PROTOCOL 3V2640-CLIN-005

A PHASE 2, MULTI-CENTER, SINGLE-BLIND, RANDOMIZED, PLACEBO CONTROLLED STUDY OF TVB-2640 IN SUBJECTS WITH NON-ALCOHOLIC STEATOHEPATITIS

IND Number:	138506
Protocol Version:	7.0 (US Only)
Date:	17 March 2021
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Sagimet Biosciences
[redacted]

INVESTIGATOR'S AGREEMENT

I have received and read the Investigator's Brochure for TVB-2640. I have read the protocol for Study 3V2640-CLIN-005 and agree to conduct the study as outlined. I agree to maintain the confidentiality of all information received or developed in connection with this protocol.

_____

Printed Name of Investigator

_____

Signature of Investigator

_____

Date

[redacted]

Table 1 Emergency Contact Information

[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
[redacted]	[redacted]	[redacted]
Serious Adverse Event (SAE) Reporting	*SAEs are to be reported within 24 hours of discovery via the electronic data capture (EDC) system.*	

2. SYNOPSIS

Name of Sponsor/Company:
Sagimet Biosciences

Name of Investigational Product:
TVB-2640

Name of Active Ingredient:
TVB-2640

Title of Study:
A Phase 2, Multi-Center, Single-Blind, Randomized, Placebo-Controlled Study of TVB-2640 in Subjects With Non-Alcoholic Steatohepatitis

Study center(s):

Coordinating Investigator:
This study will be conducted at approximately 15 study centers in the United States (US) and China. The Coordinating Investigator is Rohit Loomba, MD, University of California, San Diego, CA.

Studied period (years):	**Phase of development:**
Estimated date first subject enrolled: April 2019 Estimated date last subject completed: August 2021	2

Objectives:

Primary:

- To determine the effect of once daily (QD) TVB-2640 for 12 weeks versus placebo on the change in hepatic fat fraction by proton density fat fraction magnetic resonance imaging (MRI-PDFF) from baseline in subjects with non-alcoholic steatohepatitis (NASH).
- To determine the safety of QD TVB-2640 versus placebo in subjects with NASH, including the effects on alanine aminotransferase (ALT) and aspartate aminotransferase (AST).

Secondary:

- To determine the effect of once-daily (QD) TVB-2640 for 12 weeks versus placebo in subjects with NASH on:
  - Lipid and lipoprotein parameters, including low-density lipoprotein cholesterol (LDL-C), non-LDL-C, high-density lipoprotein cholesterol (HDL-C), non-HDL-C, total cholesterol, triglycerides, apolipoprotein B (ApoB), and lipoprotein(a) (Lp[a]) particles.
  - NASH and fibrosis biomarkers including cytokeratin-18 (CK-18), fibrosis-4 (FIB-4), and enhanced liver function (ELF) or FIBROSpect 2 or ProC3 test.
  - Eicosanoid panel.

Exploratory:

- To determine the effect of QD TVB-2640 for 12 weeks versus placebo in subjects NASH on clinical measures, including:
  - Liver fibrosis, as determined by vibration-controlled transient elastography (VCTE).
  - Metabolic and inflammatory parameters, including fasting glucose, insulin, homeostatic model assessment insulin resistance (HOMA2-IR), glycated hemoglobin (HbA1c), non-

esterified fatty acid (NEFA), adipose tissue insulin resistance (adipo-IR), adiponectin, resistin, Interleukin 6 (IL-6), and gamma-glutamyl transpeptidase (GGT).
  - Anthropometric parameters, including weight, waist and hip circumference, waist-hip ratio, and blood pressure.
  - Lipidomic analyses for de novo lipogenesis (DNL).
- Explore the relationship of plasma drug exposure to changes in efficacy and safety biomarkers.
- Explore possible relationships between genomic markers of NASH and responses to treatment.

Methodology:

This is a multi-center, randomized, single-blind, placebo-controlled, dose-escalation study to evaluate the safety and efficacy of TVB-2640 in subjects with NASH. Male and female subjects aged ≥18 years with either biopsy-proven NASH within 2 years before randomization and MRI-PDFF ≥8% or, if prior liver biopsy was not performed or results are not available, magnetic resonance elastography (MRE) ≥2.5 kpa and MRI-PDFF ≥8% during screening are planned to be enrolled. Approximately 117 unique subjects will be enrolled and randomized, with at least 90 subjects enrolled in the US and approximately 27 evaluable subjects (18 active, 9 placebo) enrolled in China. In the US, subjects will be enrolled and randomized to achieve 90 evaluable subjects (30 active and 15 placebo in each of 2 dose cohorts). The 50 mg cohort will enroll and randomize approximately 27 additional subjects from China. The US-only 75 mg cohort will enroll approximately 12 subjects (in order to ensure 10 active, evaluable subjects) who may have previously received placebo in the 25 or 50 mg cohort. A subject is defined as evaluable if they receive study drug for at least 8 weeks and have a baseline and at least 1 post-baseline MRI-PDFF assessment on or after Week 8.

Subjects will be screened for study eligibility within 60 days before randomization. Subjects must meet all of the inclusion criteria and none of the exclusion criteria to participate in the study. Subjects who are determined to be eligible for the study, based on screening assessments, are to be randomized into the study within 24 hours before baseline (Day 1, the first day of study drug administration). Subjects who are randomized are considered to be enrolled in the study. Initially, 2 TVB-2640 dose levels are planned to be evaluated in a sequential fashion: 25 mg and, if study suspension criteria are not met at the 25 mg dose level (see Section 7.2), 50 mg. At each of the 2 initial dose levels, subjects will be randomly assigned to TVB-2640 or placebo at a 2:1 ratio, with randomization stratified by type 2 diabetes mellitus status. The 50 mg dose level will also be stratified by country:

- Cohort 1, 25 mg dose level: TVB-2640 (Planned N=30) and Placebo (Planned N=15)
- Cohort 2, 50 mg dose level: US: TVB-2640 (Planned N=30) and Placebo (Planned N=15); China: TVB-2640 (Planned N=18) and Placebo (Planned N=9); Total: TVB-2640 (Planned N=48) and Placebo (Planned N=24)

Subjects will receive TVB-2640 or placebo QD PO for 12 weeks, or until the time of the final on-treatment MRI-PDFF if after Week 12, but for no longer than 16 weeks, with the first dose administered on Day 1. During the 12-week Treatment period, subjects are to attend study center visits at Week 1, Days 1 and 2, and Weeks 2, 4, 8, and 12. After completion of the 12-week Treatment period, subjects are to attend a Follow-up visit at Week 16 for post-treatment safety and efficacy assessments.

All subjects must complete the 12-week treatment period at the 25 mg dose level, with review of all safety data and written approval by the Safety Review Committee and reviewed by the Food and Drug Administration before enrollment and treatment of subjects at the 50 mg dose level may commence.

After completion of Cohorts 1 and 2, and again if no stopping criteria are met upon review by the Independent Safety Review Committee (SRC), subjects assigned to placebo in either cohort are eligible to crossover to receive TVB-2640 75 mg in Open-label Cohort 3 (planned N=12 in order to obtain 10 evaluable subjects). Cohort 3 will be conducted at select sites in the US only.

The Sponsor notes that the coronavirus disease-2019 (COVID-19) pandemic may impact the conduct of the 50 mg and 75 mg cohorts, with challenges potentially arising from quarantines and travel limitations affecting study subjects as well as clinic and imaging facility closures. Accordingly, safety assessments scheduled for the Week 12 and 16 visits, as identified in Table 2, including sample collection for clinical laboratory tests, physical examinations, and documentation of adverse events (AEs), may be performed at an alternate study center or by home health care visit. Furthermore, if the subject is unable to attend the Week 12 visit at the study center, one MRI-PDFF may be performed at any time between Week 11 and 16. If this scan is performed after Week 12, the subject must continue to receive study drug until the final scan is performed; however, study drug is not to be continued for >16 weeks. The safety of subjects who continue dosing beyond 12 weeks will be assessed during this continued dosing period by planning for blood sample collection at an alternate site or home visit at Week 12 and blood sample collection and physical examination on the last day of dosing. A 4-week off treatment safety assessment is to occur after the last dose, regardless of whether a subject receives treatment for 12 or 16 weeks.

In Cohorts 1 and 2 study drug will be dispensed to subjects in a single-blinded fashion. All other study personnel, with the exception of personnel conducting/interpreting imaging studies (see Section 11.1) at each study center, will be unblinded to the subject's treatment assignment. In Cohort 3, study drug will be dispensed in an open-label fashion.

In Cohorts 1 and 2, subjects will undergo MRI-PDFF prior to the start of treatment (pre-dose), at the end of the initial 12 week Treatment Period (between 11 and 12 weeks after the start of treatment), and at Week 16. In accordance with the Food and Drug Administration (FDA) guidance for the conduct of clinical trials of medicinal products during the COVID-19 pandemic, if the subject is unable to attend the Week 12 visit at the study center, one MRI-PDFF may be performed at any time between Week 11 and 16. If this scan is performed after Week 12, the subject must continue to receive study drug until the final scan is performed. However, study drug is not to be continued for >16 weeks. In Cohort 3, subjects will undergo MRI-PDFF prior to the start of treatment (pre-dose) and at the end of the initial 12 week Treatment Period (between 11 and 12 weeks after the start of treatment).

During the study, safety will be assessed by vital signs (oral temperature, pulse, respiratory rate, and blood pressure), 12-lead electrocardiogram (ECG), physical examination, including ophthalmologic examination, and clinical laboratory testing (hematology, chemistry, and urinalysis). Subjects will be evaluated for AEs and concomitant medication use throughout the study.

Efficacy variables include MRI-PDFF as well as measurement of liver aminotransferases, lipid parameters, and markers of NASH and liver fibrosis. The primary efficacy endpoint is percent change from baseline in liver fat at Week 12, as determined by MRI-PDFF. The percentage of subjects with at least a 30% reduction in the primary efficacy endpoint is a key secondary efficacy endpoint.

[redacted]

Number of subjects (planned):

Approximately 117 unique subjects will be enrolled and randomized in a 2:1 fashion to TVB-2640 or placebo in Cohorts 1 and 2. This includes at least 45 US subjects in the 25 mg cohort, and at least 45 US subjects and approximately 27 Chinese subjects in the 50 mg cohort. Up to 12 subjects assigned to placebo in Cohorts 1 and 2 will be eligible to crossover and receive TVB-2640 75 mg in an open-label fashion in Cohort 3 in the US.

Diagnosis and main criteria for inclusion:

The study population is male and female subjects aged ≥18 years with either biopsy-proven NASH within 2 years before randomization and MRI-PDFF ≥8% OR, if prior liver biopsy was not performed or results are not available, MRE ≥2.5 kpa and MRI-PDFF ≥8% during screening.

Note that subjects who initially fail to meet all entrance criteria based on screening assessments (i.e., screen failures) may be submitted for re-screening once (see Section 8.5 for details).

Inclusion Criteria:

1. Must be willing to participate in the study and provide written informed consent.
2. Aged ≥18 years with a body mass index (BMI) ≤40 kg/m^2.
3. If a female subject of child-bearing potential, must have a negative serum pregnancy (beta-human chorionic gonadotropin [β-HCG]) test during screening and is not breastfeeding, does not plan to become pregnant during the study, and agrees to use two forms of birth control, one being a barrier method (i.e., condoms, diaphragm, non-hormonal intrauterine device [IUD]) throughout the study and for at least 3 months after the final study drug dose.

   Hormonal contraception (estrogens stable ≥3 months) and hormonal IUDs are permitted if used with a secondary barrier birth control method (e.g., condoms).
4. If a female subject of non-child-bearing potential, must have documentation of surgical sterility (bilateral oophorectomy, hysterectomy, or tubal ligation) or natural sterility (>12 consecutive months without menses).
5. If male, agrees to use adequate birth control throughout the study and for at least 3 months after the final study drug dose.
6. Prior liver biopsy within 24 months of randomization with fibrosis Stage 1 to 3 and a non-alcoholic fatty liver disease (NAFLD) activity score (NAS) of ≥4 with at least a score of 1 in each of the following NAS components:
   - Steatosis.
   - Ballooning degeneration.
   - Lobular inflammation.

   AND

   - Confirmation of ≥8% liver fat content on MRI-PDFF.

   OR, if prior biopsy is not available:

   - Either overweight (BMI ≥25 and <30 kg/m^2) or obese (BMI ≥30 kg/m^2) or diabetic or ALT ≥30 U/L for men or ≥19 U/L for women or fatty liver on ultrasound and at least one more feature of metabolic syndrome by Adult Treatment Panel III (ATP III) criteria.

   AND

   - MRE ≥2.5 kpa (Cohorts 1 and 2 only) and MRI-PDFF ≥8% during screening.

For Cohort 3 Only

7. Subject may have previously participated in this study in Cohort 1 or 2 and is known to have received placebo by single-blind treatment assignment.

Exclusion Criteria

1. History of significant alcohol consumption for a period of more than 3 consecutive months within 1 year prior to screening, as assessed by the Investigator.

   Note: Significant alcohol consumption is defined as average of >20 g/day in female subjects and >30 g/day in male subjects. For reference, 14 grams of alcohol are contained in the following:

   - 12 fl oz regular beer containing ~5% alcohol.
   - 8-9 fl oz malt liquor containing ~7% alcohol.
   - 5 fl oz of table wine containing ~12% alcohol.
   - 1.5 fl oz of distilled spirits (e.g., gin, rum, vodka, whiskey) containing ~40% alcohol.
2. Inability to reliably quantify alcohol consumption based upon judgment of the Investigator.
3. Use of drugs historically associated with NAFLD (amiodarone, methotrexate, systemic glucocorticoids, tetracyclines, tamoxifen, estrogens at doses greater than those used for hormone replacement, anabolic steroids, valproic acid, and other known hepatotoxins) for more than 2 weeks in the year prior to screening.
4. Weight gain or loss >5% in the 6 months prior to randomization or >10% in the 12 months prior to screening.
5. Prior or planned (during the study period) bariatric surgery (e.g., gastroplasty, roux-en-Y gastric bypass).
6. Type 1 diabetes.
7. Uncontrolled Type 2 diabetes defined as:
   - HbA1c ≥9.5% during screening. (Subjects with HbA1c ≥9.5% may be retested during screening).
   - Basal insulin dose adjustment >10% within 60 days prior to enrollment.
   - Requirement for glucagon-like peptide analogue or a complex oral anti-diabetic (OAD) regimen (3 or more OADs) within 6 months of screening.
   - History of severe hypoglycemia (symptomatic hypoglycemia requiring outside assistance to regain normal neurologic status) within the previous year.

   Note: Individual diabetes regimens will be reviewed by Investigator and may be adjusted based on American Diabetes Association guidelines.
8. Presence of cirrhosis on liver biopsy (Stage 4 fibrosis) or imaging (MRE ≥4.67 kpa).
9. Platelet count $<150\times10^9$/L.
10. Clinical evidence of hepatic decompensation as defined by the presence of any of the following abnormalities:
    - Serum albumin ≤3.5 g/dL,
    - International normalized ratio (INR) ≥1.3 (a single retest is permitted if laboratory error is suspected),
    - Total bilirubin >1.8 mg/dL and/or direct bilirubin >0.8 mg/dL (unless due to Gilbert's syndrome, as documented in the subject's medical history/records), or
    - History of esophageal varices, ascites, or hepatic encephalopathy.

11. Evidence of other forms of chronic liver disease including the following:
    - Hepatitis B, as defined by presence of hepatitis B surface antigen (HBsAg) at screening,
    - Hepatitis C, as defined by presence of hepatitis C virus (HCV) antibody (anti-HCV), and HCV ribonucleic acid (RNA). Subjects with positive anti-HCV who test negative for HCV RNA at screening will be allowed to participate in the study,
    - Evidence of ongoing autoimmune liver disease,
    - Primary biliary cirrhosis,
    - Primary sclerosing cholangitis,
    - Wilson's disease,
    - Homozygous alpha-1-anti-trypsin deficiency,
    - History of hemochromatosis or iron overload,
    - Drug-induced liver disease,
    - Known bile duct obstruction,
    - Suspected or proven liver cancer, or
    - Any other type of liver disease other than NASH.
12. Serum ALT >5 × the upper limit of normal (ULN).
13. Liver function tests (ALT/AST) > 5 × ULN at screening. One repeat test may be allowed within 7 days at the discretion of the Investigator.
    - If AST or ALT is > than ULN at screening visit an additional ALT, AST, INR, and total bilirubin will be obtained at least 2 weeks after the screening laboratories are obtained. If the abnormal parameters increase by > 30% at the retesting visit, the subject may not be randomized. The test may then be repeated in 2-4 weeks and if there is no further increase in the abnormal laboratory parameter, the subject may be randomized.
14. Estimated glomerular filtration rate <60 mL/min/1.73 m^2, as determined using the Modification of Diet in Renal Disease Study equation.
15. History of biliary diversion.
16. Positive for human immunodeficiency virus infection.
17. Active, serious medical disease with likely life expectancy <2 years.
18. Active substance abuse, including inhaled or injected drugs, within 1 year prior to screening. (Note that recreational cannabis/tetrahydrocannabinol use is permissible.)
19. Use of any excluded medications listed in Section 9.8.1.
20. Participation in an investigational new drug study in the 30 days prior to randomization (with the exception of Cohort 3 in which participants are prior placebo recipients in Cohort 1 and 2 of this study).
21. History of clinically significant dry eye (xerophthalmia) or other corneal abnormality or, if a contact lens wearer, does not agree to abstain from contact lens use from baseline through the last study drug dose.
22. Evidence of a clinically significant abnormality on slit-lamp examination or other clinically significant ophthalmologic finding, as determined by an ophthalmologist.
23. Known allergy or hypersensitivity to components of TVB-2640.
24. Prior history of hypersensitivity or drug/radiation-induced or other immune-mediated pneumonitis.
25. Prior history of palmar-plantar erythrodysesthesia syndrome.

26. Any contraindication to MRI (e.g., claustrophobia, metal implants).
27. Positive severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) polymerase chain reaction (PCR) test within 30 days before Baseline, history of hospitalization for COVID-19, or history of use of oxygen due to COVID-19. Note that previous COVID-19 infection alone is not exclusionary and vaccination against SARS-CoV-2 is allowed, but must be documented.
28. Any other condition which, in the opinion of the Investigator, would impede compliance, hinder completion of the study, compromise the well-being of the subject, or interfere with the study outcomes.

Investigational product, dosage and mode of administration:

TVB-2640 is supplied by the Sponsor as 25 and 50 mg strength tablets.

Subjects randomly assigned to TVB-2640 will receive study drug PO QD. Each QD dose of study drug is to be taken at the same time of day with food; the study drug dose is to be taken in the morning on Days 1 to 8 and then in the evening on each day thereafter, with each dose separated by 24 hours (±4 hours). (The Day 8 and 9 doses will be separated by 36 hours.)

Duration of treatment:

Subjects will receive treatment with TVB-2640 or placebo daily for 12 weeks according to their cohort and treatment assignment.

Reference therapy, dosage and mode of administration:

Subjects randomly assigned to placebo will receive placebo tablets PO QD under the same conditions and frequency as described for TVB-2640.

Criteria for evaluation:

Efficacy:

Efficacy variables include hepatic fat fraction by MRI-PDFF (primary) as well as measurement of liver aminotransferases, lipid parameters, markers of NASH and liver fibrosis, and an eicosanoid panel. Metabolic and anthropometric parameters will be assessed as exploratory measures.

[redacted]

Safety:

Safety variables to be assessed include safety laboratory tests (hematology, clinical chemistry, and urinalysis), vital signs and anthropometrics, 12-lead ECGs, vital sign, physical examination findings, including ophthalmologic examination findings, AEs, and concomitant medications.

Sample size:

The sample size is based on a fixed-sequence strategy which tests for a treatment difference in the primary endpoint (percent change from baseline in liver fat at Week 12, as determined by MRI-PDFF) between each randomized TVB-2640 dose group (25 mg and 50 mg) versus placebo. The fixed-sequence strategy will start with the highest dose group comparison. If the high dose comparison is statistically significant, then testing will proceed to the low dose group comparison. Each randomized TVB-2640 dose group will be compared to the pooled placebo group using an F-test test from an analysis of covariance (ANCOVA) model with fixed effects for diabetes status, treatment group, and baseline MRI-PDFF value as a covariate. The non-parametric Wilcoxon rank-sum test is conservatively used, instead of the F-test, for power calculations.

The sample size is based on a conservative assumption that the primary analysis is going to be performed in the US population only, resulting in 30 evaluable subjects in the placebo group, and 30 evaluable subjects in each of the randomized TVB-2640 dose levels. Based on Patel (2016), it is conservatively assumed that the primary endpoint has a standard deviation of 30. Power calculations assume the primary endpoint is lognormally distributed with a standard deviation of 30, there are at least 30 evaluable subjects in each treatment group, and the two-sided Wilcoxon rank sum test will be used to test each pairwise treatment difference at the 0.05 Type I error level. Under the fixed-sequence strategy which maintains an overall 0.05 Type I error rate, the study has at least 80% overall power to detect both treatment differences (i.e., both high dose [50 mg] versus placebo and low dose [25 mg] versus placebo), if each mean treatment difference is at least 24. If the study does not proceed to the high-dose level, (i.e., there are 30 evaluable subjects treated with 25 mg TVB-2640 and only 15 with placebo), then the study has at least 77% power to detect a mean treatment difference of at least 24. The study will have more power if the subjects from China are included in the primary analysis and the same assumptions of treatment difference and variability hold.

Statistical methods:

The modified intent-to-treat population (mITT) is defined as all subjects who are randomized, receive study drug for at least 8 weeks, and have a baseline and at least 1 post-baseline MRI-PDFF assessment on or after Week 8. The mITT is the primary population for analysis of MRI-PDFF assessments. The intent-to-treat population (ITT) is defined as all subjects who are randomized and received at least 1 dose of study drug. The ITT will be used for analysis of secondary efficacy endpoints and supportive and sensitivity analyses of MRI-PDFF assessments.

A test for country-by-treatment interaction with respect to the primary efficacy endpoint, percent change from baseline in hepatic fat fraction at Week 12, as determined by MRI-PDFF, will be performed in the 50 mg cohort. An ANCOVA model with fixed effects for the stratification factor (diabetes presence/absence), country (US/China), treatment group (TVB-2640 50 mg and pooled placebo), and country-by-treatment interaction and with the baseline MRI-PDFF value as a covariate including all subjects from US and China will be performed. If there is no interaction detected ($p \geq 0.05$), the primary efficacy endpoint will be analyzed using an ANCOVA model with fixed effects for the stratification factor (diabetes presence/absence) and treatment group (i.e., randomized TVB-2640 dose groups and pooled placebo) and with the baseline MRI-PDFF value as a covariate including all subjects from US and China. If the country-by-treatment interaction is significant ($p<0.05$) then the primary analysis will be conducted on US subjects and will maintain sufficient power with the 90 evaluable subjects in the US (30 active in each cohort and 30 combined placebo). A separate analysis would be performed on subjects from China in the 50 mg cohort. This analysis is not powered to detect a significant difference but would be supportive/descriptive in nature to evaluate trends.

The primary efficacy analyses will be based on an F-test from the ANCOVA model in the mITT to compare each randomized TVB-2640 dose group individually with the placebo group. The fixed-

sequence method, as described below, will be used to maintain an overall 0.05 Type I error rate. Analyses with transformed data or rank-based methods may be substituted if required to meet model assumptions. Summary statistics will be displayed by treatment group along with the difference in least squares means (and the associated 95% confidence interval and p-value) for each randomized TVB-2640 dose group to pooled placebo comparison.

Differences between each randomized TVB-2640 dose group (25 mg, 50 mg) and pooled placebo for the key secondary efficacy endpoint, the percentage of subjects achieving a 30% or greater reduction of liver fat as determined by MRI PDFF (i.e., a response), will be analyzed using Cochran-Mantel-Haenszel (CMH) methods. The key secondary efficacy analyses will be based on a CMH test in the mITT to compare each randomized TVB-2640 dose group with placebo, adjusting for the stratification factor (diabetes presence/absence). If a country-by-treatment interaction is found in the primary efficacy endpoint, the subjects enrolled in the US and in China in the 50 mg cohort may be analyzed separately for the key secondary efficacy endpoint and analysis of the subjects from China will be viewed as supportive/descriptive. The estimated response rates and the corresponding exact 95% confidence interval based on a binomial distribution will be calculated for each treatment group. The exact 95% confidence limit for the CMH estimate of the common risk difference will also be obtained.

The primary and key secondary efficacy analyses will be tested using a fixed-sequence strategy to maintain the overall Type I error rate at 0.05. Each test in the fixed-sequence, uses a 2-sided test at the 0.05 level of significance and starts with the primary efficacy analyses. The primary efficacy analyses start with a comparison of the highest randomized dose-group (50 mg) to placebo and if statistically significant, testing will proceed to the low dose group comparison. If both comparisons of the primary efficacy endpoint are significant, then testing will proceed to the key secondary efficacy endpoint. The key secondary efficacy analyses similarly start with a comparison of the highest dose-group with placebo and if statistically significant, testing will proceed to the low dose group comparison.

Missing Week 12 values for MRI-PDFF, in the primary and key secondary analyses, will be imputed with the last post-baseline observation on or after Week 8 in the mITT. Secondary and sensitivity analyses using other methods for missing data and in different analysis populations will be described in the Statistical Analysis Plan (SAP). For other continuous secondary efficacy endpoints measured at multiple post-baseline visits, the changes at 12 weeks will be summarized for each treatment group and compared between each randomized TVB-2640 dose group and pooled placebo using a linear mixed-effects model for repeated measures in the ITT. The model will include the stratification factor (diabetes presence/absence), treatment group, visit, and treatment-by-visit interaction as the fixed effects, and baseline value as a covariate. The point estimates for the least-squares mean of the pairwise difference for each randomized TVB-2640 dose group and pooled placebo comparison at each visit and the corresponding 95% confidence interval and 2-sided p-value will be summarized. Additional details for handling of data from subjects enrolled in the US and in China will be described in the SAP.

Similarly, a generalized linear mixed-effects model for repeated measures based on a logit link function will be used for comparing other categorical efficacy endpoints or derived categorical response rates for continuous endpoints at 12 weeks between each randomized TVB-2640 dose group and pooled placebo group in the ITT.

No formal statistical testing will be conducted for the safety analyses.

Data from Cohort 3 will be summarized descriptively. Further details will be provided in the SAP. Fewer samples will be collected for potential biomarker assays in the 75 mg cohort, as outlined in the Schedule of Events.

Subset analyses will be performed on all safety and efficacy endpoints for subjects enrolled in China to evaluate the consistency of results between subjects enrolled in each country.

Schedule of events:
The schedule of events is presented in Table 2.

Table 2 Schedule of Events

Evaluation [redacted]	[redacted]	Study Visit [redacted]							
	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Screening/baseline assessments									
Informed consent	X								
Inclusion and exclusion criteria review	X	X							
Demographics	X								
Medical and social history	X								
Screening/baseline pregnancy test	X[3]	X[3]							
Alcohol breath test or blood alcohol test	X	X							
Confirmation of NASH diagnosis	X[4,5]								
Screening serology	X[6]								
Height, BMI, waist and hip circumference, and waist-hip ratio	X								
Safety Assessments									
ECG	X	X[7]	X	X	X	X	X	X	X
Vital signs[8]	X	X		X	X	X	X	X	X
Complete physical examination	X						X	X	

Evaluation	Study Visit								
	Screening (Day -60 to -1)	**Baseline (Day 1)[1]**	**Day 2**	**Week 2**	**Week 4**	**Week 8**	**Week 12[2]**	**Early Termination Visit[2]**	**Week 16 (Follow-up Visit)[2]**
Abbreviated physical examination, including examination of focused liver signs, skin, and extremities		X		X	X	X			X
Ophthalmologic examination[9]	X				X	X	X	X	X
Safety Laboratory tests									
Hematology and coagulation studies[10]	X	X		X	X	X	X	X	X
Liver aminotransferases[11]	X[12]	X		X	X	X	X	X	X
Clinical chemistry[13]	X	X		X	X	X	X	X	X
Urinalysis[14]	X	X					X	X	
Urine pregnancy testing[15]				X	X	X	X	X	
Adverse events		AEs occurring from screening (i.e., after the ICF is signed) through 28 days after the last study drug dose are to be documented.							
Prior / concomitant medications	X	All medications taken within 28 days prior to the first study drug dose through 28 days after the last study drug dose are to be documented.							
Efficacy, PD, and PK assessments									
MRI-PDFF	X[16]						X[17]	X[17]	X[17] *(Cohorts 1 & 2 Only)*
Weight	X	X		X	X	X	X	X	X
Waist and hip circumference and waist-hip ratio		X					X	X	

Evaluation		Study Visit							
	Screening (Day -60 to -1)	**Baseline (Day 1)[1]**	**Day 2**	**Week 2**	**Week 4**	**Week 8**	**Week 12[2]**	**Early Termination Visit[2]**	**Week 16 (Follow-up Visit)[2]**
MRE (Cohorts 1 and 2 only)	X[16,18]								
MRI / FibroScan™		X[16]					X	X	
Blood sample collection for:									
Lipid panel[19]	X	X		X	X	X	X	X	X
HbA1c	X	X					X	X	
Metabolic panel[20]		X		X	X	X	X	X	X
NASH / fibrosis biomarkers[21]		X					X	X	
Eicosanoid panel *(Cohorts 1 and 2 only)*[22]		X			X		X	X	
Inflammatory biomarkers (TNFα, IL-6, IL-8, Hs-CRP, MCP-1) *(Cohorts 1 and 2 only)*[22]		X			X		X	X	
Adiponectin, resistin, leptin *(Cohorts 1 and 2 only)*[22]		X			X		X	X	
Lipoprotein particle size analysis (NMR) *(Cohorts 1 and 2 only)*[22]		X					X	X	
Lipidomic analyses for DNL		X			X *(Cohort 3 Only)*		X	X	
Genomics pertaining to the disease under study[23]		X							
[redacted]		[redacted]	[redacted]	[redacted]					

Evaluation	[redacted]	Study Visit							
	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]	[redacted]
Exploratory PD biomarkers *(Cohorts 1 and 2 only)*[22]		X					X	X	
Lymphocyte subsets in peripheral blood mononuclear cells *(Cohorts 1 and 2 only)*[22,26]		X					X		
Sample for storage[22,27]		X					X	X	
Spot urine sample collection for:									
Exploratory PD biomarkers[22]		X					X	X	
Study Drug Administration									
Randomization		X[28]							
Study drug dispensation		X			X	X			
Study drug administration		QD PO from Day 1 through Week 12							
Study drug accountability				X	X	X	X	X	

Table footnotes appear on the following page.

1 Baseline evaluations are to be performed pre-dose, unless otherwise specified.

2 At the time of this amendment, subject participation in the 25 mg cohort is complete, with participation in the 50 mg cohort fully enrolled but with subject participation ongoing. The Sponsor notes that the COVID-19 pandemic may impact the conduct of the 50 and 75 mg cohorts, with challenges potentially arising from quarantines and travel limitations affecting study subjects as well as clinic and imaging facility closures. Accordingly, safety assessments scheduled for the Week 12 and 16 visits, including sample collection for clinical laboratory tests, physical examinations, and documentation of AEs, may be performed at an alternate study center or by home health care visit. Furthermore, if the subject is unable to attend the Week 12 visit at the study center, one MRI-PDFF may be performed at any time between Week 11 and 16. If this scan is performed after Week 12, the subject must continue to receive study drug until the final scan is performed; however, study drug is not to be continued for >16 weeks. The safety of subjects who continue dosing beyond 12 weeks will be assessed during this continued dosing period by planning for blood sample collection at an alternate site or home visit at Week 12 and blood sample collection and physical examination on the last day of dosing. A 4-week off treatment safety assessment is to occur after the last dose, regardless of whether a subject receives treatment for 12 or 16 weeks.

For subjects in Cohort 3, Week 16 assessments may serve as screening assessments if within the window before the Day 1 visit.

3 Serum beta-human chorionic gonadotropin (β-HCG) pregnancy testing is to be performed for all female subjects of child-bearing potential during screening and serum or urine β-HCG pregnancy testing is to be performed within 48 hours before first dose on Day 1; subjects with positive results are not eligible for study participation. Pregnancy testing is to be repeated during the study any time pregnancy is suspected; study drug is to be discontinued for any subject with a positive result.

4 Results of liver biopsy conducted within 24 months before randomization, with findings consistent with NASH diagnosis, must be collected and documented.

5 If prior biopsy was not performed or is not available, subjects must have a suspected diagnosis of NASH based on clinical presentation/historical assessments (see protocol Section 8.2) and must have MRE ≥2.5 kpa (Cohorts 1 and 2 only) and MRI-PDFF ≥8% during screening.

6 Screening serologies include HIV RNA, HBsAg, anti-HCV, and HCV RNA. Subjects with positive anti-HCV who test negative for HCV RNA at screening will be allowed to participate in the study. A PCR test for SARS-Cov-2 (the virus that causes COVID-19) also is to be performed; subjects with positive results are not eligible for study participation.

7 ECGs are to be performed before and 4 hours after the first study drug dose on Day 1.

8 Vital signs include oral temperature and supine pulse, systolic/diastolic blood pressure, and respiration rate. Measurements are to be made after the subject has been resting in a supine position for a minimum of 5 minutes.

9 Slit lamp and near and far visual acuity testing is to be performed. Baseline examinations may be performed within 28 days before Day 1. If a subject experiences a treatment-emergent ophthalmologic abnormality, an ophthalmologic examination should be performed within 24 to 48 hours or next business day after symptom onset to evaluate the abnormality.

10 Hematology parameters include hemoglobin, hematocrit, platelet count, red blood cell (RBC) count, white blood cell (WBC) count with differential, and absolute neutrophil count (ANC), and prothrombin time (PT), international normalized ratio (INR), and activated partial thromboplastin time (aPTT) and fibrinogen.

11 Liver aminotransferases include AST and ALT. Liver aminotransferases collected as part of the clinical chemistry panel for safety are also applicable for efficacy assessments.

12 Baseline liver aminotransferases are to be the mean of at least 2 values taken at least 4 to 12 weeks apart; the first measurement may be a historical measurement.

13 Fasting clinical chemistries include chloride (Cl), carbon dioxide (CO_2), sodium (Na), potassium (K), blood urea nitrogen (BUN), calcium, magnesium, creatinine, albumin, alkaline phosphatase (ALP), total bilirubin, indirect and direct bilirubin, and total protein. Creatine phosphokinase (CPK) (total and fractionated) is to be calculated during screening and at baseline.

14 Urinalysis includes specific gravity, pH, blood, glucose, protein, ketones, and microscopic examination of sediment.

15 For female subjects of child-bearing potential only; subjects with positive results are to discontinue study drug immediately.

16 Baseline imaging studies may be performed within 28 days before Day 1.

17 Due to the COVID-19 pandemic, if the subject is unable to attend both the Week 12 and Week 16 visit, one MRI-PDFF may be performed at any time between Week 11 and 16. If this scan is performed after Week 12, the subject must continue to receive study drug until the final scan is performed. However, study drug is not to be continued for >16 weeks.

18 Note that subjects without a biopsy performed within 24 months of randomization MUST have MRE performed during screening.

19 Fasting lipid panel includes LDL-C, non-LDL-C, HDL-C, non-HDL-C, total cholesterol, triglycerides, ApoB, and Lp(a) particles.

20 Fasting metabolic panel includes glucose, insulin, NEFA, and GGT. HOMA2-IR and adipo-IR are to be calculated from fasting insulin, fasting glucose, and fasting NEFA, as applicable.
21 NASH and fibrosis biomarkers include CK-18, FIB-4 Index, ELF (TIMP-1, PIIINP, HA), PROC3, and FIBROSpect 2. For Cohort 3 (75 mg), only PROC3 sample will be collected.
22 For Cohort 3 (75 mg), these samples will not be collected as dedicated blood collections. Instead, assays for some of these parameters may be conducted retrospectively using additional aliquots collected from the "NASH/fibrosis markers sample" or from the "lipidomic analyses for DNL sample".
23 The genomics sample collection is optional for Cohort 3 (75 mg).
24 [redacted]
25 [redacted]
26 Blood samples are to be collected from a subset of subjects enrolled at selected study centers at baseline and Week 12 for exploratory evaluation of change in CD4/Th17/regulatory T-cell populations and, potentially, natural killer-T and epigenetic or activation changes in peripheral blood mononuclear cells.
27 Additional blood samples will be collected at specified time points and stored for potential future analysis for biomarkers relevant to NAFLD/NASH. These samples will neither be used for genetic analysis nor creation of cell lines.
28 Randomization may be performed within 24 hours before baseline.

3. TABLE OF CONTENTS, LIST OF TABLES, AND LIST OF FIGURES

TABLE OF CONTENTS

LIST OF TABLES

4. LIST OF ABBREVIATIONS AND DEFINITIONS OF TERMS

Abbreviation or Specialist Term	Explanation
15-HETE	15-LOX–derived 15-hydroxyeicosatetraenoic acid
ACC	Acetyl-CoA carboxylase
Adipo-IR	Adipose tissue insulin resistance
AE	Adverse event
ALT	Alanine aminotransferase
ANCOVA	Analysis of covariance
ANOVA	Analysis of variance
ApoB	Apolipoprotein B
APRI	AST to platelet ratio
AST	Aspartate aminotransferase
ATP III	Adult Treatment Panel III
BMI	Body mass index
CAP	Controlled attenuation parameter
CFDA	China Food and Drug Administration
CK-18	Cytokeratin-18
CMH	Cochran-Mantel-Haenszel
CoA	Coenzyme A
COVID-19	Coronavirus disease-2019
CRA	Clinical research associate
CTCAE	Common Terminology Criteria for Adverse Events
CYP3A4	Cytochrome P450 3A4
DILI	Drug-induced liver injury
DLT	Dose-limiting toxicity
DNL	De novo lipogenesis
ECG	Electrocardiogram
eCRF	Electronic case report form
EDC	Electronic data capture
ELF	Enhanced liver function
ELISA	Enzyme-linked immunosorbent assay
FASN	Fatty acid synthase
FIB-4	Fibrosis-4

Abbreviation or Specialist Term	Explanation
GCP	Good Clinical Practice
GGT	Gamma-glutamyl transpeptidase
GLP	Good Laboratory Practice
HbA1c	Glycated hemoglobin
HBsAg	Hepatitis B surface antigen
HCV	Hepatitis C virus
HDL-C	High-density lipoprotein cholesterol
HOMA2-IR	Homeostatic model assessment insulin resistance
Hs-CRP	High-sensitivity C-reactive protein
ICH	International Council for Harmonisation
IL	Interleukin
IL-6	Interleukin 6
INR	International normalized ratio
IRB	Institutional Review Board
IST	Investigator-sponsored trial
ITT	Intent-to-treat population
IUD	Intrauterine device
LDL-C	Low-density lipoprotein cholesterol
Lp(a)	Lipoprotein(a)
LTB4	5-LOX–derived leukotriene B4
LXA4	Lipoxin A4
MCP-1	Monocyte chemoattractant protein-1
mITT	Modified intent-to-treat population
MRE	Magnetic resonance elastography
MRI-PDFF	Proton density fat fraction by magnetic resonance imaging
MRS	Magnetic resonance spectroscopy
MTD	Maximum tolerated dose
NADPH	Nicotinamide adenine dinucleotide phosphate
NAFLD	Non-alcoholic fatty liver disease
NAS	NAFLD activity score
NASH	Non-alcoholic steatohepatitis
NCI	National Cancer Institute

Abbreviation or Specialist Term	Explanation
NEFA	Non-esterified fatty acid
NMR	Nuclear magnetic resonance
NOAEL	No observed adverse effect level
OAD	oral anti-diabetic
PCR	Polymerase chain reaction
PD	Pharmacodynamic
PGE2	prostaglandin E2
PK	Pharmacokinetic
PO	Orally
PPE	Palmar-plantar erythrodysesthesia
QD	Once daily
RNA	Ribonucleic acid
SAP	Statistical analysis plan
SARS-CoV-2	Severe acute respiratory syndrome coronavirus-2
SRC	Safety Review Committee
TEAE	Treatment-emergent adverse event
TG	Triglycerides
TNFα	Tumor necrosis factor-alpha
ULN	Upper limit of normal
US	United States
US FDA	United States Food and Drug Administration
VCTE	Vibration-controlled transient elastography
β-HCG	Beta-human chorionic gonadotropin

5. INTRODUCTION

Fatty acid synthase (FASN) is a key enzyme in the de novo lipogenesis (DNL) pathway and catalyzes the biosynthesis of palmitate from acetyl-coenzyme A (CoA) and malonyl-CoA substrates, which can then undergo further modifications into other fatty acids and complex lipids. Because FASN catalyzes the last step in the fatty acid biosynthetic pathway, it is believed to be a determinant of the maximal liver capacity to synthesize fatty acids by DNL (Postic, 2008).

Dysregulation of FASN activity is found in a number of different disease states including cancer (Buckley, 2017) and metabolic diseases.

Metabolic diseases spanning the continuum of metabolic syndrome, non-alcoholic fatty liver disease (NAFLD) and the more advanced disease of non-alcoholic steatohepatitis (NASH) can progress to significant liver diseases, including cirrhosis and hepatocellular carcinoma. In addition, NAFLD is associated with increased comorbidities, including cardiovascular diseases (Widya et al. 2016; Targher, Day, and Bonora 2010) and type 2 diabetes (Cusi 2016). Obesity and the metabolic syndrome are two key risk factors for NAFLD, which are characterized as an imbalance in energy utilization and storage (Grundy, 2005). This imbalance may lead to dysregulated metabolic pathways and inflammatory responses that drive further changes leading to liver damage and comorbid conditions. Over the next 5 years, NASH will overtake hepatitis as the most prevalent liver disease in the United States (US).

Normal responses to feeding carbohydrate in meals include a transient increase in hepatic DNL, a FASN-dependent pathway, followed by a return to baseline levels upon fasting. In subjects with NAFLD, however, hepatic DNL is increased (Donnelly et al. 2005; Lambert et al. 2014), and FASN gene expression has been shown to be elevated in liver biopsies from NAFLD patients (Kohijimi, 2007; Mitsuyoshi, 2009), contributing to elevated liver stores of triglycerides (TG) and saturated fatty acid species, which in turn may contribute to liver inflammation (Wei 2006), tissue damage, and fibrosis.

There are currently no drugs licensed for the treatment of NASH. Inhibiting lipogenesis, with an FASN inhibitor for example, may be a viable clinical strategy to address NASH. Pharmacological inhibition of FASN with TVB-3664, a chemical surrogate of TVB-2640, improved hepatic steatosis, inflammation and fibrosis in a diet-induced mouse model of NASH (Duke 2017). Moreover, studies in NASH subjects have shown that inhibition of DNL with acetyl-CoA carboxylase (ACC) inhibitor GS-0976 (Stiede, 2017) results in a significant reduction of intrahepatic fat and surrogate markers of fibrosis and cell death (Loomba, 2017). Also, pioglitazone use has been shown to improve advanced fibrosis in NASH (Musso, 2017), which may be associated with a reduction in DNL (Beysen, 2008). Further studies are needed, however, to evaluate the full spectrum of treatment effects elicited by these drugs and management of their side-effects.

Sagimet Biosciences (formerly 3-V Biosciences) has developed an oral, once-daily (QD) FASN inhibitor, TVB-2640. This drug has been evaluated in approximately160 humans to date including patients with solid tumors and healthy adults, some of whom had characteristics of metabolic syndrome. The safety, pharmacokinetic (PK) and pharmacodynamic (PD) parameters of TVB-2640 have been measured. Preliminary results demonstrated that once-daily, oral administration of TVB-2640 for 10 days decreased hepatic DNL in a dose-dependent manner and improved other markers of metabolic dysfunction in otherwise healthy subjects with

characteristics of the metabolic syndrome (Study 2006432, Syed-Abdul, 2019). These observations form the basis for evaluating the TVB-2640 as a novel therapy for subjects with NASH.

5.1. TVB-2640

The clinical safety profile to date for TVB-2640 is based primarily on one completed uncontrolled open-label Phase 1 study in humans (Study 3V2640-CLIN-002) in 136 patients with solid tumors, 76 treated with monotherapy (at doses ranging from 60 mg/m^2 to a dose of 240 mg/m^2 (i.e., up to 600 mg) and 60 treated with TVB-2640 in combination with a taxane (paclitaxel or docetaxel).

Overall, TVB-2640 demonstrated a favorable tolerability profile. Most treatment-emergent adverse events (TEAEs) were Grade 1 or 2 in intensity, non-serious, and manageable.

As anticipated, based on nonclinical study findings, the principal toxicities associated with TVB-2640 as monotherapy were skin and ocular effects. With monotherapy, most skin events were Grade 1 or 2 in intensity, and all were non-serious.

The maximum tolerated dose (MTD) of TVB-2640 monotherapy in patients with solid tumors was determined to be 100 mg/m^2. Dose-limiting toxicities (DLTs) associated with TVB-2640 monotherapy were skin and ocular events, including corneal edema, keratitis, and palmar-plantar erythrodysesthesia (PPE) syndrome.

As monotherapy, the most common individual TEAEs were alopecia (61%), PPE syndrome (46%), fatigue (37%), decreased appetite (26%), and dry skin (22%).

When TVB-2640 was administered in combination with a taxane (paclitaxel or docetaxel), skin and ocular effects also were common. Most skin events were Grade 1 or 2 in intensity, and all but 1 skin event were non-serious.

As was the case with monotherapy, the MTD of TVB-2640 in combination with paclitaxel was determined to be 100 mg/m^2. DLTs associated with TVB-2640 in combination included PPE syndrome and uveitis. The most common TEAEs with TVB-2640 in combination with paclitaxel were fatigue (53%), alopecia (46%), PPE syndrome (46%), nausea (40%), and peripheral neuropathy (36%). Peripheral neuropathy is known to be associated with paclitaxel.

When TVB-2640 was administered in combination with paclitaxel, 6 episodes of serious pneumonitis were experienced by 5 patients, with all assessed by the Investigator as related to both TVB-2640 and paclitaxel. (No cases of pneumonitis were reported among monotherapy patients.) Given the occurrence of these events, respiratory events were identified as adverse events of special interest for patients receiving TVB-2640 in combination.

Overall, among patients with solid tumors treated with TVB-2640, no apparent trends were seen over time or across TVB-2640 dose groups with regard to change from baseline in clinical laboratory or vital sign parameters. Furthermore, based on review of electrocardiogram (ECG) and Holter monitoring data, TVB-2640 was shown to have no clinically relevant QTc prolonging effect.

A total of 12 subjects with features of the metabolic syndrome have been treated with TVB-2640 at flat doses of 50 to 150 mg QD orally (PO) for 10 days in an Investigator-sponsored trial (IST).

TVB-2640 was well tolerated in this subject population. All TEAEs were Grade 1 in intensity and non-serious and none led to study drug discontinuation. The only TEAEs reported for >1 subjects were alopecia and PPE syndrome (each 2 subjects). In this study, QD PO administration of TVB-2640 for 10 days was shown to decrease hepatic DNL in a dose-dependent manner and improved other markers of metabolic dysfunction in this population of otherwise healthy subjects with characteristics of the metabolic syndrome. These observations form the basis for evaluating the TVB-2640 as a novel therapy for subjects with NASH.

Refer to the Investigator's Brochure for more detailed information regarding TVB-2640.

6. OBJECTIVES AND PURPOSE

6.1. Primary Objectives

The primary objectives are:

- To determine the effect of QD TVB-2640 for 12 weeks versus placebo on the change in hepatic fat fraction by proton density fat fraction by magnetic resonance imaging (MRI-PDFF) from baseline in subjects with NASH.
- To determine the safety of QD TVB-2640 versus placebo in subjects with NASH, including the effects on alanine aminotransferase (ALT) and aspartate aminotransferase (AST).

6.2. Secondary Objective

The secondary objective is:

- To determine the effect of QD TVB-2640 for 12 weeks versus placebo in subjects with NASH on:
  - Lipid and lipoprotein parameters, including low-density lipoprotein cholesterol (LDL-C), non-LDL-C, high-density lipoprotein cholesterol (HDL-C), non-HDL-C, total cholesterol, TG, apolipoprotein B (ApoB), and lipoprotein(a) (Lp[a]) particles.
  - NASH and fibrosis biomarkers including cytokeratin-18 (CK-18), fibrosis-4 (FIB-4), and enhanced liver function (ELF) or FIBROSpect 2 or ProC3 test.
  - Eicosanoid panel.

6.3. Exploratory Objectives

The exploratory objectives are:

- To determine the effect of QD TVB-2640 for 12 weeks versus placebo in subjects NASH on clinical measures, including:
  - Liver fibrosis, as determined by vibration-controlled transient elastography (VCTE).
  - Metabolic parameters, including fasting glucose, insulin, homeostatic model assessment insulin resistance (HOMA2-IR), HbA1c, non-esterified fatty acid (NEFA), adipose tissue insulin resistance (adipo-IR), adiponectin, resistin, Interleukin 6 (IL-6), and gamma-glutamyl transpeptidase (GGT).
  - Anthropometric parameters, including weight, waist and hip circumference, waist-hip ratio, and blood pressure.
  - Lipidomic analyses for DNL.
- Explore the relationship of plasma drug exposure to changes in efficacy and safety biomarkers.

- Explore possible relationships between genomic markers of NASH and responses to treatment.

6.4. Endpoints

6.4.1. Efficacy Endpoints

The primary efficacy endpoint is:

- Percent change from baseline in liver fat at Week 12, as determined by MRI-PDFF.

The key secondary efficacy endpoint is:

- The percentage of subjects with at least a 30% reduction in liver fat at Week 12, as determined by MRI-PDFF.

Additional efficacy endpoints are:

- Percent change from baseline in liver fat at Week 16, as determined by MRI-PDFF.
- Percentage of subjects with at least a 30% reduction in liver fat at Week 16, as determined by MRI-PDFF.
- Change from baseline over the 12-week treatment period in:
  - Liver aminotransferases.
  - Lipid and lipoprotein parameters.
  - NASH and fibrosis markers.
  - Eicosanoids.

Exploratory efficacy endpoints are:

- Change from baseline over the 12-week treatment period in:
  - Liver fibrosis, as determined by VCTE.
  - Metabolic parameters.
  - Anthropometric parameters.
  - Lipidomic analyses for DNL.
- Proportion of subjects with <5% liver fat (normalized) at Week 12.

6.4.2. Safety Endpoints

Safety endpoints are:

- Proportion of subjects experiencing treatment-emergent adverse events (TEAEs), Grade 3 or 4 TEAEs, and serious adverse events (SAEs).
- Change from baseline in vital sign measurements, 12-lead electrocardiogram (ECG) findings, and clinical laboratory test results.
- Proportion of subjects experiencing treatment-emergent ophthalmologic abnormalities.

7. INVESTIGATIONAL PLAN

7.1. Overall Study Design

This is a multi-center, randomized, single-blind, placebo-controlled, dose-escalation study to evaluate the safety and efficacy of TVB-2640 in subjects with NASH. Male and female subjects aged ≥18 years with either biopsy-proven NASH within 2 years before randomization and MRI-PDFF ≥8% or, if prior liver biopsy was not performed or results are not available, magnetic resonance elastography (MRE) ≥2.5 kpa (Cohorts 1 and 2 only) and MRI-PDFF ≥8% during screening are planned to be enrolled. Approximately 117 unique subjects will be enrolled and randomized, with at least 90 subjects enrolled in the US and approximately 27 evaluable subjects enrolled in China (18 active and 9 placebo). In the US, subjects will be enrolled and randomized to achieve 90 evaluable subjects (30 active and 15 placebo in each of 2 dose cohorts). The 50 mg cohort will enroll and randomize approximately 27 additional subjects from China. The US-only 75 mg cohort will enroll approximately 10 active, evaluable subjects who previously received placebo in the 25 or 50 mg cohort. A subject is defined as evaluable if they receive study drug for at least 8 weeks and have a baseline and at least 1 post-baseline MRI-PDFF assessment on or after Week 8.

Subjects will be screened for study eligibility within 60 days before randomization. Subjects must meet all of the inclusion criteria and none of the exclusion criteria to participate in the study. Note that subjects who initially fail to meet all entrance criteria based on screening assessments (i.e., screen failures) may be submitted for re-screening once (see Section 8.5 for details).

Subjects who are determined to be eligible for the study, based on screening assessments, are to be randomized into the study within 24 hours before baseline (Day 1, the first day of study drug administration). Subjects who are randomized are considered to be enrolled in the study. Initially, 2 TVB-2640 dose levels are planned to be evaluated in a sequential fashion: 25 mg and, if study suspension criteria are not met at the 25 mg dose level (see Section 7.2), 50 mg. At each of the 2 initial dose levels, subjects will be randomly assigned to TVB-2640 or placebo at a 2:1 ratio, with randomization stratified by type 2 diabetes mellitus status. The 50 mg dose level will also be stratified by country:

- 25 mg dose level: TVB-2640 (Planned N=30) and Placebo (Planned N=15)
- 50 mg dose level: US: TVB-2640 (Planned N=30) and Placebo (Planned N=15); China: TVB-2640 (Planned N=18) and Placebo (Planned N=9); Total: TVB-2640 (Planned N=48) and Placebo (Planned N=24)

Subjects will receive TVB-2640 or placebo QD PO for 12 weeks, or until the time of the final on-treatment MRI-PDFF if after Week 12, but for no longer than 16 weeks. During the 12-week Treatment period, subjects are to attend study center visits at Week 1, Days 1 and 2, and Weeks 2, 4, 8, and 12. After completion of the 12-week Treatment period, subjects are to attend a Follow-up visit at Week 16 for post-treatment safety and efficacy assessments.

All subjects must complete the 12-week treatment period at the 25 mg dose level, with review of all safety data and written approval by the Safety Review Committee (SRC) and reviewed by the

Food and Drug Administration (FDA) before enrollment and treatment of subjects at the 50 mg dose level may commence.

After completion of Cohorts 1 and 2, and again if no stopping criteria are met upon review by the Independent SRC, subjects assigned to placebo in either cohort are eligible to crossover to receive TVB-2640 75 mg in Open-label Cohort 3 (planned N=12 in order to obtain 10 evaluable subjects). Cohort 3 will be conducted at select sites only.

The Sponsor notes that the coronavirus disease-2019 (COVID-19) pandemic may impact the conduct of the 50 mg and 75 mg cohorts, with challenges potentially arising from quarantines and travel limitations affecting study subjects as well as clinic and imaging facility closures. Accordingly, safety assessments scheduled for the Week 12 and 16 visits, as identified in Table 2, including sample collection for clinical laboratory tests, physical examinations, and documentation of adverse events (AEs), may be performed at an alternate study center or by home health care visit. Furthermore, if the subject is unable to attend the Week 12 visit at the study center, one MRI-PDFF may be performed at any time between Week 11 and 16. If this scan is performed after Week 12, the subject must continue to receive study drug until the final scan is performed; however, study drug is not to be continued for >16 weeks. The safety of subjects who continue dosing beyond 12 weeks will be assessed during this continued dosing period by planning for blood sample collection at an alternate site or home visit at Week 12 and blood sample collection and physical examination on the last day of dosing. A 4-week off treatment safety assessment is to occur after the last dose, regardless of whether a subject receives treatment for 12 or 16 weeks.

In Cohorts 1 and 2, study drug will be dispensed to subjects in a single-blinded fashion. All other study personnel, with the exception of personnel conducting/interpreting imaging studies (see Section 11.1) at each study center, will be unblinded to the subject's treatment assignment. In Cohort 3, study drug will be dispensed in an open-label fashion.

Efficacy variables include MRI-PDFF as well as measurement of liver aminotransferases, lipid parameters, and markers of NASH and liver fibrosis. The primary endpoint is percent change from baseline in liver fat from baseline at Week 12, as determined by MRI-PDFF. The percentage of subjects with at least a 30% reduction in the primary endpoint is a key secondary efficacy endpoint.

In Cohorts 1 and 2, subjects will undergo MRI-PDFF prior to the start of treatment (pre-dose), at the end of the initial 12-week Treatment Period (between 11 and 12 weeks after the start of treatment), and at Week 16. Due to the COVID-19 pandemic, if the subject is unable to attend the Week 12 visit, one MRI-PDFF may be performed at any time between Week 11 and 16. If this scan is performed after Week 12, the subject must continue to receive study drug until the final scan is performed. However, study drug is not to be continued for >16 weeks. In Cohort 3, MRI-PDFF will undergo MRI-PDFF prior to the start of treatment (pre-dose) and at the end of the initial 12 week Treatment Period (between 11 and 12 weeks after the start of treatment).

During the study, safety will be assessed by vital signs (oral temperature, pulse, respiratory rate, and seated blood pressure), 12-lead ECG, physical examination, including ophthalmologic examination, and clinical laboratory testing (hematology, chemistry, and urinalysis). Subjects will be evaluated for AEs and concomitant medication use throughout the study.

Blood samples will be collected at specified times for the assessment of PK.

An SRC will oversee the study to ensure subject safety and to advise if any dosing alterations are recommended. The SRC will review safety including liver-related events (i.e., clinically meaningful elevations in ALT, AST, and bilirubin), and other efficacy (lipid parameters) and safety data as needed.

7.2. Study Suspension Criteria

Treatment of ongoing subjects and enrollment of new subjects at a given dose level will be suspended in the event of any of the following:

- One subject experiences a Grade 5 TEAE (i.e., TEAE with a fatal outcome).
- Two subjects experience the same Grade 4 TEAE.
- Three subjects experience the same Grade 3 TEAE.

In such cases, the SRC will review unblinded data to determine the relationship of the TEAE(s) to TVB-2640 and make a determination whether enrollment and treatment of subjects may be resumed or is to be permanently discontinued.

7.3. Criteria for Study Termination by the Sponsor

The Sponsor reserves the right to terminate the study or a particular study center at any time. If the Sponsor or Investigator discovers conditions arising during the study that suggest the study should be halted, then study termination can occur only after appropriate consultation between the Sponsor and Investigators. Conditions that may warrant study or study center termination include, but are not limited to:

- The discovery of any unexpected, significant, or unacceptable risk to the subjects enrolled in the study.
- Failure of the Investigator to enter subjects at an acceptable rate.
- Insufficient adherence to the protocol requirements.
- A decision on the part of the Sponsor to suspend or discontinue development of study drug.

Should the study be closed prematurely, all study materials (study drug, etc.) must be returned to the Sponsor or designee (or disposed of as directed by the Sponsor or designee).

8. SELECTION AND WITHDRAWAL OF SUBJECTS

8.1. Number of Subjects

Approximately 117 unique subjects will be enrolled and randomized across 2 dosing cohorts (25 and 50 mg). Up to 12 subjects assigned to placebo in Cohorts 1 and 2 will be eligible to crossover and receive TVB-2640 75 mg in an open-label fashion in Cohort 3. The study plans to enroll male and female subjects aged ≥18 years with either biopsy-proven NASH within 24 months before randomization and MRI-PDFF ≥8% or, if prior liver biopsy was not performed or results are not available, MRE ≥2.5 kpa (Cohorts 1 and 2 only) and MRI-PDFF ≥8% during screening.

8.2. Subject Inclusion Criteria

Subjects meeting all of the following criteria are eligible for enrollment in the study.

1. Must be willing to participate in the study and provide written informed consent.
2. Aged ≥18 years with a body mass index (BMI) ≤40 kg/m^2.
3. If a female subject of child-bearing potential, must have a negative serum pregnancy (beta-human chorionic gonadotropin [β-HCG]) test during screening and is not breastfeeding, does not plan to become pregnant during the study, and agrees to use two forms of birth control, one being a barrier method (i.e., condoms, diaphragm, non-hormonal intrauterine device [IUD]) throughout the study and for at least 3 months after the final study drug dose.

   Hormonal contraception (estrogens stable ≥3 months) and hormonal IUDs are permitted if used with a secondary barrier birth control method (e.g., condoms).
4. If a female subject of non-child-bearing potential, must have documentation of surgical sterility (bilateral oophorectomy, hysterectomy, or tubal ligation) or natural sterility (>12 consecutive months without menses).
5. If male, agrees to use adequate birth control throughout the study and for at least 3 months after the final study drug dose.
6. Prior liver biopsy within 24 months of randomization with fibrosis Stage 1 to 3 and a NAFLD activity score (NAS) of ≥4 with at least a score of 1 in each of the following NAS components:

   - Steatosis.
   - Ballooning degeneration.
   - Lobular inflammation.

   AND

   - Confirmation of ≥8% liver fat content on MRI-PDFF.

OR, if prior biopsy is not available:

- Either overweight (BMI ≥25 and <30 kg/m^2) or obese (BMI ≥30 kg/m^2) or diabetic or ALT ≥30 U/L for men or ≥19 U/L for women or fatty liver on ultrasound and at least one more feature of metabolic syndrome by Adult Treatment Panel III (ATP III) criteria.

AND

- MRE ≥2.5 kpa (Cohorts 1 and 2 only) and MRI-PDFF ≥8% during screening.

For Cohort 3 Only

7. Subject may have previously participated in this study in Cohort 1 or 2 and is known to have received placebo by single-blind treatment assignment.

8.3. Subject Exclusion Criteria

Subjects meeting any of the following criteria are not eligible for enrollment in the study.

1. History of significant alcohol consumption for a period of more than 3 consecutive months within 1 year prior to screening, as assessed by the Investigator.

   Note: Significant alcohol consumption is defined as average of >20 g/day in female subjects and >30 g/day in male subjects. For reference, 14 grams of alcohol are contained in the following:

   - 12 fl oz regular beer containing ~5% alcohol.
   - 8-9 fl oz malt liquor containing ~7% alcohol.
   - 5 fl oz of table wine containing ~12% alcohol.
   - 1.5 fl oz of distilled spirits (e.g., gin, rum, vodka, whiskey) containing ~40% alcohol.
2. Inability to reliably quantify alcohol consumption based upon judgment of the Investigator.
3. Use of drugs historically associated with NAFLD (amiodarone, methotrexate, systemic glucocorticoids, tetracyclines, tamoxifen, estrogens at doses greater than those used for hormone replacement, anabolic steroids, valproic acid, and other known hepatotoxins) for more than 2 weeks in the year prior to screening.
4. Weight gain or loss >5% in the 6 months prior to randomization or >10% in the 12 months prior to screening.
5. Prior or planned (during the study period) bariatric surgery (e.g., gastroplasty, roux-en-Y gastric bypass).
6. Type 1 diabetes.
7. Uncontrolled Type 2 diabetes defined as:

   - HbA1c ≥9.5% during screening. (Subjects with HbA1c ≥9.5% may be retested during screening).

- Basal insulin dose adjustment >10% within 60 days prior to enrollment.
- Requirement for glucagon-like peptide analogue or a complex oral anti-diabetic (OAD) regimen (3 or more OADs) within 6 months of screening.
- History of severe hypoglycemia (symptomatic hypoglycemia requiring outside assistance to regain normal neurologic status) within the previous year.

Note: Individual diabetes regimens will be reviewed by Investigator and may be adjusted based on American Diabetes Association guidelines.

8. Presence of cirrhosis on liver biopsy (Stage 4 fibrosis) or imaging (MRE ≥4.67 kpa).
9. Platelet count $<150\times10^9$/L.
10. Clinical evidence of hepatic decompensation as defined by the presence of any of the following abnormalities:
    - Serum albumin ≤3.5 g/dL,
    - International normalized ratio (INR) ≥1.3 (a single retest is permitted if laboratory error is suspected),
    - Total bilirubin >1.8 mg/dL and/or direct bilirubin >0.8 mg/dL (unless due to Gilbert's syndrome, as documented in the subject's medical history/records), or
    - History of esophageal varices, ascites, or hepatic encephalopathy.
11. Evidence of other forms of chronic liver disease including the following:
    - Hepatitis B, as defined by presence of hepatitis B surface antigen (HBsAg) at screening,
    - Hepatitis C, as defined by presence of hepatitis C virus (HCV) antibody (anti-HCV), and HCV ribonucleic acid (RNA). Subjects with positive anti-HCV who test negative for HCV RNA at screening will be allowed to participate in the study,
    - Evidence of ongoing autoimmune liver disease,
    - Primary biliary cirrhosis,
    - Primary sclerosing cholangitis,
    - Wilson's disease,
    - Homozygous alpha-1-anti-trypsin deficiency,
    - History of hemochromatosis or iron overload,
    - Drug-induced liver disease,
    - Known bile duct obstruction,
    - Suspected or proven liver cancer, or
    - Any other type of liver disease other than NASH
12. Serum ALT >5 × the upper limit of normal (ULN).

13. Liver function tests (ALT/AST) > 5 × ULN at screening. One repeat test may be allowed within 7 days at the discretion of the Investigator.
    - If AST or ALT is > than ULN at screening visit an additional ALT, AST, INR, and total bilirubin will be obtained at least 2 weeks after the screening laboratories are obtained. If the abnormal parameters increase by > 30% at the retesting visit, the subject may not be randomized. The test may then be repeated in 2-4 weeks and if there is no further increase in the abnormal laboratory parameter, the subject may be randomized.
14. Estimated glomerular filtration rate <60 mL/min/1.73 m^2, as determined using the Modification of Diet in Renal Disease Study equation.
15. History of biliary diversion.
16. Positive for human immunodeficiency virus infection.
17. Active, serious medical disease with likely life expectancy <2 years.
18. Active substance abuse, including inhaled or injected drugs, within 1 year prior to screening. (Note that recreational cannabis/tetrahydrocannabinol use is permissible.)
19. Use of any excluded medications listed in Section 9.8.1.
20. Participation in an investigational new drug study in the 30 days prior to randomization (with the exception of Cohort 3 in which participants are prior placebo recipients in Cohort 1 and 2 of this study).
21. History of clinically significant dry eye (xerophthalmia) or other corneal abnormality or, if a contact lens wearer, does not agree to abstain from contact lens use from baseline through the last study drug dose.
22. Evidence of a clinically significant abnormality on slit-lamp examination or other clinically significant ophthalmologic finding, as determined by an ophthalmologist.
23. Known allergy or hypersensitivity to components of TVB-2640.
24. Prior history of hypersensitivity or drug/radiation-induced or other immune mediated pneumonitis.
25. Prior history of PPE syndrome.
26. Any contraindication to MRI (e.g., claustrophobia, metal implants).
27. Positive severe acute respiratory syndrome coronavirus-2 (SARS-CoV-2) polymerase chain reaction (PCR) test within 30 days before Baseline, history of hospitalization for COVID-19, or history of use of oxygen due to COVID-19. Note that previous COVID-19 infection alone is not exclusionary and vaccination against SARS-CoV-2 is allowed, but must be documented.
28. Any other condition which, in the opinion of the Investigator, would impede compliance, hinder completion of the study, compromise the well-being of the subject, or interfere with the study outcomes.

8.4. Source of Subjects

This will be a multi-center study. Each study center is required to obtain Institutional Review Board (IRB) approval to conduct the study before enrollment of subjects may commence. Subjects meeting the entry criteria who are known by or referred to the study center will be eligible for enrollment.

8.5. Subject Identification and Randomization

Subjects who are candidates for enrollment into the study will be assigned a sequential and unique subject number by the Investigator after the subject has provided written informed consent. Once a subject number has been assigned, it cannot be reused.

Subjects who have provided written informed consent will be evaluated for eligibility by the Investigator to ensure that the entry criteria (see Section 8.2 and Section 8.3) have been satisfied and that the subjects is eligible for participation in this clinical study.

Note that subjects who initially fail to meet all entrance criteria based on screening assessments (i.e., screen failures) may be submitted for re-screening once, with each case handled on an individual subject basis. The subject must be submitted for review via electronic mail and only entered into the electronic data capture (EDC) system as a screening candidate with written approval from the medical monitor. Subjects who initially fail screening because of clinical laboratory tests may have laboratory tests repeated during screening, at the Investigator's discretion, without being re-submitted for screening.

Three TVB-2640 dose levels are planned to be evaluated in a sequential fashion: 25 mg and, if study suspension criteria are not met at the 25 mg dose level (see Section 7.2), 50 mg, and then, in an open-label fashion, 75 mg.

At the 25 and 50 mg dose levels, subjects will be randomly assigned to TVB-2640 or placebo at a 2:1 ratio, with randomization stratified by type 2 diabetes mellitus status. The 50 mg dose level will also be stratified by country:

- Cohort 1: 25 mg dose level: TVB-2640 (Planned N=30) and Placebo (Planned N=15)
- Cohort 2: 50 mg dose level: US: TVB-2640 (Planned N=30) and Placebo (Planned N=15); China: TVB-2640 (Planned N=18) and Placebo (Planned N=9); Total: TVB-2640 (Planned N=48) and Placebo (Planned N=24)

Subjects are to receive their first study drug dose on Day 1 within 24 hours after randomization.

At both dose levels, subjects will receive TVB-2640 or placebo QD PO for 12 weeks, or until the time of the final on-treatment MRI-PDFF if after Week 12, but for no longer than 16 weeks.

After completion of Cohorts 1 and 2, subjects assigned to placebo in either cohort are eligible to crossover to receive TVB-2640 75 mg in Open-label Cohort 3 (Planned N=12 in order to obtain 10 evaluable subjects).

8.6. Subject Withdrawal Criteria

Subjects will be informed that they have the right to withdraw from the study at any time for any reason, without prejudice to their medical care. Study drug must be permanently discontinued

and the subject withdrawn from the study for any of the following TEAEs, with the subject managed as indicated:

- **Palmar-plantar Erythrodysesthesia (PPE)**: For mild to moderate symptoms of PPE (e.g., redness, pain, or scaling of hands or feet), study drug should be interrupted and symptomatic measures, including hand cream such as "Udder Cream", instituted immediately. Study drug may be resumed after resolution of symptoms. Study drug should be permanently discontinued at the first sign of significant/severe, redness, pain, or scaling of hands or feet, and symptomatic measures instituted immediately.
- **Dyspnea or suspected pneumonitis**: Study drug should be permanently discontinued for subjects with shortness of breath or other respiratory symptoms that are otherwise unexplained. The subject is to be carefully monitored for improvement. In the event that symptoms worsen or present as more than mild intensity, immediate referral to a qualified pulmonologist for definitive diagnosis and treatment should be undertaken.
- **Visual symptoms (including keratitis)**: Study drug should be permanently discontinued at the first sign of significant, redness, pain, or dryness of the eyes. Symptomatic measures, including moisturizing eye drops should be instituted. In the event that symptoms worsen or present as more than mild, immediate referral to a qualified ophthalmologist should be undertaken.
- **Evidence of drug-induced liver injury (DILI)**; Study drug should be permanently discontinued for subjects with evidence of DILI; see Appendix A for details.

Refer to the Study Manual for additional details regarding the assessment and management of these events.

Furthermore, the Investigator also has the right to withdraw subjects from the study for any of the following reasons:

- Progression of underlying disease that, in the opinion of the Investigator, precludes further study treatment.
- Occurrence of any other unacceptable AE.
- Study drug interruption for any reason for >7 days.
- Subject requires use of a prohibited concomitant medication or therapy.
- General or specific changes in the subject's condition unacceptable for further treatment within the study parameters, in the judgment of the Investigator.
- Non-compliance.
- Lost to follow-up.
- Subject withdrawal of consent.
- Sponsor request.

At the time of withdrawal, all study procedures outlined for the Early Termination visit should be completed. The primary reason for a subject's withdrawal from the study is to be recorded in the electronic case report form (eCRF).

8.7. Safety Review Committee

An SRC will oversee the study to ensure subject safety and to advise if any dosing alterations are recommended. The SRC will review safety including liver-related events (i.e., clinically meaningful elevations in ALT, AST, and bilirubin), and other efficacy (lipid parameters) and safety data as needed.

If study suspension criteria are met at any time (see Section 7.2), the SRC will review the relevant study data and make a determination whether enrollment and treatment of subjects may continue or be permanently terminated. In addition, the SRC will review all safety data from the 25 mg dose level after all subjects at that dose level complete the 12-week treatment period. Enrollment and treatment of subjects at the 50 mg dose level may commence only with the written approval of the SRC and with US FDA review.

8.8. Investigator Compliance

Study centers that deviate significantly from the protocol without prior approval from the Sponsor and regulatory authorities may be discontinued from the study. The Investigator at each study center is responsible for ensuring the accuracy and completeness of all research records, the accountability of study drug, and the conduct of clinical and laboratory evaluations as outlined in the protocol.

8.9. Subject Adherence to Protocol Schedule

All subjects are required to adhere to the protocol-specified visit schedule.

Subjects will be evaluated for study eligibility during the screening period, within 60 days before randomization. All subjects must provide written informed consent before any study-related samples are collected or evaluations performed in this study.

Subjects who are determined to be eligible for the study will be randomized within 24 hours before baseline (Day 1, the first day of study drug administration). Thereafter, subjects are to attend study center visits at Day 2 and Week 2, 4, 8, and 12. Subjects who discontinue study drug before completion of the Week 12 visit are to attend an Early Termination visit within 1 week after the last study drug dose. All study visits are to be conducted on an out-patient basis. Additional study center visits may be scheduled, as deemed necessary based on the subject's clinical status.

Permissible visit windows are specified in Table 2.

A Follow-up visit will be conducted at Week 16 (±1 week) for MRI-PDFF, measurement of liver aminotransferases, and safety assessments. Additional safety follow-up may be required thereafter if drug-related AEs have not resolved at that time.

Failure to attend scheduled study visits within the protocol-specified windows may result in discontinuation from the study.

The Sponsor notes that the COVID-19 pandemic may impact the conduct of the 50 mg and 75 mg cohorts, with challenges potentially arising from quarantines and travel limitations affecting study subjects as well as clinic and imaging facility closures. Accordingly, safety assessments scheduled for the Week 12 and 16 visits, as identified in Table 2, including sample

collection for clinical laboratory tests, physical examinations, and documentation of AEs, may be performed at an alternate study center or by home health care visit. Furthermore, if the subject is unable to attend the Week 12 visit at the study center, one MRI-PDFF may be performed at any time between Week 11 and 16. If this scan is performed after Week 12, the subject must continue to receive study drug until the final scan is performed; however, study drug is not to be continued for >16 weeks. The safety of subjects who continue dosing beyond 12 weeks will be assessed during this continued dosing period by planning for blood sample collection at an alternate site or home visit at Week 12 and blood sample collection and physical examination on the last day of dosing. A 4-week off treatment safety assessment is to occur after the last dose, regardless of whether a subject receives treatment for 12 or 16 weeks.

9. TREATMENT OF SUBJECTS

9.1. Description of Study Drug

9.1.1. TVB-2640

TVB-2640 is a small-molecule, orally-bioavailable, reversible inhibitor of the human FASN enzyme. TVB-2640 has a molecular formula of $C_{27}H_{29}N_5O$ and a molecular weight of 440.

TVB-2640 immediate-release formulation will be supplied for oral administration as 25 and 50 mg strength tablets.

9.1.2. Placebo

A placebo tablet is available which conforms to the appearance, shape and size of the TVB-2640 50-mg strength tablet.

9.2. Study Drug Packaging and Labeling

TVB-2640 will be packaged in bulk, screw-top plastic bottles.

TVB-2640 will be labeled in accordance with applicable regulatory requirements. Study drug labels will not bear any statement that is false or misleading in any manner or represents that the study drug is safe or effective for the purposes for which it is being investigated.

9.3. Study Drug Storage

TVB-2640 drug product and placebo should be stored in a secure, limited access storage area at room temperature.

9.4. Study Drug Accountability

The US FDA, Chinese Food and Drug Administration (CFDA) and other applicable regulatory authorities require accounting of all investigational drug received by each study center. Records of drug disposition required include the date received by the center, date administered, quantity administered, and the subject to whom study drug was administered. The Investigator is responsible for the accountability of all used and unused study drug containers and unused study drug.

Each study center is to use a study drug accountability log to document study drug disposition. All items on this form are to be completed in full. The Sponsor's clinical research associate (CRA) is to approve the area where study drug is to be stored and accountability records are to be maintained.

The investigator identification number and subject initials (as allowed by local regulations) and identification number are to be recorded on each study drug accountability log. Each time study personnel dispense study drug for a subject, he or she is to record the date dispensed, amount of study drug dispensed, and his or her initials. Study personnel are to monitor the inventory of clinical supplies and maintain a count of all used and unused study drug. The CRA is to review study drug accountability records and remaining drug supplies during routine monitoring visits.

9.5. Study Drug Dose and Administration

All subjects will receive study drug QD PO for 12 weeks, or until the time of the final on-treatment MRI-PDFF if after Week 12, but for no longer than 16 weeks, with the first dose administered on Day 1.

Each QD dose of study drug is to be taken at the same time of day with food; the study drug dose is to be taken in the morning on Days 1 to 8 and then in the evening on each day thereafter, with each dose separated by 24 hours (±4 hours). (The Day 8 and 9 doses will be separated by 36 hours.) Subjects will receive one of the following each day, based on their cohort and treatment assignment:

- Placebo: 1 placebo tablet
- TVB-2640 25 mg: 1 25-mg TVB-2640 tablet
- TVB-2640 50 mg: 1 50-mg TVB-2640 tablet
- TVB-2640 75 mg: 1 50-mg TVB-2640 tablet and 1 25-mg TVB-2640 tablet

9.6. Dose Reductions

Dose reductions for the management of other study drug-related toxicities are permissible for subjects assigned to TVB-2640 50 mg or 75 mg with the approval of the Medical Monitor.

- Subjects assigned to 50 mg may have the dose reduced to 25 mg. In such cases, subjects will receive 1 25-mg tablet.
- Subjects assigned to 75 mg may have the dose reduced to 50 mg. In such cases, subjects will receive 1 50-mg tablet.

Subjects for whom the study drug dose is reduced are to continue treatment at the reduced dose for the remainder of the treatment period. At the time the dose is reduced, all study drug is to be returned and study drug will be redispensed according to the reduced dose.

Subjects receiving 25 mg for whom a dose reduction is deemed necessary should be discontinued from the study.

9.7. Rationale for the Doses Selected

The following 3 dose levels have been selected for this study: 25 mg, 50 mg, and 75 mg, delivered in QD PO tablet(s) of strengths 25 mg and 50 mg.

These doses are expected to span a range of effects on the primary endpoint, percentage of subjects achieving a 30% or greater reduction of liver fat determined by MRI-PDFF, as well as overall reduction of liver fat. This range also is expected to enable determination of minimal effective dose for future studies. In addition, the AE profile of the doses will be collected and used to assess dose optimization for future studies.

In a Phase 1 study of 12 otherwise healthy men with characteristics of the metabolic syndrome (Study 2006432), 6 subjects received 50 mg of QD TVB-2640 for 10 days, the lowest dose tested. This 50 mg dose reduced hepatic lipogenesis by an average of 24% (95% confidence interval -4%, -39%, p=0.02) and reduction was observed in 100% of the 6 subjects. These data,

and similar observations with other hepatic lipogenesis inhibitors, suggest that 12 weeks of QD dosing with 50 mg or 75 mg of TVB-2640 will result in at least a 30% reduction of intrahepatic fat in a large percentage of NASH subjects.

The safety of the 50 mg and 75 mg doses is supported by nonclinical Good Laboratory Practice (GLP) toxicology studies and prior human experience.

[redacted] Additionally, none of the 6 men with metabolic syndrome who received 50 mg of TVB-2640 experienced a TEAE.

In Study 3V2640-CLIN-002, cancer patients received daily doses of 100 mg or greater of TVB-2640 and the blood exposure was reported to be linearly proportional to the oral dose. PK data from the Phase 1 study of 12 men with metabolic syndrome are consistent with this relationship. [redacted].

Refer to the Investigator's Brochure for a summary of safety data with TVB-2640.

The existing nonclinical and clinical studies of TVB-2640 support the safety of the doses selected as well as the potential to differentiate the activity of the drug on the percent of subjects achieving a 30% or greater reduction of liver fat and an overall reduction of liver fat. These data will enable guidance of a minimal effective dose for future studies.

Due to the COVID-19 pandemic, if the subject in the 50 mg and 75 mg cohorts is unable to attend the Week 12 visit, one MRI-PDFF may be performed at any time between Week 11 and 16. If this scan is performed after Week 12, the subject must continue to receive study drug until the final scan is performed. However, study drug is not to be continued for >16 weeks.

Nonclinical and clinical data obtained to date support daily dosing with TVB-2640 for 16 weeks, as follows:

- Complete safety data from 30 subjects who received TVB-2640 25 mg for 12 weeks and available safety data from 30 subjects who received TVB-2640 50 mg for 4 to 12 weeks (ongoing cohort) as reviewed by the SRC, support continued dosing beyond Week 12 and up to and including Week 16. Safety and PK data from these 2 doses demonstrate a proportional and expected plasma exposure of TVB-2640 in subjects, further providing confidence in continued dosing.

- Safety data are available from 136 patients with cancer who received either TVB-2640 alone or in combination with a taxane of whom 72 received TVB-2640 at doses of 100 to 600 mg per day, with a mean and maximum time on treatment of 62 days (9 weeks) and 324 days (46 weeks), respectively. These doses and duration used in CLIN-002 far exceed those in the current study.
- In addition to 2 previously completed 13-week GLP toxicity studies, the Sponsor is currently conducting 2 chronic toxicology studies. ([redacted])
  a. The in-life portion of a 6-month chronic toxicity study in rats has been completed (SN19-1112); adverse in-life or clinical pathology toxicities were not observed in this study. Toxicokinetic results from this study were consistent with expected exposures. The AUC of TVB-2640 at the highest dose in this study [redacted], over 2.5-times higher than the 50 mg dose in humans.
  b. The in-life portion for the 9-month toxicity study in dogs has been completed through Week 36 of dosing (SN19-1113), with no in-life or clinical pathology toxicities observed through that time. The AUC of TVB-2640 at the highest dose was [redacted], over 2.4-times higher than the 50 mg dose in humans.

9.8. Concomitant Medications

All prescription and non-prescription medications and therapies, including pharmacologic doses of vitamins, herbal medicines, or other non-traditional medicines, taken from 28 days prior to the first dose of TVB-2640 through the Final Study Visit must be recorded in the eCRF.

9.8.1. Excluded Medications

The following medications and treatments are prohibited during study participation.

- Any investigational agent or device other than TVB-2640.
- Strong inhibitors or inducers of cytochrome P450 3A4 (CYP3A4). (Refer to the following for examples: http://www.fda.gov/drugs/developmentapprovalprocess/developmentresources/druginteractionslabeling/ucm093664.htm.)
- Drugs historically associated with NAFLD (amiodarone, methotrexate, systemic glucocorticoids, tetracyclines, tamoxifen, estrogens at doses greater than those used for hormone replacement, anabolic steroids, valproic acid, and other known hepatotoxins).
- Glucagon-like peptide analogue or a complex oral OAD regimen (3 or more OADs).
- Insulin dose adjustment >10%. (Subjects receiving insulin at baseline who require a >10% dose adjustment for the management of Type 2 diabetes mellitus are to be discontinued from the study.)

- Introduction of new or changes in continuing oral diabetic medications (e.g., pioglitazone) is prohibited. Concomitant oral diabetic medications are permitted only if the dose has been stable for at least 3 months before screening and remains stable throughout study participation.

Subjects requiring such treatment during study participation are to be discontinued.

9.8.2. Permitted Medications

Medications and treatments other than those specified in Section 9.8.1 are permitted during the study. Subjects should be closely monitored, and treatment is to be instituted for disease-related symptoms, as appropriate.

9.8.3. Contraception

[redacted] females of childbearing potentia must use two forms of birth control, one being a barrier method (i.e., condoms, diaphragm, non-hormonal IUD) throughout the study and for at least 3 months after the final study drug dose. Hormonal contraception (estrogens stable ≥3 months) and hormonal IUDs are permitted if used with a secondary barrier birth control method (e.g., condoms).

9.9. Blinding

In Cohorts 1 and 2, study drug will be dispensed to subjects in a single-blinded fashion. All other study personnel, with the exception of personnel conducting/interpreting imaging studies (see Section 11.1) at each study center, will be unblinded to the subject's treatment assignment. In Cohort 3, TVB-2640 will be dispensed in an open-label fashion.

10. BASELINE ASSESSMENTS

All subjects must provide written informed consent before the performance of any study-related procedures.

10.1. Demographics

Subject demographics, including age, sex, race, and ethnicity, are to be documented during screening.

10.2. Medical and Social History

A complete medical history is to be documented during screening and updated at baseline, prior to administration of the first study drug dose.

The medical history also is to include NASH related history, including the date of diagnosis, and as available, NAS score, APRI, NAFLD fibrosis score, and/or enhanced liver function (ELF) score, as well as abbreviated weight history, family history (siblings and subjects; particularly of obesity and liver disease), past medical/surgical events and illness history (including diabetes, gestational diabetes, hypertension, lipodystrophy, polycystic ovarian syndrome, and any history of Gilbert's disease) and other associated co-morbid conditions including previously diagnosed lipid and metabolic disease-related conditions (hypercholesterolemia, hypertriglyceridemia, diabetes mellitus), and all diagnoses related to previous liver disease as well as other diagnoses of major organ systems including cardiac disease, renal disease, endocrine disease, hypertension, gout or other arthropathies, disturbances of vision, peripheral neuropathy, myopathy, pancreatitis, and cholelithiasis.

History of previous COVID-19 infection or vaccination against SARS-CoV-2 should also be documented.

Social history, including alcohol, tobacco, and caffeine use, also will be documented.

10.3. Screening Serology

Screening serologies include HIV RNA, HBsAg, anti-HCV, and HCV RNA. A PCR test for SARS-CoV-2 (the virus that causes COVID-19) also is to be performed.

Subjects with positive anti-HCV who test negative for HCV RNA at screening will be allowed to participate in the study.

10.4. Pregnancy Testing

For women of child-bearing potential, a serum β-hCG pregnancy test will be performed during screening and a serum or urine β-hCG pregnancy test will be performed within 48 hours before baseline. Results must be available and confirmed to be negative before the subject is eligible for randomization in the study. Pregnancy testing is to be repeated during the study any time pregnancy is suspected.

All subjects of reproductive potential must agree to adequate birth control from the time the screening pregnancy test is performed through 3 months after the final study drug dose.

Study drug is to be discontinued for any subject with positive pregnancy test findings; see Section 12.2.6 for details regarding management of any pregnancies during the study.

Women of non-child-bearing potential are to have this documented as part of the medical history.

11. EFFICACY, PHARMACOKINETIC, AND PHARMACODYNAMIC ASSESSMENTS

11.1. Efficacy and Pharmacodynamic Assessments

11.1.1. Magnetic Resonance Imaging Studies

All imaging studies will be done by experienced research personnel according to a standard imaging protocol. Personnel reading imaging studies will be blinded to the subject's treatment group allocation and clinical and biochemical data. A trained image analyst will assess all images at each study center; ideally, the same reviewer should review all scans at that study center for the duration of the study. All imaging studies also will be read centrally; the central reader also will be blinded to the subject's treatment group allocation and will not have access to the subject's clinical and biochemical data. The results of the central read will be used in the analysis of imaging data.

11.1.1.1. Proton-density Fat Fraction by Magnetic Resonance Imaging (MRI-PDFF)

MRI-PDFF is a non-invasive, quantitative, and accurate measure of liver fat content to assess treatment response in early-phase of NASH studies (Caussy 2018). MRI-PDFF is to be determined at the time points designated in Table 2. Visceral adipose tissue measurement by MRI will be performed for all eligible subjects. Subjects who are randomized will have repeat visceral adipose tissue and liver fat measurements performed.

11.1.1.2. Liver Stiffness by Magnetic Resonance Elastography

MRE has been shown to be a robust, accurate biomarker for the quantitative, noninvasive evaluation of liver stiffness as a surrogate for liver fibrosis and for NASH detection. MRE is to be determined during screening, as per Table 2. Note that subjects without a biopsy performed within 24 months of randomization MUST have MRE performed during screening.

11.1.2. Liver Fibrosis by FibroScan

The degree of liver fibrosis, as indicated by the kpa, will be assessed using Fibroscan at the time points designated in Table 2.

11.1.3. Clinical Laboratory Tests

Refer to the Laboratory Manual for details regarding clinical efficacy/pharmacodynamic laboratory sample collection, processing, storage, and shipment. Note that in Cohort 3 (75 mg), some of the exploratory biomarker samples will not be collected or may be collected on an optional basis, as indicated in Table 2.

11.1.3.1. Lipid and Metabolic Panel, Glycated Hemoglobin, and Lipoprotein Particle Size

A fasting blood sample for lipid and metabolic panels as well as HbA1c is to be collected at the time points designated in Table 2. The following parameters are to be determined:

- **Lipid Panel**: LDL-C, non-LDL-C, HDL-C, non-HDL-C, total cholesterol, TG, ApoB, and Lp(a) particles

- **Metabolic Panel**: Glucose, insulin, NEFA, and GGT. HOMA2-IR and adipo-IR are to be calculated from fasting insulin, fasting glucose, and fasting NEFA, as applicable
- **Hb1Ac**
- **Lipoprotein Particle Size Analysis** by nuclear magnetic resonance (NMR)

Subjects are to fast overnight for at least 9 hours prior to collection of the fasting sample. (Water is permissible.)

11.1.3.2. NASH / Fibrosis Markers

A blood sample for NASH / fibrosis markers, including CK-18, FIB-4 Index, ELF (TIMP-1, PIIINP, HA), PROC3, and FIBROSpect 2, is to be collected at the time points designated in Table 2. Instructions regarding the calculation of indices are provided in the Operations Manual.

11.1.3.3. Eicosanoid Panel

A blood sample for an eicosanoid panel, including 5-LOX–derived leukotriene B4 (LTB4), COX-derived prostaglandin E2 (PGE2), and 15-LOX–derived 15-hydroxyeicosatetraenoic acid (15-HETE) and lipoxin A4 (LXA4), is to be collected at the time points designated in at the time points designated in Table 2.

11.1.3.4. Inflammatory Biomarkers

A blood sample for inflammatory biomarkers, including tumor necrosis factor-alpha (TNFα), interleukin (IL)-6, IL-8, high-sensitivity C-reactive protein (Hs-CRP), monocyte chemoattractant protein-1 (MCP-1), is to be collected according to the time points in Table 2.

11.1.3.5. Adiponectin, Resistin, Leptin

A blood sample for determination of adiponectin, resistin, and leptin, is to be collected according to the time points in Table 2, with these parameters determined using enzyme-linked immunosorbent assay (ELISA).

11.1.4. Lipidomic Analyses

In the current study, a fasting blood sample for fatty acid analyses will be collected according to the time points in Table 2. Plasma TG will be isolated from the blood. The quantity and composition of fatty acids in these TGs will be evaluated. The quantity and composition of fatty acids in these TGs will be evaluated. The quantity of palmitate (C16:0) in this fraction, representing hepatic synthesized lipid, will be compared to the quantity of omega-6-linoleic acid (C18:2n6), an essential fatty acid obtained through dietary sources. Decreased ratios of C16:0 / C18:2n6 are indicative of decreased hepatic lipogenesis (Hudgins, 1996).

11.1.5. Genomics

A blood sample for genomics pertaining to the disease under study will be collected at the time points designated in Table 2; subjects will be asked to provide separate written informed consent for the collection of such samples.

11.1.6. Sample Collection for Storage

Blood and spot urine samples for storage will be collected at the time points designated in Table 2. These samples will be stored for the duration of the TVB-2640 clinical development program and used for analysis of additional pharmacodynamic biomarkers relevant to NAFLD and/or NASH when such biomarkers are identified and/or relevant assays are developed.

These samples will not be used for genetic analysis nor creation of cell lines. The biomarker assessments performed may include tests to determine the level of certain proteins, RNA or other disease related molecules and whether they change after TVB-2640 treatment.

The samples will be coded and de-identified, in the same way all other study samples are coded and de-identified. Every study subject has a unique study number that will be used to track the samples. The subject's confidentiality will be maintained and will not be made publicly available to the extent permitted by the applicable laws and regulations.

The Sponsor and/or its designee and the Regulatory Authorities will have access to the samples/data if they are ever used for a future research.

The collection of blood and spot urine samples for storage is not voluntary and will be collected for all subjects randomized to the study.

11.1.7. Anthropometric Assessments

Anthropometric assessments are to be performed at the time points designated in Table 2, including:

- Weight (without shoes or heavy clothing) (kg), with calculation of BMI using screening height and weight.
- Waist circumference at umbilicus (cm)
- Hip circumference (cm)
- Waist-hip ratio

11.1.8. Lymphocyte Subsets in Peripheral Blood Mononuclear Cells

Blood samples are to be collected from a subset of subjects enrolled at selected study centers at baseline and Week 12 for exploratory evaluation of change in CD4/Th17/regulatory T-cell populations and, potentially, natural killer-T and epigenetic or activation changes in peripheral blood mononuclear cells.

11.2. Pharmacokinetic Assessments

[redacted]

[redacted]

The times of study drug administration, the most recent meal prior to blood sample collection, and of blood sample collection ares to be documented in the eCRF.

[redacted]

12. ASSESSMENT OF SAFETY

12.1. Safety Parameters

12.1.1. Vital Signs

Vital signs, including oral temperature (ºC), pulse (bpm), systolic and diastolic blood pressure (mmHg), and respiration rate (breaths/minute), are to be measured and documented in the eCRF at the time points designated in Table 2. Measurements are to be made after the subject has been resting in a supine position for a minimum of 5 minutes.

12.1.2. Physical Examination and Height

A complete physical examination will be conducted for all subjects at the time points designated in Table 2. The complete physical is to include examination of general appearance, head/ears/eyes/nose/throat, lungs/chest, heart, abdomen, lymph nodes, musculoskeletal, extremities, skin, and neurological examination. Liver signs (jaundice, spider angiomata, palmar erythema, hepatomegaly, splenomegaly, asterixis) also are to be assessed.

During screening, the physical examination is to include measurement of height (cm).

Abbreviated (i.e., symptom-directed) physical examinations, including assessment of focused liver signs (presence or absence of edema, ascites, hepatomegaly, splenomegaly, asterixis), skin, and extremities, will be conducted at the time points designated in Table 2 to address any complaints or concerns verbalized by the subject at all other study visits.

Any abnormal findings are to be documented as AEs.

12.1.3. Electrocardiogram (ECG)

Single 12-lead ECGs are to be performed for all subjects at the time points designated in Table 2.

On Day 1, ECGs are to be performed before and 4 hours after the first study drug dose, with the ECG performed prior to PK blood sample collection.

12.1.4. Ophthalmologic Examinations

Ophthalmologic examinations are to be performed for all subjects at the time points designated in Table 2.

Near and far visual acuity is to be assessed by an ophthalmologist using standard measures (e.g., Early Treatment Diabetic Retinopathy Study or similar) in each eye. If the subject wears corrective lenses (e.g., glasses), then visual acuity is to be checked first with corrective lenses and then without correction. (A best-corrected examination is not required.) Note that contact lens wearers are to abstain from contact lens use during study drug treatment.

A slit lamp examination (biomicroscopy), including examination of the eyelids, conjunctiva, cornea, anterior chamber, iris, and lens, is to be performed after completion of visual acuity testing for the determination of corneal health.

If a subject experiences a treatment-emergent ophthalmologic abnormality, an ophthalmologic examination should be performed within 24 to 48 hours or next business day after symptom onset to evaluate the abnormality.

12.1.5. Safety Laboratory Assessments

Blood and urine samples are to be collected for hematology, clinical chemistry, and urinalysis according to the time points designated in Table 2. The following safety laboratory parameters are to be measured:

Hematology	
Hematocrit	Platelet count
Hemoglobin	White blood cell count with differential
Red blood cell count	Absolute neutrophil count
Coagulation Studies	
Prothrombin time	Activated partial thromboplastin time
INR	
Chemistry (Fasting)	
Chloride	Carbon dioxide
Sodium	Potassium
Blood urea nitrogen	Calcium
Creatinine	Magnesium
Albumin	Glucose
Total protein	Alkaline phosphatase
AST*	ALT*
Indirect and direct bilirubin	
Creatine phosphokinase (total and fractionated) (screening and baseline only)	
*AST and ALT collected as part of the clinical chemistry panel are also applicable for efficacy assessments.	
Urinalysis	
Specific gravity	Protein
pH	Ketones
Blood	Microscopic examination of sediment
Glucose	

LFTs determined as part of the clinical chemistry panel will be used for the assessment of the secondary efficacy endpoint.

Clinical laboratory evaluations are to be repeated as necessary during treatment at a schedule determined by the Investigator, based on the subject's clinical status.

Laboratory abnormalities that are considered by the Investigator to be clinically significant for a particular subject are to be reported as an AE.

12.2. Adverse and Serious Adverse Events

12.2.1. Definition of Adverse Events

12.2.1.1. Adverse Events (AE)

An AE is any untoward medical occurrence in a patient or clinical investigation subject 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 abnormal laboratory findings), symptom, or disease temporally associated with the use of an investigational product, whether or not related to the investigational product.

An unexpected AE is any event for which the nature or severity is not consistent with the information in the current Investigator's Brochure.

All AEs that occur from screening (i.e., after the ICF is signed) through 28 days after the last dose of study drug, whether or not they are related to the study drug, are to be reported in the eCRF.

12.2.1.2. Serious Adverse Event (SAE)

An AE or suspected adverse reaction is considered serious if, in the view of either the Investigator or Sponsor, it:

- Results in death.
- Is life-threatening. Life-threatening means that the subject was at immediate risk of death from the reaction as it occurred, i.e., it does not include a reaction which hypothetically might have caused death had it occurred in a more severe form.
- Requires in-patient hospitalization or prolongation of existing hospitalization. Hospitalization admissions and/or surgical operations scheduled to occur during the study period, but planned prior to study entry are not considered AEs if the illness or disease existed before the subject was enrolled in the study, provided that it did not deteriorate in an unexpected manner during the study (e.g., surgery performed earlier than planned). Additional exclusions to SAE reporting include hospitalizations for:
  - Elective procedures.
  - Social/administrative reasons in the absence of an AE.
  - Expected deterioration caused by progression of the underlying disease.
- Results in persistent or significant disability/incapacity. Disability is defined as a substantial disruption of a person's ability to conduct normal life functions.
- Is a congenital anomaly/birth defect.

- Is an important medical event. An important medical event is an event that may not result in death, be life-threatening, or require hospitalization but may be considered an SAE when, based upon appropriate medical judgment, it may jeopardize the subject and may require medical or surgical intervention to prevent one of the outcomes listed in the definitions for SAEs. Examples of such medical events include allergic bronchospasm requiring intensive treatment in an emergency room or at home, blood dyscrasias or convulsions that do not result in in-patient hospitalization, or the development of drug dependency or drug abuse.

All SAEs that occur after any subject has been enrolled, before treatment, during treatment, or within 28 days following the cessation of treatment, whether or not they are related to the study drug, must be recorded on forms provided by Sagimet Biosciences, or designee.

12.2.2. Adverse Event Assessment

12.2.2.1. Intensity

The intensity of each AE is to be assessed by the Investigator according to the National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE), Version 5.0 (see https://ctep.cancer.gov/protocoldevelopment/electronic_applications/docs/CTCAE_v5_Quick_Reference_5x7.pdf). If the AE is not included in the NCI CTCAE, then the Investigator is to determine the intensity of the AE according to the following criteria:

Mild (Grade 1): AE that disappears or is easily tolerated on continuation of study drug.

Moderate (Grade 2): AE sufficiently discomforting to cause interference with usual work activities.

Severe (Grade 3): AE that is incapacitating, with inability to work or perform daily activities.

Life-Threatening (Grade 4): AE that is *potentially* life-threatening.[1]

Death (Grade 5): Death related to AE.

12.2.2.2. Relationship to Study Drug

The causal relationship of each AE to study drug will be determined by the Investigator according to best medical judgment, as follows:

Definitely related: This category applies when, after careful medical consideration, there is almost no consideration of other causation.

Probably related: There is a clinically plausible time sequence between onset of the AE and study drug administration. The AE is unlikely to be caused by a concurrent and/or underlying illness, other drugs, or

[1]. If a life-threatening (Grade 4) adverse event is *immediately* life-threatening, the event is, by definition, serious and is to be reported as described in Section 12.2.4.

	procedures. If applicable, the AE follows a clinically consistent resolution pattern upon withdrawal of study drug.
Possibly related:	There is a clinically plausible time sequence between onset of the AE and study drug administration, but the AE could also have been caused by the concurrent/underlying illness, other drugs, or procedures. Information regarding study drug withdrawal may be lacking or unclear. "Possible" should be used when study drug administration is one of several biologically plausible causes of the AE.
Unlikely related:	The AE is most likely due to a non-study drug-related cause. However, association with the study drug cannot be completely ruled out.
Unrelated:	Another cause of the AE is most plausible and a clinically plausible temporal sequence is inconsistent with the onset of the AE and study drug administration and/or a causal relationship is considered biologically implausible.

If the relationship between the AE/SAE and study drug is determined to be "possible", "probable", or "definite", the event will be considered to be treatment-related for the purposes of expedited regulatory reporting and safety analyses.

12.2.2.3. Seriousness

AEs that meet the criteria specified in Section 12.2.1.2 are to be considered serious.

12.2.3. Recording Adverse Events

Each subject must be carefully monitored for the development of any AEs. This information should be obtained in the form of non-leading questions (e.g., "How are you feeling?") and from signs and symptoms detected during each examination, observations of study personnel, and spontaneous reports from subjects.

All AEs (serious and non-serious) spontaneously reported by the subject and/or in response to an open question from study personnel or revealed by observation, physical examination, or other diagnostic procedures will be documented in the subject's source documents and recorded in the eCRF. Any clinically relevant (as determined by the Investigator) deterioration in laboratory assessments or other clinical findings is considered an AE and must be recorded in the subject's source documents and in the eCRF.

Information about AEs will be collected from screening (i.e., after the ICF is signed) through 28 days after the last dose of study drug. The AE term should be reported in standard medical terminology when possible. Also when possible, signs and symptoms indicating a common underlying pathology should be noted as one comprehensive event. For each AE, the investigator will evaluate and report the onset date, resolution date, intensity, causality, action taken, serious outcome (if applicable), and whether or not it caused the subject to discontinue the study.

12.2.4. Reporting Serious Adverse Events

All SAEs (related and unrelated) occurring from screening through 28 days after the last study drug dose are to be reported. Any suspected case of pneumonitis, considered an adverse events of special interest, should be reported in the same manner as an SAE.

The Investigator must report all SAEs within 24 hours of discovery via the EDC system, which will trigger email notifications to the ProSciento, Inc., Medical Monitor/Safety team. (In the event the EDC system is not available, then SAE reports must be emailed to 3VBIOSafety@ProSciento.com.)

The Investigator must complete and electronically sign and date the SAE pages of the eCRF and verify the accuracy of the information recorded on the SAE pages with the corresponding source documents.

Additional follow-up information, if required or available, should be submitted within the EDC system within one business day of receipt and this should be completed on a follow-up SAE form and placed with the original SAE information and kept with the appropriate section of the eCRF and/or study file.

Sagimet Biosciences is responsible for notifying the relevant regulatory authorities of certain events. It is the Investigator's responsibility to notify the IRB of all SAEs that occur at his or her study center. Investigators will also be notified of all unexpected, serious, drug-related events (7/15 Day Safety Reports) that occur during the clinical study. Each study center is responsible for notifying its IRB of these additional SAEs.

12.2.5. Follow-Up of Adverse Events

The Investigator must continue to follow all treatment-emergent SAEs and non-serious AEs considered to be at least possibly related to study drug either until resolution or the event is clearly determined to be stable or due to a subject's stable or chronic condition or inter-current illness(es). This follow-up may extend after the end of the study.

12.2.6. Pregnancy

Pregnancies occurring in the subject or subject's partner while the subject is receiving study drug or within 3 months after the subject's last dose of study drug will not be considered serious, but are to be reported using the same procedures as for SAEs described in Section 12.2.4.

Study drug must be discontinued immediately in the event of a pregnancy in the subject. The subject should be referred to an obstetrician/gynecologist experienced in reproductive toxicity for further evaluation and counseling.

The Investigator will follow the subject / subject's partner until completion of the pregnancy, and must notify the Medical Monitor of the outcome within 5 days. The Investigator will provide this information as a follow-up to the initial report.

If the outcome of the pregnancy meets the criteria for immediate classification as an SAE (i.e., spontaneous abortion [any congenital anomaly detected in an aborted fetus is to be documented], stillbirth, neonatal death, or congenital anomaly), then the Investigator should report it as such.

Furthermore, all neonatal deaths that occur within 28 days of birth should be reported, without regard to causality, as SAEs. In addition, any infant death after 28 days that the Investigator suspects is related to the in utero exposure to the study drug should also be reported.

12.2.7. Overdose

Signs and symptoms of an overdose should be reported as AEs. Overdoses will not be considered SAEs unless the outcome of the overdose meets seriousness criteria (see Section 12.2.1.2).

12.2.8. Protocol Deviations Due to an Emergency or Adverse Event

Departures from the protocol will be determined as allowable on a case-by-case basis and only in the event of an emergency. The Investigator or other physician in attendance in such an emergency must contact the Medical Monitor as soon as possible to discuss the circumstances of the emergency.

The Medical Monitor, in conjunction with the Investigator, will decide whether the subject should continue to participate in the study. All protocol deviations and reasons for such deviations must be documented in the subject's source records.

13. STATISTICS

13.1. Determination of Sample Size

The sample size is based on a fixed-sequence strategy which tests for a treatment difference in the primary endpoint (percent change at Week 12 in liver fat from baseline as determined by MRI-PDFF) between each randomized TVB-2640 dose group (25 and 50 mg) versus placebo. The fixed-sequence strategy will start with the highest dose group comparison. If statistically significant, then testing will proceed to the low dose group comparison. Each TVB-2640 dose group will be compared to the pooled placebo group using an F-test test from an analysis of covariance (ANCOVA) model with fixed effects for the stratification factor (diabetes presence/absence) and treatment group (i.e., TVB-2640 dose groups and pooled placebo) and with the baseline MRI-PDFF value as a covariate. Since the primary efficacy endpoint may not be normally distributed, the non-parametric Wilcoxon rank-sum test is conservatively used, instead of the F-test, for power calculations.

The sample size is based on a conservative assumption that the primary analysis is going to be performed in the US population only, resulting in 30 evaluable subjects in the placebo group, and 30 evaluable subjects in each of the randomized TVB-2640 dose levels. Based on Patel (2016), it is conservatively assumed that the primary endpoint has a standard deviation of 30. Power calculations assume the primary endpoint is lognormally distributed with a standard deviation of 30, there are at least 30 evaluable subjects in each treatment group, and the two-sided Wilcoxon rank sum test will be used to test each pairwise treatment difference at the 0.05 Type I error level. Under the fixed-sequence strategy which maintains an overall 0.05 Type I error rate, the study has at least 80% overall power to detect both treatment differences (i.e., both high dose [50 mg] versus placebo and low dose [25 mg] versus placebo), if each mean treatment difference is at least 24. If the study does not proceed to the high-dose level, (i.e., there are 30 evaluable subjects treated with 25 mg TVB-2640 and only 15 with placebo), then the study has at least 77% power to detect a mean treatment difference of at least 24. The study will have more power if the subjects from China are included in the primary analysis and the same assumptions of treatment difference and variability hold.

Power calculations for the key secondary endpoint (percentage of subjects with at least a 30% reduction in the primary endpoint) are based on pairwise comparisons with the placebo group using Fisher's two-sided exact test [equivalent to Cochran-Mantel-Haenszel (CMH) without stratification] at the 0.05 significance level. Based on Loomba (2018), it is expected there will be less than 5 responses out of 30 evaluable subjects in the placebo group (i.e., less than 16.7%). With 30 evaluable subjects in each treatment group, there is at least 80% power to detect at least a 36.7% increase in the response rate (TVB-2640 treatment group minus placebo) when the placebo response rate is at most 16.7%. If the study does not proceed to the high-dose level, (i.e., 30 evaluable subjects treated with 25 mg TVB-2640 and only 15 with placebo), then the study has at least 63% power to detect at least a 36.7% increase in the response rate (25 mg TVB-2640 treatment group minus placebo) when the placebo response rate is at most 2 out of 15 evaluable subjects (i.e., 13.34%).With 30 subjects in each TVB-2640 treatment arm, there is at least a 95% probability of detecting at least 1 AE when the AE rate is 10% or more in the underlying population. The open label 75mg cohort is being assessed for safety only.

13.2. Statistical Analyses

13.2.1. Analysis Populations

The modified intent-to-treat population (mITT) is defined as all subjects who are randomized, receive study drug for at least 8 weeks, and have a baseline and at least 1 post-baseline MRI-PDFF assessment on or after Week 8. The mITT is the primary population for analysis of MRI-PDFF assessments and subjects will be analyzed according to the randomized treatment assignment.

The intent-to-treat population (ITT) is defined as all subjects who are randomized and received at least 1 dose of study drug. The ITT will be used for analysis of secondary efficacy endpoints and supportive and sensitivity analyses of MRI-PDFF assessments. Subjects will be analyzed according to the randomized treatment assignment. Details will be provided in the Statistical Analysis Plan (SAP).

The safety population is defined as all subjects who are randomized and received at least 1 dose of study drug and will be used for all analysis of safety. Subjects will be analyzed according to the treatment received. If all subjects were dosed according to randomized treatment assignment, then the safety population and ITT are identical.

13.2.2. Efficacy Analysis

A test for country-by-treatment interaction with respect to the primary efficacy endpoint, percent change from baseline in hepatic fat fraction at Week 12, as determined by MRI-PDFF, will be performed in the 50 mg cohort. An ANCOVA model with fixed effects for the stratification factor (diabetes presence/absence), country (US/China), treatment group (TVB-2640 50 mg and pooled placebo), and country-by-treatment interaction and with the baseline MRI-PDFF value as a covariate including all subjects from US and China will be performed. If there is no interaction detected ($p \geq 0.05$), the primary efficacy endpoint, percent change from baseline in hepatic fat fraction at Week 12, as determined by MRI PDFF, will be analyzed using an analysis of covariance (ANCOVA) model with fixed effects for the stratification factor (diabetes presence/absence) and treatment group (i.e., randomized TVB-2640 dose groups and pooled placebo) and with the baseline MRI-PDFF value as a covariate including all subjects from US and China. If the country-by-treatment interaction is significant ($p<0.05$) then the primary analysis will be conducted on subjects enrolled in the US and will maintain sufficient power with the 90 evaluable subjects in the US (30 active in each cohort and 30 combined placebo). A separate analysis would be performed on subjects enrolled in China in the 50 mg cohort. This analysis is not powered to detect a significant difference but would be supportive/descriptive in nature to evaluate trends.

The primary efficacy analyses will be based on a F-test from the ANCOVA model in the mITT to compare each randomized TVB-2640 dose group individually with the placebo group. The fixed-sequence method, as described below, will be used to maintain an overall 0.05 Type I error rate. Data normality will be assessed and the appropriate data transformation will be applied to fulfill the underlying assumption of the ANCOVA model; if a suitable transformation cannot be determined the corresponding rank ANCOVA will become the primary analysis for this endpoint. Treatment interaction will be evaluated among the treatment groups, and if needed it

will be considered supportive of the primary efficacy analyses. Summary statistics will be displayed by treatment group along with the difference in least squares means (and the associated 95% confidence interval and p-value) for each randomized TVB-2640 dose group to pooled placebo comparison.

Differences between each randomized TVB-2640 dose group (25 mg, 50 mg) and pooled placebo for the key secondary efficacy endpoint, the percentage of subjects achieving a 30% or greater reduction of liver fat, as determined by MRI-PDFF (i.e., a response), will be analyzed using CMH methods. The key secondary efficacy analyses will be based on a CMH test in the mITT to compare each randomized TVB-2640 dose group with placebo, adjusting for the stratification factor (diabetes presence/absence). If a country-by-treatment interaction is found in the primary efficacy endpoint, the subjects from the US and China in the 50 mg cohort may be analyzed separately for the key secondary efficacy endpoint and analysis of the subjects from China will be viewed as supportive/descriptive. To check for other possible interactions, i.e., diabetes strata-dependent treatment effects, the Breslow-Day test will be performed; if the p-value of the Breslow-Day test is $\geq$0.05, the treatment-by-strata interaction is deemed not significant. However, if the interaction effect is statistically significant (i.e., p-value <0.05), then the Gail and Simon test will be used to test for the qualitative interaction at a significance level of 0.05 and provided as an aid for interpretation of the CMH results. The estimated response rates and the corresponding exact 95% confidence interval based on a binomial distribution will be calculated for each treatment group. The exact 95% confidence limit for the CMH estimate of the common risk difference will also be obtained; a Wald asymptotic approximation with continuity correction is an acceptable substitute for the exact limits in the event of computational difficulties. Response level definitions other than 30% may be explored in additional supportive analyses.

The primary and key secondary efficacy analyses will be conducted using a fixed-sequence strategy to maintain the overall Type I error rate at 0.05. The primary efficacy analyses will compare the primary efficacy endpoint between each randomized TVB-2640 dose group with the pooled placebo group, each using a 2-sided test at the 0.05 level of statistical significance with the goal of identifying the minimum effective dose. The primary efficacy analyses start with the highest dose-group comparison and if statistically significant, testing will proceed to the low dose group comparison. If both comparisons of the primary efficacy endpoint are significant, then testing will proceed to the key secondary efficacy analyses. The key secondary efficacy analyses will similarly compare the key secondary efficacy endpoint between each randomized TVB-2640 dose group with pooled placebo group, each using a 2-sided test at the 0.05 level of statistical significance. The key secondary efficacy analyses start with the highest dose-group comparison and if statistically significant, testing will proceed to the low dose group comparison.

In the primary and key secondary analyses, missing Week 12 values for MRI-PDFF will be imputed with the last post-baseline observation on or after Week 8 in the mITT. Secondary and sensitivity analyses using other methods for missing data methods and in different analysis populations will be described in the SAP.

For other continuous secondary efficacy endpoints measured at multiple post-baseline visits, the changes at 12 weeks will be summarized for each treatment group and compared between each randomized TVB-2640 dose group and pooled placebo using a linear mixed-effects model for repeated measures in the ITT. The model will include the stratification factor (diabetes

presence/absence), treatment group, visit, and treatment-by-visit interaction as the fixed effects, and baseline value as a covariate using an unstructured variance-covariance matrix. The point estimates for the least-squares mean of the pairwise difference for each randomized TVB-2640 dose group and pooled placebo comparison at each visit and the corresponding 95% confidence interval and two-sided p-value will be summarized. Other variance-covariance structures may be substituted if convergence problems arise.

Similarly, a generalized linear mixed-effects model for repeated measures based on a logit link function will be used for comparing other categorical efficacy endpoints at Week 12 between each randomized TVB-2640 dose group and pooled placebo in the ITT. The model will include the stratification factor (diabetes presence/absence), treatment group, visit, treatment-by-visit interaction as fixed effects and, depending on the nature of the data, the baseline result as fixed effect or covariate with an unstructured variance-covariance matrix. The point estimates for the least-squares mean of the pairwise difference for each randomized TVB-2640 dose group and pooled placebo comparison at each visit and the corresponding 95% confidence interval and two-sided p-value will be summarized. Other variance-covariance structure may be substituted if convergence problems arise.

Subset analyses will be performed on all efficacy endpoints for subjects enrolled in China to evaluate the consistency of results between subjects enrolled in each country.

Efficacy data will be summarized descriptively for Cohort 3.

Details for handling of data from the US and Chinese subjects will be described in the SAP. Additional details of analysis of other secondary efficacy endpoints and the analysis of exploratory and safety endpoints and pharmacokinetics will be described in the SAP.

13.2.3. Safety Analysis

No formal statistical testing will be conducted for the safety analyses. Analysis of safety will be detailed in the SAP. Subset analyses will be performed on all safety endpoints for subjects enrolled in China to evaluate the consistency of results between subjects enrolled in each country.

14. ETHICS

14.1. Good Clinical Practice Compliance

The Sponsor and any third party to whom aspects of the study management or monitoring have been delegated will undertake their assigned roles for this study in compliance with all applicable industry regulations and International Council for Harmonisation (ICH) Good Clinical Practice (GCP) Guideline E6.

The Investigator also must undertake to perform the study in accordance with ICH GCP Guideline E6 and applicable regulatory requirements and guidelines.

ICH GCP Guideline E6 is available at:

https://www.ich.org/fileadmin/Public_Web_Site/ICH_Products/Guidelines/Efficacy/E6/E6_R2__Step_4_2016_1109.pdf

14.2. Institutional Review Board (IRB)

The IRB will review all appropriate study documentation in order to safeguard the rights, safety, and well-being of the subjects. The study will only be conducted at study centers where IRB approval has been obtained. The protocol, Investigator's Brochure, informed consent, advertisements (if applicable), written information given to the subjects (including diary cards), safety updates, annual progress reports, and any revisions to these documents will be provided to the IRB by the Investigator.

The final study protocol, including the final version of the Informed Consent Form, must be approved or given a favorable opinion in writing by an IRB as appropriate. Written IRB approval must be received by Sagimet Biosciences or designee before a site can enroll any subject into the study.

The Investigator is responsible for informing the IRB of any amendment to the protocol in accordance with local requirements. In addition, the IRB must approve all advertising used to recruit subjects for the study. The protocol (and other amended study documents) must be re-approved by the IRB upon receipt of amendments and annually, as local regulations require. The Investigator is also responsible for providing the IRB with reports of any reportable serious adverse drug reactions from any other study conducted with the investigational product. Sagimet Biosciences will provide this information to the Investigator.

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

To ensure compliance with GCP and all applicable regulatory requirements, Sagimet Biosciences or designee may conduct a quality assurance audit. Please see Section 15.5 for more details regarding the audit process.

14.3. Ethical Conduct of the Study

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

14.4. Written Informed Consent

The Investigator(s) at each center will ensure that the subject is given full and adequate oral and written information about the nature, purpose, possible risk and benefit of the study. Subjects must also be notified that they are free to discontinue from the study at any time. The subject should be given the opportunity to ask questions and allowed time to consider the information provided. This process should be recorded in the subject's source documentation.

The subject's signed and dated informed consent must be obtained before conducting any study procedures. Documentation of the consenting process must be recorded in the subject's source documents.

The Investigator(s) must maintain the original, signed Informed Consent Form. A copy of the signed Informed Consent Form must be given to the subject, and this must be documented in the subject's source documents.

14.5. Subject Confidentiality

In order to maintain subject privacy, all eCRFs, study drug accountability records, study reports, and communications will identify the subject by initials (as allowed by local regulations) and the assigned subject number. The Investigator will grant monitor(s) and auditor(s) from the Sponsor or its designee and regulatory authority(ies) access to the subject's original medical records for verification of data gathered on the eCRFs and to audit the data collection process. The subject's confidentiality will be maintained and will not be made publicly available to the extent permitted by the applicable laws and regulations.

15. ADMINISTRATIVE REQUIREMENTS

15.1. Study Monitoring

Monitoring and auditing procedures developed by the Sponsor or designee will be followed, in order to comply with GCP guidelines.

Before a study center can enter a subject into the study, a representative of Sagimet Biosciences or designee will visit the study center to:

- Determine the adequacy of the facilities.
- Discuss with the Investigator(s) and other personnel their responsibilities with regard to protocol adherence, and the responsibilities of Sagimet Biosciences or its representatives. This will be documented in a Clinical Study Agreement between Sagimet Biosciences and the Investigator.

During the study, a monitor from Sagimet Biosciences or designee will have regular contacts with the study center, for the following:

- Provide information and support to the Investigator(s).
- Confirm that facilities remain acceptable.
- Confirm that the investigational team is adhering to the protocol, that data are being accurately recorded in the source documents and eCRFs, and that investigational product accountability checks are being performed.
- Perform source data verification. This includes a comparison of the data in the eCRFs with the subject's medical records at the hospital or practice, and other records relevant to the study. This will require direct access to all original records for each subject (e.g., clinic charts).
- Record and report any protocol deviations not previously sent to Sagimet Biosciences.
- Confirm AEs and SAEs have been properly documented in the eCRFs and confirm any SAEs have been forwarded to the Sponsor or designee, and those SAEs that met criteria for reporting have been forwarded to the IRB.

The monitor will be available between visits if the Investigator(s) or other staff needs information or advice.

15.2. Case Report Form Completion

The Sponsor or designee will provide the study centers with eCRFs for each subject.

eCRFs will be completed for each study subject. It is the Investigator's responsibility to ensure the accuracy, completeness, and timeliness of the data reported in the subject's eCRF. Source documentation supporting the eCRF data should indicate the subject's participation in the study and should document the dates and details of study procedures, AEs, and subject status.

The Investigator, or designated representative, should complete the eCRF as soon as possible after information is collected, preferably on the same day that a subject is seen for an

examination, treatment, or any other study procedure. Any outstanding entries must be completed immediately after the final examination. An explanation should be given for all missing data.

The Investigator must electronically sign and date the Investigator's Statement at the end of the eCRF to endorse the recorded data.

15.3. Computerized Systems / Medical Records as Source Data

All study data recorded on source documents are to be transcribed into the eCRFs. Any electronic study data are to be entered into a secure, validated data processing system and a backup maintained. Any changes to electronic study data will be documented.

15.4. Retention of Records

The Investigator will maintain all study records according to ICH GCP and applicable regulatory requirement(s). Records will be retained for at least 2 years after the last marketing application approval or 2 years after formal discontinuation of the clinical development of the investigational product or according to applicable regulatory requirement(s). If the Investigator withdraws from the responsibility of keeping the study records, custody must be transferred to a person willing to accept the responsibility. The Sponsor must be notified immediately by telephone or e-mail and the notification confirmed in writing if a custodial change occurs.

15.5. Audits and Inspections

Authorized representatives of Sagimet Biosciences or designee, a regulatory authority, or IRB may visit the study center to perform audits or inspections, including source data verification. The purpose of an audit or inspection is to systematically and independently examine all study-related activities and documents to determine whether these activities were conducted, and data were recorded, analyzed, and accurately reported according to the protocol, ICH GCP, and any applicable regulatory requirements.

The investigator should contact Sagimet Biosciences or designee immediately if contacted by a regulatory agency about an inspection.

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16. LIST OF REFERENCES

Andrade P, Rodrigues S, Rodrigues-Pinto E, Gaspar R, Lopes J, Lopes S, Macedo G. Diagnostic Accuracy of Controlled Attenuation Parameter for Detecting Hepatic Steatosis in Patients with Chronic Liver Disease. GE Port J Gastroenterol. 2017;24(4):161-168.

Beysen C, Murphy EJ, Nagaraja H, Decaris M, Riiff T, Fong A, et al. A pilot study of the effects of pioglitazone and rosiglitazone on de novo lipogenesis in type 2 diabetes. J Lipid Res 2008;49(12):2657-63.

Buckley D, Duke G, Heuer TS, O'Farrell M, Wagman AS, McCulloch W, Kemble G. Fatty acid synthase - Modern tumor cell biology insights into a classical oncology target. Pharmacol Ther. 2017;177:23-31.

Caussy C, Reeder SB, Sirlin CB, Loomba R. Non-invasive, quantitative assessment of liver fat by MRI-PDFF as an endpoint in NASH trials. Hepatology. 2018;68(2):763-772.

Clinical Study Report for Project 2006432, “Evaluation of TVB-2640, a FASN Inhibitor, to reduce de novo lipogenesis in subjects with characteristics of the metabolic syndrome”.

Cusi K. Treatment of patients with type 2 diabetes and non-alcoholic fatty liver disease: current approaches and future directions. Diabetologia. 2016;59(6):1112-20.

Donnelly KL, Smith CI, Schwarzenberg SJ, Jessurun J, Boldt MD, Parks EJ. Sources of fatty acids stored in liver and secreted via lipoproteins in patients with nonalcoholic fatty liver disease. J Clin Invest 2005;115(5):1343-51.

Duke G, Wagman AS, Buckley D, McCulloch W, Feigh M, Veidal S, et al. Fatty acid synthase inhibitor TVB-3664 reduces collagen accumulation in bleomycin-induced murine skin fibrosis and reverses multiple components of diet-induced and biopsy-confirmed nonalcoholic steatohepatitis in mice treated with or without co-administered pirfenidone and reduces collagen accumulation in bleomycin-induced murine skin fibrosis. Abstract 1994. AASLD, 2017.

Dulai PS, Sirlin CB, Loomba R. MRI and MRE for non-invasive quantitative assessment of hepatic steatosis and fibrosis in NAFLD and NASH: Clinical trials to clinical practice. J Hepatol. 2016 Nov;65(5):1006-1016.

Grundy SM, Cleeman JI, Daniels SR, Donato KA, Eckel RH, Franklin BA, et al. Diagnosis and management of the metabolic syndrome: an American Heart Association/National Heart, Lung, and Blood Institute scientific statement: Executive Summary. Crit Pathw Cardiol 2005;4(4):198-203.

Hudgins LC, Hellerstein M, Seidman C, Neese R, Diakun J, Hirsch J. Human fatty acid synthesis is stimulated by a eucaloric low fat, high carbohydrate diet. J Clin Invest. 1996;97(9):2081-91.

Kohjima M, Enjoji M, Higuchi N, Kato M, Kotoh K, Yoshimoto T, et al. Re-evaluation of fatty acid metabolism-related gene expression in nonalcoholic fatty liver disease. Int J Mol Med. 2007;20(3):351-8.

Lambert JE, Ramos-Roman MA, Browning JD, Parks EJ. Increased de novo lipogenesis is a distinct characteristic of individuals with nonalcoholic fatty liver disease. Gastroenterology. 2014;146(3):726-35.

Loomba et al. Oral Abstracts (Abstracts 1–263). Hepatology 201766:1–148.

Loomba R, Kayali Z, Noureddin M, Ruane P, Lawitz EJ, Bennett M, et al. GS-0976 Reduces Hepatic Steatosis and Fibrosis Markers in Patients With Nonalcoholic Fatty Liver Disease. Gastroenterology. 2018 Nov;155(5):1463-1473.e6.

Mitsuyoshi H, Yasui K, Harano Y, Endo M, Tsuji K, Minami M, et al. Analysis of hepatic genes involved in the metabolism of fatty acids and iron in nonalcoholic fatty liver disease. Hepatol Res. 2009;39(4):366-73.

Musso G, Cassader M, Paschetta E, Gambino R, Thiazolidinediones and advanced liver fibrosis in nonalcoholic steatohepatitis: a meta-analysis. JAMA Intern Med 2017;177(5):633-640.

Patel J, Bettencourt R, Cui J, Salotti J, Hooker J, Bhatt A, et al. Association of noninvasive quantitative decline in liver fat content on MRI with histologic response in nonalcoholic steatohepatitis. Ther Adv Gastroenterol 2016;9(5):692-701.

Postic C, Girard J. Contribution of de novo fatty acid synthesis to hepatic steatosis and insulin resistance: lessons from genetically engineered mice. J Clin Invest. 2008;118(3):829-838.

Stiede K, Miao W, Blanchette HS, Beysen C, Harriman G, Harwood HJ, et al. Acetyl-coenzyme A carboxylase inhibition reduces de novo lipogenesis in overweight male subjects: A randomized, double-blind, crossover study. Hepatology 2017;66:324–334.

Syed-Abdul MM, Parks EJ, Gaballah AH, Bingham K, Hammoud GM, Kemble G, et al. First-in-class fatty acid synthase inhibitor TVB-2640 reduces hepatic de novo lipogenesis in males with metabolic abnormalities. Hepatology. 2019 Oct 20. doi: 10.1002/hep.31000. [Epub ahead of print].

Targher G, Day CP, Bonora E. Risk of cardiovascular disease in patients with nonalcoholic fatty liver disease. N Engl J Med. 2010 Sep 30;363(14):1341-50.

Wei Y, Wang D, Topczewski F, Pagliassotti MJ. Saturated fatty acids induce endoplasmic reticulum stress and apoptosis independently of ceramide in liver cells. Am J Physiol Endocrinol Metab. 2006;291(2):E275-81.

Widya RL, de Mutsert R, den Heijer M, le Cessie S, Rosendaal FR, Jukema JW, et al. Association between Hepatic Triglyceride Content and Left Ventricular Diastolic Function in a Population-based Cohort: The Netherlands Epidemiology of Obesity Study. Radiology. 2016;279(2):443-50.

Wilke MS, French MA, Goh YK, Ryan EA, Jones PJ, Clandinin MT. Synthesis of specific fatty acids contributes to VLDL-triacylglycerol composition in humans with and without type 2 diabetes. Diabetologia. 2009;52(8):1628-37.

17. APPENDICES

Appendix A: Algorithm for Evaluating Subjects for a Potential Drug-induced Liver Injury (DILI)

The mean of at least 2 liver aminotransferase values taken at least 4 to 12 weeks apart during screening should be used to establish baseline values; the first measurement may be a historical measurement.

During the study, aminotransferase elevations are to be managed as follows:

1. Isolated aminotransferase elevation (i.e., total bilirubin - normal):

ALT or AST	Event during the study	Action
Baseline ALT and AST within normal range	Increase ≥5 × ULN	Repeat liver profile (AST, ALT, bilirubin and PT/INR) within 2 to 3 days. Monitor the subject as per "close observation" definition* in the DILI guidance.
ALT or AST above normal range at baseline	increase ≥3 × baseline levels and no liver related symptoms	
ALT or AST above normal range at baseline	increase ≥3x baseline + subject experiences liver related symptoms (nausea, vomiting, right upper quadrant pain)	Interrupt study drug. Initiate potential DILI evaluation for alternative etiologies and repeat liver profile and PT/INR within 48-72 hours and closely observe the subject.

*Close observation as defined in the FDA DILI guidance i.e., laboratory testing and physical examination 2-3 times per week. Study drug is to be restarted only if a firm alternative etiology is "identified" and the liver tests return to baseline.

2. Elevation of aminotransferase and total bilirubin or cholestatic markers:

Analyte	Event during the study	Action
TB + AST or ALT	↑ TB >2 mg/dL and ↑ ALT or ALT ≥3x baseline or 5x ULN (whichever comes first)	Interrupt study drug. Initiate potential DILI evaluation for alternative etiologies. Repeat liver profile and PT/INR within 48-72 hours and place subject under "close observation" as defined in the DILI guidance. Study drug can be restarted only if a firm competing etiology is identified and the liver tests return to baseline.
TB, ALP, or GGT	↑ TB >2 mg/dL and ALP or GGT ↑ >2X ULN	

Analyte	Event during the study	Action
TB OR INR + symptoms (regardless of any magnitude AST/ALT elevation)	Elevation ↑ TB >2 mg/dL OR INR ≥1.5 and Indicators of immunological reaction (i.e., rash or >5% eosinophilia), or appearance of nausea, vomiting, right upper quadrant pain (symptoms consistent with clinical hepatitis)	Permanently discontinue study drug. Initiate potential DILI evaluation for alternative etiologies. Repeat liver profile and PT/INR within 48-72 hours and place subject under "close observation" as defined in the DILI guidance.

Note the following:

1. If a subject lives in a remote area, they can be tested locally, with the results communicated to the Investigator promptly.
2. For subjects with Gilbert's syndrome, a doubling of direct bilirubin instead of total bilirubin should be used for close monitoring and TVB-2640 discontinuation.
3. If the following criteria are met, study drug must be discontinued and the subject must be followed until the clinical and laboratory abnormalities stabilize or normalize:
   - If close monitoring of a subject is not possible.
   - In the presence of total bilirubin elevation (>2 × ULN or >1.5 × baseline); with any degree of aminotransferase elevation; AND if there is appearance of symptoms i.e., fatigue, nausea, vomiting, right upper quadrant pain or tenderness, fever, rash, and/or eosinophilia (>5%).
   - If any degree of total bilirubin, ALT, or AST elevation recurs following re-challenge with study drug.