A PHASE 2A, RANDOMIZED, DOUBLE-BLIND, PLACEBO-CONTROLLED, DOSE-RANGING, PARALLEL GROUP STUDY TO EVALUATE SAFETY, TOLERABILITY, AND PHARMACODYNAMICS OF PF-05221304 ADMINISTERED DAILY FOR 16-WEEKS TO ADULT SUBJECTS WITH NONALCOHOLIC FATTY LIVER DISEASE

Investigational Product Number:	PF-05221304
Investigational Product Name:	Not applicable
United States (US) Investigational New Drug (IND) Number:	[redacted]
European Clinical Trials Database (EudraCT) Number:	2017-001156-55
Protocol Number:	C1171002
Phase:	2a

[redacted]

Document History

Document	Version Date	Summary of Changes and Rationale
Amendment 2	03 October 2017	This amendment is making the following substantial changes to align with the Pfizer Enterprise-level revision to appropriate measures to prevent pregnancy in the population of childbearing potential enrolled who are sexually active ***and*** align with the Clinical Trial Facilitation Group (CTFG) 2014 European Guidance– • Section 4.4.6 revised to – 1. Remove mandate to use highly effective contraception in female partners of male subjects of childbearing potential; CCI [redacted] 2. Use of male or female condom alone is viewed as an effective method of contraceptive but must continue to be used with another highly effective method of contraceptive; • Language mandating use with spermicide has been removed since the intent is to combine use of condom with another highly-effective method of contraception; • The following administrative changes to enhance clarity and ensure intended interpretation of protocol content have also been incorporated – • Section 4.2, exclusion criterion #9, correct inconsistency and permit at both Pre-Qualification and Screen 1 visits (not only Screen 1) repeat laboratory-related assessments to assess eligibility and depict intent to exclude those who appear to have been cured of Hepatitis C viral infection by pharmacological intervention; • Section 5.8.5 updated to remove use of symbol and replace with text to delineate that list of medications taken within (*not* prior to) 8 weeks of Visit 1 are prohibited; • Section 5.8.5 updated to clarify that anti-convulsants specifically prescribed for management of seizures are *not* permitted (though use for other indications *may* be permitted); • Section 5.8.5 updated to clarify that β-blockers and calcium channel blockers are permitted, while other antiarrhythmic agents (eg, those that function primarily as sodium and potassium channel blockers) are *not* permitted; • Typographical error in Section 7.2.1 corrected to capture that Visit 11 is Week 16 (*not* Week 11); CCI [redacted] • Figure 2 and Section 7.9.1 further clarified (beyond Enterprise-level text in Section 8.2.2) and by extension in Appendix 2, that in those subjects with observed platelet count below lower limit of normal by Week 8, additional (unplanned) visits every two weeks should continue while on double-blinded investigational product (ie, add unplanned visits at Weeks 10, and 14, in addition to already scheduled visits at Weeks 12, and 16) with blood samples collected

Document	Version Date	Summary of Changes and Rationale
		to permit safety-related laboratory parameters including hematology by sponsor identified central laboratory; • Appendix 2 and Appendix 3 updated to reflect removal of "scheduled" as visits for assessment could also be unplanned.
Amendment 1	24 July 2017	This amendment is making the following substantial changes as requested by the United States Food and Drug Administration (US FDA) as part of their review of the original protocol submitted on 11May2017 – • In addition to conducting standard full and brief physical examinations, targeted physical examinations of skin, eyes, and pulmonary system were added at predose at Weeks 2, 8, and 16 – refer to Table 1 and Section 7.2.1CCI [redacted] • Section 3.2 updated to include explicit cross-reference to detailed Enterprise-level standardized management of AEs, SAEs, and cases of DILI; • Section 3.3 updated to offer instances wherein unblinded safety review by IRC will be triggered based on-going blinded safety reviews by selected members of the Sponsors' study team; • Section 5.8.5 updated to exclude CYP3A4/5 substrates with narrow therapeutic indices CCI [redacted] • Section 7.9.3 expanded to include language expecting subjects with type 2 diabetes mellitus to share the low blood glucose information sheet which delineates the symptoms of hypoglycemia along with treatment options with their family member(s) / roommate(s) to help in their management of hypoglycemia; • While PF-05221304 is not known to carry the potential risk of hypoglycemia, many of the agents used for the management of glycemic control in patients with T2DM carry this risk - this rationale this added to Section 1.3.2; • *In addition*, based on determination by the Central IRB for sites in Canada and USA, that informed consent must be offered by the participating subject *only*, via this amendment, at the study-level, a requirement is being instituted that informed consent for both pre-qualification and main study, be offered by the participating subject, ***only*** • The provision that permits subjects' legally acceptable representative to offer informed consent has been deleted (in Sections 4.1, 8.1.2, 8.1.4, and 12.3). • Administrative clarifications added to Section 7.1.2 regarding FibroScan® operators; and Appendix 4 to capture time of onset and resolution of hypoglycemic event rather than time to recovery to match Sponsors' new standard for capture of these events.
Original protocol	04 May 2017	Not applicable

This amendment incorporates all revisions to date, including amendments made at the request of country health authorities and institutional review boards (IRBs)/ethics committees (ECs).

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

APPENDICES

PROTOCOL SUMMARY

Background and Rationale: The current study is the first clinical trial proposed with PF-05221304 in adult subjects with NAFLD. The investigational product will be administered orally, once-daily for up to 16 weeks. It is designed as a dose-ranging trial with placebo and 4 active doses of PF-05221304 to assess the safety, tolerability and the effect of PF-05221304 on liver fat. In addition, assessment of the effect of PF-05221304 on other pharmacodynamics/ exploratory parameters is planned.

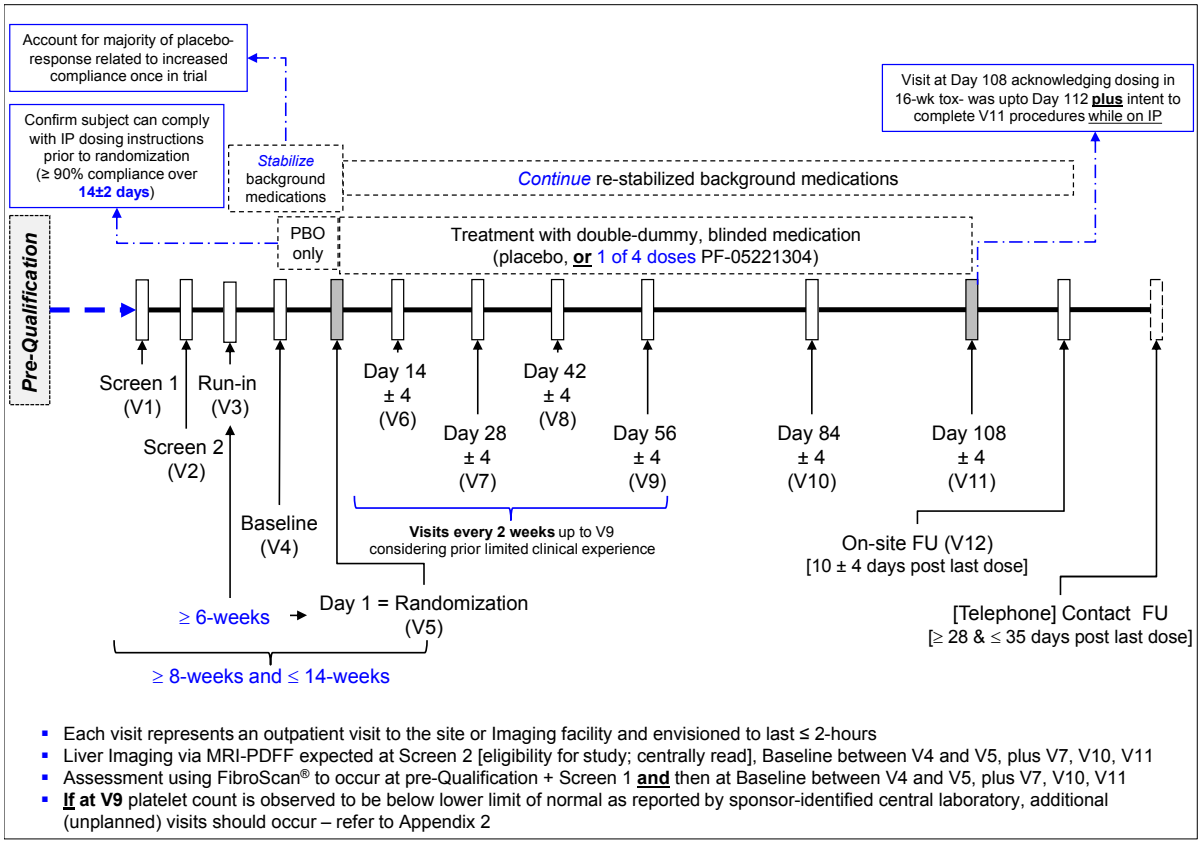
Objectives and Endpoints:

Primary Objective:	**Primary Endpoint:**
To evaluate the dose-response for the effect on ***liver fat*** with a range of PF-05221304 doses administered daily in adults ***with nonalcoholic fatty liver disease (entire study population)***	Percent change from baseline in liver fat, as assessed using MRI-PDFF, at Week 16
Secondary Objective(s):	**Secondary Endpoint(s):**
To evaluate the dose-response for the effect on ***ALT*** with a range of PF-05221304 doses administered daily in the 1st tier stratification comprising of adults ***with diagnosed/presumed nonalcoholic steatohepatitis, only***	Percent change from baseline in ALT, at Week 16
To evaluate the ***safety and tolerability*** of a range of PF-05221304 doses administered daily in adults ***with nonalcoholic fatty liver disease (entire study population)***	Assessment of TEAEs, safety-related clinical laboratory tests, vital signs, and 12-lead ECGs
Tertiary Objective(s):	**Tertiary Endpoint(s):**
To evaluate the dose-response for the effect on liver fat, ***over time***, with a range of PF-05221304 doses administered daily in adults ***with nonalcoholic fatty liver disease (entire study population)***	Percent change from baseline in liver fat, as assessed using MRI-PDFF, over time up to Week 16
CCI [redacted]	
To evaluate the dose-response for the effect on key liver function tests and NASH-related biomarkers ***over time***, with a range of PF-05221304 doses administered daily in the 1st tier stratification comprising of adults ***with diagnosed/presumed nonalcoholic steatohepatitis, only***	Percent change from baseline, over time up to Week 16, for: • ALT, AST, Alkaline Phosphatase, GGT • CK18-M30 and CK18-M65 CCI [redacted] • Pro-C3 & Pro-C6

Tertiary Objective(s):	Tertiary Endpoint(s):
CCI	
To evaluate the dose-response for the effect on other potentially mechanism-related parameters and metabolic parameters, ***over time***, with a range of PF-05221304 doses administered daily in adults with ***nonalcoholic fatty liver disease (entire study population)***	Percent change from baseline, over time up to Week 16, for: • Potentially mechanism-related parameters – – Serum apolipoprotein A1, B (total), C3, E CCI • Metabolic parameters – – HbA1C – Fasting lipid panel (fasting total cholesterol, direct LDL-C, HDL-C, triglycerides, VLDL) CCI
To evaluate the dose-response for the effect on glycemic parameters, ***over time***, with a range of PF-05221304 doses administered daily in the 2nd tier stratification comprising of ***adults with T2DM, only***	Change from baseline, over time up to Week 16, for: • HbA1C • FPG • FPI • HOMA-IR
CCI	

For ***all*** endpoints, baseline defined as result closest prior to dosing at Visit 5 (Day 1); for list of terms corresponding to the abbreviations used herein, refer to Appendix 1.

Study Design: This is a randomized, double-blind, placebo-controlled, 5-arm (placebo, plus 4 active doses of PF-05221304), parallel-group study. The overall study design is summarized in the figure below:



Study Treatments: Each dose will consist of 3 tablets (1 tablet from each of the 3 bottles dispensed) as outlined below:

Regimen	Regimen Description (dosed once-daily)	Number of PF-05221304 tablets				Number of PF-05221304-matching placebo tablets	
		1 mg	5 mg	25 mg	50 mg	1 / 5 mg	25 / 50 mg
A	Placebo	-	-	-	-	2	1
B	PF-05221304 – 2 mg	2	-	-	-	-	1
C	PF-05221304 – 10 mg	-	2	-	-	-	1
D	PF-05221304 – 25 mg	-	-	1	-	2	-
E	PF-05221304 – 50 mg	-	-	-	1	2	-

Study Population: In the Phase 2a study, the entire study population will have NAFLD (liver fat ≥ 8% by MRI-PDFF as part of screening assessment) and features of metabolic syndrome. The population enrolled will be stratified into two sub-populations as follows:

- **Subjects with diagnosed or presumed NASH:** These subjects would either have been diagnosed with NASH on liver biopsy (within the prior ≤ 24 months) **or** would meet imaging and clinical characteristics highly suggestive of NASH in the absence of a liver biopsy;

- **Subjects with NAFLD with likely minimal inflammation and fibrosis:** These subjects will be classified based on evidence of ALT ≤ 1.25x upper limit of normal reference range (ULN) for the sponsor-identified central laboratory, and no/minimal evidence of liver fibrosis (as measured by Vibration-Controlled Transient Elastography [VCTE™] and reported as liver stiffness measure [LSM, in kilopascals, kPa] using FibroScan®). This population is considered a distinct, albeit related population, to those categorized as diagnosed/presumed NASH; and included as a means to more broadly assess general safety (including potential mechanistic effects on platelets) and additional potential beneficial effects of ACC inhibition (such as improvement in glycemic control), then could be readily achieved via limiting the population to only subjects with NASH.

***In each arm*,** ≥ 50% of subjects randomized will be deemed to be eligible for the sub-population classified as diagnosed/presumed NASH; with the remaining subjects per arm eligible for the stratum defined as NAFLD with likely minimal inflammation and fibrosis.

Statistical Methods: The proposed sample size for this study is approximately 360 subjects randomized (72 subjects per arm) to ensure at least 300 subjects (60 subjects per arm) complete the study. The sample size of a minimum of 60 completers per arm, was calculated to yield ≥ 80% power to detect a placebo-adjusted, percent change from baseline of ≥ 30% in liver fat (in entire study population *and* in n=30/arm in each sub-population of the 1st tier stratification), using a 1-sided, t-test at level 10%. For the secondary endpoint of ALT in the 1st tier stratification sub-population of diagnosed/presumed NASH, the sample size of a minimum of 30 completers per arm was calculated to yield ≥ 80% power to detect a placebo-adjusted, percent change from baseline in ALT of ≥ 35%, using a 1-sided, t-test at level 10%.

For the primary and key secondary endpoint of placebo-adjusted, percent change from baseline in liver fat via MRI-PDFF (in entire study population) and ALT (in the sub-population of diagnosed/presumed NASH) at Week 16, a mixed model repeated measure (MMRM) analysis will be performed with treatment, time and treatment-by-time interaction as fixed effects, and subject as a random effect. Descriptive summaries of the observed values and percent change from baseline in liver fat in each treatment group at each time point will be produced. For statistical analyses, 80% confidence intervals will be produced for the least-square mean differences in each treatment group as compared to placebo. One sided p-values will be displayed for the comparison against placebo with no multiplicity adjustment made.

Similar analyses will be applied to continuous secondary and tertiary endpoints, as appropriate to the data. Descriptive summaries will be displayed for categorical secondary and tertiary endpoints.

SCHEDULE OF ACTIVITIES: TABLE 1 and **TABLE 2** provide an overview of the protocol visits and procedures. Refer to Section 6 and Section 7 of the protocol for detailed information on each procedure and assessment required for compliance with the protocol. The investigator may schedule unplanned visit(s) *in addition* to those listed in **Tables 1** and **2**, in order to conduct evaluations or assessments required to protect the well-being of the subject.

Table 1. Study-Related Procedures in C1171002

Visit Identifier [for abbreviations refer to Appendix 1]	**Pre-Qualification**	**Screen 1**	**Screen 2**	**Run-in**	**Baseline**	**Treatment Phase** [all procedures *before* morning dose of blinded IP, except liver fat & stiffness assessments]							**Follow-up**		**Early Termination**
Weeks Relative to Dosing on Day 1					**-2**	**0**	**2**	**4**	**6**	**8**	**12**	**16**	**17-18**	**20[a]**	
Days relative to Dosing on Day 1					**-14± 2**	**1**	**14±4**	**28±4**	**42±4**	**56±4**	**84±4**	**108±4**	**--**	**--**	
Visit		**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**--**	
Informed consent (x2[b]) & demography	x	x													
(Update) Medical & Medication history	x	x		x	x	x	x	x	x	x	x	x	x	x	x
Liver fat & stiffness (via FibroScan®)[c]	x	x			x[d]			x			x	x[d]			
Liver fat (via MRI-PDFF)[e]			x[f]		x[d]			x			x	x[d]			
Physical Exam[g]	x	x			x		x			x		x	x		x
Assess alcohol intake (AUDIT questionnaire)	x	x				x[h]						x			
Assess correct use of contraception				x	x	x	x	x	x	x	x	x	x	x	x
Counseling on diet/exercise guidelines				x		x									
Train on use and dispense glucometer & ancillary supplies (T2DM subjects, *only*)				x											
Open-ended inquiry for adverse events	x	x	x	x	x	x	x	x	x	x	x	x	x	x	x
Body weight (+ hula-hoop test at pre-Q)	x	x			x	x	x	x	x	x	x	x	x		
Single supine 12-lead ECG	x	x			x	x	x	x	x	x	x	x	x		x
Single seated vitals (BP & pulse rate)	x	x			x	x	x	x	x	x	x	x	x		x
Registration in trial (via IRT)	x			x											
Randomization in trial (via IRT)						x									
Dispensation[i] (via IRT) of IP					x	x		x		x	x				
Witnessed dosing on site of IP					x	x[h]	x	x	x	x	x	x			
Compliance via pill count *on site* of ***returned*** IP						x[h]		x		x	x	x			
Continued administration of IP					x[i]	x[i]	→	→	→	→	→	x[j]			x[j]

a. To occur via telephone contact (unless local regulations dictate on-site visit); ***regardless***, on-site visit to be considered for follow-up of open TEAEs and/or abnormal laboratory tests
b. Subjects to provide consent for pre-qualification first; and once deemed likely eligible requested to provide ***separate*** consent for main study (V1 and on)
c. Assessment to be performed following ≥ 4-hr fast, at site (as part of site visit) or separate visit(s) to Imaging facility; attempts to be made to stay within ± 2-hr window of V1 time
d. Assessment to be performed on any day between V4 & V5 (ie, Baseline); and while subject is taking blinded IP with previous dose ≤ 24-hr prior (at V11)
e. Assessment to be performed following ≥ 4-hr fast, via separate visit(s) to Imaging facility; attempts to be made to stay within ± 2-hr window of V2 time
f. A single repeat assessment permitted as determined by sponsor-identified central imaging vendor; subject must ***not*** progress to V3 until results from screen-repeat confirm eligibility
g. Includes arm circumference (at pre-Q); height *plus* waist circumference (at pre-Q & V1); targeted PE at V6, V9, V11 and brief PE for open AEs/abnormal tests, at investigator discretion
h. Review of AUDIT questionnaire responses must be deemed acceptable, **and** ≥ 90% compliance (with pills in *each* bottle), required to progress to randomization
i. At V4, reflects single-blind placebo; from V5 onwards, reflects double-blind randomized regimen
j. At V11 and ET visits reflects return to site of any remaining unused double-blind randomized regimen; subjects will *not* be offered IP post last dose in this trial

Table 2. Study-Related Blood and Urine Collections in C1171002

Visit Identifier [for abbreviations refer to Appendix 1]	**Pre-Qualification**	**Screen 1**	**Screen 2**	**Run-in**	**Baseline**	**Treatment Phase** [all procedures *before* morning dose of blinded IP, except when specified otherwise]							**Follow-up**		**Early Termination**
Weeks Relative to Dosing on Day 1					**-2**	**0**	**2**	**4**	**6**	**8**	**12**	**16**	**17-18**	**20**[a]	
Days relative to Dosing on Day 1					**-14± 2**	**1**	**14±4**	**28±4**	**42±4**	**56±4**	**84±4**	**108±4**	**--**	**--**	
Visit		**1**	**2**	**3**	**4**	**5**	**6**	**7**	**8**	**9**	**10**	**11**	**12**	**--**	
Blood collection, after ≥ 8-hour overnight fast, for – [refer also to Table 4 for comprehensive list of analytes]															
– Hematology, chemistry, coagulation, HbA1C	x[b]	x[b]			x	x	x	x	x	x	x	x	x	x[d]	x
– Fasting lipid panel	x[b]	x[b]			x[c]	x	x	x	x	x	x	x	x	x[d]	x
– Pregnancy (females only)	x[b]	x[b]			x[c]			x		x	x	x	x	x[d]	x
– FSH (females only), Serology (HBsAg, HBcAb, HCVAb, HIV), α1-antitrypsin, ceruloplasmin	x[b]	x[b]													
– % carbohydrate deficient transferrin	x[b]	x[b]			x	x						x			
CCI					CCI										
– Apolipoproteins					x	x		x		x	x	x			
– CK18-M30, CK18-M65, Pro-C3, and Pro-C6					x	x		x		x	x	x			
CCI					CCI										
Spot urine collection for –															
– Urine drug test	x[b]	x[b]			x[c]										
– Urinalysis (and microscopy, as appropriate)	x[b]	x[b]			x	x	x	x	x	x	x	x	x	x[d]	x
– On-site pregnancy test (WOCBP, only)					x[g]	x[g]		x[g]		x[g]	x[g]	x[g]	x	x[d]	x

No Blood *or* Urine Collections

a. To occur via telephone contact (unless local regulations dictate on-site visit); ***regardless***, on-site visit to be considered for follow-up of open AEs and/or abnormal laboratory tests
b. Test results must be reviewed by a medically qualified site staff and deemed acceptable *before* progression to next visit (pre-Q results prior to V1; and V1 results prior to V2)
c. Test results must be reviewed by a medically qualified site staff and deemed acceptable *before* progression to randomization and first dose of double-blinded IP, at V5
d. Collections to occur only if visit is an on-site visit; collections to be skipped if visit is via telephone contact

CCI

f. If not collected on the designated collection day, collect at the next available time point when biospecimens are being collected in conjunction with a subject visit
g. *At each of these visits*, the test result must be reviewed by a medically qualified site staff and deemed acceptable, in order to continue participation in study

1. INTRODUCTION

Acetyl-CoA carboxylase (ACC) is a biotin carboxylase that catalyzes the adenosine triphosphate (ATP) dependent condensation of acetyl-CoA and carbonate to form malonyl-CoA.[1] The malonyl-CoA produced by ACC serves two major physiologic functions. It is an essential and rate-limiting substrate for *de novo* lipogenesis (DNL) and it acts as an allosteric inhibitor of the enzyme carnitine-palmitoyltransferase 1 (CPT-1). CPT-1 is responsible for the transport of long-chain fatty acyl-CoAs across the mitochondrial membrane into the mitochondria where they become available for fatty acid oxidation. The transport step is rate-determining for this process. Thus, ACC is positioned as a key physiologic switch regulating the transition from oxidative to lipogenic metabolism. Reducing malonyl-CoA production via ACC inhibition is expected to inhibit simultaneously the *de novo* synthesis of fatty acids and to increase flux through CPT-1 leading to increased β-oxidation of long-chain fatty acids. There are two closely related isoforms of ACC, ACC1, and ACC2, encoded by separate gene products that differ in tissue and cellular distribution.

PF-05221304 is a potent, selective, orally bioavailable, and reversible dual ACC1/2 inhibitor designed to have asymmetric distribution to the liver, with ≥ 100 fold asymmetric hepatic distribution demonstrated in both rats and monkeys; as such PF-05221304 is expected to inhibit DNL and stimulate fatty acid oxidation in the liver to a greater extent than in peripheral tissues. In humans, administration of PF-05221304 has been shown to suppress hepatic DNL in a Phase 1 trial in healthy subjects; in addition, the drug is expected to stimulate hepatic fatty acid oxidation, and consequently reduce fat accumulation in the liver. This inhibition of hepatic DNL is postulated to result in a decrease and normalization of the excessive DNL observed in nonalcoholic fatty liver disease (NAFLD). In addition, the inhibition of ACC via administration of PF-05221304 also has the potential for anti-inflammatory effects in nonalcoholic steatohepatitis (NASH).

The current study is the first clinical trial proposed with PF-05221304 in adult subjects with NAFLD. The investigational product will be administered orally, once-daily for up to 16 weeks. It is designed as a dose-ranging trial with placebo and 4 active doses of PF-05221304 to assess the safety, tolerability and the effect of PF-05221304 on liver fat. In addition, assessment of the effect of PF-05221304 on other pharmacodynamics/exploratory parameters is planned.

1.1. Mechanism of Action/Indication

PF-05221304 is a potent, selective, orally bioavailable, and reversible dual ACC1/2 inhibitor, designed to have asymmetric distribution to the liver. It is being developed for the treatment of NASH with fibrosis.

1.2. Background

The World Health Organization lists NAFLD and NASH as the most important of conditions contributing to the global health burden due to liver diseases, with NASH acknowledged as a potentially fatal condition leading to cirrhosis, liver failure, and hepatocellular carcinoma (HCC).[2] NASH is diagnosed clinically by liver biopsy demonstrating steatosis, inflammation, and cytological ballooning of liver hepatocytes, often with varying degrees of

fibrosis. NASH progresses with increasing degrees of fibrosis, with cirrhosis developing in a subset of patients[3] with the most common complication of cirrhosis being HCC.[4] Patients with NASH may be asymptomatic or have non-specific symptoms such as fatigue, despite having significant disease on liver biopsy and associated risk for progression to cirrhosis and liver-related mortality.

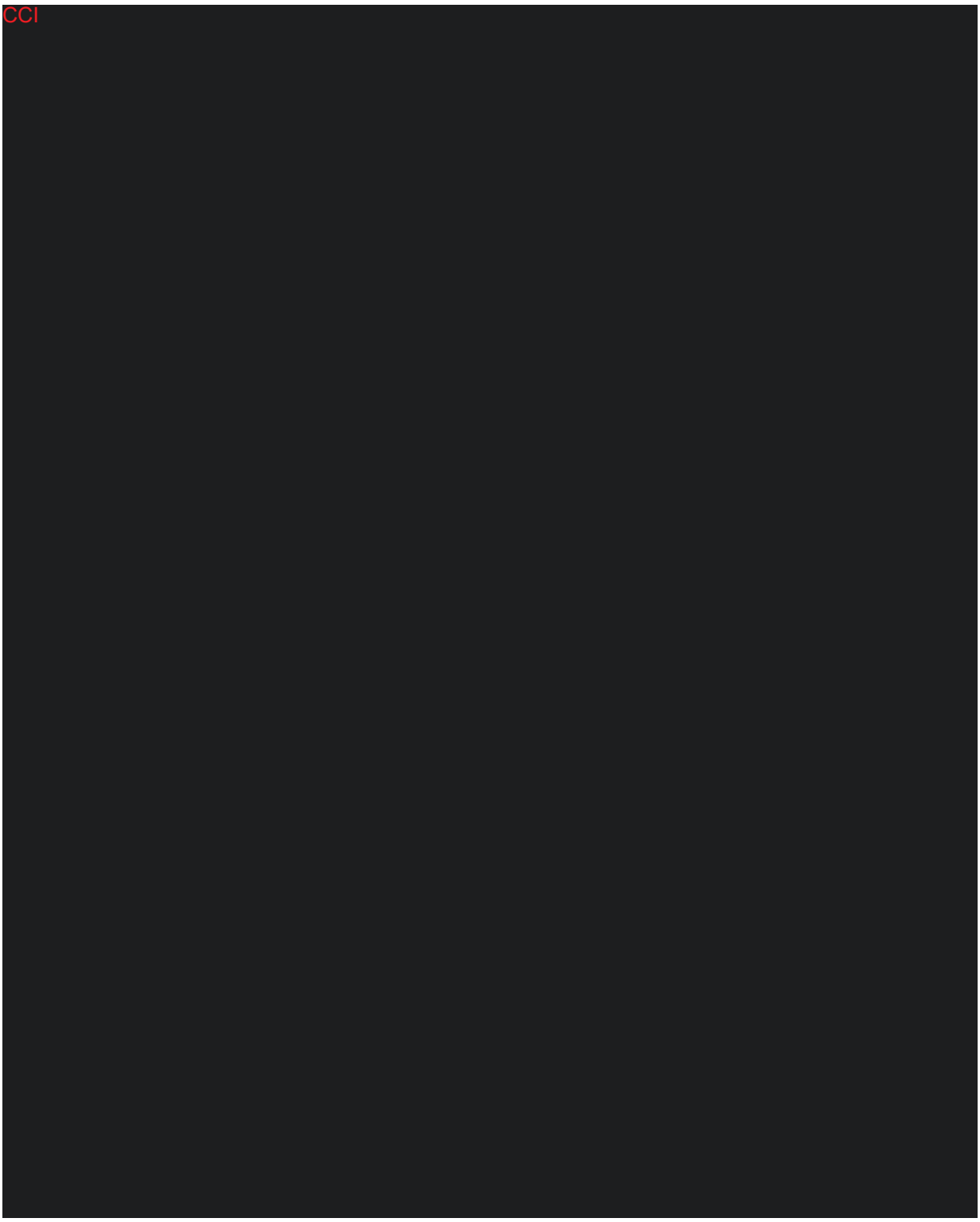
NASH is a clinical and histological subset of NAFLD (defined as presence of ≥ 5% hepatic steatosis) that is associated with increased all-cause mortality, cirrhosis and end-stage liver disease, increased cardiovascular mortality, and increased incidence of both liver related and non-liver related cancers.[3] In a recent meta-analysis, the global prevalence of NAFLD is estimated at 25%, with the prevalence of NASH in the subset with biopsy-proven NAFLD assessed at 59%.[5] The majority of the population with NAFLD has simple steatosis which has, in general, a benign clinical course. A proportion of patients with NAFLD progress to having hepatocellular ballooning and lobular inflammation – taking close to a decade to progress from 1 stage to the next and 30-40 years to develop cirrhosis; however, a smaller subset of patients progress very rapidly (within 10 years) to liver cirrhosis from NAFLD.[6] The 5-year (67%) and 10-year (38%) survival rates in patients with NASH is significantly different than in those with NAFLD.[7] The pooled liver-specific and overall mortality incidence rate estimates among those with NAFLD were calculated at 0.8 and 15.4, respectively, per 1,000 person-years. In contrast, amongst the population with NASH, the incidence rate estimates were 11.8 (liver-specific) and 25.6 (overall) mortality.[5]

Alterations in lipid metabolism have been hypothesized to contribute to the molecular pathogenesis of NAFLD and NASH. Elevated rates of hepatic DNL have been reported to be a distinctive characteristic of NAFLD (defined as liver fat ≥ 5%).[8] Humans with elevated liver fat have a greater than 3-fold increase in the rate of hepatic DNL relative to subjects with normal liver fat, but differences between the groups were not detected in adipose free fatty acid (FFA) flux or in production of very low density lipoprotein (VLDL) from FFAs. Consequently, when comparing human subjects with steatosis to those with normal liver fat, the contribution of hepatic DNL was most significantly elevated in subjects with high liver fat.[8] ACC inhibition is expected to suppress hepatic DNL and stimulate hepatic fatty acid oxidation, and consequently reduce steatosis.[9] Partial inhibition of this enzyme activity would be expected to lead to such improvements, with 70% hepatic DNL inhibition normalizing the elevated flux in subjects with NAFLD to that observed in individuals with normal liver fat.[8] Consistent with this, beneficial effects of pharmacological ACC inhibition on liver fat in rodent models of NAFLD/NASH have been reported.[9,10,11,12]

Emerging data also suggest that suppression of DNL through ACC inhibition may also directly reduce inflammation by restraining the formation of the inflammatory interleukin-17 secreting T cells of the T helper 17 lineage (Th17) cells which in turn promote the development of anti-inflammatory Foxp3(+) regulatory T (Treg) cells.[13] Studies comparing patients with NASH versus those with simple steatosis or healthy liver have demonstrated that progression from NAFLD to NASH is marked by a higher frequency of Th17 cells in the liver and increase in the ratio of Th17 cells to Treg cells in peripheral blood and in the liver.[14]

CCI

CCI



1.2.2. Clinical Experience with PF-05221304

As of May-2017, a single clinical study (C1171001) has been conducted with PF-05221304. A total of 96 (93 male and 3 female) healthy adult subjects were randomized in this one study. Overall, 82 subjects (85%) have been exposed to at least a single oral dose of PF-05221304; with 56 of these subjects exposed to repeated doses of PF-05221304 for up to 14-days. Of these, 16 subjects received single oral doses of PF-05221304 ranging from 1 mg to 240 mg – with each single dose of PF-05221304 evaluated in at least 6 subjects. An additional 70 subjects were exposed to up to 14-days of repeated oral dosing of PF-05221304 (n=56 subjects; up to 8 subjects per dose) or placebo (n=14 subjects, total) ranging from total daily doses of 2 mg/day to 200 mg/day – administered every 12 hours (Q12H) in 5 cohorts and once-daily (QD) in 2 cohorts – to assess safety, pharmacokinetics (PK), and steady-state effect on marker of target engagement (ie, inhibition of DNL). Ten more subjects received single oral dose (100 mg) in 2 periods with and without a high-fat/high-caloric morning meal to assess the effect of food on plasma exposure of PF-05221304.

In Study C1171001, single oral doses (up to 240 mg) and repeated total daily doses (up to 200 mg/day) were found to be well tolerated with an acceptable safety profile. Over this dose range evaluated, while the maximum tolerated dose (MTD) was *not* identified, higher doses were not assessed either because the a priori identified PK stopping limit was achieved (single dose) *or* near complete inhibition of hepatic DNL was observed (upon repeated dosing).

In the clinical program to date, there have been no serious adverse events (SAEs), no treatment-emergent adverse events (TEAEs) of 'severe' intensity, and no apparent dose-related increase in frequency or severity of TEAEs across the 240-fold (single) and 100-fold (repeated) dose range studied. All of the TEAEs were 'mild' in intensity except 1 TEAE of 'platelet count decreased' deemed to be of 'moderate' intensity; this resulted in premature withdrawal of 1 subject following 13 (of 14) days of PF-05221304 dosing at 100 mg Q12H (200 mg/day), the highest repeated dose evaluated. This 'moderate' intensity TEAE comprised of a gradual decline in platelet count from a baseline of 167,000/mm^3 to 114,000/mm^3 at 24-hours post last dose (on Day 13). This subject was asymptomatic with no signs or symptoms of platelet dysfunction and the decrease in platelet count did not meet criteria for thrombocytopenia (defined as < 100,000/mm^3). The platelet count increased towards baseline by 72 hours post last dose and was back to baseline by 10 days post last dose.

Upon repeated dosing, the only TEAE reported by more than 2 subjects exposed to PF-05221304, across the entire dose range evaluated, was headache. CCI [redacted] Of note, no dose related trends in venipuncture bruising-related TEAEs (eg, contusions) were noted despite 51 direct venipunctures (for serial blood collections) performed per subject during the inpatient stay phase of the repeated dosing portion of this study.

Across the other safety-related data collected, some asymptomatic, drug-related, laboratory parameter changes were observed. This included a gradual increase in fasting and post-prandial serum triglycerides with repeated doses ≥ 40 mg/day with effect plateauing over the 14-day dosing period. CCI [redacted] At the doses ≥ 60 mg/day, a gradual decrease in platelet count was observed with repeated doses over the 14 day dosing period. This observation was not noted at PF-05221304 doses up to 40 mg/day and no signs or symptoms of platelet dysfunction were apparent based on the safety assessments performed. There were no apparent abnormalities noted in the other safety-related assessments such as blood pressure, and cardiac conduction intervals [assessed on 12-lead electrocardiograms (ECGs)].

The plasma PK of PF-05221304 following oral administration suggests a moderate rate of absorption with a median time to maximum plasma concentration (T_{max}) ranging from 3 to 5 hours, following repeated dosing with the morning meal. Food (high-fat/high-caloric meal) had no clinically relevant effect on exposure which decreased slightly (area-under-the-curve to infinity (AUC_{inf}) by ~10% and C_{max} by ~25%, while rate of absorption was delayed with T_{max} of 2.5 hours (dosing in fasted state) moving out to 5 hours (when dosing occurred with food). After attainment of C_{max}, the disposition of PF-05221304 showed a decline with the half-life of PF-05221304 being independent of dose, and ranging from approximately 13 to 18 hours. With Q12H dosing frequency, an approximate 2-fold accumulation was observed which reduced to approximately 1.5-fold with QD dosing frequency. Across the range of repeated doses evaluated (2 mg/day to 200 mg/day), the plasma exposure increased approximately dose-proportionally with increasing dose. Less than 1% of the oral dose of PF-05221304 was excreted unchanged in the urine indicating that renal clearance is not a major clearance mechanism for PF-05221304.

1.3. Rationale for Study

The current study is the first trial specifically designed to evaluate the effect of PF-05221304 (compared to placebo) on liver fat, as assessed via magnetic resonance imaging proton density fat fraction (MRI-PDFF), and other pharmacodynamic/exploratory parameters in adults with NAFLD. The investigational product will be administered orally, once-daily for up to 16 weeks.

1.3.1. Rationale for Design

Along with an assessment of effect of PF-05221304 on liver fat, this study is designed to assess the safety and tolerability of a range of oral doses of PF-05221304 compared to placebo. Eligible population is defined as having liver fat ≥ 8% as measured by MRI-PDFF at Screen 2 ***and*** evidence of concomitant medical conditions, including features of metabolic syndrome, albeit stable with intent to stratify the population enrolled (refer to Section 1.3.2 and Section 4.3 for details). While NAFLD is typically defined by liver fat ≥ 5%, entry criteria at Screen 2 is proposed at ≥ 8% to account for a potential drift in liver fat during the run-in/stabilization period and negate the need for a randomization criteria related

to liver fat assessment between Visit 4 (Day -14) and Visit 5 (Day 1; randomization). A run-in period of ≥ 6-weeks post confirmation of eligibility and prior to randomization (ie, Visit 3 to Visit 5) is specifically included to permit stabilization of background/concomitant medications and achievement of pre-randomization baseline.

Overall, this study includes two distinct phases to confirm eligibility before subjects are progressed to Screen 2 to determine eligibility based on MRI-PDFF-assessed liver fat. As such, two separate informed consents will be obtained – 1st limited to pre-qualification visit only; and 2nd for the main study only in those deemed to meet eligibility criteria based on pre-qualification procedures. Between the pre-qualification visit and Visit 1 (Screen 1), many of the procedures and clinical laboratory tests assessed are identical – this is motivated by intent to confirm that the population progressed to Screen 2 is stable and consistently characterized in one of the two pre-specified 1st tier stratification sub-populations of diagnosed/presumed NASH versus NAFLD with minimal inflammation and fibrosis. As a measure of additional diligence and attempt to remove variability due to differences from laboratory-to-laboratory, the clinical laboratory tests assessed at pre-qualification and Visit 1 (Screen 1) are proposed to be analyzed by the sponsor-identified central laboratory. Furthermore, assessment of liver fat via ultrasound-based, but more quantitative FibroScan® and reported as controlled attenuation parameter (CAP™) is included at pre-qualification and Visit 1 (Screen 1), in the entire study population, with a threshold of ≥ 280 decibels per meter (dB/m), is designed to help identify the likely eligible population based on MRI-PDFF via a more logistically feasible, and overall more efficient approach. [redacted] The use of generic abdominal ultrasound to detect liver fat is not advocated given the limited utility of this assessment for liver fat less than 20% although there is high specificity for detecting ≥ 20-30% liver steatosis.[15]

For the assessment of liver fat (either via FibroScan® or MRI-PDFF), subjects will be required to fast (water permitted) for ≥ 4-hours given the ability of food to impact the results. As an additional measure to limit variability, attempts will be made to standardize the nominal time, for FibroScan® assessment and, especially related to the primary endpoint (MRI-PDFF). The clock time of day when these assessments are made should fall within a practical window (± 2-hours) relative to clock time at Visit 1 (Screen 1) for FibroScan® and Visit 2 (Screen 2) for MRI-PDFF. Serial assessments post randomization via MRI-PDFF and [redacted] are planned to permit a mixed-model-repeated-measure analysis thereby allowing for evaluation of drug effect longitudinally and, at each visit, including end of dosing with the investigational product (IP) – ie, Visit 11 (Week 16).

To date, the clinical experience with PF-05221304 is limited to a single Phase 1 trial in healthy subjects with repeated, in-patient/witnessed dosing up to 14-days. Transitioning to the current study, with dominantly outpatient dosing in a patient population, increased variability in exposure and drug-response is plausible. Hence, an added feature of the proposed study design is the frequent (every 2-week), outpatient visits during the first 8-weeks post randomization and monthly visits thereafter. Furthermore, at least 1 interim analysis (IA) is planned [refer to Section 9.5] to permit ongoing review of safety of the population enrolled, amongst the planned activities.

Plasma exposure of PF-05221304 following a high-fat/high-caloric breakfast was similar relative to administration of PF-05221304 following an overnight fast – suggesting that investigational product can be administered without regard to food. However, in this study, PF-05221304 administration will be requested to occur with the morning meal – given the importance of a morning meal to the standard-of-care diet counselling for the planned population.

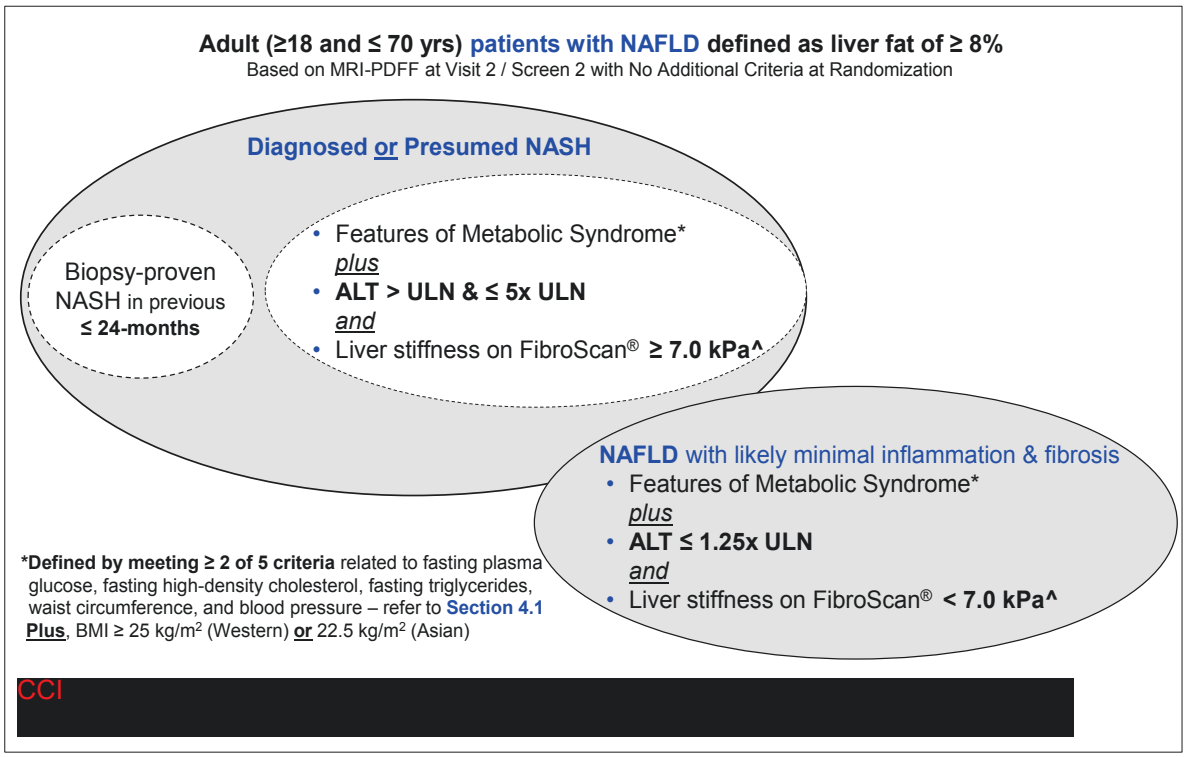
Total duration of dosing of investigational product in this study is proposed as up to 16 weeks[redacted]. This duration should permit an assessment of the primary pharmacology of PF-05221304 (ie, anti-steatosis), as well as potential improvement in liver function (as denoted by reduction in alanine transaminase (ALT) in the 1st tier stratification comprising of subjects with diagnosed or presumed NASH – a sub-population selected to have elevated ALT at Visit 1 (Screen 1).

A fixed, single-blind, 2-week baseline period (ie, Visit 4 to Visit 5) is included in this study with the explicit purpose of familiarizing the subjects with the dosing instructions for the blinded IP (refer to Table 3), and exclude subjects who are *not* compliant with the blinded placebo prior to randomization in an attempt to minimize medications errors post randomization.

1.3.2. Rationale for Population Enrolled

NAFLD is a condition marked by excessive (≥ 5%) fat accumulation in the liver with a subgroup progressing to having hepatocellular injury and inflammation (ie, NASH), and cirrhosis, and a smaller proportion of patients developing hepatic cellular carcinoma (HCC).[7] In this study, while the entire study population will have evidence of liver fat ≥ 8% at Screen 2 and features of metabolic syndrome, the population enrolled will be stratified as denoted in Figure 1 as a means to permit an opportunity to evaluate potential for changes in measures of hepatic inflammation and fibrosis [refer to Figure 3 for additional detail regarding stratification].

Figure 1. Overview of C1171002 Study Population



In the 1st tier stratification, the two sub-populations will be categorized as having diagnosis or presumed NASH versus NAFLD with likely minimal inflammation and fibrosis. Those with diagnosed/presumed NASH comprise of a subset with biopsy-proven diagnosis of NASH *or* a constellation of clinical and liver imaging characteristics which are highly suggestive of a NASH diagnosis (in the absence of a liver biopsy). The other sub-population of NAFLD with likely minimal inflammation and fibrosis will include those with alanine aminotransferase (ALT) ≤ 1.25x upper limit of reference range (ULN) for the sponsor-identified central laboratory, and no/minimal evidence of liver fibrosis (as measured by Vibration-Controlled Transient Elastography (VCTE™) and reported as liver stiffness measure (LSM, in kilopascals, kPa) using Fibroscan® and as such considered a distinct albeit related, population to those labeled as diagnosed/presumed NASH. This NAFLD with likely minimal inflammation and fibrosis population is included in this Phase 2a, dose-ranging study, as it provides an opportunity to more broadly assess general safety (including potential mechanistic effect on platelet count and serum triglycerides) and additional potential metabolic benefits of ACC inhibition (such as improvement in glycemic control), than could be readily achieved via limiting the population to only subjects with NASH. In addition, this study aims to permit those subjects within identified thresholds for ALT *in combination* with liver stiffness in the F0 to F3 grades of fibrosis. In doing so, population stratification acknowledges that elevated ALT alone is an imperfect marker of NASH *and* the technology used (FibroScan®) to assess liver stiffness lacks the specificity to categorize F0-F2 grades of fibrosis with a great deal of precision. Even with these caveats, the combination of these

screening parameters *may* aide in characterizing the population in the absence of a liver biopsy – the current gold standard[7] despite its inherent limitations such as its invasive assessment with procedural risks, potential for sampling errors, and inconsistency in interpretation of the histopathology.

The liver stiffness cut-off for the sub-population categorized as presumed NASH was motivated by the intent to include those with F2-F3 grades of fibrosis though noted to permit likely F1 (but not F0) grades of fibrosis; similarly, the liver stiffness cut-off for the NAFLD with likely minimal inflammation and fibrosis reflects the intent to limit population to dominantly F1-F0 grades of fibrosis. Considering that many patients diagnosed with NASH on liver biopsy have ALT ≤ ULN, and many with elevated ALT do not have NASH on liver biopsy,[16] it is acknowledged that the approach taken to stratify the study population is likely unable to definitively rule-out patients with NASH from the sub-population categorized as NAFLD with likely minimal inflammation and fibrosis.

Beyond the categorization of the enrolled population into diagnosed/presumed NASH versus NAFLD with likely minimal inflammation and fibrosis, this study will apply a 2nd tier stratification to identify the population with and without a diagnosis of type 2 diabetes mellitus (T2DM) based on glycated hemoglobin (HbA1C) result > 6.5% and/or current use of agents for glycemic control (Section 5.8.1). This is to permit an assessment of whether PF-05221304 demonstrates insulin sensitization properties. While PF-05221304 is not known to carry the potential risk of hypoglycemia, many of the agents used for the management of glycemic control in patients with T2DM carry this risk and hence a standardized approach to capture of such events will be undertaken – refer to Section 7.9.3.1.1 and Appendix 4.

Together, the study is designed to understand the effect of PF-05221304 on liver fat, ALT and other NASH-related biomarkers, glycemic parameters, fasting lipids, and effect on other parameters [ie, Apolipoprotein A1, B (total), C3, E, CCI [redacted] as well as assess effect of PF-05221304 on metabolic parameters. NASH-related markers include: cytokeratin18-M30 (CK18-M30) and CK18-M65, markers of apoptotic activity; CCI [redacted] ProC3, the N-terminal pro-peptide of type III procollagen which is a potential predictor of fast progressors and responders to therapy at baseline; and ProC6, the C-terminal fragment of α3 chain of procollagen type VI (also known as Endotrophin) which is a potential predictor of responders to insulin sensitizers at baseline. CCI [redacted]

In this study, based on a combination of thorough medical history (including assessment of ethanol intake and administration of the Alcohol Use Disorders Identification Test (AUDIT) 10-item questionnaire, and clinical laboratory tests (including gamma glutamyl transferase [GGT] and percent carbohydrate deficient transferrin relative to total transferrin [%CDT]),

medically-qualified site-staff will be required to rule-out alcohol-based as well as other causes of hepatic steatosis, and ensure that the population enrolled is confirmed, as best as possible, to have non-alcoholic fatty liver disease.[18] These assessments (AUDIT and %CDT) are re-assessed prior to randomization and at end of dosing to confirm that while in the study alcohol intake remained in moderation. Subjects with previously confirmed genetic polymorphisms, for example, patatin like phospholipase domain containing protein 3 (PNPLA3) carriers, are eligible given these polymorphisms are known to modify/increase risk for steatohepatitis but do not by themselves cause steatohepatitis.

An explicit intent in this study is to exclude those with severe hepatic impairment (eg, ascites, hepatic encephalopathy, cirrhosis, and HCC) and severe renal impairment (ie, estimated glomerular filtration rate [eGFR] ≤ 30 mL/min) via specific eligibility criteria (refer to Section 4.1 and Section 4.2). This is in line with: (1) the clinical data to date obtained in a trial enrolling healthy subjects with the current trial being the first in the patient population; (2) the mechanism of action (ie, primarily anti-steatotic) is unlikely to display anti-fibrotic action in a trial with up to 16 weeks of dosing; and (3) at present, lack of Phase 1 studies to describe the plasma PK of PF-05221304 in subjects with hepatic and/or renal impairment. CCI [redacted] Thus, the inclusion/exclusion criteria have been defined to help limit pharmacokinetic variability while also capturing a population that is characteristic of the intended patient population. CCI [redacted]

CCI [redacted] In order to minimize PF-05221304 pharmacokinetic variability due to drug-drug interactions (DDIs), medications that have been shown to perpetrate OATP mediated clinically significant DDIs are excluded (notably gemfibrozil, cyclosporine). Furthermore, concomitant use of potent inhibitors (eg, ketoconazole or itraconazole) or inducers (eg, rifampin) of CYP3A4/5 is to be avoided. CCI [redacted]

[redacted]

CCI [redacted] Amongst the agents for glycemic control, thiazolidinediones (TZDs) and glucagon-like peptide-1 receptor (GLP-1r) agonists are not permitted given their potent anti-steatotic effect[18,19] and hence the potential to confound the primary endpoint of this study. Refer to Section 5.8 for complete details regarding permitted and excluded concomitant medications.

Given the observed effects of PF-05221304 on platelet count, and serum triglycerides (both fasting and post-prandial) in the Phase 1 study (C1171001), in the current study, eligibility criteria for platelet count and fasting triglycerides is included (refer to Section 4) along with a monitoring strategy for each aspect – refer to Section 7.9.1 and Appendix 2 (platelet count), as well as Section 7.9.2 and Appendix 3 (fasting serum triglycerides).

For the postulated improvement in insulin sensitivity with PF-05221304 administration in this study, diligence will be applied via monitoring of fasting plasma glucose (FPG), fasting plasma insulin (FPI), HOMA-IR, HbA1C, and in patients with T2DM the standardized capture of any hypoglycemic events (HAEs) along with hyperglycemia (refer to Section 7.9.3). To ease subjects' burden, at Visit 3 (Run-in), the sponsor will provide all eligible subjects with T2DM glucometer and associated ancillary supplies to permit at-home self-monitoring – at least once daily.

Both females of childbearing potential as well as those who are of non-childbearing potential will be enrolled CCI [redacted] refer to Schedule of Activities (Table 1 and Table 2) as well as Section 4.4.6 and Section 7.3.1]. Measures to prevent pregnancy in female partners of male subjects are suggested to include a simple barrier method CCI [redacted]

1.3.3. Rationale for Doses Selected

In addition, to placebo, the PF-05221304 doses selected for this study (2, 10, 25 and 50 mg once daily) are based on the observed safety, tolerability, pharmacokinetic, and pharmacodynamic data from Study C1171001 in healthy adult subjects. In Study C1171001, PF-05221304 was found to be well tolerated with an acceptable safety profile with single doses up to 240 mg and repeated doses up to 200 mg/day) administered and the MTD *not* identified. At the three highest doses (≥ 60 mg/day), a gradual decrease in platelet count, was observed with repeated doses over the 14 day dosing period, though none of the values met the criteria for thrombocytopenia. In addition, a gradual increase in fasting and post-prandial triglycerides was observed with repeated doses ≥ 40 mg/day with effect plateauing over the 14-day dosing period.

Following repeated dosing in healthy adult subjects (Study C1171001), change from baseline in fructose-stimulated hepatic DNL was assessed via incorporation of deuterated water tracer into triglyceride-palmitate over a 10-hour period where fructose doses were given every 30-minutes to stimulate DNL. Inhibition in DNL, a marker of target engagement, was observed in a dose and concentration-dependent manner with ≥ 90% DNL inhibition at PF-05221304 total daily doses ≥ 40 mg/day. CCI [redacted] Given the projected degree of target engagement (ie, DNL inhibition) with the planned dosing frequency (ie, once-daily), PF-05221304 dosing frequency is proposed as once-daily (rather than Q12H evaluated on 5 of the 7 dose-levels in the Phase 1 Study C1171001 in healthy subjects). The selected dose range spans a range of expected DNL inhibition, allowing for assessment of the relationship between DNL and the primary endpoint (liver fat), key secondary endpoint (ALT), as well as safety and other parameters.

CCI [redacted]

The overall dose selection of PF-05221304 was also influenced by the aim to identify not only a minimally-effective dose but also characterize the dose-response using the tablet strengths available (1, 5, 25, and 50 mg – with the 2 lower strengths looking identical but different than the 2 higher strength tablets) as efficiently as possible. As proposed, the dose-range planned will necessitate each subject to self-administer 3 tablets per day in order to maintain the double-blind design of this study.

CCI [redacted]

[redacted]

CCI [redacted]

1.4. Summary of Benefit Risk Assessment

Study C1171002 is the first time that PF-05221304 is being administered to patients with NAFLD. For the subjects participating in this study, close monitoring of their medical condition and safety, will occur as outlined in this protocol. Those randomized to PF-05221304 may potentially derive benefit from reduction of DNL and its potential associated effects including reduction in hepatic steatosis, and potential reduction in liver inflammation. Those randomized to placebo are not expected to obtain any specific benefit, beyond close monitoring of their medical condition and safety. As of the issuance of this protocol, identified adverse drug reactions that may be encountered with the administration of PF-05221304 include elevation in blood triglycerides and reduction of platelet count.

Any adverse clinical impact of these findings are expected to depend on the severity of the potential reaction, and are minimized through – (1) the proposed frequent visits to site to permit close oversight of subjects' safety via monitoring of a range of clinical assessments including laboratory tests, 12-lead ECGs, vitals, assessment of AEs; (2) inclusion of provisions for temporary discontinuation of dosing and permanent withdrawal from the study as outlined in Section 3.2; (3) institution of an Independent Review Committee (IRC) to undertake unblinded review of the safety data, while the study is on-going, and make recommendation(s) as to whether 1 or more doses should be paused/discontinued (refer to Section 3.3 and Section 9.5), amongst the planned activities.

2. STUDY OBJECTIVES AND ENDPOINTS

Primary Objective:	**Primary Endpoint:**
To evaluate the dose-response for the effect on ***liver fat*** with a range of PF-05221304 doses administered daily in adults ***with nonalcoholic fatty liver disease (entire study population)***	Percent change from baseline in liver fat, as assessed using MRI-PDFF, at Week 16
Secondary Objective(s):	**Secondary Endpoint(s):**
To evaluate the dose-response for the effect on ***ALT*** with a range of PF-05221304 doses administered daily in the 1st tier stratification comprising of adults ***with diagnosed/presumed nonalcoholic steatohepatitis, only***	Percent change from baseline in ALT, at Week 16
To evaluate the ***safety and tolerability*** of a range of PF-05221304 doses administered daily in adults ***with nonalcoholic fatty liver disease (entire study population)***	Assessment of TEAEs, safety-related clinical laboratory tests, vital signs, and 12-lead ECGs

Tertiary Objective(s):	Tertiary Endpoint(s):
To evaluate the dose-response for the effect on liver fat, ***over time***, with a range of PF-05221304 doses administered daily in adults ***with nonalcoholic fatty liver disease (entire study population)***	Percent change from baseline in liver fat, as assessed using MRI-PDFF, over time up to Week 16
CCI	
To evaluate the dose-response for the effect on key liver function tests and NASH-related biomarkers ***over time***, with a range of PF-05221304 doses administered daily in the 1st tier stratification comprising of adults ***with diagnosed/presumed nonalcoholic steatohepatitis, only***	Percent change from baseline, over time up to Week 16, for: • ALT, AST, Alkaline Phosphatase, GGT • CK18-M30 and CK18-M65 CCI • Pro-C3 & Pro-C6
CCI	
To evaluate the dose-response for the effect on other potentially mechanism-related parameters and metabolic parameters, ***over time***, with a range of PF-05221304 doses administered daily in adults with ***nonalcoholic fatty liver disease (entire study population)***	Percent change from baseline, over time up to Week 16, for: • Potentially mechanism-related parameters – – Serum apolipoprotein A1, B (total), C3, E CCI • Metabolic parameters – – HbA1C – Fasting lipid panel (fasting total cholesterol, direct LDL-C, HDL-C, triglycerides, VLDL) CCI
To evaluate the dose-response for the effect on glycemic parameters, ***over time***, with a range of PF-05221304 doses administered daily in the 2nd tier stratification comprising of ***adults with T2DM, only***	Change from baseline, over time up to Week 16, for: • HbA1C • FPG • FPI • HOMA-IR
CCI	

Tertiary Objective(s):	Tertiary Endpoint(s):
CCI [redacted]	

For ***all*** endpoints, baseline defined as result closest prior to dosing at Visit 5 (Day 1); for list of terms corresponding to the abbreviations used herein, refer to Appendix 1.

3. STUDY DESIGN

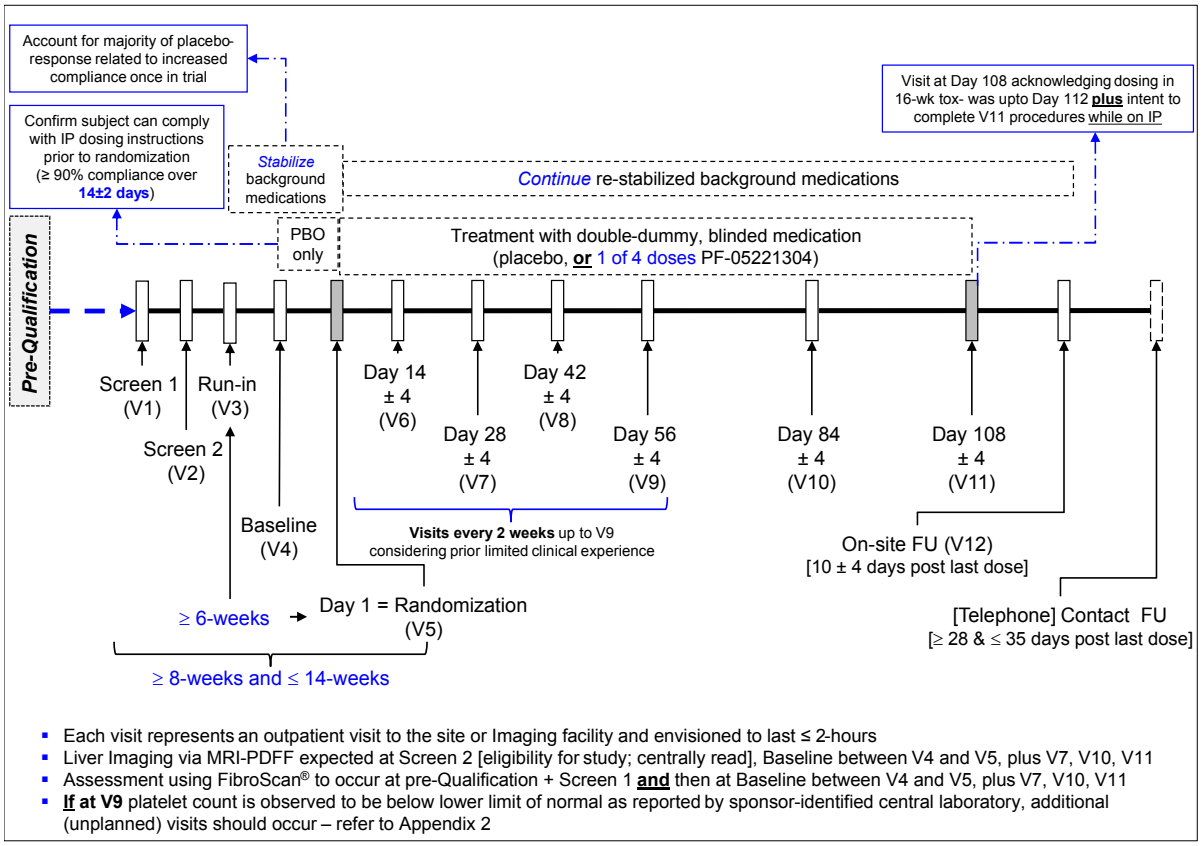
3.1. Study Overview

This is a randomized, double-blind, placebo-controlled, 5-arm (placebo, plus 4 active doses of PF-05221304), parallel-group study.

Determination of eligibility for this study will occur via a sequential, 3-step process – starting with pre-qualification. Subjects identified to be eligible based on pre-qualification will then undergo Visit 1 (Screen 1) with only those confirmed to continue to be eligible progressing to Visit 2 (Screen 2). For a given subject, this 3-step eligibility assessment (from pre-qualification to Screen 2, inclusive) may take up to 2-months.

Once confirmed to be eligible based on Screen 2 procedure(s), subjects will progress to a run-in phase when concomitant treatment(s) will be stabilized, and baseline-related visits will occur. At Visit 5 (Day 1), subjects will be randomized to receive 1 of 5 blinded regimens for a duration of up to 16 weeks (ie, 108±4 days). Excluding the pre-qualification visit, this study includes a total of 12 *scheduled* outpatient visits ***in the morning***, CCI [redacted] ***and*** a potential for up to at least 3 visits for repeat assessments (for procedures/tests at Visit 1, Visit 2, or 2nd Follow-up visit). The total participation, from Visit 1 (Screen 1) to 2nd Follow-up visit, will be 27-weeks (minimum) to 35-weeks (maximum) – refer to Figure 2.

Figure 2. C1171002 Study Design



At least 360 subjects (72 per arm) will be randomized at approximately 120 sites to ensure a minimum of 300 subjects (60 per arm) complete the study. ***In each arm***, ≥ 50% of subjects randomized will be deemed to be eligible for 1st tier stratification of diagnosed/presumed NASH; with the remaining subjects per arm eligible for 1st tier stratification of NAFLD with likely minimal inflammation and fibrosis – refer to Section 4.3.

Subjects who are noted to have non-evaluable baseline MRI-PDFF, as determined by the sponsor-identified central imaging vendor, may be replaced, at sponsor discretion – refer to Section 7.1.4; otherwise, there are no plans to replace subjects who are prematurely withdrawn. To ensure that the required minimum number of subjects complete the study, the plan is to overenroll by approximate 20% to account for premature early withdrawal.

3.2. Stopping Rules for Dosing in Individual Subjects

At investigator discretion, ***for subjects' safety,*** dosing with double-blinded investigational product (ie, post randomization), may be stopped in an individual subject – either temporarily **or** permanently. In such cases, any open TEAEs must be followed to resolution or until such time that the event is viewed to have stabilized – refer to Section 8 for details – specifically, Section 8.1 which outlines the requirements for reporting adverse events, Section 8.2.3 detailing definition of serious adverse events, and Section 8.4.2 summarizing management of potential cases of drug-induced liver injury which are categorized as potentially important medical events.

An example where **temporary discontinuation of dosing** with investigational product, may be considered appropriate includes hospitalization with subsequent discharge within ≤ 7-days in stable medical condition, as determined by medically-qualified investigator. Of note, in this study, re-challenge/re-initiation of dosing with double-blinded investigational product should **not** be attempted in subjects who meet threshold for withdrawal based on platelet count (Appendix 2) *or* fasting serum triglycerides (Appendix 3).

Examples where **permanent discontinuation of dosing** and withdrawal of the subject from the study is deemed appropriate, as determined by investigators' medical judgment, include:

- Hospitalization due to serious adverse event (SAE) resulting in clinical decompensation of the subject *and* necessitating continued inpatient stay for more than 7-days;
- Subjects observed to have a decline over time ultimately reaching the threshold of < 75,000/mm^3 in platelet count – refer to Section 7.9.1 and Appendix 2 for additional guidance;
- Individual subjects with consistently increasing fasting serum triglycerides over time ultimately reaching the threshold of ≥ 800 mg/dL (9 mmol/L) – refer to Section 7.9.2 and Appendix 3 for additional guidance;
- Of note, in both the above cases of pre-identified laboratory thresholds, 1 additional unplanned assessment may be warranted ***before*** permanent withdrawal if the threshold is observed to be met as an isolated occurrence relative to all prior results ***and*** subject is asymptomatic.

3.3. Considerations for Pausing or Stopping Active Dose(s) Based on Observed Safety

The decision to pause *or* stop dosing at a study-level, for 1 or more active dose(s) of PF-05221304 may be considered based on recommendation from the Internal Review Committee (IRC) according to their review of unblinded, study-level emerging, *observed* safety data – refer to Section 9.5.

In addition, during study conduct, after approximately ***every*** 20% of the total planned randomizations [for example: 72 subjects (20%), 145 subjects (40%), 215 subjects (60%), and 290 subjects (81%) of planned 360 subjects], based on blinded safety review by the selected members of the Sponsor's study team, unblinded review of the safety data by the IRC will be triggered and a determination of the safety to continue the study documented if post randomization ***either*** of the below conditions are met –

- More than 20% of subjects, develop a moderate or severe AE in the same system organ class (SOC);
- More than 15 % of subjects meet the individual permanent discontinuation of dosing rules (Section 3.2).

4. SUBJECT ELIGIBILITY CRITERIA

This study can fulfill its objectives only if appropriate subjects are enrolled. The following eligibility criteria are designed to select subjects for whom participation in the study is considered appropriate. All relevant medical and nonmedical conditions should be taken into consideration when deciding whether a particular subject is suitable for this protocol.

Subject eligibility should be reviewed and documented by an appropriate member of the investigator's study team before subjects are included in the study.

4.1. Inclusion Criteria

Subjects must meet **all** of the following inclusion criteria to be eligible for enrollment into the study:

1. Male or female subjects between the ages of 18 (*or* the minimum country-specific age of consent if > 18) and 70 years, inclusive, at Visit 1 (Screen 1):

   - **Male and female subjects of childbearing potential** must agree to use highly effective method(s) of contraception [refer to Section 4.4.6]. A subject is of childbearing potential if, in the opinion of the investigator, he/she is biologically capable of having children **and** is sexually active.

     - Female subjects of childbearing potential must **not** be pregnant, breastfeeding, or planning to become pregnant for the duration of their participation in this study and **within 28-days** following last dose of blinded investigational product.

   - **Female subjects of non-childbearing potential must meet at least 1 of the following criteria:**

     a. Achieved postmenopausal status, defined as follows: cessation of regular menses for at least 12 consecutive months with no alternative pathological or physiological cause; **and** have a serum follicle-stimulating hormone (FSH) level confirming the postmenopausal state, with a single repeat assessment, via the sponsor-identified central laboratory, permitted to assess postmenopausal state, if needed;

     b. Have undergone a documented hysterectomy and/or bilateral oophorectomy;

     c. Have medically confirmed ovarian failure;

     All other female subjects (including female subjects with tubal ligations) are considered to be of childbearing potential.

   - At Pre-Qualification *and* Visit 1 (Screen 1), **in all female subjects,** serum beta human chorionic gonadotropin (β-hCG) level, with a single repeat assessment, via the sponsor-identified central laboratory, permitted to assess non-pregnant state, if needed.

2. At Pre-Qualification, pass the hula-hoop test [refer to Section 7.1.3] with a single repeat assessment, permitted, if needed, to assess eligibility.

3. At Pre-Qualification, have an arm circumference compatible with the largest blood pressure cuff size available at individual sites [refer to Section 7.2.4] with a single repeat assessment, permitted, if needed, to assess eligibility.

4. At Pre-Qualification *and* Visit 1 (Screen 1), total body weight of > 50 kg (110 lbs) **with** BMI ≥ 25 kg/m^2 (for sites in Africa, Europe, North America) or BMI ≥ 22.5 kg/m^2 (for sites in Asia), with a single repeat assessment of body weight and/or BMI permitted to assess eligibility, if needed, at each of these 2 visits.

5. At Pre-Qualification *and* Visit 1 (Screen 1), meet the following criteria for **liver fat**, based on assessment via FibroScan®, with a single repeat permitted to assess eligibility, if needed, at each of these 2 visits:

   - CAP™ ≥ 280 dB/m;

   - *NOTE*: if Pre-Qualification and Visit 1 values are discordant stratifying a given subject, 1 more assessment via FibroScan® ***on a separate day*** is permitted, with 2 of the 4 assessments (at least 1 between pre-qualification and repeat pre-qualification, ***plus*** at least 1 between Screen 1 and repeat Screen 1) required to meet above threshold ***before progressing*** to Screen 2.

6. At Pre-Qualification *and* Visit 1 (Screen 1), meet the following criteria, as assessed by sponsor-identified central laboratory; with a single repeat on any of these parameters permitted to assess eligibility, if needed, at each of these 2 visits:

   - International Normalized Ratio (INR) ≤ 1.3;

   - Albumin ≥ lower limit of normal (LLN);

   - Platelet count ≥ 0.95x LLN;

   - Fasting triglycerides ≤ 400 mg/dL (4.5 mmol/L);

   - HbA1C ≤ 9.5%;

   - Fasting plasma glucose ≤ 270 mg/dL (15.0 mmol/L);

   - Total bilirubin < 1.5x upper limit of normal (ULN) **and** direct bilirubin ≤ ULN.

     - NOTE: Subjects with a history of Gilbert syndrome would be eligible for this study provided direct bilirubin level is ≤ ULN **plus** ALT met criteria for applicable 1st tier stratification **plus** alkaline phosphatase, hemoglobin, and reticulocyte count are all ≤ ULN.

7. Documented liver biopsy ≤ 24-months prior to Visit 1 (Screen 1) with histological evidence of NASH in order to be classified in the sub-population with biopsy-proven NASH, ***only***.

8. In those *without* biopsy-proven NASH, ***only***, at Pre-Qualification *and* Visit 1 (Screen 1), meet the following criteria **for liver stiffness**, based on assessment via FibroScan®, with a single repeat permitted to assess eligibility, if needed, at each of these 2 visits:

   - LSM ≥ 7.0 kPa – classified as presumed NASH;
   - LSM < 7.0 kPa – classified as NAFLD with likely minimal inflammation and fibrosis;
   - *NOTE*: if Pre-Qualification and Visit 1 values are discordant in stratifying a given subject, 1 more assessment via FibroScan® on ***a separate day*** is permitted, with 2 of the 4 assessments (at least 1 between pre-qualification and repeat pre-qualification, ***plus*** at least 1 between Screen 1 and repeat Screen 1) required to meet above threshold ***before progressing*** to Screen 2.

9. In those *without* biopsy-proven NASH, ***only***, at Pre-Qualification *and* Visit 1 (Screen 1), meet the following criteria, as assessed by the sponsor-identified central laboratory, with a single repeat permitted to assess eligibility, if needed, at each of these 2 visits:

   - ALT > ULN and ≤ 5x ULN – classified as presumed NASH;
   - ALT ≤ 1.25x ULN – classified as NAFLD with likely minimal inflammation and fibrosis;
   - *NOTE*: if Pre-Qualification and Visit 1 values are discordant in stratifying a given subject, 1 more blood collection on ***a separate day*** is permitted, with 2 of the 4 assessments (at least 1 between pre-qualification and repeat pre-qualification, ***plus*** at least 1 between Screen 1 and repeat Screen 1) required to meet above threshold ***before progressing*** to Screen 2.

10. In those *without* biopsy-proven NASH, ***only***, at Pre-Qualification *and* Visit 1 (Screen 1), meet **≥ 2** of the following 5 criteria for metabolic syndrome [*for laboratory parameters*, results must be as assessed by the sponsor-identified central laboratory, with a single repeat permitted to assess eligibility, if needed, at each of these 2 visits]:

    - FPG ≥ 100 mg/dL (5.6 mmol/L), or on pharmacological agents *with explicit purpose* of improving glycemic control (refer to Section 5.8.1 for acceptable versus prohibited medications);
    - Fasting serum HDL-C < 40 mg/dL (1 mmol/L) for males and < 50 mg/dL (1.3 mmol/L) for females, or on pharmacological agents *with explicit purpose* to increase HDL-C (refer to Section 5.8.2 for acceptable versus prohibited medications);

- Fasting serum triglyceride (TG) ≥ 150 mg/dL (1.7 mmol/L), or on pharmacological agents *with explicit purpose* to decrease TG (refer to Section 5.8.2 for acceptable versus prohibited medications);

- Seated blood pressure (BP) ≥ 130 / 85 mm Hg, or on pharmacological agents *with explicit purpose* for BP control (refer to Section 5.8.4 for acceptable versus prohibited medications);

- Waist circumference ≥ 40 inches (102 cm) for males and ≥ 35 inches (89 cm) for females.

11. At Visit 2 (Screen 2), liver fat of ≥ 8%, using MRI-PDFF acquisition protocol at the sponsor-qualified Imaging facility in close proximity to the site, confirmed via a single repeat, if deemed necessary by the sponsor-identified central imaging vendor of MRI-PDFF images – refer to Section 7.1.4.

12. At Pre-Qualification, evidence of a personally signed and dated informed consent document indicating that the subject has been informed of all pertinent aspects of the pre-qualification procedures of this protocol.

    - *NOTE*: for subjects who qualify based on Pre-Qualification procedures, at Visit 1 (Screen 1), evidence of a ***separate*** personally signed and dated informed consent document indicating that the subject has been informed of all pertinent aspects of the main study, is required;

13. Subjects willing and able to comply with scheduled visits, treatment plan, laboratory tests, and other study-related procedures as outlined herein.

4.2. Exclusion Criteria

Subjects with ***any*** of the following characteristics/conditions will *not* be included in the study:

1. Subjects with known prior participation (ie, randomized and received at least 1 dose of investigational product) in a trial involving PF-05221304.

2. Participation in other studies involving investigational drug(s) within **30-days** prior to Visit 1 (Screen 1) and during study participation (ie, up to 2nd Follow-up).

3. At Pre-Qualification *and* Visit 1 (Screen 1), history of regular alcohol consumption exceeding 14 drinks/week for females **or** 21 drinks/week for males [1 drink = 5 ounces (150 mL) of wine or 12 ounces (360 mL) of beer or 1.5 ounces (45 mL) of hard liquor)] within the previous 6 months.

4. At Pre-Qualification *and* Visit 1 (Screen 1), a total score of ≥ 8 on the AUDIT questionnaire, indicating harmful or hazardous ethanol consumption.

5. At Pre-Qualification *and* Visit 1 (Screen 1), a positive urine drug test for illicit drugs.

6. At Pre-Qualification *and* Visit 1 (Screen 1), a persistent severe, uncontrolled hypertension; for example: ***seated*** systolic blood pressure (SBP) ≥180 mm Hg and/or diastolic blood pressure (DBP) ≥105 mm Hg after ≥5-minute of rest, with a single repeat permitted to assess eligibility, if needed, at each of these 2 visits:

    - For subjects with SBP ≥ 160 mm Hg **or** DBP ≥ 100 mm Hg, the Run-in Period must be used to refine the doses of the agents used for management of blood pressure (refer to Section 5.8.3) with the aim to have SBP ≤ 159 mm Hg **and** DBP ≤ 99 mm Hg at the time of randomization [refer to Section 4.3];

7. At Pre-Qualification *and* Visit 1 (Screen 1), a ***supine*** 12-lead ECG demonstrating QTc interval >450 msec **or** a QRS interval >120 msec:

    - If QTc interval exceeds 450 msec, or QRS interval exceeds 120 msec, the ECG should be repeated two more times and the *average of the three* QTc intervals or QRS intervals should be used to determine the subject's eligibility.

8. At Pre-Qualification *and* Visit 1 (Screen 1), clinical evidence of hepatic decompensation, including, but not limited to esophageal varices, ascites or hepatic encephalopathy.

9. At Pre-Qualification *and* Visit 1 (Screen 1), evidence of other forms of chronic liver disease including but not limited to the entities listed below; evidence may include laboratory tests, as assessed by the sponsor-identified central laboratory, with a single repeat permitted to assess eligibility, if needed, at each of these 2 visits:

    - Hepatitis B virus (HBV), defined by presence of hepatitis B surface antigen (HBsAg);
    - Hepatitis C virus (HCV), defined by presence of hepatitis C antibody (HCVAb);
        - *irrespective of* HCV RNA result (when reflexed based on a positive result for HCVAb);
    - Human Immunodeficiency Virus (HIV) infection, defined as presence of HIV antibody;
    - Known diagnosis of primary biliary cirrhosis, primary sclerosing cholangitis, autoimmune hepatitis, or overlap syndrome;
    - Alcoholic liver disease, as evaluated via history along with laboratory tests including serum % carbohydrate deficient transferrin result ≥ 1.5x ULN deemed to be highly suggestive to alcohol abuse;
    - History of genetic reason for fatty liver;
    - Wilson's disease, defined as ceruloplasmin level < LLN;

- Known diagnosis of hemochromatosis;
- Alpha-1-antitrypsin (A1AT) deficiency, defined as alpha-1-antitrypsin level < LLN;
- Prior known drug-induced liver injury;
- Known or suspected Hepatocellular Carcinoma or other liver cancer;
- History of liver transplant, current placement on a liver transplant list, or current model of end-stage liver disease (MELD) score >12;
- Histological presence of cirrhosis on prior biopsy.

10. At Pre-Qualification *and* Visit 1 (Screen 1), subjects with an estimated glomerular filteration rate (eGFR) of < 30 mL/min using the Modification of Diet in Renal Disease (MDRD) equation, and serum creatinine (SCr), as assessed by the sponsor-identified central laboratory, with a single repeat permitted to assess eligibility, if needed, at each of these 2 visits.

11. At Pre-Qualification *and* Visit 1 (Screen 1), subjects with **any** of the following medical conditions:

    - Any condition possibly affecting drug absorption (eg, prior bariatric surgery, gastrectomy, ileal resection);
        - NOTE: subjects who have undergone cholecystectomy and/or appendectomy are eligible for this study so long as the surgery occurred more than 6 months prior to Screen 1;
    - Diagnosis of type 1 diabetes mellitus;
    - In those with diagnosis of type 2 diabetes mellitus, history of:
        - Episodes of hypoglycemia of 'severe' intensity *either* ≥ 1 in previous 1 month ***or*** ≥ 3 in previous 3 months;
        - Treatment with excluded agents for glycemic control – refer to Section 5.8.1;
    - Subjects with prior history of pancreatitis;
    - Recent (ie, within the previous 6 months) history of congestive heart failure (New York Heart Association, NYHA, class III or IV) or unstable angina;
    - A history of myocardial infarction, stroke, **or** transient ischemic attack, in the previous 6 months;
    - ≥ 5% weight loss within the previous 3 months;

- Any malignancy not considered cured (except basal cell carcinoma and squamous cell carcinoma of the skin); a subject is considered cured if there has been no evidence of cancer recurrence in the previous 5 years;
- Active placement of medical devices in/on thoracic cavity – such as pacemakers, defibrillators [as these interfere with use of FibroScan®].

12. Subjects meeting criteria for contraindication for MRI – including the following:
    - History of severe claustrophobia impacting ability to perform MRI during the study *even despite* mild sedation/treatment with an anxiolytic;
    - Subjects with metal implants, devices, paramagnetic objects contained within the body and excessive **or** metal-containing tattoos;
    - Subjects unable to lie still within the environment of the MRI scanner or maintain a breath hold for the required period to acquire images *even despite* mild sedation/treatment with an anxiolytic;
13. At Pre-Qualification *and* Visit 1 (Screen 1), subjects on any prohibited concomitant medication(s) ***or*** those unwilling/unable to switch to permitted concomitant medication(s) – refer to Section 5.8.
14. Subjects with any anatomical or pathological abnormality that would either preclude or tend to confound the analysis of study data, including any clinically significant abnormal findings on MRI obtained at Visit 2 (Screen 2), by the sponsor-identified central imaging vendor of MRI-PDFF images [refer to Section 7.1.4].
15. Male subjects with partners who are currently pregnant, pregnant female subjects, breastfeeding female subjects, as well as fertile male subjects and female subjects of childbearing potential who are unwilling or unable to use highly effective methods of contraception as outlined in Section 4.4.6 for the duration of the study and for **at least 28 days** after the last dose of blinded investigational product.
16. Blood donation (excluding plasma donations) of approximately 1 pint (500 mL) or more within 56 days prior to Visit 1 (Screen 1) **or** during the study until Visit 12 (on-site Follow-up).
17. Investigator site staff members directly involved in the conduct of the study and their family members, site staff members otherwise supervised by the investigator, or subjects who are Pfizer employees, including their family members, directly involved in the conduct of the study.
18. At Pre-Qualification *and* Visit 1 (Screen 1), other acute or chronic medical or psychiatric condition including recent (within the past year) or active suicidal ideation or behavior or laboratory abnormality that may increase the risk associated with study participation or investigational product administration or may interfere with the interpretation of study results and, in the judgment of the investigator, would make the subject inappropriate for entry into this study.

4.3. Randomization Criteria

- *At Pre-Qualification*, following completion of informed consent, subjects will be assigned a single 8-digit study-specific subject identification (SSID) number by the IRT (interactive response technology) system which will be retained throughout the duration of participation in the study;

- ***At Visit 5 (Day 1)***, IRT will be used to assign each eligible subject a randomization number, with this number recorded on the electronic Case Report Form (e-CRF).

  - A computer-generated randomization code using the method of random permuted blocks will be utilized to randomize subjects 1:1:1:1:1 (across placebo or 1 of 4 active doses of PF-05221304) at Visit 5 (Day 1) prior to the first dose of the double-blinded investigational product (PF-05221304 or matching placebo) provided subjects' satisfy **all** the following criteria:

    - Eligibility criteria outlined in Section 4.1 and Section 4.2;

    - A negative urine drug test for illicit drugs for sample collected at Visit 4 (Day -14) and results as reported by sponsor-identified central laboratory;

    - Fasting (≥ 8 hours overnight, with water permitted) serum triglyceride result of ≤ 400 mg/dL (4.5 mmol/L), for sample collected at Visit 4 (Day -14), as reported by sponsor-identified central laboratory;

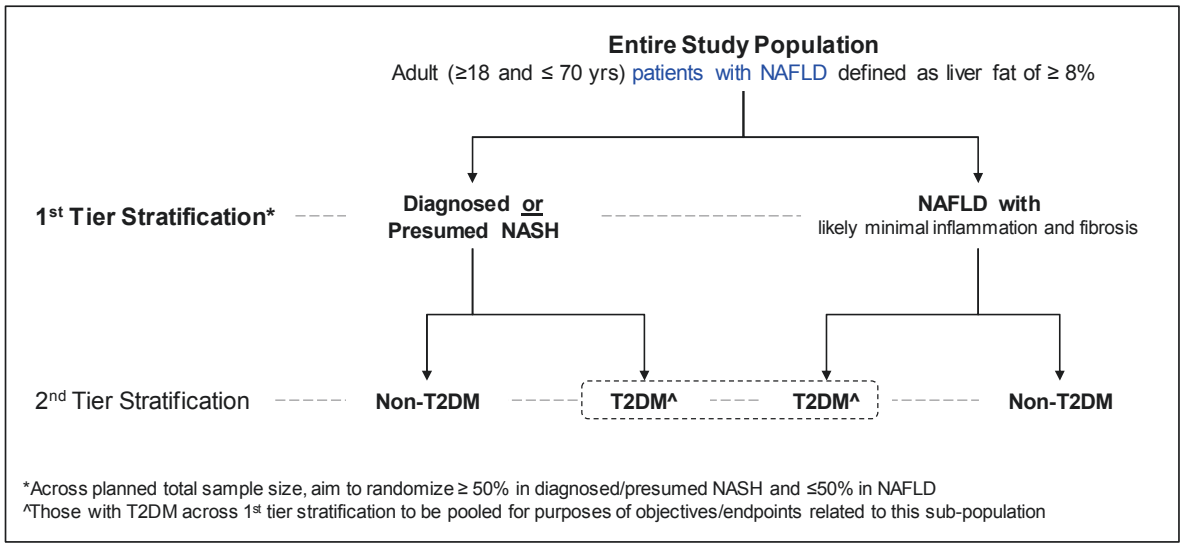
    - In females, *serum* pregnancy test at Visit 4 (Day -14) as reported by sponsor-identified central laboratory **and** *urine* pregnancy test *on-site* (in females of childbearing potential) at Visit 5 (Day 1) as reported on-site using supplies offered by sponsor-identified central laboratory, are negative for pregnancy – refer to Section 7.3.1;

    - Noted to be ≥ 90% compliant (based on pill count) with the single-blind placebo administered from Visit 4 (Day -14) to *1 day before* Visit 5 (Day 1), inclusive, for a total duration of 14 ± 2 days;

  - Total score of < 8 on the AUDIT questionnaire, as assessed at Visit 5 (Day 1).

- Study will employ 2-tier stratification as outlined in Figure 3.

Figure 3. Two-Tiered Stratification in Study C1171002



Via randomization, an attempt will be made to balance the number of subjects assigned to receive each active dose of PF-05221304 and placebo within the 1st tier stratification. However, an imbalance in the number of subjects assigned to receive each active dose of PF-05221304 versus placebo between the two-tiered stratification is allowed.

4.4. Lifestyle Requirements

After confirmation of eligibility at Visit 2 (Screen 2), ***and*** starting at Visit 3 (Run-in), subjects will be instructed to maintain the guidelines described below for the duration of participation in the study. These guidelines must be reiterated at Visit 5 (Day 1).

4.4.1. Dietary Restrictions

- Subjects must abstain from all food and drink (except water) for **≥ 8-hours** prior to any blood sample collections for clinical laboratory tests, fasting glucose monitoring, CCI [redacted]
- Water may be consumed as desired (ad libitum);
- Blinded investigational product must be administered every day with the morning meal;
- On scheduled outpatient visits to the site, ***in the morning***, from Visit 4 (Day -14) through Visit 11 (Week 16), subjects must be instructed to arrive **without** having morning meal/breakfast, self-administration of the blinded investigational product, *and* morning dose of concomitant medication for the control of glycemia, dyslipidemia, and BP, if applicable, for the given day;

- At Visit 4 (Day -14) through Visit 11 (Week 16), inclusive, the morning meal will be consumed with above mentioned medications at the site [with the meal *either* provided by the site *or* the subject provided a voucher (or similar) by the site to purchase the meal before arriving to the site for each visit];

CCI [redacted]

- Subjects will be counseled on appropriate dietary and lifestyle guidelines, at Visit 3 (Run-in) and re-iterated at Visit 5 (Day 1):

  - Subjects will be asked to maintain these guidelines throughout participation in the study (ie, Visit 12, the on-site Follow-up);

  - Counseling on dietary guidelines should be individualized in accordance with local medical standards of care for these patients and appropriate for the concomitant medical condition(s) of each subject.

4.4.2. Alcohol, Caffeine, and Tobacco

- Intake of alcohol is permitted in moderation [refer to Exclusion Criterion 3 for limits on amount of alcohol consumption];

- Consumption of caffeinated drinks and nicotine-containing products is permitted during participation in the study; however, there may be a need for brief interruption while at the site and/or Imaging facility, depending on local policy.

4.4.3. Activity

- Subjects will **not** be permitted to engage in physically strenuous exercise (for example: heavy lifting, weight training, calisthenics, and aerobics) within **48-hours** before each blood sample collection for clinical laboratory tests for the duration of participation in the study; physical activity at an individual subject's normal pace is permitted.

4.4.4. Confinement to Site

- Each outpatient visit to the site will occur *with the subject arriving at the site* between approximately 6 AM and 10 AM with each visit envisioned to last ≤ 2-hours;

- Visit to the Imaging facility for MRI-PDFF [± FibroScan®] assessment will have restrictions related to prior meals/food consumption and clock time of day [refer to Schedule of Activities (Table 1) and Section 7.1].

4.4.5. Confinement at Imaging facility

- Each outpatient visit to the Imaging facility (for MRI-PDFF) will occur following a fast (water permitted) of ≥ 4-hours and same time of day (± 2 hours) relative to MRI-PDFF completed at Visit 2 (Screen 2); refer to Section 7.1 for additional details;
    - Visits to Imaging facility can occur on the same day as the visit to the site either prior **or** after the Site visit;
    - Visit to Imaging facility is also permitted to be on days separate from visits to site; but in this case must still occur within the pre-defined visit window [refer to Schedule of Activities (Table 1)].
- For assessment via FibroScan®, visits will occur either to the site **or** the Imaging facility – depending on where the FibroScan® device is located – however, the location of the device must be kept consistent at individual sites throughout the study execution;
    - Similar to MRI-PDFF, each assessment via FibroScan® to occur following a fast (water permitted) of ≥ 4-hours and same time of day (± 2 hours) relative to FibroScan® assessment completed at Visit 1 (Screen 1).

4.4.6. Contraception

In this study, male subjects who are able to father children, and female subjects who are of childbearing potential have an 80% chance of being randomized to receive PF-05221304; CCI [redacted]

Female subjects who, ***in the opinion of the investigator*** are sexually active and at risk for pregnancy must agree to use highly effective contraception throughout the study and continue use for **at least 28 days** after the last dose of blinded investigational product. The investigator or his or her designee, in consultation with the subject, will confirm that the subject has selected the appropriate method of contraception for the individual subject from the list of permitted contraception methods below and instruct the subject in their consistent and correct use. At time points indicated in the Schedule of Activities (Table 1), the investigator or designee will inform the female subject of the need to use the highly effective method of contraception consistently and correctly and document such conversation, and the subject's affirmation, in the subject's chart.

Highly effective methods of contraception are those that, alone or in combination, result in a failure rate of less than 1% per year when used consistently and correctly (ie, perfect use). **One** of the below options must be used to satisfy the requirement for highly effective contraception in female subjects participating in this study:

1. Established use of hormonal methods of contraception associated with inhibition of ovulation (eg, inserted, injected, implanted; *and* including oral, as well as transdermal

methods unless prohibited by local regulations or ethics committee decision), combined with *either*:

a. Male *or* female condom;

b. Male sterilization.

2. Correctly placed copper-containing intrauterine device (IUD) or intrauterine system combined with *either*:

   a. Male *or* female condom;

   b. Male sterilization.

3. Male sterilization with absence of sperm in the postvasectomy ejaculate in the female subjects' partner combined with *either*:

   a. Male *or* female condom;

   b. Bilateral tubal ligation/bilateral salpingectomy or bilateral tubal occlusive procedures.

4. Bilateral tubal ligation/bilateral salpingectomy or bilateral tubal occlusive procedure (provided that occlusion has been confirmed in accordance with the device's label) **combined with one** of the following:

   a. IUD *or* intrauterine system;

   b. Male *or* female condom;

   c. Male sterilization.

All sexually active male subjects must agree to prevent potential transfer to and exposure of female partner(s) to drug through ejaculate by using a condom consistently and correctly, beginning with the first dose of double-blinded investigational product at Visit 5 (Day 1) and continuing for **at least 28 days** after the last dose of blinded investigational product.

In female subjects, pregnancy testing will be performed at time points indicated in the Schedule of Activities (Table 2). The specific contraceptive method(s) in the females of childbearing potential will be documented, and ***at every visit*** their consistent and correct use will be ascertained and documented in the subject's source document. For additional details, refer to Section 7.3.1.

In addition, the investigator or designee will instruct *all subjects of childbearing potential* to call immediately if the selected contraception method is discontinued or if pregnancy is known or suspected in the female subject or male subjects' female partner.

4.5. Sponsor's Qualified Medical Personnel

The contact information for the sponsor's appropriately qualified medical personnel for the study is documented in the study contact list located in the study portal.

To facilitate access to appropriately qualified-medical personnel on study-related medical questions or problems, subjects are provided with a contact card. The contact card contains, at a minimum, protocol and investigational product identifiers, subject study numbers, contact information for the investigator site, and contact details for a contact center in the event that the investigator site staff cannot be reached to provide advice on a medical question or problem originating from another healthcare professional not involved in the subject's participation in the study. The contact number can also be used by investigator staff if they are seeking advice on medical questions or problems; however, it should be used only in the event that the established communication pathways between the investigator site and the study team are not available. It is therefore intended to augment, but not replace, the established communication pathways between the investigator site and the study team for advice on medical questions or problems that may arise during the study. The contact number is not intended for use by the subject directly, and if a subject calls that number, he or she will be directed back to the investigator site.

5. STUDY TREATMENTS

For the purposes of this study, and per International Conference on Harmonisation (ICH) guidelines, investigational product is defined as a pharmaceutical form of an active ingredient or placebo being tested or used as a reference/comparator in a clinical trial, including a product with a marketing authorization when used or assembled (formulated or packaged) in a way different from the approved form, or when used for an unapproved indication, or when used to gain further information about an approved use (ICH E6 1.33).

For this study, the investigational products are tablets for oral administration in the following strengths:

- 1 mg PF-05221304;
- 5 mg PF-05221304;
- 25 mg PF-05221304;
- 50 mg PF-05221304;
- Placebo matching PF-05221304 tablet strengths of 1 mg and 5 mg;
- Placebo matching PF-05221304 tablet strengths of 25 mg and 50 mg.

Note that the drug substance is denoted as PF-05221304-82. All descriptions using this term are equivalent to PF-05221304 for the purpose of identifying the active moiety relevant to this study. An investigational product manual will be provided to the sites prior to the initiation of randomization of subjects into the study.

5.1. Allocation to Treatment

In this study, at least 360 subjects will be randomized to receive 1 of the regimens described in Table 3. Each dose will consist of 3 tablets (1 tablet from *each of the 3* bottles dispensed).

Table 3. Randomized Regimens in Study C1171002

Regimen	Regimen Description (dosed once-daily)	Number of PF-05221304 tablets				Number of PF-05221304-matching placebo tablets	
		1 mg	5 mg	25 mg	50 mg	1 / 5 mg	25 / 50 mg
A	Placebo	-	-	-	-	2	1
B	PF-05221304 – 2 mg	2	-	-	-	-	1
C	PF-05221304 – 10 mg	-	2	-	-	-	1
D	PF-05221304 – 25 mg	-	-	1	-	2	-
E	PF-05221304 – 50 mg	-	-	-	1	2	-

Allocation of subjects to dosing regimens will occur through the use of an interactive response technology (IRT) system (interactive Web-based response [IWR]). The site personnel (study coordinator or specified designee) will be required to enter or select information including but not limited to the user's identification (ID) and password, the protocol number, and the subject number. The IRT system will provide a confirmation report containing the subject number, randomization number, and container number(s) assigned. The confirmation report must be stored in the site's files.

The study-specific IRT reference manual will provide the contact information and further details on the use of the IRT system.

5.2. Breaking the Blind

The study will be subject and investigator blinded.

At the initiation of the study, the investigator site will be instructed on the method for breaking the blind. The method will be an electronic process. Blinding codes should be broken only in exceptional circumstances when knowledge of the actual treatment code is absolutely essential for further management of the subject. Investigators are encouraged to discuss with a member of the study team if they believe that unblinding is necessary. When the blinding code is broken, the reason must be fully documented and entered on the case report form (CRF).

5.3. Subject Compliance

The number of pills returned by the subjects at Visit 5 (Day 1), Visit 7 (Week 4), Visit 9 (Week 8), Visit 10 (Week 12), and Visit 11 (Week 16) will be used to assess subject compliance. Compliance (as assessed by pill count) will be defined as self-administration, by the subjects, of:

- ≥ 90% compliance with pills in *each bottle* of the study-supplied blinded placebo administered from Visit 4 (Day -14) to **1 day prior** to Visit 5 (Day 1), inclusive;

- ≥ 80% compliance with pills in *each bottle* for the double-blinded IP from Visit 5 (Day 1) to Visit 11 (Week 16), inclusive; investigators must closely follow non-compliant, randomized subjects in order to enhance their adherence to IP;

    - *Post randomization*, at each dispensation visit [refer to Schedule of Activities (Table 1)], subjects who are < 80% compliant must be re-educated on the importance of daily administration of investigational product, as much as possible; with the overall aim to maintain ≥ 80% compliance over the 16-week duration of dosing.

5.4. Investigational Product Supplies

5.4.1. Dosage Form(s) and Packaging

Blinded investigational products (PF-05221304 and its matched placebo) will be provided as tablets for oral administration. The 1-mg, 5-mg, 25-mg, and 50-mg strength tablets of PF-05221304 and matching placebo tablets will be supplied in separate bottles and labeled according to local regulatory requirements.

PF-05221304 / matching-placebo for dispensation at the visits denoted in the Schedule of Activities (Table 1) will be packaged with each bottle containing a sufficient number of tablets to permit dosing for 14 (+2) days during single-blind period from Visit 4 (Day -14) to 1 day prior to Visit 5 (Day 1) **or** 28 (+4) days from Visit 5 (Day 1) and onwards.

5.4.2. Preparation and Dispensing

The investigational product will be dispensed using an IRT drug management system from Visit 4 (Day -14) to Visit 10 (Week 12) at selected visits [refer to Schedule of Activities – Table 1]. A qualified staff member will dispense the investigational product via unique container numbers in the bottles provided, in quantities appropriate for the study visit schedule. **Subjects should be instructed to maintain the IP in the bottle(s) provided throughout the course of dosing at the labelled storage conditions** and return with the dispensed bottles to the site **at each visit** starting at Visit 5 (Day 1) and through Visit 11 (Week 16). The instructions for dosing will be provided to the sites and individual subjects (refer to Section 5.5). The number of bottles dispensed and the number of tablets administered per dose will be identical for all randomized regimens.

5.5. Administration

Subjects will be instructed to take the blinded investigational product at the same time of day with the morning meal each day. In addition, subjects will be instructed to delay the self-administration of the blinded investigational product as well as their medications for glycemic control, dyslipidemia, and BP control, if applicable, on the days of their outpatient visit to the site; on these days, these medications will be administered at the site with the morning meal – refer to Section 4.4.1.

Subjects will swallow the investigational product whole, and will not manipulate or chew the investigational product prior to swallowing.

Subjects will be provided dosing instructions at Visit 4 (Day -14) to aide in remembering the correct steps to self-administer blinded investigational product.

5.6. Investigational Product Storage

The investigator **or** an approved representative (eg, pharmacist, study coordinator) will ensure that all investigational products are stored in a secured area with controlled access under required storage conditions and in accordance with applicable regulatory requirements.

Investigational products should be stored in their original containers and in accordance with the labels.

Any storage conditions stated in the SRSD will be superseded by the storage conditions stated on the product label.

Site systems must be capable of measuring and documenting (for example, via a log), at a minimum, daily minimum and maximum temperatures for all site storage locations (as applicable, including frozen, refrigerated, and/or room-temperature products). This should be captured from the time of investigational product receipt throughout the study. Even for continuous-monitoring systems, a log or site procedure that ensures active evaluation for excursions should be available. The intent is to ensure that the minimum and maximum temperature is checked each business day to confirm that no excursion occurred since the last evaluation and to provide the site with the capability to store or view the minimum/maximum temperature for all non-working days upon return to normal operations. The operation of the temperature monitoring device and storage unit (for example, refrigerator), as applicable, should be regularly inspected to ensure they are maintained in working order.

Any excursions from the product label storage conditions should be reported to Pfizer upon discovery. The site should actively pursue options for returning the product to the storage conditions described in the labeling, as soon as possible. Deviations from the storage requirements, including any actions taken, must be documented and reported to Pfizer.

Once an excursion is identified, the investigational product must be quarantined and not used until Pfizer provides permission to use the investigational product. It will not be considered a protocol deviation if Pfizer approves the use of the investigational product after the temperature excursion. Use of the investigational product prior to Pfizer approval will be considered a protocol deviation. Specific details regarding information the site should report for each excursion will be provided to the site prior to the initiation of Visit 4 (Day -14) in this study.

Site staff will instruct subjects on the proper storage requirements for take home investigational products. The site staff should refer to the Investigational Product Manual for additional guidance on storage conditions and actions to be taken when conditions are outside of the specified range.

5.7. Investigational Product Accountability

The investigator site must maintain adequate records documenting the receipt, use, loss, or other disposition of the investigational product supplies. All investigational products will be accounted for using a drug accountability form/record.

All bottles of blinded investigational product must be returned to the investigator by the subject at every dispensation visit and at the end of the study [refer to Schedule of Activities – Table 1].

5.7.1. Destruction of Investigational Product Supplies

At the end of the study, the sponsor or designee will provide guidance on the destruction of *unused* investigational product (eg, at the site). If destruction is authorized to take place at the investigator site, the investigator must ensure that the materials are destroyed in compliance with applicable environmental regulations, institutional policy, and any special instructions provided by Pfizer, and all destruction must be adequately documented.

5.8. Concomitant Treatment(s)

Given the duration of the dosing phase in this study (ie, up to 16 weeks), ***whenever possible***, attempts must be made to ***not*** alter the doses and regimens of the background/concomitant medications after randomization [ie, Visit 5 (Day 1)], and until Visit 12 (on-site Follow-up).

All concomitant medications taken during the study must be recorded with, at a minimum, the indication for use. In addition, for diabetes, lipid-modifying, BP control medications, daily dose, and start and stop dates of administration must also be captured. All subjects must be questioned about concomitant medication at each outpatient visit.

Medications taken within **6-weeks** before **Visit 5 (Day 1)** will be documented as a prior medication. Medications taken after dosing of double-blind IP at Visit 5 (Day 1) and until the 2nd Follow-up visit (or telephone contact) will be documented as concomitant medications.

5.8.1. Medications for Glycemic Control

The use of the following classes of agents is **not** permitted within **8-weeks prior to Visit 1 (Screen 1)** and until Visit 12 (on-site Follow-up):

- TZDs such as pioglitazone and rosiglitazone;
- Subcutaneously administered agents for glycemic control (eg, insulin, exenatide, liraglutide, pramlintide).

Subjects are permitted to be on stable doses of up to a ***maximum of 3 oral agents for glycemic control,*** starting at ≥ **8-weeks prior to Visit 1 (Screen 1)** and until Visit 12 (on-site Follow-up), across the country-specific, approved classes of agents including (but not limited to) the following:

- Biguanide such as metformin;
- Sulphonylureas such as acetohexamide, chlorpropamide, tolazamide, tolbutamine, glimepiride, glipizide, glyburide;
- Meglitinide analogues such as repaglinide, nateglinide;
- Dipeptidyl peptidase-IV inhibitors (DPP-IVi) such as sitagliptin, saxagliptin, vildagliptin;
- α-glucosidase inhibitors such as acarbose, miglitol;
- Sodium-glucose cotransporter 2 inhibitors such as canagliflozin, dapagliflozin, empagliflozin.

5.8.2. Lipid-modifying medications

The use of monoclonal antibodies inhibiting proprotein convertase subtilisin/kexin type 9 (PCSK9) such as alirocumab, evolocumab is ***not*** permitted **within 12-weeks prior to Visit 1 (Screen 1)** and until Visit 12 (on-site Follow-up).

Subjects are permitted to be on stable doses of up to a ***maximum of 3 lipid-modifying agents,*** starting at ≥ **8-weeks prior to Visit 1 (Screen 1)** and until Visit 12 (on-site Follow-up), across the country-specific, approved classes of agents including (but not limited to) the following –

- Statins such as atorvastatin, simvastatin, rosuvastatin, pravastatin;
- Bile acid sequestrants such as cholestyramine, colestipol;
- Fenofibrate , a fibric acid derivative;
- Nicotinic acid / niacin;
- Ezetimibe;
- Doses and agent(s) are permitted to be adjusted at Visit 3 (Run-in), with stable doses for **≥ 6-weeks** before Visit 5 (Day 1) required for continuing participation in this study.

- The use of the following selected agents is **not** permitted starting at Visit 3 (Run-in) and until Visit 12 (on-site Follow-up):
    - Gemfibrozil (a potent OATP inhibitor);
    - Colesevalam.
    - *Subjects on either of the above two oral agents*, at Visit 3 (Run-in), must be willing to switch to other acceptable agents and on stable doses of the acceptable agents for **≥ 6-weeks** before Visit 5 (Day 1) in order to participate in this study.

5.8.3. Medications for Controlling Blood Pressure

Across the many classes of agents for management of hypertension, subjects are permitted to be on stable doses of up to a ***maximum of 3 agents,*** starting at ≥ **8-weeks prior to Visit 1 (Screen 1)** and until Visit 12 (on-site Follow-up), across the country-specific, approved classes of agents;

- Starting at Visit 3 (Run-in), doses of medications (and additional medications can be added to a maximum of 3) in order to control BP and meet criteria needed to progress to randomization [refer to Section 4.2, Exclusion Criterion 6].

5.8.4. Other Acceptable Concomitant Medications

Any subject on the following list of medications ***must be*** on stable doses [ie, **≥ 8-weeks prior to Visit 1 (Screen 1)** and until Visit 12 (on-site Follow-up), ***where possible***]:

- Non-steroidal anti-inflammatory medications (NSAIDs) such as ibuprofen, ketoprofen, diclofenac, naproxen, indomethacin, meloxicam; and celecoxib;
    - Also permitted is *intermittent* use of acetaminophen/paracetamol at doses of up to 1 gram per day ***and*** intermittent use of NSAIDs (*for example*: for short-term pain management);
- Inhaled and topical corticosteroids:
    - *NOTE*: Intercurrent treatment with systemic steroids, during participation in the study, may be permitted if treatment does/will not exceed 14-days;
- Thyroid replacement therapy, as well as acceptable contraception medication and hormone replacement therapy (in eligible female subjects), etc;
- Anti-psychotic medications such as olanzapine, risperidone;
- Antidepressant medications such as tricyclic agents, selective serotonin reuptake inhibitors, and serotonin/norepinephrine reuptake inhibitors;

- Vitamin E, including high doses so long as regimen *is stable*;
- Certain herbal supplements ***but only*** following consultation with sponsor.

5.8.5. Prohibited Medications

Across the agents used for glycemic control, lipid-modification, and BP control, certain medications are *not* permitted – refer above to Section 5.8.1 to Section 5.8.3.

Also, use of drugs historically associated with fatty liver are prohibited within **any ≥ 4-week interval** in the previous 12 months prior to Visit 1 (Screen 1) –

- *Examples* include: amiodarone, methotrexate, systemic glucocorticoids (such as prednisone, dexamethasone, triamcinolone, budesonide, betamethasone), anabolic steroids, tetracyclines, tamoxifen, estrogens at doses greater than those used for hormone replacement, valproic acid, other known hepatotoxins;

In addition, subjects using the following medications within **8-weeks prior to** Visit 1 (Screen 1) **or** needing them at any time until Visit 12 (on-site Follow-up) should **not** participate in this study –

- *Chronic use* of immunosuppressants such as cyclosporine and tacrolimus;
- Pharmacological agents with *approved indication* for weight loss such as orlistat and sibutramine;
- Over-the-counter appetite- simulant or appetite- suppressant, as advertised;
- (Medical-grade) marijuana, regardless of medical indication;
- Specific classes of agents given current study is 1st study in patient population –
    - Coumadin-type anticoagulants *or* other anticoagulants (eg, dabigatran); *though* aspirin at doses ≤ 325 mg/day is permitted
    - Anticonvulsants – specifically when prescribed for the explicit management of seizures (though use of some agents for other indications *may* be permitted; *for example*: use of benzodiazepines for management of anxiety);
    - Antiarrhythmics – specifically those agents whose primary mechanism of action is sodium or potassium channel blockade (eg, procainamide, phenytoin, quinidine, propafenone; as well as amiodarone, dofetilide, sotalol);
        - **NOTE:** β-adrenergic receptor blocking agents (eg, atenolol, metoprolol) and calcium channel blockers (eg, diltiazem, amlodipine, nifedipine) are permitted;

CCI [redacted]

- Use of chronic agents which are clinically significant OATP inhibitors (eg, cyclosporine, gemfibrozil, rifampin).

5.9. Rescue Medication

There is no rescue therapy to reverse any adverse events (AEs) observed with administration of investigational product; standard medical supportive care must be provided to manage the AEs including administration of carbohydrates to treat hypoglycemic events.

Subjects with T2DM with ***repeated*** **fasting** glucose values > 270 mg/dL (15.0 mmol/L), as reported by the sponsor-identified central laboratory **or** self-checking conducted by subjects via sponsor-provided glucometers, must be withdrawn.

6. STUDY PROCEDURES

Prior to the conduct of any study-specific procedures – at the Pre-Qualification Visit ***and separately*** at Visit 1 (Screen 1) – each subject must provide informed consent.

- Request for the subjects to arrive at the site following an overnight fast of ≥ 8-hours for either the Pre-Qualification *or* Screen 1 visit will **not** be considered study-specific; such instruction is classified as routine standard-of-care for the targeted population for this study.

6.1. Proposed Chronology of Procedures

For the procedures described below, where multiple procedures are scheduled at the same timepoint(s) relative to dosing, the following chronology of events should be adhered to, ***where possible***:

- *12-lead ECG:* obtain prior to vital signs assessment, blood samples, and prior to dosing [refer to Section 7.2.3];
- *Vital Signs (blood pressure, pulse rate)*: obtain after 12-lead ECG collection but prior to obtaining blood samples and prior to dosing [refer to Section 7.2.4];
- *Fasting blood samples* [for safety (Section 7.3), CCI after assessment of 12-lead ECG and vital signs but prior to dosing;

CCI

- *Other pre-dose procedures*: should be obtained/performed as close as possible to the scheduled time, but may be obtained before or after blood specimen collection;
- *Dosing*: must occur with the morning meal; and where applicable, after any predose blood sample collection(s).

6.2. Pre-Qualification

Refer to the Schedule of Activities (Table 1 and Table 2) for the study procedures to be completed at the pre-qualification visit. Subjects can only proceed to Visit 1 (Screen 1) of the main study when deemed to meet eligibility criteria [refer to Section 4.1 and Section 4.2] for the selected procedures completed as part of pre-qualification.

6.3. Screening (Visits 1 and 2)

Refer to the Schedule of Activities (Table 1 and Table 2) for the study procedures to be completed at the 1st and 2nd Screening visits. **The two screening visits are to be performed sequentially**.

- At Visit 1 (Screen 1), all screening procedures *except* assessment of liver fat by MRI-PDFF, will be undertaken to determine eligibility – refer to Section 4.1 and Section 4.2;
- **Only** after subjects are identified to be otherwise eligible, should they proceed with the Visit 2 (Screen 2) to the Imaging facility to undergo liver fat assessment via MRI-PDFF acquisition to assess whether they meet Inclusion Criterion #11.

Subjects will be screened **within 8-weeks (minimum)** *and* **14-weeks (maximum)** prior to the first dose of the blinded investigational product to confirm that they meet the subject selection criteria for the study. The investigator (or an appropriate delegate at the investigator site) will obtain informed consent from each subject.

In ***rare*** cases, subjects may be re-screened; however, this is permitted ***only*** when, due to ***logistical constraints***, the maximum period between Visit 1 to Visit 5 (ie, Screen 1 to Day 1), of **14 weeks,** is exceeded. In such cases, all pre-screening ***and*** screening procedures must be repeated and the subject assigned a new 8-digit SSID number. Subjects must be deemed to meet all the eligibility criteria including assessment of liver fat via MRI-PDFF at Screen 2 visit, under the new 8-digit SSID.

To prepare for study participation, subjects will be instructed on the Lifestyle Requirements (Section 4.4) and Concomitant Treatments (Section 5.8).

6.4. Visit 3 (Run-in) through to Visit 11 (Week 16)

Refer to the Schedule of Activities (Table 1 and Table 2) for the study procedures to be completed starting at Visit 3 (Run-in) through Visit 11 (Week 16).

6.5. Follow-up Visit

In this study, there are two Follow-up visits –

- **1st is an on-site visit** where the subjects will return to the site 10 ± 4 days post last dose of blinded investigational product – Visit 12;
- **2nd is a telephone contact visit** (unless local regulations dictate on-site visit) to occur ≥ 28- to ≤ 35-days, inclusive, following the last dose of blinded investigational product
  - *NOTE*: this visit can be considered as on-site in order to permit follow-up of open TEAEs and/or abnormal laboratory tests from prior visit(s), as needed.
- Refer to the Schedule of Activities (Table 1 and Table 2) for the study procedures to be completed at each of the two Follow-up visits.

6.6. Subject Withdrawal / Early Termination

Subjects may withdraw from the study at any time at their own request, or they may be withdrawn at any time at the discretion of the investigator or sponsor for safety (refer to Section 8.1.3) or behavioral reasons, or the inability of the subject to comply with the protocol-required schedule of study visits or procedures at a given study site.

In this study, any subject who discontinues participation in the trial post the pre-qualification visit (eg, viewed as ineligible based on procedures completed), **or** discontinues based on Screen 1 or Screen 2 procedures (ie, Screen Fail) will have no additional procedures completed. **However, an early termination visit is to be considered** for all subjects who were randomized *and* received at least 1 dose of the investigational product, *and then* are prematurely withdrawn from the study. Subjects should return to the site for final safety assessments **to be scheduled as early as practically feasible following the decision to withdraw but ≤ 14-days after last dose of blinded investigational product.** Subjects should be questioned regarding their reason for withdrawal. At the early withdrawal visit, every effort must be made to complete the assessments outlined in the Schedule of Activities (Table 1 and Table 2). Lack of completion of all or any of the early termination procedures will not be viewed as protocol deviations so long as the subject's safety was preserved.

Withdrawal of consent:

Subjects who request to discontinue receipt of study treatment will remain in the study and must continue to be followed for protocol specified follow-up procedures. The only exception to this is when a subject specifically withdraws consent for any further contact with him or her or persons previously authorized by the subject to provide this information.

Subjects should notify the investigator in writing of the decision to withdraw consent from future follow-up, whenever possible. The withdrawal of consent should be explained in detail in the medical records by the investigator, as to whether the withdrawal is only from further receipt of investigational product or also from study procedures and/or posttreatment study follow-up, and entered on the appropriate CRF page. In the event that vital status (whether the subject is alive or dead) is being measured, publicly available information should be used to determine vital status only as appropriately directed in accordance with local law.

Lost to follow-up:

All reasonable efforts must be made to locate subjects to determine and report their ongoing status. This includes follow-up with persons authorized by the subject as noted above. Lost to follow-up is defined by the inability to reach the subject after a minimum of 2 documented phone calls, faxes, or e-mails as well as lack of response by the subject to 1 registered mail letter. All attempts should be documented in the subject's medical records. If it is determined that the subject has died, the site will use locally permissible methods to obtain the date and cause of death. If the investigator's use of a third-party representative to assist in the follow-up portion of the study has been included in the subject's informed consent, then the investigator may use a sponsor-retained third-party representative to assist site staff with obtaining the subject's contact information or other public vital status data necessary to complete the follow-up portion of the study. The site staff and representative will consult publicly available sources, such as public health registries and databases, in order to obtain updated contact information. If, after all attempts, the subject remains lost to follow-up, then the last-known-alive date as determined by the investigator should be reported and documented in the subject's medical records.

If a subject does not return for a scheduled visit, every effort should be made to contact the subject. All attempts to contact the subject and information received during contact attempts must be documented in the subject's medical record. In any circumstance, every effort should be made to document subject outcome, if possible. The investigator should inquire about the reason for withdrawal, request that the subject return all unused investigational product(s), request that the subject return for a final visit, if applicable, and follow up with the subject regarding any unresolved adverse events (AEs).

If the subject withdraws from the study, and also withdraws consent for disclosure of future information, no further evaluations should be performed, and no additional data should be collected. The sponsor may retain and continue to use any data collected before such withdrawal of consent.

Subjects withdrawn from the study, after receiving at least 1 dose of the double-blinded IP (ie, after dosing at Visit 5 [Day 1]), will **not** be replaced – except those with non-evaluable baseline MRI-PDFF assessment – refer to Section 7.1.4.

7. ASSESSMENTS

Every effort should be made to ensure that the protocol-required tests and procedures are completed as described. However, it is anticipated that from time to time there may be circumstances outside of the control of the investigator that may make it unfeasible to perform the test. In these cases the investigator will take all steps necessary to ensure the safety and well-being of the subject. When a protocol-required test cannot be performed, the investigator will document the reason for this and any corrective and preventive actions that he or she has taken to ensure that normal processes are adhered to as soon as possible. The study team will be informed of these incidents in a timely manner.

For samples being collected and shipped, detailed collection, processing, storage, and shipment instructions and contact information will be provided to the investigator site prior to initiation of the study.

7.1. Liver Assessments using FibroScan® and MRI-PDFF

7.1.1. Background on Liver Assessment Techniques

FibroScan® is an ultrasound based technique permitting an assessment of liver fat (via controlled attenuation parameter - CAP™) and liver stiffness (via vibration controlled transient elastography - VCTE™ and reported as liver stiffness measure – LSM), developed by EchoSens. These measurements are performed on the right lobe of the liver through intercostal spaces with the subject lying in dorsal decubitus with the right arm in maximal abduction. A transducer probe delivers a painless, mechanical impulse to the liver and measures the resulting shear wave. The LSM is directly proportional to the velocity of the shear wave. Simultaneously, the CAP™ measures the ultrasonic attenuation coefficient in the forward and return path of the radio-frequency pulse through the liver. To account for varying subcutaneous fat layers, the device is equipped with *at least two probes*; the **M+ probe** for lean/overweight adult subjects, and the **XL+ probe** for overweight/obese adults. After a total of 10 valid, serial measures are acquired per assessment, the device calculates the median LSM and CAP™ values and reports these values simultaneously.

MRI-PDFF technique further improved upon the conventional Dixon in- and out-of-phase (IOP) method to give a quantitative, standardized, and objective MRI measurement of fat content. MRI-PDFF utilizes a gradient echo sequence with low flip angle (FA) to minimize T_1 bias, corrects T_2* decay (due to iron overload) via modeling of the fat signal as a superposition of multiple frequency components from five different lipid types, and is applied in each of the 9 Couinaud segments. This technique improves fat quantification accuracy for the entire liver permitting quantification of small differences/changes following pharmacological intervention. Relative to the reference standard of magnetic resonance spectroscopy (MRS), MRI-PDFF has the advantages of permitting fat mapping for the entire liver via an imaging-based technique versus a biochemical-based technique that provides data on liver fat content in a small region of interest in the liver.

7.1.2. Assessment of Liver Fat and Stiffness – using FibroScan®

In this study, assessment of liver fat and liver stiffness using FibroScan® will occur at *scheduled* visits outlined in the Schedule of Activities (Table 1) and Section 4.4.5. The results for liver fat (via CAP™) in decibels per meter (dB/m) and liver stiffness (via LSM) in kilopascals (kPa) to determine whether an individual subject qualifies at pre-qualification to progress to Screen 1 (or at Screen 1 to progress to Screen 2) will be displayed on the FibroScan® device at the end of each assessment. These results do *not* need independent over-read but steps to ensure that acquisition was complete and accurate are required – per training to be offered to **at least 2 site staff** (who must be sonographers or comparable) by EchoSens, ***and*** certified as operators based on this training by EchoSens, at the start of the study. *As much as practically possible*, attempts will be made to ensure each individual subjects' assessment is performed by the same site staff.

Each acquisition will require the subjects to be in a supine position with the right arm placed flat above the head and will take approximately 10 minutes. Each assessment must be undertaken **following a fast (except water) of ≥ 4-hours**, and ***as much as practically possible***, at the same time (± 2-hours) of the day relative to assessment at Visit 1 (Screen 1).

Complete details regarding acquisitions using FibroScan® to assess CAP™ and LSM will be offered by EchoSens before the initiation of pre-qualification portion of this study. All images acquired must be saved by the site until the conclusion of the study; the summary of numerical results (including quality-related outputs) must be printed and saved by the site (or Imaging facility, as applicable) as part of each subject's source documents.

7.1.3. Assessment of Liver Fat Using MRI-PDFF Acquisition and Analysis

In this study, at *scheduled* visits [refer to the Schedule of Activities (Table 1) and Section 4.4.5] liver fat will be assessed via MRI, using the PDFF acquisition protocol. In order to ensure that the population identified for this trial can be appropriately imaged via an MRI machine, **at the pre-qualification visit,** subjects will undergo a **hula-hoop test –**

- Subject will be asked to be in the standing position with arms either to the side **or** vertically up above the head, feet positioned close together, and weight evenly distributed across both feet;
    - *NOTE:* as much as practically possible, the same position of the arms must be used for all MRI-PDFF assessments in this trial;
- Using a sponsor-approved hula-hoop, an attempt will be made to slide the hoop over the subjects head and determine if the hoop can pass over the subjects' trunk and hips;
- Hula-hoop will be passed while the subject is at the end of a *normal* expiration (when lungs are at their residual capacity);
- **If** the hula-hoop slides easily from the head down to the feet, the subject is eligible and confirmed to fit inside the site-specific MRI machine and its respective bore size;

- If the hula-hoop cannot slide down to the feet (eg, fits snug at the waist or hips), the subject is deemed ineligible for this study;
- Additional details on steps for identification of the appropriate hula-hoop circumference, which depends on the individual Imaging facility's MRI machine opening (ie, bore diameter) along with bed thickness, and coils selected, will be provided in an Imaging Manual offered to the sites prior to the start of the study.

Transportation of the subjects to the Imaging facility does ***not*** need to be supervised by the site staff. Each assessment will require the subjects to be in a supine position in the confined space of the MRI scanner for approximately 25 minutes with the image acquisition undertaken **following a fast (except water) of ≥ 4-hours**, and ***as much as practically possible***, at the same time (± 2-hours) of the day relative to assessment at Visit 2 (Screen 2).

Across the sites selected for this study, the sponsor-identified central imaging vendor will train the staff at the Imaging facility on the MRI-PDFF acquisition protocol, on just-in-time review of the acquired images for assessment of images being deemed evaluable, and on transfer (preferably electronically) of the images to the sponsor-identified central imaging vendor for analysis and quantification of liver fat. **Only the staff members at the Imaging site who are trained by the sponsor-identified central imaging vendor are permitted to acquire images in the subjects who consent for this study;** *in rare/limited situations, exceptions may be granted with written approval of the sponsor*. Complete details on the MRI-PDFF acquisition protocol, determination of quality of images, and transmission of data to sponsor-identified central imaging vendor will be provided in an Imaging Manual offered to the sites prior to the start of the study.

As much as practically possible, analysis of the MRI-PDFF images acquired from baseline (ie, between Visit 4 and Visit 5) to Visit 11 (Week 16) will be undertaken by a single colleague at the sponsor-identified central imaging vendor ***who will be blinded*** to individual subjects' clinical data, as well as randomization and stratification assignment. However, this colleague will be unblinded to Visit of each assessment (ie, Screen 2, Baseline, etc).

7.1.4. Analysis of MRI-PDFF Images Including Determination of Eligibility

Subjects' eligibility for this study based on liver fat as assessed via MRI-PDFF at Screen 2 will be made by the sponsor-identified central imaging vendor, *only*. The individual subjects' liver fat result will ***not*** be communicated to the site [though results from Screen 2 and Baseline *may* be known to the sponsor]. In the case of the Screen 2 MRI, sites will *only* be informed whether the subjects meet eligibility criteria or if the screening MRI should be repeated once – as determined by the sponsor-identified central imaging vendor. For ***all*** subsequently scheduled MRI-PDFF assessments, sites will *only* be informed whether the images are deemed evaluable (or not); of note, subjects with non-evaluable baseline images [as determined by the sponsor-identified central imaging vendor] *may* be withdrawn and replaced, at the discretion of the sponsor.

Management of incidental findings

An incidental finding is one unknown to the subject that has potential health or reproductive importance, which is discovered unexpectedly in the course of a research study, but is unrelated to the purpose and beyond the aims of the study.

The MR images will be reviewed by the sponsor-identified central imaging vendor. The purpose of this review is to evaluate images for the amount of fat in the liver. Central image review is not a complete medical review of the subject. If, during the central review process, an unexpected observation is identified and this finding could, in the opinion of the central reviewer, have a significant health or reproductive consequence, this finding may be shared with the study sponsor for disclosure to the principal investigator. All follow-up testing and final diagnosis will be left to the discretion of the medical professionals at the site or those with an existing physician-patient relationship. The principal investigator will be responsible for reporting any AEs identified from incidental findings as described in the Adverse Event Reporting section. Identification of such incidental findings during the central review process should not be expected, and the site maintains responsibility for performing a general safety review of all images as per site protocols.

CCI

[redacted]

7.2. Safety-Related Assessments

7.2.1. Physical Examination

In this study, physical examinations are to be performed at nominal time points specified in the Schedule of Activities (Table 1).

Physical examinations may be conducted by a physician, trained physician's assistant, or nurse practitioner as acceptable according to local regulation. A **full physical examination** will include head, ears, eyes, nose, mouth, skin, heart and lung examinations, lymph nodes, gastrointestinal, musculoskeletal, and neurological systems. A **targeted physical examination** specifically covering visual examination of skin, examination of the eyes using an ophthalmoscope, as well as auscultation of the pulmonary system, will be conducted at Visit 6 (Week 2), Visit 9 (Week 8) and Visit 11 (Week 16). *In addition*, this targeted physical examination will be incorporated into the full physical examinations prior to the first dose of the investigational product. The **brief physical examination** will be focused on general appearance, the respiratory and cardiovascular systems, as well as towards subject reported symptoms.

7.2.1.1. Measurement of Waist Circumference

Waist circumference will be measured at the nominal time points specified in the Schedule of Activities (Table 1) using a flexible anthropometric tape and ideally, reporting the measurement in centimeters with accuracy to the nearest 0.1 centimeter (or 1/16th inch). Measurement will be undertaken as follows:

- Either before or after the assessment of body weight;
- While subject is in a standing position with arms resting comfortably at the side;
- Under standard conditions including post-void, with the tape touching the skin (not clothing);
- At the end of a normal expiration (when lungs are at their residual capacity).

And the measurement will consider the following anatomical features as benchmarks:

- Circumference of the narrowest part of the torso as viewed from the anterior aspect **or;**
- If the narrowest part of the torso cannot be identified, the measurement must be made of the smallest horizontal circumference in the area between the ribs and the iliac crest.

7.2.2. Body Weight

In this study, assessment of body weight will occur at the nominal time points specified in the Schedule of Activities (Table 1) per the following specifications –

- Weight will be recorded using a scale placed on a stable, flat surface in a (semi)-private area;
- Same scale, ***as much as practically possible***, will be used with the scale reporting weight in kilograms or pounds, and accuracy to the **nearest 0.1 kg [or 0.2 pounds]** - ie, the device used for this study must be able to distinguish a difference between 68.4 kg and 68.3 kg.
- Measurement must be undertaken:
  - At approximately the same time of the day at each nominal time point;
  - After the subject has been asked to void (ie, forced void);
  - Under standard conditions (eg, subjects must wear light clothing with content of their pockets emptied or hospital gown **and not** be wearing shoes or bulky layers of clothing/jackets);

- With subjects remaining still while on the weighing scale.

7.2.3. 12-lead Electrocardiogram

In this study, assessment of cardiac conduction via 12-lead ECGs will occur at the nominal time points specified in the Schedule of Activities (Table 1) per the following specifications:

- All *scheduled* 12-lead ECGs should be performed after the subject has rested quietly for **≥ 10-minutes** in a **supine** position;
- **Single supine 12-lead ECGs** will be obtained; the ECG collected at Visit 5 (Day 1) will serve as each subject's baseline;
- Refer to Section 6.1 for proposed chronology of procedures for nominal time points when 12-lead ECG assessments coincide with other procedures.

In some cases, it may be appropriate to repeat abnormal 12-lead ECGs to rule out improper lead placement as contributing to the ECG abnormality. It is important that leads are placed in the same positions each time in order to achieve precise ECG recordings. If a machine-read QTc value is prolonged, as defined above, repeat measurements may not be necessary if a qualified physician's interpretation determines that the QTc values are in the acceptable range.

7.2.4. Blood Pressure and Pulse Rate

In this study, assessment of vital signs (including seated blood pressure, and pulse rate) will occur at the nominal time points specified in the Schedule of Activities (Table 1) per the following specifications –

- *At the pre-qualification visit*, the subjects' arm circumference [eg, using a flexible anthropometric tape] should be measured at the midpoint of the length of the upper arm and the appropriate cuff selected and used throughout the study to measure BP/pulse rate via an automated device using an oscillometric method (not auscultation):
  - Subjects with arm circumference greater than the largest cuff size available at each site are not eligible.
- **Single seated** blood pressure/pulse rate will be measured with the subject's arm supported at the level of the heart, and recorded to the nearest mm Hg, following a rest of **≥ 5-minutes**; the assessment at Visit 5 (Day 1) will serve as each subject's baseline;
- Same arm (preferably the dominant arm) will be used for blood pressure/pulse rate assessment throughout the study;

- Blood pressure/pulse rate assessment should not be taken from the arm with an intravenous catheter, if placed;

- Subjects should be instructed not to speak during blood pressure/pulse rate measurements;

- Refer to Section 6.1 for proposed chronology of procedures for nominal time points when vital sign assessments coincide with other procedures.

Additional collection times, or changes to collection times of blood pressure /pulse rate will be permitted, as necessary, to ensure appropriate collection of safety data.

7.3. Clinical Laboratory Tests

The blood/urine samples for the clinical laboratory tests [summarized in Table 4] related to safety and exploratory biomarkers will be collected at the nominal time points specified in the Schedule of Activities (Table 2).

Table 4. All Planned Safety-Related Laboratory Tests and Exploratory Biomarkers in Study C1171002

Hematology	Chemistry	Urinalysis	Other
−Hemoglobin −Hematocrit −RBC count −Reticulocyte count (Abs) −MCV −MCH −MCHC −Platelet count −WBC count −Total neutrophils (Abs) −Eosinophils (Abs) −Monocytes (Abs) −Basophils (Abs) −Lymphocytes (Abs)	−BUN −Creatinine [***and*** eGFR using MDRD] −*Plasma* glucose (fasting) −Calcium −Sodium −Potassium −Chloride −Total CO_2 (Bicarbonate) −AST (SGOT) −ALT (SGPT) −Alkaline phosphatase −GGT −Total Bilirubin −Direct (conjugated) bilirubin[a] −Indirect (unconjugated) bilirubin[a] −Total bile acids −Creatine Kinase[b] −Uric acid −Albumin −Total protein	−pH −Glucose (qual) −Protein (qual) −Blood (qual) −Ketones −Nitrites −Leukocyte esterase −Urobilinogen −Urine bilirubin −Microscopy[c]	−Plasma aPTT, PT, INR −Serum FSH[d] −Serum and urine pregnancy test [refer to Section 7.3.1] −Urine drug test[e] −Serology[f]: HBsAg, HBcAb, HCVAb (and if positive, reflex HCV RNA), HIV −α1-antitrypsin[f] −Ceruloplasmin[f] −Serum % carbohydrate deficient transferrin[f] −HbA1C[g] −Fasting Lipid Panel[h]
Additional Tests (Needed for instances of suspected Hy's law) −AST, ALT −Total bilirubin −Albumin −Alkaline phosphatase −Direct bilirubin −Indirect bilirubin −Creatine kinase −GGT −PT/INR −Total bile acids −Acetaminophen drug levels and/or protein adduct level		**Additional exploratory biomarker assessments[i] include:** −Serum apolipoproteins A1, B(total), C3, E CCI [redacted] −Plasma Insulin (fasting) −Calculate HOMA-IR using fasting glucose & insulin CCI [redacted] −CK18-M30, CK18-M65 −ProC3 & ProC6 CCI [redacted]	

a. Direct + indirect bilirubin measured at Pre-qualification, Screen 1, V4, and V5; after initiation of investigational product, direct + indirect bilirubin measured only when total bilirubin is >ULN
b. Test to be assessed at Pre-qualification, Screen 1, V4, and V5; after initiation of investigational product, this test measured only when ALT or AST is >ULN
c. Only if urine dipstick is positive for blood, protein, nitrites or leukocyte esterase
d. At Pre-qualification, Screen 1, ONLY; and in females, only
e. At Pre-qualification, Screen 1, V4, ONLY; minimum requirement for urine drug test include cocaine, tetrahydrocannabinol (THC), opiates/opioids, benzodiazepines and amphetamines
f. At Pre-qualification, Screen 1, ONLY [*in addition*, %CDT also assessed at V4, V5, *and* V11]
g. HbA1C = glycosylated hemoglobin (NGSP certified method)
h. Includes total cholesterol, triglycerides, VLDL, HDL-C, and *direct* LDL-C
i. As part of clinical laboratory tests collected at V4 though V11, only – refer to Table 2

For list of terms corresponding to the abbreviations used herein, refer to Appendix 1

- **All** blood samples for *scheduled* **clinical laboratory tests** must be obtained following an overnight fast (except water) of **≥ 8-hours**.

Additional laboratory results may be reported on these samples as a result of the method of analysis **or** the type of analyzer used by the sponsor-identified central laboratory or secondary laboratory; **or** as derived from calculated values. These additional tests would *not* require additional collection of blood.

Unscheduled clinical laboratory tests ***for subject safety*** may be obtained at any time during the study to assess any perceived safety concerns and shipped to the sponsor-identified central laboratory for analysis.

Additional details regarding sample collection, processing, and shipment will be offered in the study-specific central laboratory manual to be provided prior to the start of the study.

7.3.1. Pregnancy Testing

All pregnancy tests (serum *and* urine) used in this study must have a sensitivity of **at least 25 mIU/mL,** and must be performed using supplied offered by the sponsor-identified central laboratory certified laboratory.

For female subjects of childbearing potential, following a negative pregnancy test result ***and*** after confirmed eligibility based on Visit 2 (Screen 2) tests(s), appropriate contraception must be commenced and the second negative pregnancy test result will then be required prior to randomization. *In addition*, *at a minimum*, 2 negative *serum* pregnancy tests (ie, negative pregnancy test at Visit 1 and Visit 4) ***plus*** a negative *urine* at Visit 4 (Day -14) and Visit 5 (Day 1; randomization) are required before receiving double-blind IP – starting on Day 1. In the absence of regular menstrual bleeding, the female subject should have used highly effective method of contraception (refer to Section 4.4.6) for **at least 28-days** before Visit 4 (Day -14).

Pregnancy tests will be performed at times defined in the Schedule of Activities (Table 2) to confirm that the subject has not become pregnant during the study. Pregnancy tests will also be done whenever 1 menstrual cycle is missed during the active treatment period (ie, Day 1 through Week 16, inclusive) and when potential pregnancy is otherwise suspected, and may be repeated if requested by institutional review boards (IRBs)/ethics committees (ECs) or if required by local regulations. In the case of a positive confirmed pregnancy, the subject will be withdrawn from administration of investigational product and from the study.

7.4. Rater Qualifications

In this study, the AUDIT questionnaire will be completed by the site staff based on responses offered by the subjects – at the visits outlined in the Schedule of Activities (Table 1). Training for the site staff completing this questionnaire will be offered by the sponsor as part of the protocol-specific training ahead of the initiation of the study.

CCI [redacted]

CCI

[redacted]

7.6. Exploratory Biomarkers

Blood samples for exploratory biomarkers – including NASH-related biomarkers, potential mechanism-related parameters, metabolic parameters, and exploratory markers of liver function – will be collected at the nominal time points defined in the Schedule of Activities (Table 2). Refer to list of endpoints outlined in Section 2 that encompass these exploratory biomarkers.

- These samples must be processed and shipped as indicated in the study-specific laboratory manual provided to the investigator site, prior to initiation of study, to maintain sample integrity:

  - Any deviations from the sample handling procedure (eg, sample collection and processing steps, interim storage or shipping conditions), including any actions taken, must be documented and reported to the sponsor. On a case-by-case basis, the sponsor may make a determination as to whether sample integrity has been compromised. Any deviation from the specified sample handling procedure resulting in compromised sample integrity will be considered a protocol deviation;
  - Any *scheduled* collection prior to next dose of blinded investigational product, if undertaken post dose, will be captured as a protocol deviation even if results are deemed evaluable;
- Refer to Section 6.1 for proposed chronology of procedures for nominal time points when these collections coincide with other procedures;
- Samples will be analyzed using a validated analytical method (which need **not** meet Good Laboratory Practice (GLP) standards, especially for these exploratory endpoints) but in all cases, the method will be in compliance with Pfizer standard operating procedures.

CCI

CCI

CCI

7.8. Blood Volume

In this study, the total blood sampling volume for individual subjects will be approximately **400 mL.** This includes approximately **40 mL** blood drawn across the clinical laboratory tests at the pre-Qualification; and the remaining approximately **360 mL** blood drawn from Visit 1 (Screen 1) through the On-site Follow-up visit, inclusive.

Additional blood samples may be taken ***for safety assessments, only***, provided the total volume taken during the study does ***not*** exceed 550 mL.

7.9. Specific Subject Safety Scenarios

7.9.1. Potential Cases of Thrombocytopenia

While thrombocytopenia (defined as platelet count below 100,000/mm^3) has *not* been observed in the completed Study C1171001, in the current study, platelet count as reported by the sponsor-identified central laboratory, with or without any accompanying signs/symptoms, will be assessed versus the threshold outlined in Appendix 2 every two weeks post randomization (on Day 1 / Visit 5) up to Visit 9 (Week 8) and monthly thereafter (Visit 10 / Week 12 and Visit 11 / Week 16).

- In subjects ***observed*** to have platelet count below the LLN at Visit 9 (Week 8), unplanned visits at Week 10 and Week 14 (*in addition* to the scheduled Visit 10 [Week 12] and Visit 11 [Week 16]) should be used to –
    - Inquire about adverse events via open-ended inquiry;
    - *And* collect blood sample to permit safety-related laboratory assessments included hematology panel assessment by the sponsor-identified central laboratory.

Subjects observed to have a decline over time ultimately reaching the threshold of < 75,000/mm^3 in platelet count as outlined in Appendix 2 should be withdrawn with the primary reason for withdrawal captured as an adverse event of thrombocytopenia.

7.9.2. Potential Cases of Hypertriglyceridemia

In Study C1171001, gradual increases in fasting and post-prandial serum triglycerides have been observed – refer to Section 1.2.2. Hence, in the current study, fasting serum triglycerides as reported by the sponsor-identified central laboratory, with or without any accompanying signs/symptoms, will be assessed versus the threshold outlined in Appendix 3.

Individual subjects with consistently increasing fasting serum triglycerides over time ultimately reaching the threshold of ≥ 800 mg/dL (9 mmol/L) as outlined in Appendix 3 should be withdrawn with have the primary reason for withdrawal captured as an adverse event of hypertriglyceridemia.

7.9.3. Management of Glycemic Control in Patients with T2DM

Events of hypoglycemia (HAE) and fasting plasma glucose will be routinely monitored during participation in the study, in subjects with T2DM. At Visit 3 / Run-in, subjects will be given the following instructions/guidance to be complied with for the duration of participation in the study:

- Subjects will be provided the following (as provided to the sites by the sponsor).
- To aide in management of their T2DM the following (or similar) items:

- Home glucose monitoring supplies including a sponsor-provided, plasma-referenced, home glucometer and accompanying supplies;
- Diary to be completed at home for any symptomatic episodes and brought to **each** visit to the site along with the glucometers;
- Instructions on use and documentation of home glucose monitoring results;
- Low blood glucose information sheet which delineates the symptoms of hypoglycemia along with treatment options ***and*** instructions to share with their family member(s) / roommate(s) to help in their management of hypoglycemia;
- Recommended frequency for routine home glucose monitoring will be determined for each subject by the investigator, however, a frequency of **at least once per day** should be *emphasized*;
- Home glucose monitoring must be undertaken in the event subjects' experience symptoms of HAE, whenever possible; these symptoms must be captured in the diary.

- Routine safety monitoring (eg, liver function monitoring) for any background medications, including anti-diabetic agents, is the responsibility of the investigator and should be consistent with guidelines found in prescribing information for the respective agents;
- ***Wherever possible***, new agents for glycemic control *or* change in doses of concomitant agents for glycemic control (refer to Section 5.8.1) should be avoided during participation in the study unless protocol-specific criteria for inadequate glycemic control are met – refer to Section 7.9.3.1.

7.9.3.1. Protocol-Specific Criteria for Inadequate Glycemic Control

Investigators must review the diary completed by the subjects and the readings stored in the glucometer devices at **each** visit after the Run-in Visit. Based on this information, as well as review of the results reported by the central laboratory, an assessment of any symptomatic and asymptomatic occurrence of hypo- or hyper- glycemia must be undertaken.

7.9.3.1.1. Management of Hypoglycemia (HAE)

Any episode of HAE must be captured with sufficient detail as outlined in Appendix 4. For definition of HAE and severity categorization, refer to Section 7.9.3.1.2, below.

7.9.3.1.2. Definition and Severity Categorization of HAE

Based on review of the subject completed diary at **each** visit to the site as well as results reported by the central laboratory, the investigator must assess the glucose values as well as any symptoms documented.

HAE is defined as **one** of the following:

1. **Asymptomatic hypoglycemia:** An event *not* accompanied by typical symptoms of HAE but a plasma glucose reading of ≤ 49 mg/dL (2.7 mmol/L) using sponsor-provided, plasma-referenced, home glucometers (or sponsor-identified central laboratory);

2. **Documented symptomatic hypoglycemia:** An event during which typical symptoms of HAE are accompanied *with* a glucose reading of < 70 mg/dL (3.9 mmol/L) using sponsor-provided, plasma-referenced, home glucometers (or sponsor-identified central laboratory) ***and*** the clinical picture includes prompt resolution with food intake, subcutaneous glucagon, or intravenous glucose;

3. **Probable symptomatic hypoglycemia:** An event during which symptoms of HAE are *not* accompanied by a plasma glucose determination, but was presumably caused by a plasma glucose concentration of < 70 mg/dL (3.9 mmol/L) using sponsor-provided, plasma-referenced, home glucometers (or sponsor-identified central laboratory) ***and*** the clinical picture includes prompt resolution with food intake, subcutaneous glucagon, or intravenous glucose.

***NOTE*: Pseudo-hypoglycemia:** An event during which symptoms consistent with HAE are reported along *with* a plasma glucose reading ≥ 70 mg/dL (3.9 mmol/L) using sponsor-provided, plasma-referenced, home glucometers (or sponsor-identified central laboratory) *may* be reported as an adverse event (AE) of HAE but will **not** be considered as having met the protocol definition of HAE offered above.

Each episode of HAE must be characterized with respect to severity. In order to characterize the event as 'severe', **all three (3)** criteria below must be met:

1. The subject was unable to treat him/herself. Neurologic impairment, and not the age of the subject, is the explanation for why the subject could not treat him/herself and required the assistance of another person.

2. The subject exhibited at least **one** of the following neurological symptoms:

   - Memory Loss;
   - Confusion;
   - Uncontrolled behavior;
   - Irrational behavior;
   - Unusual difficulty in awakening;
   - Suspected seizure;

- Seizure;
- Loss of consciousness.

3. Either:

- **If** blood glucose was measured and was ≤ 49 mg/dL (2.7 mmol/L) using sponsor-provided, plasma-referenced, home glucometers (or central laboratory) **or**
- **If** blood glucose was **not** measured, the clinical manifestations were reversed by oral carbohydrates, subcutaneous glucagon, or intravenous glucose.

Events that do **not** meet **all** the criteria above for 'severe' HAE are characterized as 'mild' or 'moderate' in severity.

7.9.3.1.3. Management of Hyperglycemia

Hyperglycemia is defined as a glucose reading of > 270 mg/dL (15 mmol/L) using sponsor-provided, plasma-referenced, home glucometers (or sponsor-identified central laboratory).

Subjects noted to have a **fasting** plasma glucose value (during home glucose monitoring using sponsor-provided, plasma-referenced, home glucometers **or** as reported by the sponsor-identified central laboratory) meeting the above definition of hyperglycemia must be instructed to repeat the measurement the next day (following **≥ 8-hour fast** except water). If the second measurement also meets the above definition, subjects must be asked to return to the site in the next **5 ± 2 days,** following **≥ 8-hour fast** except water, and have blood collected for fasting plasma glucose (and shipped to sponsor-identified central laboratory for analysis).

If the results from the sponsor-identified central laboratory confirm the readings using sponsor-provided, plasma-referenced, home glucometers **or** a previous result reported by the sponsor-identified central laboratory, the subject must be withdrawn from the study – with the reason for withdrawal captured as an adverse event of hyperglycemia.

8. ADVERSE EVENT REPORTING

In this study, the active collection period for adverse events (serious **and** non-serious) will start at the time individual subjects' consent to pre-Qualification and continue through to a **minimum of 28 calendar days** after the last dose of the investigational product. Treatment-emergent adverse events will be defined as those noted to have an onset *or* increase in severity after first dose of double-blinded investigational product (at Visit 5 / Day 1).

8.1. Requirements

The table below summarizes the requirements for recording safety events on the CRF and for reporting safety events on the Clinical Trial (CT) Serious Adverse Event (SAE) Report Form to Pfizer Safety. These requirements are delineated for 3 types of events: (1) SAEs; (2) non-serious adverse events (AEs); and (3) exposure to the investigational product under study during pregnancy or breastfeeding, and occupational exposure.

Safety Event	**Recorded on the CRF**	**Reported on the CT SAE Report Form to Pfizer Safety Within 24 Hours of Awareness**
SAE	All	All
Non-serious AE	All	None
Exposure to the investigational product under study during pregnancy or breastfeeding, and occupational exposure	All (regardless of whether associated with an AE), **except occupational exposure**	Exposure during pregnancy, exposure via breastfeeding, occupational exposure (regardless of whether associated with an AE)

All observed or volunteered events regardless of treatment group or suspected causal relationship to the investigational product(s) will be reported as described in the following paragraphs.

Events listed in the table above that require reporting to Pfizer Safety on the CT SAE Report Form within 24 hours of awareness of the event by the investigator **are to be reported regardless of whether the event is determined by the investigator to be related to an investigational product under study**. In particular, if the SAE is fatal or life-threatening, notification to Pfizer Safety must be made immediately, irrespective of the extent of available event information. This time frame also applies to additional new (follow-up) information on previously forwarded reports. In the rare situation that the investigator does not become immediately aware of the occurrence of an event, the investigator must report the event within 24 hours after learning of it and document the time of his/her first awareness of the event.

For each event, the investigator must pursue and obtain adequate information both to determine the outcome and to assess whether it meets the criteria for classification as an SAE (see Section 8.2.3 below). In addition, the investigator may be requested by Pfizer Safety to obtain specific follow-up information in an expedited fashion. This information is more detailed than that recorded on the CRF. In general, this will include a description of the event in sufficient detail to allow for a complete medical assessment of the case and independent determination of possible causality. Any information relevant to the event, such as concomitant medications and illnesses, must be provided. In the case of a subject death, a summary of available autopsy findings must be submitted as soon as possible to Pfizer Safety. Any pertinent additional information must be reported on the CT SAE Report Form; additional source documents (eg, medical records, CRF, laboratory data) are to be sent to Pfizer Safety **ONLY** upon request.

As part of ongoing safety reviews conducted by the sponsor, any non-serious AE that is determined by the sponsor to be serious will be reported by the sponsor as an SAE. To assist in the determination of case seriousness, further information may be requested from the investigator to provide clarity and understanding of the event in the context of the clinical study.

8.1.1. Additional Details on Recording Adverse Events on the CRF

All events detailed in the table above will be recorded on the AE page(s) of the CRF. It should be noted that the CT SAE Report Form for reporting of SAE information is not the same as the AE page of the CRF. When the same data are collected, the forms must be completed in a consistent manner. AEs should be recorded using concise medical terminology and the same AE term should be used on both the CRF and the CT SAE Report Form for reporting of SAE information.

8.1.2. Eliciting Adverse Event Information

The investigator is to record on the CRF all directly observed AEs and all AEs spontaneously reported by the study subject. In addition, each study subject will be questioned about the occurrence of AEs in a non-leading manner.

8.1.3. Withdrawal from the Study Due to Adverse Events (see also Section 6.6)

Withdrawal due to AEs should be distinguished from withdrawal due to other causes, according to the definition of AE noted below, and recorded on the CRF.

When a subject withdraws from the study because of an SAE, the SAE must be recorded on the CRF and reported, as appropriate, on the CT SAE Report Form, in accordance with Section 8.1 above.

8.1.4. Time Period for Collecting AE/SAE Information

The time period for actively eliciting and collecting AEs and SAEs ("active collection period") for each subject begins from the time the subject provides informed consent, which is obtained before the subject's participation in the study (ie, before undergoing any study-related procedure and/or receiving investigational product), through and including a **minimum of 28 calendar days** after the last administration of the investigational product.

For subjects who fail during the pre-qualification phase or after Screen 1, the active collection period ends when the subject is deemed to be **not** suitable / eligible for this study (ie, screen failure status).

The time period for actively eliciting and collecting AEs and SAEs ("active collection period") for each subject begins from the time the subject provides informed consent, which is obtained before the subject's participation in the study (ie, before undergoing any study-related procedure and/or receiving investigational product), through and including the 2nd Follow-up Visit. At the 2nd Follow-up Visit, the subject will be contacted by telephone (unless local regulations dictate on-site visit) to inquire about SAEs, including hospitalizations and newly diagnosed chronic medical conditions since previous on-site visit.

8.1.4.1. Reporting SAEs to Pfizer Safety

All SAEs occurring in a subject during the active collection period are reported to Pfizer Safety on the CT SAE Report Form.

SAEs occurring in a subject after the active collection period has ended are reported to Pfizer Safety if the investigator becomes aware of them; at a minimum, all SAEs that the investigator believes have at least a reasonable possibility of being related to investigational product must be reported to Pfizer Safety.

Follow up by the investigator continues throughout and after the active collection period and until the event or its sequelae resolve or stabilize at a level acceptable to the investigator, and Pfizer concurs with that assessment.

8.1.4.2. Recording Non-serious AEs and SAEs on the CRF

During the active collection period, both non-serious AEs and SAEs are recorded on the CRF.

Follow-up by the investigator may be required until the event or its sequelae resolve or stabilize at a level acceptable to the investigator, and Pfizer concurs with that assessment.

8.1.5. Causality Assessment

The investigator's assessment of causality must be provided for all AEs (serious and non-serious); the investigator must record the causal relationship on the CRF, and report such an assessment in accordance with the SAE reporting requirements, if applicable. An investigator's causality assessment is the determination of whether there exists a reasonable possibility that the investigational product caused or contributed to an AE; generally the facts (evidence) or arguments to suggest a causal relationship should be provided. If the investigator does not know whether or not the investigational product caused the event, then the event will be handled as "related to investigational product" for reporting purposes, as defined by the sponsor. If the investigator's causality assessment is "unknown but not related" to investigational product, this should be clearly documented on study records.

In addition, if the investigator determines that an SAE is associated with study procedures, the investigator must record this causal relationship in the source documents and CRF, and report such an assessment in the dedicated section of the CT SAE Report Form and in accordance with the SAE reporting requirements.

8.1.6. Sponsor's Reporting Requirements to Regulatory Authorities

AE reporting, including suspected unexpected serious adverse reactions, will be carried out in accordance with applicable local regulations.

8.2. Definitions

8.2.1. Adverse Events

An AE is any untoward medical occurrence in a study subject administered a product or medical device; the event need not necessarily have a causal relationship with the treatment or usage. Examples of AEs include, but are not limited to:

- Abnormal test findings;
- Clinically significant signs and symptoms;
- Changes in physical examination findings;
- Hypersensitivity;
- Progression/worsening of underlying disease;
- Drug abuse;
- Drug dependency.

Additionally, AEs may include signs and symptoms resulting from:

- Drug overdose;
- Drug withdrawal;
- Drug misuse;
- Drug interactions;
- Extravasation;
- Exposure during pregnancy (EDP);
- Exposure via breastfeeding;
- Medication error;
- Occupational exposure.

8.2.2. Abnormal Test Findings

Abnormal objective test findings should be recorded as AEs when any of the following conditions are met:

- Test result is associated with accompanying symptoms; and/or

- Test result requires additional diagnostic testing or medical/surgical intervention; and/or

- Test result leads to a change in study dosing (outside of any protocol-specified dose adjustments) or discontinuation from the study, significant additional concomitant drug treatment, or other therapy; and/or

- Test result is considered to be an AE by the investigator or sponsor.

Merely repeating an abnormal test, in the absence of any of the above conditions, does not constitute an AE. Any abnormal test result that is determined to be an error does not require recording as an AE.

8.2.3. Serious Adverse Events

A serious adverse event is any untoward medical occurrence at any dose that:

- Results in death;

- Is life-threatening (immediate risk of death);

- Requires inpatient hospitalization or prolongation of existing hospitalization;

- Results in persistent or significant disability/incapacity (substantial disruption of the ability to conduct normal life functions);

- Results in congenital anomaly/birth defect.

Or that is considered to be:

- An important medical event.

Medical and scientific judgment is exercised in determining whether an event is an important medical event. An important medical event may not be immediately life-threatening and/or result in death or hospitalization. However, if it is determined that the event may jeopardize the subject, **or** may require intervention to prevent one of the other AE outcomes, the important medical event should be reported as serious.

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

8.2.4. Hospitalization

Hospitalization is defined as any initial admission (even less than 24 hours) in a hospital or equivalent healthcare facility, or any prolongation of an existing admission. Admission also includes transfer within the hospital to an acute/intensive care unit (eg, from the psychiatric wing to a medical floor, medical floor to a coronary care unit, or neurological floor to a

tuberculosis unit). An emergency room visit does not necessarily constitute a hospitalization; however, the event leading to the emergency room visit is assessed for medical importance.

Hospitalization does not include the following:

- Rehabilitation facilities;
- Hospice facilities;
- Respite care (eg, caregiver relief);
- Skilled nursing facilities;
- Nursing homes;
- Same-day surgeries (as outpatient/same-day/ambulatory procedures).

Hospitalization or prolongation of hospitalization in the absence of a precipitating clinical AE is not in itself an SAE. Examples include:

- Admission for treatment of a preexisting condition not associated with the development of a new AE or with a worsening of the preexisting condition (eg, for workup of a persistent pretreatment laboratory abnormality);
- Social admission (eg, subject has no place to sleep);
- Administrative admission (eg, for yearly physical examination);
- Protocol-specified admission during a study (eg, for a procedure required by the study protocol);
- Optional admission not associated with a precipitating clinical AE (eg, for elective cosmetic surgery);
- Hospitalization for observation without a medical AE;
- Preplanned treatments or surgical procedures. These should be noted in the baseline documentation for the entire protocol and/or for the individual subject;

Diagnostic and therapeutic noninvasive and invasive procedures, such as surgery, should not be reported as SAEs. However, the medical condition for which the procedure was performed should be reported if it meets the definition of an SAE. For example, an acute appendicitis that begins during the reporting period should be reported if the SAE requirements are met, and the resulting appendectomy should be recorded as treatment of the AE.

8.3. Severity Assessment

If required on the AE page of the CRF, the investigator will use the adjectives MILD, MODERATE, or SEVERE to describe the maximum intensity of the AE. For purposes of consistency, these intensity grades are defined as follows:	
MILD	Does not interfere with subject's usual function.
MODERATE	Interferes to some extent with subject's usual function.
SEVERE	Interferes significantly with subject's usual function.

Note the distinction between the severity and the seriousness of an AE. A severe event is not necessarily an SAE. For example, a headache may be severe (interferes significantly with the subject's usual function) but would not be classified as serious unless it met one of the criteria for SAEs, listed above.

8.4. Special Situations

8.4.1. Protocol-Specific Serious Adverse Events

There are no protocol-specified SAEs in this study. All SAEs will be reported to Pfizer Safety by the investigator as described in previous sections, and will be handled as SAEs in the safety database.

8.4.2. Potential Cases of Drug-Induced Liver Injury

Humans exposed to a drug who show no sign of liver injury (as determined by elevations in transaminases) are termed “tolerators,” while those who show transient liver injury, but adapt are termed “adaptors.” In some subjects, transaminase elevations are a harbinger of a more serious potential outcome. These subjects fail to adapt and therefore are "susceptible" to progressive and serious liver injury, commonly referred to as drug-induced liver injury (DILI). Subjects who experience a transaminase elevation above 3 times the upper limit of normal (xULN) should be monitored more frequently to determine if they are an “adaptor” or are “susceptible.”

In the majority of DILI cases, elevations in aspartate aminotransferase (AST) and/or alanine aminotransferase (ALT) precede total bilirubin (TBili) elevations (>2x ULN) by several days or weeks. The increase in TBili typically occurs while AST/ALT is/are still elevated above 3x ULN (ie, AST/ALT and TBili values will be elevated within the same lab sample). In rare instances, by the time TBili elevations are detected, AST/ALT values might have decreased. This occurrence is still regarded as a potential DILI. Therefore, abnormal elevations in either AST OR ALT in addition to TBili that meet the criteria outlined below are considered potential DILI (assessed per Hy’s law criteria) cases and should always be considered important medical events, even before all other possible causes of liver injury have been excluded.

The threshold of laboratory abnormalities for a potential DILI case depends on the subject's individual baseline values and underlying conditions. Subjects who present with the following laboratory abnormalities should be evaluated further as potential DILI (Hy's law) cases to definitively determine the etiology of the abnormal laboratory values:

- Subjects with AST/ALT *and* TBili baseline values within the normal range who subsequently present with AST **or** ALT values >3x ULN **and** a TBili value >2x ULN with no evidence of hemolysis and an alkaline phosphatase value <2 xULN *or* not available;

- For subjects with baseline AST **or** ALT **or** TBili values above the ULN, the following threshold values are used in the definition mentioned above, as needed, depending on which values are above the ULN at baseline:

  - Preexisting AST or ALT baseline values above the normal range: AST or ALT values > 2 times the baseline values **and** > 3x ULN **or** >8x ULN (whichever is smaller).

  - Preexisting values of TBili above the normal range: TBili level increased from baseline value by an amount of at least > ULN **or** if the value reaches >3x ULN (whichever is smaller).

Rises in AST/ALT and TBili separated by more than a few weeks should be assessed individually based on clinical judgment; any case where uncertainty remains as to whether it represents a potential Hy's law case should be reviewed with the sponsor.

The subject should return to the investigator site and be evaluated as soon as possible, preferably **within 48 hours** from awareness of the abnormal results. This evaluation should include laboratory tests, detailed history, and physical assessment.

In addition to repeating measurements of AST and ALT and TBili, laboratory tests should include albumin, creatine kinase (CK), direct and indirect bilirubin, gamma-glutamyl transferase (GGT), prothrombin time (PT)/international normalized ratio (INR), total bile acids, alkaline phosphatase and acetaminophen drug and/or protein adduct levels – refer to Table 4. Consideration should also be given to drawing a separate tube of clotted blood and an anticoagulated tube of blood for further testing, as needed, for further contemporaneous analyses at the time of the recognized initial abnormalities to determine etiology. A detailed history, including relevant information, such as review of ethanol, acetaminophen (either by itself or as a coformulated product in prescription or over-the-counter medications), recreational drug, supplement (herbal) use and consumption, family history, sexual history, travel history, history of contact with a jaundiced person, surgery, blood transfusion, history of liver or allergic disease, and potential occupational exposure to chemicals, should be collected. Further testing for acute hepatitis A, B, C, D, and E infection and liver imaging (eg, biliary tract) may be warranted.

All cases demonstrated on repeat testing as meeting the laboratory criteria of AST/ALT and TBili elevation defined above should be considered potential DILI (Hy's law) cases if no other reason for the liver function test (LFT) abnormalities has yet been found. **Such potential DILI (Hy's law) cases are to be reported as SAEs, irrespective of availability of all the results of the investigations performed to determine etiology of the LFT abnormalities.**

A potential DILI (Hy's law) case becomes a confirmed case only after all results of reasonable investigations have been received and have excluded an alternative etiology.

8.4.3. Exposure to the Investigational Product During Pregnancy or Breastfeeding, and Occupational Exposure

Exposure to the investigational product under study during pregnancy or breastfeeding and occupational exposure are reportable to Pfizer Safety **within 24 hours** of investigator awareness.

8.4.3.1. Exposure During Pregnancy

For both unapproved/unlicensed products and for marketed products, an exposure during pregnancy (EDP) occurs if:

- A female becomes, or is found to be, pregnant either while receiving or having been exposed (eg, because of treatment or environmental exposure) to the investigational product; or the female becomes or is found to be pregnant after discontinuing and/or being exposed to the investigational product;
    - An example of environmental exposure would be a case involving direct contact with a Pfizer product in a pregnant woman (eg, a nurse reports that she is pregnant and has been exposed to chemotherapeutic products).
- A male has been exposed (eg, because of treatment or environmental exposure) to the investigational product prior to or around the time of conception and/or is exposed during his partner's pregnancy.

If a subject or subject's partner becomes or is found to be pregnant during the subject's treatment with the investigational product, the investigator must report this information to Pfizer Safety on the CT SAE Report Form and an EDP supplemental form, regardless of whether an SAE has occurred. In addition, the investigator must submit information regarding environmental exposure to a Pfizer product in a pregnant woman (eg, a subject reports that she is pregnant and has been exposed to a cytotoxic product by inhalation or spillage) to Pfizer Safety using the EDP supplemental form. This must be done irrespective of whether an AE has occurred and **within 24 hours** of awareness of the exposure. The information submitted should include the anticipated date of delivery (see below for information related to termination of pregnancy).

Follow-up is conducted to obtain general information on the pregnancy and its outcome for all EDP reports with an unknown outcome. The investigator will follow the pregnancy until completion (or until pregnancy termination) and notify Pfizer Safety of the outcome as a follow-up to the initial EDP supplemental form. In the case of a live birth, the structural integrity of the neonate can be assessed at the time of birth. In the event of a termination, the reason(s) for termination should be specified and, if clinically possible, the structural integrity of the terminated fetus should be assessed by gross visual inspection (unless pre-procedure test findings are conclusive for a congenital anomaly and the findings are reported).

If the outcome of the pregnancy meets the criteria for an SAE (ie, ectopic pregnancy, spontaneous abortion, intrauterine fetal demise, neonatal death, or congenital anomaly [in a live-born baby, a terminated fetus, an intrauterine fetal demise, or a neonatal death]), the investigator should follow the procedures for reporting SAEs.

Additional information about pregnancy outcomes that are reported to Pfizer Safety as SAEs follows:

- Spontaneous abortion includes miscarriage and missed abortion;
- Neonatal deaths that occur within 1 month of birth should be reported, without regard to causality, as SAEs. In addition, infant deaths after 1 month should be reported as SAEs when the investigator assesses the infant death as related or possibly related to exposure to the investigational product.

Additional information regarding the EDP may be requested by the sponsor. Further follow-up of birth outcomes will be handled on a case-by-case basis (eg, follow-up on preterm infants to identify developmental delays). In the case of paternal exposure, the investigator will provide the subject with the Pregnant Partner Release of Information Form to deliver to his partner. The investigator must document in the source documents that the subject was given the Pregnant Partner Release of Information Form to provide to his partner.

8.4.3.2. Exposure During Breastfeeding

Scenarios of exposure during breastfeeding must be reported, irrespective of the presence of an associated SAE, to Pfizer Safety **within 24 hours** of the investigator's awareness, using the CT SAE Report Form. An exposure during breastfeeding report is not created when a Pfizer drug specifically approved for use in breastfeeding women (eg, vitamins) is administered in accord with authorized use. However, if the infant experiences an SAE associated with such a drug's administration, the SAE is reported together with the exposure during breastfeeding.

8.4.3.3. Occupational Exposure

An occupational exposure occurs when, during the performance of job duties, a person (whether a healthcare professional or otherwise) gets in unplanned direct contact with the product, which may or may not lead to the occurrence of an AE.

An occupational exposure is reported to Pfizer Safety **within 24 hours** of the investigator's awareness, using the CT SAE Report Form, regardless of whether there is an associated SAE. Since the information does not pertain to a subject enrolled in the study, the information is not recorded on a CRF; however, a copy of the completed CT SAE Report Form is maintained in the investigator site file.

8.4.4. Medication Errors

Other exposures to the investigational product under study may occur in clinical trial settings, such as medication errors.

Safety Event	Recorded on the CRF	Reported on the CT SAE Report Form to Pfizer Safety Within 24 Hours of Awareness
Medication errors	All (regardless of whether associated with an AE)	Only if associated with an SAE

Medication errors may result from the administration or consumption of the investigational product by the wrong subject, or at the wrong time, or at the wrong dosage strength.

Medication errors include:

- Medication errors involving subject exposure to the investigational product;
- Potential medication errors or uses outside of what is foreseen in the protocol that do or do not involve the participating subject.

Such medication errors occurring to a study participant are to be captured on the medication error page of the CRF, which is a specific version of the AE page.

In the event of a medication dosing error, the sponsor should be notified immediately.

Whether or not the medication error is accompanied by an AE, as determined by the investigator, the medication error is recorded on the medication error page of the CRF and, if applicable, any associated AE(s), serious and non-serious, are recorded on an AE page of the CRF.

Medication errors should be reported to Pfizer Safety **within 24 hours** on a CT SAE Report Form **only when associated with an SAE**.

9. DATA ANALYSIS/STATISTICAL METHODS

Detailed methodology for summary and statistical analyses of the data collected in this study is outlined here and further detailed in a statistical analysis plan (SAP), which will be maintained by the sponsor. The SAP may modify what is outlined in the protocol where appropriate; however, any major modifications of the primary endpoint definitions or their analyses will also be reflected in a protocol amendment.

9.1. Sample Size Determination

The sample size for this study was based on interest to assess a dose-response for effect of PF-05221304 on percent change from baseline in liver fat (entire study population as well as separately in each sub-population in 1st tier stratification) and percent change from baseline in ALT in the 1st tier stratification sub-population of diagnosed/presumed NASH.

The proposed sample size for this study is approximately 360 subjects randomized (72 subjects per arm) to ensure at least 300 subjects (60 subjects per arm) complete the study.

The sample size of a minimum of 60 completers per arm, was calculated to yield ≥ 80% power to detect a placebo-adjusted, percent change from baseline of ≥ 30% in liver fat (in entire study population *and* in n=30/arm in each sub-population of the 1st tier stratification), using a 1-sided, t-test at level 10%. The calculation assumes that the standard deviation for liver fat is 7.8% (between-subject) and 6.2% (within-subject).

For the secondary endpoint of ALT in the 1st tier stratification sub-population of diagnosed/presumed NASH, the sample size of a minimum of 30 completers per arm was calculated to yield ≥ 80% power to detect a placebo-adjusted, percent change from baseline in ALT of ≥ 35%, using a 1-sided, t-test at level 10%. The calculation assumes that the standard deviation for the parameter of placebo-adjusted, percent change from baseline in ALT is 55% (between-subject).

Overall, via the planned interim analysis (refer to Section 9.5), if the observed standard deviation for the liver fat (MRI-PDFF) and ALT data, based on unblinded review, is greater than the assumptions above, the sample size for this study *may* be adjusted upward, at sponsor discretion, to a maximum of 500 subjects randomized (ie, 100 subjects per arm).

9.2. Efficacy Analysis

9.2.1. Analysis of the Primary Endpoint

The primary endpoint is placebo-adjusted, percent change from baseline in liver fat (via MRI-PDFF) at Week 16, for the entire study population. A mixed model repeated measure (MMRM) analysis will be performed with treatment, time and treatment-by-time interaction as fixed effects, and subject as a random effect.

Descriptive summaries of the observed values and percent change from baseline in liver fat in each treatment group at each time point will be produced.

For statistical analyses, 80% confidence intervals will be produced for the least-square mean differences in each treatment group as compared to placebo. One sided p-values will be displayed for the comparison against placebo with no multiplicity adjustment made.

Dose-response modeling of the percent change from baseline in liver fat will be explored with total daily dose and time as a continuous variable to characterize and evaluate the dose-response relationship.

9.2.2. Analysis of Secondary Endpoint

One of the key secondary endpoint is percent change from baseline in ALT at Week 16 in the 1st tier stratification sub-population of diagnosed/presumed NASH. An MMRM analysis will be performed with treatment, time and treatment-by-time interaction as fixed effects, and subject as a random effect.

Descriptive summaries of the observed values and percent change from baseline in ALT in each treatment group at each time point will be produced.

For statistical analyses, 80% confidence intervals will be produced for the least-square mean differences in each treatment group as compared to placebo. One sided p-values will be displayed for the comparison against placebo with no multiplicity adjustment made.

Dose-response modeling of the percent change from baseline in ALT, the 1st tier stratification sub-population of diagnosed/presumed NASH, will be explored with total daily dose and time as a continuous variable to characterize and evaluate the dose-response relationship; assessment of dose-response for effect on ALT for the entire entire study population, may be considered, depending on the data.

9.3. Analysis of Tertiary Endpoints

CCI

9.3.2. Other Tertiary Endpoints

For continuous endpoints, a MMRM analysis will be performed with treatment, time and treatment-by-time interaction as fixed effects, and subject as a random effect. Two-sided p-values and 80% confidence intervals will be displayed for the least-square mean difference or ratio (for natural log-transformed endpoints) against placebo. Descriptive summaries of the observed values and change from baseline in each treatment group at each time point will also be produced.

Depending on the observed data at the end of the study and when applicable, dose-response modeling of the tertiary endpoints may be explored with total daily dose as a continuous variable to characterize and evaluate the dose-response relationship.

Descriptive summary (number and percentages) will be displayed for categorical endpoints.

9.4. Safety Analysis

Treatment-emergent AEs, as well as SAEs/deaths, clinical safety-related laboratory tests, 12-lead ECG data, as well as vital signs will be summarized descriptively as appropriate for Pfizer data standards. Statistical inference may be determined for safety measures of clinical concern. Safety data will be presented in tabular and/or graphical format and summarized descriptively, as appropriate to Pfizer standards.

Physical exam information collected during the course of the study will not be captured for inclusion into the study database, unless otherwise noted. However, any untoward findings identified on physical exams conducted after the administration of the first dose of blinded investigational product will be captured as an adverse event, if those findings meet the definition of an adverse event as defined in Section 8.

CCI [redacted]

9.5. Interim Analysis

Interim analysis (IA) will be performed **at least once** while the study is on-going. This review will include an assessment of the observed safety, and observed (versus assumed) variability for the primary endpoint (liver fat via MRI-PDFF) as well as key secondary endpoint (ALT). The 1 planned IA is proposed after **at least one-third** of the total planned randomized subjects (ie, approximately 120 subjects), complete through **at least one-third** of the total dosing duration (ie, **Visit 8 [Week 6]**) of the study. Interim analysis results may be used for conducting a sample size re-estimation, stopping a dose-level for observed safety, facilitate CCI [redacted] and internal business decisions regarding planning of future trials with PF-05221304. Before any interim analysis is instigated, the details of the objectives, decision criteria, dissemination plan, and method of maintaining the study blind as per Pfizer's standard operating procedures (SOPs) will be documented and approved in an IRC charter. In addition, the analysis details will be documented and approved in an interim analysis SAP or final SAP.

9.6. Data Monitoring Committee

This study will **not** use a data monitoring committee.

An IRC to undertake unblinded review of the safety data and unblinded review of the observed standard deviation for the liver fat (MRI-PDFF) and ALT data, is planned while the study is on-going – refer to Section 9.5.

10. QUALITY CONTROL AND QUALITY ASSURANCE

Pfizer or its agent will conduct periodic monitoring visits during study conduct to ensure that the protocol and Good Clinical Practices (GCPs) are being followed. The monitors may review source documents to confirm that the data recorded on CRFs are accurate. The investigator and institution will allow Pfizer monitors/auditors or its agents and appropriate regulatory authorities direct access to source documents to perform this verification. This verification may also occur after study completion.

During study conduct and/or after study completion, the investigator site may be subject to review by the IRB/EC, and/or to quality assurance audits performed by Pfizer, or companies working with or on behalf of Pfizer, and/or to inspection by appropriate regulatory authorities.

The investigator(s) will notify Pfizer or its agents immediately of any regulatory inspection notification in relation to the study. Furthermore, the investigator will cooperate with Pfizer or its agents to prepare the investigator site for the inspection and will allow Pfizer or its agent, whenever feasible, to be present during the inspection. The investigator site and investigator will promptly resolve any discrepancies that are identified between the study data and the subject's medical records. The investigator will promptly provide copies of the inspection findings to Pfizer or its agent. Before response submission to the regulatory authorities, the investigator will provide Pfizer or its agents with an opportunity to review and comment on responses to any such findings.

It is important that the investigator(s) and their relevant personnel are available during the monitoring visits and possible audits or inspections and that sufficient time is devoted to the process.

11. DATA HANDLING AND RECORD KEEPING

11.1. Case Report Forms/Electronic Data Record

As used in this protocol, the term CRF should be understood to refer to either a paper form or an electronic data record or both, depending on the data collection method used in this study.

A CRF is required and should be completed for each included subject. The completed original CRFs are the sole property of Pfizer and should not be made available in any form to third parties, except for authorized representatives of Pfizer or appropriate regulatory authorities, without written permission from Pfizer.

The investigator has ultimate responsibility for the collection and reporting of all clinical, safety, and laboratory data entered on the CRFs and any other data collection forms (source documents) and ensuring that they are accurate, authentic/original, attributable, complete, consistent, legible, timely (contemporaneous), enduring, and available when required. The CRFs must be signed by the investigator or by an authorized staff member to attest that the data contained on the CRFs are true. Any corrections to entries made in the CRFs or source documents must be dated, initialed, and explained (if necessary) and should not obscure the original entry.

In most cases, the source documents are the hospital or the physician subject chart. In these cases, data collected on the CRFs must match the data in those charts.

In some cases, the CRF may also serve as the source document. In these cases, a document should be available at the investigator site and at Pfizer that clearly identifies those data that will be recorded on the CRF, and for which the CRF will stand as the source document.

11.2. Record Retention

To enable evaluations and/or inspections/audits from regulatory authorities or Pfizer, the investigator agrees to keep records, including the identity of all participating subjects (sufficient information to link records, eg, CRFs and hospital records), all original signed informed consent documents, copies of all CRFs, safety reporting forms, source documents, and detailed records of treatment disposition, and adequate documentation of relevant correspondence (eg, letters, meeting minutes, and telephone call reports). The records should be retained by the investigator according to the ICH guidelines, according to local regulations, or as specified in the clinical study agreement (CSA), whichever is longer.

If the investigator becomes unable for any reason to continue to retain study records for the required period (eg, retirement, relocation), Pfizer should be prospectively notified. The study records must be transferred to a designee acceptable to Pfizer, such as another investigator, another institution, or an independent third party arranged by Pfizer.

Investigator records must be kept for **a minimum of 15 years** after completion or discontinuation of the study or for longer if required by applicable local regulations.

The investigator must obtain Pfizer's written permission before disposing of any records, even if retention requirements have been met.

12. ETHICS

12.1. Institutional Review Board/Ethics Committee

It is the responsibility of the investigator to have prospective approval of the study protocol, protocol amendments, informed consent documents, and other relevant documents, eg, recruitment advertisements, if applicable, from the IRB/EC. All correspondence with the IRB/EC should be retained in the investigator file. Copies of IRB/EC approvals should be forwarded to Pfizer.

The only circumstance in which an amendment may be initiated prior to IRB/EC approval is where the change is necessary to eliminate apparent immediate hazards to the subjects. In that event, the investigator must notify the IRB/EC and Pfizer in writing immediately after the implementation.

12.2. Ethical Conduct of the Study

The study will be conducted in accordance with the protocol, legal and regulatory requirements and the general principles set forth in the International Ethical Guidelines for Biomedical Research Involving Human Subjects (Council for International Organizations of Medical Sciences 2002), ICH Guideline for Good Clinical Practice, and the Declaration of Helsinki.

12.3. Subject Information and Consent

All parties will ensure protection of subject personal data and will not include subject names or other identifiable data in any reports, publications, or other disclosures, except where required by law.

When study data are compiled for transfer to Pfizer and other authorized parties, subject names, addresses, and other identifiable data will be replaced by numerical codes based on a numbering system provided by Pfizer in order to de-identify study subjects. The investigator site will maintain a confidential list of subjects who participated in the study, linking each subject's numerical code to his or her actual identity. In case of data transfer, Pfizer will maintain high standards of confidentiality and protection of subjects' personal data consistent with applicable privacy laws.

The informed consent documents and any subject recruitment materials must be in compliance with ICH GCP, local regulatory requirements, and legal requirements, including applicable privacy laws.

The informed consent documents used during the informed consent process and any subject recruitment materials must be reviewed and approved by Pfizer, approved by the IRB/EC before use, and available for inspection.

The investigator must ensure that each study subject is fully informed about the nature and objectives of the study and possible risks associated with participation.

The investigator, or a person designated by the investigator, will obtain written informed consent from each subject before any study-specific activity is performed. The investigator will retain the original of each subject's signed consent document.

At the end of the study, the investigational product in this study will ***not*** be provided to the subjects who participated.

12.4. Reporting of Safety Issues and Serious Breaches of the Protocol or ICH GCP

In the event of any prohibition or restriction imposed (ie, clinical hold) by an applicable regulatory authority in any area of the world, or if the investigator is aware of any new information that might influence the evaluation of the benefits and risks of the investigational product, Pfizer should be informed immediately.

In addition, the investigator will inform Pfizer immediately of any urgent safety measures taken by the investigator to protect the study subjects against any immediate hazard, and of any serious breaches of this protocol or of ICH GCP that the investigator becomes aware of.

13. DEFINITION OF END OF TRIAL

13.1. End of Trial in a Member State

End of trial in a Member State of the European Union is defined as the time at which it is deemed that a sufficient number of subjects have been recruited and completed the study as stated in the regulatory application (ie, clinical trial application [CTA]) and ethics application in the Member State. Poor recruitment (recruiting less than the anticipated number in the CTA) by a Member State is not a reason for premature termination but is considered a normal conclusion to the study in that Member State.

13.2. End of Trial in All Other Participating Countries

End of trial in all *other* participating countries is defined as last subject last visit (LSLV) reflected by completion of the 2nd Follow-up visit for the last subject randomized in the study.

14. SPONSOR DISCONTINUATION CRITERIA

Premature termination of this study may occur because of a regulatory authority decision, change in opinion of the IRB/EC, or investigational product safety problems, or at the discretion of Pfizer. In addition, Pfizer retains the right to discontinue development of **PF-05221304** at any time.

If a study is prematurely terminated, Pfizer will promptly notify the investigator. After notification, the investigator must contact all participating subjects and the hospital pharmacy (if applicable) within **28 calendar days**. As directed by Pfizer, all study materials must be collected and all CRFs completed to the greatest extent possible.

15. PUBLICATION OF STUDY RESULTS

15.1. Communication of Results by Pfizer

Pfizer fulfills its commitment to publicly disclose clinical trial results through posting the results of studies on www.clinicaltrials.gov (ClinicalTrials.gov), the European Clinical Trials Database (EudraCT), and/or www.pfizer.com, and other public registries in accordance with applicable local laws/regulations.

In all cases, study results are reported by Pfizer in an objective, accurate, balanced, and complete manner and are reported regardless of the outcome of the study or the country in which the study was conducted.

www.clinicaltrials.gov

Pfizer posts clinical trial US Basic Results on www.clinicaltrials.gov for Pfizer-sponsored interventional studies (conducted in patients) that evaluate the safety and/or efficacy of a Pfizer product, regardless of the geographical location in which the study is conducted. US Basic Results are submitted for posting within 1 year of the primary completion date (PCD) for studies in adult populations or within 6 months of the PCD for studies in pediatric populations.

PCD is defined as the date that the final subject was examined or received an intervention for the purposes of final collection of data for the primary outcome, whether the clinical study concluded according to the prespecified protocol or was terminated.

EudraCT

Pfizer posts European Union (EU) Basic Results on EudraCT for all Pfizer-sponsored interventional studies that are in scope of EU requirements. EU Basic Results are submitted for posting within 1 year of the PCD for studies in adult populations or within 6 months of the PCD for studies in pediatric populations.

www.pfizer.com

Pfizer posts Public Disclosure Synopses (clinical study report synopses in which any data that could be used to identify individual patients has been removed) on www.pfizer.com for Pfizer-sponsored interventional studies at the same time the US Basic Results document is posted to www.clinicaltrials.gov.

15.2. Publications by Investigators

Pfizer supports the exercise of academic freedom and has no objection to publication by the principal investigator (PI) of the results of the study based on information collected or generated by the PI, whether or not the results are favorable to the Pfizer product. However, to ensure against inadvertent disclosure of confidential information or unprotected inventions, the investigator will provide Pfizer an opportunity to review any proposed publication or other type of disclosure of the results of the study (collectively, "publication") before it is submitted or otherwise disclosed.

The investigator will provide any publication to Pfizer at least 30 days before it is submitted for publication or otherwise disclosed. If any patent action is required to protect intellectual property rights, the investigator agrees to delay the disclosure for a period not to exceed an additional 60 days.

The investigator will, on request, remove any previously undisclosed confidential information before disclosure, except for any study- or Pfizer product-related information necessary to the appropriate scientific presentation or understanding of the study results.

If the study is part of a multicenter study, the investigator agrees that the first publication is to be a joint publication covering all investigator sites, and that any subsequent publications by the PI will reference that primary publication. However, if a joint manuscript has not been submitted for publication within 12 months of completion or termination of the study at all participating sites, the investigator is free to publish separately, subject to the other requirements of this section.

For all publications relating to the study, the institution will comply with recognized ethical standards concerning publications and authorship, including Section II - "Ethical Considerations in the Conduct and Reporting of Research" of the Uniform Requirements for Manuscripts Submitted to Biomedical Journals, http://www.icmje.org/index.html#authorship, established by the International Committee of Medical Journal Editors.

Publication of study results is also provided for in the CSA between Pfizer and the institution. In this section entitled Publications by Investigators, the defined terms shall have the meanings given to them in the CSA.

If there is any conflict between the CSA and any attachments to it, the terms of the CSA control. If there is any conflict between this protocol and the CSA, this protocol will control as to any issue regarding treatment of study subjects, and the CSA will control as to all other issues.

16. REFERENCES

1. Saggerson D. Malonyl-CoA, a key signaling molecule in mammalian cells. Annu Rev Nutr 2008, 28, 253-72.

2. LaBrecque D, Abbas Z, Anania F, et al. Nonalcoholic fatty liver disease and nonalcoholic steatohepatitis. In: WGO Global Guidelines NAFLD/NASH (long version). Milwaukee, WI: World Gastroenterology Organisation; 2012: 29 pages.

3. Sanyal AJ, Friedman SL, McCullough AJ, et al. Challenges and opportunities in drug and biomarker development for nonalcoholic steatohepatitis: findings and recommendations from an American Association for the Study of Liver Diseases-U.S. Food and Drug Administration Joint Workshop. Hepatology 2015; 61(4):1392-405.

4. Khan F, Perumpail R, Wong R, et al. Advances in hepatocellular carcinoma: Nonalcoholic steatohepatitis-related hepatocellular carcinoma. World J Hepatol 2015; 7(18):2155-61.

5. Younossi Z, Koenig A, Abdelatif D, et al. Global epidemiology of nonalcoholic fatty liver disease – meta-analytic assessment of prevalence, incidence, and outcomes. Hepatology 2016;64(1):73-84.

6. Noureddin M, Anstee Q, Loomba R. Review article: emerging anti-fibrotic therapies in the treatment of non-alcoholic steatohepatitis. Aliment Pharmacol Ther 2016;43(11):1109-23.

7. LaBrecque D, Abbas Z, Anania F, et al. Nonalcoholic fatty liver disease and nonalcoholic steatohepatitis. In: WGO Global Guidelines NAFLD/NASH (long version). Milwaukee, WI: World Gastroenterology Organisation; 2012: 29 pages.

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

9. Savage DB, Choi CS, Samuel VT, et al. Reversal of diet-induced hepatic steatosis and hepatic insulin resistance by antisense oligonucleotide inhibitors of acetyl-CoA carboxylases 1 and 2. J Clinical Invest 2006; 116(3):817-24.

10. Schreurs M, van Dijk TH, Gerding A, et al. Soraphen, an inhibitor of the acetyl-CoA carboxylase system, improves peripheral insulin sensitivity in mice fed a high-fat diet. Diabetes Obes Metab 2009; 11(10):987-91.

11. Esler WP, Turner, SM, Amor PA, et al. Effects of acetyl-CoA carboxylase inhibition on lipid metabolism in rodents and human subjects. In: American Diabetes Association 75th Scientific Session; 5-9 Jun 2015; Boston, MA, 2015. Abstr.

12. Harriman G; Greenwood J, Bhat S, et al. ND-630, a potent and liver-directed acetyl-CoA carboxylase inhibitor, reduces hepatic steatosis and improves dyslipidemia in diet-induced obese and diabetic rat models of non-alcoholic fatty liver disease. In: Keystone Symposia; 01-06 Mar 2015; British Columbia, Canada. 2015: Abstr.

13. Berod L, Friedrich C, Nandan A, et al. De novo fatty acid synthesis controls the fate between regulatory T and T helper 17 cells. Nat Med 2014; 20(11):1327–33.

14. Rau M, Schilling AK, Meertens J, et al. Progression from nonalcoholic fatty liver to nonalcoholic steatohepatitis is marked by a higher frequency of Th17 cells in the liver and an increased Th17/resting regulatory T cell ratio in peripheral blood and in the liver. J Immunol 2016; 196(1):97–105.

15. Hernaez R, Lazo M, Bonekamp S, et al. Diagnostic accuracy and reliability of ultrasonography for the detection of fatty liver: A meta-analysis. Hepatology 2011;54:1082-90.

16. Neuschwander-Tetri BA, Clark JM, Bass NM, et al. Clinical, Laboratory and Histological Association in Adults with Nonalcoholic Fatty Liver Disease. Hepatology 2010;52:913-24.

17. Schomaker S, Warner R, Bock J, et al. Assessment of emerging biomarkers of liver injury in human subjects. Toxicol Sci 2013;132(2):276-83.

18. Chalasani N, Younossi Z, Lavine JE, et al. The diagnosis and management of non-alcoholic fatty liver disease: practice Guideline by the American Association for the Study of Liver Diseases, American College of Gastroenterology, and the American Gastroenterological Association. Hepatology. 2012; 55(6):2005-23.

19. Petit JM, Cercueil JP, Loffroy R, et al. Effect of liraglutide therapy on liver fat content in patients with inadequately controlled type 2 diabetes. The Lira-NAFLD study. J Clin Endocrinol Metab 2017; doi:10.1210/jc.2016-2775.

Appendix 1. Abbreviations

This following is a list of the abbreviations that may be used in this document.

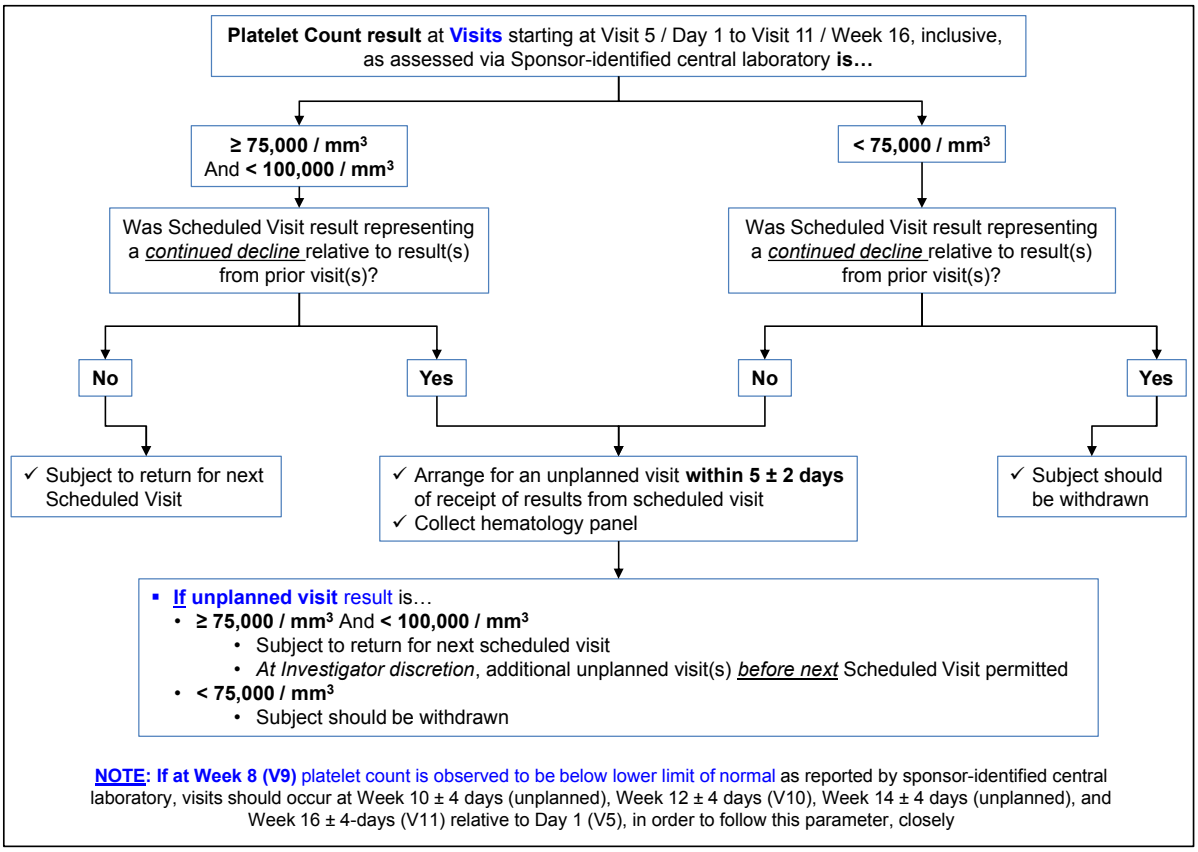
Abbreviation	Term
%CDT	Percent Carbohydrate deficient transferrin relative to total transferrin
β-hCG	Beta-human chorionic gonadotropin
A1AT	Alpha-1-antitrypsin
Abs	absolute
ACC	acetyl-CoA carboxylase
AE	adverse event
ALT / SGPT	alanine aminotransferase / serum glutamic pyruvic transaminase
AST / SGOT	aspartate aminotransferase / serum glutamic oxalo-acetic transaminase
ATP	adenosine triphosphate
AUC	area under the curve (used when referring to PK)
AUC_{24}	area under the concentration-time curve from time 0 to 24 hours
AUC_{inf}	area under the concentration-time curve from time 0 to infinity
AUC_{last}	area under the concentration-time curve from time 0 to the time of the last quantifiable concentration
AUC_{tau}	area under the concentration-time curve from time 0 to time t
AUDIT	Alcohol use disorders identification test
AUEC	Area under effect curve (used when referring to drug response)
CCI	
BID	bis in die
BLQ	below the limit of quantification
BMI	body mass index
BP	blood pressure
BUN	blood urea nitrogen
CAP™	Controlled attenuation parameter
CK	creatine kinase
CK18-M30	cytokeratin-18-M30 sub-species
CK18-M65	cytokeratin-18-M65 sub-species
C_{max}	peak or maximum observed concentration
CO_2	carbon dioxide (bicarbonate)
CPT-1	carnitine-palmitoyl transferase 1
CSA	clinical study agreement
CSR	clinical study report
CT	clinical trial
CTA	clinical trial application
CYP3A	Cytochrome P-450 3A
CCI	
D_2O / $^2H^2O$	deuterium oxide / deuterated water
dB/m	decibels per meter
DBP	diastolic blood pressure
DCT	data collection tool
DDI	Drug-drug interaction
DILI	drug-induced liver injury
DMC	data monitoring committee
DNA	deoxyribonucleic acid
DNL	de novo lipogenesis
DPP-IVi	Dipeptidyl peptidase-IV inhibitors
DU	Dispensing Unit
EC	ethics committee
ECG	electrocardiogram
e-CRF	Electronic case report form
EDP	exposure during pregnancy
EDTA	edetic acid (ethylenediaminetetraacetic acid)

Abbreviation	Term
CCI	
eGFR	Estimated glomerular filtration rate
EIU	exposure in utero
ET	Early Termination
EU	European Union
EudraCT	European Clinical Trials Database
FA	flip angle
FDA	Food and Drug Administration
FFA	free fatty acid
CCI	
FIH	first-in-human
FPG	Fasting plasma glucose
FPI	Fasting plasma insulin
FSH	follicle-stimulating hormone
FU	Follow-up
GCP	Good Clinical Practice
GGT	Gamma-glutamyl transferase
GLDH	Glutamate dehydrogenase
GLP	Good Laboratory Practice
GLP-1r	glucagon-like peptide-1 receptor
HAE	Hypoglycemia
HbA1C	Glycated hemoglobin
HBsAg	hepatitis B surface antigen
HBV	Hepatitis B virus
HCC	Hepatic Cellular Carcinoma
HCV	Hepatitis C virus
HCV RNA	Hepatitis C virus genetic material (ie, Ribonucleic acid)
HCVAb	hepatitis C antibody
HDL-C	high density lipoprotein cholesterol
HIV	human immunodeficiency virus
HOMA-IR	Homeostatic model assessment of insulin resistance
hr	hour
CCI	
ht	height
IA	Interim Analysis
IB	Investigator's brochure
ICH	International Conference on Harmonisation
ID	identification
IND	investigational new drug application
INR	international normalized ratio
IOP	in- and out-of-phase
IP	investigational product
IRB	institutional review board
IRC	internal review committee
IRT	interactive response technology
IUD	intrauterine device
IWR	interactive web response
CCI	
kPa	kilopascals
LDL-C	low density lipoprotein-cholesterol
LFT	liver function test
LLN	lower limit of normal
LSLV	last subject last visit
LSM	Liver stiffness measure
CCI	
MCH	mean corpuscular hemoglobin
MCHC	mean corpuscular hemoglobin concentration

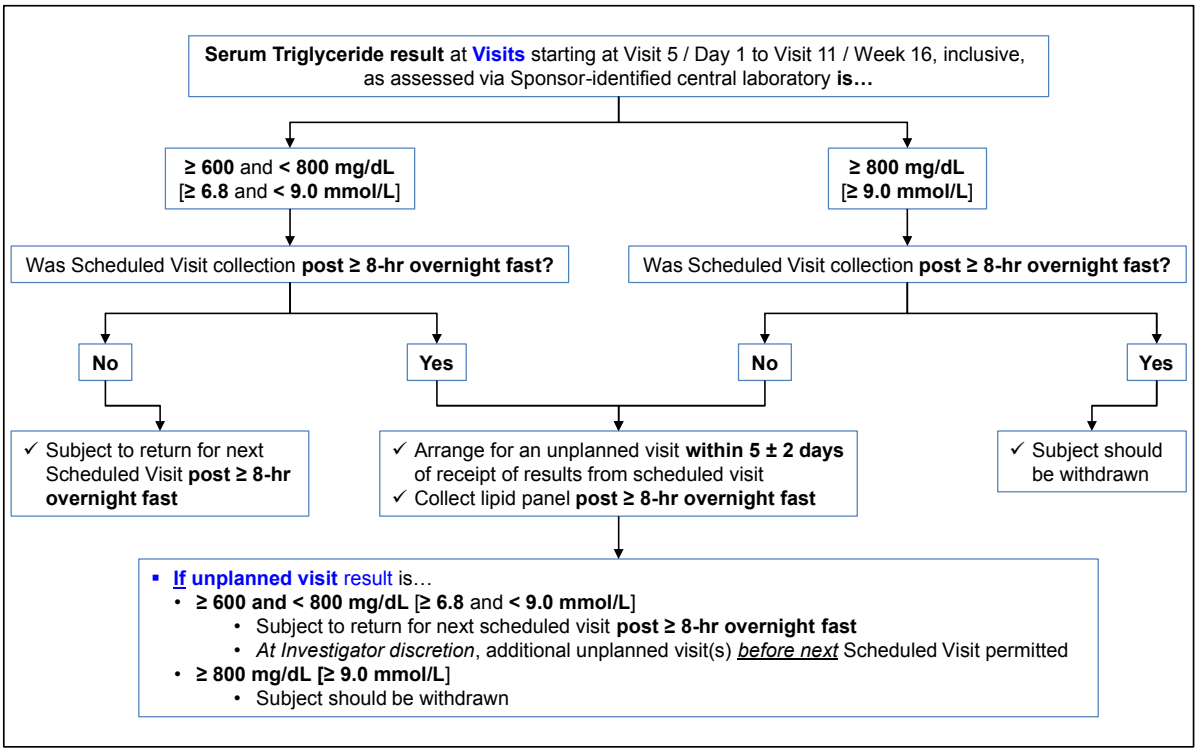
Abbreviation	Term
MCV	mean corpuscular volume
MDR1	multi-drug resistance protein-1
MDRD	Modification of Diet in Renal Disease
MEC	molar extinction coefficient
MedDRA	medical Dictionary for Regulatory Activities
MELD	model of end-stage liver disease
min	minute
MMRM	Mixed model repeated measure
MRI-PDFF	Magnetic resonance imaging using proton density fat fraction acquisition
MRS	Magnetic resonance spectroscopy
MSDS	material safety data sheets
MTD	Maximum tolerated dose
N/A	not applicable
NAFLD	nonalcoholic fatty liver disease
NASH	Nonalcoholic steatohepatitis
CCI	
NGSP	National Glycohemoglobin Standardization Program
NOAEL	no observed adverse effect level
NOEL	no observed effect level
NSAIDS	Non-steroidal anti-inflammatory medications
NYHA	New York Heart Association
CCI	
OATP	organic anion-transporting polypeptide
CCI	
OEB5	occupational exposure banding 5
PBPK	physiologically based pharmacokinetic
PCD	primary completion date
PCSK9	proprotein convertase subtilisin/kexin type 9
PD	Pharmacodynamics(s)
PE	physical exam
CCI	
PI	Principal investigator
PK	pharmacokinetics
CCI	
PNPLA3	Patatin like phospholipase domain containing protein 3
pre-Q	Pre-qualification visit
ProC3	N-terminal pro-peptide of type III procollagen
ProC6	C-terminal fragment of α3 chain of procollagen type VI (also known as Endotrophin)
PT	prothrombin time
PTR	peak to trough ratio
Q12H	Every 12 hours (eg, dosing)
QC	quality control
QD	quaque die
QTc	corrected QT
RBC	red blood cell
RNA	ribonucleic acid
SAE	serious adverse event
SAP	statistical analysis plan
SBP	systolic blood pressure
SC	subcutaneous
SCr	serum creatinine
SEM	standard error of mean
SOP	standard operating procedure
SRSD	single reference safety document
SSID	Study-specific Subject identification
SUSAR	suspected unexpected serious adverse reaction

Abbreviation	Term
$T_{1/2}$	terminal half-life
T2DM	type 2 diabetes mellitus
TBili	total bilirubin
TEAE	Treatment-emergent adverse event
TG	triglycerides
Th17	T helper 17
THC	tetrahydrocannabinol
T_{max}	time to reach maximum concentration
Treg	regulatory T
TZDs	thiazolidinediones
CCI	
ULN	upper limit of normal
US	United States
V	Visit
VCTE	Vibration-Controlled Transient Elastography
VLDL	very low density lipoprotein
V_z/F	apparent oral volume of distribution
w/v	weight per volume
WBC	white blood cell
WOCBP	Women of childbearing potential
wt	Weight

Appendix 2. Guidance to Investigators – Management of Individual Subjects With Decrease in Platelet Count



Appendix 3. Guidance to Investigators – Management of Individual Subjects With Elevation in Fasting Serum Triglycerides



Appendix 4. Guidance to Investigators – Capturing Details for Episodes of Hypoglycemia in Patients with T2DM

The following checklist must be **completed by the site staff** during each outpatient visit to the site based on responses provided by the subject and where applicable, the adequate medical care provided to treat episodes of hypoglycemia.

Did the subject have any symptoms of hypoglycemia since their last visit to the site?

□ YES □ NO

If yes, did the subject require assistance of another person to actively administer treatment for the event? □ YES □ NO

Did the subject exhibit any of the CNS symptoms of severe hypoglycemia? These include: memory loss, confusion, uncontrollable behavior, irrational behavior, unusual difficulty in awakening, suspected seizure, seizure, loss of consciousness. □ YES □ NO

Was blood glucose measured? □ NO □ YES

If yes, what was the result? ____ □ mmol/L □ mg/dL; **and** did the result meet threshold for severe hypoglycemia? □ YES □ NO

Severe hypoglycemia is defined as subject was unable to treat him/her self AND subject exhibited CNS symptoms of severe hypoglycemia mentioned above AND *either* blood glucose when measured was ≤ 49 mg/dL (or 2.7 mmol/L) *or* blood glucose was not measured by clinical manifestation reversed by oral carbohydrates, subcutaneous glucagon or intravenous glucose.

If no, did symptoms resolve when treated with carbohydrates, or glucagon? □ YES □ NO

Did the event awaken the subject while asleep for the night? □ YES □ NO

What time did the hypoglycemic event start? What time did it resolve?

When was the last meal (of ≥ 400 kcals) consumed? ____ Date; ____ Time

When was the last dose of concomitant medication for glycemic control taken?

____ Date; ____ Time

Were any actions taken? □ YES □ NO

If yes, dosing with investigational product – choose only **one** and enter details in concomitant treatment section.

□ No action taken □ Trial drug stopped temporarily □ Trial drug stopped permanently

Concomitant treatment dosing – choose only **one** and enter details in concomitant treatment section.

□ No action taken □ Dose increased □ Dose reduced

□ Drug stopped temporarily □ Drug stopped permanently

Subject was: choose **all relevant options** and enter details in concomitant treatment section if applicable.

□ Withdrawn from Trial

□ Treatment given (specify details in concomitant treatment section

□ Other (specify):

□ No Action

Does Serious Criteria apply? Serious is defined as fatal, life-threatening, inpatient hospitalization or prolongation of existing hospitalization, persistent or significant diability/incapability, congenital anomaly/birth defect, important medical event (ie, may jeopardize subject and may require medical/surgical intervention to prevent above listed outcomes). □ YES □ NO

Was the event a result of a study medication error? □ YES / UNKNOWN □ NO

If yes, complete Medication Error Log.

The following elements must be completed at the time the event resolves or at the end of the trial:

Causality: Is there a reasonable possibility the adverse event is related to trial treatment?

□ YES / UNKNOWN □ NO

If no, what is the most likely cause of the hypoglycemic adverse event? Choose **one** of the following:

□ Concomitant treatment – drug or nondrug (specify);

□ Other (specify).

If subject experienced symptoms of hypoglycemia, choose **all** that apply:

□ Hunger □ Palpitations □ Blurred Vision □ Headache □ Tachycardia □ Disorientation

□ Dizziness □ Tremulousness □ Loss of Consciousness □ Light Headedness □ Irritability

□ Seizures □ Sweats □ Other (specify)

What type of treatment was administered?

Oral carbohydrates □ YES □ NO

Parenteral □ YES □ NO

Was a precipitating event identified? □ YES □ NO

If yes, choose **all relevant options** from the list below:

□ Missed Meal/missed snack □ Diabetes Medication Error – wrong dose

□ Delayed Meal/delayed snack □ Diabetes Medication Error – wrong time

□ Exercise □ Other precipitating event (specify)

□ Comments