Nektar Therapeutics

CLINICAL STUDY PROTOCOL

A PHASE 1 DOUBLE-BLIND, DOUBLE-DUMMY, PARALLEL-GROUP, RANDOMIZED, POSITIVE CONTROL STUDY USING FUNCTIONAL MAGNETIC RESONANCE IMAGING TO EVALUATE THE EFFECT OF NKTR-181 ON BRAIN ACTIVITY IN HEALTHY, NON-PHYSICALLY DEPENDENT RECREATIONAL OPIOID USERS

Protocol Number: 18-181-26

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INVESTIGATOR SIGNATURE PAGE

Nektar Therapeutics

TITLE:	A Phase 1 Double-Blind, Double-Dummy, Parallel-Group, Randomized, Positive Control Study Using Functional Magnetic Resonance Imaging to Evaluate the Effect of NKTR-181 on Brain Activity in Healthy, Non-physically Dependent Recreational Opioid Users
PROTOCOL NUMBER:	18-181-26
PHASE OF STUDY:	1
PROTOCOL DATE:	24 October 2019
STUDY SPONSOR:	Nektar Therapeutics 455 Mission Bay Boulevard South San Francisco, CA 94158 USA

PRINCIPAL INVESTIGATOR COMMITMENT:

I, the undersigned Principal Investigator, submit this statement of commitment as evidence that I understand my responsibilities pursuant to the Code of Federal Regulations (21 CFR § 312.60 through § 312.70, 21 CFR § 11, 50, 54, 56) and ICH E6 Good Clinical Practice guidelines, as well as with any and all applicable federal, state and/or local laws and regulations, and agree to conduct the study in accordance with the protocol referenced herein.

Principal Investigator Printed Name

Principal Investigator Signature **Date**

PROTOCOL APPROVAL PAGE

A Phase 1 Double-Blind, Double-Dummy, Parallel-Group, Randomized, Positive Control Study Using Functional Magnetic Resonance Imaging to Evaluate the Effect of NKTR-181 on Brain Activity in Healthy, Non-physically Dependent Recreational Opioid Users

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LIST OF STUDY CONTACTS

Study Contact	Name	Contact Information
Nektar		

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

ABBREVIATIONS

Abbreviation or Term	Definition
AE	adverse event
ALP	alkaline phosphatase
ALT (SGPT)	alanine aminotransferase (serum glutamic pyruvic transaminase)
AST (SGOT)	aspartate aminotransferase (serum glutamic oxaloacetic transaminase)
AT	aminotransferase
ATC	Anatomical, Therapeutic, or Chemical
AUC	area under the curve
$AUC_{0\text{-last}}$	area under the concentration time curve from time zero to the last measurable concentration
AUE	area under effect
BMI	body mass index
BUN	blood urea nitrogen
CARI	Collaborative Advanced Research Institute
CFR	Code of Federal Regulations
C_{max}	maximum concentration
CNS	central nervous system
COWS	clinical opiate withdrawal scale
CRSU	Clinical Research Services Unit
CYP3A4	cytochrome P 450 enzyme isoform 3A4
DCIs	data collection instruments
DEA	Drug Enforcement Administration
DILI	drug induced liver injury
DSM-5	Diagnostic and Statistical Manual of Mental Disorders 5th Edition
ECG	electrocardiogram
eCRF	electronic case report form
EOS	End of Study
ET	Early Termination
$ETCO_2$	End tidal CO_2 monitoring
FDA	Food and Drug Administration
fMRI	functional magnetic resonance imaging
GCP	Good Clinical Practice
GGT	gamma-glutamyl transferase
GLM	general linear model
HAP	human abuse potential
HDPE	high-density polyethylene
IB	Investigator's Brochure
ICF	informed consent form

Abbreviation or Term	Definition
ICH	International Council for Harmonisation
IEC	independent ethics committee
IND	investigational new drug
IR	immediate release
IRB	institutional review board
kg	kilogram
LDH	lactate dehydrogenase
m^2	meters squared
MedDRA	Medical Dictionary for Regulatory Activities
mg	milligram
MINI	Mini-International Neuropsychiatric Interview 7.0.2 for DSM-5
mL	milliliter
MRI	magnetic resonance imaging
OTC	over-the-counter
oxycodone IR	oxycodone immediate release
PD	pharmacodynamic
Pgp	P-glycoprotein
PK	pharmacokinetics
PT	preferred term
Q12h	every 12 hours
QTcF	corrected QT interval (Fridericia)
RR	respiratory rate
SAE	serious adverse event
SOC	system organ class
SOP	standard operating procedure
SpO_2	peripheral blood oxygen saturation
SUSAR	suspected unexpected serious adverse reaction
$t_{1/2}$	terminal elimination phase half-life
TBL	total bilirubin
TEAE	treatment-emergent adverse event
THC	tetrahydrocannabinol
T_{max}	time to maximum concentration
ULN	upper limit of normal

1.0 STUDY SYNOPSIS

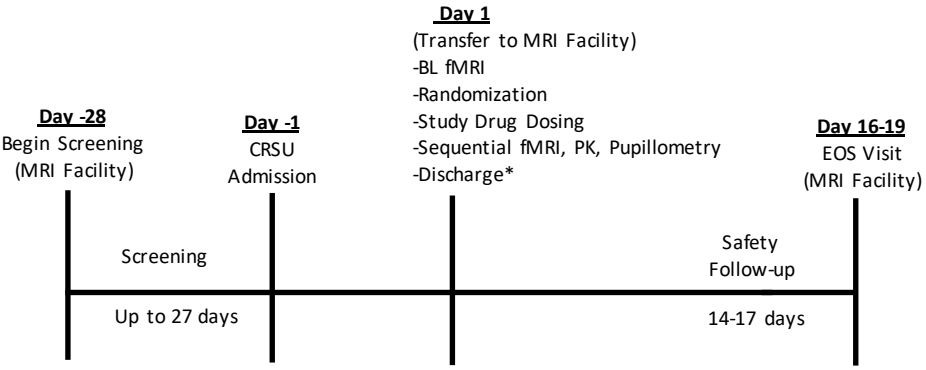
Name of Sponsor:	Nektar Therapeutics
Name of Finished Product:	NKTR-181
Name of Active Ingredient:	NKTR-181
Title of Study:	A Phase 1 Double-Blind, Double-Dummy, Parallel-Group, Randomized, Positive Control Study Using Functional Magnetic Resonance Imaging to Evaluate the Effect of NKTR-181 on Brain Activity in Healthy, non-physically Dependent Recreational Opioid Users
Duration of Study:	47 days, including a 27-day Screening period and a minimum 14-day Safety Follow-Up period
Phase of Development:	1
Principal Investigators:	[redacted]
Objectives:	Primary: • To evaluate the effects of oral administration of NKTR-181 on brain activity via functional magnetic resonance imaging (fMRI) Secondary: • To evaluate safety in subjects administered NKTR-181 or oxycodone immediate release (oxycodone IR) • To evaluate the objective drug effects of NKTR-181 and oxycodone IR on pupil diameter • To evaluate the pharmacokinetic profile of NKTR-181 and of oxycodone IR Exploratory: • To describe the difference between NKTR-181 and oxycodone IR with respect to fMRI signaling in brain reward circuitry
Study Population:	Male and female healthy non-physically dependent recreational opioid users, age 18 to 65 years, inclusive
Number of Subjects:	Approximately 24 subjects completing all MRI scans
Number of Study Sites:	One
Countries:	USA
Study Design:	This study is a single-center, double-blind, double-dummy, parallel-group, randomized, positive control trial in healthy, non-physically dependent recreational opioid users. Subjects will enter a screening period between Day -28 and Day -2. Upon meeting all criteria for enrollment, on Day -1 subjects will enter the Clinical Research Services Unit (CRSU) for a an overnight confinement. On Day 1, following baseline pharmacokinetic (PK) sample collection and pupillometry procedures, subjects will undergo a baseline fMRI. Upon fMRI completion, subjects will be randomized to NKTR-181 or oxycodone IR.

	Treatment Groups: Group 1: NKTR-181 400 mg and oxycodone IR placebo Group 2: Oxycodone IR 40 mg and NKTR-181 placebo Following randomization, a single dose of study drug and matched alternate-treatment placebo will be administered. Subjects will undergo a series of three fMRIs post dose (at hours 1, 2, and 4). At post-dose hours 0.5, 1, 2, 3, 4, 5, 6, and 8, pupillometry will be performed and PK blood samples will be drawn. When occurring at times of fMRI, these assessments will be performed prior to the scans. Following a 14- to 17-day safety follow-up period, subjects will return to the research facility clinic for the End of Study (EOS) visit (Day 16-19).
Key Eligibility Criteria:	1. Male and female healthy, non-physically dependent recreational opioid users, age 18 to 65 years, inclusive. A recreational opioid user is defined as one who is not currently physically dependent on opioids but has experience in the use of opioids for non-therapeutic purposes (i.e., for psychoactive effects) on at least 10 occasions within the last year and at least once in the 8 weeks prior to the screening visit. 2. Subjects must be free from significant medical conditions including convulsive disorders, and from current clinically significant psychiatric disorders including claustrophobia 3. Females must be surgically sterile (by means of bilateral tubal ligation, bilateral salpingectomy, hysterectomy or bilateral oophorectomy), or be post-menopausal (defined as spontaneous cessation of menses for at least 1 year). If of childbearing potential, subjects must be practicing abstinence or using a medically acceptable form of contraception (e.g., hormonal contraception, intrauterine device, double barrier method [condom, diaphragm or cervical/vault cap with spermicidal foam/gel/film/cream/suppository]) throughout the duration of the study and continued until 2 months following the last dose of study drug. 4. Males with female partners of child-bearing potential must be practicing abstinence or must agree to use a double barrier form of contraception (e.g., condom with spermicidal foam/gel/film/cream/suppository) throughout the duration of the study and continued until 2 months following the last dose of study drug. This criterion may be waived for male subjects who have had a vasectomy > 6 months before signing the Informed Consent Form (ICF).
Test Product, Dose and Mode of Administration:	Two NKTR-181 200 mg tablets administered orally.
Reference Therapy:	• Two oxycodone IR 20 mg tablets, over-encapsulated, administered orally. • NKTR-181 placebo tablets, oral administration • Oxycodone IR placebo capsule, oral administration
Safety:	Adverse event (AE) data will be collected. Subjects will be monitored pre- and post-dose for systolic and diastolic blood pressure, heart rate, peripheral blood oxygen saturation (SpO_2), temperature, and respiratory rate. Pre- and post-dose electrocardiograms (ECGs) will be obtained. Clinical laboratory testing will be performed. Subjects will be under continuous pulse oximetry monitoring from pre-dose on Day 1 until discharge from the CRSU.
Pharmacokinetics:	PK blood samples will be collected pre-dose and through 8 hours post-dose for assessment of NKTR-181 (and selected metabolites) and oxycodone concentrations in plasma.

Pharmacodynamics:	Pupil diameter measurements and fMRI scans will be obtained pre-dose and at specified times through 8 hours post-dose.
Statistical Methods:	Sample size: No formal sample size calculation has been made. Based on experience from previous studies, a total of 24 evaluable subjects (12 per arm) is considered sufficient for the study. An evaluable subject is defined as a subject who completes sufficient MRI scans and PK/PD sample collections for evaluating the effects of oral administration of NKTR-181 400 mg on brain activity Analysis populations: The safety population will consist of all subjects who receive a dose of study drug. There will be two PD populations, one for pupillary endpoints and the other for fMRI endpoints. The pupillary PD population will consist of all subjects who received study drug and have at least one pupillary determination. The fMRI PD population will consist of all subjects who received study drug and have sufficient MRI data for NKTR-181 or oxycodone IR treatment to allow description of modulation of brain circuitry. The PK population will consist of subjects who have sufficient plasma concentration data to facilitate the calculation of at least one PK parameter such as area under the curve (AUC) or maximum concentration (C_{max}), as determined by the pharmacokineticist. Subject Characteristics and Disposition: Subject demographics, baseline characteristics, and disposition will be summarized. Pharmacokinetic analyses: Individual subject plasma PK concentration data and parameters will be tabulated and summarized using descriptive statistics. The PK parameters C_{max}, time to maximum concentration (T_{max}), and area under the concentration time curve from time zero to the last measurable concentration ($AUC_{0\text{-last}}$) will be estimated from plasma concentration-time data where possible. Pharmacodynamic analyses: The effective connectivity analyzed will be summarized for NKTR-181 and oxycodone. The effects of medication at each time point through DCM analysis will be summarized and included in data listings. Perfusion differences between treatments over time after drug administration may be modeled using ROI-based measurements made in regions in the brain, if appropriate. Data from pupillometry will be summarized by treatment using descriptive statistics. The time course of treatment mean of PD endpoints will be displayed graphically to show the visual differences between treatment groups over time. If it is appropriate, the peak effect for pharmacodynamical endpoint may be explored. Safety analyses: Treatment-emergent AEs will be presented by Medical Dictionary for Regulatory Activities (MedDRA) system organ class (SOC) and preferred term (PT) and will be summarized by severity and relationship to treatment. Data from other safety assessments will be summarized descriptively. An informal data analysis may be performed prior to study completion.

1.1 Study Schematics

Figure 1: Study Schema



BL = Baseline; CRSU = Clinical Research Services Unit; EOS = End of Study; fMRI = Functional MRI; PK = Pharmacokinetic

* If the site deems the subject not appropriate for discharge, the subject will be transported from the MRI facility back to the CRSU.

1.2 Schedule of Assessments

Table 1: Schedule of Assessments

	Screening[1]	Day -1	Day 1										Day 2	EOS (Day16-19)/ET
			Pre	0	0.5h	1h	2h	3h	4h	5h	6h	8h	Post-Hour 8		
Informed consent	X														
Demographic information	X														
MINI 7.0.2 for DSM-5	X														
Eligibility review	X	X													
Medical/medication history	X	X													X
Complete physical examination	X														X
Brief physical examination		X										X		X[7]	
Body weight and height[2]	X	X													
Substances of abuse screen (urine)	X	X	X												
Pregnancy test[4,11]	X	X	X												
Alcohol breath test[3]	X	X	X												
Clinical laboratory tests[4]	X	X										X[7]		X[7]	X
12-lead electrocardiogram	X	X	X									X		X[7]	X
Vital sign measurements[5]	X	X	X			X	X					X		X[7]	X
Study drug administration				X											
Randomization			X												
Training on PD measures		X													
fMRI[6,10]			X			X	X[12]		X						
Pupillometry[6]			X		X	X	X	X	X	X	X	X			
$ETCO_2$[10]			X			X	X		X			X			
PK sample collection			X		X	X	X	X	X	X	X	X			
COWS	X	X	X												
Discharge from clinic[7]													X[7]	X[7]	
Continuous $ETCO_2$ monitoring[10]			←	—	—	—	—	—	—	—	—	→			
Continuous SpO_2 monitoring[8]			←	—	—	—	—	—	—	—	—	→	X	X	
Concomitant meds/AEs	←	—	—	—	—	—	—	—	—	—	—	—	—	—	→
Confinement to CRSU[7,9]		↔											↔		

Abbreviations: AE = adverse event; BP = blood pressure; CO_2 = carbon dioxide; COWS = clinical opiate withdrawal scale; CRSU = clinical research services unit; DSM = Diagnostic and Statistical Manual of Mental Disorders; EOS = end of study; ET = early termination; $ETCO_2$ = end tidal carbon dioxide; fMRI = functional magnetic resonance imaging; h = hours; HR = heart rate; MINI = mini-international neuropsychiatric interview; MRI = magnetic resonance imaging; PD = pharmacodynamics; PK = pharmacokinetic; SpO_2 = saturation of peripheral oxygen

[1] Screening occurs during the 28 days prior to Day -1.

[2] Height measured at screening only.

[3] Alcohol breath test will be performed at the Screening visit, prior to admission to the CRSU on Day -1, and at the MRI facility prior to baseline MRI scanning.

[4] Clinical laboratory tests include chemistry, hematology and urinalysis.

[5] Vital signs include BP, HR and SpO2; temperature taken at screening and Day -1 only.

[6] Prior to each fMRI scan, pupillometry will be performed and PK blood samples will be drawn at the scheduled time.

[7] Discharge following at least 8 hours of monitoring post-dosing of study drug. Following an intoxication assessment. Subjects who cannot safely be discharged after 8 hours per PI discretion will return to the CRSU for a second overnight of safety monitoring. ECG and laboratory testing will be performed at the Investigator's discretion. If the patient is held overnight through Day 2, Day 1 Hour 8 laboratory testing may be done on Day 2 instead, per the Investigator's discretion.

[8] Continuous pulse oximetry from pre-dose while at the MRI facility on Day 1 until approximately 8 hours post-dose. Only pulse-ox values at the time of vital sign measurement will be recorded.

[9] Subjects will be confined to CRSU except for time spent in transport to and from, and at, the MRI facility on Day 1.

[10] End tidal CO2 (ETCO2) will be recorded before each MRI pulse sequence during each of the scans. ETCO2 will be captured continuously from pre-dose until Hour 8.

[11] All females will undergo serum pregnancy tests at screening and on Day -1. They will undergo a urine pregnancy test on Day 1 prior to dose administration.

[12] The 2-hour fMRI may begin any time during the 30-minute period beginning 2 hours post-dose.

2.0 INTRODUCTION

2.1 Background

Opioid analgesics are effective agents for the relief of moderate to severe pain. These compounds are highly effective as analgesics, both acutely and chronically; however, their clinical utility is often significantly limited by adverse effects, particularly those involving the central nervous system (CNS) (Labianca, 2012). Sedation, cognitive impairment, and respiratory depression often limit the doses that patients can tolerate, resulting in less than adequate pain control. In addition, the CNS effects experienced as pleasurable by some users of opioid drugs have led to a well-documented and much publicized issue of opioid misuse, abuse, and diversion, including criminality, which have recently been identified as a public health emergency. Although sustained-release formulations and combination products designed to make access to the active opioid ingredient more difficult have been developed and approved, the safety mechanisms built into these drug products can be subverted and/or are not optimal in preventing misuse.

2.1.1 NKTR-181

NKTR-181 is a novel mu-opioid receptor agonist being developed for the treatment of moderate to severe chronic low back pain with the goal of providing effective analgesia while reducing CNS-mediated side effects, including opioid abuse potential. NKTR-181 has been designed to have a slow rate of entry into the brain to reduce the attractiveness of the molecule as a target of abuse. NKTR-181 is a new molecular entity (NME) with low abuse potential properties resulting from its novel molecular structure and pharmacodynamic properties. NKTR-181 is not a prodrug, reformulation, or drug product formulated with sustained release of an existing opioid.

Modulation of NKTR-181 entry into the CNS is achieved via physicochemical properties at the molecular level, rather than through formulation. The primary therapeutic hypothesis of NKTR-181 is that clinically meaningful analgesia can be achieved with concurrent reduction of acute CNS effects such as euphoria, respiratory depression, and sedation by reducing the rate of opioid entry into the brain. Furthermore, by reducing the pleasurable adverse CNS effects associated with acute opioid exposure, reduced psychological dependence and abuse potential is expected with NKTR-181. This outcome would represent an advancement over currently marketed opioids and may fill an increasingly important role in the treatment of moderate to severe chronic pain conditions for which NKTR-181 is targeted. At this stage of development, NKTR-181 is a Drug Enforcement Administration (DEA) Schedule II compound based on the Controlled Substances Act. However, NKTR-181 has been shown to have greatly reduced abuse potential compared with oxycodone in animal models and in two human abuse potential (HAP) studies in healthy, non-physically dependent recreational opioid users.

To date, Nektar Therapeutics has not identified any conventional chemical or physical methods for altering the NKTR-181 molecule to modulate or accelerate the entry of this mu-opioid agonist into the CNS. Efforts to chemically manipulate the molecule have degraded the pharmacophore, rendering it inactive as a mu-opioid agonist. NKTR-181 is therefore intended to

provide clinically meaningful analgesia with an improved CNS side effect profile and an attenuated potential for misuse and abuse.

2.1.1.1 NKTR-181 Pharmacokinetic Profile

Following oral administration, NKTR-181 was absorbed readily with maximum concentration (C_{max}) achieved in less than 4 hours after single doses of 10 mg to 1200 mg, and after multiple 100 mg to 400 mg doses given twice daily. The mean terminal elimination phase half-life ($t_{½}$) values ranged between 11 to 14 hours. Across the range of doses evaluated, exposure to NKTR-181 increased in a dose-proportional manner. Plasma concentrations reached steady state after Day 3 of twice-daily dosing. No gender differences in NKTR-181 pharmacokinetics (PK) have been observed.

2.1.1.2 NKTR-181 Efficacy in Phase 3 Clinical Trial

The Phase 3 SUMMIT-07 study (Study 14-181-07) evaluated four analgesic doses of NKTR-181 (100 mg, 200 mg, 300 mg, and 400 mg) in over 600 opioid-naïve patients with moderate to severe chronic low back pain. The primary efficacy endpoint demonstrated significantly improved chronic back pain with NKTR-181 compared to placebo (p=0.0019), and an average pain score reduction of over 65% (from a mean of 6.73 at Screening to a mean of 2.32 at randomization) was seen during the dose titration period. Key secondary endpoints of the study also achieved high statistical significance.

2.1.1.3 NKTR-181 Abuse Potential

An initial HAP study (Study 12-181-05) of 42 non-opioid-dependent recreational opioid users was conducted to evaluate abuse potential of NKTR-181 versus oxycodone. In subjects administered NKTR-181 doses of 100 mg, 200 mg, and 400 mg versus oxycodone 40 mg, NKTR-181 more closely resembled placebo than oxycodone HCl at most timepoints. At all NKTR-181 dose levels (100 mg, 200 mg, and 400 mg), scores on all measures of abuse potential were significantly lower for NKTR-181 compared with oxycodone HCl. These measures included the primary endpoint of Drug Liking and secondary endpoints from the Drug Effects Questionnaire (Feel Drug Effects, Have Good Effects, Have Bad Effects, Have Nausea, Feel Sick, Make Dizzy, and Make Sleepy) as well as pupillometry. While NKTR-181 was detected in blood within 15 minutes of dosing, the onset of pupil constriction, a CNS-mediated effect of mu-opioid drugs, was delayed, and maximal constriction occurred 2 to 3 hours after plasma concentrations were maximal and had begun to decline.

A second HAP study (Study 15-181-15) assessed the relative oral abuse potential of NKTR-181 in 54 healthy non-physically dependent recreational drug users. NKTR-181 administered at 400 mg, 600 mg, and at a 1200 mg supratherapeutic dose was compared to oxycodone 40 mg and 60 mg. For the primary endpoint of Drug Liking, NKTR-181 (400 mg and 600 mg) rated less likable compared to oxycodone 40 mg and 60 mg (p<0.0001), and a supratherapeutic dose of NKTR-181 (1200 mg) rated less likable than oxycodone 60 mg (p=0.0071). Key secondary

endpoints of Area Under Effect (AUE) for Drug Liking (0-1 hours, 0-2 hours, 0-3 hours), Drug High, and Take Drug Again also met statistical significance for all doses of NKTR-181 (400 mg, 600 mg,1200 mg) compared to oxycodone (60 mg).

2.1.1.4 NKTR-181 Safety and Tolerability

Under Nektar-Sponsored studies through 08 March 2018, a total of 2223 subjects have received at least one dose of NKTR-181. NKTR-181 has been generally well tolerated as single doses ranging from 10 mg to 1200 mg and as multiple doses from 100 mg every 12 hours to 600 mg every 12 hours.

Adverse events (AEs) attributed to NKTR-181 in clinical trials include nausea, dizziness, constipation, somnolence, pruritus, decreased appetite, vomiting, dry mouth, euphoric mood, urinary retention, abnormal dreams, feeling abnormal, blurred vision, oxygen desaturation, and feeling of relaxation. The most common AEs associated with NKTR-181 in Phase 3 studies are consistent with those seen with an opiate agonist: constipation, nausea, headache, somnolence, and vomiting. These were generally of mild or moderate severity and were manageable.

One death was reported in the Phase 3 clinical study, a cerebrovascular accident considered unrelated to study drug. The subject was a 64-year-old male with a history of hypertension and hyperlipidemia, who had been receiving NKTR-181 100 mg at the time of the event. Few serious adverse events (SAEs) have been reported. SAEs considered by the Investigator to be related to study drug include single instances of transient blindness and angioedema; in both cases, subjects were taking concomitant medications known for their potential to cause each event (tadalafil in the case of transient blindness and lisinopril in the case of angioedema).

A Phase 3 study in patients with moderate to severe chronic low back pain demonstrated a favorable safety profile consistent with that observed in the earlier studies.

Refer to the NKTR-181 Investigator's Brochure (IB) for further information on nonclinical data (including toxicology results) and clinical results of prior studies (Nektar Therapeutics, 2018).

2.2 Study Population

This study will enroll and randomize approximately 24 healthy non-physically dependent recreational opioid users, age 18 to 65 years, inclusive.

Given the modulatory effects of both chronic pain and chronic opioid exposure on reward pathways in the brain, subjects with the presence of drug (except nicotine or caffeine) or alcohol dependence as determined by structured clinical interview (including subjects who are seeking treatment for such a disorder) will be excluded from this study, based on responses on the Mini-International Neuropsychiatric Interview 7.0.2 (MINI) for Diagnostic and Statistical Manual of Mental Disorders 5th edition (DSM-5) and Clinical Opiate Withdrawal Scale (COWS).

2.3 Study Rationale

Functional magnetic resonance imaging (fMRI) assessments in subjects administered opioids such as morphine, buprenorphine, and nalbuphine have shown drug-induced signaling changes in reward structures such as the nucleus accumbens, orbitofrontal cortex, and hippocampus, as well as changes in the functional connectivity of reward circuitry (Becerra, 2006; Upadhyay, 2012; Gear, 2013). Visualization of the effects of NKTR-181, a centrally-acting full mu-opioid receptor agonist, on modulation of brain circuitry, may provide further insight into the central effects of the novel opioid.

Oxycodone immediate release (oxycodone IR), a Schedule II opioid, will be used as a positive control. PK and pharmacodynamic (PD) data will be used for descriptive correlation with brain activation patterns seen on serial fMRI scans. In the SUMMIT-07 Phase 3 study (Study 14-181-07), the NKTR-181 400 mg every 12 hour (Q12h) dose was the highest administered dose evaluated and was both efficacious and well-tolerated in opioid-naïve patients with chronic low back pain. NKTR-181 400 mg was also the dose common to both HAP studies (Studies 12-181-05 and 15-181-15), as was the comparator oxycodone IR dose of 40 mg.

2.4 Risks/Benefits

A single oral administration of NKTR-181 400 mg is not expected to be of benefit to the subjects in this study.

A single oral administration of oxycodone IR 40 mg is not expected to be of benefit to the subjects in this study.

Risks associated with oral administration of NKTR-181 400 mg have been described in the IB (Nektar Therapeutics, 2018).

Risks associated with oral administration of oxycodone IR include subjects' experiencing adverse events common to opioid medications. See oxycodone IR prescribing information for safety information.

3.0 STUDY OBJECTIVES

3.1 Primary Objective

- To evaluate the effects of oral administration of NKTR-181 on brain activity via fMRI

3.2 Secondary Objectives

- To evaluate safety in subjects administered NKTR-181 or oxycodone IR
- To evaluate the objective drug effects of NKTR-181 and oxycodone IR on pupil diameter
- To evaluate the pharmacokinetic profile of NKTR-181 and of oxycodone IR

3.3 Exploratory Objective

- To describe the difference between NKTR-181 and oxycodone IR with respect to fMRI signaling in brain reward circuitry

4.0 STUDY DESIGN

4.1 Summary of Study Design

This study is a single-center, double-blind, double-dummy, parallel-group, randomized, positive control trial in healthy, non-physically dependent recreational opioid users.

Subjects will enter a Screening period between Day -28 and Day -2. Upon meeting all criteria for enrollment, on Day -1 subjects will enter the clinical research services unit (CRSU) for a 2-day confinement.

On Day 1, following pre-dose collection of blood for assessment of PK parameters and pupillometry procedures, subjects will undergo a baseline fMRI. Upon completion of MRI assessment, subjects will be randomized to NKTR-181 or oxycodone IR.

Treatment Groups:
Group 1: NKTR-181 400 mg and oxycodone placebo
Group 2: Oxycodone IR 40 mg and NKTR-181 placebo

Following randomization, a single dose of the randomized study drug as well as matching placebo of the other study drug will be administered. Post-dose, subjects will undergo a series of three fMRIs (at hours 1, 2, and 4). At post-dose hours 0.5, 1, 2, 3, 4, 5, 6, and 8, pupillometry will be performed and PK blood samples will be drawn. When occurring at times of fMRI, these assessments will be performed prior to the scans.

At least 8 hours post-dosing of study drug, subjects will be discharged from the CARI Clinic. Following a 14- to 17-day safety follow-up period, subjects will return to the CARI Clinic for the End of Study (EOS) visit (Day 16-19).

A schematic of the study design is presented in Section 1.1; the Schedule of Assessments is in Section 1.2.

5.0 SELECTION OF STUDY POPULATION

5.1 Inclusion Criteria

Subjects must meet the following criteria to be enrolled in this study:

1) Willing and able to provide written documentation of informed consent.
2) Healthy male and female recreational opioid users, 18 to 65 years of age, inclusive, at the time of signing the informed consent form (ICF). A recreational opioid user is defined as one who is not currently physically dependent on opioids but has experience in the use of opioids for non-therapeutic purposes (i.e., for psychoactive effects) on at least 10 occasions within the last year and at least once in the 8 weeks prior to the Screening visit.
3) Body mass index (BMI) between 19.0 to 45.0 kg/m^2, inclusive.
4) Females must be surgically sterile (by means of bilateral tubal ligation, bilateral salpingectomy, hysterectomy or bilateral oophorectomy), or be post-menopausal (defined as spontaneous cessation of menses for at least 1 year), or, if of childbearing potential, must be practicing abstinence or using a medically acceptable form of contraception (e.g., hormonal contraceptives, intrauterine device, double barrier method [condom, diaphragm or cervical/vault cap with spermicidal foam/gel/film/cream/suppository]) throughout the duration of the study and continued until 2 months following the last dose of study drug.
5) Males with female partners of child-bearing potential must be practicing abstinence or must agree to use a double barrier form of contraception (e.g., condom with spermicidal foam/gel/film/cream/suppository) throughout the duration of the study and continued until 2 months following the last dose of study drug. This criterion may be waived for male subjects who have had a vasectomy > 6 months before signing the ICF.
6) Able to communicate with the Investigator and study site personnel, and willing and able to understand and comply with all study procedures, including swallowing oral medications.
7) Have at least one urine drug screen positive for opioids during Screening to confirm recreational opioid use. Subjects testing positive for methadone or buprenorphine prescribed for treatment, will be excluded.

5.2 Exclusion Criteria

1) Any metal fragments or other bodily metal (e.g., pacemaker, hip replacement) that would pose a risk to subjects during MRI scanning as determined by the MRI technologist and/or MRI physicist

2) Any clinically significant disease or condition (medical or surgical) that might compromise the cardiovascular, hematological, renal, hepatic, pulmonary (including chronic asthma), endocrine (e.g., diabetes, Addison's disease, hypothyroidism), central nervous, or gastrointestinal (including gastric ulcer or paralytic ileus) systems or other conditions that may interfere with the absorption, distribution, metabolism, or excretion of the study drug or would place the subject at increased risk unless jointly considered not clinically significant by the Investigator and Sponsor.
3) Elevated aspartate aminotransferase (AST), alanine aminotransferase (ALT), gamma-glutamyl transferase (GGT), alkaline phosphatase (ALP), lactate dehydrogenase (LDH), total bilirubin (TBL), blood urea nitrogen (BUN), or creatinine levels (> 10% above the upper limit of normal) at Screening. unless considered not clinically significant by both the Investigator and Sponsor.
4) Corrected QT interval, Fridericia's correction (QTcF) > 450 msec for males and > 470 msec for females, or any clinically significant abnormality on the 12-lead electrocardiogram (ECG) at Screening or Day -1.
5) History of clinically significant acute asthma or other obstructive airway disease (e.g., chronic obstructive pulmonary disease) requiring daily controller medication (e.g., inhaled corticosteroids or bronchodilators) or any condition that may increase the risk for respiratory depression (e.g., sleep apnea), judged as clinically significant by the Investigator.
6) Current neurologic conditions such as convulsive disorders, or history of severe head injury, judged as clinically significant by the Investigator.
7) Any current DSM-5 psychiatric disorder, including but not limited to bipolar disorder, major depressive disorder, attention deficit hyperactivity disorder, or schizophrenia, or a neurological disorder requiring ongoing treatment.
8) Current Substance Use Disorder (moderate to severe), other than Opioid, Nicotine, THC (tetrahydrocannabinol), Cocaine or Caffeine, as defined by the DSM-5.
9) Physical dependence for Opioids (as assessed by the MINI).
10) History of claustrophobia or any other psychiatric disorder that would preclude subject tolerance of MRI procedures.
11) Current suicidal or homicidal ideation or a suicide attempt within the past 6 months.
12) Inability to commit to abstinence from use of any tobacco- or nicotine-containing products during the period from admission to discharge from the CRSU or CARI Clinic, despite being administered a nicotine transdermal patch while confined in the CRSU and while in the CARI Clinic.
13) Known contraindication, sensitivity (including nausea and/or vomiting), or allergy to any opioid analgesic.

14) Use of any prescription medication (other than recreational opioid use) within 14 days prior to Day 1, or use of over-the-counter [OTC] products, such as antacids, aspirin, vitamins, minerals, dietary/herbal preparations, and nutritional supplements during the period from admission until discharge from the CRSU or CARI Clinic, unless jointly approved by the Investigator and Sponsor. Use of medications that are inhibitors or inducers of cytochrome P 450 enzyme isoform 3A4 (CYP3A4) and P-glycoprotein (Pgp) or that prolong the QT interval are prohibited within 5 half-lives or 2 weeks (whichever is longer) prior to Day 1, unless jointly approved by the Investigator and Sponsor.
15) Current use of any medication that could affect central nervous system blood flow (e.g. certain cardiovascular medications, triptan migraine medications), unless approved by the Investigator.
16) Consumption of grapefruit, grapefruit juice, Seville oranges, Seville orange marmalade, or other products containing grapefruit or Seville oranges within 14 days prior to Day 1.
17) Currently participating or have previously participated in another investigational drug or investigational biologic study within 30 days prior to signing the ICF and through study completion.
18) Evidence of poor venous access in the opinion of the Investigator.
19) Receipt of blood or blood products within 45 days prior to Day 1.
20) Previous exposure to NKTR-181.
21) Any condition including medical, emotional, psychiatric, or logistical that, in the opinion of the Investigator, would preclude the subject from adhering to the protocol.
22) Involvement in the planning and/or conduct of the study (applies to both Sponsor personnel and personnel at the study site).
23) Clinical Opiate Withdrawal Scale (COWS) score of greater than 5 during screening or prior to first scan.
24) Positive urine drug screen for buprenorphine or methadone immediately prior to the first scan.
25) Females who are lactating, pregnant or planning to become pregnant within 2 months of receiving their dose of study drug.

5.3 Removal of Subjects from Study Therapy or Assessment

Subjects may choose to discontinue the trial at any time, for any reason.

Subjects may be stopped from participation in or be withdrawn from the study for any of the following reasons:

- The subject entered the study in violation of this protocol

- Safety reasons as judged by the Investigator and/or Sponsor, including AEs
- The subject requires the use of an unacceptable concomitant medication (see Section 6.4)
- Significant subject noncompliance to the protocol as judged by the Investigator and/or Sponsor
- In the Investigator's opinion, it is not in the best interest of the subject to continue
- Sponsor decision to end the study
- If, on Day -1 or prior to dosing on Day 1, an ECG is obtained and reveals that the subject's QTcF interval is out of the range specified in the eligibility criteria (> 450 msec for males and > 470 msec for females) and is confirmed with a second ECG within 30 minutes, study medication will not be dosed and the subject will be terminated from the study.

In the event that a subject is withdrawn or does not complete the study for any reason, the subject will complete End of Treatment (ET) procedures and be discharged from the CRSU or CARI Clinic and from the study. The date and the reason for discontinuation will be documented. If a subject has an ongoing AE, the event will be followed until resolution or for 2 weeks after the last visit, whichever comes first. If a subject refuses to complete ET procedures, this information will be documented.

The decision to enroll replacement subjects will be made at the discretion of the Sponsor.

If a subject is "lost to follow-up" (i.e., fails to return for study visits), a reasonable effort will be made to contact him/her to determine the reason for the failure to return and these contacts will be documented in the source documents.

6.0 TREATMENT PLAN

Each clinical study visit and the clinical study confinement period will take place at a Phase 1 CRSU or the MRI facility.

6.1 Screening Period (Visit 1)

Each subject will attend a Screening visit (Visit 1) at the MRI facility within 28 days prior to Day -1. Refer to the Schedule of Assessments (Section 1.2) for the procedures performed. Subjects will provide written informed consent to participate in the study before completing any protocol-specified procedures or evaluations.

6.2 Treatment Period (Confinement Period) (Visit 2)

At Visit 2 (Day -1), subjects will have their eligibility re-confirmed, based on Day -1 assessments outlined on the Schedule of Assessments (Section 1.2). AEs will also be assessed. Subjects who remain eligible will be admitted to the CRSU. A urine drug screen and alcohol breath test will be repeated on the CRSU.

Water will be restricted from 1 hour prior to dosing through 1 hour post-dose, except for the water required for dosing study drug.

On Day 1, subjects will be transported from the CRSU to the MRI facility. After arrival at the facility, concomitant medications and AEs will be assessed per the Schedule of Assessments (Section 1.2) and an alcohol breath test will be administered. Subjects will have a catheter placed in a right or left arm vein for blood draws.

Baseline (pre-dose) PK and pupillometry assessments will be performed. A urine pregnancy test will also be performed. Continuous pulse oximetry will be initiated, which will continue for at least 8 hours post-dose. $ETCO_2$ will be captured from pre-dose until Hour 8. The pre-dose fMRI scan will then be obtained. Following the fMRI, subjects will be randomized to either the NKTR-181 or oxycodone IR treatment arm and receive the randomized study drug dose and matching alternate treatment placebo.

Following administration of the study drug, ECG, vital signs, PK collection, and pupillometry will be conducted per the Schedule of Assessments (Section 1.2).

Following at least 8 hours of monitoring post-dosing of study drug per the Schedule of Assessments (Section 1.2), subjects will undergo a brief physical exam and be discharged from the CARI clinic. If the site deems the subject not appropriate for discharge, the subject will be transported from the MRI facility back to the CRSU.

6.3 Safety Follow-up Period (Visit 3/End of Study [EOS])

Following discharge, subjects will return to the CARI Clinic for the EOS visit between Day 16 and Day 19 for assessment of safety parameters. Refer to the Schedule of Assessments (Section 1.2) for the procedures performed.

6.4 Prior and Concomitant Medications

The administration of concomitant medications (prescription or OTC) is prohibited from admission to the CRSU until discharge, unless jointly approved by the Investigator and Sponsor. Use of medications that are inhibitors or inducers of CYP3A4 and Pgp or that prolong the QT interval are prohibited within 5 half-lives or 2 weeks (whichever is longer) before Day 1, unless jointly approved by the Investigator and Sponsor. A separate document will be provided as a reference that lists the common drugs that may affect CYP3A4/Pgp metabolism.
The Investigator should refer to the package insert for any medications to determine unacceptable interactions and/or contact the Sponsor.

The short-term use of an OTC medication for a self-limiting indication (e.g., acetaminophen for a moderate to severe headache) during the period from admission to the CRSU until discharge may be authorized by the Investigator in consultation with the Sponsor. The Investigator must make the decision to authorize use of such a medication only after consideration of the clinical situation, the potential for masking symptoms of a more significant underlying event, and whether use of the medication will compromise the outcome or validity of the study. Upon entering the study, each subject will be instructed to report the use of any medication to the Investigator. Subjects will also be instructed about the importance of not taking any medication (including OTC medications) without consulting the Investigator.

6.5 Prohibited and Restricted Medications

See Section 6.4.

6.6 Permitted Medications

See Section 6.4.

6.7 Adequate Forms of Birth Control

Females must be surgically sterile (by means of bilateral tubal ligation, bilateral salpingectomy, hysterectomy or bilateral oophorectomy), or be post-menopausal (defined as spontaneous cessation of menses for at least 1 year), or, if of childbearing potential, must be practicing abstinence or using a medically acceptable form of contraception (e.g., hormonal contraceptives, intrauterine device, double barrier method [condom, diaphragm or cervical/vault cap with spermicidal foam/gel/film/cream/suppository]) throughout the duration of the study and continued until 2 months following the last dose of study drug.

Males with female partners of child-bearing potential must be practicing abstinence or must agree to use a double barrier form of contraception (e.g., condom with spermicidal foam/gel/film/cream/suppository) throughout the duration of the study and continued until 2 months following the last dose of study drug. This criterion may be waived for male subjects who have had a vasectomy > 6 months before signing the ICF.

6.8 Restrictions on Study Subjects

Water will be restricted (except for water required for study drug dosing) from 1 hour pre-dose through 1 hour post-dose, but will otherwise be permitted ad libitum.

Subjects will be instructed to refrain from consuming grapefruit, grapefruit juice, Seville oranges, Seville orange marmalade, and other products containing grapefruit or Seville oranges from 14 days prior to the first dose administration and for the duration of the study.

Subjects will be instructed to refrain from consuming poppy seeds during the Screening period and while residing in the CRSU. Subjects will be instructed to refrain from consuming caffeine (including caffeine pills, beverages, or any other caffeinated product) for at least 2 hours prior to the first MRI scan until after the last MRI scan is completed.

Subjects will be instructed to refrain from consuming alcohol for 48 hours prior to Screening and throughout their admission to the CRSU.

Subjects are to refrain from smoking (or use of any nicotine-containing substance) throughout their admission to the CRSU. Subjects who feel they will not be able to refrain from nicotine use while confined to the CRSU will be administered a transdermal nicotine patch at a dose consistent with their current nicotine use, which will also be administered between each MRI scan.

Subjects will be instructed to refrain from excess physical activity for the duration of the study.

Subjects will be advised to refrain from blood donation for 30 days after study completion.

6.9 Randomization and Blinding

6.9.1 Assigning Subject Numbers

Each subject will be assigned a unique subject number at Screening after signing the ICF. This subject number will be used on all subjects' study documentation. Subject numbers will not be reassigned. The first 4 digits will be the site number (XXXX) followed by a 4-digit Screening number (YYYY), which together will be the subject number (e.g., XXXX-YYYY). The second set of 4 digits (YYYY) will be sequential, starting with 0001.

6.9.2 Method of Randomization

Treatment assignments will be based on computer-generated randomization schedules prepared by the Sponsor or designee prior to study start. Subjects will be assigned a randomization number on Day 1 prior to dosing and will enter one of the following groups:

Group 1: NKTR-181 and oxycodone IR placebo
Group 2: Oxycodone IR and NKTR-181 placebo

If a subject discontinues from the study, the randomization and/or subject numbers will not be re-used and the subject will not be allowed to re-enter the study, and that subject may be replaced at the discretion of the Sponsor. The replacement subject will follow the same treatment as the original subject randomized. The replacement subject will have the same randomization number as the original subject plus 1000. For example, if the subject with treatment randomization number 1008 discontinued, the replacement subject's randomization number would be 2008 and this subject would follow the treatment assigned to 1008.

6.9.3 Blinding

All study staff, other than the study pharmacist, will be blinded to treatment assignment. The Sponsor will also be blinded to treatment assignment.

Every effort will be made to maintain the integrity of the blind. In certain rare circumstances, wherein knowledge about the study drug received is required to adequately manage serious side effects or AEs a subject experiences, it may be necessary to unblind a subject's treatment regimen before all outcome assessments have been performed. If the Investigator believes that the study blind needs to be broken for a subject, he or she should contact the Medical Monitor, unless there is a medical emergency, **before** the blind is broken. Unblinding will follow the Sponsor's internal procedures for unblinding and associated documentation. If the blind is broken, details surrounding the breaking of the blind (e.g., date, time, and reason) are to be recorded in the subject's study file.

7.0 INVESTIGATIONAL PRODUCT(S)/STUDY MEDICATIONS

7.1 Description and Formulation

Each subject will be administered one capsule of overencapsulated oxycodone IR or placebo and 2 tablets of NKTR-181 or placebo at the time of dosing.

NKTR-181 will be administered orally in a single dose of 400 mg (2 x 200 mg tablet).

Oxycodone IR will be administered orally in a single overencapsulated dose of 40 mg (2x 20 mg tablet).

Refer to the Pharmacy Manual for full dosing instructions.

7.1.1 Packaging and Labeling

NKTR-181 tablets will be provided in blister packs (Phase 3 formulation until 30 November 2019) or To Be Marketed Formulation. The blister packs are inserted into wallets. The wallets are packaged in an aluminum foil pouch containing a desiccant. The wallets and pouches will be labeled with the drug name, quantity and storage conditions. The To Be Marketed formulation will be provided in 60-count high-density polyethylene (HDPE) bottles, induction-sealed with Child Resistant caps; the bottles will be labeled with the drug name, quantity and storage conditions. Pack and bottles will be compliant with United States Food and Drug Administration (FDA) and DEA regulations for a Schedule II, investigational drug product. NKTR-181 matching placebo tablets will be provided in 20-count (until 30 November 2019), or 60-count HDPE bottles, induction-sealed with Child Resistant caps. The bottles will be labeled with the drug name, quantity and storage conditions, and will be compliant with US FDA regulations.

Oxycodone IR tablets will be supplied in the original commercial packaging.

Oxycodone IR matching placebo will be prepared by the pharmacist or designee at the research facility (Section 7.3).

7.2 Study Drug Storage

NKTR-181 is classified as a DEA Schedule II controlled substance. NKTR-181 tablets must be handled, stored, and accounted for in accordance with all federal, state, and local regulations for a Schedule II compound.

NKTR-181 tablets are to be stored at controlled room temperature, i.e., 20°C to 25°C (68°F to 77°F); excursions are allowed from 15°C to 30°C (59°F to 86°F).

Oxycodone IR tablets shall be stored according to the manufacturer's requirements, and handled, stored, and accounted for in accordance with all federal, state, and local regulations for a Schedule II compound.

7.3 Preparation

The pharmacist or designee at the clinical research facility will prepare the individual treatments from the supplies of study drug. The individual treatments may be prepared up to 24 hours in advance of dose administration to the subjects.

7.3.1 NKTR-181 and Placebo Preparation and Dosing

Once removed from the blister pack or bottle, NKTR-181 tablets or matched placebo will be protected from ambient humidity and moisture prior to administration to the subjects.

Each dose will be administered orally with 240 mL of room temperature plain drinking water and the subject will be instructed to drink all of the water. The subjects will be instructed not to crush or chew the NKTR-181 or NKTR-181 placebo tablets. A mouth and hand check will be performed after dose administration to ensure the subject has ingested the study drug.

Subjects should remain in an upright position (e.g., semi-recumbent, sitting, standing, or ambulatory) for at least 1 hour following each dose administration unless another position is required by study procedures. If a restroom visit is needed, subjects should be escorted to the restroom during this period.

7.3.2 Oxycodone IR and Placebo Preparation and Dosing

Oxycodone IR tablets will be overencapsulated within an appropriate size gelatin capsule with microcrystalline cellulose overfill (Avicel, PH102). Each capsule will contain 2 Oxycodone IR 20 mg tablets. The matching oxycodone IR placebo capsule will contain only microcrystalline cellulose.

Additional instructions for preparation of the capsules will be provided in separate pharmacy instructions. The individual doses will be labeled according to standard procedures.

Each dose will be administered orally with 240 mL of room temperature plain drinking water and the subject will be instructed to drink all of the water. The subjects will be instructed not to crush or chew the placebo or overencapsulated oxycodone IR tablets. A mouth and hand check will be performed after dose administration to ensure the subject has ingested the study drug.

Subjects should remain in an upright position (e.g., semi-recumbent, sitting, standing, or ambulatory) for at least 1 hour following each dose administration unless another position is required by study procedures. If a restroom visit is needed, subjects should be escorted to the restroom during this period.

7.4 Accountability and Reconciliation

The Investigator is responsible for ensuring accountability of all study medications supplied and appropriate storage and allocation of these supplies.

The Investigator is responsible for ensuring that all study drugs received at the site are inventoried and accountability performed and that dispensed study drug is recorded in both the electronic case report form (eCRF) and the study drug accountability logs. The Investigator or designee will verify study drug accountability with subjects during site visits. Any discrepancies should be investigated. The Investigator will not supply study drug to any person except those who are subjects in this study and will not dispense study drug from any site other than those listed on Form FDA 1572. Study drug may not be relabeled or reassigned for use by other subjects.

All unused or damaged study drug will be returned and unit counts will be performed whenever medication is returned. The site must account for all medication received. The site will retain and store all study drug until inventoried by the study monitor. All returned, unused, and damaged study drug will be returned to the Sponsor or designee for destruction.

8.0 PHARMACOKINETIC AND PHARMACODYNAMIC ASSESSMENTS

8.1 Pharmacokinetic Assessments

8.1.1 Pharmacokinetic Blood Sample Collection

Pharmacokinetic assessments for NKTR-181 and metabolites oxycodol and oxycodone after NKTR-181 administration and oxycodone after oxycodone IR administration will be performed for all subjects who are dosed in the study. Samples of 4 mL of venous blood will be obtained according to CRSU procedure and collected into dipotassium ethylenediaminetetraacetic acid (K_2 EDTA) tubes at the times indicated in the Schedule of Assessments (Section 1.2). The total amount of blood for PK assessments and safety is as follows (Table 2):

Table 2: Volume of Blood to be Collected during the Study

	Sample Volume (mL)	Number of Samples	Total Volume (mL)
Screening			
Chemistry and pregnancy	8.5	1	8.5
Hematology	4	1	4
Study Period			
Chemistry and pregnancy	8.5	2	17
Hematology	4	2	8
PK samples	4	9	36
End of Study/Early Termination			
Chemistry and pregnancy	8.5	1	8.5
Hematology	4	1	4
Total			**86**

Samples collected within 10% of the specified times will not be considered protocol deviations; however, every attempt will be made by the clinical research facility to collect samples at the specified times. The pre-dose sample should be obtained within 30 minutes prior to dose administration. The exact sample collection times will be recorded to the nearest minute.

8.1.2 Pharmacokinetic Blood Sample Storage and Shipment

All plasma samples will be stored frozen (at -70°C or below with an upper temperature limit of -50°C) until they are shipped to the bioanalytical facility. Prior to shipping, the samples will be packed into thermal insulated containers containing a temperature monitor and packed in sufficient dry ice to assure they remain frozen, and are protected from breakage during shipment. Samples will be shipped by overnight, priority courier with appropriate documentation to identify the samples. The samples will be divided into 2 shipments, each containing 1 aliquot of

plasma for each time point. After receipt of verification that the first shipment was received by the bioanalytical facility, the second shipment (plasma) will be processed and shipped.

Samples will be shipped to the bioanalytical facility according to instructions provided by the Sponsor. The shipping address and contact information will be provided in a separate document. The study site personnel will notify the bioanalytical facility by telephone and/or email prior to the shipment of any samples.

8.1.3 Pharmacokinetic Analysis

A validated liquid chromatography-tandem mass spectrometry (LC-MS/MS) method will be used for the determination of concentrations of NKTR-181 (and selected metabolites) and oxycodone from the plasma samples.

A description of the planned PK analyses will be provided in the Analysis Plan. Details of the Bioanalytical Analysis Plan will be included in the final study report.

8.2 Pharmacodynamic Assessments

8.2.1 MRI assessments

See Section 9.0.

8.2.2 Pupillometry

Pupillometry is used to measure change in pupil size as an indicator of central nervous system mu-opioid pharmacological effects. Single pupil measurements at scheduled times will be performed using a pupillometer per manufacturer's specification. The light in a specifically designed measurement room will be maintained at 3.6 to 4.3 lux, using a Lutron light meter. Subjects will be escorted to the designated room and their eyes will be allowed to adjust to the room lighting for at least 3 minutes prior to any measurement. Pupillometry will be measured using the right eye for all subjects, unless otherwise determined by the Investigator. The same eye will be used for all measurements in the study.

The timing of all pupillometry pharmacodynamic measurements is detailed in the Schedule of Assessments (Section 1.2).

Pupillometry, an objective measure of pupil constriction demonstrating pharmacological effect, will also be performed. Pupillometry measurements may be performed within 10 minutes prior to the scheduled times for assessments up to and including the 1-hour post-dose time point. Pupillometry measurements after the 1-hour time point may be performed within ± 10 minutes of the scheduled time.

Pre-dose PD assessments will occur within 1 hour prior to treatment administration.

9.0 STUDY ASSESSMENTS

9.1 Imaging and Imaging Analysis

MRI data will be collected on Philips Ingenia 3.0T scanner (Best, The Netherlands). Peripheral blood oxygen saturation (SpO_2) will be monitored with an MRI-compatible finger-clip pulse oximeter during the MRI session. In addition, $ETCO_2$ monitoring will be performed via nasal cannula as specified on Table 1. $ETCO_2$ values will be recorded to document any changes, which could influence MRI measurements. fMRI data will be collected with a single-shot gradient echo-echo planar imaging (EPI) sequence (10 minutes in length). Prior to each fMRI run, two EPI images with the same echo time and spacing as the main run are acquired with opposite phase-encode directions (less than 60 second duration). From this scan pair, susceptibility-induced off-resonance field correction is estimated using the method implemented in FSL "topup" software. In addition, peripheral pulse rate and respiratory rate (using standard MRI respiratory chest belt) will be recorded during the fMRI scans for removing artefactual physiological signals using the “retroicor” module of AFNI software. The peripheral pulse rate and respiratory rate will be collected and electronically recorded continuously during the fMRI scans. The “topup” and “retroicor” steps will be conducted using FSL and AFNI because these two signal processing steps are useful for correction of scanning artefacts, but these two steps are not available in the SPM software package. Imaging parameters for perfusion will be done using a pseudocontinuous arterial spin labeling (pCASL) sequence with a single-shot gradient-echo EPI for scan duration of approximately 4.5 minutes (in addition to approximately 5 minutes for MR angiography scans to normalize the pCASL). MR spectroscopy will be performed on the nucleus accumbens region and will include a J-resolved Point-Resolved Spectroscopic Sequence (PRESS). The MRS scan is approximately 4.5 minutes. T1-W structural images (6 min) will be acquired to anatomically register with the sequences outlined above. In addition, a FLuid-Attenuated Inversion Recovery (FLAIR) will be acquired at baseline (3 minutes). FLAIR as well as the T1-W structural imaging will be read by a board-certified neuroradiologist to rule out incidental pathology from the MRI scan. Total scan time at baseline is approximately 37 min with subsequent scans at approximately 34 minutes.

Based on previous PK data for NKTR-181 and oxycodone IR, one MRI scan will be performed during the ascending phase of drug concentration on the PK curve (within the first hour), while the second and third MRI scans will be performed to capture resting-state activity and perfusion at the plateau phase or near C_{max} (2.5-3 hours post-administration). A fourth MRI scan will be performed during the descending phase of drug concentration on the PK curve.

The pCASL data is arterial spin labeling perfusion data (not fMRI data) and will be analyzed using FSL (FMRIB [Functional Magnetic Resonance Imaging of the Brain] Analysis Group, Oxford University, United Kingdom), because SPM does not have the capability of analyzing arterial spin labeling perfusion data. In addition, a normalization strategy outlined in (Aslan, 2010) will be used incorporating phase-contrast velocity MRI to find the labeling efficiency of the technique. MRS data is magnetic resonance spectroscopy data (not fMRI data), and the MRS analysis will be performed by using the Java-based Magnetic Resonance User

Interface (jMRUI) v. 5.0 (http://www.mrui.uab.es/mrui/) because SPM does not have the capability of analyzing magnetic resonance spectroscopy data.

9.1.1 Resting-State Functional Magnetic Resonance Imaging (rsfMRI) Data Preprocessing

The rsfMRI data will be preprocessed using the CONN toolbox (http://www.nitrc.org/projects/conn) (Whitfield-Gabrieli and Nieto-Castanon, 2012), based on Statistical Parametric Mapping 12 (SPM12) software (http://www.fil.ion.ucl.ac.uk/spm/), and Matlab (Mathworks Inc. Sherborn MA, USA).

9.1.2 Dynamic Causal Modeling (DCM) Based Effective Connectivity Analysis

fMRI-based DCM is a biophysical model of how the neuronal connectivity generates the observed fMRI signal (Friston, 2003). In brief, DCM is a system of dynamical bilinear differential state equations with coefficients (in units of Hz) (Friston, 2003). The endogenous connectivity measures the effective connectivity (EC) strengths between nodes, regardless of the moment-to-moment switching on and off of inputs. DCM for resting state fMRI is a special case of above task-based DCM, in which the dynamics of neuronal activity in a DCM node is driven by the neuronal activity in this DCM node, and the neuronal activity in other DCM nodes through the endogenous connectivities. Thus, DCM for resting state fMRI measures ECs which are internal to the system, without effects from external inputs. Spectral DCM (Friston, 2014), which was designed to model resting-state fMRI, will be used to analyze ECs in resting state networks (RSNs).

The candidate nodes used for the DCM EC analysis will be selected based on the reward circuit summarized by Kim (Kim, 2016) from previous studies of morphine addiction and which are within the empirically detected functional connectivity networks for oxycodone resulting from whole brain analysis of the oxycodone data using the SPM12 CONN toolbox (http://www.nitrc.org/projects/conn) (Whitfield-Gabrieli and Nieto-Castanon, 2012), based on Statistical Parametric Mapping 12 (SPM12) software (http://www.fil.ion.ucl.ac.uk/spm/), and Matlab (Mathworks Inc. Sherborn MA, USA). In addition, candidate nodes will be included which are within the empirically detected functional connectivity networks for NKTR-181 resulting from the whole brain analysis of the NKTR-181 data using the SPM12 CONN toolbox. Both the whole brain oxycodone functional connectivity CONN analysis and the whole brain NKTR-181 functional connectivity CONN analysis will use the AICHA atlas (atlas of intrinsic connectivity of homotopic areas) (Joliot, 2015), which contains 384 nodes throughout the whole brain.

Group comparison of the ECs at baseline. The Parametric Empirical Bayes (PEB) approach (Friston, 2016), as implemented in SPM12, will be used to evaluate group effects in the DCM parameters (e.g., if a parameter is different from zero in a single group, or if a parameter is different between groups). PEB allows for empirical updating of priors for each subject during the DCM analysis. After specifying a second level (group) general linear model (GLM) and a

design matrix (with ones for the first subject group and minus ones for the second subject group), the second level (group) PEB model is estimated using the SPM12 code (spm_dcm_peb.m). The estimated PEB model contains all parameters representing group effects on each DCM parameter. After that, the group effects are identified using the SPM12 code (spm_dcm_peb_bmc.m), which prunes away any parameters from the PEB model which don't contribute to the model evidence.

Group comparison of the study drug-related altered ECs over time. In order to measure the effects of medication at each time point, the baseline rsfMRI and the rsfMRI scans for timepoint x are concatenated. After that, an experimental condition (regressor) called "medication_x – baseline" is created and used as a single modulator of EC. Here x refers to the x-th subsequent rsfMRI scan. The effect of medication will be reflected by the estimated value of the x-baseline modulator (DCM B matrix). The DCM analysis will be repeated for each x-th rsfMRI scan. The remaining steps are the same as those described in the previous paragraph to compare the value of the x-baseline modulator between medication groups.

10.0 ASSESSMENT OF SAFETY AND ADVERSE EVENTS AND SERIOUS ADVERSE EVENTS

10.1 Adverse Events and Serious Adverse Events

10.1.1 Adverse Event Definition and Assessment

An AE is defined as any untoward medical occurrence in a subject who is administered a medicinal product and that does not necessarily have a causal relationship with this treatment.

An AE can therefore be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of a medicinal product, regardless of whether it is considered related to the medicinal product. An AE can also arise from any use of the drug and from any route of administration, formulation, or dose. This definition includes intercurrent illnesses or injuries, and exacerbation of preexisting conditions as well as events attributed to protocol-mandated procedures. Clinical laboratory abnormalities will only be reported as AEs if they are deemed clinically significant by the Investigator and/or are associated with signs and symptoms, require treatment, or require follow-up.

An AE does not include the following:

- A medical or surgical procedure (e.g., surgery, endoscopy, tooth extraction, or transfusion); an AE is the underlying condition that leads to the procedure.
- Preexisting diseases or conditions present or detected before the start of study drug administration that do not worsen or increase in severity or frequency after the administration of study drug.
- Situations where an untoward medical occurrence has not occurred (e.g., hospitalization for elective surgery for a condition that has not worsened on study, social and/or convenience admissions to grant families a respite in caring for a subject).

10.1.2 Monitoring Adverse Events

All AEs will be assessed by the Investigator and recorded on the eCRF, including but not limited to the following: the event term, the date of onset and resolution, seriousness, severity, relationship to study drug, outcome, treatment of the event, and action taken with the study drug. Adverse events will be reported starting immediately after the subject has provided written informed consent through completion of the EOS visit.

Any AE attributed to a protocol-mandated procedure occurring after the subject has provided informed consent but prior to the first dose of study drug should be recorded on the eCRF as an AE.

Adverse events that are not attributed to protocol-mandated procedures occurring after signing of the informed consent but before the first dose of study drug will be reported as medical history.

Example 1:

Thrombophlebitis associated with a blood draw for assessments required prior to dosing per protocol is an event that is related to protocol-mandated procedures. In this scenario, the event of "thrombophlebitis" will be captured as an AE on the eCRF page, and it will be documented as being "unrelated" to study drug as applicable.

Example 2:

An ankle sprain following an unexpected fall from a flight of stairs while at home, after the subject has provided informed consent, but before the first dose of study drug, is clearly unrelated to any protocol-mandated procedures and would therefore be captured as medical history in the medical history section of the eCRF.

10.1.3 Grading of Adverse Events

The severity of an event and the seriousness are not to be considered synonymous. The severity is grading the intensity of an event. The seriousness of an event is based on the subject/event outcome (Section 10.1.6). All AEs will be assessed for severity by the Investigator using the following criteria:

- Mild: The event results in mild or transient discomfort, not requiring or needing only minimal intervention or treatment; does not limit or interfere with daily activities (e.g., insomnia, mild headache);
- Moderate: The event is sufficiently discomforting so as to limit or interfere with daily activities; may require interventional treatment (e.g., fever requiring antipyretic medication); and
- Severe: The event results in significant symptoms that prevent normal daily activities; may require hospitalization or invasive intervention.

Adverse events will be reported with an individual start and stop date for each level of severity.

10.1.4 Causality Relationship of Adverse Events

The relationship of each AE to the study drug will be evaluated by the Investigator using the following definitions:

- Not related: The AE is considered not related to the study drug(s). The AE can be reasonably explained by other factors such as the subject's preexisting medical condition, underlying disease, concurrent illness, or concomitant medications/therapies;

- Related: There is reasonable possibility that an AE is caused by the study drug(s). A plausible temporal sequence exists between the time of administration of the study drug(s) and the development of the AE. The AE cannot be reasonably explained by the known characteristics of the subject's clinical state or other concomitant therapies or interventions administered to the subject.

10.1.5 Adverse Event Reporting and Follow-up

All ongoing AEs must be followed until resolution or until the EOS visit, whichever comes first. If the AE has not completely resolved by the EOS visit, the final outcome of these ongoing AEs will be captured as "Not Recovered/Not Resolved" or "Recovering/Resolving", whichever is applicable.

For specific instructions on identifying and reporting SAEs, see Sections 10.1.6 and 10.1.7.

10.1.6 Serious Adverse Event Definition

An SAE is defined as any untoward medical occurrence that at any dose:

- Results in death;
- Is life-threatening, i.e., in the opinion of the Investigator or Sponsor, the AE places the subject at immediate risk of death from the event as it occurred; 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 that occurs during the course of a subject's participation in a clinical study, except for those that occur due to any of the following:
  - A surgery or procedure that was planned or anticipated before the subject entered the study and that is part of the planned study procedure; or
  - Nonmedical reasons, in the absence of an AE;
- Results in persistent or significant disability or incapacity;
- Is a congenital anomaly or birth defect; or
- Is an important medical event that, based upon appropriate medical judgment, may jeopardize the subject and may require medical or surgical intervention to prevent one of the other outcomes listed above.

Death is an outcome of an AE, and not an AE in itself. All fatal events regardless of causality must be reported. "Life-threatening" means that the subject was at immediate risk of death from the event as it occurred. This does not include an event that might have led to death, if it had occurred with greater severity. "Inpatient hospitalization" means that the subject has been admitted to a hospital for medical reasons for any length of time. The Investigator should attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information.

In such cases, the diagnosis should be documented as the AE and/or SAE and not the individual signs/symptoms.

10.1.7 Serious Adverse Event Reporting

All SAEs regardless of causality attribution occurring during the conduct of the study should be reported to Nektar Therapeutics Drug Safety or its designee within **24 hours** of when the site becomes aware of the event.

Follow-up reports and any additional records (such as hospital records, consultant reports, and autopsy findings) should be emailed or faxed to Nektar Therapeutics Drug Safety or designee within **24 hours** of when the site becomes aware of the additional information.

The Investigator must complete the SAE form, assess the causality relationship to the study drug, as applicable, and send the completed SAE form via email or fax to Nektar Therapeutics Drug Safety or designee.

Any medication or other therapeutic measures used to treat the event will be recorded.

All SAEs will be followed as described in Section 10.1.8.

Reporting of SAEs to the Institutional Review Board (IRB) will be done in accordance with the standard operating procedures (SOPs) and policies of the IRB. Adequate documentation must be provided to the Sponsor, showing that the IRB was properly notified. Serious AEs will be reported by the Sponsor or designee to the regulatory authorities, per local regulations.

10.1.8 Serious Adverse Event Follow-up

All SAEs will be followed until resolution, stabilization of condition, return to baseline, or until follow-up is no longer possible.

Any SAE occurring after the end of the study will be captured only if assessed by the Investigator as related to study drug.

10.1.9 Expedited Reporting of Serious Adverse Events

A suspected unexpected serious adverse reaction (SUSAR) is an SAE that is considered "unexpected" and is assessed by the Investigator or the Sponsor as related to the study drug. All SAEs deemed related to the study drug and not expected based on the most current NKTR-181 Investigator's Brochure or oxycodone IR package insert are subject to expedited reporting to the applicable regulatory authorities by the Sponsor. Therefore, the Investigator or site personnel must report all SAEs to Nektar Therapeutics Drug Safety or its designee within **24 hours** of first becoming aware of the event.

Fatal or life-threatening SUSARs will be reported to the regulatory authorities by the Sponsor as soon as possible but no later than 7 calendar days after the Sponsor or its designee has first

knowledge of the minimum criteria for expedited reporting, with a full written report within 8 calendar days later. Nonfatal or nonlife-threatening SUSARs will be reported to the regulatory authorities, IRBs, and Investigators as soon as possible but no later than 15 calendar days after the Sponsor or its designee has first knowledge of the SUSAR.

Reporting of SUSARs to all applicable regulatory authorities will be made by Nektar Therapeutics Drug Safety or its designee as per local country and regional regulations.

Notification of SUSARs to the central IRBs will be made by Nektar Therapeutics Clinical Operations or its designee in accordance with the SOPs and policies of the IRBs. Reporting to local IRBs will be made by the applicable study site personnel per their institutional guidelines. Adequate documentation must be provided to Nektar Therapeutics Clinical Operations or its designee showing that the local IRB was properly notified.

Reporting of SUSARs to all participating clinical Investigators will be done by Nektar Therapeutics Drug Safety or its designee per local regulations.

10.2 Special Reporting Situations

10.2.1 Pregnancy

The Sponsor must be notified within **24 hours** of the initial report and any follow-up reports of a female subject or male subject's female partner becoming pregnant during the course of the study and for 2 months after the last dose of the study drug. Pregnancy, although reportable, is not considered an AE/SAE unless a female subject or male subject's female partner experiences signs or symptoms of pregnancy complications; however, the contact information for pregnancy reporting is the same as for SAE reporting and listed in List of Study Contacts. Females who become pregnant will be followed every trimester until the outcome of the pregnancy is known. Pregnancy follow-up should describe the outcome of the pregnancy, including any voluntary or spontaneous termination, details of the birth, and the presence or absence of any congenital abnormalities or birth defects in the offspring.

10.2.2 Potential Drug Induced Liver Injury

Wherever possible, timely confirmation of initial liver-related laboratory abnormalities should occur prior to the reporting of a potential drug induced liver injury (DILI) event. All occurrences of potential DILIs, meeting the defined criteria, must be reported as SAEs (see Section 10.1.7 for reporting details).

Potential drug induced liver injury is defined as:
Aminotransaminases (ALT or AST) elevation > 3 times upper limit of normal (ULN)
AND
total bilirubin > 2 times ULN, without clinical jaundice or initial findings of cholestasis (e.g., elevated serum alkaline phosphatase),
AND

No other immediately apparent possible causes of AT elevation and hyperbilirubinemia, including, but not limited to, viral hepatitis, pre-existing chronic or acute liver disease, or the administration of other drug(s) known to be hepatotoxic.

10.3 Clinical Laboratory Tests

Clinical laboratory tests will be conducted according to the Schedule of Assessments (Section 1.2). Clinical laboratory tests will be performed by the clinical site's laboratory. The site will be provided instructions for specimen collection, processing, storage, packaging and shipment. Laboratory tests results will be provided to the Sponsor by the site.

Clinical laboratory test data will be reviewed by the Investigator or qualified sub-Investigator. Additional clinical laboratory tests may be ordered at the Investigator's or qualified sub-Investigator's discretion. All additional testing will be performed by the designated central laboratory.

The Investigator or qualified sub-Investigator will review all lab results for clinical significance. Any laboratory result deemed clinically significant will be recorded as an AE as described in Section 10.1.

See Table 2 for blood volumes collected in the study.

Clinical laboratory tests that will be conducted for this study are listed in Table 3.

Table 3: Clinical Laboratory Tests

Hematology	Chemistry	Urinalysis
White blood cell (WBC) count Red blood cell (RBC) count Hemoglobin Hematocrit Mean corpuscular volume (MCV) Mean corpuscular hemoglobin (MCH) Mean corpuscular hemoglobin concentration (MCHC) Platelet count WBC differential: • Neutrophils (absolute) • Lymphocytes (absolute) • Monocytes (absolute) • Eosinophils (absolute) • Basophils (absolute)	Glucose Blood urea nitrogen (BUN) Creatinine Sodium Potassium Chloride Carbon dioxide (CO_2) content or bicarbonate Calcium Total protein Albumin Total bilirubin Alkaline phosphatase Aspartate aminotransferase (AST) Alanine aminotransferase (ALT) Gamma-glutamyl transferase (GGT) Lactate dehydrogenase (LDH)	Specific gravity pH WBC esterase Protein Glucose Ketones Blood Bilirubin Nitrites For positive protein, WBC esterase, or blood, a microscopic examination will include the following: • WBC • RBC • Epithelial cells • Bacteria • Crystals • Casts
Substances of Abuse Screen	**Pregnancy**	
Urine Amphetamines (including methamphetamine and Ecstasy) Barbiturates Benzodiazepines Buprenorphine Cannabinoids Cocaine Methadone Phencyclidine (PCP) Opiates Oxycodone BreathEthanol (Breathalyzer)	Serum β-hCG (quantitative) at screening and Day -1 Urine pregnancy test on Day 1	

10.4 Vital Signs and Continuous Pulse Oximetry

Vital sign measurement will be recorded at Screening and daily while subjects are admitted to the CRSU or CARI Clinic. Vital signs include systolic and diastolic blood pressure, heart rate, respiratory rate, and SpO_2. Blood pressure and heart rate measurements will be taken after the subject has been supine for at least 5 minutes. Continuous pulse oximetry monitoring will begin

approximately 1 hour prior to first study drug dose and will continue for approximately 8 hours after study drug administration. Continuous pulse oximetry (which is separate from episodic pulse oximetry collected with other vital sign measurements according to the Schedule of Assessments), will be performed for monitoring purposes or for processing of MRI data only; these data will not be collected in the electronic data capture system but will be captured and saved with MRI data at the site. Pulse oximetry data used to estimate brain perfusion will be captured by the site in the electronic data capture system. If a subject experiences oxygen desaturation, additional pulse oximetry values and/or vital signs will be captured as unscheduled assessments. $ETCO_2$ will be captured from pre-dose until Hour 8 to document changes which could influence the results. Any AEs associated with abnormal vital sign or pulse oximetry findings will be recorded.

10.5 Twelve-Lead Electrocardiograms

Twelve-lead Screening ECGs will be performed on a calibrated 12-lead machine according to the Schedule of Assessments (Section 1.2). Subjects must be resting quietly in the supine position for at least 5 minutes before the 12-lead ECG. The Investigator or qualified sub-Investigator will review all ECG interpretations and interval duration measurements for clinical significance. Any males with QTcF > 450 msec and females with QTcF > 470 msec will not be enrolled to the study.

10.6 Medical History

A detailed medical, surgical, and medication history will be recorded for each subject to confirm eligibility for the study. Clinically significant medical conditions or conditions relevant to the study that are ongoing will be recorded. The medication history must include any known drug allergies, present abuse or history of drug abuse, and use of acute or chronic medications. Demographic information, i.e., birth date, sex, and race/ethnicity, will be recorded for all subjects.

10.7 Drug and Alcohol Screening

Subjects will undergo an alcohol breath test at the Screening visit, prior to admission to the CRSU, and at the MRI facility prior to MRI scanning. Subjects who have a positive alcohol breath test immediately prior to admission to the CRSU will not be allowed to participate further and will be removed from the study. Disposable, individual use alcohol breathalyzers will be provided by the Sponsor.

Urine samples will be collected for drug screening at Screening and on Day -1. Samples will be screened for multiple substances of abuse (see Table 3). Dipstick urine drug screen tests will be provided by the Sponsor.

10.8 Unblinding of Study Drug

Every effort will be made to maintain the integrity of the blind. In certain rare circumstances, wherein knowledge about the study drug received is required to adequately manage serious side effects or AEs a subject experiences, it may be necessary to unblind a subject's treatment regimen before all outcome assessments have been performed. If the Investigator believes that the study blind needs to be broken for a subject, he or she should contact the Medical Monitor, unless there is a medical emergency, before the blind is broken. Unblinding will follow the Sponsor's internal procedures for unblinding and associated documentation. If the blind is broken, details surrounding the breaking of the blind (e.g., date, time, and reason) are to be recorded in the subject's study file.

10.9 Subject Replacement

The decision to enroll replacement subjects will be made at the discretion of the Sponsor. If any subjects discontinue from the study prior to completion of Day 1 MRI, PK, and PD procedures, enrollment will continue to ensure 24 subjects complete these procedures. The decision made to add replacement subjects will be documented by the Sponsor prior to adding these subjects.

10.10 Precautions

The dose levels of NKTR-181 and oxycodone IR used in this study could cause respiratory depression. This effect is antagonized by naloxone (Narcan®). The clinical research facility must have an adequate supply of naloxone readily available on-site in anticipation of the simultaneous occurrence of respiratory depression in multiple subjects. More than 1 dose of naloxone may be necessary since the duration of action of NKTR-181 or oxycodone IR will likely exceed that of the antagonist. In the event of an emergency, equipment to ventilate subjects, including a bag valve mask, must also be available. Additional information regarding AEs observed in other clinical studies for NKTR-181 is provided in the NKTR-181 IB (Nektar Therapeutics, 2018).

11.0 STATISTICAL METHODS PLANNED IN THE PROTOCOL AND DETERMINATION OF SAMPLE SIZE

11.1 General Considerations

Generally, if not specified, continuous data will be summarized by descriptive statistics, including sample size, mean, standard deviation, median, and range. Categorical data will be summarized by the number and percentage of subjects. Data analysis will be performed using SAS® version 9.4 or greater.

Details of the analysis will be defined in the Statistical Analysis Plan and PK Analysis Plan.

An informal data analysis may be performed prior to study completion.

11.2 Determination of Sample Size

No formal sample size calculation has been made. Based on experience from previous studies, a total of 24 evaluable subjects (12 per arm) is considered sufficient for the study. An evaluable subject is defined as a subject who completes sufficient fMRI and PK/PD measurements for NKTR-181 or oxycodone IR to allow analysis.

11.3 Analysis Populations

The safety population will consist of all subjects who receive a dose of study drug.

There will be two PD populations, one for pupillary endpoints and the other for fMRI endpoints. The pupillary PD population will consist of all subjects who received study drug and have at least one pupillary determination. The fMRI PD population will consist of all subjects who received study drug and have sufficient MRI data for NKTR-181 or oxycodone IR treatment to allow description of modulation of brain circuitry.

The PK population will consist of all subjects who have sufficient plasma concentration data to facilitate the calculation of at least one PK parameter such as area under the curve (AUC) or C_{max} as determined by the pharmacokineticist.

11.4 Subject Disposition

Subject disposition will be summarized for subjects who complete the study or discontinue early. The overall summary will comprise the percentage of subjects who completed the study and discontinued early with reasons for discontinuation.

11.5 Demographic and Baseline Disease Characteristics

Demographics including age, race, ethnicity, weight, height, and BMI will be summarized and also listed in by-subject listings.

11.6 Prior and Concomitant Medications

Prior and concomitant medications will be reported at each visit and coded to Anatomical, Therapeutic, or Chemical (ATC) level and preferred term (PT) according to the World Health Organization drug dictionary. Prior and concomitant medications will be summarized by treatment group, ATC classification, and PT.

11.7 Pharmacokinetic Analysis

All PK summaries and analyses will be performed on the PK population. The PK parameters such as C_{max}, time to maximum concentration (T_{max}), and area under the concentration time curve from time zero to the last measurable concentration ($AUC_{0\text{-}last}$) will be estimated for NKTR-181 and its metabolites and oxycodone from the plasma concentration-time data using standard non-compartmental methods. Actual sample times (hours, relative to the corresponding administration time) will be used in the computation of the PK variables, rather than scheduled times.

Individual subject plasma PK concentration data and PK parameters will be tabulated and summarized by treatment group using descriptive statistics.

11.8 Functional MRI Analysis

The effective connectivity analyzed will be summarized separately for NKTR-181 and oxycodone.

The effects of study drug (NKTR-181 or oxycodone IR) at each time point through DCM analysis will be summarized separately for NKTR-181 and oxycodone and included in data listings.

Perfusion differences between treatments over time after drug administration may be modeled using ROI-based measurements made in regions in the brain, if appropriate.

11.9 Pupillometry Data Analysis

Data from pupillometry will be summarized by treatment using descriptive statistics. The time course of treatment mean of PD endpoints will be displayed graphically to assess differences between treatment groups over time. If appropriate, the peak effect for change in pupil size may be explored.

11.10 Safety Analysis

The safety analysis will be based on the Safety population. A treatment-emergent adverse event (TEAE) is defined as an AE whose onset is on or after the initiation of the first dose of the treatment phase. The frequency of treatment-emergent AEs will be tabulated using the Medical Dictionary for Regulatory Activities (MedDRA) by system organ class (SOC) and PTs as well as

by severity and relationship to treatment. All AEs, serious AEs and AEs resulting in study discontinuation will be included in the data listings.

Vital sign measurements, ECGs, and clinical laboratory tests will be summarized using descriptive statistics. Individual laboratory test results that are outside the normal range for that test will be flagged as high (H) or low (L), as appropriate.

11.11 Missing Data and Imputation

No missing data imputation is planned unless otherwise specified in the Statistical Analysis Plan.

12.0 STUDY OR STUDY SITE TERMINATION

The Sponsor has the right to suspend or terminate the study at any time. The study site may be suspended or terminated for any reason.

13.0 QUALITY CONTROL AND QUALITY ASSURANCE

The Sponsor will implement and maintain quality control and quality assurance procedures with written SOPs to ensure that the study is conducted and data are generated, documented, and reported in compliance with the protocol, Good Clinical Practice (GCP), and applicable regulatory requirements.

All protocol deviations and the reasons for such deviations are to be documented in the source documents and reported to the Sponsor.

13.1 Changes to the Protocol

The Investigator may not deviate from the protocol without a formal protocol amendment having been established and approved by an appropriate IRB, except when necessary to eliminate immediate hazards to the subject. Any deviation may result in the subject having to be withdrawn from the study and rendering that subject nonevaluable.

13.2 Monitoring

In accordance with Code of Federal Regulations 21 CFR 312.56, International Council for Harmonisation (ICH) GCP and local regulations, the clinical monitor will periodically inspect all eCRFs, study documents, research facilities, and clinical laboratory facilities associated with this study at mutually convenient times during and after completion of the study. As required by 21 CFR 312 Subpart D (Responsibilities of Sponsors and Investigators), ICH GCP, and local regulations, the monitoring visits provide the Sponsor with the opportunity to evaluate the progress of the study; verify the accuracy and completeness of eCRFs; ensure that all protocol requirements, applicable FDA, ICH GCP, and local regulations, and Investigator's obligations are being fulfilled; and resolve any inconsistencies in the study records. This includes inspection of all documents and records that are required to be maintained by the Investigator, including but not limited to medical records (office, clinic, or hospital) for the subjects in this study. The names and identities of all research subjects will be kept in strict confidence and will not appear on eCRFs or other records provided to or retained by the Sponsor. The Investigational New Drug Application (IND) regulations also require the Investigator to allow authorized representatives of the Sponsor, IRB/IEC, and FDA to inspect and make copies of the same records. The names and identities of the subjects need not be divulged to the Sponsor; however, the records must nevertheless be inspected. This can be accomplished by blacking out the subject's name and replacing the name with the subject's study identification number. If these requirements are in conflict with local regulatory restrictions or institutional requirements, the Investigator must inform the Sponsor of these restrictions before initiation of the study.

13.3 Direct Access to Source Data/Documents for Audits and Inspections

Members of the Sponsor's GCP Quality Assurance Department or designees may conduct an audit of a clinical site at any time during or after completion of the study. The Investigator will be informed if an audit is to take place and advised as to the scope of the audit. Inspections and

audits are typically carried out during the clinical and reporting phases of this study to ensure that the study is conducted and data are generated, documented, and reported in compliance with the protocol, GCP, written SOPs and applicable laws, rules, and regulations.

Representatives of the FDA or other regulatory agencies, including IRB/IEC representatives may also conduct an audit of the study. If informed of such an inspection, the Investigator should notify the Sponsor immediately. The Investigator will ensure that the auditors have access to the clinical supplies, study site facilities, and laboratory, and that all data (including original source documentation) and all study files are available, if requested.

14.0 ETHICS

This study will be conducted to be consistent with the principles that have their origin in the Declaration of Helsinki and in accordance with FDA regulations (21 CFR § 11, 50, 54, 56, and 312), with the ICH GCP guidelines (ICH E6), as well as with any and all applicable federal, state, and/or local laws and regulations.

14.1 Institutional Review Board/Independent Ethics Committee Approval

Before enrollment of subjects into the study, as required by federal regulations (21 CFR § 56), ICH GCP, and local regulations, the current protocol and ICF will be reviewed and approved by an appropriate IRB or IEC. A letter documenting the IRB or IEC approval must be received by the Sponsor before the initiation of the study at a clinical site. Amendments to the protocol will be subject to the same requirements as the original protocol.

The Investigator will submit a progress report at least once yearly to the IRB or IEC. However, the frequency of these reports will depend on IRB or IEC requirements. As soon as possible after completion or termination of the study, the Investigator will submit a final report to the IRB or IEC per the IRB or IEC requirements, and in compliance with FDA regulations and ICH GCPs.

The Investigator, the Sponsor, or designee shall promptly notify the IRB or IEC of any SAEs, or any other information that may affect the safe use of the study drug during the course of the study, per the IRB or IEC local requirements, and in compliance with FDA regulations and ICH GCPs.

14.2 Written Informed Consent

Written informed consent must be obtained from each subject or subject's legal representative before entering the study. Subjects/legal representatives will be informed of the nature of the study, and the ICF must be presented to each subject/legal representative in the language in which the subject/legal representative is fluent.

Informed consent will be obtained and documented by each subject or subject's legal representative prior to any protocol-specified procedures. Signed ICFs will be retained by the Investigator with the study records. Each subject/legal representative will be given a copy of the signed and dated ICF.

15.0 DATA HANDLING AND RECORD KEEPING

15.1 Data Collection Instruments and Source Documents

15.1.1 Study Records

During the study, the Investigator will maintain adequate records for the study, including a record of potential subjects screened and medical records, and records detailing the progress of the study for each enrolled subject, laboratory reports, eCRFs, signed ICFs, drug accountability records, correspondence with the IRB or IEC and regulatory agencies' AE reports, and information regarding subject discontinuation and completion of the study.

15.1.2 Data Collection Instruments

Data collection instruments (DCIs) (e.g., eCRFs, electronic clinical outcomes assessments [eCOA], paper forms) will be used in this study. These instruments are used to transmit the information collected during the performance of this study to the Sponsor or Sponsor's designee and regulatory authorities. The Investigator must review the DCIs for completeness and accuracy and must approve all data, including any changes made. Furthermore, the Investigator retains full responsibility for the appropriateness and accuracy of all data collected in the DCIs.

15.2 Retention of Essential Documents

All records and documents pertaining to the study including, but not limited to, those outlined above (see Section 15.1.1) will be maintained by the Investigator for a period of at least 2 years after FDA approval of the drug or at least 2 years after withdrawal of the IND under which this study was conducted, whichever is longer. To avoid any possible errors, the Investigator will contact the Sponsor before transferring or destroying any study records. The Investigator will also promptly notify the Sponsor in the event of accidental loss or destruction of any study records.

15.3 Confidentiality

Subject confidentiality will be maintained per local legal and regulatory requirements and applicable US federal regulations and ICH GCP guidelines. To comply with GCP guidelines and requirements, subject records will be reviewed during monitoring visits and audits conducted by the Sponsor, Sponsor's representatives, or health authorities. During these activities, every reasonable effort will be made to keep medical information, including subject identifying information, as confidential as possible as required by law.

15.4 Security Measures

Sites will employ both technical and organizational measures (such as, but not limited to, controlling access to personal subject data to only those with a need to know such data, data encryption, data anonymization and pseudonymization, and so forth) to ensure subject and

subject data privacy. Sites will adhere to a “privacy by design” and “privacy by default” approach in collecting, storing, and processing personal subject data.

In the event of a breach of the security measures used by the site to ensure subject and subject data privacy, the site will immediately notify the Sponsor.

16.0 PUBLICATION POLICY

All data are the property of the Sponsor. However, it is intended that the results of the study will be published and/or presented at scientific meetings. Any formal presentation or publication of data from this study will be considered as a joint publication by the Investigator(s) and appropriate Sponsor personnel. Authorship will be determined by mutual agreement.

The Sponsor must receive copies of any intended communication in advance of submission for publication (at least 14 days for an abstract or oral presentation and 30 days for a journal submission). The Sponsor will review the communications for accuracy (thus avoiding potential discrepancies with submissions to health authorities), verify that confidential information is not being inadvertently divulged, and provide any relevant supplementary information.

The Investigator may be required to sign the clinical study report if it is to be used in a registration submission to the health authorities of some countries.

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