Cover Page for Protocol

Sponsor name:	Dicerna Pharmaceuticals, Inc.
NCT number	NCT05845398
Sponsor trial ID:	DCR-AUD-102
Official title of study:	A Phase 1b, Double-blind, Placebo-controlled, Repeat-dose, Safety, Tolerability, Pharmacokinetic, and Pharmacodynamic Study of DCR-AUD in Healthy Volunteers
Document date*:	17-January-2023

*Document date refers to the date on which the document was most recently updated.

Dicerna™

Protocol Title:	A Phase 1b, Double-blind, Placebo-controlled, Repeat-dose, Safety, Tolerability, Pharmacokinetic, and Pharmacodynamic Study of DCR-AUD in Healthy Volunteers
Protocol Number:	DCR-AUD-102
Compound:	DCR-AUD
Study Phase:	1b
Short Title:	Phase 1b Repeat Dose Study of DCR-AUD in Healthy Volunteers
Pre-IND Number:	152401
Sponsor Name:	Dicerna Pharmaceuticals, a Novo Nordisk Company 75 Hayden Avenue Lexington, MA 02421 US (617) 621-8097
Version and Date:	2.0, 17-Jan-2023

[redacted]

Sponsor Signature Page

Title: A Phase 1b, Double-blind, Placebo-controlled, Repeat-dose, Safety, Tolerability, Pharmacokinetic, and Pharmacodynamic Study of DCR-AUD in Healthy Volunteers

Protocol Number: DCR-AUD-102

Version: 2.0

Date: 17-Jan-2023

DocuSigned by:
[redacted]
17-Jan-2023
Date

DocuSigned by:
[redacted]
18-Jan-2023
Date

DocuSigned by:
[redacted]
17-Jan-2023
Date

Medical Monitor and Pharmacovigilance Contact Information

SAE Submission:

Novo Nordisk:

Email: soclin@novonordisk.com

24-hour Safety Fax: +45 44427787

Clinical Research Organization Medical Monitor:

[redacted]

Sponsor Medical Lead and Monitor:

[redacted]

PROTOCOL AMENDMENT SUMMARY OF CHANGES

DOCUMENT HISTORY	
Document	**Date**
Amendment 1	17-Jan-2023
Original Protocol	06-Dec-2022

Amendment 1 (17-Jan-2023)

Overall Rationale for the Amendment:

The protocol was amended in response to queries from the US Food and Drug Administration. Additional updates and clarifications were also included. A summary of all changes follows in the table below.

Description of Change	Brief Rationale	Section # and Name
Clarified that, following an SAE, additional EIAs will not be performed if the SAE may have been related to study intervention and that repeat EIAs may pose a recurrent risk to the participant.	To enhance participant safety and allow for PI discretion in the conduct of EIAs following an SAE.	7.3.1 Serious Adverse Event
Decrease from 3 to 2 the number of participants experiencing a Grade 3 CTCAE event that would trigger a study stopping rule.	To enhance participant safety.	7.4. Study Stopping Rules
Require that participants remain in clinic until the 24-hour postdose PK blood sample is collected (increases in-clinic admission by one night). Also results in change of timing for distribution of alcohol use symptom diary.	To simplify clinical operations and reduce the likelihood of missed sample collection.	1.3. Schedule of Activities
Changed pre-admission alcohol testing from urine to breath.	Breath-alcohol testing provides a more accurate assessment of recent alcohol intake than does urine testing.	1.3. Schedule of Activities 5.3. Lifestyle Considerations 8.6.1. Ethanol Interaction Assessments 10.2. Clinical Laboratory Tests
Added that a positive urine drug screen on Day −1 (in addition to at Screening) will result in study exclusion.	To ensure that participants are not users of illicit drugs.	1.3. Schedule of Activities 5.2. Exclusion Criteria
Updated text in Exclusion Criterion #3 (history of suicidal ideation or behavior).	To provide clarification on the timeframe during which positive answers in the C-SSRS would result in study exclusion.	5.2. Exclusion Criteria

Description of Change	Brief Rationale	Section # and Name
Added that metabolites of DCR-AUD may also be analyzed	To provide for a more complete PK profile of DCR-AUD	8.5 Pharmacokinetics
Changed sampling times for blood draws for ADA detection from Days 29 and 57 to Days 42 and 70.	To provide for ADA sample collection 14 days after each dose administration.	1.3. Schedule of Activities
Changed timing of *ALDH2* genotyping from Screening to Day −1.	To allow that only eligible participants undergo genotyping.	1.3. Schedule of Activities

TABLE OF CONTENTS

Table of Tables

Table of Figures

1. PROTOCOL SUMMARY

1.1. Synopsis

Protocol Title: A Phase 1b, Double-blind, Placebo-controlled, Repeat-dose, Safety, Tolerability, Pharmacokinetic, and Pharmacodynamic Study of DCR-AUD in Healthy Volunteers

Short Title: Phase 1b Repeat Dose Study of DCR-AUD in Healthy Volunteers

1.1.1. Rationale

DCR-AUD is being developed for the treatment of alcohol use disorder (AUD) in adults using an RNA interference (RNAi) technology platform. DCR-A1203, the drug substance of DCR-AUD, is a synthetic double-stranded (annealed duplex) RNA oligonucleotide conjugated to GalNAc ligands that enable subcutaneous delivery of stable, highly specific, and long-acting knockdown of mRNA in the liver via hepatic GalNAc carbohydrate receptors. The target of DCR-AUD is aldehyde dehydrogenase 2 (ALDH2), one of the primary enzymes required for alcohol metabolism. Alcohol metabolism occurs predominantly in the liver via a 2-step process in which alcohol is first converted to acetaldehyde via alcohol dehydrogenase and then to acetic acid by ALDH2. ALDH2 enzyme inhibition can result in unpleasant symptoms after alcohol consumption such as flushing, headache, lightheadedness, palpitations, nausea, and vomiting, as a result of unmetabolized acetaldehyde accumulation. Unpleasant reactions to alcohol consumption in humans with naturally occurring *ALDH2* mutations are thought to be protective against AUD and account for its low prevalence in individuals who are heterozygous or homozygous for *ALDH2* allele mutations causing slower metabolism of acetaldehyde. The magnitude and duration of acetaldehyde accumulation is proportional to both the degree of ALDH2 enzyme inhibition (e.g., partial vs total) and the amount of alcohol consumed.

DCR-AUD is designed to selectively reduce *ALDH2* mRNA and ALDH2 activity in the liver, and thereby decrease the conversion of acetaldehyde to acetic acid after ethanol ingestion. In cynomolgus monkeys, a single SC dose of 3 mg/kg DCR-AUD resulted in reduction of *ALDH2* mRNA expression in liver up to 112 days postdose, with maximum reduction observed at 4 weeks postdose.

This repeat-dose study is being conducted to characterize the safety and tolerability and PK profiles of DCR-AUD when administered monthly, and to determine if a PD effect (resulting from cumulative *ALDH2* mRNA knock down) can be observed during Ethanol Interaction Assessments (EIA) in a population of social drinkers after repeat doses of DCR-AUD.

1.1.2. Objectives and Endpoints

Objectives	Endpoints
Primary	
To evaluate the safety and tolerability of repeat doses of DCR-AUD administered to adult HVs	• Incidence and severity of AEs and SAEs • Changes from baseline in vital signs, 12-lead ECG, clinical laboratory tests, and physical examination findings
Secondary	
1. To characterize the PD symptom profile of repeat doses of DCR-AUD in adult HVs	Change in the occurrence and/or severity of 6 symptoms related to plasma acetaldehyde accumulation observed during in-clinic EIAs over the course of the study.
2. To characterize the plasma PD of repeat doses of DCR-AUD	Change in plasma PD biomarker assessments during serial EIA o acetaldehyde o acetate o ethanol
3. To characterize the PK of repeat doses of DCR-AUD in adult HVs	Plasma PK parameters of DCR-AUD
Exploratory	
To explore change in alcohol consumption following treatment with DCR-AUD in adult HVs	• Change in alcohol consumption from baseline using TLFB • Change from baseline in phosphatidylethanol levels

Abbreviations: AE: adverse event; EIA: ethanol interaction assessment; HV: healthy volunteer; PD: pharmacodynamics; PK: pharmacokinetic; SAE: serious adverse event; TLFB: Timeline Follow Back

1.1.3. Overall Design

This is a double-blind, randomized, placebo-controlled study of DCR-AUD to evaluate the safety, tolerability, PK, and PD of repeat doses of DCR-AUD in adult HVs.

Participants will receive 3 injections of study intervention (Day 1, Day 29, and Day 57), and have assessment visits through Week 24. Up to 10 EIAs will be conducted over the 24-week period to characterize the PD of DCR-AUD. For each EIA, participants will consume 4 standard drinks over the course of 80 minutes. Participants will be admitted to the clinic overnight for each EIA. At the discretion of the Investigator, participants who present with increasing positive ethanol reaction symptoms at the Day 169 EIA (flushing, headache, lightheadedness, palpitations, nausea, or vomiting) will be followed for an additional 4 weeks (through Day 197).

Table 1 presents the schedule of activities for the Screening, Dosing, End of Study, and Conditional Follow-up Visits. Table 2 presents the schedule of activities for interim visits at which no study drug is administered. Table 3 presents the schedule of activities for the EIA.

Number of Participants:

16 participants are planned for enrollment.

Intervention Groups and Duration:

Participants will be randomized 3 to 1 to DCR-AUD or placebo, administered at a dose of 480 mg once every 4 weeks at Days 1, 29, and 57.

Individual participants will be screened for a period of up to 4 weeks. Randomization and administration of the first dose of study intervention will occur on Day 1. Participants will return to the site for safety, tolerability, PK, and PD monitoring at specified time points as per the Schedule of Activities. Participants will be followed for approximately 24 weeks after the first injection of study intervention.

Study Population Key Criteria:

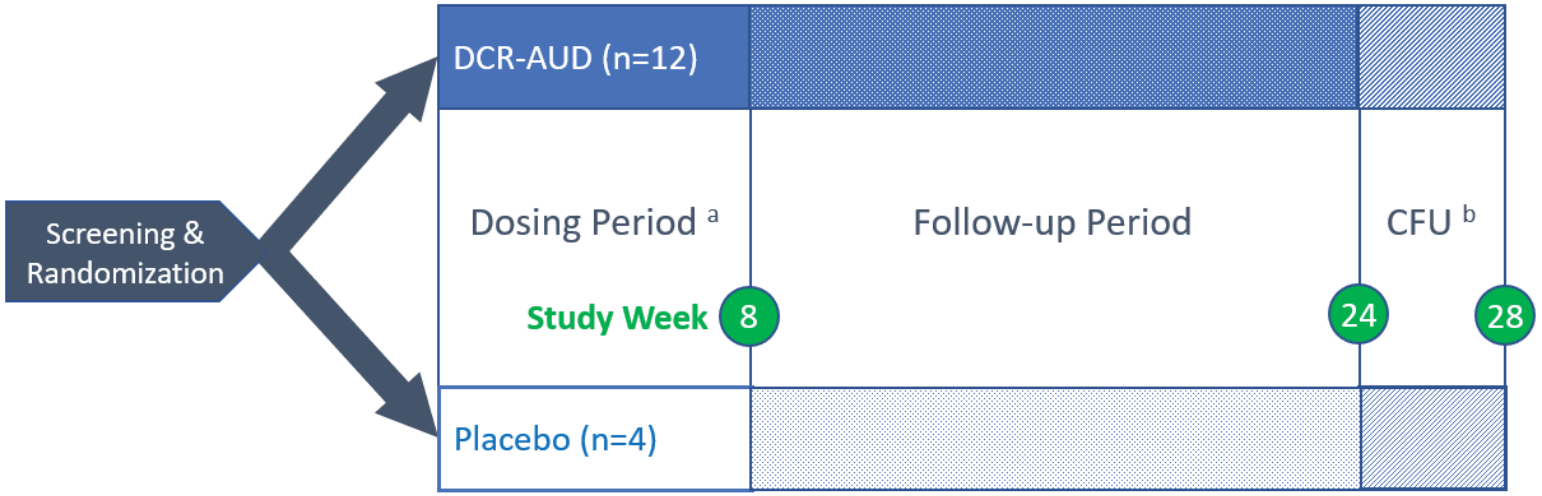
- Overtly healthy men and women aged 21 to 65 years.
- No diagnosis of AUD within the preceding 12 months per Diagnostic and Statistical Manual of Mental Disorders (DSM-5).
- Social drinkers who consume, on average, 5 to 20 drinks per week for men or 5 to 14 drinks per week for women over the 4 weeks prior to screening, and not more than 8 drinks in a single day.
- No evidence of overt or sub-clinical hepatic pathology, as manifest by serologic tests demonstrating hepatic inflammation or compromised hepatic synthetic function.
- No history of substance use disorder (SUD) or illicit drug use (excluding cannabis) within the preceding 12 months.
- No history of alcohol withdrawal symptoms including delirium tremens or alcohol-related seizures.

Treatment Blinding:

Participants and site study staff will be blinded to the randomization. Some members of the Sponsor team will be unblinded. Complete details will be presented in the Study Blinding Plan.

1.2. Overall Study Schema

Figure 1: Study Schematic for DCR-AUD-102



a. DCR-AUD (480 mg) or placebo administered on Days 1, 29, and 57
b. 4-Week conditional follow-up (CFU) conducted at the discretion of the Investigator in participants experiencing symptoms at the End-of-Study (Week 24) visit.

1.3. Schedule of Activities (SoA)

Table 1: Schedule of Activities – Screening, Dosing, EOS, and CFU Visits (see Table 2 for non-dosing visits)

	Screen	Dosing Visits														EOS/ET [a]		CFU[b]
Study Week		—					4					8				24		28
Study Day	−28 to −2	−1	1		2	4	28	29		30	32	56	57		58	168	169	197
Window (days) [c]		–	–		–	–	–	±2		–	–	–	±2		–	–	±7	±7
Procedure/Assessment			Pre	Post				Pre	Post				Pre	Post				
Informed consent	X																	
Inclusion/exclusion criteria	X	X																
Demographic characteristics	X																	
General medical history	X																	
Medication history	X																	
Psychological history	X																	
Urine drug testing [d]	X																	
Screening laboratory testing [e]	X																	
ALDH2 genotyping [f]		X																
MINI Screen	X																	
C-SSRS [g]	X	X					X					X				X		
Alcohol TLFB [h]	X	X					X					X				X		X
Pregnancy test [i]	X	X					X					X				X		X
SARS-CoV-2 test [j]		X					X					X				X		
Drug/alcohol screen [k]		X					X					X				X		
Clinic site admission [l]		X					X					X				X		

Table 1: Schedule of Activities – Screening, Dosing, EOS, and CFU Visits (see Table 2 for non-dosing visits) (Continued)

	Screen	Dosing Visits														EOS/ET [a]		CFU[b]
Study Week		—					4					8				24		28
Study Day	−28 to −2	−1	1		2	4	28	29		30	32	56	57		58	168	169	197
Window (days) [c]		–	–		–	–	–	±2		–	–	–	±2		–	–	±7	±7
Procedure/Assessment			Pre	Post				Pre	Post				Pre	Post				
Clinic site discharge [m]					X					X					X		X	
EIA [n]		X					X					X				X		
Randomize			X															
Administer study intervention			X					X					X					
12-lead ECG [o]	X		X	X				X	X				X	X			X	
Vital signs	X [p]		X	X [q]				X	X [q]				X	X [q]			X	
Physical examination [r]	X	X					X					X				X		
Blood sample for PEth [s]		X										X				X		
Hematology, chemistry, UA [t]		X					X					X				X		
Coagulation panel [u]		X					X					X				X		
Complement panel [v]			X	X	X								X	X	X			
Blood sample for ADA [w]			X													X		
Blood sample for PK [x]			X	X	X	X		X	X	X	X		X	X	X	X		
Alcohol use symptom diary [y]		X			X		X			X		X			X	X		X
Record AEs & SAEs [z]	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Record concomitant meds		X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X

Abbreviations: ADA=antidrug antibody; AE=adverse event; *ALDH2*=aldehyde dehydrogenase 2 gene; CBC=complete blood count; CFU=conditional follow-up visit; C-SSRS=Columbia-Suicide Severity Rating Scale; ECG=electrocardiogram; EIA=ethanol interaction assessment; EOS=end of study; ET=early termination; FSH=follicle-stimulating hormone; HbA1c=hemoglobin A1c; ICF=informed consent form; MINI=Mini-International Neuropsychiatric Interview; PEth=phosphatidylethanol; PK=pharmacokinetic; RBC=red blood cell; SAE=serious adverse event; SARS-CoV-2=severe acute respiratory syndrome coronavirus 2; SIB=suicidal ideation or behavior; TLFB=Timeline Follow Back; UA=urinalysis

[a] At the time of discontinuation for the study, an early termination (ET) visit should be conducted, if possible.

[b] Participants with significant symptoms at the Day 168 EIA will be followed for an additional 4 weeks at the discretion of the Investigator.
[c] Visits without a specified window are to be conducted relative to the associated scheduled visit. For example, Day 28 will always be the day prior to the Day 29 visit, regardless of where Day 29 falls within the ± 2-day visit window
[d] Screening tests positive for cannabis are not exclusionary.
[e] To include hematology (CBC with differential and RBC indices), clinical chemistry, coagulation panel, urinalysis, viral serology, HbA_{1c}, and FSH (as indicated). See Section 10.2 for a complete list of parameters.
[f] Baseline genetic testing for *ALDH2* mutations. Participation is optional; participants who do not wish to undergo genotyping may still participate in the study.
[g] The Baseline version will be administered at Screening and at subsequent visits the "C-SSRS Since Last Visit" version will be administered. Participants who exhibit signs of Suicidal Ideation or Behavior (SIB) should undergo a risk assessment. All factors contributing to SIB should be evaluated and consideration should be given to discontinuation of study intervention.
[h] TLFB will be completed for the 2-week period that precedes each study visit, including Screening.
[i] Pregnancy testing required only in female participants. If a urine pregnancy test is positive, confirm with serum pregnancy test. Local laboratory may be used. Female participants must have a negative pregnancy test prior to overnight admission.
[j] Testing to be performed at the site upon admission. Participants with a positive SARS-CoV-2 test will not be admitted to clinic and will not receive study intervention or undergo EIA. Other assessments not requiring clinic admission (i.e., safety assessments) will be performed. Testing will be performed per site standard processes.
[k] Study participants are to refrain from drinking alcohol and using cannabis for 24 hours prior to each EIA. Participants with a positive urine-drug or breath-alcohol test will not be admitted to clinic and will not undergo EIA. Participants with a positive urine drug test on Day −1 will be excluded from the study.
[l] Participants must be fasting for at least 3 hours prior to admission to the clinic.
[m] Participants may be discharged from clinic following collection of the 24-hour postdose PK sample. Participants will return to clinic for collection of PK samples at 72 hours postdose on Days 4 and 32.
[n] EIA will be conducted on the first day of the visit once all other assessments for that day have been completed. A total of 10 EIAs will be conducted. The timing of assessments and ethanol administration are detailed in Table 3.
[o] On dosing days, ECGs will be conducted approximately 30 minutes predose and 15-to-30 minutes postdose. If multiple assessments are due, the order of assessment should be ECG, vitals, PK, and then other assessments; with PK sample collection occurring at the nominal time. At Screening and EOS, one ECG will be performed. Screening ECGs may be repeated one time for determination of eligibility.
[p] Blood pressure, if out of range at Screening, may be repeated one time for determination of eligibility.
[q] Blood pressure and pulse rate to be recorded 15 (±5) minutes postdose.
[r] Height will be measured and recorded at Screening. Weight will be measured and recorded at every visit.
[s] Sample to be collected prior to ethanol administration.
[t] Hematology to include complete blood count with differential and RBC indices. See Section 10.2 for a complete list of parameters.
[u] See Section 10.2 for a complete list of parameters.
[v] Blood samples for complement testing to be collected predose and 2, 8, and 24 hours postdose on Day 1 and Day 57.
[w] Blood sample for ADA testing to be drawn at same time as predose PK sample on Day 1.
[x] Plasma samples for PK analysis to be collected predose and at 1, 2, 4, 6, 8, 24, and 72 hours postdose (exception: no 72-hour PK sample is required at Day 57). A single sample will be collected on Day 168. If multiple assessments are due, the order of testing should be ECG, vitals, PK, and then other assessments, with the PK sampling performed preferably at the nominal time point. A ±15-minute window will be allowed for collections at 1, 2, 4, 6, and 8 hours postdose. A ±30-minute window will be allowed for collection at 24 hours postdose. A ±60-minute window will be allowed for collection at 72 hours postdose.
[y] Participants to record any symptoms associated with use of alcohol outside of in-clinic EIAs. Diary to be issued at time of discharge from clinic and collected and reviewed upon return to clinic.
[z] AEs and SAEs will be recorded from time ICF is signed. SAEs will be followed until resolution, stabilization, the event is otherwise explained, or the participant is lost to follow-up.

Table 2: Schedule of Activities – Non-dosing Visits (see Table 1 for Screening, Dosing, and EOS Visits)

Week	2		6		10		12		16		20	
Study Day	14	15	42	43	70	71	84	85	112	113	140	141
Visit Window (days) [a]	–	±2	–	±2	–	±3	–	±5	–	±7	–	±7
Procedure/Assessment												
Drug/alcohol screen [b c]	X		X		X		X		X		X	
Pregnancy test [d]	X		X		X		X		X		X	
SARS-CoV-2 test [e c]	X		X		X		X		X		X	
Clinical site admission [f]	X		X		X		X		X		X	
Physical examination [g]	X		X		X		X		X		X	
C-SSRS [h]							X					
Hematology, chemistry, UA [i j]							X					
Coagulation panel [j]							X					
Blood sample for ADA [k]	X		X		X							
Blood sample for PK [k]	X		X		X							
Blood sample for PEth [k]									X			
EIA [l]	X		X		X		X		X		X	
Alcohol TLFB [m]	X		X		X		X		X		X	
Alcohol use symptom diary [n]	X	X	X	X	X	X	X	X	X	X	X	X
Record AEs and SAEs [o]	X	X	X	X	X	X	X	X	X	X	X	X
Record concomitant meds	X	X	X	X	X	X	X	X	X	X	X	X
Clinical site discharge [p]		X		X		X		X		X		X

Abbreviations: ADA=antidrug antibody; AE=adverse event; C-SSRS=Columbia-Suicide Severity Rating Scale; EIA=ethanol interaction assessment; PEth=phosphatidylethanol; PK=pharmacokinetics; SAE=serious adverse event; SARS-CoV-2=severe acute respiratory syndrome coronavirus 2; SIB=suicidal ideation or behavior; TLFB=Timeline Follow Back; UA=urinalysis

[a] Visits without a specified window are to be conducted relative to the associated scheduled visit. For example, Day 14 will always be the day prior to the Day 15 visit, regardless of where Day 15 falls within the ± 2-day visit window.
[b] Study participants are to refrain from drinking alcohol and using cannabis for 24 hours prior to each EIA. Participants with a positive urine-drug or breath-alcohol test will not be admitted to clinic and will not undergo EIA. Other assessments not requiring clinic admission (i.e., safety assessments) should be performed.
[c] Other assessments not requiring clinic admission (i.e., safety assessments) should be performed.
[d] Pregnancy testing required only in female participants. If a urine pregnancy test is positive, confirm with serum pregnancy test. Local laboratory may be used. Female participants must have a negative pregnancy test prior to overnight admission.
[e] Participants with a positive SARS-CoV-2 test will not be admitted to clinic and will not undergo EIA. Testing will be performed per site standard processes.
[f] Participants must be fasting for at least 3 hours prior to admission to the clinic.
[g] Weight will be measured and recorded at each visit.
[h] The "C-SSRS Since Last Visit" version will be administered. Participants who exhibit signs of SIB should undergo a risk assessment.
[i] Hematology to include complete blood count.
[j] See Section 10.2 for complete list of testing parameters.
[k] Sample to be collected prior to ethanol administration for EIA.
[l] EIA will be conducted once all other assessments for that day have been completed. The timing of assessments and ethanol administration are detailed in Table 3.
[m] TLFB will be completed for the 2-week period that precedes each study visit.
[n] Participants to record any symptoms associated with use of alcohol outside of in-clinic EIAs. Diary to be issued at time of discharge from clinic and collected and reviewed upon return to clinic.
[o] AEs and SAEs will be recorded from time ICF is signed. SAEs will be followed until resolution, stabilization, the event is otherwise explained, or the participant is lost to follow-up.
[p] Participants will be discharged in the morning when the PI deems it medically appropriate to do so.

Table 3: Schedule of Activities - 4-Hour Ethanol Interaction Assessments

	Pre-EIA		EIA Test Period										
Time (minutes)	**-50**	**-20**	**0** [a]	**20**	**30**	**40**	**60**	**90**	**120**	**150**	**180**	**210**	**240**
Window (minutes)			**-**	**±5**	**±5**	**±5**	**±10**	**±10**	**±10**	**±10**	**±10**	**±10**	**±10**
Procedure:													
Standard meal [b]	X												
Venous catheter placement [c]		X											
12-lead ECG [d]			X				X						X
Vital signs [d e]			X	X		X	X	X	X		X	X	X
Plasma biomarker sample [d f]			X		X		X	X	X		X		X
Questionnaire of 6 EIA symptoms [d g]			X		X		X [d]		X		X		
Ethanol consumption [h]			X	X		X	X						
EIA AEs [i]			X	X		X	X	X	X	X	X	X	X

Abbreviations: AE=adverse event; ECG=electrocardiogram; EIA=ethanol interaction assessment; PK=pharmacokinetics

a "Time 0" is immediately before the first sip of ethanol is consumed. All Time 0 assessments must be performed prior to initiation of alcohol intake.

b Participants will arrive at the site after a minimum 3-hour fast, then will be provided a standardized meal before receiving the first drink. Participants will be given 30 minutes to consume the meal.

c Catheter placement may be performed earlier in the day if preferred, but participants must be given at least a 20-minute rest period following placement of the venous access catheter prior to recording predose vital signs and ECG. For EIAs conducted prior to a dosing day, the venous catheter will be left in place for PK sample collections.

d If multiple assessments are due at the same timepoint, the order of assessments should be ECG, vitals, blood draw, then any other assessments. The 60-minute symptom assessment should be conducted before the start of the 4th drink.

e To include pulse rate and blood pressure.

f Blood samples will be collected via venous access device. To include acetaldehyde, acetate, and ethanol.

g Participants will be questioned by site staff to assess the 6 symptoms (flushing, headache, lightheadedness, palpitations, nausea, and vomiting) associated with increased blood acetaldehyde levels.

h Ethanol will be consumed in 4 aliquots over an 80-minute period (20 minutes for each aliquot). Time windows do not apply to ethanol consumption. The ethanol will be mixed in a liquid that has no other alcohol, no caffeine, and is low in sugar. The dose of ethanol will be 0.8 g/kg (to a maximum of 56 g) for men and 0.68 g/kg (to a maximum of 48 g) for women. See Section 8.6.1.3 of the protocol for ethanol administration stopping rules.

i AEs associated with administration of ethanol will include events spontaneously reported by participants beginning with the first sip of ethanol (symptoms elicited under direct questioning [footnote g] are not considered AEs).

2. INTRODUCTION

2.1. Study Rationale

DCR-AUD is being developed for the treatment of AUD in adults using an RNAi technology platform. DCR-A1203, the drug substance of DCR-AUD, is a synthetic double-stranded (annealed duplex) RNA oligonucleotide conjugated to GalNAc ligands that enable specific hepatic access and uptake after subcutaneous (SC) administration. Once inside the hepatocyte, the antisense sequence of DCR-AUD is designed to target and reduce the expression of ALDH2 mRNA in the liver.

The target of DCR-AUD is ALDH2, one of the primary enzymes required for alcohol metabolism. Alcohol metabolism occurs predominantly in the liver via a 2-step process in which alcohol is first converted to acetaldehyde via alcohol dehydrogenase and then to acetic acid by ALDH2. DCR-AUD is designed to selectively reduce *ALDH2* mRNA and ALDH2 activity in the liver, and thereby decrease the conversion of acetaldehyde to acetic acid after ethanol ingestion. ALDH2 enzyme inhibition can result in unpleasant symptoms after alcohol consumption such as flushing, headache, lightheadedness, palpitations, nausea, and vomiting as a result of unmetabolized acetaldehyde accumulation. Unpleasant reactions to alcohol consumption in humans with naturally occurring *ALDH2* mutations are thought to be protective against AUD and account for its low prevalence in individuals who are heterozygous or homozygous for *ALDH2* allele mutations. The magnitude and duration of acetaldehyde accumulation is proportional to both the degree of ALDH2 enzyme inhibition (e.g., partial vs total) and the amount of alcohol consumed.

Interim results of the ongoing Study DCR-AUD-101, which was the first study of DCR-AUD in humans, have shown DCR-AUD to be safe and well tolerated at doses ranging from 80 to 960 mg, but no PD effects (sufficient elevation of plasma acetaldehyde levels or clinical symptoms) have been observed.

The aims of this repeat-dose study are to characterize the safety and tolerability and PK profiles of DCR-AUD when administered monthly, and to determine if a PD effect (resulting from cumulative *ALDH2* mRNA knock down) can be observed during EIAs in a population of social drinkers after repeat doses of DCR-AUD.

2.1.1. Overview of Alcohol Use Disorder

Alcohol use disorder is a chronic condition characterized by compulsive alcohol use, loss of control over alcohol use, and a negative emotional state when not using alcohol. A range of medical, psychological, social, economic, and personal problems are associated with AUD. Diagnosis is defined as meeting ≥ 2 of 11 criteria in the Diagnostic and Statistical Manual of Mental Disorders-5 (DSM 5) during the same 12-month period. A survey of over 36,000 adults found AUD had a 12-month prevalence of 13.9% adults in the US in 2013 (Grant et al., 2015). AUD often goes untreated, possibly due to the lack of adequate treatment options. It is estimated that < 10% of US adults with AUD seek help or treatment, and that similarly low percentages of those treated receive pharmacotherapy (Grant et al., 2015; SAMHSA, 2014; Litten et al., 2014; Han et al., 2015; Knudsen et al., 2011).

There are 3 FDA-approved medications for AUD: disulfiram, oral and long-acting injectable naltrexone, and acamprosate. However, many individuals show limited or no response to these drugs, with poor compliance a factor (EMA, 2010; Gastfriend, 2014; Chick et al., 1992; Jørgensen et al., 2011; Bryson et al., 2011). Thus, there remains an unmet need for an effective AUD treatment.

2.1.2. Nonclinical Overview of DCR-AUD

The pharmacology of DCR-A1203 was characterized in in vivo studies in mice and monkeys to demonstrate the ability to reduce levels of the hepatic *ALDH2* mRNA. The potential for off-target gene silencing was assessed using in silico evaluation of the human transcriptome. The sequence is conserved in mouse, monkey, and human, but not rabbit. Therefore, DCR-A1203 is active in both general toxicology species evaluated, mouse and monkey. In addition, a stand-alone comprehensive safety pharmacology study in cynomolgus monkeys was conducted.

Overall, the data support the hypothesis that DCR-AUD has potential to yield meaningful and robust clinical responses across the AUD patient population based upon the following:

- A single SC dose of up to 100 mg/kg in mice resulted in a dose-dependent decrease in *Aldh2* mRNA expression in liver, persistent for 14 days postdose, followed by dose-dependent recovery.
- Following a single 3 mg/kg SC dose of DCR-A1203, liver *ALDH2* mRNA expression levels were reduced by > 75% in cynomolgus monkeys, with the nadir at 28 days postdose, and > 50% reduction in *ALDH2* mRNA expression up to 112 days postdose.
- Repeat SC administration of DCR-A1203 at 30 to 300 mg/kg resulted in significant reduction of hepatic *Aldh2* mRNA in mice (> 97%) and hepatic *ALDH2* in monkeys (> 86%) at all dose levels evaluated.
- Based on *in silico* analysis, potential off-target complementarities with 3 or fewer mismatches and a full seed-region match, were completely absent from the human transcripts analyzed, demonstrating that DCR-A1203 has a high degree of specificity for hepatic *ALDH2* mRNA.

The safety of DCR-A1203 has been characterized in a program of nonclinical studies. Subcutaneous repeat-dose toxicity studies have been conducted at doses up to 300 mg/kg for up to 6 months in mice and 9 months in monkeys. DCR-A1203 has been evaluated for potential genotoxicity in a bacterial reverse mutation assay and in vitro and in vivo micronucleus studies. In addition, preliminary reproductive toxicity studies were conducted in mice and rabbits.

- DCR-A1203 administration had no adverse effects on cardiovascular, respiratory, or central nervous system function in conscious, radiotelemetry-implanted cynomolgus monkeys following a single SC injection of up to 300 mg/kg.
- The administration of DCR-A1203 in repeat-dose studies for up to 6 months in mice and 9 months in monkeys resulted in no DCR-A1203-related, adverse findings. The NOAEL in the mouse and monkey repeat-dose toxicity studies was considered to be 300 mg/kg, the highest dose level administered.

- In a preliminary embryofetal development (EFD) study in mice with daily SC dosing, no DCR-A1203-related toxicity was observed at dose levels up to 1000 mg/kg (highest dose administered). In a preliminary EFD study in rabbits with daily SC dosing (dose levels from 2 to 50 mg/kg), maternal toxicity (lower body weight gains and lower food consumption) in dams and embryofetal effects (higher post implantation loss and lower mean fetal weights) were observed at all DCR-A1203 dose levels. The human relevance of these contradictory observations in mice and rabbits is unclear, as DCR-A1203 is not pharmacologically active in rabbits and no such observations were noted in the EFD study in the pharmacological responsive mouse with DCR-A1203.
- DCR-A1203 was not genotoxic in the bacterial reverse mutation or in vitro and in vivo micronucleus assays.

Overall, the nonclinical safety program of DCR-A1203 did not identify any major safety issues. The human relevance of preliminary nonclinical reproductive studies is unclear, and participants will follow contraception guidance. A detailed description of the chemistry and pharmacology of DCR-AUD is provided in the Investigator's Brochure.

2.2. Benefit/Risk Assessment

The summary of potential risks provided below is based on results of the ongoing FIH study (DCR-AUD-101) and on in-depth review of literature relating to the proposed approach and insights from the nonclinical pharmacology and toxicology studies.

The antisense sequence of DCR-AUD is designed to target and reduce the expression of *ALDH2* mRNA in the liver. Potential off-target complementarities at the level of a single mismatch were completely absent from the human transcripts analyzed in in-silico studies. These findings demonstrate that DCR-AUD has a high degree of specificity for the *ALDH2* mRNA. The sequence is conserved in mouse, monkey, and human, and therefore, DCR-AUD is active in all nonclinical general toxicology species evaluated. To date, DCR-A1203 has been associated with no adverse effects in nonclinical studies.

Clinical experience with the Dicerna GalXC™ siRNA platform in more than 200 patients and HVs suggests a favorable safety profile with little-to-no evidence of systemic immune stimulation or toxicities. Mild, self-limiting injection site reactions have been reported (Dicerna data on file). In addition, in the on-going DCR-AUD-101 trial, single doses of DCR-AUD (range: 80 to 960 mg) administered to 24 participants have shown good tolerability. Specifically, no SAEs have been observed, and the vast majority of AEs in the trial have been mild in severity.

As part of the study's planned mitigation strategy, all study participants will be monitored for signs or symptoms of systemic toxicity, decline in liver or kidney function, abnormalities in hematologic parameters (including platelets and coagulation parameters), and abnormalities in vital signs, ECGs, complement activation, or other relevant safety parameters.

2.2.1. Risks Related to DCR-AUD Mechanism of Action

DCR-AUD, at the proposed dose, is designed to induce mild symptoms resulting from substantial alcohol consumption (e.g., ≥ 4 drinks/occasion) by selective reduction of *ALDH2* mRNA and ALDH2 activity in the liver, and thereby a decrease in the conversion of

acetaldehyde to acetic acid after ethanol ingestion. In sufficient amounts, acetaldehyde accumulation is associated with unpleasant effects, potentially including flushing, headache, lightheadedness, palpitations, nausea, and vomiting. More severe reactions to acetaldehyde accumulation can include copious vomiting, sweating, chest pain, palpitation, dyspnea, hypotension, syncope, marked uneasiness, weakness, vertigo, blurred vision, and confusion. The intensity of the reaction varies with each individual but is generally proportional to the amounts of alcohol ingested and the degree of ALDH2 inhibition. The duration of the reaction varies from 30 to 60 minutes and up to several hours in the more severe cases if there is prolonged alcohol exposure in the blood. Symptoms are typically self-limited and managed with watchful observation and supportive care. More severe acetaldehyde reactions have been documented with disulfiram treatment and in individuals who are homozygous for *ALDH2*-gene–inactivating mutations compared with *ALDH2* mutation heterozygotes, proportional to the magnitude of systemic ALDH2 inactivation.

Study participants will be clearly informed of these potential risks. Symptoms that develop with concomitant alcohol consumption are expected to be self-limited and manageable with supportive care and can be avoided by limiting or eliminating alcohol consumption.

In the absence of direct measurement of ALDH2 activity in the liver (e.g., via liver biopsy), EIAs are necessary to evaluate and confirm DCR-AUD PD activity. The EIAs in this protocol (4 standard drinks [approximately 56 g for men and 48 g for women] administered over an 80-minute window) are designed to reflect a plausible level of consumption for frequent drinkers and patients with AUD. Possible signs and symptoms of alcohol intolerance will be recorded in the clinic under close observation and controlled conditions.

2.2.2. Risk Assessment

Potential Risk of Clinical Significance	Summary of Data/ Rationale for Risk	Mitigation Strategy
Study Intervention: DCR-AUD		
Injection site reactions	Subcutaneous administration of the study intervention may cause a specific local reaction originating around the injection site, such as erythema, itching, discomfort, and pain.	Most ISRs are mild and self-limiting. Participants will be monitored for the development of ISRs (Section 10.3.3)
Potential reproductive effects	In a preliminary EFD study in rabbits with daily SC dosing, maternal toxicity in dams and embryofetal effects were observed at all dose levels. The human relevance of these preliminary nonclinical reproductive studies is unclear.	All participants of childbearing potential are required to practice highly effective contraception consistently throughout the study and for 24 weeks after the last dose of study intervention (Section 10.4).
Severe symptoms associated with acetaldehyde accumulation following consumption of alcohol	In sufficient amounts, acetaldehyde accumulation is associated with unpleasant effects, including flushing, headache, lightheadedness, palpitations, nausea, and vomiting. More severe reactions to acetaldehyde accumulation can include copious vomiting, sweating, chest pain, palpitation, dyspnea, hypotension, syncope, marked uneasiness, weakness, vertigo, blurred vision, and confusion.	If significant or severe adverse symptoms occur in ≥3 participants treated with DCR-AUD during the trial, additional DCR-AUD dosing (if scheduled) and subsequent EIAs will be discontinued. In the event this occurs, all study participants will be advised to avoid alcohol for the entire study observation period.
Suicidal ideation or behavior (SIB)	Patients diagnosed with AUD may be at risk for SIB. Because DCR-AUD is being developed for the treatment of AUD, HV will be screened and monitored for the presence or emergence of SIB while on study.	There is no evidence to suggest that HV exposed to DCR-AUD are at risk for SIB, however, all participants will be monitored for the emergence of SIB via the C-SSRS at regular intervals throughout the study (Section 8.2.5.1).
Study Procedures		
EIA: alcohol administration may cause intoxication and is expected to cause mild, unpleasant effects in participants receiving DCR-AUD	Required to assess and confirm DCR-AUD PD activity, including magnitude and duration of effects associated with concomitant alcohol interaction	During EIAs, alcohol to be administered as one standard drink every 20 minutes for 4 drinks (i.e., over 80 minutes) in a Phase 1 unit clinical setting under close observation. Eligible participants will have drinking histories suggesting tolerability to the amount of ethanol to be administered in the EIAs.

Potential Risk of Clinical Significance	Summary of Data/ Rationale for Risk	Mitigation Strategy
Blood sampling, including cannulation. Risks include acute pain, bleeding, vessel injury, and, in rare instances, arterial vessel blockage, potentially leading to infection.	Blood sampling is required to assess the safety, PK, and PD of DCR-AUD.	The total volume of blood to be withdrawn for research purposes has been limited to the minimum required for a complete assessment.
Other		
Prolonged PD activity of DCR-AUD may cause alcohol intolerance for up to 3 months or more after last dose in study participants. Adverse effects are not expected but potentially possible with concomitant small or incidental amounts of alcohol during DCR-AUD treatment.	Adverse effect(s) resulting from high levels of alcohol intake during DCR-AUD treatment is an intentional design element of the DCR-AUD target product profile and predicted based on the core RNAi/GalXC drug platform. The clinical significance is expected to be limited to high or very high alcohol consumption, but adverse effects may occur at lower ingested amounts.	Ensure risks related to DCR-AUD potential PD effects and duration are clearly communicated in the informed consent. Adverse effects related to alcohol consumption are self-limited, reversible, and avoidable.

2.2.3. Benefit Assessment

There is no expected benefit for HV participants in the study. Data generated in this study will contribute to the development of a potential new therapy for AUD, an area of significant unmet medical need, and FDA-approved treatment options.

The siRNA-mediated reduction of *ALDH2* via DCR-AUD is designed to offer significant safety advantages relative to agents such as disulfiram, which inhibits multiple aldehyde dehydrogenases as well as other enzymes (Koppaka et al., 2012). siRNA-mediated reduction will only target *ALDH2* in the liver, leaving other aldehyde dehydrogenases able to function. Further, DCR-AUD reduction of ALDH2 will be liver-specific, leaving non-liver ALDH2 unaffected.

2.2.4. Overall Benefit:Risk Conclusion

With the measures planned to minimize participant risk in this study, the potential risks identified in association with DCR-AUD are justified by possible future benefits that may be afforded to patients with AUD.

3. OBJECTIVES AND ENDPOINTS

Objectives	Endpoints
Primary	
To evaluate the safety and tolerability of repeat doses of DCR-AUD administered to adult HVs	• Incidence and severity of AEs and SAEs • Changes from baseline in vital signs, 12-lead ECG, clinical laboratory tests, and physical examination findings
Secondary	
1. To characterize the PD symptom profile of repeat doses of DCR-AUD in adult HVs	Change in the occurrence and/or severity of 6 symptoms related to plasma acetaldehyde accumulation observed during in-clinic EIAs over the course of the study.
2. To characterize the plasma PD of repeat doses of DCR-AUD	Change in plasma PD biomarker assessments during serial EIA o acetaldehyde o acetate o ethanol
3. To characterize the PK of repeat doses of DCR-AUD in adult HVs	Plasma PK parameters of DCR-AUD
Exploratory	
To explore change in alcohol consumption following treatment with DCR-AUD in adult HVs	• Change in alcohol consumption from baseline using TLFB • Change from baseline in phosphatidylethanol levels

Abbreviations: AE: adverse event; EIA: ethanol interaction assessment; HV: healthy volunteer; PD: pharmacodynamics; PK: pharmacokinetic; SAE: serious adverse event; TLFB: Timeline Follow Back

4. STUDY DESIGN

4.1. Overall Design

This is a double-blind, randomized, placebo-controlled study of DCR-AUD to evaluate the safety, tolerability, PK, and PD of repeat doses of DCR-AUD in adult HVs.

Each participant will be screened for up to 28 days. Participants may undergo rescreening one time at the discretion of the Investigator or Sponsor. Screening assessments will include a psychological history and a history of alcohol consumption using the TLFB method to identify participants who are HVs and who meet the minimum inclusion/exclusion alcohol consumption requirements. TLFB is a validated method to retrospectively assess drinking behavior history. Randomization and administration of the first dose of study intervention will occur on Day 1.

Repeated doses of 480 mg study intervention will be administered to 16 adult HVs (12 active and 4 placebo). Each participant will receive 3 SC doses of study intervention (on Day 1, Day 29, and Day 57). Up to 10 EIAs will be conducted over the 24-week period to characterize the PD of DCR-AUD.

All participants will be given a diary to track AEs and drinking history for the study duration. Quantitative drinking history will be captured by TLFB questionnaire completed every 2 weeks. Participants will be trained on the use of the diary prior to discharge from the clinic and will report on the diary at each visit.

At the discretion of the Investigator, participants who present with increasing positive ethanol reaction symptoms at the Day 168 EIA (e.g., flushing, headache, lightheadedness, palpitations, nausea, or vomiting) will be followed for an additional 4 weeks (through Day 197).

4.2. Scientific Rationale for Study Design

DCR-AUD is designed to selectively reduce *ALDH2* mRNA and ALDH2 activity in the liver, and thereby decrease the conversion of acetaldehyde to acetic acid after ethanol ingestion. In cynomolgus monkeys, a single SC dose of 3 mg/kg DCR-AUD resulted in reduction of *ALDH2* mRNA expression in liver up to 112 days postdose, with maximum reduction observed at 4 weeks postdose.

Study DCR-AUD-101 was designed to evaluate the safety, tolerability, PK, and PD of single ascending doses of DCR-AUD in healthy adult volunteers who abstained from drinking alcohol during the study (outside of the study-mandated EIAs). In that first-in-human (FIH) study, DCR-AUD, at doses ranging from 80 to 960 mg, was found to be safe and well tolerated. However, while the plasma PK was well characterized, no clear pattern of plasma biomarkers or physiologic symptoms during EIAs indicative of target engagement was established at any dose.

In addition to further characterizing the safety and tolerability and PK profiles of DCR-AUD when administered monthly, this repeat-dose study is being conducted to determine if a PD effect (resulting from cumulative *ALDH2* mRNA knock down) can be observed in a population of social drinkers. Knockdown of ALDH2 activity by DCR-AUD that is sufficient to increase plasma acetaldehyde levels and induce symptoms may require both repeat doses and several months to be manifest, supporting the 24-week follow-up observation period in this trial.

The up-to-10 EIAs will permit evaluation of the onset of DCR-AUD effect and when this effect wanes or extinguishes. The amount of ethanol administered during EIAs in this study is double that given during Study DCR-AUD-101. The 4 drinks over 80 minutes were included to reflect the drinking patterns experienced by patients with AUD.

4.3. Justification for Dose

The dose level of 480 mg was selected following evaluation of nonclinical PK/PD and safety data and based on results of the on-going DCR-AUD-101 study evaluating single ascending doses up to 960 mg.

Administration of DCR-A1203 in the repeat-dose studies for up to 6 months in mice and 9 months in monkeys resulted in no adverse findings related to DCR-A1203. The NOAEL in the mouse and monkey chronic repeat-dose toxicity studies was considered to be 300 mg/kg, the highest dose level administered.

Interim results from DCR-AUD-101 have shown single doses of DCR-AUD to be safe and well tolerated at doses ranging from 80 to 960 mg. No clear pattern of plasma biomarkers or physiologic symptoms indicative of target engagement was established at any dose.

See the DCR-AUD IB for an overview of safety margins.

4.4. End of Study Definition

A participant is considered to have completed the study if he/she has completed all of the Day 169 EOS assessments, as shown in the SoA (Section 1.3). The conditional follow-up visit is not required of all participants and is conducted only at the discretion of the Investigator, based on the occurrence of substantial symptoms during the Day 169 EIA, or a reported adverse event profile reported at that visit that warrants further follow-up.

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

5. STUDY POPULATION

Sixteen participants are planned to be enrolled in this study: 12 will receive DCR-AUD and 4 will receive placebo.

Protocol waivers or exemptions are not allowed. Participants may undergo rescreening one time at the discretion of the Investigator or Sponsor (Section 5.4)

5.1. Inclusion Criteria

Participants are eligible to be included in the study only if all the following criteria apply.

Age

1. 21 to 65 years, inclusive, at the time of signing informed consent.

Type of Participant and Disease Characteristics

2. Overtly healthy volunteers, as determined by medical evaluation including medical history, physical examination, and laboratory testing.
3. No diagnosis of AUD within the preceding 12 months per Diagnostic and Statistical Manual of Mental Disorders (DSM-5).
4. Social drinkers who consume, on average, 5 to 20 drinks per week for men or 5 to 14 drinks per week for women over the 4 weeks prior to Screening, and not more than 8 drinks in a single day.
5. No evidence of overt or sub-clinical hepatic pathology, as manifest by serologic tests demonstrating hepatic inflammation or compromised hepatic synthetic function (i.e., AST, ALT, GGT, total bilirubin < 1.5 times the ULN at Screening and Day −1).
6. eGFR ≥ 60 mL/min/1.73 m^2 at Screening.
7. No history of significant adverse reaction(s) to alcohol. Participant should be expected to tolerate the amount of alcohol administered during EIAs.
8. Willing to participate in up to 10 EIAs.
9. Has a negative test for SARS-CoV-2 infection on Day −1.
10. Systolic BP in the range of 80 to 140 mmHg and diastolic BP in the range of 50 to 95 mmHg at Screening. If out of range, BP may be repeated once at the discretion of the Investigator.

Sex and Contraceptive Requirements

11. Male or female
    - Male participants with partners of childbearing potential must agree to use contraception from Screening through at least 24 weeks after the last dose of study intervention and refrain from donating sperm during this period (see Section 10.4).
    - Female participants may not be pregnant or breastfeeding, and at least one of the following conditions must apply:
      - Is not a woman of childbearing potential (WOCBP) **or**

- If a WOCBP, must agree to follow the contraceptive guidance (see Section 10.4), beginning at consent and the first Screening visit and for at least 24 weeks after the last dose of study intervention.

Weight

12. BMI within the range 18.0 to 32.0 kg/m^2 (inclusive).

Informed Consent

13. Capable of giving signed informed consent, which includes compliance with the requirements and restrictions listed in the ICF and in this protocol.

5.2. Exclusion Criteria

Participants are excluded from the study if any of the following criteria apply.

Medical Conditions

1. History of any medical condition that may interfere with the metabolism of study intervention or with the clinical and laboratory assessments in this study.
2. History of serious, persistent medical conditions, including liver, gastrointestinal, pulmonary, renal, or cardiovascular abnormalities.
3. History of suicidal attempt at any time or an answer of "yes" on any of the following items in the C-SSRS at Screening:
   a. Items 1 or 2 of the Suicidal Ideation section, if ideation occurred in the previous 12 months.
   b. Items 4 or 5 of the Suicidal Ideation section, in lifetime.
   c. Any item of the Suicidal Behavior section of the C-SSRS, in lifetime.
4. Any history of severe or recent clinically significant depression, anxiety, bipolar disorder, schizophrenia, or other neuropsychiatric disorder that, in the judgement of the Investigator, represents a safety risk to the participant were they to participate in the trial, as informed by the participant's medical history and/or responses to the MINI Screen Questionnaire.
5. History of substance use disorder (SUD) or illicit drug use (excluding cannabis) within the preceding 12 months.
6. History of alcohol withdrawal symptoms including delirium tremens or alcohol-related seizures.
7. Any condition that, in the opinion of the Investigator, would make the participant unsuitable for participation or could interfere with participation in or completion of the study, including:
   a. Poorly controlled or unstable hypertension.
   b. Diabetes mellitus treated with insulin or hypoglycemic agents (including metformin) or $HbA_{1C} > 7\%$.

c. Asthma requiring hospital admission within the preceding 12 months. NOTE: Persons with clinically stable asthma who have not been hospitalized in the prior year and are treated only with orally inhaled medications are not excluded.

d. Currently poorly controlled endocrine conditions, except for hypothyroidism that is stable (no treatment change in prior 6 months).

e. Significant infection or known systemic inflammatory process ongoing at Screening.

f. History of chronic or recurrent UTI, or UTI within 1 month prior to Screening.

8. History of malignancy within the preceding 5 years requiring treatment, with the exception of excised low grade basal cell skin neoplasms.

9. History of any concomitant medical condition for which alcohol consumption is prohibited or advised against by the participant's physician or health care provider.

10. SARS-CoV-2 infection in the 14 days prior to randomization.

11. Clinically significant illness within the 7 days prior to the first administration of study intervention.

Prior/Concomitant Therapy

12. History of multiple drug allergies or a history of allergic reaction to an oligonucleotide-based therapy.

13. Use of prescription medications (except for hormonal replacement/contraceptive medication for women and inhaled medication for treatment of clinically stable asthma) within 14 days or 5 half-lives (whichever is longer) prior to administration of study intervention. Participants being treated for hypothyroid disease must be on stable treatment (no treatment changes in the preceding 6 months).

14. Receipt of any vaccine (including COVID-19) within 14 days prior to the first administration of study intervention.

15. Regular use of OTC medications, including NSAID (periodic or occasional NSAID use to control temporary pain is not exclusionary).

Prior/Concurrent Clinical Study Experience

16. Previously participated in Dicerna Study DCR-AUD-101.

17. Has received an investigational agent within 30 days or 5 half-lives (whichever is longer) prior to dosing or is in follow-up of another clinical study prior to initial dosing with the study intervention.

Diagnostic Assessments

18. Clinically significant abnormalities in vital signs at Screening: pulse rate (< 40 bpm or > 90 bpm), respiratory rate, or temperature.

19. Clinically significant abnormalities in 12-lead ECG at Screening or predose on Day 1, including QTcF > 470 msec in females and > 450 msec in males.

20. Positive urine drug test at Screening or Day −1. Tests positive for cannabis are not exclusionary.

21. Seropositive for antibodies to HIV, HBV, or HCV at Screening (historical testing may be used if performed within the 3 months prior to screening). NOTE: In participants with previous treatment for hepatitis C with direct-acting HCV medication and seropositivity for HCV, or in participants with prior infection and spontaneous resolution, HCV RNA must be undetectable (at least 2 negative HCV RNA tests at least 12 weeks apart), and the HCV infection must have been resolved or cured > 3 years prior to initial dosing with the investigational medication.
22. Safety laboratory test result at Screening considered clinically unacceptable for study participation by the Investigator.

Other Exclusions

23. History of intolerance to SC injection(s) or significant abdominal scarring that could potentially hinder study intervention administration or evaluation of local injection site tolerability.
24. Scheduled for an elective surgical procedure during the conduct of this study.
25. Donation of > 500 mL of blood within the 2 months prior to administration of study intervention or donation of plasma within 7 days prior to Screening.

5.3. Lifestyle Considerations

Alcohol, Tobacco, and Cannabis

1. Study participants are to refrain from drinking alcohol for 24 hours prior to each EIA and must have a negative breath-alcohol test on the day of the study visit.
2. Study participants will be advised to avoid consuming more than 4 drinks during any drinking session outside of in-clinic EIAs.
3. Study participants will be instructed to stop drinking at the onset of any ethanol reaction symptoms during outside drinking sessions.
4. Tobacco/nicotine use is not restricted.
5. Study participants are to refrain from using cannabis for 24 hours prior to each EIA and must have a negative urine-drug test on the day of the study visit.

Activity

1. Study participants should abstain from strenuous exercise for 24 hours before each blood collection for clinical laboratory tests. Participants may participate in light recreational activities (e.g., walking at a pace < 3 miles per hour, shopping, watering plants) in that 24-hour time period.

Meals and Dietary Restrictions

1. For each EIA study visit, participants will be required to fast for at least 3 hours and then will be fed a standardized meal in the clinic prior to administration of ETOH for the EIA.

5.4. Screen Failures

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

Individuals who do not meet the criteria for participation in this study (screen failure) may be rescreened. Participants may undergo rescreening one time at the discretion of the Investigator or Sponsor. Among blood tests, only pregnancy, hematology, chemistry, and coagulation tests may be repeated. Blood pressure, heart rate, and ECG may be repeated for participants who have abnormal readings but who are otherwise healthy, and, in the opinion of the Investigator, the abnormal values reflect a transient departure from the range permitted for eligibility.

6. STUDY INTERVENTION

Study intervention is defined as any investigational intervention(s), including placebo, intended to be administered to a study participant according to the study protocol.

6.1. Study Intervention(s) Administered

The clinical dose is expressed in terms of the weight of the DCR-AUD free acid, rather than that of the sodium salt (Table 4).

Table 4: Study Intervention Administered

Arm Name	Active	Placebo
Intervention Name	DCR-AUD	Physiologic saline (0.9% aqueous sodium chloride) for injection
Intervention Type	Drug	Drug
Dose Formulation	solution for injection	n/a
Unit Dose Strength	160 mg/mL	n/a
Dose Level(s) and Frequency	480 mg Q4W × 3	Volume to match active, Q4W × 3
Route of Administration	SC injection (thigh or abdomen)	SC injection (thigh or abdomen)
Use	Experimental	Placebo comparator
IMP or NIMP	IMP	n/a
Sourcing	Provided centrally by the Sponsor or designee	Provided locally by the study site (Pharmacy staff is not blinded)
Packaging and Labeling	DCR-AUD will be provided in vials. Each vial will be labeled as required per country.	n/a

Abbreviations: IMP: investigational medicinal product; n/a: not applicable; NIMP: non-investigational medicinal product; Q4W: once every 4 weeks; SC: subcutaneous

6.2. Preparation/Handling/Storage/Accountability

6.2.1. Handling and Accountability

The Investigator or designee must confirm appropriate temperature conditions have been maintained during transit for all study intervention received and that any discrepancies are reported and resolved before use of the study intervention.

Only participants enrolled in the study may receive study intervention, and only authorized site staff may supply or administer study intervention.

The Investigator, institution, or the head of the medical institution (where applicable) is responsible for study intervention accountability, reconciliation, and record maintenance (i.e., receipt, reconciliation, and final disposition records).

Further guidance and information for the final disposition of unused study intervention are provided in the Pharmacy Manual.

6.2.2. Storage

DCR-AUD is to be stored at or below 30°C (inclusive). Do not freeze.

All study intervention must be stored in a secure, environmentally controlled and monitored (manual or automated) area in accordance with the labeled storage conditions, with access limited to the Investigator and authorized site staff.

The placebo comparator should be stored in accordance with the product labeling.

6.2.3. Preparation and Administration

Study intervention will be prepared by the unblinded pharmacist or designee. A second medically qualified person must check the dose preparation prior to administration to be sure the correct dose has been prepared.

If stored at 2°C to 8°C, DCR-AUD should be allowed to warm to room temperature for approximately 1 hour before administration.

DCR-AUD is administered as a SC injection into the thigh or abdomen. The maximum volume of a single SC injection should not exceed 1.5 mL; the total dose will be administered as 2 SC injections of equal volume at separate injection sites, preferably in separate quadrants.

Placebo will be administered as an SC injection of physiologic saline (0.9% aqueous sodium chloride) in the thigh or abdomen at a volume equal to that of DCR-AUD.

All used DCR-AUD vials will be retained and quarantined in the pharmacy. Once drug accountability is recorded to be accurate, the vials will be destroyed on site per clinic SOP. Complete details will be provided in the Pharmacy Manual.

6.3. Measures to Minimize Bias: Randomization and Blinding

This is a randomized, placebo-controlled, double-blind study, where the Investigators, study site personnel (excluding pharmacy staff), CRO staff, and study participants will be blinded for the duration of the study. Some members of the Sponsor team will be unblinded for the duration of the study. This enables the Sponsor to immediately and comprehensively evaluate the safety of the study participants. Complete details will be presented in the Study Blinding Plan.

The overall DCR-AUD to placebo ratio is 3:1. All participants will be centrally assigned to randomized study intervention using an Interactive Voice/Web Response System (IVRS/IWRS). Before the study is initiated, the telephone number and call-in directions for the IVRS and/or the log in information and directions for the IWRS will be provided to each site. To maintain the study blind, the unblinded site pharmacy staff will be responsible for the preparation and dispensing of all study intervention.

In the event of a Quality Assurance audit, the auditor(s) will be allowed access to unblinded study intervention records at the site(s) to verify that randomization/dispensing has been done accurately.

The IVRS/IWRS will be programmed with blind-breaking instructions. In case of an emergency, the Investigator has the sole responsibility for determining if unblinding of a participant's treatment assignment is warranted. Participant safety must always be the first consideration in making such a determination. If the Investigator decides that unblinding is warranted, the

Investigator should make every effort to contact the Medical Monitor prior to unblinding a participant's treatment assignment, unless this could delay emergency treatment of the participant. If a participant's treatment assignment is unblinded, the Medical Monitor must be notified within 24 hours after breaking the blind. The date and reason that the blind was broken must be recorded in the source documentation and case report form, as applicable.

Potentially unblinded study results will not be shared with blinded study participants or site personnel.

6.4. Study Intervention Compliance

At the site, study intervention will be administered by the Investigator or designee. The date and time of each dose administered in the clinic will be recorded in the source documents and recorded in the eCRF. The dose of study intervention and study participant identification will be confirmed at the time of dosing by a member of the study site staff other than the person administering the study intervention.

6.5. Dose Modification

Not applicable. See Section 7.3 for individual participant stopping rules.

6.6. Continued Access to Study Intervention after the End of the Study.

Not applicable.

6.7. Concomitant Therapy

Study participants must abstain from taking prescription or nonprescription drugs within 14 days or 5 half-lives (whichever is longer) prior to the first administration of study intervention until EOS, with the following exceptions:

- Regular use of vitamins is acceptable.
- Hormonal replacement/contraceptive medication for women is acceptable.
- Inhaled medication for treatment of clinically stable asthma is acceptable.
- Participants being treated for hypothyroid disease must be on stable treatment (no treatment changes in the preceding 6 months).
- Occasional use of NSAIDs for pain management is acceptable. Regular use of OTC medications (including NSAIDs) is not allowed.
- Participants must not receive any vaccine from Day −14 through Day 71 of the study.

Any medication or vaccine (including OTC or prescription medicines, recreational drugs [with the exception of cannabis], vitamins, and herbal supplements) that the participant is receiving at the time of enrollment or receives during the study must be recorded along with:

1. Reason for use.
2. Dates of administration, including start and end dates.
3. Dosage information, including dose and frequency.

6.7.1. Rescue Medicine

Not applicable.

7. DISCONTINUATION OF STUDY INTERVENTION AND PARTICIPANT DISCONTINUATION/WITHDRAWAL

7.1. Participant Discontinuation/Withdrawal from the Study

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

If a participant has a significant adverse reaction to alcohol at the Day –1 visit (before administration of DCR-AUD), the participant will be discontinued from the study. The participant will be replaced.

If possible, at the time of discontinuing from the study, an early termination (ET) visit should be conducted, as shown in the SoA (Table 1).

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

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

The reason for participant discontinuation or withdrawal from the study will be recorded on the eCRF. Participants who sign the ICF and are randomized but do not receive the study intervention may be replaced. Participants who sign the ICF, are randomized and do receive the study intervention and subsequently withdraw or are withdrawn or discontinued from the study may be replaced at the discretion of the Sponsor.

7.2. Lost to Follow Up

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

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

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

- Should the participant continue to be unreachable, he/she will be considered to have withdrawn from the study.

7.3. Individual Participant Stopping Rules

7.3.1. Serious Adverse Event

If a participant experiences an SAE (Section 10.3.2), unless it is clearly the result of a non-study-related event, further dosing of the participant will be paused while the SRC reviews the event. Based on this review, a decision will be made as to whether the participant may continue to receive doses of study intervention. In participants who have completed dosing, continued follow-up with protocol-specified assessments, including subsequent EIAs, will be pursued if the participant is willing and the PI determines it is safe to do so. Additional EIAs will not be performed if, in the clinical judgment of the PI and/or SRC, the SAE could be related to study intervention and that repeat EIAs pose a recurrent risk to the participant, or if the participant declines these additional assessments. Participants will continue to be followed for other study procedures as detailed in the SoA.

7.3.2. Abnormal Liver Function Tests

Discontinuation of study intervention is required when a participant meets one of the conditions outlined in Section 10.6. Discontinuation of study intervention may also be considered in the presence of other abnormal liver function tests, if the Investigator believes that it is in best interest of the participant.

7.3.3. Electrocardiogram Findings

If a clinically significant finding is identified (including, but not limited to, the QTcF findings below) after enrollment, the Investigator or qualified designee will determine if the participant can continue in the study and if any change in participant management is needed. This review of the ECG printed at the time of collection must be documented. Any new clinically relevant finding should be reported as an AE.

- Absolute QTcF > 500 msec and/or
- QTcF change of > 60 msec from baseline and >450 msec.
- New and clinically significant arrhythmia or rhythm abnormality.

7.3.4. Significant Negative Reaction to Alcohol Ingestion

Any participant who experiences a significant negative reaction to alcohol ingestion that includes severe or serial vomiting or hemodynamic instability (syncope or significant pre-syncope, or BP decline of more than 25 mmHg from Baseline with some lightheaded symptoms, or HR increases to ≥ 110 bpm with some symptoms during an EIA) will be discontinued from additional dosing and will be advised to not drink outside of the clinic for the duration of the study. The participant will continue to be followed in the study for other protocol-specified procedures. The PI will determine if additional/subsequent protocol-specified EIAs will be conducted, based on the nature and severity of symptoms observed.

See Section 8.6.1.3 regarding full details on stopping rules for conduct of an EIA.

7.4. Study Stopping Rules

All further dosing in the study will be stopped if any of the following conditions occur AND the SRC recommends stopping the study:

- 2 participants develop a Grade 3 CTCAE event, unless it is clearly the result of a non-study-related event.
- 1 participant develops a ≥ Grade 4 CTCAE event, unless it is clearly the result of a non-study-related event.

All further DCR-AUD dosing and subsequent EIAs will be stopped if the following occurs AND the SRC recommends stopping the study:

- 3 participants treated with DCR-AUD experience significant or severe adverse symptoms associated with alcohol ingestion. In the event this occurs, all study participants will be advised to avoid alcohol for the remainder of the study period.

In the event a study stopping rule is triggered and the SRC calls a halt on dosing, the SRC may request that an external, independent, unblinded DSMC be convened to review the decision and make a recommendation to the SRC as to whether the study should stop or continue with no changes or continue with appropriate modifications to address the safety concern (see Section 10.1.5). Should the DSMC recommend stopping the study, all ongoing participants will continue to be followed for safety assessments as indicated in the schedule of activities.

The Sponsor will promptly notify the site if study stopping rules are triggered.

8. STUDY ASSESSMENTS AND PROCEDURES

Study procedures and their timing are summarized in Section 1.3. Adherence to the study design requirements, including those specified in Table 1, Table 2, and Table 3 (Section 1.3) is essential and required for study conduct.

Protocol waivers or exemptions are not allowed. Participants may undergo rescreening one time at the discretion of the Investigator or Sponsor (Section 5.4).

Urgent safety concerns should be discussed with the Sponsor immediately upon occurrence or awareness.

Participants will be given a diary to track AEs associated with alcohol intake for the study duration. Participants will be trained on the use of the diary prior to discharge from the clinic for Week 1 and will report on the diary at each visit.

If multiple assessments are due, the order of priority should be ECG, vitals, blood draws, and then other assessments.

The maximum amount of blood drawn from each participant (including PK, biomarker, and safety laboratory testing) will not exceed 550 mL over an 8-week period.

8.1. Efficacy Assessments

This is a Phase 1 study with the primary objective of evaluating safety. Pharmacokinetic and PD assessments are detailed in Section 8.5 and Section 8.6, respectively.

As the study participants are HV not diagnosed with AUD, no true assessment of efficacy can be made. However, study participants may be inclined to change their alcohol intake should they experience physiologic symptoms associated with increased acetaldehyde while drinking. As such, 2 assessments of alcohol intake, TLFB and PEth, will be considered in exploratory analyses.

8.1.1. Alcohol Consumption via Timeline Follow Back

Participants will complete the TLFB at screening and throughout the study period to capture any change in drinking behavior. At every visit, the TLFB will be used to assess each participant's alcohol consumption for the 2-week period that precedes the study visit.

The TLFB was developed as a procedure to aid recall of past drinking (Sobell & Sobell, 1992). Currently, self-reports are the only viable method for retrospectively measuring drinking with any precision. Alternative methods exist, but they are either impractical (e.g., continuous direct observation), fraught with problems (e.g., alcohol sweat patches), or they only measure very recent drinking (e.g., blood alcohol tests). TLFB stands as the most exhaustively evaluated method that can be used to retrospectively assess drinking behavior over time.

8.1.2. Blood Phosphatidylethanol Levels

Hepatic ethanol oxidation is the prevalent route of human ethanol metabolism, eliminating more than 90% of ingested ethanol. Several nonoxidative routes of ethanol metabolism that result in the enzymatic conjugation of ethanol to endogenous metabolites have also been described. Due to slower elimination rates, these nonoxidative ethanol metabolites persist in body fluids and tissues for much longer than ethanol itself, thus enabling the retrospective assessment of ethanol intake even when ethanol itself is no longer present in the body.

Phosphatidylethanol (PEth) is a phospholipid produced in erythrocyte membranes after alcohol exposure, first described by Alling et al. (1983). In drinking experiments, PEth production began as soon as ethanol was consumed and peaked 1 to 3 hours after alcohol consumption (Gnann et al., 2012; Heier et al., 2016). With frequent alcohol consumption, and a half-life of approximately 5 days, PEth accumulates in whole blood (Javors et al., 2016). Because PEth can be detected in blood for up to one month after one-time alcohol intake, it can be used as a biomarker to determine both current and recent consumption (Kummer et al., 2016).

Ulwelling & Smith (2018) proposed 3 ranges of PEth values likely to describe a person's average recent alcohol consumption/drinking behavior:

- <20 ng/mL Abstinence or light drinking (0 to <2 drinks/day)
- 20 – 199 ng/mL Moderate drinking (2 to 4 drinks/day for several days a week)
- > 200 ng/mL Heavy drinking (at least 4 drinks/day for several days a week)

Thus, PEth may be used to identify and/or monitor individuals with chronic excessive drinking behavior (Viel et al., 2012).

8.2. Safety Assessments

Planned time points for all safety assessments are provided in the SoA (Section 1.3). Safety assessments will be conducted in all participants at every study visit unless otherwise specified.

Site personnel will be provided training in the use of all study-specific instruments (see the Study Reference Manual).

8.2.1. Screening Tests

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

8.2.1.1. Mini-International Neuropsychiatric Interview

The MINI Screen is a brief structured diagnostic interview to meet the need for a short but accurate structured psychiatric interview for multicenter clinical trials (Sheehan et al., 1998). This validated tool can be administered in a short period of time (approximately 15 minutes) and will be administered at screening to exclude participants with major psychiatric disorders. The MINI Screen will be administered by a Health Care Provider.

8.2.1.2. Laboratory Tests to Determine Eligibility

In addition to the hematology, clinical chemistry, coagulation, and urinalysis panels indicated in Table 6, the following tests will be performed during screening:

- Follicle-stimulating hormone (as needed to confirm menopausal status)
- Viral serology (human immunodeficiency virus antibody, hepatitis B surface antigen, and hepatitis C virus antibody)
- Hemoglobin A_{1c}
- Urine drug screen (to include, at a minimum, amphetamines, barbiturates, cocaine, opiates, and benzodiazepines)

8.2.2. Physical Examinations

Complete physical examinations will be conducted at every visit. Physical examinations will include, at a minimum, assessments of the Cardiovascular, Respiratory, Gastrointestinal, Neurological, and Skin systems. Height will be measured and recorded at Screening. Weight will be measured and recorded at every visit.

- Investigators should pay special attention to clinical signs related to previous serious illnesses or AEs.
- All physical examinations should include inspection of the injection site.

8.2.3. Vital Signs

Temperature by skin refraction, pulse rate, respiratory rate, and blood pressure will be assessed, unless otherwise indicated in the SoA.

Blood pressure and pulse measurements will be assessed in the seated position with a completely automated device. Manual techniques will be used only if an automated device is not available.

Blood pressure and pulse measurements should be preceded by at least 5 minutes of rest for the participant in a quiet setting without distractions (e.g., television, cell phones).

Temperature will be obtained in degrees Celsius (°C), pulse rate will be counted for a full minute and recorded in beats per minute, and respirations will be counted for a full minute and recorded in breaths per minute.

Note: In addition to standard vital sign measurements for safety, vital signs will be recorded as part of the EIA and results will be identified as such (e.g., HR for EIA) in the eCRF

8.2.4. Electrocardiograms

Standard 12-lead ECGs will be performed in the supine position after the participant has rested comfortably for 5 minutes. ECGs will be obtained using an ECG machine that automatically calculates the heart rate and measures PR, QRS, QT, and corrected QT interval (QTcF, Fridericia correction). Refer to Section 7.3.3 for QTc withdrawal criteria and any additional readings that may be necessary. Screening ECGs may not be repeated for determination of eligibility.

The Investigator or designee is responsible for reviewing the ECGs to assess whether the results are within normal limits and to determine the clinical significance of the results. These assessments will be recorded on the CRF.

ECGs also will be conducted for EIA assessments. These ECG results will be recorded as part of the EIA and will be identified as such (e.g., ECG for EIA) in the CRF.

Note: If multiple assessments are due, the order of priority should be ECG, vitals, blood draws, and then other assessments, with the PK sampling being performed preferably at the nominal time point.

8.2.5. Suicidal Ideation and Behavior Risk Monitoring

Patients diagnosed with AUD may be at risk for SIB. Because DCR-AUD is being developed for the treatment of AUD, HV will be screened and monitored for the presence or emergence of SIB while on study. Consideration should be given to discontinuing study intervention in participants who experience signs of SIB, following a risk assessment.

8.2.5.1. Columbia-Suicide Severity Rating Scale

The C-SSRS is a suicidal ideation rating scale created by researchers at Columbia University (Posner et al., 2011). It rates an individual's degree of suicidal ideation on a scale, ranging from “wish to be dead” to “active suicidal ideation with specific plan and intent.”

The scale identifies behaviors that may be indicative of an individual's intent to commit suicide. The C-SSRS is used extensively across primary care, clinical practice, surveillance, research, and institutional settings and is part of a national and international public health initiative involving the assessment of suicidal ideation and behavior. The C-SSRS requires no mental health or medical training to administer it. The C-SSRS will be administered by site personnel who are trained in its administration.

The Baseline version will be administered at Screening and at subsequent visits the “C-SSRS Since Last Visit” version will be administered.

8.2.6. Clinical Safety Laboratory Assessments

See Section 10.2 for the list of clinical laboratory tests to be performed and the SoAs (Section 1.3) for the timing and frequency.

The Investigator must review the laboratory report, document this review in the eCRF, and record any clinically relevant changes occurring during the study in the AE section of the eCRF. The laboratory reports must be filed with the source documents.

All laboratory tests with values considered clinically significantly abnormal during participation in the study should be repeated (central laboratory) until the values return to normal or stabilize at a level no longer considered clinically significant by the Investigator or Medical Monitor.

- If such values do not return to normal/baseline within a period of time judged reasonable by the Investigator, the etiology should be identified, and the Sponsor notified.
- All protocol-required laboratory assessments, as defined in Section 10.2, must be conducted in accordance with the Laboratory Manual and the SoA (Section 1.3).
- If laboratory values from non-protocol specified laboratory assessments performed at the institution's local laboratory require a change in participant management or are considered clinically significant by the Investigator (e.g., SAE or AE), then the results must be recorded in the eCRF.

8.2.7. Pregnancy Testing

WOCBP should only be included after a confirmed menstrual period and a negative highly sensitive urine or serum pregnancy test. If a urine test is positive, it must be confirmed with a serum test. See Section 8.3.5 for reporting of pregnancy in a study participant or the partner of a male participant.

Additional pregnancy testing should be performed as indicated in SoA (Section 1.3).

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

Pregnancy testing with a sensitivity of at least 25 mIU/mL will be performed.

8.3. Adverse Events, Serious Adverse Events, and Other Safety Reporting

The definitions of AEs, SAEs, and adverse events of special interest (AESI) can be found in Section 10.3.1, Section 10.3.2, and Section 10.3.3, respectively.

The Investigator and any qualified designees are responsible for detecting, documenting, and recording events that meet the definition of an AE or SAE and remain responsible for following up AEs that are severe or serious, and considered related to the study intervention or study procedures, or that caused the participant to discontinue study intervention or the study.

8.3.1. Time Period and Frequency for Collecting Adverse Event Information

All AEs and SAEs will be collected from the time the ICF is signed through the last follow-up visit at the time points specified in the SoAs (Section 1.3).

All SAEs will be recorded and reported to the Sponsor or designee within 24 hours, as indicated in Section 10.3.5. The Investigator will submit any updated SAE data to the Sponsor within 24 hours of it being available.

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

8.3.2. Method of Detecting AEs and SAEs

The method of recording, evaluating, and assessing causality of AE and SAE and the procedures for completing and transmitting SAE reports are provided in Section 10.3.

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

Injection sites will be examined by study staff for assessment of ISRs (Section 10.3.3).

8.3.3. Follow-up of AEs and SAEs

After the initial AE/SAE report, the Investigator is required to proactively follow each participant at subsequent visits/contacts. All SAEs and AESIs (as defined in Section 10.3.3), will be followed until resolution, stabilization, the event is otherwise explained, or the participant is lost to follow-up (as defined in Section 7.2). Further information on follow-up procedures is provided in Section 10.3.4. If symptoms related to ethanol ingestion that the Investigator considers relevant are present at the end of study, the Sponsor may request continued contact with participants in order to collect further data.

8.3.4. Regulatory Reporting Requirements for SAEs

Prompt notification by the Investigator to the Sponsor of an SAE is essential so that legal obligations and ethical responsibilities towards the safety of participants and the safety of a study intervention under clinical investigation are met.

The Sponsor has a legal responsibility to notify both the local regulatory authority and other regulatory agencies about the safety of a study intervention under clinical investigation. The Sponsor will comply with country-specific regulatory requirements relating to safety reporting to the regulatory authority, IRBs/IECs, and Investigators.

Investigator safety reports must be prepared for suspected unexpected serious adverse reactions (SUSAR) according to local regulatory requirements and Sponsor policy and forwarded to Investigators as necessary.

An Investigator who receives an Investigator safety report describing a SAE or other specific safety information (e.g., summary or listing of SAEs) from the Sponsor will review and then file

it with the Investigator's Brochure and will notify the IRB/IEC, if appropriate, according to local requirements.

8.3.5. Pregnancy

Details of all pregnancies in female participants and female partners of male participants will be collected after the start of study intervention and until 24 weeks after the last dose.

If a pregnancy is reported, the Investigator should inform the Sponsor within 24 hours of learning of the pregnancy and should follow the procedures outlined in Section 10.4.3.

While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication or elective termination of a pregnancy for medical reasons will be reported as an AE or SAE. Abnormal pregnancy outcomes (e.g., spontaneous abortion, fetal death, stillbirth, congenital anomalies, ectopic pregnancy) are considered SAEs.

The participant or pregnant partner will be followed to determine the outcome of the pregnancy. The Investigator will collect follow-up information on the participant or pregnant partner and the neonate, and the information will be forwarded to the Sponsor.

Any post-study pregnancy-related SAE considered reasonably related to the study intervention by the Investigator will be reported to the Sponsor as described in Section 8.3.4. While the Investigator is not obligated to actively seek this information in former study participants, he or she may learn of an SAE through spontaneous reporting.

8.4. Treatment of Overdose

For this study, any dose of DCR-AUD greater than 960 mg will be considered an overdose.

The Sponsor does not recommend specific treatment for an overdose.

In the event of an overdose, the Investigator should:

- Keep the participant under observation at least overnight. The Investigator will use his or her judgment to determine if it is safe for the participant to undergo the EIA scheduled for that visit.
- Closely monitor the participant for any AEs or laboratory abnormalities until DCR-AUD PD effects can no longer be detected.
- Contact the CRO Medical Monitor immediately.
- Document the quantity of the excess dose.

It should be noted that an overdose is not an AE, in and of itself. See Section 10.3.1 for details.

8.5. Pharmacokinetics

Blood samples (2 mL) will be collected for measurement of plasma concentrations of DCR-AUD and its metabolites as specified in the SoAs (see Section 1.3).

Instructions for the collection and handling of biological samples will be provided by the Sponsor in the Laboratory Manual and the Laboratory Specification document. The actual date and time (24-hour clock time) of each sample will be recorded.

Each plasma sample will be divided into 2 aliquots (one for PK and one backup). Samples collected for analyses of plasma concentrations of DCR-AUD and its metabolites may also be used to evaluate safety or efficacy aspects related to concerns arising during or after the study.

Pharmacokinetic parameters to be determined may include area under the concentration curve (AUC), minimum observed concentration (C_{min}), maximum observed concentration (C_{max}), time to maximum concentration (T_{max}), terminal elimination half-life ($t_{1/2}$), apparent clearance (CL/F), and apparent volume of distribution (Vd/F). Additional parameters may be estimated if deemed appropriate.

Any changes in the timing or addition of time points for any planned study assessments must be documented and approved by the relevant study team member and then archived in the Sponsor and site study files but will not constitute a protocol amendment. The IRB/IEC will be informed of any safety issues that require alteration of the safety monitoring scheme or amendment of the ICF.

Drug concentration information that may be unblinding will not be reported to investigative sites or blinded personnel until the study has been unblinded.

8.6. Pharmacodynamics

DCR-AUD is designed to selectively reduce ALDH2 activity in the liver and the conversion of acetaldehyde to acetic acid after alcohol consumption. The build-up of acetaldehyde causes unpleasant effects after drinking (e.g., flushing, headache, lightheadedness, palpitations, nausea, or vomiting). As such, the PD activity of DCR-AUD will be assessed during EIAs via these 6 symptoms, blood biomarkers (ethanol, acetaldehyde, and acetate), and heart rate.

Published literature consistently demonstrate that administration of low/moderate alcohol intake (0.1 to 0.8 g/kg) in human *ALDH2* mutation homo- or heterozygotes results in clearly differentiated increases in blood acetaldehyde and heart rate compared with ALDH2 normal alleotypic controls (Bae et al., 2012; Peng et al., 1999; Peng et al., 2007; Nishimura et al., 2002; Enomoto et al., 1991).

8.6.1. Ethanol Interaction Assessments

Serial EIAs will be performed to assess the indirect PD effects of ALDH2 reduction (acetaldehyde increase). EIAs will be conducted in accordance with the SoA in Section 1.3. Should an EIA be missed, due to COVID-19 or other circumstances, the missed EIA may be conducted at an unscheduled visit at the discretion of the Investigator and the Sponsor.

Participants will be required to be fasted for at least 3 hours prior to administration of a standardized meal and must have a negative breath alcohol test on the day of the study visit prior to the EIA. Prior to each EIA, venous catheters will be placed at least 20 minutes before the first EIA vital signs are assessed at time 0. For EIAs conducted prior to a dosing day, the venous catheter will be left in place for PK sample collections.

Participants will consume a fixed dose of ethanol mixed in a liquid that has no additional alcohol, no caffeine, and is low in sugar. The ethanol dose will be 0.8 g/kg (to a maximum of 56 g) for male participants and 0.68 g/kg (to a maximum of 48 g) for female participants. Ethanol will be consumed in 4 equal aliquots over an 80-minute period (20 minutes for each

aliquot). During the EIA, participants will receive nothing by mouth (remain NPO) except water ad libitum. Assessments will be made over an estimated 4-hour period, as detailed in Table 3.

Participants will remain in the Phase 1 unit for over-night observation. The Investigator will not discharge any participant experiencing ongoing effects of ethanol administration and will not do so until it is deemed medically safe to do so.

8.6.1.1. EIA Blood Biomarkers

Blood biomarkers to be assessed include measurement of acetaldehyde, acetate, and ethanol. Blood samples (2 mL) will be collected via a venous access device at the times indicated in Table 3. Additional details regarding blood sample collection and processing will be detailed in the Laboratory Manual.

As plasma biomarker data may be unblinding, results will not be reported to investigative sites or blinded personnel until the study has been unblinded.

8.6.1.2. EIA Physiologic Signs and Symptoms

EIA assessments will include the direct questioning of participants as to the presence of ethanol reaction symptoms (flushing, headache, lightheadedness, palpitations, nausea, and vomiting) at the times indicated in Table 3. Note: the presence of these ethanol reaction symptoms will not be considered AEs.

A composite 6-symptom score for each participant during each EIA will be determined as follows:

- At each of the timepoints during the EIA, each symptom will be given a numeric score of 0 (not present) to 3 (present and severe), with the exception of vomiting, which will be given a score of 3.
- The composite symptom score for each of the EIA timepoints will be the summed value for each of the 6 symptoms at that timepoint (range 0 – 18).
- The Peak score for a given day's EIA will be the participant's highest post-alcohol ingestion timepoint score minus the participant's pre-alcohol score.

Vital signs (BP and heart rate) and standard 12-lead ECGs will be recorded at the times indicated in Table 3.

8.6.1.3. Stopping Rules for an EIA

During the conduct of an EIA, administration of alcohol to a participant may be stopped for the following physiologic responses, though follow-up EIA assessments will be conducted:

- Any vomiting or severe nausea (as judged by Investigator)
- Systolic blood pressure increase of > 25 mmHg (or to a level > 160 mmHg) or decrease of > 15 mmHg to < 70 mmHg or any decline accompanied by symptomatic hypotension (pre-syncope, syncope, lightheadedness)
- Diastolic blood pressure decrease to < 50 mmHg or any decline accompanied by symptomatic hypotension (pre-syncope, syncope, lightheadedness)

- HR increase of > 20 beats per minute and to a level > 100 beats per minute
- Temperature increase of > 1°Celsius
- Any clinically meaningful change in affect or clinical status, per Investigator or supervising designee's clinical judgment

8.6.1.4. Follow-up of Participants Who Continue to have Positive Ethanol Reaction Symptoms at Day 168 (Conditional Follow-up)

All participants who have ethanol reaction symptoms during the Day 168 EIA will be instructed to continue limit drinking to 4 drinks or less during any drinking session.

Participants who present clinically significant positive ethanol reaction symptoms at the Day 168 EIA (flushing, headache, lightheadedness, palpitations, nausea, or vomiting) that the PI considers severe and/or warranting further follow-up, will be followed for an additional 4 weeks (through Day 197; see Table 1). If symptoms related to ethanol ingestion that the Investigator considers relevant are present at Day 197, the Sponsor may request continued contact with participants in order to collect further data.

8.6.2. Presence of Physiologic Symptoms Associated with Consumption of Ethanol Outside of EIAs

Participants will be allowed to continue drinking alcohol in the windows between study visits, but will be advised not to drink more than 4 drinks in any one drinking session. Participants will be given a diary to report the occurrence and severity of any physical symptoms that occur with alcohol consumption outside of the in-clinic EIAs.

Participants will be instructed to stop drinking at the onset of any symptoms during outside drinking sessions.

8.7. Genetics

Unpleasant reactions to alcohol consumption in humans with naturally occurring *ALDH2* mutations are thought to be protective against AUD and account for its low prevalence in individuals who are heterozygous or homozygous for *ALDH2* allele mutations. A 6 mL sample of whole blood for DNA isolation will be collected from participants who have consented to *ALDH2* genotyping. Participation is optional; participants who do not wish to undergo genotyping may still participate in the study.

In the event of DNA extraction failure, a replacement genetic blood sample may be requested from the participant. Signed informed consent will be required to obtain a replacement sample, unless it was included in the original consent.

See Section 10.5 for information regarding the use of DNA samples. Details on processes for collection and shipment and destruction of these samples can be found in the Laboratory Manual.

8.8. Immunogenicity Assessments

Serum samples for the detection and characterization of antibodies to DCR-AUD will be stored until a validated assay method is available.

Serum samples will be collected at times specified in the SoA (Section 1.3). Additionally, serum samples should also be collected at the final visit from participants who discontinued study intervention or were withdrawn from the study.

Serum samples will be screened for antibodies binding to DCR-AUD, and the titer of confirmed positive samples will be reported. Other analyses may be performed to verify the stability of antibodies to DCR-AUD and/or further characterize the immunogenicity of DCR-AUD.

Antibodies may be further characterized and/or evaluated for their ability to neutralize the activity of DCR-AUD.

8.9. Health Economics

Health Economics parameters are not planned for this study.

9. STATISTICAL CONSIDERATIONS

9.1. Statistical Hypotheses

Statistics will be descriptive. No tests of statistical inference are planned.

9.2. Sample Size Determination

No formal sample size estimations were performed. The sample size of 16 participants (12 active and 4 placebo) was considered sufficient to provide an initial assessment of the safety profile of DCR-AUD in HVs, and adequate for the purpose of describing PK/PD data in HVs.

The sample size is based on clinical rather than statistical considerations.

9.3. Populations for Analyses

For purposes of analysis, the following populations are defined.

Population	Description
Enrolled	All participants who sign the ICF.
Randomized	All participants who sign the ICF and who are randomized to study intervention.
Pharmacokinetic	All participants randomized to study intervention and who receive at least 1 dose of DCR-AUD and have at least 1 postdose PK assessment.
Pharmacodynamic	All participants randomized to study intervention and who receive at least 1 dose of DCR-AUD or placebo and have at least 1 postdose PD assessment
Safety	All participants randomized to study intervention and who receive at least 1 dose of DCR-AUD or placebo. Participants will be analyzed according to the initial dose received.

9.4. Statistical Analyses

The statistical analysis plan (SAP) will be finalized prior to database lock. The SAP will include a more technical and detailed description of the statistical analyses described in this section. This section is a summary of the planned statistical analyses of the most important endpoints including primary and key secondary endpoints.

9.4.1. General Considerations

In general, summary statistics (n, mean, SD, median, minimum, and maximum for continuous variables, and number and percentage of participants in each category for categorical variables) will be provided for all variables.

Baseline is defined as the last non-missing measurement prior to the date and time of the dose of study intervention.

All analyses will be based on observed data without imputation for missing values. Except for outlier analyses, measurements collected from unscheduled visits will not be included in the summary tables or figures but will be included in the participant listings. Source data for the summary tables and statistical analyses will be presented as participant data listings.

9.4.2. Primary Endpoint(s)

The primary endpoints of the study are the assessment of the incidence and severity of AEs and SAEs, and the changes from baseline in vital signs, 12-lead ECG, clinical laboratory tests, and physical examination findings.

All safety analyses will be performed on the Safety Population. Adverse events will be defined as treatment-emergent AEs if they have a start date on or after the administration of study intervention during the treatment period. Frequency and percentages by treatment group will be used to summarize AEs, SAEs, AEs of special interest, and AEs by relationship.

Descriptive statistics and changes from baseline will be provided for vital signs, 12-lead ECG, clinical laboratory tests, and physical examination findings by treatment group and visit. Laboratory parameter shifts from baseline to abnormal post-baseline values will be presented by treatment group. Mean laboratory values over time will be plotted by dose group.

By-participant listings will be provided for all the safety parameters.

9.4.3. Secondary Endpoints

PD analyses will be performed on the Pharmacodynamic Population, and PK analyses will be performed on Pharmacokinetic Population. Actual sampling times will be used for all parameter estimations. The principal secondary endpoint of interest will be the change from baseline in the composite 6-symptom score after EIAs (as described in Section 8.6.1.2).

9.4.3.1. EIA Composite Score

EIAs will include an active assessment of the 6 key symptoms (flushing, headache, palpitations, lightheadedness, nausea, and vomiting) that DCR-AUD may elicit, in order to determine if the protocol-specified DCR-AUD dose regimen gives the proper constellation of these symptoms. These symptoms will be collected by active questioning of each participant at different time points during each EIA.

The following symptom scoring framework would be described as an arbitrary point system, where:

Symptom	No Symptom	Mild symptom	Moderate symptom	Severe symptom
Score	0	1	2	3

Each participant at each timepoint in an EIA after alcohol administration will be given a composite score for that timepoint as sum of all 6 symptom scores.

Baseline is defined as last non-missing value prior to alcohol administration.

Descriptive statistics of EIA composite score (n, mean, median, IQR [Inter Quartile Range], minimum and maximum) for absolute values and changes from baseline will be presented by treatment, placebo, visit, and time point.

Descriptive statistics of Peak EIA composite score (n, mean, median, IQR, minimum and maximum) for absolute values and changes from baseline will be presented by treatment, placebo, and visit (Days). The Peak EIA composite score is defined as maximum composite score at each visit.

9.4.3.2. Blood Biomarkers

Descriptive statistics and change from baseline will be provided for blood PD assessments, including acetaldehyde accumulation (absolute values and AUC) during the EIAs, where applicable.

9.4.3.3. Pharmacokinetics

Plasma concentrations and PK parameters will be listed and summarized by group and by time point with descriptive statistics. The arithmetic mean, SD, CV%, geometric mean, geometric CV%, minimum, median, and maximum will be presented.

Additional PK parameters may be estimated if deemed appropriate. Details will be provided in the PK Analysis Plan. Analysis of PK parameters may be reported in a supplement to the CSR.

PK analyses will be conducted with noncompartmental and/or compartmental models to determine PK parameter estimates. Population PK and Population PK-PD analyses may be performed and will be reported in a separate report.

9.4.4. Exploratory Endpoint(s)

The exploratory endpoint of change in drinking amounts will be assessed using the TLFB and by analysis of blood levels of phosphatidylethanol.

Average daily drinking amounts over each 2-week period after the start of dosing will be compared to the amount during the 2-week period before first dose. Change in drinking amounts for each postdose period will be evaluated in connection with changes in EIA-induced peak symptoms and plasma acetaldehyde levels during the preceding EIA.

Other exploratory analyses will be conducted as deemed appropriate and will be detailed in the SAP.

9.5. Interim Analysis

In order to enable decision making for the future clinical development of DCR-AUD, an interim analysis may be conducted after all participants have received all 3 doses of study intervention.

Variables to be analyzed may include assessments of safety (AEs, laboratory testing, physical findings), pharmacokinetics, and pharmacodynamics (blood biomarkers and physiologic signs and symptoms). Should the interim analysis be conducted, complete details will be provided in the SAP.

9.6. Safety Review Committee and Data Safety Monitoring Committee

An SRC will conduct periodic reviews and will review all cumulative safety, tolerability, and available PD data on participants. Participant safety will be continuously monitored by the SRC by ongoing review of AEs, laboratory results, and other protocol-specified assessments, to allow safety signal detection throughout the study.

Table 5 shows the minimal required timing of reviews by the SRC, as well as the decisions that the SRC may undertake with respect to the possible occurrence of study stopping rules (see Section 7.4).

At its discretion, and based on clinical judgment, the SRC may suspend DCR-AUD dosing or EIAs during the study at any time, even for reasons not explicitly stated elsewhere, and refer to the DSMC for any potential concern regarding participant safety or wellbeing. In addition, the DSMC will be convened on an ad hoc basis if an AE does trigger a stopping rule. The DSMC will review data and advise on whether the study should be stopped to ensure the safety of study participants.

Complete details on the responsibilities of the Safety Committees will be detailed in the committee charters. For additional details on the composition of the SRC and DSMC, refer to Section 10.1.5.

Table 5: Minimal Timing of the Safety Review Committee Data Reviews

Occurrence of a significant or severe adverse symptom	Upon the occurrence of a significant or severe adverse symptom (see Section 7.3), all dosing and enrollment into the study will be interrupted until safety data can be reviewed by the SRC. The SRC will review the events associated with the significant or severe adverse symptom as well as the cumulative safety data collected on all participants previously dosed. The SRC will determine whether dosing, enrollment, and subsequent EIAs may resume.
Occurrence of a safety issue that, in the SRC's clinical judgment, could affect participant safety or study conduct	Upon the occurrence of a situation or safety question that could affect participant safety or study conduct, the SRC will meet to discuss the issue. Actions to be taken include, but are not limited to: • Discontinuation of an individual participant • Discontinuation of dosing • Discontinuation of additional EIAs • Referral to the DSMC for review and recommendation, should a pattern of AEs of concern be identified, regardless of severity
Occurrence of a potential study stopping rule	Upon the occurrence of a potential study stopping rule (see Section 7.4), all dosing and enrollment will be interrupted until safety data can be reviewed by the SRC. The SRC will review the events associated with the triggering of the stopping rule as well as the cumulative safety data collected on all participants previously dosed. The SRC will determine whether dosing and enrollment may resume. The SRC may elect to refer the issue that triggered the study interruption to the DSMC for an independent review and recommendation regarding study resumption.

Abbreviations: AE: adverse event; AUD: alcohol use disorder; DSMC: Data Safety Monitoring Committee; EIA: ethanol interaction assessment; SRC: Safety Review Committee

10. SUPPORTING DOCUMENTATION AND OPERATIONAL CONSIDERATIONS

10.1. Regulatory, Ethical, and Study Oversight Considerations

10.1.1. Regulatory and Ethical Considerations

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

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

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

Any amendments to the protocol will require IRB/IEC approval before implementation of changes made to the study design, except for changes necessary to eliminate an immediate hazard to study participants.

The Investigator will be responsible for the following:

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

10.1.2. Financial Disclosure

Investigators and sub-Investigators will provide the Sponsor with sufficient, accurate financial information as requested to allow the Sponsor to submit complete and accurate financial certification or disclosure statements to the appropriate regulatory authorities. Investigators are responsible for providing information on financial interests during the course of the study and for 1 year after completion of the study.

10.1.3. Informed Consent Process

Each interested prospective participant (or legally authorized representative) will receive an informed consent document that contains study information and be given ample time to read the information and consider participation. The Investigator or his/her representative will explain the nature of the study and its risks and potential benefits to the prospective participant or his/her legally authorized representative and answer all questions regarding the study.

Prospective participants must be informed that the study involves research, their participation is voluntary, and that the choice not to participate or to discontinue the study at any time will involve no penalty or loss of benefits to which the individual is otherwise entitled. Consenting participants, or their legally authorized representatives, will be required to sign a statement of informed consent that meets the requirements of 21 CFR 50, local regulations, ICH guidelines, Health Insurance Portability and Accountability Act (HIPAA) requirements, where applicable, and the IRB/IEC or study center.

The medical record must include a statement that written informed consent was obtained before the participant was enrolled in the study, and the date the written consent was obtained. The authorized person obtaining the informed consent must also sign the ICF.

Participants must be re-consented to the most current version of the ICF(s) during their participation in the study.

A copy of the ICF(s) must be provided to the participant or the participant's legally authorized representative.

10.1.4. Data Protection

Participants will be assigned a unique identifier by the Sponsor. Any participant records or datasets that are transferred to the Sponsor will contain the identifier only; participant names or any information which would make the participant identifiable will not be transferred.

The participant must be informed that his/her personal study-related data will be used by the Sponsor in accordance with local data protection law. The level of disclosure must also be explained to the participants, who will be required to give consent for their data to be used as described in the informed consent

The participant must be informed that his/her medical records may be examined by Clinical Quality Assurance auditors or other authorized personnel appointed by the Sponsor, by appropriate IRB/IEC members, and by inspectors from regulatory authorities.

10.1.5. Committees Structure

10.1.5.1. Safety Review Committee

The SRC will comprise the PI and/or sub-Investigator, the Sponsor Medical Monitor, and the CRO Medical Monitor.

See Section 9.5 for further details regarding the SRC.

10.1.5.2. Data Safety Monitoring Committee

The independent DSMC will comprise appropriately qualified clinicians who are not involved in the study. The DSMC will provide a comprehensive review of study safety and tolerability data on an ad hoc basis. Further operational details will be prespecified in the DSMC charter.

10.1.6. Dissemination of Clinical Study Data

Data collected during the study will be shared with Investigators whose proposed use of the data has been approved by an independent review committee. A tabular summary of study results will be posted to www.ClinicalTrials.gov within 1 year of study completion.

10.1.7. Data Quality Assurance

[redacted] will use an electronic data capture system to completely collect all medical data during this trial. ClinBase™ is a software tool designed similarly to an electronic medical record for the documentation of e.g., medical history, demographics, vital signs, and AEs. Unlike a system (which is relying on data from other sources like paper records and is setup just to digitize these data for transmission), ClinBase ensures quality assurance and enables comprehensive capture of all relevant medical information gathered during clinical studies. Through a system provided workflow that includes barcode scanning and interfaces to medical equipment, trial operations performance is controlled and captured in real time. Thus, the system provides 100% transparency for monitors and is at any time accessible to the Investigators and clinical personnel for all medical documentation processes.

ClinBase meets ICH GCP requirements as data are attributable, legible, contemporaneous, original, accurate, and complete (ALCOA-C). The system is fully Code of Federal Regulations (CFR) Title 21 Part 11 compliant.

The Investigator will ensure the accuracy, completeness and timeliness of the data reported to the Sponsor. System supported data collection processes and procedures are validated to ensure completeness, accuracy, reliability, and consistency. A complete audit trail is maintained of all data changes. The Investigator or designee will cooperate with the Sponsor's representative(s) for the periodic review of trial documents to ensure the accuracy and completeness of the data capture system at each scheduled monitoring visit.

Electronic consistency checks (plausibility and completeness) will be used along with the Investigator review to identify any errors or inconsistencies in the data. Data clarification requests will be provided to the trial team by means of electronic or manual queries.

During the trial setup, adequate and accurate clinical procedure forms are used to generate medical records, digital ECGs, AE and concomitant medication reporting, raw data collection forms, etc., which are designed as protocol to record all observations and other pertinent data for each subject receiving trial medication. The only regularly used paper-based trial document is the ICF because it requires wet-ink signatures. The ICFs will be archived in paper form at the end of the trial. Should additional paper forms be used, the recorded data must be transferred into ClinBase accurately and within due time. In pre-stabilized intervals or at the end of the trial, data will be exported from ClinBase data management systems for further processing.

The Investigator will allow Sponsor representatives, contract designees, authorized regulatory authority inspectors and the IRB to have direct access to all electronic records pertaining to the trial.

All data should be recorded, handled, and stored in a way that allows its accurate reporting, interpretation, and verification.

Monitoring details describing strategy (e.g., risk-based initiatives in operations and quality such as Risk Management and Mitigation Strategies and Analytical Risk-Based Monitoring), methods, responsibilities, and requirements, including handling of noncompliance issues and monitoring techniques (central, remote, or on-site monitoring) are provided in the Monitoring Plan.

The Sponsor or designee is responsible for the data management of this study, including quality checking of the data.

The Sponsor assumes accountability for actions delegated to other individuals (e.g., CROs).

Study monitors will perform ongoing source data verification to confirm that data entered into the eCRF by authorized site personnel are accurate, complete, and verifiable from source documents; that the safety and rights of participants are being protected; and that the study is being conducted in accordance with the currently approved protocol and any other study agreements, ICH GCP, and all applicable regulatory requirements.

Records and documents, including signed ICFs, pertaining to the conduct of this study must be retained by the Investigator for 5 years after study completion unless local regulations or institutional policies require a longer retention period. No records may be destroyed during the retention period without the written approval of the Sponsor. No records may be transferred to another location or party without written notification to the Sponsor.

10.1.8. Study and Site Start and Closure

The study start date is the date on which the clinical study will be open for recruitment of participants.

The first act of recruitment is site activation and will be the study start date.

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

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

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

- Investigator noncompliance with the protocol, GCP, or regulatory requirements
- Unsatisfactory enrollment with respect to quantity or quality
- Incomplete data collection; inaccurate or knowingly false data submission
- The PI is no longer capable of performing the tasks of the PI, and no replacement can be found.
- The SRC determines that termination of the study is in the best interest of the research participants.
- The Sponsor, Investigator or IRB/IEC determines that continuation of the study will not serve any scientific purpose.

- Circumstances beyond the control of the Sponsor or Investigator make it unreasonable to require the study's continuation.
- A request to discontinue the study by a regulatory or health authority.

If the study is prematurely terminated or suspended, the Sponsor shall promptly inform the Investigators, the IECs/IRBs, the regulatory authorities, and any CRO(s) used in the study of the reason for termination or suspension, as specified by the applicable regulatory requirements. The Investigator shall promptly inform the participants and should assure appropriate participant therapy and/or follow-up. The complete set of stopping rules is detailed in Section 7.

10.1.9. Publication Policy

Publication of a summary of the results of the study is permissible according to the Sponsor and is not inconsistent with the preceding affirmation. Scientific dissemination of the results of this study is encouraged. Any formal publication of data collected as a result of the study will be considered a joint publication by the Investigator and the appropriate personnel of the Sponsor or their designees. Authorship will be determined by mutual agreement and in line with International Committee of Medical Journal Editors authorship requirements.

No submission of a manuscript may be made until the results from the study site have been received and analyzed by Sponsor, or the study has been terminated at all centers. A separate, individual publication of the results of the study will be delayed until initial publication of the results of the multi-center study, or a decision not to publish is made. Please refer to clinical study agreement for further details.

For any publication or presentation, a manuscript of the paper or abstract must be received and approved by the Sponsor prior to outside submission. Whether or not there is a Sponsor author of the publications or presentations, a manuscript will be forwarded to the Sponsor for review and approval at least 30 days prior to submission of a journal publication, or at least 10 days prior to submission of an abstract.

Note: Any language relating to these issues appearing in the clinical study agreement will supersede that which is outlined in this section.

10.2. Clinical Laboratory Tests

Table 6 summarizes the laboratory tests to be conducted. Full details for the testing are provided in the laboratory manual. Protocol-specific requirements for inclusion or exclusion of participants are detailed in Section 5.

Additional tests may be performed during the study as determined necessary by the Investigator or required by local regulations.

Investigators must document their review of each laboratory safety report.

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

Table 6: Protocol-Required Safety Laboratory Assessments

Laboratory Assessments	Parameters	
Hematology	Complete Blood Cell Count: red blood cells hemoglobin hematocrit platelets white blood cells	Complete Blood Cell Count with differential and RBC indices: red blood cells hemoglobin hematocrit platelets reticulocytes mean corpuscular volume (MCV) mean corpuscular hemoglobin (MCH) mean corpuscular hemoglobin concentration (MCHC) white blood cells: lymphocytes, abs and % monocytes, abs and % eosinophils, abs and % neutrophils, abs and % basophils, abs and %
Coagulation parameters	International normalized ratio (INR), prothrombin time (PT), partial thromboplastin time (PTT)	
Clinical chemistry	alanine transaminase (ALT) aspartate transaminase (AST) gamma-glutamyl transferase (GGT) alkaline phosphatase (ALP) bilirubin (total and direct) lactate dehydrogenase (LDH) total protein albumin	creatine kinase (CK) sodium chloride potassium creatinine blood urea nitrogen (BUN) fasting blood glucose
Urinalysis	specific gravity, pH, glucose, protein, blood, ketones, bilirubin, urobilinogen, nitrite, leukocyte esterase by dipstick microscopic examination (if blood or protein is abnormal)	
Complement panel	Bb, C3a, C5a	
Immunogenicity	Antidrug antibodies	
Laboratory tests for admission to clinic	SARS-CoV-2 (local laboratory testing) Breath alcohol test (local laboratory testing) Urine drug screen (to include at minimum: amphetamines, barbiturates, cannabis, cocaine, opiates, and benzodiazepines)	
Pregnancy testing	Highly sensitive urine hCG pregnancy test (in female participants). Any positive urine pregnancy test will be confirmed with a serum pregnancy test (local laboratory testing)	
Other tests performed at Screening	Follicle-stimulating hormone (as needed to confirm menopausal status) Viral serology (human immunodeficiency virus antibody, hepatitis B surface antigen, and hepatitis C virus antibody) Hemoglobin A1c Urine drug screen (to include at minimum: amphetamines, barbiturates, cocaine, opiates, and benzodiazepines)	

10.3. Adverse Events: Definitions and Procedures for Recording, Evaluating, Follow-up, and Reporting

10.3.1. Definition of Adverse Event

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

Events Meeting the AE Definition

- Any abnormal laboratory test results (hematology, clinical chemistry, or urinalysis) or other safety assessments (e.g., ECG, radiological scans, vital signs measurements), including those that worsen from baseline, considered clinically significant in the medical and scientific judgment of the Investigator.
- Exacerbation of a chronic or intermittent pre-existing condition, including either an increase in frequency and/or intensity of the condition.
- New condition detected or diagnosed after study intervention administration, even though it may have been present before the start of the study.
- Signs, symptoms, or the clinical sequelae of a suspected drug-drug interaction.
- Signs, symptoms, or the clinical sequelae of a suspected overdose of either study intervention or a concomitant medication. Overdose per se will not be reported as an AE/SAE unless it is an intentional overdose taken with possible suicidal/self-harming intent. Such overdoses should be reported regardless of sequelae.

Events NOT Meeting the AE Definition

- During the conduct of EIAs, participants will be questioned by site staff to assess the 6 symptoms (flushing, headache, lightheadedness, palpitations, nausea, and vomiting) associated with increased blood acetaldehyde levels. These symptoms solicited via active questioning do not meet the AE definition.
- Medical or surgical procedure (e.g., endoscopy, appendectomy): the condition that leads to the procedure is the AE.
- Situations in which an untoward medical occurrence did not occur (social and/or convenience admission to a hospital).
- Anticipated day-to-day fluctuations of pre-existing disease(s) or condition(s) present or detected at the start of the study that do not worsen.

10.3.2. Definition of Serious Adverse Event

If an event is not an AE per definition above, then it cannot be an SAE even if serious conditions are met.

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

- **Results in death**
- **Is life-threatening**

The term 'life-threatening' in the definition of 'serious' refers to an event in which the participant was at risk of death at the time of the event. It does not refer to an event that hypothetically might have caused death if it were more severe.

- **Requires inpatient hospitalization or prolongation of existing hospitalization**

In general, hospitalization signifies that the participant has been detained (usually involving at least an overnight stay) at the hospital or emergency ward for observation and/or treatment that would not have been appropriate in the physician's office or outpatient setting. Complications that occur during hospitalization are AEs. If a complication prolongs hospitalization or fulfills any other serious criteria, the event is serious. When in doubt as to whether "hospitalization" occurred or was necessary, the AE should be considered serious.

Hospitalization for elective treatment of a pre-existing condition that did not worsen from baseline is not considered an AE. Pre-planned hospitalizations scheduled prior to signing the ICF, for an elective medical/surgical procedure do not meet this criterion.

- **Results in persistent disability/incapacity**

The term disability means a substantial disruption of a person's ability to conduct normal life functions.

This definition is not intended to include experiences of relatively minor medical significance such as uncomplicated headache, nausea, vomiting, diarrhea, influenza, and accidental trauma (e.g., sprained ankle), which may interfere with or prevent everyday life functions but do not constitute a substantial disruption.

- **Is a congenital anomaly/birth defect**
- **Other situations:**

Medical or scientific judgment should be exercised in deciding whether SAE reporting is appropriate in other situations, such as important medical events that may not be immediately life-threatening or result in death or hospitalization but may jeopardize the participant or may require medical or surgical intervention to prevent one of the other outcomes listed in the above definition. These events should usually be considered serious.

Examples of such events include invasive or malignant cancers, intensive treatment in an emergency room or at home for allergic bronchospasm, blood dyscrasias or convulsions that do not result in hospitalization, or development of drug dependency or drug abuse.

10.3.3. Definition of Adverse Events of Special Interest

Per the Council for International Organizations of Medical Sciences Cumulative Pharmacovigilance Glossary (CIOMS VI, 2021), an AESI (serious or non-serious) is one of scientific and medical concern specific to the Sponsor's product or program, for which ongoing monitoring and rapid communication by the Investigator to the Sponsor may be appropriate. Such events may require further investigation in order to characterize and understand them.

Events Meeting the AESI Definition

Injection Site Reaction

An ISR is characterized by an intense adverse reaction (usually immunologic) developing at the site of an injection after 4 or more hours post-injection. Subcutaneous administration of the study intervention may cause a specific local reaction originating around the injection site, such as erythema, itching, discomfort and pain, and could include more severe manifestations such as ulceration or necrosis.

Potential ISRs will be evaluated and recorded as follows:

Individual signs or symptoms at the injection site with an onset of less than 4 hours from the time of study intervention administration will be recorded as individual AEs (bruising, itching, transient erythema, swelling, etc.) and not as ISRs.

Signs or symptoms at the injection site with a time to onset of 4 or more hours post-injection will be recorded as an ISR if any of the CTCAE criteria for ISR detailed in Table 7 are met. Signs or symptoms at the injection site that occur ≥ 4 hours post-injection but are not specified in Table 7 should be recorded as AEs.

Table 7: Grading of Injection Site Reactions, CTCAE v 5.0

Grade 1	Grade 2	Grade 3	Grade 4	Grade 5
Tenderness with or without associated symptoms (e.g., warmth, erythema, itching)	Pain, lipodystrophy; edema; phlebitis	Ulceration or necrosis; severe tissue damage; operative intervention indicated	Life-threatening consequences; urgent intervention indicated	Death

10.3.4. Recording and Follow-up of AEs and/or SAEs

10.3.4.1. AE and SAE Recording

When an AE/SAE occurs, it is the responsibility of the Investigator to review all documentation (e.g., hospital progress notes, laboratory reports, diagnostics reports) related to the event.

The Investigator will then record all relevant AE/SAE information in the eCRF.

It is not acceptable for the Investigator to send photocopies of the participant's medical records to the Sponsor in lieu of completion of the AE/SAE CRF page.

There may be instances when copies of medical records for certain cases are requested by the Sponsor. In this case, all participant identifiers, with the exception of the participant number, will be redacted on the copies of the medical records before submission to the Sponsor.

The Investigator will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. Whenever possible, the diagnosis (not the individual signs/symptoms) will be documented as the AE/SAE.

10.3.4.2. Assessment of Intensity

The Investigator will make an assessment of each AE and SAE reported during the study. The grade of each event will be determined using CTCAE v 5.0 criteria.

10.3.4.3. Assessment of Causality

The Investigator is obligated to assess the relationship between study intervention and each occurrence of each AE/SAE.

A "reasonable possibility" of a relationship conveys that there are facts, evidence, and/or arguments to suggest a causal relationship, rather than a relationship cannot be ruled out.

The Investigator will use clinical judgment to determine the relationship. Alternative causes, such as concomitant therapy and other risk factors, as well as the temporal relationship of the event to study intervention administration will be considered and investigated. The Investigator will also consult the Investigator Brochure (IB) and/or Product Information (for marketed products) in his/her assessment.

For each AE/SAE, the Investigator must document in the medical notes that he/she has reviewed the AE/SAE and has provided an assessment of causality.

There may be situations in which an SAE has occurred, and the Investigator has minimal information to include in the initial report. However, it is very important that the Investigator always make an assessment of causality for every event before the initial transmission of the SAE data to SAE Coordinator.

The Investigator may change his/her opinion of causality in light of follow-up information and send a SAE follow-up report with the updated causality assessment.

The causality assessment is one of the criteria used when determining regulatory reporting requirements.

The following definitions will be used in assessing causality:

- **Not Related:** Event for which sufficient evidence exists to conclude that the etiology is unrelated to study intervention.
- **Possibly Related:** There is some temporal relationship between the event and the administration of the study intervention, and the event is unlikely to be explained by the participant's medical condition or other therapies.
- **Probably Related:** The temporal relationship between the event and administration of the study intervention is suggestive and the event is unlikely explained by the participant's medical condition or other therapies.
- **Definitely Related:** The event follows reasonable temporal sequence from administration of the study intervention, follows a known or suspected response pattern to the study intervention, is confirmed by improvement upon stopping the study intervention, and reappears upon repeated exposure, if that occurs.

10.3.4.4. Follow-up of AEs and SAEs

The Investigator is obligated to perform or arrange for the conduct of supplemental measurements and/or evaluations as medically indicated or as requested by the Sponsor to elucidate the nature and/or causality of the AE or SAE as fully as possible. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals.

If a participant dies during participation in the study or during a recognized follow-up period, the Investigator will provide the Sponsor with a copy of any post-mortem findings including histopathology.

New or updated information will be recorded in the originally completed CRF.

The Investigator will submit any updated SAE data to the Sponsor within 24 hours of receipt of the information.

10.3.5. Reporting of SAEs

Facsimile or electronic transmission of the SAE Report Form is the preferred method to transmit this information to the SAE coordinator.

In rare circumstances and in the absence of facsimile equipment, notification by telephone is acceptable with a copy of the SAE data collection tool sent by overnight mail or courier service.

Initial notification via telephone does not replace the need for the Investigator to complete and sign the SAE Report Form within the designated reporting time frames.

Contacts for SAE reporting can be found on the Medical Monitor and Pharmacovigilance Contact Information page.

10.4. Contraceptive Guidance and Collection of Pregnancy Information

10.4.1. Definitions

Woman of Childbearing Potential (WOCBP)

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

If fertility is unclear (e.g., amenorrhea in athletes) and a menstrual cycle cannot be confirmed before administration of study intervention, additional evaluation should be considered.

Women in the following categories are not considered WOCBP

1. Premenarchal
2. Premenopausal with one of the following:
    a. Documented hysterectomy
    b. Documented bilateral salpingectomy
    c. Documented bilateral oophorectomy

For individuals with permanent infertility due to an alternate medical cause other than the above, (e.g., müllerian agenesis, androgen insensitivity), Investigator discretion should be applied to determining study entry.

Note: Documentation can come from the site personnel's: review of the participant's medical records, medical examination, or medical history interview.

3. Postmenopausal

   a. A postmenopausal state is defined as no menses for 12 months without an alternative medical cause.

      A high follicle stimulating hormone (FSH) level in the postmenopausal range may be used to confirm a postmenopausal state in women not using hormonal contraception or hormonal replacement therapy (HRT). However, in the absence of 12 months of amenorrhea, confirmation with more than one FSH measurement is required.

   b. Participants on HRT and whose menopausal status is in doubt will be required to use one of the non-estrogen hormonal highly effective contraception methods if they wish to continue their HRT during the study. Otherwise, they must discontinue HRT to allow confirmation of postmenopausal status before study enrollment.

10.4.2. Contraception Guidance

Male Participants

Male participants with female partners of childbearing potential are eligible to participate if they agree to ONE of the following from Day 1 through the end of study (EOS) visit:

- Are abstinent from penile-vaginal intercourse as their usual and preferred lifestyle (abstinent on a long-term and persistent basis) and agree to remain abstinent.
- Agree to use a male condom plus partner use of a contraceptive method with a failure rate of < 1% per year as described in Table 8 when having penile-vaginal intercourse with a WOCBP who is not currently pregnant.

Male participants must refrain from donating sperm for the duration of the study and following the last dose of study intervention for at least 24 weeks.

Male participants with a pregnant or breastfeeding partner must agree to remain abstinent from penile-vaginal intercourse or use a male condom during each episode of penile penetration during the protocol-defined time frame.

Female Participants

Female participants of childbearing potential are eligible to participate if they agree to use a highly effective method of contraception consistently and correctly as described in Table 8.

Table 8: Highly Effective Contraceptive Methods

Highly Effective Contraceptive Methods That Are User Dependent [a] ***(Failure rate of <1% per year when used consistently and correctly)***
Combined (estrogen- and progestogen-containing) hormonal contraception associated with inhibition of ovulation • Oral • Intravaginal • Transdermal Progestogen-only hormonal contraception associated with inhibition of ovulation • Oral • Injectable Sexual abstinence Sexual abstinence is considered a highly effective method only if defined as refraining from heterosexual intercourse during the entire period of risk associated with the study intervention. The reliability of sexual abstinence needs to be evaluated in relation to the duration of the study and the preferred and usual lifestyle of the participant.
Highly Effective Methods That Are User Independent [a]
• Implantable progestogen-only hormonal contraception associated with inhibition of ovulation • Intrauterine device (IUD) • Intrauterine hormone-releasing system (IUS) • Bilateral tubal occlusion • Vasectomized partner A vasectomized partner is a highly effective contraception method provided that the partner is the sole male sexual partner of the WOCBP, and the absence of sperm has been confirmed. If not, an additional highly effective method of contraception should be used.

10.4.3. Collection of Pregnancy Information

10.4.3.1. Male Participants with Partners Who Become Pregnant

The Investigator will attempt to collect pregnancy information on any male participant's female partner who becomes pregnant while the male participant is in this study. This applies only to male participants who receive DCR-AUD.

After obtaining the necessary signed informed consent from the pregnant female partner directly, the Investigator will record pregnancy information on the appropriate form and submit it to the Sponsor within 24 hours of learning of the partner's pregnancy. The female partner will also be followed to determine the outcome of the pregnancy. Information on the status of the mother and

child will be forwarded to the Sponsor. Generally, the follow-up will be no longer than 6 to 10 weeks following the estimated delivery date. Any termination of the pregnancy will be reported regardless of fetal status (presence or absence of anomalies) or indication for the procedure.

10.4.3.2. Participants Who Become Pregnant

The Investigator will collect pregnancy information on any participant who becomes pregnant while participating in this study. The initial Information will be recorded on the appropriate form and submitted to the Sponsor within 24 hours of learning of a participant's pregnancy.

The participant will be followed to determine the outcome of the pregnancy. The Investigator will collect follow-up information on the participant and the neonate, and the information will be forwarded to the Sponsor. Generally, follow-up will not be required for longer than 6 to 10 weeks beyond the estimated delivery date. Any termination of pregnancy will be reported, regardless of fetal status (presence or absence of anomalies) or indication for the procedure.

While pregnancy itself is not considered to be an AE or SAE, any pregnancy complication or elective termination of a pregnancy for medical reasons will be reported as an AE or SAE. A spontaneous abortion (occurring at < 22 weeks gestational age) or still birth (occurring at > 22 weeks gestational age) is always considered to be an SAE and will be reported as such. Any post-study pregnancy-related SAE considered reasonably related to the study intervention by the Investigator will be reported to the Sponsor as described in Section 8.3.4. While the Investigator is not obligated to actively seek this information in former study participants, he or she may learn of an SAE through spontaneous reporting.

Any participant who becomes pregnant while participating in the study will discontinue study intervention.

10.5. Genetics

Use/Analysis of DNA

- DNA samples will be analyzed for *ALDH2* genotyping. Additional exploratory analyses may be conducted if it is hypothesized that this may help further understand the clinical data.
- The results of genetic analyses may be reported in the CSR or in a separate study summary.
- The Sponsor will store the DNA samples in a secure storage space with adequate measures to protect confidentiality.
- The samples will be retained while research on DCR-AUD continues, but no longer than 5 years or other period as per local requirements.

10.6. Liver Safety: Suggested Actions and Follow-up Assessments

Phase 1 liver chemistry stopping criteria are designed to ensure participant safety and to evaluate liver event etiology:

Liver Chemistry Stopping Criteria (Applies both to additional DCR-AUD dosing and further EIAs)	
ALT/AST-absolute	ALT or AST increases to ≥ 3×ULN If ALT or AST ≥ increases to >2×ULN **AND** total bilirubin ≥ 2×ULN (> 35% direct bilirubin) or INR > 1.5, report to Sponsor in expedited manner.[a, b] See additional actions and follow-up assessments below

[a] Serum bilirubin fractionation should be performed if testing is available. If serum bilirubin fractionation is not immediately available, discontinue study intervention if ALT ≥ 3×ULN and total bilirubin ≥ 2×ULN. Additionally, if serum bilirubin fractionation testing is unavailable, record the absence/presence of detectable urinary bilirubin on dipstick which is indicative of direct bilirubin elevations suggesting liver injury.

[b] All events of ALT ≥ 3×ULN and total bilirubin ≥ 2×ULN (> 35% direct bilirubin) or ALT ≥ 3×ULN and INR > 1.5 may indicate severe liver injury (possible 'Hy's Law') and must be reported to sponsor in an expedited manner and as an SAE if SAE criteria met (excluding studies of hepatic impairment or cirrhosis).

Required Actions, Monitoring, and Follow-up Assessments

Actions:

- Immediately discontinue study intervention
- Report the event to the CRO Medical Monitor **within 24 hours**
- Complete an SAE data collection tool if the event also met the criteria for an SAE [b]
- Perform liver function follow-up assessments as described in the Follow Up Assessments below.
- Do not restart or rechallenge participant with study intervention (neither IMP nor additional EIAs).
- Monitor the participant until liver function test abnormalities resolve, stabilize, or return to baseline (as detailed below).

Monitoring:

If ALT or AST ≥ 3×ULN AND total bilirubin ≥ 2×ULN or INR > 1.5

- Repeat liver function tests (include ALT, AST, alkaline phosphatase, total bilirubin, and INR) and perform liver function follow-up assessments within **24 hours.**
- Monitor participant twice weekly until liver function test abnormalities resolve, stabilize, or return to baseline.
- A specialist or hepatology consultation is recommended.

If ALT or AST ≥ 3×ULN AND total bilirubin < 2×ULN and INR ≤ 1.5:

- Perform liver function tests (include ALT, AST, alkaline phosphatase, total bilirubin, and INR) and perform liver function follow-up assessments within **24 to 72 hours**
- Monitor participants weekly until liver function abnormalities resolve, stabilize, or return to baseline

Follow-Up Assessments:

- Viral hepatitis serology (includes Hepatitis A IgM antibody; HBsAg and HBcAb; hepatitis C RNA; cytomegalovirus IgM antibody; Epstein-Barr viral capsid antigen IgM antibody [or if unavailable, heterophile antibody or monospot testing]; and hepatitis E IgM antibody)
- Obtain serum CK, LDH, GGT, GLDH, and serum albumin
- Fractionate bilirubin, if total bilirubin ≥ 2×ULN
- Obtain complete blood count with differential to assess eosinophilia
- Record the appearance or worsening of clinical symptoms of liver injury, or hypersensitivity, on the eCRF
- Record use of concomitant medications or ingestions (including wild mushrooms, acetaminophen, herbal remedies, recreational drugs, and other over-the-counter medications). Note any concomitant medications that have been recently initiated or for which doses have increased.
- Record alcohol use on the eCRF

If ALT or AST ≥ 3×ULN AND total bilirubin ≥ 2×ULN or INR > 1.5 obtain the following in addition to the assessments listed above**:**

- Anti-nuclear antibody, anti-smooth muscle antibody, Type 1 anti-liver kidney microsomal antibodies, and quantitative total immunoglobulin G (IgG) or gamma globulins
- Liver imaging (ultrasound, magnetic resonance, or computerized tomography) and/or liver biopsy to evaluate liver disease
- Liver biopsy may be considered and discussed with local specialists if available, for instance:
  - When serology raises the possibility of autoimmune hepatitis (AIH)
  - When suspected DILI progresses or fails to resolve on withdrawal of study intervention
  - In participants with acute or chronic atypical presentation.

10.7. Abbreviations

Abbreviation	Description
ADA	antidrug antibody(ies)
AE	adverse event
AESI	AE of special interest
ALDH2	aldehyde dehydrogenase 2
ALDH2	gene for aldehyde dehydrogenase 2
ALP	alkaline phosphatase
ALT	alanine aminotransferase
AST	aspartate aminotransferase
AUC	area under the curve
AUD	alcohol use disorder
BAC	blood alcohol concentration
BMI	body mass index
BP	blood pressure
bpm	beats per minute
BUN	blood urea nitrogen
°C	degrees Celsius
CFR	Code of Federal Regulations
CFU	conditional follow-up
CIOMS	Council for International Organizations of Medical Sciences
CK	creatine kinase (creatine phosphokinase)
CL	clearance
C_{max}	maximum observed concentration
C_{min}	minimum observed concentration
CONSORT	Consolidated Standards of Reporting Trials
CRO	contract research organization
CSR	clinical study report
C-SSRS	Columbia-Suicide Severity Rating Scale
CTCAE	Common Terminology Criteria for Adverse Events
CV%	coefficient of variation (percent)
DCR-A1203	drug substance for this IMP

Abbreviation	Description
DS	drug substance
DSMC	Data Safety Monitoring Committee
ECG	electrocardiogram
eCRF	electronic case report form
e.g.	for example (*exempli gratia*)
EIA	ethanol interaction assessment
EOS	end of study
EOT	end of treatment
ETOH	ethyl alcohol
FDA	Food and Drug Administration
FIH	first-in-human
FSH	follicle stimulating hormone
GABA	gamma-aminobutyric acid
GalNAc	*N*-acetylgalactosamine
GCP	Good Clinical Practice
GGT	gamma glutamyl transferase
GI	gastrointestinal
GLP	Good Laboratory Practice
HbA1c	hemoglobin A1c
HBsAg	hepatitis B surface antigen
hCG	human chorionic gonadotropin
HCV	hepatitis C virus
HIPPA	Health Insurance Portability and Accountability Act
hr	hour(s)
HIV	human immunodeficiency virus
HRT	hormone replacement therapy
HV	healthy volunteer
IA	interim analysis
IB	Investigator's Brochure
ICF	informed consent form
ICH	The International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use

Abbreviation	Description
i.e.	that is (*id est*)
IEC	Independent Ethics Committee
IMP	Investigational Medicinal Product
INR	international normalized ratio
IRB	Institutional Review Board
ISR	injection site reaction
ITT	Intent to treat
IUD	intrauterine device
IVRS/IWRS	Interactive Voice/Web Response System
IUS	intrauterine hormone-releasing system
kg	kilogram(s)
LDH	lactate dehydrogenase
LFT	liver function test
MCH	mean corpuscular hemoglobin
MCHC	mean corpuscular hemoglobin concentration
MCV	mean corpuscular volume
mg	milligram(s)
min	minute(s)
MINI	Mini International Neuropsychiatric Interview
mL	milliliter(s)
mRNA	messenger ribonucleic acid
NOAEL	no observed adverse effect level
NPO	nothing by mouth
NSAID	nonsteroidal anti-inflammatory drug
OTC	over the counter
PD	pharmacodynamic(s)
PEth	phosphatidylethanol
PK	pharmacokinetic
PI	Principal Investigator
QTcF	QT interval (Fridericia correction)
RNA	ribonucleic acid
RNAi	RNA interference

Abbreviation	Description
SAE	serious adverse event
SAP	statistical analysis plan
SC	subcutaneous
SD	standard deviation
SIB	suicidal ideation or behavior
siRNA	small interfering RNA
SoA	schedule of activities
SRC	Safety Review Committee
SUD	substance abuse disorder
SUSAR	suspected unexpected serious adverse reaction
t1/2	terminal elimination half-life
TBD	to be determined
TLFB	Timeline Follow Back
T_{max}	time to maximum observed concentration
ULN	upper limit of normal
UTI	urinary tract infection
Vd/F	apparent volume of distribution
WFI	water for injection
WOCBP	woman(en) of childbearing potential

10.8. Protocol Amendment History

Amendment 1 (17-Jan-2023)

Overall Rationale for the Amendment

The protocol was amended in response to queries from the US Food and Drug Administration. Additional updates and clarifications were also included.

The Protocol Amendment Summary of Changes Table for the current amendment is located directly before the Table of Contents.

10.9. Investigator Signature Page

A Phase 1b, Double-blind, Placebo-controlled, Repeat-dose, Safety, Tolerability, Pharmacokinetic, and Pharmacodynamic Study of DCR-AUD in Healthy Volunteers

Protocol Number: DCR-AUD-102

Version: 2.0

Date: 17-Jan-2023

I have read this protocol and agree to conduct this trial in accordance with all stipulations of the protocol and in accordance with Good Clinical Practice (GCP) guidelines, the Declaration of Helsinki, and local regulations (as applicable).

Signature:	**Date:**
Name:	**Title:**
Institution:	**Site Number:**

11. REFERENCES

Alling C, Gustavsson L, Anggard E: An abnormal phospholipid in rat organs after ethanol treatment. FEBS Lett 1983; 152: 24–8.

Bae KY, Kim SW, Shin HY, Kim JM, Shin IS, Kim SJ, et al. The acute effects of ethanol and acetaldehyde on physiological responses after ethanol ingestion in young healthy men with different ALDH2 genotypes. Clin Tox. 2012 Apr 1;50(4):242-9.

Bryson WC, McConnell J, Korthuis PT, McCarty D. Extended-release naltrexone for alcohol dependence: persistence and healthcare costs and utilization. Am J Manag Care. 2011 Jun;17 Suppl 8:S222-34.

Chick J, Gough K, Falkowski W, Kershaw P, Hore B, Mehta B, et al. Disulfiram treatment of alcoholism. Br J Psychiatry. 1992 Jul;161:84-9.

CIOMS Cumulative Pharmacovigilance Glossary, Geneva, Switzerland: Council for International Organizations of Medical Sciences (CIOMS), 2021.

EMA/CHMP/EWP/20097/2008 (previously EMEA/CHMP/EWP/20097/2008). Committee for Medicinal Products for Human Use (CHMP). Guideline on the development of medicinal products for the treatment of alcohol dependence. 18 February 2010.

Enomoto N, Takase S, Yasuhara M, Takada A. Acetaldehyde metabolism in different aldehyde dehydrogenase-2 genotypes. Alcohol Clin Exp Res. 1991;15(1):141-4.

Gastfriend DR. A pharmaceutical industry perspective on the economics of treatments for alcohol and opioid use disorders. Ann NY Acad Sci. 2014 Oct;1327:112-30.

Gnann H, Weinmann W, Thierauf A. Formation of phosphatidylethanol and its subsequent elimination during an extensive drinking experiment over 5 days. Alcohol Clin Exp Res. 2012 Sep;36(9):1507-11.

Grant BF, Goldstein RB, Saha TD, Chou SP, Jung J, Zhang H, et al. Epidemiology of DSM-5 Alcohol Use Disorder: Results from the National Epidemiologic Survey on Alcohol and Related Conditions III. JAMA Psychiatry. 2015 Aug;72(8):757-66.

Han B, Hedden SL, Lipari R, Copello EA, Kroutil LA. Receipt of services for behavioral health problems: Results from the 2014 National Survey on Drug Use and Health. Rockville, MD: Substance Abuse and Mental Health Services Administration. 2015 Sep. Available from: https://www.samhsa.gov/data/report/receipt-services-behavioral-health-problems-results-2014-national-survey-drug-use-and-health.

Heier C, Xie H, Zimmermann R: Nonoxidative ethanol metabolism—from biomarkers to bioactive lipids. Int Union Biochem Mol Biol 2016; 68:916–23.

Javors MA, Hill-Kapturczak N, Roache JD, Karns-Wright TE, Dougherty DM. Characterization of the pharmacokinetics of phosphatidylethanol 16: 0/18: 1 and 16: 0/18: 2 in human whole blood after alcohol consumption in a clinical laboratory study. Alcohol Clin Exp Res. 2016 Jun;40(6):1228-34.

Jørgensen CH, Pedersen B, Tønnesen H. The efficacy of disulfiram for the treatment of alcohol use disorder. Alcohol Clin Exp Res. 2011 Oct;35(10):1749-58.

Koppaka V, Thompson DC, Chen Y, Ellermann M, Nicolaou KC, Juvonen RO, et al. Aldehyde dehydrogenase inhibitors: a comprehensive review of the pharmacology, mechanism of action, substrate specificity, and clinical application. Pharmacol Rev. 2012 Jul;64(3):520-39.

Knudsen HK, Abraham AJ, Roman PM. Adoption and implementation of medications in addiction treatment programs. J Addict Med. 2011 Mar;5(1):21-7.

Kummer N, Ingels AS, Wille SM, Hanak C, Verbanck P, Lambert WE, et al. Quantification of phosphatidylethanol 18:0/18:1, 18:1/18:1, 16:0/16:0 in venous blood and venous and capillary dried blood spots from patients in alcohol withdrawal and control volunteers. Anal Bioanal Chem 2016; 408: 825–38.

Litten RZ, Falk D, Ryan M, Fertig J. Research opportunities for medications to treat alcohol dependence: addressing stakeholders' needs. Alcohol Clin Exp Res. 2014 Jan;38(1):27-32.

Nishimura FT, Fukunaga T, Kajiura H, Umeno K, Takakura H, Ono T, Nishijo H. Effects of aldehyde dehydrogenase-2 genotype on cardiovascular and endocrine responses to alcohol in young Japanese subjects. Auton Neurosci. 2002;102:60-70.

Peng G-S, Chen Y-C, Tsao T-P, Wang M-F, Yin S-J. Pharmacokinetic and pharmacodynamic basis for partial protection against alcoholism in Asians, heterozygous for the variant *ALDH2*2* gene allele. Pharmacogenet Genomics. 2007;17(10):845-55.

Peng G-S, Wang M-F, Chen C-Y, Luu S-U, Chou H-C, Li T-K, Yin S-J. Involvement of acetaldehyde for full protection against alcoholism by homozygosity of the variant allele of mitochondrial aldehyde dehydrogenase gene in Asians. Pharmacogenetics. 1999;9:463-76.

Posner K, Brown GK, Stanley B, Brent DA, Yershova KV, Oquendo MA, et al. The Columbia-Suicide Severity Rating Scale: initial validity and internal consistency findings from three multisite studies with adolescents and adults. Am J Psychiatry. 2011; 168(12):1266-77.

Sheehan DV, Lecrubier Y, Sheehan KH, Amorim P, Janavs J, Weiller E, et al. The Mini-International Neuropsychiatric Interview (M.I.N.I.): the development and validation of a structured diagnostic psychiatric interview for DSN-IV and ICD-10. J Clin Psychiatry. 1998; 59[Suppl 20]:22-33.

Sobell LC, Sobell MB. Timeline follow-back: A technique for assessing self-reported alcohol consumption. In: Litten RZ, Allen JP, editors. Measuring alcohol consumption: psychosocial and biochemical methods. Washington DC: American Psychological Association.1992. pp. 41-72.

Substance Abuse and Mental Health Services Administration, Results from the 2013 National Survey on Drug Use and Health: Summary of National Findings, NSDUH Series H-48, HHS Publication No. (SMA) 14-4863. Rockville, MD: Substance Abuse and Mental Health Services Administration, 2014. Available from: https://www.samhsa.gov/data/sites/default/files/NSDUHresultsPDFWHTML2013/Web/NSDUHresults2013.pdf.

Ulwelling W, Smith K. The PEth blood test in the security environment: what it is; why it is important; and interpretative guidelines. J Forensic Sci. 2018 Nov;63(6):1634-40.

Viel G, Boscolo-Berto R, Cecchetto G, Fais P, Nalesso A, Ferrara S: Phosphatidylethanol in blood as a marker of chronic alcohol use: a systematic review and meta-analysis. Int J Mol Sci 2012; 13:14788–812.