PROTOCOL NUMBER:	AROHSD1001
STUDY TITLE:	A Phase 1/2a Single and Multiple Dose-Escalating Study to Evaluate the Safety, Tolerability, Pharmacokinetics and Pharmacodynamic Effects of ARO-HSD in Normal Healthy Volunteers as well as in Patients with NASH or Suspected NASH
STUDY TREATMENT (Active):	ARO-HSD
ROUTE:	Subcutaneous Injection
SPONSOR'S RESPONSIBLE MEDICAL MONITOR:	[redacted]
SPONSOR:	Arrowhead Pharmaceuticals, Inc. 177 East Colorado Blvd., Suite 700 Pasadena, CA 91105 Telephone: +1 626 304 3400 Facsimile: +1 626 304 3401
PROTOCOL VERSION	7.0
PROTOCOL DATE	12 March 2021

1. PROTOCOL SYNOPSIS

Study Title: A Phase 1/2a Single and Multiple Dose-Escalating Study to Evaluate the Safety, Tolerability, Pharmacokinetics and Pharmacodynamic Effects of ARO-HSD in Normal Healthy Volunteers as well as in Patients with NASH or Suspected NASH
Study Number: AROHSD1001
Phase: Phase 1/2a
Number of Sites: One or more sites in Australia, New Zealand, and Asia
Study Treatments: There will be two study treatments; one active (Test Formulation) and one placebo (Reference Formulation). Test Formulation: The test formulation is active ARO-HSD Injection (also referred to as ARO-HSD). The active pharmaceutical ingredient (API) contained in ARO-HSD is a synthetic, double-stranded, small interfering RNA (siRNA) duplex conjugated to an N-acetyl-galactosamine targeting ligand to facilitate hepatocyte delivery. Reference Formulation: The reference formulation is placebo (PBO): normal saline (0.9%) administered subcutaneously, volume matched to the corresponding ARO-HSD dose volume.
Study Objective and Endpoints: • The objective of the study is to assess the safety and tolerability of ARO-HSD in normal healthy volunteers (NHVs) and patients with NASH or suspected NASH.

Primary Endpoint:

- The incidence and frequency of adverse events (AEs) possibly or probably related to treatment after single doses in healthy volunteers and after multiple doses in patients with NASH or suspected NASH

Secondary Endpoints

- Pharmacokinetics of ARO-HSD

Exploratory Endpoints

(All Cohorts)

- Change in fasting LDL-C, Total Cholesterol, non-HDL-C, HDL-C, VLDL-C, and triglycerides compared to baseline

(Patient cohorts 1b, 3band 4b)

- Change in ALT and in AST as a marker of ARO-HSD activity compared to baseline
- Change in cytokeratin-18 compared to baseline
- Change in IL-1β, IL-6, IL-10, INF-gamma, and TNF-α compared to baseline
- Change in retinol and retinol binding protein compared to baseline
- Change in liver fat using MRI-PDFF compared to baseline
- Change in blood exosomal HSD17β13 mRNA levels (if feasible) compared to baseline
- Change in blood exosomal PNPLA3 mRNA levels (if feasible) compared to baseline
- Change in Enhanced Liver Fibrosis (ELF) Score at EOS compared to baseline
- Change in Pro-C3 at EOS compared to baseline
- Liver biopsy derived HSD17β13 mRNA expression compared to baseline
- Liver biopsy derived HSD17β13 protein expression compared to baseline in response to multiple doses of ARO-HSD (if scientifically feasible)
- Liver biopsy derived change in PNPLA3 mRNA expression compared to baseline
- Liver biopsy derived change in PNPLA3 protein expression compared to baseline in response to multiple doses of ARO-HSD (if scientifically feasible)

Study Population/Patient Number: This study will be conducted in adult males and females, aged 18 through 65 years (minimum of 19 years of age for NASH or suspected NASH patients) with BMI between 18.0 and 40.0 kg/m² enrolled and dosed as follows (see also Cohort Summary below):

NHV Cohorts 1, 2, 3, and 4: Each double-blind cohort will enroll eight (8) subjects (4 active: 4 PBO) with all cohorts planned to receive single escalating doses of ARO-HSD or placebo at dose levels of 25 mg (Cohort 1), 50 mg (Cohort 2), ≤ 100 mg (Cohort 3), and ≤ 200 mg (Cohort 4). These dose levels represent the highest dose that may be used in the cohort but after Cohort 1, the Data Safety Committee (DSC) may adjust the dose lower if indicated.

- Patient Cohorts 1b, 3band 4b: All patients will have suspected NASH and meet patient entry criteria. Cohorts are open label with up to 6 patients per cohort. All subjects will receive two doses of ARO-HSD at dose levels of 25 mg (Cohort 1b), ≤ 100 mg (Cohort 3b) and ≤ 200 mg (Cohort 4b). These dose levels represent the highest dose that may be used in the cohort but after Cohort 1, the DSC may adjust the dose lower if indicated.

  *Any NASH/Suspected NASH patient already in Screening when a patient cohort has reached capacity (defined as up to six patients who have completed D1 in Cohort 3b and up to six patients who have completed D1 in Cohort 4b and up to six patients who have completed D1 in Cohort 1b), may participate in the study up to an additional two patients per cohort.

Cohort Summary

Cohort	Population	Blinding	# Subjects	Dosing Schedule
1	NHVs	Double blind	8 (4 active: 4 placebo)	25 mg Day 1 only
1b	Patients with suspected NASH (biopsies on screen and day 71)	Open Label	up to 6 (all active)	25 mg Days 1, 29
2	NHVs	Double blind	8 (4 active: 4 placebo)	50 mg Day 1 only
3	NHVs	Double blind	8 (4 active: 4 placebo)	≤ 100 mg Day 1 only
4	NHVs	Double blind	8 (4 active: 4 placebo)	≤ 200 mg Day 1 only
3b	Patients with suspected NASH (biopsies on screen and day 71)	Open Label	up to 6 (all active)	≤100 mg Days 1, 29
4b	Patients with suspected NASH (biopsies on screen and day 71)	Open Label	up to 6 (all active)	≤200 mg Days 1, 29

Approximately 50 subjects may be enrolled in the study (not including replacements or optional expansion cohort).

Number of Doses per Treatment: Single dose (Cohorts 1, 2, 3, 4) or two doses (Cohorts 1b, 3b and 4b) dosed 28 days apart

Study Duration: For subjects in Cohorts 1 through 4, the duration of the study clinic visits is approximately 158 days from screening to the End-of-Study visit. For subjects in Cohorts 1b, 3b and 4b, the duration of the study clinic visits is approximately up to 173 days from screening to the End-of-Study visit. Additionally, all subjects will have a 90-day post-end of study visit follow-up phone call assessing compliance with contraception.

Study Confinement: For all NHV cohorts (1-4), clinical facility confinement will be approximately 3 days for dose administration (Day -1 through 24-hour assessments) with discharge on Day 2. Subjects will return to the clinical facility for outpatient visits per the Schedule of Assessments. Patients in Cohorts 1b, 3b and 4b will be at the facility for approximately 4 hours (2 hours before dosing and 2 hours after dosing) on dosing days.

Study Design/Methods:

Participants who have signed an EC/IRB-approved informed consent form and have met all the protocol eligibility criteria during screening may be enrolled into the study in a double-blind or open label fashion depending on the cohort. Cohorts 1 through 4 (NHVs) will begin with administration of ARO-HSD or PBO to two sentinel participants (one ARO-HSD, one PBO). Following the Day 3 evaluation in these participants, if there are no significant safety concerns, the remaining participants in the cohort will be treated at the discretion of the Principal Investigator (PI). Dosing of participants will be staggered by at least 30 minutes such that no two participants will be dosed simultaneously. Sentinel dosing is not required in NASH/suspected NASH patient cohorts.

Dose levels by cohort are outlined in the Cohort Summary above and Dose Escalation Schedule below. NHV Cohorts 1 through 4 will enroll sequentially with patient cohorts opening for enrollment in a step-wise manner. Screening for all cohorts may begin once Cohort 1 has been opened for enrollment.

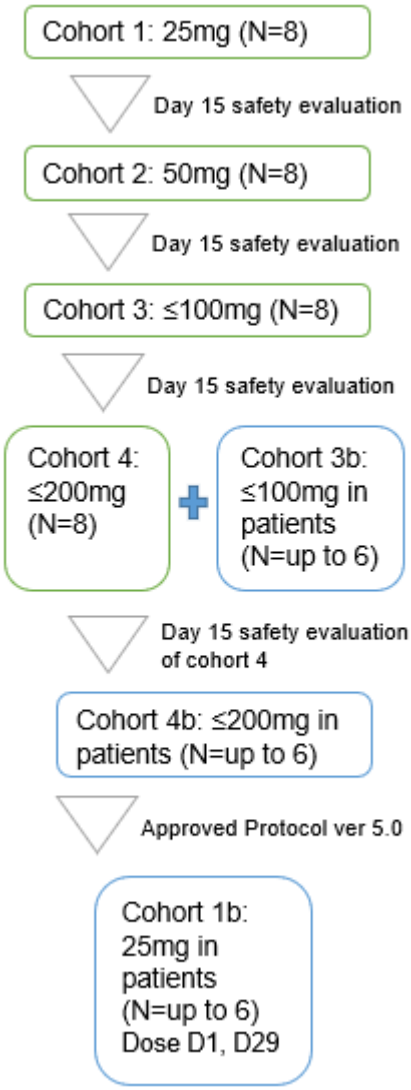
Dose escalation will require approval by the DSC based on all cumulative available safety data for prior cohorts, and through at least Day 15 of the current cohort as shown in Figure 1. Based on this cumulative safety data the DSC will vote to approve dose escalation to the next single dose NHV cohort as well as vote to open a NASH/suspected NASH patient cohort at the dose most recently tested in NHVs. Escalation to the next dose level will proceed until the highest planned dose level is completed, or the trial is halted prematurely by the PI, DSC, or Sponsor due to safety or other reasons. Where specified, dose levels represent the highest dose which may be used. The DSC may adjust doses downward if indicated based on review of available safety data and pharmacodynamic data (if available). All subjects who withdraw from the study prior to their End of Study (EOS) visit, for reasons other than an AE, may be replaced.

Cohort 1b (25 mg on Days 1 and 29) will enroll sequentially after Cohorts 3b and 4b are enrolled and will not require DSC approval to initiate enrollment as the dose is lower than the 200 mg top dose used in cohorts 4 and 4b which have already been opened by the DSC.

In double-blind cohorts, treatment un-blinding may occur, at the PI's discretion, only where deemed necessary for treatment of an AE or for a decision to be made regarding trial continuation. After all subjects in double-blinded cohorts have completed their EOS visit (not including the 90-day follow-up call), all blinded cohorts may be unblinded.

Single and multiple doses of ARO-HSD will be evaluated in a sequential manner as shown in the Dose Escalation Schedule.

Dose Escalation Schedule



AE monitoring

Safety assessments will include: AEs/SAEs, physical examinations, vital sign measurements (blood pressure, heart rate, temperature, and respiratory rate), ECGs, clinical laboratory tests, concomitant medications/therapy, and reasons for treatment discontinuation. Safety assessments will be performed at specified time points and prior to study completion.

The AE/SAE reporting period for an enrolled participant begins when the participant provides informed consent. Treatment-emergent AEs/SAEs are defined as those following study drug administration or a pre-existing condition exacerbated by study drug. All AEs that occur during the AE reporting period specified in the protocol must be reported to Arrowhead via electronic case report forms within approximately 48 hours. All SAEs that occur during the reporting period, in addition to reporting via electronic case report forms, must also be reported to Arrowhead via the SAE report form within 24 hours of awareness. All AEs/SAEs will be followed until resolution, until the condition stabilizes, until the event is otherwise explained, or until the participant is lost to follow-up. If the PI learns of any SAE, including a death, at any time after a patient has been discharged from the study, and he/she considers the event reasonably related to the investigational product, the PI will promptly notify the Sponsor. Laboratory abnormalities will

be reported as AEs if considered clinically significant by the PI. Laboratory abnormalities not reported as AEs are not to be reported as clinically significant (CS) in the study database.

Treatment Stopping Rules:
Escalation to the next cohort will proceed according to the study design until the highest planned dose cohort is completed, unless the trial is stopped early by the DSC, PI or Sponsor. A decision to stop the trial early or discontinue drug in an individual subject, group of subjects or to halt enrollment temporarily or permanently **may** be indicated based on any of the following:

1. A serious adverse reaction (i.e., a serious AE [SAE] considered at least possibly related to the IMP administration) in ONE subject; or severe non-serious adverse reactions (i.e. severe non-serious AEs considered as, at least, possibly related to the IMP administration) in TWO subjects in the same cohort, independent of whether or not the events occur within the same system-organ-class.

2. One of the following abnormal results at least possibly related to ARO-HSD:

- In NHV cohorts:
  - Treatment emergent AST and/or ALT > 8X ULN which must be confirmed by repeat blood draw within 48 hours of initial results; OR
  - A treatment emergent AST or ALT >3X ULN (must be confirmed on repeat) with a total bilirubin >2X ULN (must be confirmed on repeat); OR
  - A treatment emergent ALT or AST >3X ULN (must be confirmed on repeat) with symptoms of RUQ pain, fever, rash or eosinophilia; OR
  - A treatment emergent AST or ALT >3X ULN (must be confirmed on repeat) with an INR > 1.5 (must be confirmed on repeat) and which must not be explained by another plausible cause
- See Appendix 16.1 for treatment modification guidelines in subjects/patients (NASH/Suspected NASH patients) with baseline/pre-dose elevated ALT or AST.

If such serious adverse reactions or severe non-serious adverse reactions occur, enrollment and dosing in healthy volunteers will be paused pending a review of safety data and a decision on how to proceed by the DSC.

Sponsor or PI can discontinue any subject at any time with or without DSC consultation. If such events (as described in #1, #2 above) occur and the subject is not discontinued from the study, the reason for not discontinuing the subject will be included in DSC meeting minutes. Including, but not limited to the events listed above, the DSC **may** pause the study to additional dosing or dose escalation to provide time to evaluate safety data and recommend the action to be taken, which may include, but is not limited to, one of the following:

- Discontinuation of a subject or group of subjects from the study
- The study is stopped immediately with no further dosing
- The study will continue until the current cohort is completed
- The study will continue, but the next dose escalation will be to a level midway between the current level and the next level specified in the Dose Escalation Schedule above
- The study will continue as planned

Study Assessments:

Safety Assessments:

Safety assessments will be performed at specified time points per the Schedule of Assessments and will include the following:

- Vital signs: Resting heart rate, semi-supine systolic/diastolic blood pressure, respiratory rate, oxygen saturation and temperature

- Clinical laboratory measurements (e.g., chemistry including lipase and lipids, hemoglobin A1C, hematology, coagulation, and urinalysis)
- Resting ECG measurements (measured after participant is semi-supine for at least 3 $minutes$)
- At each visit, participants will be asked about concomitant medications/therapy and will be instructed to volunteer any information regarding AEs and SAEs that they may have experienced. Any known untoward event that occurs beyond the AE reporting period that the PI considers an SAE and possibly related to study treatment will be reported to Arrowhead.
- 90-day post-end of study visit pregnancy follow-up phone call assessing compliance with contraception.

Pharmacodynamic assessments:

Pharmacodynamic assessments will be performed at specified time points per the Schedule of Assessments and will include fasting (at least 8 hours) LDL-C, total cholesterol, non-HDL-C, HDL-C, VLDL-C, triglycerides, retinol, retinol binding protein, cytokines, cytokeratin-18, blood exosomal HSD17β13 and PNPLA3 mRNA (if feasible), liver biopsy tissue PNPLA3 mRNA, liver biopsy tissue HSD17β13 mRNA, liver biopsy tissue PNPLA3 and HSD17β13 protein levels (if feasible), hemoglobin A1C, serum glucose, PRO-C3, ELF, MRI-PDFF and Fibroscan. All serum tests will be completed after an 8-hour fast unless as otherwise specified. Results, percent change, and duration of response (when applicable) from baseline to 4 weeks (or longer as necessary) will be analyzed and summarized by dose cohort and treatment group. For lipid related and serum pharmacodynamic assessments, baseline is defined as the pre-dose value obtained nearest to the first dose.

Immunogenicity:

Samples will be collected for anti-drug antibody testing for cohorts (NASH patients only) at pre-dose, day 57, and EOS, or at Early Termination as per Schedule of Assessments.

Pharmacokinetics & Urine/Plasma Drug Metabolites:

Blood and urine samples will be collected for pharmacokinetic and drug metabolite analysis per the Schedule of Assessments.

Table 1 Schedule of Assessments for Cohorts 1-4 Single Dose: NHV

Assessment	Screening (Day -45 to Day -1)	Pre-dose	Day 1									24 hr post-dose	48 hr	Day 8	Day 15	Day 29	Day 43	Day 57, 71	Day 85, 99	End of Study (or Early Term) Day 113	90 days after EOS
			0 min	15 min	30 min	1 hr	2 hr	4 hr	8 hr	12 hr	18 hr										
Visit Window		-2hrs	±2 minutes PK only, ±10 minutes other procedures									±1 hr		±2d	±2d	±2d	±2d	±2d	±2d	±2d	±7d
Informed Consent	X																				
Eligibility Criteria	X																				
Demographics/Medical Hx	X																				
Physical Exam (See Section 10.3.2)	X	X										as needed		as needed	as needed	as needed	as needed	as needed	as needed	X	
BMI	X	X																		X	
Concomitant Meds/Therapies	X	X										X	X	X	X	X	X	X	X	X	
Electrocardiogram (See Section 10.3.3)	X	X				X	X					X	X			X				X	
Urine Pregnancy Test (WOCP) **confirmed negative pre-dose**	X	X																		X	
FSH (post-menopause)	X																				
Urine Drug Screen	X																				
Vitals (BP, temp, HR, RR, O_2)	X	X			X	X	X	X	X			X	X	X	X	X	X	X	X	X	
HIV, Hep B, C	X																				
Labs (heme, coag, lipase, chem, lipids)	X	X										X	X	X	X	X	X	X	X	X	
Hemoglobin A1c	X	X																		X	
Blood PK including met ID		X		X	X	X	X	X	X	X	X	X	X	X	X	X					
Urine PK including met ID		spot	Day 1 0-4 hr, 4-12 hr, 12-24 hr urine collections										spot	spot	spot	spot					
Urinalysis	X	X										X	X	X	X	X	X	X	X	X	
Administration of ARO-HSD			X																		
Adverse Events Collection	X	→																		X	
Pregnancy phone call																					X

Table 2 Schedule of Assessments for Cohorts 1b, 3b, 4b: Subjects with suspected NASH (biopsies on screen and day 71)

Assessment	Screening (Day -60 to Day -1)	Pre-dose	Day 1				24 hr post-dose	Day 15	Day 29	Day 57	Day 71	Day 85	End of Study (or Early Term) Day 113	90 days after EOS
			0 min	30 min	1 hr	2 hr								
Visit Window		-2hrs	±2min (PK only), ±10min				±1hr	±2d	±2d	±2d	±5d	±2d	±5d	±7d
Informed Consent	X													
Eligibility Criteria	X													
Demographics/Medical Hx	X													
Physical Exam (See Section 10.3.2)	X	X					as needed	as needed	as needed	as needed	as needed	as needed	X	
BMI (Height and Body Weight)	X	X[1]							X[1]		X[1]		X[1]	
Concomitant Meds/Therapies	X	X					X	X	X	X	X	X	X	
Electrocardiogram (See Section 10.3.3)	X	X			X	X	X		(0, 1, 2hr)[2]				X	
Urine Pregnancy Test (WOCP) confirmed negative pre-dose/pre-bx	X	X							pre-dose		pre-bx		X	
FSH (post-menopause)	X													
Urine Drug Screen	X													
Vitals (BP, temp, HR, RR, O_2)	X	X		X	X	X	X	X	X	X	X	X	X	
HIV, Hep B, C	X													
Labs (heme, coag, chem, lipase, lipids)	X	X					X	X	X	X	X	X	X	
Retinol, RBP, blood for exosomes, cytokine panel		X						X	X	X	X	X	X	
Hemoglobin A1c	X	X											X	
Pro-C3, ELF, Cytokeratin-18		X											X	
Blood PK		X		X	X	X	X	X	pre-dose					
Genotyping (HSD17β13, PNPLA3)	X													
Urinalysis	X	X					X	X	X	X	X	X	X	
Liver Biopsy, FibroScan, MRI-PDFF	X[3]										X			
Administration of ARO-HSD			X						X					

[1] Body weight only
[2] 10-minute window allowed
[3] Liver biopsy can be done at D1, as long it is completed prior to any D1 procedures

Anti-Drug Antibodies		X								X			X	
Adverse Events Collection	X	→	→	→	→	→	→	→	→	→	→	→	X	
Pregnancy phone call														X

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3. STUDY INFORMATION AND SIGNATURES

Investigator's Statement:

I have read and understood the information in this protocol and agree to conduct the trial according to the protocol (subject to any amendments) and in accordance with the principles of ICH E6 Good Clinical Practice guidelines. I have read and agree to comply with the Investigator obligations stated in this protocol, as well as with any and all applicable federal, state, or local laws and regulations. Any changes in procedure will only be made if necessary, to protect the safety, rights or welfare of participants.

I agree to conduct or to supervise the trial in person.

I agree to ensure that all that assist me in the conduct of the study are aware of their obligations.

Principal Investigator:

_____ _____

Signature Date

_____

Printed Name

4. LIST OF ABBREVIATIONS AND TERMS

AE	Adverse event
ALT	Alanine aminotransferase
API	Active Pharmaceutical Ingredient
AST	Aspartate transaminase
ARO	Arrowhead Pharmaceuticals, Inc.
AUC	Area under the curve
AUC_{inf}	Area under the curve from time 0 to infinity
BP	Blood pressure
cGCP	current Good Clinical Practice
cGMP	current Good Manufacturing Practice
C_{max}	Concentration maximum (peak)
CRA	Clinical Research Associate
CRF	Case Report Form
CRO	Contract Research Organization
CS	Clinically significant
CTN	Clinical Trial Notification
CVA	Cerebrovascular accident
dL	deciliter
DSC	Data Safety Committee
EC	Ethics Committee
ECG	Electrocardiogram
eCRF	Electronic Case Report Form
ELF	Enhanced Liver Fibrosis (Score)
EOS	End of Study
FDA	Food and Drug Administration
FSH	Follicle-Stimulating Hormone
GCP	Good Clinical Practice
GLP	Good Laboratory Practice
GMP	Good Manufacturing Practice
GTT	Glucose Tolerance Test
HBV	Hepatitis B virus
HIV	Human Immunodeficiency Virus
HR	Heart Rate
HSD	Hydroxysteroid dehydrogenase
ICH	International Council for Harmonisation
IRB	Institutional Review Board
Kg	Kilogram
L	Liter
LISR	Local injection site reactions
LOF	Loss of function
MCH	Mean Cell Hemoglobin
MCHC	Mean Cell Hemoglobin Concentration
MCV	Mean Cell Volume

MedDRA	Medical Dictionary for Regulatory Activities
Mg	milligram
mmHg	millimeters of mercury
MRI	Magnetic resonance imaging
NAFLD	Non-alcoholic fatty liver disease
NAS	NAFLD Activity Score
NASH	Non-alcoholic steatohepatitis
NHV	Normal healthy volunteers
NOAEL	No observed adverse event level
NYHA	New York Heart Association
OTC	Over the Counter
PBO	Placebo
PD	Pharmacodynamic
PI	Principal Investigator
PK	Pharmacokinetic
PT	Prothrombin Time or Preferred Term
PTT	Partial thromboplastin time
QRS	QRS duration (complex) – a structure on the ECG that corresponds to the depolarization of the ventricles
QT	QT interval – a measure of the time between the start of the Q wave and the end of the T wave in the heart's electrical cycle
RBP	Retinol Binding Protein
RNA	Ribonucleic acid
RNAi	RNA interference
SAE	Serious Adverse Event
SD	Standard Deviation
siRNA	Short interfering RNA oligonucleotides
SOA	Schedule of Assessments
SOC	System Organ Class
SubI	Subinvestigator
T2DM	Type 2 diabetes mellitus
$t_{1/2}$	terminal elimination half-life
TE	Treatment Emergent
TIA	Transient Ischemic Attack
ULN	Upper Limit of Normal
WOCP	Women of Childbearing Potential

5. INTRODUCTION

5.1 Background Information & Therapeutic Rationale

Nonalcoholic steatohepatitis (NASH) describes a subgroup of non-alcoholic fatty liver disease (NAFLD) in which hepatic cell injury and inflammation has developed over background steatosis. The primary risk factor is obesity. Although lifestyle modifications are reasonable therapeutic interventions, there is poor long-term adherence to weight maintenance, healthful food choices, and physical activity. The rising prevalence of NASH presents a significant health burden in many developed countries.

NAFLD is the most common chronic liver disease with a worldwide prevalence of 20-30%. NAFLD is strongly associated with metabolic syndrome, Type 2 diabetes mellitus (T2DM), and NASH. Up to 30% of those with NAFLD will go on to develop NASH which progresses to liver cirrhosis at a rate of approximately one fibrosis stage (F0-4) per seven years (Vernon et al., 2011, Ekstedt et al., 2017).

HSD17β13 is a hydroxysteroid dehydrogenase involved in the metabolism of hormones, fatty acids and bile acids. In humans, it is extensively expressed in the liver localizing to the surface of lipid droplets and acting as a retinol dehydrogenase protein. Hepatic HSD17β13 expression appears limited to hepatocytes, with no detectable expression in quiescent or activated stellate cells (Ma et al., 2018). Human genetic studies indicate that loss-of-function (LOF) mutations in HSD17β13 are protective against development of both alcohol-related and nonalcohol-related liver disease with approximately 30-50% risk reduction compared to non-carriers. Such variants result in a truncated protein that does not localize to lipid droplets and is less abundant in the liver than in non-carriers. Carriers of this variant in general show lower ALTs and ASTs compared to non-carriers. This protective effect has inspired therapeutic interest as the protective effect seen in individuals harboring loss-of-function mutations may indicate a role for HSD gene silencing in the treatment of liver disease.

HSD17β13 expression is markedly upregulated in mice and in humans with NAFLD. Specifically, the protein is associated with lipid droplets with significant up-regulation in NAFLD patient liver biopsies compared to liver biopsies from non-NAFLD patients (Su et al., 2014). Additionally, the HSD17β13 LOF variant gene is associated with low levels of PNPLA3 mRNA expression and appears to be associated with reduced risk of liver disease in individuals carrying the I148M PNPLA3 variant (Abul-Husn, et al., 2018).

5.2 Mechanism of Action of ARO-HSD

ARO-HSD, referred to as ADS-006, is an RNA interference (RNAi) based targeted therapeutic composed of a synthetic double-stranded RNAi trigger designed to target HSD17β13 for RNAi-mediated gene silencing. RNAi is a naturally occurring catalytic gene silencing mechanism that is highly specific and efficient (Fire et al. 1998; Sharp 2001). The dsRNA is conjugated to galactose containing ligands that are preferentially taken up by hepatocytes. Once internalized, ARO-HSD engages the cell's RNAi machinery to target HSD mRNA for degradation, thus reducing HSD protein synthesis.

5.3 ARO-HSD Pre-Clinical Pharmacology Studies

Further information on the pre-clinical pharmacology studies in monkeys and in mouse models is provided in the Investigator's Brochure.

5.4 ARO-HSD Pre-Clinical Pharmacokinetic and Product Metabolism Studies

PK parameters for ARO-HSD have been evaluated in both rats and monkeys. Results of these studies can be found in the Investigator's Brochure.

5.5 ARO-HSD Pre-Clinical Toxicology Studies

ARO-HSD has been clinically well tolerated in rats and in non-human primate toxicology studies. Details regarding GLP and non-GLP toxicology results are provided in the Investigator's Brochure.

5.6 Rationale for the Study

AROHSD1001 is a first in human dose-ranging study to assess the safety and tolerability of ARO-HSD in healthy volunteers and then in patients with NASH.

Treatment with ARO-HSD is expected to reduce hepatic production of the HSD17β13 protein via RNAi. The magnitude of the reduction and duration of effect will depend on the dose. Since there has not been human clinical exposure to ARO-HSD, an effective therapeutic dose to administer to patients with NASH is unknown, although in the RNAi field, non-human primate potency and duration of activity are usually similar to that seen in humans.

The study begins with single dose administration to healthy volunteers to evaluate ARO-HSD for the purpose of understanding safety profile. There are no known serum biomarkers of HSD17B13, thus no pharmacodynamic data is expected from the NHV cohorts.

NASH/suspected NASH Patients will require administration of multiple doses of ARO-HSD to sustain hepatic silencing of HSD production. Accordingly, this study uses a multiple-ascending dose (MAD) design to determine the dose required to reach and sustain maximal knockdown in HSD17β13 levels, and the dose-response relationship in patients with suspected NASH (Cohorts 1b, 3b and 4b). These patients, having clinical characteristics consistent with NASH including elevated ALT and hepatic steatosis, would often require a liver biopsy to establish a definitive diagnosis of NASH. While biopsy may be part of the standard evaluations of these patients with suspected NASH, in this study, prudent limits make it a requirement only in NASH/suspected NASH Cohorts 1b, 3b and 4b. In the patient cohorts, dose response based on gene target mRNA and protein expression changes will be measured along with imaging and plasma markers of disease. It is critical to emphasize that as there is no known or established biomarker of HSD17B13 that liver biopsy is the only way to evaluate gene target silencing as a marker of pharmacodynamic effect. Thus, liver biopsy is critical to obtaining the data from this study needed to progress into Phase 2. The cohorts employing liver biopsy plan to enroll patients most likely to require liver biopsy as part of their standard clinical evaluation for NASH.

The addition for cohorts 1b and 3c in protocol version 5.0 aimed to assess gene target silencing at lower doses (Cohort 1b, ie, ARO-HSD 25 mg on Days 1 and 29) and allow evaluation of gene

target silencing with less frequent dosing (Cohort 3c). However, Cohort 3c (ARO-HSD 100 mg on Days 1 and 85) is removed from protocol version 7.0 as the enrolled NHV cohorts as well as the three patient cohorts (3b, 4b and 1b) will provide sufficient preliminary data informing on safety, tolerability, pharmacodynamics, pharmacokinetics as well as dose response of multiple dose ARO-HSD necessary for future Phase 2 study planning.

5.7 Risk Assessment for Participants

- **Embryo-Fetal:** Limited GLP toxicology studies have been conducted. Accordingly, eligible participants enrolled in this study, both male and female (including partners), must agree to use two highly effective forms of contraception during the study and for 3 months post-dose, or agree to abstinence (acceptable only if this method is in alignment with the normal life style of the patient).

- **Hepatic:** ARO-HSD targets the liver. siRNA literature has described ALT changes associated with off-target effects of the siRNA seed region on microRNAs in the hepatocyte (Janas, et al. 2018). The siRNA sequence of the ARO-HSD sense and antisense molecules have been screened for potential mRNA and microRNA homology and sequences with homology were excluded from consideration. Thus, no such off-target effects are anticipated. However, to mitigate this risk, the proposed study protocol has built in stopping rules for ALT and AST elevation. Blood samples will be drawn frequently to evaluate liver injury and liver function. The Data Safety Committee (DSC) will review all available safety data including laboratory data prior to dose escalation. Additionally, the planned starting dose of 25 mg is approximately 1/252nd (assuming weight-based conversion and a 70kg subject) of the No-Observed-Adverse-Effect-Level (NOAEL) of 90 mg/kg based on the rat GLP study and 1/840th of the 300 mg/kg monkey NOAEL. The highest planned dose in this study of 200 mg is 1/31st of the 90 mg/kg rat NOAEL and 1/140th of the monkey NOAEL. There is a wide margin of safety between planned clinical doses and animal toxicology study NOAELs.

- **Injection Site Reactions:** Other subcutaneously administered modified siRNA drug candidates evaluated in clinical studies have been associated with mild to moderate injection site reactions (e.g. pain, erythema). This study includes a protocol for evaluation and grading of injection site reactions based on predefined criteria for mild, moderate and severe. Injection site reactions will be photographed for tracking resolution and/or progression. Additionally, steps will be taken to minimize injection site reactions such as rotating injection sites and allowing the ARO-HSD solution to come to room temperature prior to injecting.

- **Liver Biopsy:** Performance of liver biopsies is commonly part of the standard of care in patients with NASH as well as other adult liver diseases. While it is a common procedure, like any procedure it is associated with some risk. The risk of bleeding associated with biopsies requiring blood transfusion or hospitalization is 0.04% or less and the risk of less severe but clinically significant events (causing pain, tachycardia or lower blood pressure) is estimated at 0.2% (Rockey et al., 2009). Several larger studies show that complications and serious bleeding related to liver biopsy are overwhelmingly more likely in patients with serious clotting disorders, malignancy, and other serious

health conditions that are described in the exclusion criteria for this study. Measures of hemostatic function (e.g. platelets, INR) will be conducted prior to biopsy, and may be used to exclude any participant at increased risk for procedural complications. Therefore, the risk of liver biopsy related complications in this study is acceptable and similar to other studies using liver assessments as an endpoint. Monitoring and recovery of the patient following the procedure will be consistent with locally accepted standards of clinical practice.

5.8 Justification for Starting Dose in Humans

Reported pharmacokinetic properties for several oligonucleotide subclasses across species, including humans (Henry et al., 2001; Geary et al., 2003; Yu et al., 2001), indicate that the most appropriate method for extrapolating animal doses to human equivalent doses is the comparison of dose per unit body weight (mg/kg), rather than dose per surface area (mg/m^2) or plasma exposure (AUC or C_{max}). The human equivalent dose for oligonucleotide therapeutics can be extrapolated directly from monkeys to humans with a scaling factor of 1.0 on mg/kg dose administrations (Yu et al., 2015). Arrowhead has historically used 1:1 mg/kg scaling factor based on monkey NOAELs to determine starting dose in other siRNA first-in-human studies (ARO-HBV, ARO-ANG3, ARO-APOC3, and ARO-AAT first in human studies).

Extrapolated based on body weight, the proposed starting dose of 25 mg will be administered first to healthy volunteers in Cohort 1. The planned starting dose of 25 mg is approximately $1/252^{nd}$ (assuming weight-based conversion and a 70kg subject) of the No-Observed-Adverse-Effect-Level (NOAEL) of 90 mg/kg based on the rat GLP study and $1/840^{th}$ of the monkey NOAEL of 300 mg/kg. The highest planned dose in this study of 200 mg is $1/31^{st}$ of the 90 mg/kg rat NOAEL and $1/140^{th}$ of the monkey NOAEL. As ADS-006 is not cross reactive in rats or mice, but is fully cross reactive in cynomolgus and rhesus monkeys, monkeys are considered the pharmacologically relevant species. Regardless, there is a wide margin of safety between planned clinical doses and animal toxicology study NOAELs. The safety margins for each of the proposed ARO-HSD clinical doses (relative to rat or monkey NOAELs) is provided in Table 3 .

Table 3 Safety Margins of the Proposed ARO-HSD Clinical Doses (Assuming a 70 kg Subject)

ARO-HSD dose	Compared to rat NOAEL (90mg/kg)	Compared to monkey NOAEL (300mg/kg)
25 mg	1/252	1/840
50 mg	1/126	1/420
100 mg	1/63	1/210
200 mg	1/31.5	1/140

6. STUDY OBJECTIVES AND ENDPOINTS

The objective of the study is to assess the safety and tolerability of ARO-HSD in healthy normal volunteers and patients with NASH or suspected NASH.

6.1 Primary Endpoint

- The incidence and frequency of adverse events (AEs) possibly or probably related to treatment after single doses in healthy volunteers and after multiple doses in patients with NASH or suspected NASH.

6.2 Secondary Endpoint

- Pharmacokinetics of ARO-HSD

6.3 Exploratory Endpoints

All Cohorts

- Change in fasting LDL-C, total cholesterol, non-HDL-C, HDL-C, VLDL-C, and triglycerides compared to baseline

For Patient Cohorts 1b, 3b and 4b

- Change in ALT and in AST as a marker of ARO-HSD activity compared to baseline
- Change in cytokeratin-18 compared to baseline
- Change IL-1B, IL-6, IL-10, INF-gamma, and TNF-alpha compared to baseline
- Change in retinol and retinol binding protein compared to baseline
- Change in liver fat using MRI-PDFF compared to baseline
- Change in blood exosomal HSD17β13 mRNA levels (if feasible) compared to baseline
- Change in blood exosomal PNPLA3 mRNA levels (if feasible) compared to baseline
- Change in Enhanced Liver Fibrosis (ELF) Score at EOS compared to baseline
- Change in Pro-C3 at EOS compared to baseline
- Liver biopsy derived HSD17β13 mRNA expression compared to baseline in response to multiple doses of ARO-HSD as a measure of drug activity in patients with NASH
- Liver biopsy derived HSD17β13 protein expression compared to baseline in response to multiple doses of ARO-HSD as a measure of drug activity (if scientifically feasible)
- Liver biopsy derived change in PNPLA3 mRNA expression compared to baseline
- Liver biopsy derived change in PNPLA3 protein expression compared to baseline in response to multiples doses of ARO-HSD (if scientifically feasible)

7. STUDY PLAN

7.1 Study Design

Participants who have signed an EC/IRB approved informed consent form and have met all the protocol eligibility criteria during screening may be enrolled into the study in a double-blind or open label fashion depending on the cohort. Cohorts 1 through 4 will begin with administration

of ARO-HSD or PBO to two sentinel participants (one ARO-HSD, one PBO). Following the Day 3 evaluation in these participants, if there are no significant safety concerns, the remaining participants in the cohort will be treated at the discretion of the Principal Investigator (PI). Dosing of participants will be staggered by at least 30 minutes such that no two participants will be dosed simultaneously. Sentinel dosing is not required in patient cohorts.

Dose levels by cohort are outlined in Table 4 and Figure 1. Cohorts 1 through 4 will enroll sequentially with patient cohorts opening for enrollment in a step-wise manner. Screening for all Cohorts may begin once Cohort 1 has been opened for enrollment.

Clinical facility confinement will be approximately 3 days for first dose administration in NHV cohorts (Cohorts 1, 2, 3, and 4) with discharge on Day 2. Subjects will return to the clinical facility for outpatient visits per the Schedule of Assessments. Subjects in cohorts 1b, 3b and 4b will be at the facility for approximately 4 hours (2 hours before dosing and 2 hours after dosing) on dosing days.

Table 4 Cohort Summary

Cohort	Population	Blinding	# Subjects	Dosing Schedule
1	NHVs	Double blind	8 (4 active: 4 placebo)	25 mg Day 1 only
1b	Patients with suspected NASH (biopsies on screen and day 71)	Open Label	up to 6 (all active)	25 mg Days 1, 29
2	NHVs	Double blind	8 (4 active: 4 placebo)	50 mg Day 1 only
3	NHVs	Double blind	8 (4 active: 4 placebo)	≤ 100 mg Day 1 only
4	NHVs	Double blind	8 (4 active: 4 placebo)	≤ 200 mg Day 1 only
3b	Patients with suspected NASH (biopsies on screen and day 71)	Open Label	up to 6 (all active)	≤ 100 mg Days 1, 29
4b	Patients with suspected NASH (biopsies on screen and day 71)	Open Label	up to 6 (all active)	≤ 200 mg Days 1, 29

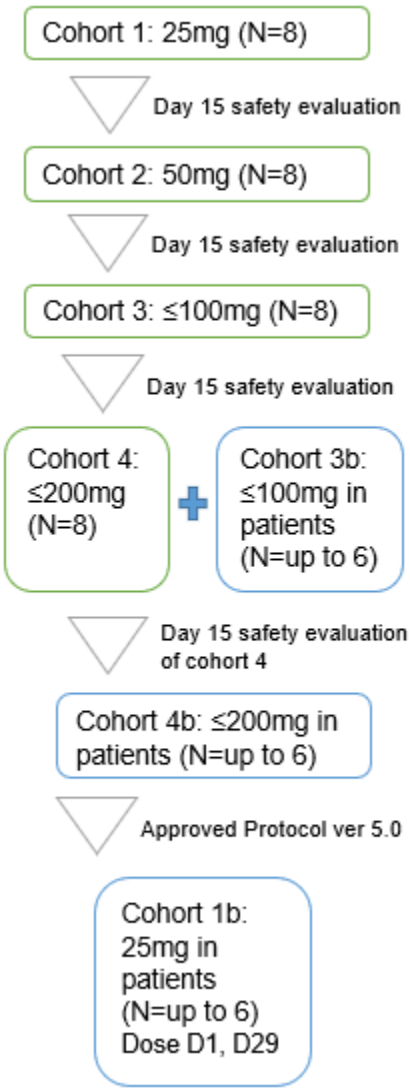
In the NHV and suspected NASH patient dose escalation part of the study, dose escalation will require approval by the DSC based on all cumulative available safety data for prior cohorts, and through at least Day 15 of the current cohort. Based on available safety data through Day 15, the DSC will vote to approve dose escalation to the next single dose NHV cohort as well as vote to open a suspected NASH patient cohort at the dose most recently tested in NHVs. DSC decisions will be based on all aggregate safety data available including all data available at least through Day 15 of the current cohort as shown in Figure 1. Escalation to the next dose level will proceed until the highest planned dose level is completed, or the trial is halted prematurely by the PI, DSC, or Sponsor due to safety or other reasons. All subjects who withdraw from the study prior to their EOS visit, for reasons other than an adverse event, may be replaced.

Cohort 1b will enroll after completion of cohorts 3b and 4b and will not require DSC approval to initiate enrollment as they use doses lower than the 200 mg top dose used in cohorts 4 and 4b which have already been opened by the DSC.

In double-blind cohorts, treatment un-blinding may occur at the PI's discretion only where deemed necessary for treatment of an AE or for a decision to be made regarding trial continuation. After all subjects in a cohort have completed the final planned study visit (not including the 90-day follow-up call), Sponsor may be unblinded at Sponsor's request. After all subjects in double blinded cohorts have completed their EOS visit (not including the 90-day follow-up call), all blinded cohorts may be unblinded.

Single and multiple doses of ARO-HSD will be evaluated in in a sequential manner as shown in Figure 1.

Figure 1: Dose Escalation Schedule



7.2 Rationale for Study Design

This first-in-human study plans to investigate ARO-HSD in normal healthy volunteers to evaluate the drug's safety and tolerability. The study initiates in healthy volunteers because the risk is considered low based on GLP animal toxicity testing. It is expected that treatment of NASH with ARO-HSD will require multiple doses; thus, this study in healthy volunteers transitions from single dose to a multi-dose study in patients with NASH/suspected NASH if safety data is acceptable to the DSC.

Cohorts 1 through 4 in the study are double-blind to limit the occurrence of conscious and unconscious bias in trial conduct and interpretation. Blinding will be achieved using a PBO (0.9% normal saline). Inclusion of participants receiving PBO will reduce bias in the assessment of drug safety and tolerability. Cohorts 1, 2, 3, and 4 are randomized with a 1:1 (active:PBO) ratio to reduce bias. Cohorts 1b, 3b and 4b are open-label and include patients with NASH or suspected NASH. The patient cohorts are proposed to understand multiple dose pharmacodynamics and pharmacokinetics which should assist with dose selection in later stage clinical studies. Liver biopsies in patient cohorts at screen and after dosing are required to measure the effect of ARO-HSD on gene expression as there currently is no other method to detect change or otherwise detect pharmacodynamic drug activity. To minimize suspected NASH patient and NHV exposure, the number of NHV and patient cohorts have been initially set at 32 NHV subjects and approximately up to 18 suspected NASH patients.

7.3 DSC and Criteria for Dose-escalation and Dose Limiting Toxicities

Escalation to the next cohort will proceed according to the study design until the highest planned dose cohort is completed, unless the trial is stopped early by the DSC, PI or Sponsor. Dose escalation will require approval by the DSC based on evaluation of all available safety and, when available, pharmacodynamic data through at least Day 15 of the most advanced NHV cohort.

The DSC will be composed of sponsor medical monitor and Head of Pharmacovigilance, the study Primary Investigator and an Independent physician medical monitor.

7.4 Stopping Rules

A decision to stop the trial early or discontinue drug in an individual subject, group of subjects or to halt enrollment temporarily or permanently may be indicated based on any of the following:

1. A serious adverse reaction (i.e. an SAE considered at least possibly related to the IMP administration) in ONE subject; or severe non-serious adverse reactions (i.e. severe non-SAEs considered as, at least, possibly related to the IMP administration) in TWO subjects in the same cohort, independent of whether or not the events occur within the same system-organ-class.

2. One of the following abnormal results at least possibly related to ARO-HSD:
   - In NHV cohorts:
     - Treatment emergent AST and/or ALT > 8X ULN which must be confirmed by repeat blood draw within 48 hours of initial results, OR

- A treatment emergent AST or ALT >3X ULN (must be confirmed on repeat) with a total bilirubin >2X ULN (must be confirmed on repeat), OR
- A treatment emergent ALT or AST >3X ULN (must be confirmed on repeat) with symptoms of RUQ pain, fever, rash or eosinophilia, OR
- A treatment emergent AST or ALT >3X ULN (must be confirmed on repeat) with an INR > 1.5 (must be confirmed on repeat) and which must not be explained by another plausible cause
- See Appendix 16.1 for treatment modification guidelines in patients with baseline/pre-dose elevated ALT or AST.

If such serious adverse reactions or severe non-serious adverse reactions occur as outlined above, enrollment and dosing in healthy volunteers will be paused pending a review of safety data and a decision on how to proceed by the DSC.

The Sponsor or PI can discontinue any patient at any time with or without DSC consultation. If such events (as described above) occur and the patient is not discontinued from the study, the reason for not discontinuing the patient will be included in DSC meeting minutes. Including, but not limited to the events listed above, the DSC **may** pause the study to additional dosing or dose escalation to provide time to evaluate safety data and recommend the action to be taken, which may include, but is not limited to, one of the following:

1. Discontinuation of a patient or group of patients from the study.
2. The study is stopped immediately with no further dosing
3. The study will continue until the current cohort is completed.
4. The study will continue, but the next dose escalation will be to a level midway between the current level and the next level specified in Table 4.
5. The study will continue as planned.

7.5 Duration of the Study

For subjects in Cohorts 1-4, the duration of the study clinic visits is approximately 158 days from Screening to the EOS visit. For patients in Cohorts 1b, 3b and 4b, the duration of the study clinic visits is approximately 173 days from Screening to the EOS visit. This does not include the 90-day post-end of study visit follow-up phone call assessing compliance with contraception.

8. PATIENT SELECTION

8.1 Number of Patients

Approximately 50 participants may be enrolled in the study (not including replacements or expansion cohort).

8.2 Inclusion Criteria

To be eligible for enrollment, the participating subject must meet all the following inclusion criteria (applicable to all cohorts):

1. Able and willing to provide written informed consent prior to the performance of any study specific procedures

2. Participants who are willing and able to comply with all study assessments and adhere to the protocol schedule

3. A 12-lead ECG at Screening that, in the opinion of the Investigator, has no abnormalities that compromise participant's safety in this study

4. Participants using highly effective contraception during the study and for 12 weeks following the last dose of ARO-HSD. Males must not donate sperm for at least 12 weeks post last dose of study treatment. Females of childbearing potential must have a negative urine pregnancy test at Screening and pre-dose on dosing days. Females not of childbearing potential must be post-menopausal (defined as cessation of regular menstrual periods for at least 12 months), confirmed by follicle-stimulating hormone (FSH) consistent with post-menopausal state based on lab reference ranges.

    - Using twice the normal protection of birth control by using a condom AND one other form of either birth control pills (The Pill), depot or injectable birth control, IUD (Intrauterine Device), birth Control Patch (e.g., Ortho Evra), NuvaRing®, OR Surgical sterilization as a single form of birth control: i.e., tubal ligation, hysterectomy, bilateral oophorectomy, vasectomy or equivalently effective surgical form of birth control

    - True subject abstinence for the duration of the study and 12 weeks after the dose of ARO-HSD is acceptable only when in line with the preferred and usual lifestyle of the subject. Periodic abstinence (calendar, symptothermal, post-ovulation methods), withdrawal (coitus interruptus), spermicides only, and lactational amenorrhoea methods are not considered "true" abstinence and are not acceptable methods of contraception

5. No abnormal finding of clinical relevance at the Screening evaluation that in the opinion of the Investigator could adversely impact subject safety during the study or adversely impact study results

6. No consistent change in diet or exercise in the month prior to screening or plans to change diet or exercise habits during the study.

7. No plans to lose weight during study

Additional Inclusion Criteria <u>for NHV Cohorts 1 through 4</u>

8. Males or females 18 to 55 years of age at Screening

9. BMI < 35 kg/m² at Screening

10. Normal ALT and AST at Screening

11. No history of or clinical suspicion of diabetes

12. No history of or clinical suspicion of liver disease

Additional Inclusion Criteria <u>for patient Cohorts 1b, 3b and 4b</u>

13. Males or females 19 to 65 years of age at Screening

14. BMI ≤ 40 kg/m² at Screening with no plans to lose weight, increase exercise or otherwise change diet during the study

15. Patients agree to have one liver biopsy during the screening period and one at the end of the treatment period for assessment of the treatment effects

16. Suspected NASH based on the presence of Screening MRI-PDFF liver fat > 8% AND ALT > ULN [for purposes of this inclusion criterion, ULN is 30 U/L for men and 19 U/L for women (Prati et al., 2002)]. Alternatively, biopsy confirmed NASH within 1 year of screen visit is acceptable for inclusion.

* 1 repeat diagnostic test is acceptable within the screening period when determining eligibility.

8.3 Exclusion Criteria

A potential subject will be excluded from the study if *any* of the following criteria apply (applicable to all cohorts):

1. Pregnant or lactating females

2. Human immunodeficiency virus infection, as shown by the presence of anti-HIV antibody (sero-positive)

3. Seropositive for HBV (HBsAg positive at Screening) or HCV (detectable HCV RNA at Screening). Cured HCV (positive antibody test without detectable HCV RNA is acceptable if HCV RNA has been negative for at least 2 years).

4. Uncontrolled hypertension (Systolic BP > 170 and diastolic BP > 100 mmHg at Screening). Patients may rescreen once BP is successfully controlled.

5. A history of torsades de pointes, ventricular rhythm disturbances (e.g., ventricular tachycardia or fibrillation), heart block (excluding first-degree block, being PR interval prolongation only), congenital long QT syndrome or new ST segment elevation or depression or new Q-wave on ECG. Participants with a history of atrial arrhythmias should be discussed with the Medical Monitor

6. Symptomatic heart failure (per NYHA guidelines), unstable angina, myocardial infarction, severe cardiovascular disease (ejection fraction < 20%. Transient ischemic attack (TIA) or cerebrovascular accident (CVA) within 6 months prior to Screening

7. History of malignancy except for adequately treated basal cell carcinoma, squamous cell skin cancer, superficial bladder tumors, or in situ cervical cancer. Participants with other curatively treated malignancies who have no evidence of metastatic disease and > 1-year disease-free interval may be entered following approval by the Medical Monitor

8. History of major surgery within 1 month prior to Screening

9. Excessive use of alcohol within three months prior to the Screening visit (i.e., > 14 units per week or > 4 units on a given day for men and > 7 units per week or > 3 units on a given day for women [1 unit = 150 mL of wine, 360 mL of beer, or 45 mL of 40% alcohol])

10. Use of illicit drugs (such as cocaine or phencyclidine [PCP]) within 1 year prior to the Screening visit or positive urine drug screen at Screening (a urine drug screen positive for benzodiazepines, amphetamines, opioids or cannabis is acceptable for enrollment if the positive test is due to a substance used for medical reasons)

11. Use of an investigational agent or device within 30 days prior to dosing or current participation in an investigational study involving a therapeutic intervention

12. Blood donation (≥ 500 mL) within 7 days prior to study treatment administration

13. Any concomitant medical or psychiatric condition or social situation that would make it difficult to comply with protocol requirements or put the participant at additional safety risk

Additional Exclusion Criteria <u>for patient Cohorts 1b, 3b and 4b</u>

14. Weight loss of more than 5% within 3 months prior to randomization

15. Cirrhosis [based on screening biopsy or historical biopsy showing definitive cirrhosis or based on Screening FibroScan > 20 kPa (one retest allowed) or if using an alternate ultrasound elastography method a value indicative of cirrhosis]

16. Other well documented causes of chronic liver disease based on medical history (e.g., Wilson's disease, alpha-1 antitrypsin deficiency, hemochromatosis, autoimmune hepatitis)

17. Homozygous HSD17βB13 rs72613567 or other loss of function variants at Screening

18. Values at Screening representing any of ALT>250 U/L; INR > 1.5, creatinine > 2.0 mg/dL (176.84 umol/L); platelets < 100,000

19. Any exclusion to undergoing liver biopsy including concomitant use of anticoagulants, antiplatelet medications, medical history of abnormal bleeding. Patients may be eligible if contraindication anticoagulant/antiplatelet medications have been discontinued for 5 half-lives prior to liver biopsy. NSAIDs must have been discontinued at least 3 days before biopsy and aspirin at least 5 days before biopsy.

* 1 repeat diagnostic test is acceptable within the screening period when determining eligibility.

8.4 Participant Withdrawal Criteria

Participants will be advised that they are free to withdraw from the study at any time for any reason or, if necessary, the PI, or medically trained designee, may withdraw a participant from the study, per the following criteria, to protect the participant's health:

- the need to take medication which may interfere with study measurements
- intolerable/unacceptable AEs
- major violation of or deviation from study protocol procedures
- non-compliance of participant with protocol
- participant unwilling to proceed and/or consent is withdrawn
- withdrawal of the participant from the study if, in the PI's judgement, it is in the participant's best interest
- confirmed pregnancy (Note: within 24 hours of awareness of the pregnancy the investigator must notify the Sponsor [see Section 11].)

The reasons for withdrawal will be recorded on the case report form (CRF) and included in the final clinical study report, along with any AEs and any necessary medical treatment.

If a participant is withdrawn from the study due to significant AE or SAE, the PI, or medically trained designee, will evaluate the urgency of the event. If the situation warrants, the PI, or medically trained designee, will take appropriate diagnostic and therapeutic measures. If the situation is not an immediate emergency, the PI, or medically trained designee, at the clinical study facility will attempt to contact the Arrowhead Pharmaceuticals, Inc. Medical Monitor or medically qualified designee for consultation. No medical help, diagnosis, or advice will be withheld from the participant due to an inability to contact the Medical Monitor. The participant will be encouraged to remain available for follow-up medical monitoring. The Sponsor will be notified as soon as possible of any participant withdrawals.

Participants who are withdrawn or discontinue prior to EOS visit for reasons other than an adverse event, may be replaced at Sponsor discretion.

8.5 Restrictions and Concomitant Medications

- ***Confinement:*** For all NHV cohorts, clinical facility confinement will be approximately 3 days for first dose administration (Day -1 through 24-hour assessments) with discharge on Day 2. Subjects will return to the clinical facility for outpatient visits per the Schedule

of Assessments. Patients in Cohorts 1b, 3b and 4b will be at the facility for approximately 4 hours (2 hours before dosing and 2 hours after dosing) on dosing days.

- ***Fasting:*** On the day of dosing or on other days with blood draws, participants will have fasted from food for at least 8 hours prior to study treatment administration or blood draw unless otherwise specified or as otherwise required by study procedures.

- ***Recreational Drugs & Alcohol:*** Participants will be instructed to abstain from consuming alcohol for at least 48 hours prior to admission, and while confined to the clinical facility. In addition, participants will be instructed to refrain from regular use of alcohol (i.e., >14 units per week or >4 units on a given day for men and >7 units per week or >3 units on a given day for women [1 Unit = 150 mL of wine, 360 mL of beer, or 45 mL of 40% alcohol]) for the study duration. Participants must abstain from use of recreational drugs throughout the study.

- ***Concomitant Medications:*** Use of other concomitant medications may be approved by sponsor medical monitor and PI. Patients will be instructed to inform the PI of the details (indication, dose and dates of administration) if they do take any medication, and these details will be recorded in the CRF. Medication wash-out to achieve study eligibility at screen constitutes a protocol procedure and will be preceded by informed consent.

9. INVESTIGATIONAL PRODUCT

9.1 Description, Identification and Dosage

Arrowhead Pharmaceuticals, Inc. is responsible for the supply of active drug supplies together with detailed instructions (in a pharmacy manual) describing preparation of ARO-HSD. The PBO (normal saline 0.9%) will be supplied by the clinical site.

Accordingly, ARO-HSD will be supplied as single sterile 2-mL vials containing ARO-HSD, with the correct dose of ARO-HSD prepared by the Pharmacy prior to dosing participants.

The PBO will be 0.9% normal saline.

Doses Administered per Dose Level:

Each single dose of either active drug (ARO-HSD) or PBO (normal saline 0.9%), will be administered by subcutaneous injection. Injections will be made into the subcutaneous tissue at an appropriate site (e.g. abdomen, thigh, upper arm, etc.) using a 25-30 Gauge, ½ inch needle. The abdomen is the preferred injection site. The injection site is to be varied (no multiple injections into the same exact site. Alternating various locations on the abdomen is acceptable). Injection site location is to be recorded in the eCRF. Prior to dose administration, the ARO-HSD vial must be allowed sufficient time to come to room temperature. Do not inject into areas of active skin disease or injury such as sunburns, skin rashes, inflammation or skin infections. Injection volume per site should not exceed approximately 1.0 mL.

There will be no dose escalation within a cohort (i.e., the same drug dose will be administered to each participant within a cohort). The randomization schedule for NHVs will be provided to each clinical site and will be maintained along with any other materials that could jeopardize the blind in a secured area of the pharmacy.

Supply, Preparation, Storage, and Labelling of ARO-HSD

ARO-HSD will be supplied as a sterile Type-1 glass 2.0-mL vial (1.2 mL nominal volume, 1.0 mL withdrawable volume).

Strength:	200 mg/mL
Volume:	1.0 mL
Appearance:	Clear, colorless to yellow solution
Inactive ingredients:	0.5 mM sodium phosphate monobasic, 0.5 mM sodium phosphate dibasic in water for injection
Shipment and storage:	Refrigerated, 2-8 ºC

ARO-HSD will be prepared, per the Pharmacy Manual, by a pharmacist or qualified pharmacy staff member at the clinical sites. Aseptic technique will be used to ensure sterility of the solution to be injected. The time of preparation for active drug must be documented and tracked to demonstrate administration within prepared drug stability boundaries. Please refer to the Pharmacy Manual for more detailed instructions.

The investigational product vials will be labelled per Good Manufacturing Practice (cGMP)/Good Clinical Practice (cGCP).

Study drug supplies will be stored at clinical sites securely under the appropriate conditions as noted in the Pharmacy Manual.

9.2 Study Drug Handling

The Sponsor will provide the PI with a sufficient quantity of clinical drug supplies. The PI must ensure that deliveries of investigational product from the Sponsor are correctly received by a responsible person, that all receipts of drug shipments are recorded on the appropriate Drug Accountability forms prepared by the pharmacy at the clinical site and that the products are

stored in a secure area under recommended storage conditions. It is also the responsibility of the PI to ensure that the integrity of packaged study product not be jeopardized prior to dispensing.

Only participants enrolled in the study may receive study drug, in accordance with all applicable regulatory requirements. Only authorized site staff may supply or administer study drug. The study drug must be stored in a secure area with access limited to the PI and authorized staff and under the physical conditions that are consistent with the study drug-specific requirements.

An authorized and trained staff member at each clinical trial site will dispense the study drug per predefined drug dispensing requirements. The dispensing will be verified by a second member of site staff.

ARO-HSD will be supplied by Arrowhead Pharmaceuticals, Inc. and labelled with the drug name, batch number, expiration date (as applicable), and storage conditions. Individual doses will be dispensed by clinical trial site staff members on the morning of dosing and recorded in the drug accountability records. A Pharmacy Manual will be prepared to define the procedures for dispensing.

Procedures delineated in the Pharmacy Manual will be followed for the receipt, handling and accountability of the study formulations.

9.3 Accountability of Study Supplies

All study related material supplied is only for use in this clinical trial and should not be used for any other purpose. The PI is responsible for the investigational product accountability, reconciliation and record maintenance at the investigational site. In accordance with all applicable regulatory requirements, the PI or designated site staff must maintain investigational product accountability records throughout the course of the study. This person will document the amount of investigational product received from Arrowhead Pharmaceuticals, Inc. and the amount administered to participants. A non-blinded Clinical Research Associate (CRA) will perform initial and ongoing study drug kit and placebo accountability. The non-blinded CRA will protect the integrity of the assignment blind and will not participate in data review for study participants. Used vials of ARO-HSD will be retained sequestered per participant and cohort (where allowable by local policy) and made available to the non-blinded CRA during study drug and placebo reconciliation.

A Drug Dispensing Log must be kept current and will contain the following information:

- the identification of the participant to whom the drug was dispensed
- the date(s) and quantity of the drug dispensed to the participant.

The date and time of dose preparation and release will be maintained to support administration of study drug. The pharmacy will dispense the study medication and the clinical site staff will administer the study medication only to participants included in this study following the procedures set out in the study protocol. Study drug administration will be documented on the CRFs and/or other study drug accountability record. The inventory must be available for inspection by the CRA during the study. Drug supplies, excluding partially used or empty

containers, will either be collected at the end of the study by the CRA or returned by the PI or designee to Arrowhead Pharmaceuticals Inc. or the designated Arrowhead approved depot.

9.4 Retention of Investigational Product Vials

For this study, used and partially used drug vials will be retained for an adequate period to allow accountability by the CRA. No additional study drug samples will be retained.

9.5 Allocation to Treatment

All potential participants who sign an informed consent at Screening will receive a unique 6-digit number (i.e. a Screening Number). The first 3 digits will represent the assigned site number and will be the same for each participant that screens at an individual site. The next 3 digits will be assigned sequentially (starting with 001). For patients who are deemed eligible, this 6-digit screening number will become the patient's permanent study ID number.

Eligible subjects in double-blind cohorts will be allocated a unique randomization number, in accordance with the randomization schedule. In each of NHV Cohorts 1-4, the first two subjects (sentinels) will be randomized separately to one active and one PBO. Each subject will be assigned to either active (ARO-HSD) or PBO treatment. The allocation of active treatment or PBO will be performed using a block randomization algorithm.

Participants who drop out prior to their EOS visit for reasons other than an adverse event, may be replaced.

9.6 Blinding and Code-Break

For blinded cohorts, blinding of study drug/PBO assignment is critical to the integrity of this clinical trial. It is expected that in most cases, AEs can be properly managed without the need for unblinding. However, in the event of a medical emergency in which knowledge of an individual participant's assignment is considered critical to the participant's well-being and management, the PI or documented designated treating physician may request permission to unblind the treatment assignment from the Arrowhead Pharmaceuticals, Inc. Medical Monitor. If the situation is not an immediate emergency, the PI should contact the responsible Medical Monitor to discuss the participant and circumstances requiring the unblinding. The blind will be broken only for the specific participant under discussion. Unblinding in situations that are not an immediate emergency may only take place with the notification and agreement of the responsible Medical Monitor. The randomization schedules will be maintained under controlled access. The personnel involved in the dispensing of investigational products will be accountable for ensuring compliance to randomization schedules. The non-blinded CRA will review the randomization schedule in comparison to the dispensing log to verify correct randomization.

If the PI considers an adverse event to be of such severity as to require immediate specific knowledge of the identity and dose of the relevant product, the treatment assignment can be determined by opening the code break envelope after discussion with the Medical Monitor and in the event of an absolute emergency. Only the Pharmacist and the PI will each have copies of the code break envelopes. The study monitor should also be informed promptly.

If a participant requires emergent unblinding (with or without a discussion between the Investigator and the Medical Monitor preceding the unblinding), the Investigator may also be required to complete a 'Drug Safety Unblinding Request/Notification Form' to document the medical rationale necessitating the unblinding. This form is then forwarded to the local Medical Monitor.

After the completion of the final study visit (not including 90-day follow-up phone call assessing compliance with contraception) for each cohort, unblinding for Sponsor analysis will occur at Sponsor discretion. After all subjects in double blinded cohorts have completed their EOS visit, all blinded cohorts may be unblinded.

Sponsor may request an interim descriptive analysis of the change from baseline in measured parameters any time after all NHV subjects planned for enrollment in each cohort have received at least one dose of ARO-HSD or PBO. This interim analysis is for planning of future studies and will not impact the conduct of this study. Sponsor will remain blinded to all subject treatment assignments. Descriptive statistics (that does not inadvertently unblind the trial) for change from baseline in pharmacodynamic measures will be calculated for all active subjects per cohort and for a pooled PBO group by an unblinded statistician and provided to Sponsor. For any AEs occurring more than once, the frequency of AEs for a specific preferred term will be calculated for pooled active and pooled PBO groups in such a way not to inadvertently break the subject-blind of the trial.

10. STUDY METHODS AND SCHEDULES

10.1 Overview of Procedures

Participants, who have consented to participate, had the screening examination and have met all of the protocol eligibility criteria will be randomized at a ratio of 4:4 (active:PBO) to receive a single subcutaneous injections of either PBO or ARO-HSD in double-blind fashion (for NHV Cohorts 1 through 4), or may receive open label ARO-HSD (Patient Cohorts 1b,3b and 4b). NHV Cohorts 1-4 will begin with administration of ARO-HSD or PBO to two sentinel participants (one ARO-HSD, one PBO). Following the Day 3 evaluation in these participants, if there are no significant safety concerns based on PI discretion, the remaining participants in the cohort may be treated.

During the study (according to the Schedule of Assessments listed in Tables 1-2), participants will undergo the following evaluations at regular intervals: medical history review, physical examinations, vital sign measurements (blood pressure, temperature, heart rate, and oxygen saturation, respiratory rate), weight measurement, AE monitoring, ECGs, urine pregnancy test (females of childbearing potential), and concomitant medication review. Blood samples will be collected for clinical laboratory analysis, and urine will be collected for urinalysis. Blood and urine samples will also be collected for pharmacokinetic and immunogenicity analysis. Patients in Cohorts 1b, 3b and 4b will have liver biopsies.

Participant visits to the clinical facility will occur as per the Schedule of Assessments. A telephone follow-up will occur 90 days after EOS to assess the patient's compliance with contraception requirements. Clinically significant changes including AEs will be followed until resolution is

achieved or considered medically stable. Refer to Schedule of Assessments (Tables 1-2) for additional information.

The PI (or medically qualified designee) will be required to remain within the clinical study facility for 2 hours after dosing on Day 1 and will remain on call for the duration of the study. Participants should refrain from strenuous physical activities throughout the study.

10.2 Selection and Screening

Prior to commencement of any screening procedures, the PI, or designee, will inform the participant about the nature and purpose of the study, including the risks and benefits involved, possible AEs, the fact that their participation is voluntary and provide a copy of the EC/IRB - approved Informed Consent Form for review. Each participant will acknowledge receipt of this information by giving written informed consent for their involvement in the study in the presence of the PI, or designee, who will also sign and date the Informed Consent Form. Time of consent will be recorded in the site's source documents and reflected in the eCRFs. The original signed consent form will be retained by the PI and a copy of the original will be given to the participant. Informed consent will be performed per the Principles of the International Conference on Harmonisation (ICH) Good Clinical Practice (cGCP) procedures.

Having given Informed Consent, potential participants will undergo procedures outlined in the Schedule of Assessments, to be performed within 60 days of the scheduled dosing date, to determine that they meet the inclusion/exclusion criteria specified in Sections 8.2 and 8.3.

10.3 On-Study Procedures/Assessments

10.3.1 Demographics/Medical History

Medical History will include previously diagnosed medical conditions, medication use over the previous 30 days, including vitamins, over-the-counter medications, prescription drugs, recreational drugs or supplements and alcohol and tobacco use.

10.3.2 Physical Exam

A complete physical exam will be performed at Screening. Areas examined include:

- Constitutional
- Skin
- HEENT
- Heart
- Lungs
- Back
- Abdomen
- Extremities
- Mental Status
- Neuro (gait/reflexes)
- Additional examination may be performed at investigator discretion

An abbreviated physical exam will be performed at pre-dose and EOS. Areas examined include:

- Constitutional
- Heart
- Lungs
- Abdomen
- Extremities
- Brief Neuro
- Additional examination may be performed at investigator discretion

At all other time points outlined in the Schedule of Assessments, a symptom-directed physical exam will be performed as needed. Height (during screening, in centimeters, without shoes) and weight (kilograms, without shoes) should be recorded to determine BMI where specified in SOA.

10.3.3 Electrocardiogram

A single 12-lead ECG measurement will be obtained at time points outlined in the Schedule of Assessments after the participant is semi-supine for at least 3 minutes. On dosing days ECGs will be performed pre-dose and 1 and 2 hours post-dose and may be performed more frequently if indicated. ECGs should be performed within a window of 10 minutes, prior to other invasive procedures such as blood draws. Any abnormal ECGs will be repeated in triplicate, with each measurement approximately 1 minute apart. Only abnormal ECG readings that are considered clinically significant in the opinion of the investigator need to be repeated in triplicate. Triplicate ECGs are three tracings approximately 1 minute apart. If the originally scheduled ECG is considered clinically significant, both the first tracing and the triplicate tracings will be entered into the EDC.

10.3.4 Vital Sign Assessments

Systolic/diastolic blood pressure, temperature, heart rate, respiratory rate (breaths/min) and oxygen levels will be obtained at time points outlined in the Schedule of Assessments after the participant is semi-supine for at least 3 minutes. Vitals signs will be obtained prior to venipuncture and other invasive procedures.

10.3.5 Clinical Laboratory Tests

Blood and urine samples will be collected to perform clinical laboratory tests. Participants will be required to fast for the screening and other pharmacodynamic sample collections.

Central labs will be utilized. However, local labs may be utilized as necessary if needed by the investigator or for emergent situations.

Within the screen period, up to 60 days prior to the first dose of study medication, a blood and urine sample will be collected for the laboratory tests detailed below to establish baseline data and eligibility for enrolment. One repeat Screening lab draw is allowed per assessment to

establish eligibility. The results will be assessed by the PI, or medically qualified designee, before study enrollment. Any abnormality in laboratory values (that are confirmed on repeat) deemed clinically significant by the PI, or medically qualified designee (i.e., those that would jeopardize the safety of the participant or impact on the validity of the study results), will result in exclusion of that participant. Clinical laboratory tests will be performed on participants' blood and urine at specified time-points listed in the Schedule of Assessments.

- ***Biochemistry*:** Sodium, potassium, chloride, bicarbonate, glucose, urea, creatinine (including calculated creatinine clearance), creatine kinase, phosphate, total calcium, albumin, total protein, total bilirubin, lipase and amylase, conjugated bilirubin, alkaline phosphatase (ALP), alanine aminotransferase (ALT), and aspartate transaminase (AST), hemoglobin A1C

- ***Hematology:*** Hemoglobin, red blood cell count (RBC), hematocrit, mean cell volume (MCV), mean cell hemoglobin (MCH), mean cell hemoglobin concentration (MCHC), platelets, white cell count, neutrophils, lymphocytes, monocytes, eosinophils and basophils

- ***Coagulation:*** Partial thromboplastin time (PTT), Prothrombin time (PT) with INR

- ***Urinalysis:*** Leucocytes, nitrites, urobilinogen, protein, pH, blood, specific gravity, ketone, bilirubin and glucose

- ***Urine drug screen:*** urine will be analyzed for benzodiazepines, amphetamines, methamphetamines, methadone, phencyclidine, barbiturates, opiates, cocaine, ecstasy and cannabinoids

- ***Microscopic urinalysis will be performed if indicated:*** white blood cells, red blood cells, epithelial cells, bacteria

- ***Serology:*** Hepatitis B surface antigen, Hepatitis C antibody and HIV antibody screen. If necessary, participants will be counseled by the PI, or medically trained designee, concerning the blood tests for Hepatitis B surface antigen, Hepatitis C and HIV antibodies, and their subsequent results.

- ***FSH:*** Post-menopausal status will be confirmed by follicle-stimulating hormone (FSH) level consistent with post-menopausal state.

- ***Pregnancy:*** Females of childbearing potential will have a urine pregnancy test at Screening and pre-dose on dosing days and biopsy days. If urine pregnancy test is positive, dosing or biopsy should not occur, and the patient will be referred to their primary care provider for follow up.

- ***Lipid Parameters:*** LDL-C, total cholesterol, non-HDL-C, HDL-C, VLDL-C, and triglycerides

- ***Retinoid pathway markers:*** retinol, retinol binding protein

- ***NASH biomarkers***: ELF [Hyaluronic acid (HA), Procollagen III amino terminal peptide (PIIINP), Tissue inhibitor of metalloproteinase 1 (TIMP-1)], cytokeratin-18, PRO-C3, IL-1β, IL-6, IL-10, INF-gamma, and TNF-α

- ***Immunogenicity:*** Specified subjects will be assessed for anti-drug antibodies.

- ***Genotype:*** All patient cohorts will be assessed for HSD17βB13 and PNPLA3 gene variants with the subject's written consent.

- ***Exosomal analysis:*** blood will be drawn for exosomal analysis of *PNPLA3* and *HSD17B13* if technically feasible.

The Day 1 (pre-dose) value will be used as each participant's baseline value for data analysis purposes or as otherwise specified. If Day 1 or as otherwise specified values are erroneous or not available and repeat blood draw is not possible, the pre-dose value closest to Day 1 (e.g., Screening) may be used as baseline.

10.3.6 Hepatic Imaging (only for patient Cohorts 1b, 3b and 4b)

Patients must fast from food for at least 4 hours prior to MRI.

Magnetic Resonance Imaging: Liver fat content via MRI-PDFF will be evaluated and quantified per the Schedule of Assessments. MRI reading for eligibility and post-dose will be completed centrally.

FibroScan®: Per the Schedule of Assessments, pre-dose and post-dose liver stiffness will be evaluated using FibroScan® standard procedures. No centralized reading is required for FibroScan. An equivalent ultrasound elastography method may be used.

Please see separate imaging manual for specific instructions related to hepatic MRI.

10.3.7 Liver Biopsy (only for Cohorts 1b, 3b and 4b)

With training and Sponsor approval, only those sites capable of processing biopsy tissue in accordance with the Laboratory and Biopsy Manuals will be required to obtain samples for protein expression analysis at the time points outlined in the Schedule of Assessments.

Samples will be used to assess effect of treatment on:

- HSD17B13 mRNA and PNPLA3 mRNA.
- Protein expression for HSD17B13 and PNPLA3 (if scientifically feasible).

The liver biopsy procedure and D1 dosing can be done on the same date. The only requirement is for the liver biopsy procedure to be completed before any other D1 procedures, especially dosing.

If the patient agrees to an additional biopsy core during Screening to confirm NASH diagnosis, the histopathology can be conducted as an option by the sites and histopathology costs will be covered by the Sponsor. This optional procedure should not be used to confirm patient eligibility.

Please see separate laboratory and biopsy manuals for specific instructions related to biopsy performance.

10.3.8 Pharmacokinetics

Samples (blood and urine) for analysis of circulating ARO-HSD will be obtained at time points following study drug administration as outlined in the Schedule of Assessments. ARO-HSD metabolites in blood and urine will be identified in samples taken at timepoints per the schedule of assessments in NHV cohorts.

Please see separate laboratory manual for specific instructions related to PK and met ID processing.

10.3.8.1 Blood Sample Collection, Processing, and Analysis

Blood samples from participants will be collected at time points outlined in the Schedule of Assessments through an indwelling cannula or through a fresh vein puncture. The actual blood collection time will be recorded in the source documents. All deviations outside the range allowed above will be documented as protocol deviations. In all such cases, appropriate time corrections, for the actual time of sample collection will be incorporated at the time of data analysis.

The actual sample times will be recorded in the eCRF and will be entered at the time of or as soon as possible after sampling. All times must be recorded in the 24-hour format. An explanation must be given for any blood sample taken outside of the set sampling times.

Whole blood will be collected and processed per the Laboratory Manual.

10.3.8.2 Urine Sample Collection, Processing and Analysis

Urine samples from participants will be collected at time points outlined in the Schedule of Assessments. For spot urine collection, the actual urine collection time and volume will be recorded. For interval sample collections, the start and stop time of urine sample collection (along with volume) will be recorded with an attempt to void at the end of the collection interval. All times must be recorded in the 24-hour format. An explanation must be given for any sample taken outside of the set sampling times.

Urine will be collected and processed per the Laboratory Manual.

10.3.9 Concomitant Medications/Therapies

Participants will be instructed to inform the PI of the details (indication, dose and dates of administration) if they do take any medication, and these details will be recorded in the CRF. If necessary, paracetamol may be used during the study as necessary.

10.3.10 Follow-Up Procedures: Pregnancy Follow-Up Telephone Call (90 days (± 7 days) post end of study)

Document telephone contact with each participant to verify compliance with contraceptive measures and absence of any known pregnancy. Information regarding any reported pregnancy should be collected for at least 1 year after birth or longer if it is decided that addition follow-up is required or until the end of the pregnancy.

10.3.11 Early Termination Procedures

The reason for Early Termination will be documented in source documents and eCRF. Procedures as outlined in the Schedule of Assessments will be completed.

10.4 Allocation of Treatment

Participants will receive study drug by assigned cohort (Table 4) as outlined in Section 7.1. Treatments will be administered per the randomized sequence (where applicable) kept by the pharmacy or in a secure place at the clinical site, under control of the un-blinded staff member.

Table 5 Injection Number and Volume per Cohort

Cohort	Dose	Concentration	Total Injection Volume
1, 1b	25 mg	200 mg/mL	0.125 mL
2	50 mg	200 mg/mL	0.25 mL
3, 3b	100 mg	200 mg/mL	0.5 mL
4, 4b	200 mg	200 mg/mL	1 mL

10.5 Study Treatment Administration

Appropriately trained employees of the clinical site will administer the study treatment. Each dose will be administered as a single subcutaneous injection. The date, time and location of administration will be recorded in the source notes and witnessed by a second person from the clinical facility. The site of injection will be marked and mapped for later observation. The preferred site of injection is the abdomen. Optional additional sites are the upper arms and thighs.

10.6 Timing of Treatments and Procedures

Actual times of procedures for each participant will vary depending on scheduling and will be recorded in the CRF.

In the event of multiple procedures scheduled at the same time, non-invasive procedures (i.e., ECGs, AE assessment) will be conducted prior to invasive procedures (i.e., blood sample collection). Timing of activities may be adjusted slightly to accommodate all procedures.

10.7 Safety Assessments

The safety of ARO-HSD will be evaluated by collection of the following measurements performed at time points as specified in the Schedule of Assessments
Tables 1-2):

- Monitoring of AEs/SAEs
- Physical examinations
- Height and weight
- Vital signs including oxygen level
- ECG measurements
- Local Injection Site Reactions
- Clinical laboratory tests (hematology, chemistry, coagulation, urinalysis)
- Concomitant medications/therapy
- Reasons for treatment discontinuation due to toxicity

- Pregnancy test for WOCP

The AE/SAE reporting period for an enrolled participant will begin when the participant provides informed consent. Treatment-Emergent AEs/SAEs will be those defined as following dose administration. All AEs/SAEs that occur during the AE reporting period specified in the protocol must be reported to Arrowhead Pharmaceuticals, Inc., regardless of the relationship of the AE to study treatment. Any known untoward event that occurs beyond the AE reporting period that the PI considers an SAE and possibly related to study treatment will be reported to Arrowhead.

11. ADVERSE EVENTS

The PI and clinical facility staff are responsible for detection, recording and reporting of events that meet the criteria and definition of various AEs as listed below. Adverse events will be recorded from time of signed consent through to EOS; only AEs that occur post-dose will be considered treatment-emergent. The PI and clinical facility staff are responsible for detection, recording and reporting of pregnancy and appropriate follow up. Information regarding any reported pregnancy should be collected for at least 1 year after birth or longer if it is decided that addition follow-up is required or until the end of the pregnancy.

11.1 Definitions

An **Adverse Event (AE)** is any untoward medical occurrence in a patient or clinical investigation patient administered a pharmaceutical product and which does not necessarily have to have a causal relationship with this treatment. An AE can therefore be any unfavorable and unintended sign (including an abnormal laboratory finding or diagnostic test), symptom, or disease temporally associated with the use of a medicinal (investigational/experimental) product,

whether related to this product or not. (Refer to ICH E2a: Clinical Safety Data Management: Definitions and Standards for Expedited Reporting, 27 October 1994).

Treatment-emergent AEs will be defined as AEs with onset after administration of the study drug, or when a pre-existing medical condition increases in severity or frequency after study drug administration.

AEs will not include:

- A medical or surgical procedure such as surgery, endoscopy, tooth extraction, or transfusion (although the condition that leads to the procedure may be an AE)
- A pre-existing disease or condition present at the start of the study that does not worsen during the study
- Any situation where an untoward medical occurrence has not occurred (for example, hospitalizations for cosmetic elective surgery or "social" admissions)
- An overdose of either the investigational product or a concurrent medication without any resulting signs or symptoms

A **Serious Adverse Event (SAE)** is an AE that:

- Results in death
- Is life-threatening (NOTE: The term 'life-threatening' in the definition of 'serious' refers to an event/reaction in which the participant was at immediate risk of death at the time of the event/reaction; it does not refer to an event/reaction which hypothetically might have caused death, if it were more severe)
- Requires inpatient hospitalization or prolongation of an existing hospitalization
- Results in persistent or significant disability/incapacity
- Is a congenital anomaly/birth defect
- Is a medically important event or reaction

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

11.2 Clinical Laboratory Abnormalities and Other Abnormal Assessments as AEs

Abnormal assessments (e.g., ECGs and vital signs) that are judged by the PI as clinically significant or result in clinical sequelae will be recorded as AEs. Laboratory abnormalities will be reported by the Investigator as AEs if the abnormality is considered clinically significant or results in clinical sequelae. Laboratory abnormalities not reported as AEs are not to be reported as clinically significant in the study database.

Clinically significant abnormal laboratory findings or other abnormal assessments that are detected during the study or are present at baseline and significantly worsen following the start of the study will be reported as AEs.

The PI (or medically qualified designee) will exercise his or her medical and scientific judgment in deciding whether an abnormal laboratory finding or other abnormal assessment is clinically significant.

11.3 Timing, Frequency, and Method of Detecting AEs

Any pre-existing conditions or signs and/or symptoms present in a participant prior to the start of the study (i.e., before informed consent) should be recorded as Medical/Surgical History.

All AEs occurring after informed consent and on or before the final visit must be reported as AEs; only AEs that occur post-dose will be considered treatment-emergent. All AEs must be recorded irrespective of whether they are considered drug-related. AEs will be collected through the EOS or through 30 days after the last dose whichever is longer.

At each visit/assessment in the period defined above, AEs will be evaluated by the PI (or medically qualified designee) and recorded.

11.4 Recording of AEs

When an AE occurs, it is the responsibility of the PI or medically qualified designee to review all documentation (e.g., hospital progress notes, laboratory, and diagnostics reports) relative to the event. The PI or medically qualified designee will then record the AE on the AE CRF. Additional reporting requirements for an AE that meets serious criteria are discussed in Section 11.7 below.

The PI or medically qualified designee will attempt to establish a diagnosis of the event based on signs, symptoms, and/or other clinical information. In all cases, when available, the diagnosis should be reported as the event and not the individual signs/symptoms. It is not acceptable for the PI to send photocopies of the patient's medical records to the Sponsor in lieu of completion of the appropriate AE eCRF pages.

11.5 Evaluating AEs

11.5.1 Assessment of Intensity

The PI or medically qualified designee will assess intensity (also known as severity) for each AE reported during the study. The assessment will be based on the PI's (or medically qualified

designee's) clinical judgment. The intensity should be assigned to one of the following categories:

- Mild: An event that is easily tolerated by the participant, causing minimal discomfort and not interfering with everyday activities.
- Moderate: An event that is sufficiently discomforting to interfere with normal everyday activities.
- Severe: An event that prevents normal everyday activities.

An AE that is assessed as severe should not be confused with an SAE. Severity is a category utilized for rating the intensity of an event; and both AEs and SAEs can be assessed as severe. An event is defined as 'serious' when it meets one of the predefined outcomes as described in Section 11.1.

11.5.2 AEs at the Injection Site and Local Injection Site Reactions (LISRs)

AEs at the injection site will be graded as either Mild, Moderate or Severe. A local injection site reaction is defined as an adverse reaction (usually immunologic) developing at the site of injection and lasting at least 48 hours. For the data reporting and analysis purposes, the definition of LISR based on duration of at least 48 hours and based on the specified MedDRA preferred times is provided in Appendix 16.2. Photographs of local reactions around injection site should be obtained at the time of reporting and at the approximate time of resolution.

- Mild: Tenderness with or without associated symptoms (e.g., warmth, erythema, itching), mild pain or mild edema
- Moderate: Pain with associated phlebitis or lipodystrophy
- Severe: Tissue ulceration or necrosis with associated severe tissue damage or if operative intervention is indicated

11.5.3 Assessment of Causality

The PI (or medically qualified designee) is obligated to assess the relationship between investigational product and the occurrence of each AE. The PI (or medically qualified designee) will use clinical judgment to determine the relationship. Alternative causes, such as natural history of the underlying diseases, concomitant therapy, other risk factors, and the temporal relationship of the event to the investigational product will be considered and investigated. The PI (or medically qualified designee) will also consult the Investigator's Brochure in the determination of his/her assessment.

There may be situations when an SAE has occurred and the PI has minimal information to include in the initial SAE report. However, it is very important that the PI (or medically qualified designee) always assess causality for every event prior to transmission of the SAE report form. The PI (or medically qualified designee) may change his/her opinion of causality considering follow-up information, amending the SAE report form accordingly. The causality assessment is one of the criteria used when determining global regulatory reporting requirements.

The PI (or medically qualified designee) will provide the assessment of causality utilizing three possible categories: Not Related, Possibly Related and Probably Related.

An AE will be considered "not related" to the use of the product if any of the following tests are met:

- An unreasonable temporal relationship between administration of the product and the onset of the AE (e.g., the event occurred either before, or too long after administration of the product for it to be considered product-related)
- A causal relationship between the product and the AE is biologically implausible (e.g., death as a passenger in an automobile accident)
- A clearly more likely alternative explanation for the AE is present (e.g., typical adverse reaction to a concomitant drug and/or typical disease-related event)

An AE will be considered "Possibly related" when there is a reasonable possibility that the incident, experience, or outcome may have been caused by the product under investigation.

An AE will be considered "Probably related" when there are facts, evidence, or arguments to suggest that the event is related to the product under investigation.

11.6 Follow-up of AEs

After the initial AE, the PI is required to proactively follow each participant and provide further information on the participant's condition as deemed appropriate.

All AEs will be followed until resolution, until the condition stabilizes, until the event is otherwise explained, or until the participant is lost to follow-up. Once resolved, the appropriate AE eCRF page and SAE report form (if event is serious) will be updated. The PI, or medically qualified designee, will ensure that follow-up includes any supplemental investigations as may be indicated to elucidate the nature and/or causality of the AE or SAE. This may include additional laboratory tests or investigations, histopathological examinations, or consultation with other health care professionals. In the event of a fatal outcome in an SAE, the PI, or medically qualified designee, will attempt to obtain postmortem findings, including histopathology, and provide all additional information in a follow up SAE report.

New or updated information regarding an SAE will be recorded on a new SAE report form marked as follow-up with the appropriate follow-up number added to the report. The follow-up report will be signed and dated by the PI.

11.7 Prompt Reporting of SAEs

AEs meeting serious criteria MUST be reported promptly to the designated Pharmacovigilance CRO and the IRB, as required.

11.7.1 Completion and Transmission of the SAE reports

Once an Investigator becomes aware that an SAE has occurred in a study participant, she/he will report the information on an SAE report form to the designated Pharmacovigilance CRO within 24 hours. The SAE report form will always be completed as thoroughly as possible with all available details of the event and signed by the PI (or medically qualified designee). If the PI does not have all information regarding an SAE, he/she will not wait to receive additional information before reporting the event. The SAE report form will be updated when additional information is received.

The PI (or medically qualified designee) will always provide an assessment of causality at the time of the initial report as described in Section 11.5.3.

Email transmission of the SAE report form are the preferred methods to transmit this information to the designated Pharmacovigilance CRO. Facsimile is acceptable if email is unavailable. In rare circumstances, notification by telephone is acceptable, with a copy of the SAE report sent by overnight mail. Initial notification via the telephone does not replace the need for the PI, or medically qualified designee, to complete and sign the SAE report form within the outlined time frames.

The Sponsor will provide a list of project contacts for SAE receipt, fax numbers, telephone numbers, and mailing addresses. Any event that in the opinion of the PI may be of immediate or potential concern for the participant's health or well-being will be reported to the Sponsor emergency contact listed below.

Sponsor Emergency Contact
[redacted]
[redacted]

11.7.2 Serious Adverse Event Reports to the IRB

The PI, or responsible person per local requirements, will comply with the applicable local regulatory requirements related to the reporting of SAEs to the appropriate IRB.

11.8 Regulatory Requirements for Reporting of SAEs

The PI (or medically qualified designee) will promptly report all SAEs in accordance with the procedures detailed in Section 11.7. Prompt notification of SAEs by the PI **is essential** so that the Sponsor may comply with its regulatory obligations.

11.9 Post-study AEs

A post-study AE is defined as any event that occurs outside of the AE detection period defined in Section 11.3.

Investigators are not obligated to actively seek AEs in former study participants. However, if the Investigator learns of any SAE at any time after a participant has been discharged from the study,

and he/she considers the event reasonably related to the investigational product, the PI will promptly notify Arrowhead.

11.10 SAEs Related to Study Participation

An SAE considered related to study participation (e.g., procedures, invasive tests, a change in existing therapy), even if it occurs during the pre- or post-treatment period, will be reported promptly (refer Section 11.7).

12. DATA ANALYSIS AND STATISTICAL CONSIDERATIONS

In general, continuous data will be summarized by descriptive statistics, including number of patients, mean, standard deviation, median, minimum, and maximum. Categorical data will be summarized by the number and percentage of subjects.

12.1 Sample Size Considerations

This study represents a proof of principle study, and as such no formal sample size calculation was performed. Results from this study will be utilized in sample size calculations for subsequent studies.

12.2 Screening Data

Demographics will be tabulated by participant and summarized by cohort and treatment group. Eligibility assessments at baseline, including medical/surgical history data and physical examination data (including height and weight), will be listed for each participant.

12.3 Safety/Tolerability Data

In general, safety analyses will be performed and the results summarized by-cohort and treatment group.

Treatment-emergent AEs will be summarized using the latest version of MedDRA by System Organ Class (SOC) and Preferred Term (PT), classified from verbatim terms. The incidence and percentage of participants with at least 1 occurrence of a PT will be included, per the most severe grade using a 3-point scale (mild, moderate, severe). The number of events per Preferred Term will also be summarized. Causality (relationship to study treatment) will be summarized separately.

The incidence and frequency of AEs, SAEs, related AEs, related SAEs and AEs leading to withdrawal, dose modification, or treatment discontinuation will be summarized by dose and treatment group per SOC and Preferred Terms. AEs will also be summarized in listings. The duration of AEs will be determined and included in listings, along with the action taken and outcome.

The incidence of laboratory abnormalities will be summarized. Results for variables that are not coded will be presented in the listings as “below, within, and above” the normal limits of the laboratory. Pregnancy test results will be summarized separately by time point.

Vital sign and oxygen level measurements will be summarized at each scheduled time point using descriptive statistics. Physical examination findings will be summarized by time point and presented in patient listings.

ECG parameter changes overall, changes from baseline and qualitative assessments will be summarized.

12.4 Immunogenicity (Anti-drug Antibodies) Data

Changes from assay negative to positive will be summarized by dose and time to positive. Descriptive statistics of immunogenicity parameters will include mean, SD, minimum, and maximum.

12.5 Pharmacokinetic Data

Plasma concentrations of ARO-HSD collected at specified time points post-dose from all participants at different dose levels will be used to calculate the following single dose pharmacokinetic parameters (Table 6):

Table 6 Definitions of Pharmacokinetic Parameters to be Assessed

AUC_{0-24}	The area under the plasma concentration versus time curve from the zero to 24 hours
AUC_{inf}	The area under the plasma concentration versus time curve from zero to infinity
C_{max}	The maximum plasma concentration will be obtained directly from the plasma concentration time profile
t_{max}	The time to maximum plasma concentration will be obtained by inspection
$t_{½}$	The half-life will be calculated by the equation $t_{½} = \ln(2)/k_{el}$

The pharmacokinetic parameters will be determined using non-compartmental method(s). Descriptive statistics of pharmacokinetic parameters will include mean, standard deviation (SD), and coefficient of variation (CV), minimum and maximum. Dose-related trends in pharmacokinetic parameters will be assessed.

Pharmacokinetic parameters will be tabulated and summarized by dose level. The concentration-time profiles for each participant and the mean concentration-time profiles by dose level will be plotted with concentration presented on both linear and logarithmic scales.

Statistical analysis will be performed on the pharmacokinetic parameters using validated statistical software.

12.6 Pharmacodynamic Data

- ***Lipid Parameters:*** LDL-C, Total Cholesterol, non-HDL-C, HDL-C, VLDL-C, and Triglycerides
- ***Retinoid pathway markers:*** retinol, retinol binding protein

- ***NASH biomarkers***: ELF [Hyaluronic acid (HA), Procollagen III amino terminal peptide (PIIINP), Tissue inhibitor of metalloproteinase 1 (TIMP-1)], cytokeratin-18, PRO-C3, IL-1β, IL-6, IL-10, INF-gamma, and TNF-α

- ***Immunogenicity:*** Specified subjects will be assessed for anti-drug antibodies.

- ***Genotype:*** All patient cohorts will be assessed for HSD17B13 and PNPLA3 gene variants with the subject's written consent.

- ***Exosomal analysis:*** Blood will be drawn for exosomal analysis of *PNPLA3* and *HSD17B13* if technically feasible.

- ***MRI-PDFF:*** Liver fat content via MRI-PDFF will be evaluated and quantified. Change from baseline will be measured.

- ***FibroScan®:*** Per the Schedule of Assessments, pre-dose and post-dose liver stiffness baseline will be measured.

- ***Liver Biopsy*:** Samples from biopsies collected following multiple doses of ARO-HSD at different dose levels will undergo analysis for HSD17β13 mRNA knockdown and evaluation in PNPLA3 mRNA expression. HSD17B13 and PNPLA3 protein expression may be assessed if scientifically feasible.

12.7 Data Recording and Quality Control

Source documents must be maintained for each participant in the study, consisting of all demographic and medical information, including clinical laboratory data, etc. A copy of the signed informed consent form must be retained. All information on the eCRFs must be traceable to these source documents in the participant's file.

Data recorded in all participants' eCRFs will be subjected to a quality control review.

13. STUDY APPROVAL AND CONDUCT

The following conditions will be met.

13.1 Regulatory Approval

The requirements for the conduct of clinical trials in accordance with local applicable regulations will be met before commencement of this study.

13.2 Ethics Committee (EC)/ Institutional Review Board (IRB) Approval

Prior to initiation of the study, written EC/IRB approval of the Protocol and Informed Consent Forms, based on the principles of ICH cGCP procedures, will be received. A copy of the signed and dated letter of approval will be provided to the clinical site and Arrowhead Pharmaceuticals, Inc. prior to study commencement. Any written information and/or advertisements to be used for volunteer recruitment will be approved by the EC/IRB prior to use. A list of the EC/IRB

voting members, their titles or occupations, FWA number (where applicable) and their institutional affiliations will be requested before study initiation.

Protocol modifications that may impact patient safety or the validity of the study will be approved by the IRB, following written agreement from the Sponsor.

13.3 Ethical Considerations

This study will be carried out per the Declaration of Helsinki 1964, as modified by the 64th World Medical Assembly, Fortaleza, Brazil, October 2013, the Notes for Guidance on Good Clinical Practice (cGCP) (2000) (CPMP/ICH/135/95), and the Principles of the ICH cGCP. The protocol will be submitted for approval to the EC/IRB, and written approval obtained before subjects are enrolled. The composition of the EC/IRB will also be provided to the Sponsor. If approval is suspended or terminated by the IRB, the PI will notify the Sponsor immediately

Where applicable, the clinical site and Arrowhead Pharmaceuticals, Inc. agree to abide by the local compensation guidelines for injury resulting from participating in a company-sponsored research project. Compensation will only be provided on the understanding that the provision of compensation does not amount to an admission of legal liability and is subject to the proposed recipient signing a full and complete release of the company from all claims, damages and costs.

13.4 Written Informed Consent

Informed consent will be obtained before the patient can participate in the study. The contents and process of obtaining informed consent will be in accordance with all applicable regulatory requirements. Study participation includes all screening procedures, as well as any wash-out of excluded medications.

It is the responsibility of the PI (or medically qualified designee) to obtain a written informed consent from everyone participating in this study after adequate explanation of the aims, methods, objectives, and potential hazards of the study. The PI (or medically qualified designee) must also explain to the participants that they are completely free to refuse to enter the study or to withdraw from it at any time. Appropriate forms for documenting a written consent will be provided by the PI or by Arrowhead Pharmaceuticals, Inc.

For this study, each eligible participant will be required to provide written informed consent before participation in the study.

All eligible participants will have the study explained by the PI or designee. They will receive a full explanation, in lay terms, of the aims of the study, the discomforts, risks and benefits in taking part as well as of insurance and other procedures for compensation in case of injury. It will be explained that the study is for research purposes only and is not expected to provide any therapeutic benefit to the individual. It will be pointed out that they can withdraw from the study at any time without prejudice. Each participant will acknowledge receipt of this information by giving written informed consent for participation in the study. The volunteer will be given a copy of the signed Informed Consent Form to retain.

13.5 Emergency Contact with Principal Investigator

Suitable arrangements will be made for participants to contact the PI or medically trained designee in the event of an emergency.

13.6 Notification of General Practitioner

It is the responsibility of the PI or designee, to notify, where applicable, with the consent of the participant, the general practitioner of the patient's participation in the trial, by sending a letter stating the nature of the trial, treatments, expected benefits or adverse events and concomitant drugs to be avoided.

13.7 Clinical Laboratory Certification and Reference Ranges

Before the initiation of this study, the PI, or designee, will obtain a copy of the certification form, with certification number and expiration date for all clinical laboratories (excluding central laboratories) used in the study. Reference ranges for each clinical laboratory test used in this study will be obtained from the appropriate laboratory, which will perform the test for the study.

13.8 Protocol Deviations

A protocol deviation is defined as any intentional or unintentional change to, or noncompliance with, the approved protocol procedures or requirements. The PI will conduct the study in compliance with the approved protocol and will not implement any deviation from or changes to the protocol without prior agreement by the Sponsor and review and documented approval from the EC/IRB of an amendment, except where necessary to eliminate an immediate hazard to study patients.

Deviations may result from the action or inaction of the participant, PI, or site staff. Examples of deviations include, but are not limited to:

- Failure to adhere to study exclusion and inclusion criteria
- Failure to comply with dispensing or dosing requirements
- Use of medications, food, drink, herbal remedies, or supplements that are specifically prohibited in the protocol
- Missed or out-of-window visits
- Drug dosing not administered within the time frame specified in the protocol
- Failure to adhere to test requirements, including vital signs, laboratory tests, physical examinations, PK blood draws, medical history, etc. – either tests not done, incorrect tests done, or not done within the time frame specified in the protocol

Protocol deviations impacting patient safety or eligibility will be reported to the Sponsor or CRO within 2 business days of occurrence and to the EC/IRB /competent regulatory authority per local regulatory requirements.

The PI is responsible for ensuring that any known protocol deviations are recorded and reported as agreed.

13.9 Termination of the Study

The Sponsor reserves the right to discontinue the trial at any time. Reasons will be provided in the event of this happening. The PI reserves the right to discontinue the study for safety reasons at any time in collaboration with the Sponsor.

14. STUDY ADMINISTRATION

14.1 Study Monitoring

Arrowhead Pharmaceuticals, Inc. is responsible for assuring the proper conduct of the study about protocol adherence and validity of the data recorded on the eCRFs. Participant confidentiality will be maintained.

In accordance with applicable regulations, cGCP, and Arrowhead Pharmaceuticals, Inc. procedures, Arrowhead Pharmaceuticals, Inc. will be responsible for assigning a study monitor (CRA) who will contact the site to organize a visit prior to participant enrolment to review the protocol and data collection procedures with site staff. In addition, the assigned study monitor will periodically contact the site, including conducting on-site visits. The extent, nature and frequency of on-site visits will be based on such considerations as the study objective and/or endpoints, the purpose of the study, study design complexity, and enrolment rate.

During these site visits, the study monitor will:

- Check the progress of the study
- Review study data collected
- Conduct source document verification
- Identify any issues and address their resolution
- Check investigational product accountability
- Review blood and urine samples and ensure they are labeled and stored correctly

This will be done to verify that the:

- Data are authentic, accurate and complete.
- Safety and rights of participants are being protected.
- Study is conducted in accordance with the currently approved protocol (and any amendments), cGCP and all applicable regulatory requirements.

The PI agrees to allow the monitor direct access to all relevant documents and to allocate his/her time and the time of his/her staff to the monitor to discuss findings and any relevant issues.

At study closure, a study monitor will conduct the following activities in conjunction with the PI or site staff as appropriate:

- Return of all study data to Arrowhead Pharmaceuticals, Inc
- Data queries
- Accountability, reconciliation and arrangements for unused investigational product(s)

- Inventory and final disposition (e.g., destruction, shipping to repository, etc.)
- Review of site study records for completeness

14.2 Quality Assurance

To ensure compliance with cGCP and all applicable regulatory requirements, Arrowhead Pharmaceuticals, Inc. may conduct a quality assurance audit of the study site. Regulatory agencies may also conduct a regulatory inspection of this study. Such audits/inspections can occur at any time during or after completion of the study. If an audit or inspection occurs, the PI and clinical site agree to notify Sponsor as soon as possible following awareness of an impending regulatory inspection. The PI and clinical site agree to allow the auditor/inspector direct access to all relevant documents and allocate his/her time and the time of his/her staff to the auditor/inspector to discuss findings and any relevant issues.

14.3 Records Retention

Following closure of the study, the PI must maintain all site study records in a safe and secure location. The records must be maintained to allow easy and timely retrieval, when needed (e.g., audit or inspection) and whenever feasible, to allow any subsequent review of data in conjunction with assessment of the facility, supporting systems and staff. When permitted by local laws/regulations or institutional policy, some of these records can be maintained in a format other than hard copy (e.g., microfiche, scanned, electronic); however, caution needs to be exercised before such action is taken. The PI must assure that all reproductions are legible and are a true and accurate copy of the original and meet accessibility and retrieval standards, including re-generating a hard copy, if required. Furthermore, the PI must ensure there is an acceptable back-up of these reproductions and that an acceptable quality control process exists for making these reproductions.

Arrowhead Pharmaceuticals, Inc. will inform the PI of the time period for retaining these records to comply with all applicable regulatory requirements. The minimum retention time will meet the strictest standard applicable to that site for the study, as dictated by any institutional requirements or local laws or regulations, or Arrowhead Pharmaceuticals, Inc. standards/procedures; otherwise, the retention period will default to 15 years.

The material to be stored shall include, but is not limited to, the following:

- Signed and dated copy of the final study protocol and any amendments
- Signed and dated letter of IRB approval, letter of constitution of the IRB and copies of any other correspondence relevant to the study with the IRB or regulatory authorities
- The IRB approved Informed Consent Form
- Current *curriculum vitae* (signed and dated) of the Principal Investigator and co-workers with major responsibilities in the trial
- Site Signature and Delegation of Responsibility Log
- FDA Form 1572 (where applicable)
- Financial Disclosure Form(s)
- Blank CRF/eCRF

- Signed participant informed consent forms
- Laboratory reference ranges (signed and dated)
- The completed CTN Application Form (where applicable)
- Clinical raw data including the Source Data Forms, all clinical laboratory report forms, patient CRFs, drug accountability forms, and dispensing records, etc

15. INFORMATION DISCLOSURE AND INVENTIONS

15.1 Ownership

[redacted]

15.2 Confidentiality

[redacted]

15.3 Publication

[redacted]

[redacted]

16. APPENDICES

16.1 Study Modification Guidelines For Subjects And Patients With Baseline Pre-Dose Evidence Of Liver Disease

Treatment-Emergent ALT	Treatment-Emergent Total Bilirubin (TBL)	Liver Symptoms	Action
Normal baseline with treatment emergent (TE) increase to ALT > 5x ULN Elevated baseline with TE increase to: ALT > 3x baseline or > 300 U/L (whichever occurs first)	Normal	None	Repeat ALT, AST, ALP, TBL, in 2–3 days Follow-up for symptoms.
Normal baseline with TE increase to ALT > 8x ULN Elevated baseline with TE increase to: ALT > 5x baseline or > 500 U/L (whichever occurs first)	Normal	None	Interrupt study drug. Initiate close observation and workup for competing etiologies. (see below) Study drug can be restarted only if an alternative etiology is identified and liver enzymes return to baseline.
Normal baseline with TE increase to ALT > 3x ULN Elevated baseline with TE increase to ALT > 2x baseline or > 200 U/L (whichever occurs first)	TBL > 2x ULN	None	Interrupt study drug. Initiate close observation and workup for competing etiologies. Study drug can be restarted only if an alternative etiology is identified and liver enzymes return to baseline.

Normal baseline with TE increase to ALT > 3x ULN Elevated baseline with TE increase to ALT > 2x baseline or > 200 U/L (whichever occurs first)	Normal or elevated	Symptoms of clinical hepatitis - severe fatigue, nausea, vomiting, right upper quadrant pain	Interrupt study drug. Initiate close observation and workup for competing etiologies. Study drug should not be restarted

Source: Adapted from Chalasani, Naga and Regev, Arie et al. Drug-Induced Liver Injury in Subjects with Preexisting Chronic Liver Disease in Drug Development: How to Identify and Manage? Gastroenterology, Volume 151, Issue 6, 1046 – 1051

Guidelines for close observation for potential drug induced liver injury:
Within 72 hours, perform a symptom directed history, physical, and liver biochemistries, including evaluation of:

- New or worsening signs and symptoms of clinical hepatitis such as fatigue, nausea, vomiting, right upper quadrant pain or tenderness, fever, rash, or eosinophilia
- Concomitant medications, including acetaminophen, dietary supplements, herbal remedies, over-the-counter medications, recreational drug use, and special diets
- Alcohol consumption
- Exposure to environmental chemical agents
- Past medical history
- Complete review of systems
- Liver biochemistries including ALT, AST, alkaline phosphatase, total bilirubin, and INR

Evaluate patients for signs and symptoms of clinical hepatitis and obtain liver biochemistries until biochemistries stabilize.

If biochemistries stabilize and the subject is asymptomatic, monitor liver biochemistries once a week until they return to baseline.

Subjects who live far from study sites may be evaluated locally for history, physical exam, and laboratories, if the results are communicated promptly to the site investigator.

16.2 Local Injection Site Reactions (LISRs)

The following MedDRA Preferred Terms determined by the Sponsor's pharmacovigilance personnel represent the local injection site reaction:

Injection site discomfort	Injection site abscess
Injection site discoloration	Injection site abscess sterile

Injection site erythema	Injection site atrophy
Injection site irritation	Injection site calcification
Injection site inflammation	Injection site cellulitis
Injection site induration	Injection site dermatitis
Injection site pain	Injection site erosion
Injection site oedema	Injection site fibrosis
Injection site pruritus	Injection site indentation
Injection site rash	Injection site necrosis
Injection site urticaria	Injection site nodule
Injection site reaction	Injection site ulcer
Injection site swelling	

LISRs will only include events that start on the day of injection and persist for at least 48 hours post injection (i.e., event onset date on the day of injection and resolution date not on the day of injection or the day after the injection) will be included. Events with onset date on the day of injection and missing resolution date will also be included in the summary.

The following calculation will be utilized to determine the percentage of injections leading to local injection site reactions:

(A/B)*, where A = number of injections with a local injection site reactions, and B = total number of injections.

17. REFERENCES

Abul-Husn, NS et al. A Protein-Truncating HSD17B13 Variant and Protection from Chronic Liver Disease. *The New England journal of medicine* vol. 378,12 (2018): 1096-1106.

Ekstedt, M., Nasr, P. & Kechagias, S. Curr Hepatology Rep. 2017;16: 391.

Fire, A., S. Xu, M. K. Montgomery, S. A. Kostas, S. E. Driver, and C. C. Mello. Potent and specific genetic interference by double-stranded RNA in C. elegans. Nature. 1998; 391:806–811.

Geary RS, Yu RZ, Watanabe T, Henry SP, Hardee GE, Chappell A, Matson J, Sasmor H, Cummins L, Levin AA. Pharmacokinetics of a tumor necrosis factor-alpha phosphorothioate 2'-O-(2-methoxyethyl) modified antisense oligonucleotide: comparison across species. Drug Metab Dispos. 2003;31(11):1419-28.

Henry SP, Geary RS, Yu R, Levin AA. Drug properties of second-generation antisense oligonucleotides: how do they measure up to their predecessors? Curr Opin Investig Drugs. 2001; 10:1444-9.

Janas MM, Schlegel MK, Harvison CE, et al. Selection of GalNAc-conjugated siRNAs with limited off-target-driven rat hepatotoxicity. Nature Comm. 2018;9:723.

Ma, Y.et al. 17-Beta Hydroxysteroid Dehydrogenase 13 Is a Hepatic Retinol Dehydrogenase Associated With Histological Features of Nonalcoholic Fatty Liver Disease. Hepatology, 2018;69:1504-1519.

Prati D, Taioli E, Zanella A, et al. Updated definitions of healthy ranges for serum alanine aminotransferase levels. Ann Intern Med. 2002; 137:1–9.

Rockey, D. C., Caldwell, S. H., Goodman, Z. D., Nelson, R. C. and Smith, A. D. Liver biopsy. Hepatology. 2009;49:1017-1044.

Sharp, P. A. 2001. RNA interference. Genes Dev. 15:485–490.

Su, W, Wang Y, Jia Xiao, et al. Role of 17β-HSD13 in NAFLD. Proceedings of the National Academy of Sciences Aug 2014;111(31):11437-11442.

Vernon, G, Baranova, A. and Younossi, Z. M. Systematic review: the epidemiology and natural history of non-alcoholic fatty liver disease and non-alcoholic steatohepatitis in adults. Alimentary Pharmacology & Therapeutics, 2011;34:274-285.

Yu, R.Z., H. Zhang, R.S. Geary, M. Graham, L. Masarjian, K. Lemonidis, R. Crooke, N.M. Dean and A.A. Levin. Pharmacokinetics and pharmacodynamics of an antisense phosphorothioate oligonucleotide targeting Fas mRNA in mice. J. Pharmacol. Exp. Ther. 2001;296: 388–395.

Yu RZ, Grundy JS, Henry SP, et al. Predictive dose-based estimation of systemic exposure multiples in mouse and monkey relative to human for antisense oligonucleotides with 2'-o-(2-methoxyethyl) modifications. Mol Ther Nucleic Acids. 2015;4(1):e218.