An Experimental Therapeutics Study of a Monoclonal Antibody Against Interleukin 17A in Patients With Treatment-Resistant Depression
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An Experimental Therapeutics Study of Ixekizumab, a Monoclonal Antibody Against Interleukin 17A, on Anhedonia, Reward Circuit Function, and Blood Brain Barrier Physiology in Patients with Treatment- Resistant Depression

Protocol Number: 21-00325

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STATEMENT OF COMPLIANCE

The trial will be conducted in accordance with International Conference on Harmonization Good Clinical Practice (ICH GCP), applicable United States (US) Code of Federal Regulations (CFR), and the Hope for Depression Research Foundation Terms and Conditions of Award. The Principal Investigator will assure that no deviation from, or changes to the protocol will take place without prior agreement from the Investigational New Drug (IND) or Investigational Device Exemption (IDE) sponsor, funding agency and documented approval from the Institutional Review Board (IRB), except where necessary to eliminate an immediate hazard(s) to the trial participants. All personnel involved in the conduct of this study have completed Human Subjects Protection and ICH GCP Training.

The protocol, informed consent form(s), recruitment materials, and all participant materials will be submitted to the IRB for review and approval. Approval of both the protocol and the consent form must be obtained before any participant is enrolled. Any amendment to the protocol will require review and approval by the IRB before the changes are implemented to the study. All changes to the consent form will be IRB approved; a determination will be made regarding whether a new consent needs to be obtained from participants who provided consent, using a previously approved consent form.

1 PROTOCOL SUMMARY

1.1 SYNOPSIS

Title: An Experimental Therapeutics Study of Ixekizumab, a Monoclonal Antibody Against Interleukin 17A, on Anhedonia, Reward Circuit Function, and Blood Brain Barrier Physiology in Patients with Treatment- Resistant Depression

Study Description: The proposed study aims to establish the feasibility and safety of ixekizumab, a monoclonal antibody (mAb) against interleukin 17A (IL-17A), for patients with treatment-resistant depression (TRD). IL-17A is a pro-inflammatory cytokine that is elevated in subgroup of patients with TRD and we hypothesize that blocking IL-17A immune signaling with ixekizumab will improve depressive symptoms in adults with TRD. We also hypothesize that use of ixekizumab will be associated with changes in other symptom and functional domains (such as anhedonia), functional neuro-circuitry of brain's reward system, permeability of blood-brain barrier, and circulating immune factors. These hypotheses will be tested in the context of an open-label trial where up to n=20 patients with TRD will be treated with ixekizumab for 4 weeks and will undergo brain scans before and after the treatment period.

Objectives:

Primary Objective

The primary objective of the current study is to test the safety and feasibility of treatment with ixekizumab in patients with TRD. Open-label ixekizumab will be given at a dose of 160 mg (two 80 mg injections) at Week 0, followed by 80 mg at Weeks 2 and 4. Treatment-related adverse effects will be assessed, and drop-out rates will be calculated.

Secondary Objectives

The study will also test the effect of ixekizumab on (1) Changes in depression severity, as assessed by the Montgomery-Asberg Depression Rating Scale (MADRS) scores from baseline to the end of treatment (EOT) visit (6-week post-treatment); (2) response and remission rates with ixekizumab treatment; (3) change in measures of clinical symptoms, including anhedonia, anxiety, and functioning, and (4) change in blood-based inflammatory markers, including CRP and other immune markers.

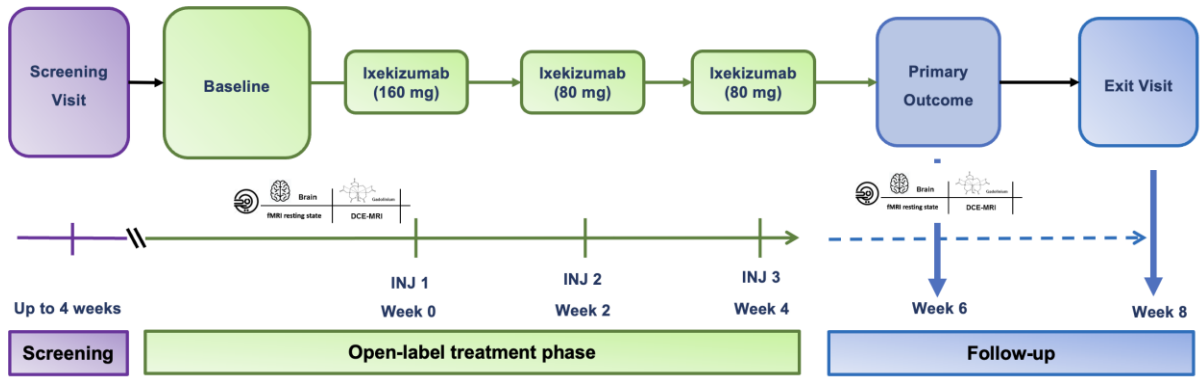
Exploratory Objectives

	The study will also test the effect of ixekizumab on changes in resting state functional connectivity (RSFC) of brain's reward circuit and change in blood brain barrier (BBB) permeability.
Endpoints:	Primary Endpoint: Completion of treatment measured as the receipt of all 3 ixekizumab injections (160 mg at week 0, and 80 mg at weeks 2 and 4. Secondary Endpoints: 1) Baseline-to-week-6 changes in depression severity as assessed by MADRS score; 2) Response and remission rates at week 6 (defined by ≥50% MADRS total score improvement and MADRS total score of ≤10, respectively); 3) Measures of global illness [Clinical Global Impression Severity (CGI-S) and Improvement (CGI-I) scale], overall depression [the Quick Inventory of Depressive Symptomatology Self-Report (QIDS-SR)], anxiety [Hamilton Anxiety Rating Scale (HAM-A)], suicidal ideation and attempt [the Columbia Suicide Severity Rating Scale (C-SSRS)], anhedonia [the Snaith Hamilton Pleasure Scale (SHAPS) and the Temporal Experience of Pleasure Scale (TEPS)], and symptoms associated with depression such as fatigue, sleep/wakefulness disturbances, anxiety, and irritability; 4) Baseline-to-week-6 changes in blood-based immune markers, like CRP and other immune markers Exploratory Endpoints: Baseline-to-week-6 changes in RSFC will be evaluated with resting-state functional magnetic resonance imaging (MRI) scans. Baseline-to-week-6 changes in BBB permeability will be evaluated with dynamic contrast enhanced MRI. Safety endpoints: 1) treatment-related adverse events; 2) Columbia-Suicide Severity Rating Scale at each time point
Study Population:	Up to n=20 adults (Aged 18-70 years; male and female) with a primary diagnosis of MDD who have failed two adequate trials of medication in the current depressive episode. Demographics reflective of the greater NYC population.
Phase:	Phase 2 study
Description of Sites/Facilities Enrolling Participants:	The human research will be conducted at the Icahn School of Medicine at Mount Sinai (ISMMS). Research will be conducted at the Depression and Anxiety Center for Discovery and Treatment, the Clinical Research Unit (CRU), the Infusion Suite of the Psychiatry Department of ISMMS. Functional MRI scans will be completed at the BioMedical Engineering and Imaging Institute (BMEII) in the Hess Center for Science and Medicine.
Description of Study Intervention:	One treatment condition in an open-label study design: ixekizumab 160 mg (two 80 mg subcutaneous injections) at Week 0, followed by 80 mg at Weeks 2 and 4.

Study Duration: The proposed study is to be completed in 24 months.

Participant Duration: The duration of an individual subject's participation in the study will be a maximum of 12 weeks including the screening (up to 4 weeks), study treatment visits (weeks 0, 2, and 4), post-treatment primary outcome visit (week 6), and study exit visit (week 8).

1.2 SCHEMA



1.3 SCHEDULE OF ACTIVITIES (SOA)

Study Phase	**Screening**	**Open-label Treatment Period (Bi-Weekly Study Visits)**			**Primary Outcome**	**Study Exit**
Study Visit	V0	V1	V2	V3	V4	V5
Week	-4 - 0	0	2	4	6	8
Screening						
Informed Consent	x					
Inclusion/Exclusion Criteria	x					
Demographics	x					
Medical History	x					
Family Psychiatric History	x					
ATRQ	x					
SCID-5-RV	x					
QIDS-C	x					
CGI-S	x					
Study drug administration						
Ixekizumab Treatment		x	x	x		
Safety assessments, concomitant medication and compliance monitoring						
Physical Exam	x				x	
Vital Signs	x	x	x	x	x	
Laboratory Tests	x				x	
U-Toxicology	x	x			x	
Pregnancy Test	x	x	x	x	x	
Adverse Events Monitoring	x	x	x	x	x	x
C-SSRS Past Month	x					
C-SSRS Since Last Visit		x	x	x	x	x
Clinical efficacy assessments						
MADRS		x	x	x	x	x
CGI-S	x	x	x	x	x	x
CGI-I		x	x	x	x	x
SHAPS		x	x	x	x	x
TEPS		x	x	x	x	x
QIDS-SR		x	x	x	x	x
HAM-A		x	x	x	x	x
Blood Markers						
Immune Markers		x			x	
Neuroimaging						
fMRI (RSFC)/DCE-MRI		x			x	

[*] Study visits 1-5 may occur 3 days before or after the scheduled visit.

	* Clinical laboratory tests include chemistry, complete blood count, liver function tests, TB test, and thyroid-stimulating hormone levels (see protocol for details). Abbreviations: ATRQ, Antidepressant Treatment Response Questionnaire; C-SSRS, Columbia-Suicide Severity Rating Scale; CGI-I, Clinical Global Impression-Improvement; CGI-S, Clinical Global Impression-Severity; fMRI, functional Magnetic Resonance Imaging; MADRS, Montgomery-Asberg Depression Rating Scale; QIDS-SR, Quick Inventory of Depressive Symptomatology-Self-Report; QIDS-C, Quick Inventory of Depressive Symptomatology-Clinician Rated; SHAPS, Snaith-Hamilton Pleasure Scale; TEPS, Temporal Experience of Pleasure Scale; HAM-A, Hamilton Anxiety Rating Scale

2 INTRODUCTION

2.1 STUDY RATIONALE

This study aims to establish the safety and feasibility of ixekizumab, a monoclonal antibody (mAb) against interleukin 17A (IL-17A), for patients with treatment-resistant depression (TRD) and evaluate its open-label effects on depression severity, other clinical symptoms, blood-based immune markers, BBB permeability, and functional brain reward circuitry. It is now clear that targeting the immune system holds enormous potential to identify novel therapeutic agents for severe and resistant forms of depression. Despite the growing evidence that immune dysregulation and depression are associated with one another, the promise of immune-modulating therapies for TRD has not been realized. In order to address this gap in knowledge, we propose to test an anti-IL-17A therapy for TRD. Based on the available clinical and preclinical work, IL-17A represents the most promising immune target for TRD that has yet to be tested in humans. If the goals of the project are achieved, the knowledge gained can be used to inform larger, placebo-controlled, well-powered trials to determine efficacy and the mechanism of action of ixekizumab for TRD. Ultimately, this can be transferred to clinical practice because marketed monoclonal antibodies, like ixekizumab, can be repurposed. The knowledge gained from this project will also stimulate new avenues in immune-based treatment discovery for depression.

2.2 BACKGROUND

2.2. A Need to develop novel antidepressant treatments.

There is a need for antidepressant medications with novel mechanisms of action, as currently available medications that target monoamine neurotransmission, are ineffective in about a third of patients with MDD.[1] There are few effective options for the subset of patients–those with treatment resistant depression (TRD)–who have failed multiple pharmacotherapy trials. The public health need of MDD in the United States is compounded by its high prevalence (1 in 5 adults during their lifetime), cost (over $200 billion per year), and burden (second highest cause of disability-adjusted life years).[2-4]

2.2. B Role of systemic inflammation in treatment-resistant depression (TRD)

Converging lines of evidence suggest that major depressive disorder (MDD) – and TRD in particular – is associated with immune dysregulation, which is thought to drive symptoms of anhedonia via dysfunction within the brain reward circuit.[5,6] Increased signaling of peripheral pro-inflammatory cytokines lead to anhedonia-like behavior in animals, and patients with depression (TRD in particular) are characterized by elevated levels of circulating cytokines, while systemic administration of pro-inflammatory cytokines triggers depressive symptoms and changes in brain responses to reward in humans.[5,7]

2.2. C Role of blood brain barrier (BBB) permeability in depression

Recent preclinical work from our team show that high levels of circulating immune factors gain access to the brain reward circuit to promote pro-depressive behaviors via increased permeability of the BBB.[8] Use of gadolinium-based contrast agents in dynamic contrast enhance magnetic resonance imaging (DCE-MRI) have facilitated quantification of BBB permeability in human for conditions ranging from multiple sclerosis and dementia.[9,10] A recent study of patients with bipolar disorder and healthy controls found that higher BBB leakage on DCE-MRI was associated with more severe depression and more chronic course of illness.[11]

2.2. D Role of interleukin 17 (IL-17) mediated immune signaling in depression

IL-17A is a highly pro-inflammatory cytokine produced by T helper 17 (Th17) cells that induces the production of numerous other inflammatory cytokines such as IL-6, IL-1β and TNF-α. Preclinical studies show that a) stress-induced pro-depressive behaviors are associated with increased levels of IL-17A or Th17cells in brain, b) peripheral infusion of IL-17A causes depression-like behaviors, and c) peripheral blockade of IL-17A with MABs directed against IL-17A is sufficient to reverse the pro-depressive phenotype.[12] Extending this work to humans, we and others have found that IL-17A levels are elevated in patients with depression versus healthy controls (HC), and higher levels of circulating IL-17A are associated with more severe depression and anhedonia.[13,14] Data from our group show that IL-17A is elevated specifically in patients with TRD ($F=5.66$; $df=2,68$; $p=0.005$), compared to HC or non-TRD forms of MDD. Critically, data show that IL-17A increases BBB permeability in preclinical models, providing a mechanism whereby peripheral IL-17A promotes depression in part by increasing BBB permeability and allowing circulating pro-inflammatory factors to gain access to mood-relevant brain circuits such as the VTA-NAc system.[5,15]

2.2. E Antidepressant effects of anti-IL-17A MABs.

The Food and Drug Administration (FDA) has approved three monoclonal antibodies (mAbs) targeting either IL-17A directly (ixekizumab and secukinumab) or its receptor (brodalumab). In phase 3 studies of psoriasis, both *ixekizumab and brodalumab were associated with marked improvement in depressive symptoms* which was not fully accounted for by improvement in psoriatic lesions. The evidence for antidepressant efficacy from clinical trial data for our study drug ixekizumab is particularly strong, wherein administration of ixekizumab every 2 weeks resulted in markedly higher improvement in overall depression versus placebo with a large effect size (Cohen's $d = 0.81$).[16]

Thus, anti-IL-17 mAbs may address the urgent need of developing novel antidepressants. This proposal will investigate ***ixekizumab*** for TRD during an 8 week open-label study. We are interested in evaluating ixekizumab and its effects on 1) depression severity; 2) other clinical symptoms; 3) the effect of IL-17A on brain function; 4) blood-based immune markers, like CRP and other immune markers; and changes in RSFC using functional magnetic resonance imaging (MRI) scans and changes in BBB permeability using dynamic contrast enhanced MRI .

2.3 Risk/Benefit Assessment

2.3.1 Known Potential Risks

2.3.1. A Risks of MRI Procedures

The functional MRI procedure is very safe and exposes subjects to minimal risk or discomfort. The MRI scanning involves the subject placing his/her head in a tube and some subjects may experience a feeling of claustrophobia while in the scanner. The subjects are always in voice contact with a researcher or technician and the scanning session can be terminated quickly if the subject so requests. Subjects with metal objects including surgical clips or metallic prostheses (such as an artificial hip or knee), or shrapnel (metal fragments) will be excluded. The risk to the subject in the event that there is susceptible metal in or on his or her body is significant and may include death. Every subject will be carefully screened using existing standard operating procedures of the Biomedical Engineering and Imaging Institute (BMEII) for the presence of susceptible metal.

MRI with Contrast: Gadolinium (MRI dye) is routinely used in medical imaging. As described in the MRI safety literature, it is a substance that is not normally absorbed into the tissues of the body. Once it is injected, it will stay in the blood stream and will be eliminated a few hours later in the urine. Some patients may have a reaction to this dye, which may be itching, nausea, breathing problems, vomiting, or a metallic taste. These reactions are very rare and usually occur in people who already have severe lung disease. There is a slight chance of an allergic reaction from the contrast which has a less than 1 in 300,000 chance that this will be severe.

A recent FDA warning has been issued that states subjects with kidney problems must not be given contrast because it may lead to debilitating and potential fatal disease that involves the skin, muscles, and internal organs. Patients with kidney disease can develop skin thickening that may prevent bending and extending joints, resulting in decreased mobility of joints. The chance of developing nephrogenic systemic fibrosis (NSF) is extremely low in individuals with normal kidney function. In addition, patients may experience scarring that has spread to other parts of the body such as the diaphragm, muscles in the thigh and lower abdomen, and the interior areas of lung.

2.3.1. B Risks of ixekizumab

The risks of ixekizumab are summarized below from the package insert provided by Ely Lilly (available at: https://uspl.lilly.com/taltz/taltz.html#pi) and are based on bi-weekly treatment in three placebo-controlled studies for adult patients with plaque psoriasis (n = 1167). Similar side effect profiles were observed in other studies that dosed ixekizumab once a month.

Most common (≥1%) adverse reactions associated with ixekizumab treatment are injection site reactions, upper respiratory tract infections, nausea, and tinea infections. Adverse reactions that occurred at rates less than 1% in the ixekizumab group and more frequently than in the placebo group during the 12-week induction period included rhinitis, oral candidiasis, urticaria, influenza, conjunctivitis, inflammatory bowel disease, angioedema, and abnormal laboratory values (complete blood count). Treatment with ixekizumab may worsen symptoms of active tuberculosis (TB) infection. Patients will be evaluated for TB infection prior to initiating treatment with ixekizumab.

In the pooled data, infections occurred in 27% of subjects treated with ixekizumab compared to 23% of subjects treated with placebo. Serious infections occurred in 0.4% of subjects treated with ixekizumab and in 0.4% of subjects treated with placebo.

There are no available data on ixekizumab use in pregnant women to inform any drug associated risks. Human IgG is known to cross the placental barrier; therefore, ixekizumab may be transmitted from the mother to the developing fetus. There are no data on the presence of ixekizumab in human milk, the effects on the breastfed infant, or the effects on milk production.

2.3.1. C Psychological Screening Risks

Answering questions related to mental health and past experiences may be stressful. Research interviews will be interrupted if subjects become distressed or object to answering questions. If in the judgment of the PI or study-affiliated psychiatrist, the patient has worsened to such a degree that further participation would put the patient at risk, then the subject will be discontinued from the study and provided appropriate clinical care. Also, at any point during the study any subject meeting DSM-5 criteria for a manic episode or psychosis based on clinical interview by the Study PI or study-affiliated psychiatrist will be discontinued and provided appropriate clinical care.

2.3.1. D Medical Screening Risks

Subjects might feel pain during blood draw or when IV is inserted, and can develop bruising, and rarely, infection. The sight of blood or insertion of IV cannula may also cause dizziness or brief loss of consciousness due to vasovagal response. Therefore, all blood draws will be conducted by trained personnel, using aseptic precautions, and in a setting where the subject is resting comfortably and the risk of fall is minimized.

2.3.1. E Privacy and Confidentiality Risks

As the research study involves collection of protected and sensitive health information, there is a potential risk of loss of confidentiality. This risk will be conveyed to subjects as part of informed consent and will be minimized by using SOPs at our Center which comply with Health Insurance Portability and Accountability Act rules.

2.3.1. F Financial risks

Subjects will not be charged for any study procedures. However, in case they experience an adverse effect and seek care, they or their insurance provider will be responsible for this clinical care. This risk will be informed to the subject as part of informed consent process.

2.3.2 KNOWN POTENTIAL BENEFITS

All study subjects will be informed that there may not be any potential benefit to them individually as a result of taking part in the study. All study subjects will receive without cost an extensive psychiatric and medical evaluation which may be of potential benefit for the subjects. Furthermore, subjects may experience improvement in symptoms, including anhedonia, with the study drug. Participating subjects may gain knowledge about the biological and psychological processes of depression, which they may find interesting and helpful in dealing with illness. No other direct benefits result from study participation.

2.3.3 Assessment of Potential Risks and Benefits

Data from clinical trials reveal that ixekizumab is well tolerated with minimal side effects during the first 12 weeks of biweekly dosing. The majority of side effects compared to placebo are injection site reactions. Ixekizumab may minimally increase the risk of infection, as the drug group had a higher rate of injections than placebo (27% vs. 23%). Overall, the cumulative risks stemming from ixekizumab treatment, psychological screening, medical screening, privacy and confidentiality, financial exposure are manageable, and the benefit of acquiring data on a novel medication for TRD outweighs the risks.

3 OBJECTIVES AND ENDPOINTS

OBJECTIVES	ENDPOINTS	JUSTIFICATION FOR ENDPOINTS
Primary		
The primary objective of the current study is to test the safety and feasibility of using ixekizumab in patients with TRD.	Completion of treatment measured as the receipt of all three Ixekizumab injections (160 mg at week 0, 80 mg at weeks 2 and 4). Drop-out rates will be calculated. The incidence and frequency of all anticipated and unanticipated serious and non-serious adverse events that occur between baseline and study exit visit will be determined. The effect of Ixekizumab will be assessed by changes in depression	Drop-out rates and treatment-related adverse events indicate the safety and tolerability of the study medication. The MADRS assessment is used as a gold-standard for assessing changes in depression

	severity as assessed with the Montgomery Asberg Depression Rating Scale (MADRS) scores from baseline to the end of treatment (EOT) visit (6-week post-treatment).	severity in clinical trials. The MADRS will be the primary endpoint of the study as we will use it to test the antidepressant effects of ixekizumab, our primary objective in the study.
Secondary		
The study will also test the effect of ixekizumab on (1) Changes in depression severity (as assessed by the MADRS from baseline to week 6 post-ixekizumab treatment); 2) response and remission rates with ixekizumab treatment; 3) change in measures of clinical symptoms, including anhedonia, anxiety, and functioning, and 4) change in blood-based inflammatory markers including CRP and other immune markers.	1) Baseline to week-6 changes in depression severity; 2) Response and remission rates at week 6 (defined as ≥50% MADRS total score improvement and MADRS total score of ≤10, respectively); 3) Measures of global illness [Clinical Global Impression Severity (CGI-S) and Improvement (CGI-I) scale], overall depression [the Quick Inventory of Depressive Symptomatology, Self-Report (QIDS-SR)], suicidal ideation and attempt [the Columbia Suicide Severity Rating Scale (C-SSRS)], anxiety [HAM-A], anhedonia [the Snaith Hamilton Pleasure Scale (SHAPS) and the Temporal Experience of Pleasure Scale (TEPS)], and symptoms associated with depression such as fatigue, sleep/wakefulness disturbances, anxiety, and irritability; 4) Baseline to week-6 changes in blood-based immune markers, including CRP and other immune markers	To collect pilot data on the potential antidepressant effects of ixekizumab and its effects on the measures taken in this study.

Exploratory		
The study will also test the effect of ixekizumab on changes in resting state functional connectivity (RSFC) of brain's reward circuit and change in blood brain barrier (BBB) permeability.	Baseline-to-week-6 changes in RSFC will be evaluated with resting-state functional magnetic resonance imaging (MRI) scans. Baseline-to-week-6 changes in BBB permeability will be evaluated with dynamic contrast enhanced MRI. \	BBB permeability and RSFC will also be assessed to determine the effects of ixekizumab on these measures.

4 STUDY DESIGN

4.1 Overall Design

We will enroll up to N=20 adults (aged 18-70 years) with TRD, defined as inadequate improvement with two or more antidepressants in the current episode. If patients are on antidepressant medications, they must be on a stable dose for >4 weeks prior to treatment. Patients will not be on any medication or nutritional supplement known to affect inflammatory status. Primary inflammatory disorder or an unstable medical condition will also be exclusionary. Subjects will receive 3 doses (160 mg at week 0, 80 mg at week 2, 80 mg at week 4) in total of ixekizumab injection, with dosing to occur every 2 weeks over a 6-week period. The primary clinical outcome will be safety and feasibility of ixekizumab as treatment for patients with TRD. The secondary clinical outcome will be change in MADRS from baseline to week 6, additional clinical outcomes, and changes in immune markers in the blood. Participants will undergo neuroimaging with DCE-MRI and rs-fMRI at baseline and at the primary outcome visit to measure the effect of treatment and change in depression severity on BBB permeability and reward circuit function.

4.2 Scientific Rationale for Study Design

We have selected to use an open-label study design for this study to determine the impact of ixekizumab in adults with TRD and no history of primary immune disorders. If successful, data from this study will be used to inform the design of larger randomized controlled clinical trials to further test the stated hypotheses.

4.3 Justification for Dose

The use of ixekizumab via subcutaneous route is consistent with the FDA-approved usage for other indications (psoriasis). The dosing regimen is based on the report by Griffiths et al.[16] which evaluated the impact of ixekizumab on depressive symptoms in three Phase 3 trials. In their report, which included

patients with psoriasis and moderate-to-severe depression, ixekizumab was started at 160 mg. Over the next 12 weeks, patients either received 80 mg of ixekizumab every 2 weeks (Q2W), 80 mg of ixekizumab every 4 weeks (Q4W), or placebo. Remission from depressive symptoms was more common in those who received ixekizumab Q2W (45.2%) as compared to ixekizumab Q4W (33.6%) or placebo (17.8%). Therefore, our planned use of ixekizumab with dose of 160 mg at baseline, and 80 mg at weeks 2 and 4 at a Q2W dosing interval is consistent with the dosing regimen found to be most effective in attaining remission of depressive symptoms in the Phase 3 clinical trials of patients with psoriasis and moderate-to-severe depression.

4.4 End of Study Definition

A participant is considered to have completed the study if he or she has completed all phases of the study including the last visit or the last scheduled procedure shown in the Schedule of Activities (SoA), Section 1.3. Alternatively, a participant may be considered exited from the study due to early withdrawal, drop-out, or screen failure. See Section 7 for more details.

5 STUDY POPULATION

5.1 Inclusion Criteria

1. Written informed consent (and assent when applicable) obtained from subject;
2. Ability for subject to comply with the requirements of the study as determined by the PI;
3. Men and women, age 18-70 years;
4. Participants must meet DSM-5 criteria for Major Depressive Disorder [MDD]) in a current major depressive episode (MDE) as determined by a study psychiatrist and confirmed using the Structured Clinical Interview for DSM-5 Research Version (SCID-5-RV);
5. Participants have had ≥ 2 adequate trials of antidepressants/augmentation strategies during current episode. (Refer to ATRQ Guidelines for Completion for guidelines on dose/duration required for a trial to be considered adequate.);
6. If patient is on antidepressant medication, they must be on a stable dose for >4 weeks prior to treatment;
7. Quick Inventory of Depressive Symptoms – Clinician Administered (QIDS-C) score ≥ 14
8. If female of childbearing potential, must agree to use of a medically accepted form of contraception, or else agree to abstinence until 6 months after the last dose of study drug.
9. Male patients, if heterosexually active with a partner who is female of childbearing potential, pregnant, or breastfeeding, must agree to barrier contraception for the treatment period and for at least 6 months after the last dose of study drug. Female partners of male participants must use at least one form of highly effective contraception starting at least one cycle prior to male patient study drug initiation until 6 months after the last dose of study drug.

5.2 Exclusion Criteria

1. A primary psychiatric diagnosis other than MDD as defined by DSM-5; [comorbid anxiety disorders (including agoraphobia, generalized anxiety disorder, social anxiety disorder and panic disorder) and posttraumatic stress disorder (PTSD) are allowed];

2. Has a history of schizophrenia or other psychotic disorder, major depressive disorder with psychotic features, or bipolar I or II disorder;

3. Diagnosis of a major neurocognitive disorder;

4. Meets criteria for a moderate or severe substance use disorder within the past 6 months, with the exception of nicotine use disorder;

5. The patient is pregnant or breastfeeding;

6. Any contraindication to MRI or gadolinium including claustrophobia, any trauma or surgery which may have left magnetic material in the body, magnetic implants or pacemakers, inability to lie still for 1 hour or more, any known allergy to gadolinium;*

7. Positive urine toxicology screen for illicit drugs at the time of screening;

8. Serious and imminent risk of self-harm or violence as determined by the PI;

9. History of suicide attempt in the past 2 years or screening CSSRS Ideation Score >2 in the past month;

10. Clinically significant abnormalities of laboratory tests or physical examination;

11. Any unstable medical illnesses including hepatic, renal, gastroenterological, respiratory, cardiovascular (including ischemic heart disease); endocrinologic, neurologic (including history of severe head injury), immunologic, or hematologic disease;

12. Presence of TB as assessed by Quantiferon Gold test at screening;

13. Concomitant treatments with other biologics or other immune-suppressant agents; PRN use of NSAIDs is permissible;

14. Female participants who are pregnant, breastfeeding, or may become pregnant, or unwilling to practice birth control during participation in the study or the 6 months following;

15. Presence of a condition or abnormality that in the opinion of the Investigator would compromise the safety of the patient or the quality of the data.

5.3 LIFESTYLE CONSIDERATIONS

Not applicable.

5.4 SCREEN FAILURES

If the patient fails to qualify for the study after signing the informed consent form, they will be considered a screen failure. Screen failures should be recorded on the electronic screening log along with the reason for disqualification. Patients who screen fail and are not randomized will be eligible to rescreen later. Patients should only be enrolled into the study once, therefore patients who have been randomized will not be eligible for future screening.

5.5 STRATEGIES FOR RECRUITMENT AND RETENTION

5.5.A Planned Recruitment Activities

The research strategy for the present study calls for the enrollment of up to N=20 adults (aged 18-70 years) with treatment-resistant depression over the two-year project period. Our total enrollment target of up to N=20 reflects individuals who are screened, eligible, and treated in the protocol. Anticipating screen fails, dropouts after enrollment and lost to follow-ups post-treatment, we plan to screen a total of N=40 individuals over the proposed 24-month recruitment period. This enrollment period provides for a highly feasible **~2 subjects screened and ~1 subject who complete baseline visits per month**, allowing for 2-month follow-up after the enrollment of last subject.

DAC has implemented a robust workflow for identifying and screening candidates for clinical research and employs direct-to-patient advertising through radio, newspaper, internet-based and other media outlets. A centralized programmatic intake process (under protocol **STUDY-10-00606; A Screening Protocol for Adult Patients with Mood and Anxiety Disorders, Chronic Medical Conditions, and Healthy Volunteers.)** is employed that includes a central phone line, a program-specific email, and a website featuring a web-based survey that patients can take to see if they qualify for studies. Individuals who contact DAC by completing the pre-screener online survey are automatically entered into a secure web-based database in order to be contacted by a member of the research staff for additional screening.

In addition to recruiting individuals through the DAC centralized programmatic intake process, recruitment is also anticipated from the following additional sources: (1) patients with MDD who are currently receiving clinical care from providers (psychiatrists and psychologists) in DAC; (2) the Mount Sinai Hospital Adult Outpatient Psychiatry Clinic; (3) self-referrals from patients who have completed other research protocols through DAC; (4) clinicians within and outside of the Mount Sinai Health System, including large departments of psychiatry at Mount Sinai Beth Israel, Mount Sinai West, Mount Sinai Morningside, and Mount Sinai South Nassau; and (5) self-referrals from individuals attending community outreach programs in the New York State and tristate area and from media advertisements. The PI will review subject enrollment at weekly project meetings with the study coordinator and team members, so that any deviation from the anticipated enrollment schedule will be detected quickly. All outreach and advertising activities will also be reviewed on a weekly basis.

5.5.B Engagement Strategies for Retention

The PI will hold weekly project meeting with the study coordinator in order to monitor enrollment and ensure that enrolled subjects are completing all study procedures as intended.

In terms of infrastructure, the data coordinating facilities at ISMMS provides a secure, validated, centralized electronic data management facility that is HIPAA and FDA-compliant (Research Electronic Data Capture, or REDCap). The REDCap system employed in this study includes several components, including a Screening Database and a Subject Tracking Database, in addition to the Study Database proper. As subjects complete visits, the date of the visit will be entered into the database. Missed visits will be coded for reason (such as ill, scheduling problem, inability to contact, etc.). If subjects terminate early, the date and reason will be coded in the Subject Tracking Database. On a weekly basis, the study research coordinator, who will also function as the data manager, will run a report on patient accrual

and completion status for review by the PI and the team. On an ongoing basis, the study data manager, the PI, and other team members will review the status of patient accrual and retention, as well as form completion, to identify and resolve any bottlenecks or problem areas that arise during the course of the study.

At DAC, the study coordinator will be the subject's point of contact during the study. Subjects will be provided with phone and email contact information for their coordinator, in addition to the contact information for the study PI.

5.5.C Strategies that will be used to ensure a diverse, representative study sample

The ISMMS is part of a large urban medical center, and benefits from a large and diverse catchment area. Located in the Manhattan borough of New York City, the racial and ethnic distribution of Mount Sinai's catchment area is 49% Hispanic, 38% African American, and 13% white non-Hispanics. The catchment area of the Mount Sinai's affiliated hospitals in Queens and the Bronx includes 22% Hispanics, 23% white non-Hispanics, 26% Asians and 24% African Americans. For the proposed clinical trial there are no special restrictions with regard to ethnicity or race. It is expected that the study sample will closely approximate the racial and ethnic composition of the greater catchment area of the Mount Sinai Health System.

5.5.D Potential recruitment/enrollment challenges and strategies that can be implemented in the event of enrollment shortfalls

Anticipated study related barriers may involve issues including staff workload and scheduling windows. This will be addressed by including coordinators who serve as back-up for the primary study coordinator responsible for the proposed project. These back-up staff members will allow the study team to screen and enroll subjects and complete study visits within subject's availability. All study staff will be trained by the PI and study procedures will be reviewed at weekly project meetings to ensure consistency of study procedures across both primary and back-up study personnel.

Some participant related barriers include appointment scheduling, transportation issues, and maintaining contact between study visits. To ensure that subjects can attend appointments, whenever possible, the visit schedule will include flexibility. To help address transportation issues, the study team will offer patients metro cards and other transportation resources available through DAC. The research staff will check voicemail and email daily to be able to respond to any patient contact and will reach out before each scheduled appointment to ensure that subjects are still available and able to complete the visit as scheduled.

6 STUDY INTERVENTION

6.1 STUDY INTERVENTION(S) ADMINISTRATION

6.1.1 STUDY INTERVENTION DESCRIPTION

Ixekizumab is a humanized immunoglobulin G subclass 4 (IgG4) monoclonal antibody (mAb) with neutralizing activity against IL-17A. Ixekizumab is produced by recombinant DNA technology in a recombinant mammalian cell line and purified using standard technology for bioprocessing. Ixekizumab is comprised of two identical light chain polypeptides of 219 amino acids each and two identical heavy

chain polypeptides of 445 amino acids each, and has a molecular weight of 146,158 Daltons for the protein backbone of the molecule.

Ixekizumab injection is a sterile, preservative free, clear and colorless to slightly yellow solution, for subcutaneous use available as 80 mg of ixekizumab in a 1 mL single-dose prefilled auto injector or a single-dose prefilled syringe. The prefilled auto injector and prefilled syringe each contain a 1 mL glass syringe with a fixed 27 gauge ½ inch needle. The ixekizumab 80 mg prefilled auto injector and prefilled syringe are manufactured to deliver 80 mg of ixekizumab. Each mL is composed of ixekizumab (80 mg); Citric Acid Anhydrous, USP (0.51 mg); Polysorbate 80, USP (0.3 mg); Sodium Chloride, USP (11.69 mg); Sodium Citrate Dihydrate, USP (5.11 mg); and Water for Injection, USP. Ixekizumab solution has a pH of 5.3 – 6.1.

Ixekizumab is a humanized IgG4 monoclonal antibody that selectively binds with the interleukin 17A (IL-17A) cytokine and inhibits its interaction with the IL-17 receptor. IL-17A is a naturally occurring cytokine that is involved in normal inflammatory and immune responses. Ixekizumab inhibits the release of proinflammatory cytokines and chemokines.

6.1.2 Dosing and Administration

Ixekizumab will be administered by subcutaneous injection. The dose is 160g (two 80 mg injections) at Week 0, followed by 80 mg at Weeks 2, and 4.

6.2 Preparation/Handling/Storage/Accountability

6.2.1 Acquisition and accountability

Ixekizumab will be obtained from commercial supplies by the Investigational Drug Service (IDS) at ISMMS.

6.2.2 Formulation, Appearance, Packaging, and Labeling

Ixekizumab injection for subcutaneous use manufactured by Eli Lilly.

Ixekizumab injection is a sterile, preservative free, clear and colorless to slightly yellow solution, for subcutaneous use available as 80 mg of ixekizumab in a 1 mL single-dose prefilled auto injector or a single-dose prefilled syringe. The prefilled auto injector and prefilled syringe each contain a 1 mL glass syringe with a fixed 27 gauge ½ inch needle. The ixekizumab 80 mg prefilled syringe are manufactured to deliver 80 mg of ixekizumab. Each mL is composed of ixekizumab (80 mg); Citric Acid Anhydrous, USP (0.51 mg); Polysorbate 80, USP (0.3 mg); Sodium Chloride, USP (11.69 mg); Sodium Citrate Dihydrate, USP (5.11 mg); and Water for Injection, USP. The ixekizumab solution has a pH of 5.3 – 6.1.

6.2.3 Product Storage and Stability

Ixekizumab injection is a sterile, preservative free, clear and colorless to slightly yellow solution available in a single-dose prefilled syringe to deliver 80 mg ixekizumab.

Ixekizumab is sterile and preservative-free. Discard any unused portion. Ixekizumab must be protected from light until use. Store refrigerated at 2°C to 8°C (36°F to 46°F). Do not freeze. Do not use Ixekizumab if it has been frozen. Do not shake. Discard the Ixekizumab single-dose syringe after use in a puncture-resistant container. Not made with natural rubber latex.

6.2.4 Preparation

Ixekizumab will be provided in a single dose prefilled syringe.

6.3 Measures to Minimize Bias: Randomization and Blinding

6.3.1 Randomization and Blinding

This study will be open-label with one treatment condition. Given that this study is open label with one treatment condition, participants cannot be blinded.

6.4 Study Intervention Compliance

An accurate and current accounting of the dispensing of study drug for each subject will be maintained on an ongoing basis by a member of the study site staff. The investigational site must keep an accurate inventory of study drug shipments received and the amount of study drug dispensed per patient on the Investigational Drug Accountability Record. At the end of the study, a full reconciliation of drug inventory will be performed. After a full reconciliation, any unused study drug will be destroyed by study personnel (if the site has the capability to do so, in accordance with applicable regulations) or sent to a designee for subsequent destruction. If no study drug remains, this will be indicated in the drug accountability log.

6.5 Concomitant Therapy

Participants will be permitted to remain on a stable dose of their psychotropic medications, per eligibility criteria. Concomitant treatments with other biologics or other immune-suppressant agents will be exclusionary. PRN use of NSAIDs is permissible.

6.5.1 Rescue Medicine

Not applicable.

7 STUDY INTERVENTION DISCONTINUATION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

7.1 Discontinuation of Study Intervention

A subject may be discontinued from study treatment at any time if the subject, the investigator, or the Sponsor feels that it is not in the subject's best interest to continue. The following is a list of possible reasons for study treatment discontinuation:

- Subject withdrawal of consent
- Subject is not compliant with study procedures

- Adverse event that in the opinion of the investigator would be in the best interest of the subject to discontinue study treatment
- Protocol violation requiring discontinuation of study treatment
- Lost to follow-up
- Sponsor request for early termination of study
- Positive pregnancy test (females)

Subjects who discontinue study treatment should come in for an early discontinuation visit as soon as possible. For patients who are lost to follow up, it may not be possible to complete early discontinuation procedures. For subjects who wish to withdraw consent, they will be asked to do so in writing following the completing of an early discontinuation visit. Some subjects may not agree and wish to withdraw consent immediately in which case these early discontinuation procedures may not occur. Reasonable attempts will be made by the investigator to provide a reason for subject discontinuation from study treatment.

7.2 Participant Discontinuation/Withdrawal from the Study

A subject may be withdrawn from the study at any time if the subject or the investigator-sponsor feels that it is not in the subject's best interest to continue. All subjects are free to withdraw from participation at any time, for any reason, specified or unspecified, and without prejudice.

Reasonable attempts will be made by the investigator to provide a reason for subject withdrawals. The reason for the subject's withdrawal from the study will be specified in the subject's source documents. As noted above, subjects who discontinue study treatment early (i.e., they withdraw prior to Visit 6) should have an early discontinuation visit.

7.3 Lost to Follow-Up

A participant will be considered lost to follow-up if he or she fails to return for any of the scheduled visits and is unable to be contacted by the study site staff.

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

- The site will attempt to contact the participant and reschedule the missed visit as well as counsel the participant on the importance of maintaining the assigned visit schedule and ascertain if the participant wishes to and/or should continue in the study.
- Before a participant is deemed lost to follow-up, the investigator or designee will make every effort to regain contact with the participant (where possible, 3 telephone calls and, if necessary, a certified letter to the participant's last known mailing address or local equivalent methods). These contact attempts should be documented in the participant's medical record or study file.
- Should the participant continue to be unreachable, he or she will be considered to have withdrawn from the study with a primary reason of lost to follow-up.

8 STUDY ASSESSMENTS AND PROCEDURES

8.1 Clinical Outcomes and safety Measures

Clinical Assessments

Demographics: Demographic information (date of birth, gender, race/ethnicity) will be recorded at screening.

Medical History: Relevant medical history, including history of current disease, other pertinent history, and information regarding underlying diseases will be recorded at screening.

Physical Examination: A complete physical examination will be performed by qualified staff (MD, NP, RN, or PA) at screening and primary outcome (week 6). New abnormal physical exam findings must be documented and will be followed by a physician or other qualified staff at the next scheduled visit. If a novel adverse event is found at study exit, subjects will be followed for up to 30 days or until resolved.

Vital Signs: Body temperature, blood pressure, pulse and respirations will be performed after resting for 5 minutes at screening, each of the three treatment visits, and at Visit 4, the primary outcome visit.

Structured Clinical Interview for DSM-5 (SCID-5). The Structured Clinical Interview for DSM-5 (First et al. 2015) is a semi-structured interview guide for making DSM-5 diagnoses. It includes an overview to obtain information about demographics, work history, chief complaint, and history of present illness, past history, treatment history, and current functioning. The main body of SCID-5 includes 9 modules that are designed to diagnose 51 mental illnesses in all. During the study screening period, the SCID-5 will be administered by a trained study team member who has completed SCID-5-RV training and who is familiar with the DSM-5 classification and diagnostic criteria. The current study will utilize the most comprehensive version of the SCID-5, the SCID-5-Research Version (RV), which contains more disorders than the Clinician Version and includes all of the relevant subtypes, severity, and course specifiers. An important feature of the SCID-5-RV is its customizability, allowing the instrument to be tailored to meet the requirements of a particular study. The SCID-5-RV comes in a standard "core" configuration that includes the disorders most researchers are likely to assess routinely for most studies, as well as in an “enhanced” configuration that includes a number of optional disorders, in addition to the disorders from the "core" configuration.

Antidepressant Treatment History Questionnaire (ATRQ): The ATRQ[17] is a self-rated scale used to determine treatment resistance in major depressive disorder (MDD) during the current episode. The ATRQ defines 6 weeks as an adequate duration of treatment and provides specific operational criteria for adequate dosage for each of the most commonly used antidepressant medications.

Montgomery-Asberg Depression Rating Scale (MADRS):[18] this is a 10
-item instrument used for the evaluation of depressive symptoms in adults and for the assessment of any changes to those symptoms. Each of the 10 items is rated on a scale of 0 to 6, with differing descriptors for each item. These individual item scores are added together to form a total score, which can range between 0 and 60 points. The MADRS provides a measure of the overall level of depression.

Snaith-Hamilton Pleasure Scale (SHAPS): The SHAPS[19] is a well-validated 14-item self-report questionnaire commonly used to assess anhedonia. Each item on the SHAPS is worded so that higher scores indicate greater pleasure capacity. A total score can be derived by summing the responses to each item. Items answered with “strongly agree” are coded as “1”, while a “strongly disagree” response was assigned a score of “4.” Total scores on the SHAPS can range from 14 to 56, with higher scores corresponding to higher levels of anhedonia.

Temporal Experience of Pleasure Scale (TEPS): The TEPS is an 18-item self-report measurement of anhedonia which consists of a series of statements that must be rated according to how accurate they are

for the individual.[20] The scale produces two sub-scores that differentiate the role of anticipatory pleasure ('wanting') from consummatory pleasure ('liking'). The anticipatory sub-score (TEPS-ant) is derived of 10 items while the consummatory sub-score (TEPS-cons) is derived from 8 items.

The Quick Inventory of Depressive Symptomatology, Self-Report (QIDS-SR): The QIDS-SR[21] is a 16-item self-rated instrument designed to assess the severity of depressive symptoms. The 16 items cover the nine symptom domains of major depression and are rated on a scale of 0-3. Total score ranges from 0 to 27, with ranges of 0-5 (normal), 6-10 (mild), 11-15 (moderate), 16-20 (moderate to severe), and 21+ (severe).

The Quick Inventory of Depressive Symptomatology, Clinician Rated (QIDS-C): The QIDS-C[21] is a 16-item instrument designed to assess the severity of depressive symptoms. The 16 items cover the nine symptom domains of major depression and are rated on a scale of 0-3. Total score ranges from 0 to 27, with ranges of 0-5 (normal), 6-10 (mild), 11-15 (moderate), 16-20 (moderate to severe), and 21+ (severe). This instrument will be used at the screening visit.

Clinical Global Impression - Severity (CGI-S): this is a widely administered clinician rated global measure of subject overall illness severity. Subjects are rated on a 1-7 scale where 1 corresponds to "Normal, Not at All Ill", 2 is "Borderline Mentally Ill", the anchor for 3 is "Mildly Ill", the anchor for 4 is "Moderately Ill", 5 is "Markedly Ill", 6 is "Severely Ill", and 7 is "Among the Most Extremely Ill Patients".

Clinical Global Impression - Improvement (CGI-I): this is a widely administered clinician rated global measure of the degree of improvement from the initial assessment in subject overall illness severity. Subjects are rated on a 1-7 scale where 1 corresponds to "Very Much Improved", 2 is "Much Improved", the anchor for 3 is "Minimally Improved", the anchor for 4 is "No Change", 5 is "Minimally Worse", 6 is "Much Worse", and 7 is "Very Much Worse".

Columbia-Suicide Severity Rating Scale (C-SSRS): The Columbia-Suicide Severity Rating Scale (C-SSRS)[22] is a comprehensive, semi-structured interview that uniquely measures the full spectrum of suicidality including passive and active suicidal ideation, suicidal intent as well as suicidal behaviors. Subjects will be assessed at every visit with the CSSRS by a qualified rater. Study participants will meet with a study physician at each study visit, which will include a review of the C-SSRS. Any subject with an increase in suicidality and all those subjects found to have a plan or intent will undergo thorough assessment by the study psychiatrist. If, at any point during the clinical trial, a participants will be assessed by the study physician with a suicidal ideation with any intent or plan, as measured by a C-SSRS score of greater than 2 during the past week and with either a CGI-S score ≥6 (indicating extreme illness severity) or a CGI-I score ≥ 6 (indicating an extreme worseness of the symptoms), the site PI, or his designee, will conduct a safety review to determine the appropriate course of action including whether acute intervention is needed and whether it is in the best interests of the subject to continue in the study.

Hamilton Anxiety Rating Scale (HAM-A): The Hamilton Anxiety Rating Scale (HAM-A) is a scale of assessments of anxiety states. The scale consists of 14 items, each defined by a series of symptoms, and measures both psychic anxiety (mental agitation and psychological distress) and somatic anxiety (physical complaints related to anxiety). Each item is scored on a scale of 0 (not present) to 4 (severe), with a total score range of 0–56, where <17 indicates mild severity, 18–24 mild to moderate severity and 25–30 moderate to severe.

Neuroimaging Measurements

Resting State fMRI (R-fMRI): Approximately 10 minutes worth of resting state fMRI data will be collected both pre- and post-treatment. Participants will be instructed to rest, eyes open and fixed on a central fixation cross, and to try to remain as still as possible. R-fMRI data will be examined primarily as an exploratory aim and additionally contributes to the common data element initiative. As there are a number of serious, potential confounds to the analysis of R-fMRI, advanced processing and analytic methods will be employed, including the use of robust artifact detection and removal.[23] We will conduct targeted analysis to explore whether treatment alters connectivity and network patterns with known abnormalities and that are specifically impacted by our treatment (e.g., fronto-striatal connectivity implicated in reward; see Heller et al., 2013[24]).

Clinical Laboratory Measurements

Hematology and Blood Chemistry Profile: Blood will be obtained at screening and primary outcome (week 6) and sent to each site's clinical hematology lab for a Hemoglobin, Hematocrit, Electrolytes, Metabolic Panel, Thyroid Function Tests, Hepatic and Renal Function Tests. At screening, blood will also be obtained to assess for existing TB infection.

Pregnancy Test: A urine pregnancy test will be obtained from female subjects who are of childbearing age prior to their participation in the study (screening visit), at Visit 1 (Week 0), Visit 2 (Week 2), Visit 3 (Week 4), and primary outcome Visit 4 (Week 6).

Urinalysis and Urine Toxicology: Urine will be obtained at the site's clinical laboratory for determination of color, urine specific gravity, osmolality, cellules, proteins and bacteria prior to their participation in the study (screening visit) and at study primary outcome (week 6). A urine toxicology screen for drugs of abuse will be performed at screening and on the day of both the pre-treatment and post-treatment scans (Visit 1 and 4).

Blood Immunological Measures: Blood will be obtained at baseline and primary outcome to be analyzed for inflammatory biomarkers and neurotrophic biomarkers. In brief, inflammatory biomarkers and neurotrophic factors will be measured using commercially available enzyme-linked immunosorbent assays or multiplex panels to analyze several biomarkers at once. Serum cytokine levels were chosen as biomarkers of inflammation because prior studies revealed a link between these biomarkers and depression.

8.2 EVALUATIONS BY VISIT

SCREENING VISIT (V0, WEEK -4 – 0)

1. Review the study with the subject and obtain written informed consent and HIPAA authorization.
2. Review inclusion and exclusion criteria.
3. Assign the subject a unique screening number.
4. Record demographics data.
5. Record medical, psychiatric, family, medication and treatment history, diagnosis date, and prior treatments.
6. Record concomitant medications.
7. Perform self and clinician administered ratings scale:

- SCID-5-RV
- CGI-S
- CSSR-S – Past month
- QIDS-C

8. Perform a complete physical examination.
9. Perform and record vital signs.
10. Urine for urinalysis, urine toxicology and pregnancy test (female subjects who are of childbearing age).
11. Collect blood for clinical laboratory tests.
12. Schedule subject for Baseline (Visit 1) within 4 weeks of Screening.

Note: The screening visit may occur on up to three separate days. The whole procedures will be completed before the treatment day and within four weeks of the first screening visit. Screening measures may be completed under a separate screening protocol at each site. As long as the assessments are completed within four weeks of the signing of consent for this protocol, the screening measures will not be repeated. Subjects who are deemed screen fails may be re-screened at a later date, based on PI discretion. If re-screened, subjects will repeat all screening procedures.

A portion of this visit, including clinician administered rating scales, may be completed remotely via a HIPAA-compliant virtual platform.

BASELINE (V1, WEEK 0)

1. Obtain interval medical history.
2. Record any AEs.
3. Record changes to concomitant medications.
4. Collect urine for urine toxicology and pregnancy test (female subjects who are of childbearing age).
5. Record vital signs
6. Perform self and clinician administered ratings scale:
    - MADRS
    - CSSRS-Since Last Visit
    - HAM-A
    - CGI-S
    - CGI-I
    - SHAPS
    - TEPS
    - QIDS-SR
7. Neuroimaging procedures.
8. Collect blood immunological measures.
9. Dispense study medication.

Note: The Baseline Visit should take place within four-weeks of the date that the subject signed consent for this study. The Baseline Visit may occur on up to two separate days within 5 business days of each other. The neuroimaging procedures must be completed before the subject is randomized and receives

the study drug.

VISIT 2 (WEEK 2)

1. Obtain interval medical history.
2. Record vital signs
3. Record any AEs.
4. Record changes to concomitant medications.
5. Collect urine for a pregnancy test, (female subjects who are of childbearing age)
6. Perform self and clinician administered ratings scale:
   - MADRS
   - CSSRS-Since Last Visit
   - HAM-A
   - CGI-S
   - CGI-I
   - SHAPS
   - TEPS
   - QIDS-SR
7. Dispense study medication.

VISIT 3 (WEEK 4)

1. Obtain interval medical history.
2. Record any AEs.
3. Record changes to concomitant medications.
4. Record vital signs
5. Collect urine for a pregnancy test, (female subjects who are of childbearing age)
6. Perform self and clinician administered ratings scale:
   - MADRS
   - CSSRS-Since Last Visit
   - HAM-A
   - CGI-S
   - CGI-I
   - SHAPS
   - TEPS
   - QIDS-SR
7. Dispense study medication.

VISIT 4 (WEEK 6)

1. Obtain interval medical history.
2. Record any AEs.
3. Record changes to concomitant medications.
4. Perform self and clinician administered ratings scale:
   - MADRS
   - CSSRS-Since Last Visit
   - HAM-A

- CGI-S
- CGI-I
- SHAPS
- TEPS
- QIDS-SR

5. Perform a complete physical examination.
6. Perform and record vital signs.
7. Urine for urinalysis, urine toxicology, and pregnancy test (female subjects who are of childbearing age).
8. Collect blood for clinical laboratory tests and immunological measures.
9. Neuroimaging Procedures.

STUDY EXIT - VISIT 5 (WEEK 8)

10. Obtain interval medical history.
11. Record any AEs.
12. Record changes to concomitant medications.
13. Perform self and clinician administered ratings scale:
    - MADRS
    - CSSRS-Since Last Visit
    - HAM-A
    - CGI-S
    - CGI-I
    - SHAPS
    - TEPS
    - QIDS-SR

Note: This visit, including clinician administered rating scales, may be completed remotely via a HIPAA-compliant virtual platform.

Note: Study Visits 2, 3, and 4 are due to take place 2, 4, and 6 weeks after the baseline visit 1 (week 0) respectively. To allow for scheduling feasibility, Visits 1-5 can take place within 5 days before or after the scheduled visit.

8.3 ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

8.3.1 DEFINITION OF ADVERSE EVENTS (AE)

An Adverse Event (AE) is any untoward medical occurrence in a study subject administered as an investigational product and that does not necessarily have a causal relationship with this treatment.

An AE therefore can be any unfavorable and unintended sign (including laboratory finding), symptom or disease temporally associated with participation in an investigational study, whether or not considered drug-related. In addition to new events, any increase in the severity or frequency of a pre-existing condition that occurs after the subject signs a consent form for participation is considered an AE. This includes any side effect, injury, toxicity, or sensitivity reaction.

Any condition, laboratory abnormality, or physical finding with an onset date prior to the subject signing consent for study participation is considered to be pre-existing in nature and part of the subject's medical history and will not be recorded as AEs.

8.3.2 Definition of Serious Adverse Events (SAE)

An SAE is defined as any AE occurring at any dose that results in any of the following outcomes:

· Death

· Life threatening experience defined as any adverse experience that places the subject, in the view of the treating physician, at immediate risk of death at the time of occurrence; i.e, it does not include a reaction that, had it occurred in a more severe form, might have caused death.

· Requires inpatient hospitalization or prolongation of an existing hospitalization (except scheduled hospitalizations for non-acute, unrelated cause such as an elective surgery)

· Results in persistent or significant disability/incapacity

· Is a congenital anomaly/birth defect in the offspring of an exposed subject

· Important medical events that may not result in death, be life threatening, or require hospitalization, may be considered an SAE when, based upon appropriate medical judgment, it jeopardizes the subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition.

· Any death occurring within 30 days of the subject receiving study drug, regardless of the subject having discontinued from the protocol, must be reported as an SAE.

8.3.3 Classification of an Adverse Event

Severity of Event

For adverse events (AEs) not included in the protocol defined grading system, the following guidelines will be used to describe severity.

- **Mild** – Events require minimal or no treatment and do not interfere with the participant's daily activities.
- **Moderate** – Events result in a low level of inconvenience or concern with the therapeutic measures. Moderate events may cause some interference with functioning.
- **Severe** – Events interrupt a participant's usual daily activity and may require systemic drug therapy or other treatment. Severe events are usually potentially life-threatening or incapacitating. Of note, the term "severe" does not necessarily equate to "serious".]

Relationship to Study INTERVENTION

All adverse events (AEs) must have their relationship to study intervention assessed by the clinician who examines and evaluates the participant based on temporal relationship and his/her clinical judgment. The degree of certainty about causality will be graded using the categories below. In a clinical trial, the study product must always be suspect.

- **Definitely Related** – There is clear evidence to suggest a causal relationship, and other possible contributing factors can be ruled out. The clinical event, including an abnormal laboratory test result, occurs in a plausible time relationship to study intervention administration and cannot be explained by concurrent disease or other drugs or chemicals. The response to withdrawal of the study intervention (dechallenge) should be clinically plausible. The event must be pharmacologically or phenomenologically definitive, with use of a satisfactory rechallenge procedure if necessary.
- **Probably Related** – There is evidence to suggest a causal relationship, and the influence of other factors is unlikely. The clinical event, including an abnormal laboratory test result, occurs within a reasonable time after administration of the study intervention, is unlikely to be attributed to concurrent disease or other drugs or chemicals, and follows a clinically reasonable response on withdrawal (dechallenge). Rechallenge information is not required to fulfill this definition.
- **Potentially Related** – There is some evidence to suggest a causal relationship (e.g., the event occurred within a reasonable time after administration of the trial medication). However, other factors may have contributed to the event (e.g., the participant's clinical condition, other concomitant events). Although an AE may rate only as "possibly related" soon after discovery, it can be flagged as requiring more information and later be upgraded to "probably related" or "definitely related", as appropriate.
- **Unlikely to be related** – A clinical event, including an abnormal laboratory test result, whose temporal relationship to study intervention administration makes a causal relationship improbable (e.g., the event did not occur within a reasonable time after administration of the study intervention) and in which other drugs or chemicals or underlying disease provides plausible explanations (e.g., the participant's clinical condition, other concomitant treatments).
- **Not Related** – The AE is completely independent of study intervention administration, and/or evidence exists that the event is definitely related to another etiology. There must be an alternative, definitive etiology documented by the clinician.]

EXPECTEDNESS

The Principal Investigator (PI) will be responsible for determining whether an adverse event (AE) is expected or unexpected. An AE will be considered unexpected if the nature, severity, or frequency of the event is not consistent with the risk information previously described for the study intervention.]

8.3.4 TIME PERIOD AND FREQUENCY FOR EVENT ASSESSMENT AND FOLLOW-UP

The occurrence of an adverse event (AE) or serious adverse event (SAE) may come to the attention of study personnel during study visits and interviews of a study participant presenting for medical care, or upon review by a study monitor.

All AEs including local and systemic reactions not meeting the criteria for SAEs will be captured on the appropriate case report form (CRF). Information to be collected includes event description, time of onset, clinician's assessment of severity, relationship to study product (assessed only by those with the training and authority to make a diagnosis), and time of resolution/stabilization of the event. All AEs occurring while on study must be documented appropriately regardless of relationship. All AEs will be followed to adequate resolution.

Any medical condition that is present at the time that the participant is screened will be considered as baseline and not reported as an AE. However, if the study participant's condition deteriorates at any time during the study, it will be recorded as an AE.

Changes in the severity of an AE will be documented to allow an assessment of the duration of the event at each level of severity to be performed. AEs characterized as intermittent require documentation of onset and duration of each episode.

The Principal Investigator will record all reportable events with start dates occurring any time after informed consent is obtained until 7 (for non-serious AEs) or 30 days (for SAEs) after the last day of study participation. At each study visit, the investigator will inquire about the occurrence of AE/SAEs since the last visit. Events will be followed for outcome information until resolution or stabilization.

8.3.5 Adverse Event Reporting

To IRB, PI, Sponsor, and DSMB

Any AE must be recorded on the appropriate CRF/eCRF. All AE/SAEs that are considered related to study drugs must be followed to resolution or stabilization if improvement is not expected. AE/SAEs that completely resolve and then recur should be recorded as a new AE/SAE. AE/SAEs that are considered related to study drug and continuing at 30 days post-last dose should have a comment in the source documents by the site PI that the event has stabilized or is not expected to improve.

Any SAE occurring during the study period (beginning with informed consent and lasting until 30 days after the last dose of study drug) must be immediately reported no later than 3 business days after learning of an SAE to the Study PI.

All AE/SAEs with an onset date after the subject signs consent for study participation must be reported to the IRB at the time of annual renewal. Details of the event must include seriousness, severity, expectedness, relationship to study drug, duration, action taken, and outcome.

All AE/SAEs that are drug-related and unexpected (not listed as treatment-related in the current Investigator's Brochure) must be reported to the PI and IRB as detailed below in Section 11.5 "Unanticipated Problem Reporting".

To FDA

A report on the MedWatch 3500A form must be sent to the FDA when the event is:

(1) serious unexpected suspected reaction (the investigator judges there is evidence to suggest a causal relationship);
(2) findings from other clinical, animal, or in-vitro studies that suggest significant human risk and
(3) a clinically important increase in the rate of a serious suspected adverse reaction no later than 15 days after determining that the information qualifies for reporting.

The relevant health authorities will be notified of any unexpected serious adverse reactions (SUSARs) within the required reporting timelines (within 7 calendar days for fatal and life-threatening SUSARs, or 15 calendar days for all other SUSARs).

8.3.6 Serious Adverse Event Reporting

The study clinician will immediately report to the sponsor any serious adverse event, whether or not considered study intervention related, including those listed in the protocol or package insert and must include an assessment of whether there is a reasonable possibility that the study intervention caused the event. Study endpoints that are serious adverse events (e.g., all-cause mortality) must be reported in accordance with the protocol unless there is evidence suggesting a causal relationship between the study intervention and the event (e.g., death from anaphylaxis). In that case, the investigator must immediately report the event to the sponsor.

All serious adverse events (SAEs) will be followed until satisfactory resolution or until the site investigator deems the event to be chronic or the participant is stable.

The study sponsor will be responsible for notifying the Food and Drug Administration (FDA) of any unexpected fatal or life-threatening suspected adverse reaction as soon as possible, but in no case later than 7 calendar days after the sponsor's initial receipt of the information. In addition, the sponsor must notify FDA and all participating investigators in an Investigational New Drug (IND) safety report of potential serious risks, from clinical trials or any other source, as soon as possible, but in no case later than 15 calendar days after the sponsor determines that the information qualifies for reporting.

8.3.7 Reporting Events to Participants

Participants will be notified of any abnormal clinical lab findings and directed to appropriate medical care. In the event that a participant’s pregnancy test returns positive, the participant will be notified immediately and exited from the study.

8.3.8 Events of Special Interest

Not applicable.

8.3.9 Reporting of Pregnancy

Subjects will be asked to immediately inform the study team and their doctor if any of the following occur:

- They become pregnant while taking the study drug
- They miss their menstrual period, or experiences unusual menstrual bleeding
- They stop using birth control
- They think, FOR ANY REASON, that they may be pregnant

Pregnancies occurring while the subject is participating in the study or within 30 days after the subject’s administration of study drug are considered expedited reportable events. The pregnancy will be considered an immediately reportable event and will be reported to the study PI within 5 business days of the site PI becoming aware of the event.

8.4 Unanticipated Problems

8.4.1 Definition of Unanticipated Problems (UP)

The Office for Human Research Protections (OHRP) considers unanticipated problems involving risks to participants or others to include, in general, any incident, experience, or outcome that meets **all** of the following criteria:

- Unexpected in terms of nature, severity, or frequency given (a) the research procedures that are described in the protocol-related documents, such as the Institutional Review Board (IRB)-approved research protocol and informed consent document; and (b) the characteristics of the participant population being studied;
- Related or possibly related to participation in the research ("possibly related" means there is a reasonable possibility that the incident, experience, or outcome may have been caused by the procedures involved in the research); and
- Suggests that the research places participants or others at a greater risk of harm (including physical, psychological, economic, or social harm) than was previously known or recognized.

8.4.2 Unanticipated Problem Reporting

The investigator will report unanticipated problems (UPs) to the reviewing Institutional Review Board (IRB) and to the Data Coordinating Center (DCC)/lead principal investigator (PI). The UP report will include the following information:

- Protocol identifying information: protocol title and number, PI's name, and the IRB project number;
- A detailed description of the event, incident, experience, or outcome;
- An explanation of the basis for determining that the event, incident, experience, or outcome represents an UP;
- A description of any changes to the protocol or other corrective actions that have been taken or are proposed in response to the UP.

8.4.3 Reporting Unanticipated Problems to Participants

Not applicable.

9 STATISTICAL CONSIDERATIONS

9.1 Statistical Hypotheses

- Primary Efficacy Endpoint(s):

The primary efficacy endpoint is the completion of treatment measured as the receipt of all three Ixekizumab (160 mg at week 0, 80 mg at weeks 2 and 4) to assess the safety and feasibility of ixekizumab in patients with TRD. The analysis of the primary endpoint for this study will be descriptive and no formal hypothesis testing with be conducted.

- Secondary Efficacy Endpoint(s):

Secondary efficacy endpoints include 1) Baseline-to-week-6 changes in depression severity, as measured by MADRS; 2) Response and remission rates at week 6; 3) changes in other clinical symptoms ;4)

Baseline-to-week-6 changes in RSFC as evaluated by fMRI scans and changes in BBB permeability; 5) Baseline-to-week-6 changes in blood-based immune markers, including CRP and other immune markers; 6) the relationship between baseline-to-week-6 changes in blood-based immune markers and RSFC with changes in depression severity scores. Analyses of secondary efficacy endpoints will be considered exploratory and hypothesis-generating. Therefore, no formal hypothesis testing will be conducted.

9.2 Sample Size Determination

In this study, up to 20 patients will be enrolled. We anticipate 80% of more of the sample will complete treatment (<20% early drop out) and the treatment will be well-tolerated with no treatment-related adverse effects. A sample size of 20 patients will produce a two-sided 95% confidence interval around the proportion who completed treatment with a width equal to 0.35 when the sample proportion is 0.8. Precision was estimated using PASS 2019 (26).

9.3 Populations for Analyses

Intent-to-Treat (ITT) Population

The ITT population will consist of all enrolled subjects regardless of whether or not they actually received ixekizumab. This sample will be used for summaries and analyses of the primary endpoint and the secondary endpoints.

Safety Analysis set

The safety analysis set will include all enrolled participants who were given at least one dose of ixekizumab, and on whom any post-dose data are available and are classified according to the treatment actually received. The safety analysis set will be used to assess safety and tolerability variables.

9.4 Statistical Analyses

9.4.1 General Approach

Continuous variables will be summarized using the following descriptive statistics: number of non-missing values, means, standard deviations, medians, interquartile range, maximum, and minimum. Categorical variables will be summarized using number of non-missing values, counts and percentages.

Rates of events will be calculated as the ratio of the total number of events recorded divided by the total patient-time. Total patient-time will be calculated by summing the time (in study time units, e.g., days or months) that patients were at risk for a specific event from the reference time point until either study exit or the end of the time period of interest. Rates and their 95% confidence intervals will be reported.

Time-to-event variables will be summarized using the Kaplan-Meier method.

For any variable measured at multiple points in time, change from baseline will be calculated as the difference between the value of the variable at a specific point in time (e.g. 6 weeks) minus the baseline value. Relative change from baseline will be calculated as the value of a parameter at a specific point in

time minus the baseline value of the parameter divided by the baseline value of the parameter. Percent change will be calculated as the relative change multiplied by 100.

All hypothesis testing will be conducted at the 0.05 two-sided significance level unless otherwise specified. P-values will be rounded to three decimal places. P-values less than 0.001 will be reported as <0.001 in tables. P-values greater than 0.999 will be reported as >0.999.

Should any of the statistical methods proposed prove unsuitable during data analysis, more appropriate methods will be used. These include data transformation (for example to a logarithmic scale) to satisfy model assumptions such as normally distributed residuals with constant variance, the application of non-parametric techniques or the use of a different link function or modeling technique. The SAP will be updated with the methods used and the justification for the change prior to data set and database lock.

9.4.2 Analysis of the Primary Efficacy Endpoint(s)

The primary efficacy endpoint is the completion of treatment measured by the receipt of all three ixekizumab injections (160 mg at week 0, 80 mg at weeks 2 and 4) to assess the safety and feasibility of ixekizumab in patients with TRD. The analysis of the primary endpoint for this study will be descriptive. The proportion of enrolled patients that complete treatment and the corresponding 95% confidence interval will be reported.

9.4.3 Analysis of the Secondary Endpoint(s)

1) Baseline-to-week-6 changes in depression severity, measured by MADRS scores

MADRS scores will be reported descriptively at each time point using the number observed, mean, standard deviation, median, and interquartile range. Individual patient trajectories will be plotted over time to explore patterns in treatment response. Mean change in MADRS scores from baseline and corresponding 95% confidence intervals will also be reported at each time point.

2) Response and remission rates at week 6

The number and proportion of patients who respond ot treatment and who remain in remission will be reported at each time point.

3) Baseline-to-week 6 changes in other clinical symptoms as measured by CGI-S, CGI-I, QIDS-SR, C-SSRS, SHAPS, TEPS, and HAM-A.

4) Baseline-to-week-6 changes in RSFC as evaluated by fMRI scans and changes in BBB permeability

RSFC and BBB permeability will be analyzed in the same manner described above for MADRS scores.

5) Baseline-to-week-6 changes in blood-based immune markers, including CRP and other immune markers

Biomarkers will be analyzed in the same manner described for MADRS scores above.

6) the relationship between baseline-to-week-6 changes in blood-based immune markers and RSFC and BBB permeability with changes in depression severity scores.

We will explore whether improving depressive symptoms correlate with changes in blood-based immune markers and RSFC and BBB permeability. MADRS scores will be plotted with biomarkers, RSFC, and BBB over time to evaluate trends descriptively. Fixed and mixed effects regression models may also be explored to estimate associations and 95% confidence intervals.

9.4.4 Safety Analyses

All safety analyses will be conducted in the safety population.

Safety and tolerability data will also be summarized descriptively. Adverse events will be categorized using the Medical Dictionary for Regulatory Activities (MedDRA) system organ class and preferred terms. For each group, number and rates of events and number and percent of patients will be tabulated by preferred term and system organ class.

All laboratory test results, vital signs, weight, and BMI will be summarized using descriptive statistics at each visit for raw numbers and change from baseline. Suicidality measures based on the C-SSRS will be summarized for each treatment group using descriptive statistics at each assessment.

9.4.5 Baseline Descriptive Statistics

Baseline characteristics will be reported descriptively. Continuous variables will be summarized using the number of non-missing values, means, standard deviations, medians, interquartile range, maximum, and minimum. Categorical variables will be summarized using number of non-missing values, counts and percentages.

9.4.6 Planned Interim Analyses

No interim analyses are planned.

9.4.7 Sub-Group Analyses

No sub-group analyses are planned.

9.4.8 Tabulation of Individual participant Data

Per NIMH policy, we will submit a final deidentified data set, listed by measure and time point, as part of the data sharing plan.

9.4.9 Exploratory Analyses

Treatment effects on R-fMRI data will be explored. It is hypothesized that Ixekizumab will result in neuroimaging changes in resting state functional connectivity, and blood brain barrier permeability, evaluated with magnetic resonance imaging (MRI) scans. As there are a number of serious, potential confounders to the analysis of R-fMRI, advanced processing and analytic methods will be employed, including the use of robust artifact detection and removal (Pruim et al., 2015). We will conduct targeted analysis to explore whether treatment alters connectivity and network patterns with known abnormalities and that are specifically impacted by our treatment (e.g., fronto-striatal connectivity implicated in reward; see Heller et al., 2013).

10 SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1 Regulatory, Ethical, and Study Oversight Considerations

10.1.1 Informed Consent Process

10.1.1.1 Consent/assent and Other Informational Documents Provided to participants

Consent forms describing in detail the study intervention, study procedures, and risks are given to the participant and written documentation of informed consent is required prior to starting intervention/administering study intervention. The consent materials are submitted with this protocol.

10.1.1.2 Consent Procedures and Documentation

Informed consent will be obtained in accordance with the Declaration of Helsinki, ICH GCP, US Code of Federal Regulations for Protection of Human Subjects (21 CFR 50.25[a,b], CFR 50.27, and CFR Part 56, Subpart A), the Health Insurance Portability and Accountability Act (HIPAA), and local regulations.

The Investigator-sponsor will prepare the informed consent form, assent and HIPAA authorization. The consent form generated by the Investigator must be approved by the IRB. The written consent document will embody the elements of informed consent as described in the International Conference on Harmonisation and will also comply with local regulations.

A properly executed, written, informed consent will be obtained from each subject prior to entering the subject into the trial. Information should be given in both oral and written form and subjects must be given ample opportunity to inquire about details of the study. If appropriate and required by the local IRB, assent from the subject will also be obtained. If a subject is unable to sign the informed consent form (ICF) and the HIPAA authorization, a legal representative may sign for the subject. A copy of the signed consent form (and assent) will be given to the subject and the original will be maintained with the subject's records.

10.1.2 Study Discontinuation and Closure

This study may be temporarily suspended or prematurely terminated if there is sufficient reasonable cause. Written notification, documenting the reason for study suspension or termination, will be

provided by the suspending or terminating party to study participants, investigator, funding agency, the Investigational New Drug (IND) sponsor and regulatory authorities. If the study is prematurely terminated or suspended, the Principal Investigator (PI) will promptly inform study participants, the Institutional Review Board (IRB), and sponsor and will provide the reason(s) for the termination or suspension. Study participants will be contacted, as applicable, and be informed of changes to study visit schedule.

Circumstances that may warrant termination or suspension include, but are not limited to:

- Determination of unexpected, significant, or unacceptable risk to participants
- Demonstration of efficacy that would warrant stopping
- Insufficient compliance to protocol requirements
- Data that are not sufficiently complete and/or evaluable
- Determination that the primary endpoint has been met
- Determination of futility

Study may resume once concerns about safety, protocol compliance, and data quality are addressed, and satisfy the sponsor, IRB and/or Food and Drug Administration (FDA).

10.1.3 Confidentiality and Privacy

Participant confidentiality and privacy is strictly held in trust by the participating investigators, their staff, and the sponsor(s) and their interventions. This confidentiality is extended to cover testing of biological samples and genetic tests in addition to the clinical information relating to participants. Therefore, the study protocol, documentation, data, and all other information generated will be held in strict confidence. No information concerning the study or the data will be released to any unauthorized third party without prior written approval of the sponsor.

All research activities will be conducted in as private a setting as possible.

The study monitor, other authorized representatives of the sponsor, representatives of the Institutional Review Board (IRB), regulatory agencies or pharmaceutical company supplying study product may inspect all documents and records required to be maintained by the investigator, including but not limited to, medical records (office, clinic, or hospital) and pharmacy records for the participants in this study. The clinical study site will permit access to such records.

The study participant's contact information will be securely stored at each clinical site for internal use during the study. At the end of the study, all records will continue to be kept in a secure location for as long a period as dictated by the reviewing IRB, Institutional policies, or sponsor requirements.

Certificate of Confidentiality

To further protect the privacy of study participants, a Certificate of Confidentiality will be issued by the National Institutes of Health (NIH). This certificate protects identifiable research information from forced disclosure. It allows the investigator and others who have access to research records to refuse to disclose identifying information on research participation in any civil, criminal, administrative, legislative,

or other proceeding, whether at the federal, state, or local level. By protecting researchers and institutions from being compelled to disclose information that would identify research participants, Certificates of Confidentiality help achieve the research objectives and promote participation in studies by helping assure confidentiality and privacy to participants.

10.1.4 Future Use of Stored Specimens and Data

Data collected for this study will be analyzed and stored on site.

During the conduct of the study, an individual participant can choose to withdraw consent to have biological specimens stored for future research. However, withdrawal of consent with regards to biosample storage may not be possible after the study is completed.

10.1.5 Key Roles and Study Governance

The Principal Investigator, Dr. James Murrough, will serve as the medical monitor for this single-site study.

Principal Investigator	**Medical Monitor**
James Murrough, MD, PhD	*James Murrough, MD, PhD*
Icahn School of Medicine	*Icahn School of Medicine*
1399 Park Avenue	*1399 Park Avenue*
212-585-4640	*212-585-4640*
james.murrough@mssm.edu	*James.murrough@mssm.edu*

10.1.6 Safety Oversight

Safety oversight will be under the direction of an Independent Safety Monitor, Dr. Sanjay Matthews. Members of the study team will meet with the ISM at least semiannually to assess safety and efficacy data on each arm of the study.

10.1.7 Clinical Monitoring

Clinical site monitoring is conducted to ensure that the rights and well-being of trial participants are protected, that the reported trial data are accurate, complete, and verifiable, and that the conduct of

the trial is in compliance with the currently approved protocol/amendment(s), with International Conference on Harmonisation Good Clinical Practice (ICH GCP), and with applicable regulatory requirement(s).

- The site PI, Dr. James Murrough, and the study team will conduct clinical monitoring on-site for the duration of the study.
- Study coordinators will comprehensive patient binder checks and review study participant's databases for accurate data entry, signatures, and accuracy of all information, on a weekly basis until the study exit of a participant.
- The study team will meet at least weekly to review participant progress, clinical safety, and data monitoring throughout the duration of the study.

10.1.8 Quality Assurance and Quality Control

Study personnel at the site will perform internal quality management of study conduct, data and biological specimen collection, documentation and completion.

Quality control (QC) procedures will be implemented beginning with the data entry system and data QC checks that will be run on the database will be generated. Any missing data or data anomalies will be communicated to the site(s) for clarification/resolution.

The ISM will verify that the clinical trial is conducted and data are generated and biological specimens are collected, documented (recorded), and reported in compliance with the protocol, International Conference on Harmonisation Good Clinical Practice (ICH GCP), and applicable regulatory requirements (e.g., Good Laboratory Practices (GLP), Good Manufacturing Practices (GMP)).

The investigational site will provide direct access to all trial related sites, source data/documents, and reports for the purpose of monitoring and auditing by the sponsor, and inspection by local and regulatory authorities.

10.1.9 Data Handling and Record Keeping

10.1.9.1 Data Collection and Management Responsibilities

Data collection is the responsibility of the clinical trial staff at the site under the supervision of the site investigator. The investigator is responsible for ensuring the accuracy, completeness, legibility, and timeliness of the data reported.

All source documents should be completed in a neat, legible manner to ensure accurate interpretation of data.

Hardcopies of the study visit worksheets will be provided for use as source document worksheets for recording data for each participant enrolled in the study. Data recorded in the electronic case report form (eCRF) derived from source documents should be consistent with the data recorded on the source documents.

Clinical data (including adverse events (AEs), concomitant medications, and expected adverse reactions data) and clinical laboratory data will be entered into Redcap. The data system includes password protection and internal quality checks, such as automatic range checks, to identify data that appear inconsistent, incomplete, or inaccurate. Clinical data will be entered directly from the source documents.

10.1.9.2 Study Records Retention

Study documents should be retained for a minimum of 2 years after the last approval of a marketing application in an International Conference on Harminosation (ICH) region and until there are no pending or contemplated marketing applications in an ICH region or until at least 2 years have elapsed since the formal discontinuation of clinical development of the study intervention. These documents should be retained for a longer period, however, if required by local regulations. No records will be destroyed without the written consent of the sponsor, if applicable. It is the responsibility of the sponsor to inform the investigator when these documents no longer need to be retained.

10.1.10 Protocol Deviations

A protocol deviation is any noncompliance with the clinical trial protocol, International Conference on Harmonisation Good Clinical Practice (ICH GCP), or Manual of Procedures (MOP) requirements. The noncompliance may be either on the part of the participant, the investigator, or the study site staff. As a result of deviations, corrective actions are to be developed by the site and implemented promptly.

These practices are consistent with ICH GCP:

- 4.5 Compliance with Protocol, sections 4.5.1, 4.5.2, and 4.5.3
- 5.1 Quality Assurance and Quality Control, section 5.1.1
- 5.20 Noncompliance, sections 5.20.1, and 5.20.2.

It is the responsibility of the site investigator to use continuous vigilance to identify and report deviations within 7 working days of identification of the protocol deviation, or within 7 working days of the scheduled protocol-required activity. All deviations must be addressed in study source documents. Protocol deviations must be sent to the reviewing Institutional Review Board (IRB) per their policies. The site investigator is responsible for knowing and adhering to the reviewing IRB requirements. Further details about the handling of protocol deviations will be included in the MOP.

10.1.11 Publication and Data Sharing Policy

This study will be conducted in accordance with the following publication and data sharing policies and regulations:

This study will comply with the Clinical Trials Registration and Results Information Submission rule. As such, this trial will be registered at ClinicalTrials.gov, and results information from this trial will be submitted to ClinicalTrials.gov. In addition, every attempt will be made to publish results in peer-reviewed journals.

10.1.12 Conflict of Interest Policy

The independence of this study from any actual or perceived influence, such as by the pharmaceutical industry, is critical. Therefore, any actual conflict of interest of persons who have a role in the design, conduct, analysis, publication, or any aspect of this trial will be disclosed and managed. Furthermore, persons who have a perceived conflict of interest will be required to have such conflicts managed in a way that is appropriate to their participation in the design and conduct of this trial.

10.2 ABBREVIATIONS

The list below includes abbreviations utilized in this template. However, this list should be customized for each protocol (i.e., abbreviations not used should be removed and new abbreviations used should be added to this list).

AE	Adverse Event
CMP	Clinical Monitoring Plan
COC	Certificate of Confidentiality
CRF	Case Report Form
FDA	Food and Drug Administration
GCP	Good Clinical Practice
GLP	Good Laboratory Practices
GMP	Good Manufacturing Practices
HIPAA	Health Insurance Portability and Accountability Act
ICH	International Conference on Harmonisation
IRB	Institutional Review Board
ISM	Independent Safety Monitor
NIH	National Institutes of Health
OHRP	Office for Human Research Protections
PI	Principal Investigator
SAE	Serious Adverse Event
SAP	Statistical Analysis Plan
SOA	Schedule of Activities
UP	Unanticipated Problem

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