the most severe type of ideation (i.e., 1-5 from above, with 1 being the least severe and 5 being the most severe). Ask about time he/she was feeling the most suicidal.

	Most Severe	Most Severe
Lifetime - *Most Severe Ideation:* Type # (1-5) _____ Description of Ideation _____ Past X Months - *Most Severe Ideation:* Type # (1-5) _____ Description of Ideation _____		
Frequency *How many times have you had these thoughts?* (1) Less than once a week (2) Once a week (3) 2-5 times in week (4) Daily or almost daily (5) Many times each day	____	____
Duration *When you have the thoughts how long do they last?* (1) Fleeting - few seconds or minutes (4) 4-8 hours/most of day (2) Less than 1 hour/some of the time (5) More than 8 hours/persistent or constant (3) 1-4 hours/a lot of time	____	____
Controllability *Could/can you stop thinking about killing yourself or wanting to die if you want to?* (1) Easily able to control thoughts (4) Can control thoughts with a lot of difficulty (2) Can control thoughts with little difficulty (5) Unable to control thoughts (3) Can control thoughts with some difficulty (0) Does not attempt to control thoughts	____	____
Deterrents *Are there things - anyone or anything (e.g., family, religion, pain of death) - that stopped you from wanting to die or acting on thoughts of committing suicide?* (1) Deterrents definitely stopped you from attempting suicide (4) Deterrents most likely did not stop you (2) Deterrents probably stopped you (5) Deterrents definitely did not stop you (3) Uncertain that deterrents stopped you (0) Does not apply	____	____
Reasons for Ideation *What sort of reasons did you have for thinking about wanting to die or killing yourself? Was it to end the pain or stop the way you were feeling (the other words you used) or was it to get attention, revenge or a reaction from others? Or both?* (1) Completely to get attention, revenge or a reaction from others (5) Mostly [illegible] living with the pain or how you were feeling] (2) Mostly to get attention, revenge or a reaction from others (3) Equally to get attention, revenge or a reaction from others and to end/stop the pain (4) Completely to end/stop the pain (you couldn't go on living with the pain or how you were feeling) (0) Does not apply	____	____

 C-SSRS—Baseline/Screening (Version 1/14/09) Page 1 of 2



SUICIDAL BEHAVIOR *(Check all that apply, so long as these are separate events; must ask about all types)*	Lifetime	Past __ Years
Actual Attempt: A potentially self-injurious act committed with at least some wish to die, *as a result of act*. Behavior was in part thought of as method to kill oneself. Intent does not have to be 100%. If there is *any* intentionality to die associated with the act, then it can be considered an actual suicide attempt. ***There does not have to be any injury or harm,*** just the potential for injury or harm. If person pulls trigger while gun is in mouth but gun is broken so no injury results, this is considered an attempt. Inferring Intent: Even if an individual denies intent/wish to die, it may be inferred clinically from the behavior or circumstances. For example, a highly lethal act that is clearly not an accident so no other intent but suicide can be inferred (e.g., gunshot to head, jumping from window of a high floor/story). Also, if someone denies intent to die, but they thought that what they did could be lethal, intent may be inferred. ***Have you made a suicide attempt?*** ***Have you done anything to harm yourself?*** ***Have you done anything dangerous where you could have died?*** ***What did you do?*** ***Did you_____ as a way to end your life?*** ***Did you want to die (even a little) when you_____?*** ***Were you trying to end your life when you_____?*** ***Or Did you think it was possible you could have died from_____?*** ***Or did you do it purely for other reasons / without ANY intention of killing yourself (like to relieve stress, feel better, get sympathy, or get something else to happen)?*** *(Self-Injurious Behavior without suicidal intent)* If yes, describe:	Yes ☐ No ☐ Total # of Attempts ___	Yes ☐ No ☐ Total # of Attempts ___
Has subject engaged in Non-Suicidal Self-Injurious Behavior?	Yes ☐ No ☐	Yes ☐ No ☐
Interrupted Attempt: When the person is interrupted (by an outside circumstance) from starting the potentially self-injurious act *(If not for that, actual attempt would have occurred).* Overdose: Person has pills in hand but is stopped from ingesting. Once they ingest any pills, this becomes an attempt rather than an interrupted attempt. Shooting: Person has gun pointed toward self, gun is taken away by someone else, or is somehow prevented from pulling trigger. Once they pull the trigger, even if the gun fails to fire, it is an attempt. Jumping: Person is poised to jump, is grabbed and taken down from ledge. Hanging: Person has noose around neck but has not yet started to hang - is stopped from doing so. ***Has there been a time when you started to do something to end your life but someone or something stopped you before you actually did anything?*** If yes, describe:	Yes ☐ No ☐ Total # of Interrupted ___	Yes ☐ No ☐ Total # of Interrupted ___
Aborted Attempt: When person begins to take steps toward making a suicide attempt, but stops themselves before they actually have engaged in any self-destructive behavior. Examples are similar to interrupted attempts, except that the individual stops him/herself, instead of being stopped by something else. ***Has there been a time when you started to do something to try to end your life but you stopped yourself before you actually did anything?*** If yes, describe:	Yes ☐ No ☐ Total # of Aborted ___	Yes ☐ No ☐ Total # of Aborted ___
Preparatory Acts or Behavior: Acts or preparation towards imminently making a suicide attempt. This can include anything beyond a verbalization or thought, such as assembling a specific method (e.g., **buying pills, purchasing a gun] or preparing for one's death by suicide**), giving things away, writing a suicide note). ***Have you taken any steps towards making a suicide attempt or preparing to kill yourself (such as collecting pills, getting a gun, giving valuables away or writing a suicide note)?*** If yes, describe:	Yes ☐ No ☐	Yes ☐ No ☐
Suicidal Behavior: Suicidal behavior was present during the assessment period?	Yes ☐ No ☐	Yes ☐ No ☐

Answer for Actual Attempts Only	Most Recent Attempt Date:	Most Lethal Attempt Date:	Initial/First Attempt Date:
Actual Lethality/Medical Damage: 0. No physical damage or very minor physical damage (e.g., surface scratches). 1. Minor physical damage (e.g., lethargic speech; first-degree burns; mild bleeding; sprains). 2. Moderate physical damage; medical attention needed (e.g., conscious but sleepy, somewhat responsive; second-degree burns; bleeding of major vessel). 3. Moderately severe physical damage; medical hospitalization and likely intensive care required (e.g., comatose with reflexes intact; third-degree burns less than 20% of body; extensive blood loss but can recover; major fractures). 4. Severe physical damage; medical hospitalization with intensive care required (e.g., comatose without reflexes; third-degree burns over 20% of body; extensive blood loss with unstable vital signs; major damage to a vital area). 5. Death	*Enter Code* ___	*Enter Code* ___	*Enter Code* ___
Potential Lethality: Only Answer if Actual Lethality=0 Likely lethality of actual attempt if no medical damage (the following examples, while having no actual medical damage, had potential for very serious lethality: put gun in mouth and pulled the trigger but gun fails to fire so no medical damage; laying on train tracks with oncoming train but pulled away before run over). 0 = Behavior not likely to result in injury 1 = Behavior likely to result in injury but not likely to cause death 2 = Behavior likely to result in death despite available medical care	*Enter Code* ___	*Enter Code* ___	*Enter Code* ___

 C-SSRS—Baseline/Screening (Version 1/14/08) Page 2 of 2

APPENDIX H. COLUMBIA – SUICIDE SEVERITY RATING SCALE – SINCE LAST VISIT VERSION

COLUMBIA-SUICIDE SEVERITY RATING SCALE (C-SSRS)

Since Last Visit

Version 1/14/09

Posner, K.; Brent, D.; Lucas, C.; Gould, M.; Stanley, B.; Brown, G.; Fisher, P.; Zelazny, J.; Burke, A.; Oquendo, M.; Mann, J.

Disclaimer:

This scale is intended to be used by individuals who have received training in its administration. The questions contained in the Columbia-Suicide Severity Rating Scale are suggested probes. Ultimately, the determination of the presence of suicidal ideation or behavior depends on the judgment of the individual administering the scale.

Definitions of behavioral suicidal events in this scale are based on those used in ***The Columbia Suicide History Form****, developed by John Mann, MD and Maria Oquendo, MD, Conte Center for the Neuroscience of Mental Disorders (CONMD), New York State Psychiatric Institute, 1051 Riverside Drive, New York, NY, 10032. (Oquendo M. A., Halberstam B. & Mann J. J., Risk factors for suicidal behavior: utility and limitations of research instruments. In M.B. First (Ed.)* Standardized Evaluation in Clinical Practice*, pp. 103 -130, 2003.)*

For reprints of the C-SSRS contact Kelly Posner, Ph.D., New York State Psychiatric Institute, 1051 Riverside Drive, New York, New York, 10032; inquiries and training requirements contact posnerk@nyspi.columbia.edu

C-SSRS Since Last Visit - United States/English - Mapi.

SUICIDAL IDEATION

Ask questions 1 and 2. If both are negative, proceed to "Suicidal Behavior" section. If the answer to question 2 is "yes", ask questions 3, 4 and 5. If the answer to question 1 and/or 2 is "yes", complete "Intensity of Ideation" section below.

Since Last Visit

1. Wish to be Dead — Yes ☐ No ☐
Subject endorses thoughts about a wish to be dead or not alive anymore, or wish to fall asleep and not wake up.
Have you wished you were dead or wished you could go to sleep and not wake up?
If yes, describe:

2. Non-Specific Active Suicidal Thoughts — Yes ☐ No ☐
General non-specific thoughts of wanting to end one's life/commit suicide (e.g., *"I've thought about killing myself"*) without thoughts of ways to kill oneself/associated methods, intent, or plan during the assessment period.
Have you actually had any thoughts of killing yourself?
If yes, describe:

3. Active Suicidal Ideation with Any Methods (Not Plan) without Intent to Act — Yes ☐ No ☐
Subject endorses thoughts of suicide and has thought of at least one method during the assessment period. This is different than a specific plan with time, place or method details worked out (e.g., thought of method to kill self but not a specific plan). Includes person who would say, *"I thought about taking an overdose but I never made a specific plan as to when, where or how I would actually do it…and I would never go through with it"*.
Have you been thinking about how you might do this?
If yes, describe:

4. Active Suicidal Ideation with Some Intent to Act, without Specific Plan — Yes ☐ No ☐
Active suicidal thoughts of killing oneself and subject reports having some intent to act on such thoughts, as opposed to *"I have the thoughts but I definitely will not do anything about them"*.
Have you had these thoughts and had some intention of acting on them?
If yes, describe:

5. Active Suicidal Ideation with Specific Plan and Intent — Yes ☐ No ☐
Thoughts of killing oneself with details of plan fully or partially worked out and subject has some intent to carry it out.
Have you started to work out or worked out the details of how to kill yourself? Do you intend to carry out this plan?
If yes, describe:

INTENSITY OF IDEATION

The following features should be rated with respect to the most severe type of ideation (i.e.,1-5 from above, with 1 being the least severe and 5 being the most severe).

Most Severe

Most Severe Ideation: _____ _____
Type # (1-5) *Description of Ideation*

Frequency —
How many times have you had these thoughts?
(1) Less than once a week (2) Once a week (3) 2-5 times in week (4) Daily or almost daily (5) Many times each day

Duration —
When you have the thoughts how long do they last?
(1) Fleeting - few seconds or minutes (4) 4-8 hours/most of day
(2) Less than 1 hour/some of the time (5) More than 8 hours/persistent or continuous
(3) 1-4 hours/a lot of time

Controllability —
Could/can you stop thinking about killing yourself or wanting to die if you want to?
(1) Easily able to control thoughts (4) Can control thoughts with a lot of difficulty
(2) Can control thoughts with little difficulty (5) Unable to control thoughts
(3) Can control thoughts with some difficulty (0) Does not attempt to control thoughts

Deterrents —
Are there things - anyone or anything (e.g., family, religion, pain of death) - that stopped you from wanting to die or acting on thoughts of committing suicide?
(1) Deterrents definitely stopped you from attempting suicide (4) Deterrents most likely did not stop you
(2) Deterrents probably stopped you (5) Deterrents definitely did not stop you
(3) Uncertain that deterrents stopped you (0) Does not apply

Reasons for Ideation —
What sort of reasons did you have for thinking about wanting to die or killing yourself? Was it to end the pain or stop the way you were feeling (in other words you couldn't go on living with this pain or how you were feeling) or was it to get attention, revenge or a reaction from others? Or both?
(1) Completely to get attention, revenge or a reaction from others (4) Mostly to end or stop the pain (you couldn't go on living with the pain or how you were feeling)
(2) Mostly to get attention, revenge or a reaction from others (5) Completely to end or stop the pain (you couldn't go on living with the pain or how you were feeling)
(3) Equally to get attention, revenge or a reaction from others and to end/stop the pain (0) Does not apply

 C-SSRS – Since Last Visit (Version 1/14/09) Page 1 of 2

SUICIDAL BEHAVIOR *(Check all that apply, so long as these are separate events; must ask about all types)*	**Since Last Visit**
Actual Attempt: A potentially self-injurious act committed with at least some wish to die, *as a result of act.* Behavior was in part thought of as method to kill oneself. Intent does not have to be 100%. If there is *any* intentionality to die associated with the act, then it can be considered an actual suicide attempt. *There does not have to be any injury or harm,* just the potential for injury or harm. If person pulls trigger while gun is in mouth but gun is broken so no injury results, this is considered an attempt. Inferring Intent: Even if an individual denies intent/wish to die, it may be inferred clinically from the behavior or circumstances. For example, a highly lethal act that is clearly not an accident (so no other intent but suicide can be inferred (e.g., gunshot to head, jumping from window of a high floor/story). Also, if someone denies intent to die, but they thought that what they did could be lethal, intent may be inferred. ***Have you made a suicide attempt?*** ***Have you done anything to harm yourself?*** ***Have you done anything dangerous where you could have died?*** ***What did you do?*** ***Did you _____as a way to end your life?*** ***Did you want to die (even a little) when you _____?*** ***Were you trying to end your life when you _____?*** ***Or Did you think it was possible you could have died from _____?*** ***Or did you do it purely for other reasons / without ANY intention of killing yourself (like to relieve stress, feel better, get sympathy, or get something else to happen)?*** (Self-Injurious Behavior without suicidal intent) If yes, describe: **Has subject engaged in Non-Suicidal Self-Injurious Behavior?**	Yes ☐ No ☐ Total # of Attempts _____ Yes ☐ No ☐
Interrupted Attempt: When the person is interrupted (by an outside circumstance) from starting the potentially self-injurious act *(if not for that, actual attempt would have occurred).* Overdose: Person has pills in hand but is stopped from ingesting. Once they ingest any pills, this becomes an attempt rather than an interrupted attempt. Shooting: Person has gun pointed toward self, gun is taken away by someone else, or is somehow prevented from pulling trigger. Once they pull the trigger, even if the gun fails to fire, it is an attempt. Jumping: Person is poised to jump, is grabbed and taken down from ledge. Hanging: Person has noose around neck but has not yet started to hang - is stopped from doing so. ***Has there been a time when you started to do something to end your life but someone or something stopped you before you actually did anything?*** If yes, describe:	Yes ☐ No ☐ Total # of interrupted _____
Aborted Attempt: When person begins to take steps toward making a suicide attempt, but stops themselves before they actually have engaged in any self-destructive behavior. Examples are similar to interrupted attempts, except that the individual stops him/herself, instead of being stopped by something else. ***Has there been a time when you started to do something to try to end your life but you stopped yourself before you actually did anything?*** If yes, describe:	Yes ☐ No ☐ Total # of aborted _____
Preparatory Acts or Behavior: Acts or preparation towards imminently making a suicide attempt. This can include anything beyond a verbalization or thought, such as assembling a specific method (e.g., buying pills, purchasing a gun) or preparing for one's death by suicide (e.g., giving things away, writing a suicide note). ***Have you taken any steps towards making a suicide attempt or preparing to kill yourself (such as collecting pills, getting a gun, giving valuables away or writing a suicide note)?*** If yes, describe:	Yes ☐ No ☐
Suicidal Behavior: Suicidal behavior was present during the assessment period?	Yes ☐ No ☐
Suicide:	Yes ☐ No ☐
Answer for Actual Attempts Only	Most Lethal Attempt Date:
Actual Lethality/Medical Damage: 0. No physical damage or very minor physical damage (e.g., surface scratches). 1. Minor physical damage (e.g., lethargic speech; first-degree burns; mild bleeding; sprains). 2. Moderate physical damage; medical attention needed (e.g., conscious but sleepy, somewhat responsive; second-degree burns; bleeding of major vessel). 3. Moderately severe physical damage; medical hospitalization and likely intensive care required (e.g., comatose with reflexes intact; third-degree burns less than 20% of body; extensive blood loss but can recover; major fractures). 4. Severe physical damage; medical hospitalization with intensive care required (e.g., comatose without reflexes; third-degree burns over 20% of body; extensive blood loss with unstable vital signs; major damage to a vital area). 5. Death	*Enter Code* _____
Potential Lethality: Only Answer if Actual Lethality=0 Likely lethality of actual attempt if no medical damage (the following examples, while having no actual medical damage, had potential for very serious lethality: put gun in mouth and pulled the trigger but gun fails to fire so no medical damage; laying on train tracks with oncoming train but pulled away before run over). 0 = Behavior not likely to result in injury 1 = Behavior likely to result in injury but not likely to cause death 2 = Behavior likely to result in death despite available medical care	*Enter Code* _____

 C-SSRS—Since Last Visit (Version 1/1 6/08) Page 2 of 2