Study Name:

SPAN: A Phase 2, Open Label, Multicenter, **P**ilot Study to **A**ssess Safety and Efficacy of an E**n**teric Microgranule Formulation of Adrulipase in Patients with Exocrine Pancreatic Insufficiency (EPI) due to Cystic Fibrosis (CF)

Protocol Number: FWB-CF-2.03

NCT #: 05719311

Document Date: 5-12-2023

CLINICAL STUDY PROTOCOL

A Phase 2, Open Label, Multicenter, Pilot Study to Assess Safety and Efficacy of an Enteric Microgranule Formulation of Adrulipase in Patients with Exocrine Pancreatic Insufficiency (EPI) due to Cystic Fibrosis (CF)

Study Name:	**SPAN:** A Pha**s**e 2, Open Label, Multicenter, **P**ilot Study to **A**ssess Safety and Efficacy of an E**n**teric Microgranule Formulation of Adrulipase in Patients with Exocrine Pancreatic Insufficiency (EPI) due to Cystic Fibrosis (CF)
Protocol Number:	FWB-CF-2.03
Investigational Product:	Adrulipase (formerly MS1819)
Phase:	Phase 2
Sponsor:	First Wave BioPharma, Inc.
Protocol Date:	May 12, 2023
Protocol Version:	2.0

1 PROTOCOL APPROVAL SIGNATURES

Medical Monitor
James Pennington, MD
Chief Medical Officer
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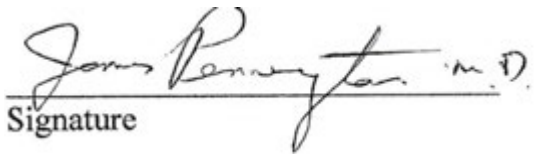

Signature

5/15/2023
Date

Sponsor Signatory

James Sapirstein
Chief Executive Officer
First Wave Biopharma, Inc.
USA

Signature

5/15/2023
Date

2 SYNOPSIS

Name of Sponsor/Company: First Wave Biopharma, Inc.	
Name of Investigational Product: Adrulipase (formerly MS1819)	
Protocol Number: FWB-CF-2.03	
Protocol Title: A Phase 2, Open Label, Multicenter, Pilot Study to Assess Safety and Efficacy of an Enteric Microgranule Formulation of Adrulipase in Patients with Exocrine Pancreatic Insufficiency (EPI) due to Cystic Fibrosis (CF)	
Study Name: SPAN: A Pha**s**e 2, Open Label, Multicenter, **P**ilot Study to **A**ssess Safety and Efficacy of an E**n**teric Microgranule Formulation of Adrulipase in Patients with Exocrine Pancreatic Insufficiency (EPI) due to Cystic Fibrosis (CF)	
Study Site(s): This study will be conducted at approximately 3 sites in the United States	
Number of Patients: Approximately **12**	**Phase of Development:** 2
Objectives To establish safety and efficacy of a new enteric microgranule formulation of adrulipase in patients with EPI due to CF.	
Study Design This is a Phase 2, single arm pilot study assessing the safety and efficacy of adrulipase in an enteric microgranule formulation. Patients with a confirmed diagnosis of cystic fibrosis who are 18 years of age or greater will be screened for eligibility if they have been clinically controlled, including minimal signs and symptoms of EPI, on a stable dose of commercial pancreatic enzyme replacement therapy (PERT) for at least one month. Patients on cystic fibrosis transmembrane conductance regulator (CFTR) modulator therapies must have been on a stable dose for at least 3 months prior to study entry, and no dose changes will be made during the study. Patients receiving gastric acid suppressants must have been on a stable dose for at least one month prior to study entry and no dose changes will be made during the study. Upon obtaining an informed consent, potentially eligible patients will receive dietary counselling during the week prior to the scheduled date of confinement for collecting stool samples for calculation of baseline coefficient of fat absorption (CFA). This counselling will emphasize the importance of dietary stability during the study. Patients found to have a CFA of 80% or greater while receiving their commercial PERT and meeting the other eligibility criteria will be enrolled into the study. The study will enroll patients into either Part 1 (under protocol version 1.0) or Part 2 (under protocol version 2.0). Under Part 1, patients will undergo a dose optimization phase during the three week treatment period where the adrulipase dose could be titrated from the low dose (1.2 g/day) up to medium (1.95 g/day) or high (2.4 g/day) doses. Under Part 2, all patients will receive 3.3 g/day of adrulipase and there will be no dose optimization, thus the treatment window will be two weeks.	

Upon study enrolment, the patient will be switched from their commercial PERT to receive adrulipase. The patient will remain on study for approximately three weeks (for Part 1 patients) or two weeks (for Part 2 patients), after which a repeat CFA will be obtained.

For Part 1, a dose titration scheme will be used for determining whether a low, medium, or high dose of adrulipase may succeed in controlling signs and symptoms of exocrine pancreatic insufficiency (EPI) and provide a CFA of 80% or greater. Patients will initially receive a low dose of adrulipase. Upon the appearance of EPI symptoms, lasting at least three days, and upon discussion with the investigator, the patient will be switched to the medium dose of adrulipase. If signs and symptoms of EPI persist for three or more days, the patient will be switched to the high dose of adrulipase. After patients complete three weeks of study treatment and their end of study CFA, they will be returned to their pre-study commercial PERT. An end of study safety visit will be scheduled for one week after finishing adrulipase therapy.

For Part 2, a dose of 3.3 g/day adrulipase will be used for all patients. After patients complete two weeks of study treatment and their end of study CFA, they will be returned to their pre-study commercial PERT. An end of study safety visit will be scheduled to occur one week after finishing adrulipase therapy.

Safety assessments will be made by collecting adverse events, safety lab assessments, and immunologic assays to assess drug induced immune responses.

Inclusion Criteria

To be eligible for study patients must satisfy all the following inclusion criteria:

1. Signed and dated informed consent form by patient as required by First Wave Biopharma, or designee and approval by an Institutional Review Board/Independent Ethics Committee (IRB/IEC)
2. Age 18 years or greater at the time of screening
3. Male or female
4. A confirmed diagnosis of cystic fibrosis, based on 2 clinical features consistent with CF, plus either a new/historic sweat chloride >60 mmol/L by quantitative pilocarpine iontophoresis (measured while not on a CFTR modulator) or genotype.
5. On stable dose of porcine PERT ≥1 month (30 days) prior to screening; stable dose is defined as dose of medication not changed during this time period, and the medication must be commercially available and be administered in the recommended dose range.
6. CFA = or > 80% at screening while on stable PERT
7. Normal nutritional status as defined by BMI ≥ 18.5 kg/M2 for patients ≥ 18 years of age.
8. Fecal elastase <100 µg/g of stool at screening
9. Clinically stable with no documented evidence of acute respiratory symptoms that would require administration of new oral or intravenous antibiotics, oxygen supplementation, or hospitalization within 30 days of screening or during the screening period.
10. Male and Female patients, if of childbearing potential, must use a reliable method of contraception during the study. A reliable method of birth control is defined as one of the following: oral or injectable contraceptives, intrauterine device, contraceptive implants, tubal ligation, hysterectomy, or a double -barrier method (diaphragm with spermicidal foam or jelly, or a condom), abstinence or vasectomy. Periodic abstinence (calendar, symptothermal, or post-

ovulation methods) is not an acceptable method of contraception. The preferred and usual lifestyle of the patient must also be evaluated in determining if sexual abstinence is a reliable method of birth control.

11. Be considered reliable and capable of adhering to the protocol, according to the judgment of the Investigator.

Exclusion Criteria

Patients will be excluded from the study if one or more of the following criteria are applicable:

1. History or diagnosis of fibrosing colonopathy
2. Total or partial gastrectomy
3. A history of solid organ transplant or significant surgical resection of the bowel; significant resection of the bowel is defined as any resection of the terminal ileum or ileocecal valve. Patients who have had qualitative, long-term changes in nutritional status after any other bowel resection (e.g., increased, or new need for pancreatic enzyme supplementation compared with preoperative status to maintain the same nutritional status) should also be excluded.
4. Any chronic diarrheal illness unrelated to pancreatic insufficiency (e.g., infectious gastroenteritis, sprue, inflammatory bowel disease)
5. Known hypersensitivity or other severe reaction to any ingredient of the investigational medicinal product (IMP)
6. Alanine aminotransferase (ALT) or aspartate aminotransferase (AST) level ≥3 ×upper limit of normal (ULN), or total bilirubin level ≥2.2 mg/dl at the Screening visit, unless in the opinion of the investigator this does not present a safety issue based on the patient's clinical status. Cases of suspected or confirmed Gilbert's syndrome should be discussed with the Medical Monito.
7. Signs and/or symptoms of liver cirrhosis or portal hypertension (e.g., splenomegaly, ascites, esophageal varices), or documented liver disease unrelated to CF
8. Patients with a known allergy to the stool marker (FD&C Blue #2)
9. Feeding via an enteral tube during 6 months before screening
10. Routine use of anti-diarrheals, anti-spasmodics, or cathartic laxatives, or a change in chronic osmotic laxatives (e.g., polyethylene glycol) regimen in the previous 3 months prior to screening
11. History of severe constipation with <1 evacuation/week under appropriate laxative therapy within the last 12 months before screening
12. Documentation of distal intestinal pseudo-obstruction syndrome within the last 12 months before screening
13. Forced expiratory volume ≤30% at the screening visit
14. Lactation or known pregnancy or positive pregnancy test at screening for women of childbearing potential (repeat at initial visit of EP)
15. Participation in another clinical study involving an investigational product within 30 days prior to screening for this study.

16. Patient's with poorly controlled diabetes according to the Investigator's judgment
17. Changes in CFTR modulator therapy during the 3 months prior to screening, for patients already on CFTR modulator therapy.
18. Changes in gastric acid suppressant therapy during the one month prior to screening for patients already on suppressant therapy.
19. Any condition that the Investigator believes would interfere with the intent of this study or would make participation not in the best interests of the patient.

Concomitant and Prohibited Medication

Standard–of–care medications are allowed (e.g., antibiotics, mucolytic agents, aerosols, CFTR modulators). Patients taking CFTR modulators must be on stable doses of the same modulator(s) for at least 3 months prior to screening. Patients should not start taking or modify doses of CFTR modulators for the duration of the study.

Gastric acid suppressants are allowed but must be a stable dosage of the same suppressant for 30 or more days before screening and must not be altered in dose or stopped during the study.

Prohibited medications during the entire clinical study will be as follows:

- Orlistat lipase inhibitor (e.g., Alli®, Xenical®)
- Laxatives consisting of mineral oil and castor oil (chronic use of osmotic laxatives is permitted)
- Symptomatic treatments of diarrhea: loperamide (e.g., loperamide generic, Imodium®, Imodium A-D®, Diamode®, Imotil®, Kao-Paverin®); atropine/diphenoxylate (Lonox®); and atropine/diphenoxylate (Lomocot®).

Investigational Product, Dosage, Duration and Mode of Administration

Adrulipase is supplied as size 00 immediate release capsules containing ~150 mg of lipase to be administered orally and taken with food. All patients in Part 1 will start at a low dose, if necessary, adjust to medium dose and if further dose optimization is necessary adjust to the high dose. All patients in Part 2 will receive a 3.3 g/day dose of adrulipase for the entirety of the study.

For Part 1, dosages will typically be fractionated as follows: 1/4 of the daily dose at each of 3 main meals, and 1/8 at each of the 2 snacks. The patients will be instructed to consume ~25 g of fat with every main meal and 12.5 g of fat with each of the 2 snacks to approximate a high fat meal of 100 g of fat/day. The 1.2 g/day dose will be fractionated as follows: 2 capsules with the morning, noontime, and evening meals, plus 1 capsule with the morning and evening snacks. The 1.95 g/day dose will be fractionated as follows: 3 capsules with each of the three main meals, and 2 capsules with each of the two snacks. The 2.4 g/day dose will be fractionated as follows: 4 capsules with each of the three main meals, and 2 capsules with each of the two snacks.

For Part 2, The 3.3 g/day dose will be fractionated as follows: 6 capsules with each of the three main meals, and 2 capsules with each of the two snacks.

Individual variations may occur if patients have discussed the variation with their PI or care team and total daily dose is achieved.

Duration of Study

The estimated study duration will be determined by when all patients complete the study. Each patient will have up to 6 weeks for screening, followed by approximately 14 or 21 days study treatment, for Part 2 or Part 1, respectively, plus a follow up visit within 7 days for a total estimated study duration of 35 to 70 days per patient.

Criteria for Evaluation

Safety endpoints:

AEs, SAEs, discontinuations due to adverse events, and laboratory values, including assays for drug induced antibodies.

Efficacy Endpoints:

Primary Endpoints:

The primary efficacy endpoint is the CFA that will be assessed at the end of the 3-week treatment period for Part 1 and at the end of the 2-week treatment period for Part 2.

Secondary Endpoints:

- Stool weights
- Signs and symptoms of malabsorption
- Coefficient of nitrogen absorption (CNA)
- Body weight
- Body mass index

Statistical Methods

Statistical methods will be further detailed in the Statistical Analysis plan (SAP).

Sample size: Based upon CFA data obtained in the recently completed Phase 2 studies (OPTION, OPTION 2), 12 patients using a dose titration scheme should provide sufficient point estimates of CFA and all other secondary efficacy endpoints. Given the acceptable safety profile obtained in the previous Phase 2 OPTION studies, a sample of 12 patients should be adequate for observation of safety.

Analysis Sets:

Safety Set: Patients receiving at least one dose of study treatment.

Efficacy Sets

- **Modified Intent-to-Treat Se**t (mITT): All enrolled patients receiving at least one dose of treatment and having a valid stool collection and CFA post baseline while receiving their study drug. The primary analysis will be performed in the mITT set.
- **Per Protocol Set**: mITT patients completing the entire study period without any major protocol deviation. A sensitivity analysis will be performed in the Per Protocol Set.

Handling Missing Data:

Missing data will not be replaced.

Safety Analysis:

Safety (AEs, SAEs, discontinuations due to adverse events, and safety laboratory values) will be assessed by descriptive methods. Assays for anti-drug antibodies will be conducted.

Efficacy Analysis:

The primary efficacy endpoint is defined as the CFA assessed at the end of the treatment period. CFA will be calculated as follows:

CFA (%) = 100 ((fat intake (g) − fat excretion (g))/ fat intake (g))

Primary Analysis:

The analysis of the primary endpoint will be done using descriptive methods.**Secondary Efficacy Endpoints:**

Coefficient of nitrogen absorption (CNA) will be calculated and analyzed in the same manner as CFA.

Stool weights, signs and symptoms of malabsorption, body weight, and BMI, will be analyzed using descriptive methods.

3 SCHEDULE OF ASSESSMENTS

Table 1: Schedule of Assessments Part 1

	SCREENING		ADRULIPASE TREATMENT				END OF
	Consent and Initial Evaluations	**CFA on PERT**	**Dispense**	**Optimize**	**Optimize**	**CFA on Adrulipase**	**STUDY/EARLY TERMINATION**
Visit Number	**1**[a]	**2**[b]	**3**	**4 (T)**[c]	**5 (T)**[c]	**6**[b]	**7**
Study Week	**-6**	**-2**	**1**	**2**	**3**	**4**	**5**
Study Days	**-42**	**-14**	**1**	**5**	**10**	**16**	**28**
Visit Window (days)	**+7**	**±7**	**±2**	**±2**	**±2**	**+5**	**±7**
Pre-Visit Instructions	**X**	**X**		**X**	**X**		
Supervised confinement		**X**				**X**	
Clinical Assessments							
Obtain informed consent	**X**						
Demographics	**X**						
Complete history and physical	**X**						
Focused physical exam[d]		**X**				**X**	**X**
Confirm CF diagnosis (Inclusion Criteria 4)	**X**						
Height/weight, vital signs (sitting)	**X**	**X**				**X**	
Inclusion/exclusion criteria review	**X**	**X**	**X**				
Concomitant medications	**X**	**X**	**X**	**X**	**X**	**X**	**X**

	SCREENING		ADRULIPASE TREATMENT				END OF
	Consent and Initial Evaluations	**CFA on PERT**	**Dispense**	**Optimize**	**Optimize**	**CFA on Adrulipase**	**STUDY/EARLY TERMINATION**
Visit Number	**1**[a]	**2**[b]	**3**	**4 (T)**[c]	**5 (T)**[c]	**6**[b]	**7**
Study Week	**-6**	**-2**	**1**	**2**	**3**	**4**	**5**
Study Days	**-42**	**-14**	**1**	**5**	**10**	**16**	**28**
Visit Window (days)	**+7**	**±7**	**±2**	**±2**	**±2**	**+5**	**±7**
Adverse events	**X**	**X**	**X**	**X**	**X**	**X**	**X**
Confirm scheduled date for next supervised confinement visit	**X**				**X**		
Study Treatment							
Instruct regarding study drug adrulipase (dose optimization, low, medium, high)			**X**			**X**	
Full study drug accountability at the end of confinement						**X**	
Return Adrulipase at the end of confinement						**X**	
Record fat and protein intake and study drug taken at all meals and snacks			**X**			**X**	
Efficacy Measures							
Malabsorption signs & symptoms		**X**	**X**	**X**	**X**	**X**	**X**
72-hour controlled diet record		**X**				**X**	

	SCREENING		ADRULIPASE TREATMENT				END OF
	Consent and Initial Evaluations	**CFA on PERT**	**Dispense**	**Optimize**	**Optimize**	**CFA on Adrulipase**	**STUDY/EARLY TERMINATION**
Visit Number	**1**[a]	**2**[b]	**3**	**4 (T)**[c]	**5 (T)**[c]	**6**[b]	**7**
Study Week	**-6**	**-2**	**1**	**2**	**3**	**4**	**5**
Study Days	**-42**	**-14**	**1**	**5**	**10**	**16**	**28**
Visit Window (days)	**+7**	**±7**	**±2**	**±2**	**±2**	**+5**	**±7**
Marker-to-marker stool collection and stool weight[f]		**X**				**X**	
Laboratory Tests							
Urinalysis	**X**					**X**	
Pregnancy test (serum for V1 screening and urine dipstick for other visits)[g]	**X**		**X**			**X**	**X**
Hematology, clinical chemistry, PT/INR, and aPTT [h]	**X**					**X**	
Fasting[i] lipids (patient to come in fasting status) and pre-albumin	**X**					**X**	
Serum samples for anti-adrulipase lipase antibodies and adrulipase concentrations		**X**				**X**	**X**
Fecal pancreatic elastase[j]	**X**						
Diagnostic Test							
Spirometry	**X**						

	SCREENING		ADRULIPASE TREATMENT				END OF
	Consent and Initial Evaluations	**CFA on PERT**	**Dispense**	**Optimize**	**Optimize**	**CFA on Adrulipase**	**STUDY/EARLY TERMINATION**
Visit Number	**1**[a]	**2**[b]	**3**	**4 (T)**[c]	**5 (T)**[c]	**6**[b]	**7**
Study Week	**-6**	**-2**	**1**	**2**	**3**	**4**	**5**
Study Days	**-42**	**-14**	**1**	**5**	**10**	**16**	**28**
Visit Window (days)	**+7**	**±7**	**±2**	**±2**	**±2**	**+5**	**±7**
Switch back to Prescribed Porcine PERT							
Switch back to prescribed porcine PERT[k]						**X**	**X**

a Screening procedures can occur up to 14 days before the first day of supervised confinement (V2). As some lab assessments require fasting status, site may utilize a pre-screening telephone consent process to obtain agreement in advance for patients to adhere fasting for at least 8 hours. Patients will also be asked to bring a stool sample to V1

b Visit 2 and Visit 6 are the first and second scheduled confinement visits and can take up to 7 days. A 5-day window is permitted around the scheduled confinement for both V2 and V6 to accommodate for scheduling. Dosing must have occurred for at least 16 days prior to the start of the scheduled confinement.

c Visits 4 and 5 are telephone visits to assess any changes to AEs and concomitant medications in addition to confirming the visit date for the next scheduled supervised confinement.

d The Focused Physical Exam will evaluate gastrointestinal tract, heart, and lungs.

e At the end of V6 (after the last stool sample has been collected), Patients will begin treatment with their prestudy porcine PERT.

f The stool samples will be sent to the central laboratory and CFA, CNA, and stool weight will be measured.

g A serum pregnancy test must be conducted in females of reproductive potential at screening Visit 1. Pregnancy status will be re-evaluated via urine pregnancy test in these Patients at Visit 3, 6 and at the End-of-Study or Early Termination visit (Visit 7).

h On the basis of laboratory safety values, unscheduled hepatic monitoring testing may be performed in patients with new, clinically meaningful increases in liver function tests occurring during the study, in consultation with study designated Medical Monitor. These tests are to be done through the central labs.

i Fasting labs should be taken after patients have been in a fasting status for at least 8 hours at Visits 1 and 6.

j Fecal pancreatic elastase will be collected at Visit 1 and sent for analysis to the central laboratory.

k At the end of the supervised confinement (once the second blue dye marker sample has been collected), patients will return to their prestudy porcine PERT dose.

Table 2: Schedule of Assessments Part 2

	SCREENING		ADRULIPASE TREATMENT				END OF
	Consent and Initial Evaluations	**CFA on PERT**	**Dispense**	**Telephone Visits**		**CFA on Adrulipase**	**STUDY/EARLY TERMINATION**
Visit Number	**1[a]**	**2[b]**	**3**	**4 (T)[c]**	**5 (T)[c]**	**6[b]**	**7**
Study Week	**-6**	**-2**	**1**	**2**	**3**	**4**	**5**
Study Days	**-42**	**-14**	**1**	**3**	**7**	**10**	**21**
Visit Window (days)	**+7**	**±7**	**±2**	**±2**	**±2**	**+3**	**±2**
Pre-Visit Instructions	**X**	**X**		**X**	**X**		
Supervised confinement		**X**				**X**	
Clinical Assessments							
Obtain informed consent	**X**						
Demographics	**X**						
Complete history and physical	**X**						
Focused physical exam[d]		**X**				**X**	**X**
Confirm CF diagnosis (Inclusion Criteria 4)	**X**						
Height/weight, vital signs (sitting)	**X**	**X**				**X**	
Inclusion/exclusion criteria review	**X**	**X**	**X**				
Concomitant medications	**X**	**X**	**X**	**X**	**X**	**X**	**X**
Adverse events	**X**	**X**	**X**	**X**	**X**	**X**	**X**

	SCREENING		ADRULIPASE TREATMENT				END OF
	Consent and Initial Evaluations	**CFA on PERT**	**Dispense**	**Telephone Visits**		**CFA on Adrulipase**	**STUDY/EARLY TERMINATION**
Visit Number	**1**[a]	**2**[b]	**3**	**4 (T)**[c]	**5 (T)**[c]	**6**[b]	**7**
Study Week	**-6**	**-2**	**1**	**2**	**3**	**4**	**5**
Study Days	**-42**	**-14**	**1**	**3**	**7**	**10**	**21**
Visit Window (days)	**+7**	**±7**	**±2**	**±2**	**±2**	**+3**	**±2**
Confirm scheduled date for next supervised confinement visit	**X**				**X**		
Study Treatment							
Instruct regarding study drug adrulipase dosing			**X**			**X**	
Full study drug accountability at the end of confinement						**X**	
Return Adrulipase at the end of confinement						**X**	
Record fat and protein intake and study drug taken at all meals and snacks			**X**			**X**	
Efficacy Measures							
Malabsorption signs & symptoms		**X**	**X**	**X**	**X**	**X**	**X**
72-hour controlled diet record		**X**				**X**	

	SCREENING		ADRULIPASE TREATMENT				END OF
	Consent and Initial Evaluations	**CFA on PERT**	**Dispense**	**Telephone Visits**		**CFA on Adrulipase**	**STUDY/EARLY TERMINATION**
Visit Number	**1**[a]	**2**[b]	**3**	**4 (T)**[c]	**5 (T)**[c]	**6**[b]	**7**
Study Week	**-6**	**-2**	**1**	**2**	**3**	**4**	**5**
Study Days	**-42**	**-14**	**1**	**3**	**7**	**10**	**21**
Visit Window (days)	**+7**	**±7**	**±2**	**±2**	**±2**	**+3**	**±2**
Marker-to-marker stool collection and stool weight[f]		**X**				**X**	
Laboratory Tests							
Urinalysis	**X**					**X**	
Pregnancy test (serum for V1 screening and urine dipstick for other visits)[g]	**X**		**X**			**X**	**X**
Hematology, clinical chemistry, PT/INR, and aPTT [h]	**X**					**X**	
Fasting[i] lipids (patient to come in fasting status) and pre-albumin	**X**					**X**	
Serum samples for anti-adrulipase lipase antibodies and adrulipase concentrations		**X**				**X**	**X**
Fecal pancreatic elastase[j]	**X**						
Diagnostic Test							
Spirometry	**X**						

	SCREENING		ADRULIPASE TREATMENT				END OF
	Consent and Initial Evaluations	**CFA on PERT**	**Dispense**	**Telephone Visits**		**CFA on Adrulipase**	**STUDY/EARLY TERMINATION**
Visit Number	**1**[a]	**2**[b]	**3**	**4 (T)**[c]	**5 (T)**[c]	**6**[b]	**7**
Study Week	**-6**	**-2**	**1**	**2**	**3**	**4**	**5**
Study Days	**-42**	**-14**	**1**	**3**	**7**	**10**	**21**
Visit Window (days)	**+7**	**±7**	**±2**	**±2**	**±2**	**+3**	**±2**
Switch back to Prescribed Porcine PERT							
Switch back to prescribed porcine PERT[k]						X	X

a Screening procedures can occur up to 14 days before the first day of supervised confinement (V2). As some lab assessments require fasting status, site may utilize a pre-screening telephone consent process to obtain agreement in advance for patients to adhere fasting for at least 8 hours. Patients will also be asked to bring a stool sample to V1

b Visit 2 and Visit 6 are the first and second scheduled confinement visits and can take up to 7 days. A 7-day and 5-day window is permitted around the scheduled confinement for V2 and V6, respectively, to accommodate for scheduling. Dosing must have occurred for at least 10 days prior to the start of the scheduled confinement.

c Visits 4 and 5 are telephone visits to assess any changes to AEs and concomitant medications in addition to confirming the visit date for the next scheduled supervised confinement.

d The Focused Physical Exam will evaluate gastrointestinal tract, heart, and lungs.

e At the end of V6 (after the last stool sample has been collected), Patients will begin treatment with their prestudy porcine PERT.

f The stool samples will be sent to the central laboratory and CFA, CNA, and stool weight will be measured.

g A serum pregnancy test must be conducted in females of reproductive potential at screening Visit 1. Pregnancy status will be re-evaluated via urine pregnancy test in these Patients at Visit 3, 6 and at the End-of-Study or Early Termination visit (Visit 7).

h On the basis of laboratory safety values, unscheduled hepatic monitoring testing may be performed in patients with new, clinically meaningful increases in liver function tests occurring during the study, in consultation with study designated Medical Monitor. These tests are to be done through the central labs.

i Fasting labs should be taken after patients have been in a fasting status for at least 8 hours at Visits 1 and 6.

j Fecal pancreatic elastase will be collected at Visit 1 and sent for analysis to the central laboratory.

k At the end of the supervised confinement (once the second blue dye marker sample has been collected), patients will return to their prestudy porcine PERT dose.

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LIST OF TABLES

5 LIST OF ABBREVIATIONS

AE	adverse event
ALP	alkaline phosphatase
ALT	alanine aminotransferase
aPTT	Activated partial thromboplastin time
AST	aspartate aminotransferase
BP	blood pressure
CF	cystic fibrosis
CFA	coefficient of fat absorption
CFTR	cystic fibrosis transmembrane conductance regulator

cGCP	current Good Clinical Practice
cGMP	current Good Manufacturing practice
CI	confidence interval
CNA	coefficient of nitrogen absorption
CRF	case report form
CRO	contract research organization
EPI	Exocrine pancreatic insufficiency
eCRF	electronic case report form
FAS	full analysis set
FDA	Food and Drug Administration
FEV_1	forced expiratory volume in 1 second
FD	freeze-dried
FDA	Food and Drug Administration
GCP	Good Clinical Practice
HDL	high density lipoprotein
ICF	informed consent form
ICH	International Council on Harmonisation of Technical Requirements for Registration of Pharmaceuticals for Human Use
IEC	independent ethics committee
IMP	investigational medicinal product
IRB	Institutional review board
LDL	low density lipoprotein
MedDRA	Medical Dictionary for Regulatory Activities
mITT	Modified Intent-To-Treat
PERT	Pancreatic Enzyme Replacement Therapy
PP	per protocol
PPI	proton pump inhibitor

SAE	serious adverse event
SAP	statistical analysis plan
SD	spray dried
SOP	standard operating procedures
ULN	upper limit of normal
WHO	World Health Organization

6 INTRODUCTION

6.1 Exocrine Pancreatic Insufficiency

The pancreas is both an endocrine and exocrine organ. As an exocrine gland, it secretes pancreatic juice containing digestive enzymes and a bicarbonate-rich fluid that neutralizes acidic gastric secretions, providing the correct pH for duodenal digestion by the pancreatic enzymes for absorption of nutrients in the small intestine. Exocrine pancreatic insufficiency (EPI) is defined as a deficiency of exocrine pancreatic enzymes which results in the inability to maintain normal digestion. The symptomatology of EPI is mainly due to pancreatic lipase deficiency. Pancreatic lipase is an enzyme that hydrolyzes triglycerides into monoglycerides and free fatty acids.

Lipid maldigestion due to lipase deficiency can lead to weight loss, steatorrhea (excess fat in the feces), fat-soluble vitamin deficiencies (e.g., vitamins A, D, E, and K), and can eventually lead to vitamin B12 deficiency and a potential decrease in quality of life (King et al, 1979). In EPI due to cystic fibrosis (CF) or chronic pancreatitis, there is also a decrease in bicarbonate output, causing a lower intestinal pH, which leads to precipitation of bile salt acids and impairment of micelle formation of fats. Fat maldigestion is further exacerbated by lower levels of pancreatic lipase and colipase leading to decreased hydrolysis of intraluminal fat (Struyvenberg et al, 2017).

6.2 Exocrine Pancreatic Insufficiency due to Cystic Fibrosis

Cystic fibrosis is an autosomal recessive chronic progressive disorder with high morbidity and a shortened life expectancy. CF affects more than 70,000 people worldwide (Cystic Fibrosis Foundation). In most Caucasian populations, CF prevalence is 7-8 cases per 100,000 inhabitants but is less frequent in other populations (Farrell, 2008; Banks et al, 2010; Cystic Fibrosis Foundation Patient Registry 2010). In the United States, there are approximately 30,000 affected individuals (Cystic Fibrosis Foundation Patient Registry, 2016). The disease occurs as a consequence of mutations in the gene encoding for the CF transmembrane conductance regulator (CFTR) protein, a plasma membrane ion channel that mediates transport of chloride, bicarbonate, and other anions. Dysfunction of the CFTR gene leads to a decrease in luminal fluid volume and decreased pH, resulting in protein precipitation within the ductal lumen and loss of normal acinar cell function. Mutation of both

alleles of CFTR results in the production of thick mucus, which causes a multisystem disease of the upper and lower respiratory tract, the digestive system, and the reproductive tract.

Cystic fibrosis is frequently associated with EPI and is usually observed at birth because of in utero exocrine pancreatic damage. A neonatal screening study found that 63% of infants with CF were exocrine insufficient, and approximately 30% of the exocrine sufficient group would progressively become insufficient over the next 36 months (Waters et al, 1990). Even individuals with less severe CFTR mutations (Class IV, V, or VI) were more likely to develop EPI later in life (Ledder et al, 2014; Bodewes et al, 2015).

6.3 Standard of Care for the Compensation of Exocrine Pancreatic Insufficiency

Porcine pancreatic enzyme replacement therapy (PERT) is the standard of care to prevent maldigestion, malnutrition, and excessive weight loss (Greenberger and Toskes 2008, Dhanasekaran and Toskes 2010, Greenberger and Toskes 2013). This therapy uses animal pancreatic extracts, which have been marketed since 1938. Porcine pancreatic extracts (porcine PERT) consist of an enzymatic mixture from pig pancreas, namely amylase, protease, and lipase, which digest starch, protein, and lipids, respectively. Therapy with porcine PERTs has demonstrated efficacy on the maldigestion symptoms due to EPI.

Despite their long-term use, major concerns have been raised by the CF Foundation, the CF community, and the Food and Drug Administration (FDA) because of the animal ingredients used for porcine PERTs and the risk of transmission of conventional and nonconventional infectious agents. Furthermore, therapy with porcine PERTs has clear limitations:

- The protease enzymatic constituents of porcine PERTs are suspected to cause fibrosing colonopathy (Kimura et al, 1998; Kimura et al. 1999), a severe adverse event (AE) observed at high doses in young patients with CF.
- To prevent this risk of fibrosing colonopathy, the CF Foundation recommended that dosages should be limited to 500-2500 lipase units/kg/meal, ≤10,000 lipase units/kg/day, or ≤4000 lipase units/g of dietary fat/day (Stallings et al, 2008). Noticeably, porcine PERTs have only mild or null effects on creatorrhea (Van Hoozen et al, 1997, Airinei et al, 2011).
- Incomplete correction of the lipid malabsorption, which can only be reached by high doses of enteric enzyme therapy in approximately 50% of patients with EPI caused by chronic pancreatitis and/or pancreatectomy [Dominguez-Munoz et al, 2006; Safdi et al, 2006]. An internal survey based on all publicly available individual data show that only 25% of patients affected by CF with EPI had fully normalized coefficient of fat absorption (CFA) (i.e., CFA higher than 93%) on porcine PERT substitution therapy. The incomplete enzymatic activity of porcine PERTs in the physiochemical environment of the upper digestive tract may explain their reduced efficacy. A number of factors are involved, including the gastric acid pH in the range of 1 to 4; the intestinal pH ranging from 4 to 6 (porcine PERTs are active at pH > 6.5 in presence of biliary salts); and the protease content of the upper digestive tract (i.e.,

gastric pepsin). Due to the acid lability of pig pancreatic lipase, commercial enzymes are formulated as enterically coated beads.

- The hazard of zoonotic pathogen transmission to humans remains possible with porcine PERTs because of their animal origins. For example, the porcine parvovirus may be present in these extracts, with (minimal) risk that it can cross species and transmit diseases to humans (Cherney, 2008).
- Some additional concerns identified with porcine PERTs include irritation of oral mucosa if chewed/retained and hyperuricemia because of the presence of purines in porcine pancreatic extracts [Package insert Creon, Pancreaze, Zenpep, Pertzye]. In addition, patients with an allergy to proteins of porcine origin may have severe allergic reactions.
- As all of the currently marketed PERTs are porcine derived, there is a supply risk (both quality and quantity) that is dependent on the availability of pig herds.

Because of the risks and limitations of porcine PERT, new drugs are demanded by pharmaceutical regulatory agencies and patient associations.

A fundamental question is the value and usefulness of a lipase-only enzyme replacement therapy, such as adrulipase for patients with EPI. For decades, the only available enzyme supplements to treat EPI have been porcine-derived pancreatic enzymes containing a mixture of lipase, protease, and amylase. While the importance of the individual components in treating maldigestion has not been prospectively assessed, it is widely agreed that the earliest and sometimes only sign of malabsorption is steatorrhea (DiMagno et al, 1993; Ferrone et al, 2007). Extrapancreatic sources of protease and amylase exist (Dominguez-Munoz, 2007; Hammer et al, 2010), whereas little lipase is available other than that secreted by the pancreas. Furthermore, some have suggested that protease deficiency can be treated in part with essential amino-acid mixtures (Engelen et al, 2014). Most agree that amylase is the least important of the 3 supplemental pancreatic enzymes (Ladas et al, 1993) perhaps because of high amylase content in salivary gland secretions. In summary, the future value of a lipase-only supplement, either as a lipase-only replacement therapy in patients with mild to moderate EPI, or as an augmenting lipase for hyporesponsive patients reaching maximal doses of porcine enzyme treatment, remains to be seen.

6.4 Investigational Product (Adrulipase)

Adrulipase alfa (adrulipase), formerly referred to as MS1819, is a preparation of a recombinant yeast lipase, designated LIP2, which is a secreted enzyme, isolated from the strictly aerobic *Yarrowia lipolytica* that is found in various foods such as cheese and olive oil. This nonpathogen micro-organism is widely used as a biocatalyst (e.g., erythritol production, a polyol used as a food additive) and is generally recognized as safe by the FDA for several industrial processes. The genetically engineered strain of *Yarrowia lipolytica* contains 5 additional copies of the LIP2 gene integrated into its genome (total of 6 including the endogenous native LIP2 gene). The genetic stability of the strain has been assessed for 100 generations, providing a safety margin as the full fermentation process corresponds to

approximately 30 generations. Both current Good Manufacturing Practice (cGMP)-compliant Master Cell Bank and Working Cell Bank have been manufactured.

The LIP2 gene product is a 334 amino-acid precursor that is released extracellularly as a 301 amino-acid protein after cleavage of its peptide signal. The protein is naturally glycosylated which provides efficient protection against the proteolysis by the gastric pepsin (Pignede et al, 2000). At least 4 secreted glycosylated isoforms of LIP2 have been characterized (Aloulou et al, 2007).

Until 2009, the active ingredient for adrulipase (recombinant yeast lipase, designated LIP2) was freeze-dried (FD) to obtain a final drug substance, MS1819-FD. From 2010 onwards, the process was changed for large scale production and the active ingredient for adrulipase was mixed with maltodextrin and then spray dried (SD) to obtain a final drug substance. In 2022, the SD process was further optimized and incorporated an enteric polymer to generic enteric microgranules.

In is this clinical study, adrulipase is supplied as size double zero capsules each containing ~150 mg lipase.

6.5 Preclinical Studies with Adrulipase

6.5.1 In Vitro Studies

The enzymatic activity of LIP2 has been extensively investigated *in vitro* and has an appropriate profile to compensate the EPI in patients with severe chronic pancreatic diseases. For example, the optimal activity of LIP2 occurs at pH 6, and works well in a range from pH 4 to 7 which is usually found in the duodenum of CF patients with decreased pancreas derived bicarbonate. In addition, LIP2 enzymatic activity is not inactivated by bile salts. Furthermore, LIP2 is active on triglycerides with a wide range of fatty-acid lengths, including long-chain triglycerides, which are the predominant forms of triglycerides in the human diet. Compared with the porcine pancreatic lipase, LIP2 is more active than the porcine pancreatic lipase with all triglycerides tested at pH ranging from 4 to 6.

6.5.2 In Vivo Studies

The efficacy of MS1819-FD has been investigated in minipigs; in which experimental pancreatitis and EPI were induced by pancreatic duct ligation. Daily doses of MS1819-FD 10.5 mg or greater administered once a day nearly completely corrected the CFA of approximately 20 kg minipigs (Aloulou 2015).

The safety of adrulipase has been investigated in 2 nonclinical regulatory trials whereby MS1819 was well tolerated at dosages up to 4700 mg/kg in rats and 1175 mg/kg in minipigs for up to 13 weeks, equivalent to 1000mg/kg and 250mg/kg of lipase respectively.

In summary, adrulipase was effective in minipigs and was nontoxic in both rodents and nonrodent species up to a maximum feasible dose over 3 months' administration.

6.6 Clinical Studies with MS1819

6.6.1 FLIP110 First-in-Man Study with MS1819-FD

The efficacy and safety of MS1819-FD have been investigated in a first-in-man study, FLIP110. This exploratory clinical trial without predefined statistical analysis objectives, was a randomized, double blind, placebo controlled, parallel study.

Patients affected with chronic pancreatitis or pancreatectomy and severe EPI, were randomly assigned to 2 phases with an allocation ratio of 2:1 to receive MS1819-FD 20 mg 3 times a day or a 'dummy' placebo treatment identical in aspect and taste. Each treatment was given for a phase of 1 week after a 1-week washout phase.

Twelve patients were randomly included: 8 in MS1819-FD phase and 4 in the placebo phase. Three patients in the MS1819-FD phase were excluded from the per protocol (PP) analysis because of severe protocol deviations (i.e., steatorrhea < 7 g/day in the baseline phase demonstrating the lack of significant EPI in these patients).

The primary endpoint of the study was defined as the relative change in steatorrhea compared with baseline. A nonstatistically significant difference of the primary endpoint was found between the 2 phases both in intent-to-treat (ITT: -14.6 % ± 26.6 in the MS1819-FD phase vs +16.9 % ± 40 in the placebo phase; not significant) and PP analysis (-15.8 % ± 20.6 in the MS1819-FD phase vs +16.9 % ± 40 in the placebo phase; not significant), respectively.

Secondary efficacy endpoints also support the efficacy of MS1819-FD compared with placebo in both ITT and PP populations, including the absolute change in CFA (ITT: +6.1 ± 13.4 vs -6.7% ± 11.0), number of daily evacuations over 7 days (ITT: -19.2% ± 22.8 vs +4.1% ± 11.7; p =.09), the weight of stools (ITT: -10.6% ± 27.4 vs +25.5% ± 48.2; not significant), and Bristol scale, which is a classification of the form of human feces to evaluate the effectiveness of treatments for diseases of the bowel (ITT: -5.13% ± 15.5 vs +2.46% ± 16.6; p<.0001).

MS1819-FD was well tolerated with no serious adverse events (SAEs). Only 2 AEs were observed: constipation (2 patients in the MS1819 group) and hypoglycemia (2 patients in the MS1819 group and 1 patient in the placebo group).

In summary, the FLIP110 study supported the continued clinical investigations of the efficacy of higher doses of MS1819 using established surrogate biomarkers for EPI correction.

6.6.2 MS1819/16/01 Phase 2a Study with MS1819 in patients with EPI due to chronic pancreatitis or pancreatectomy

An open-label, dose escalation Phase 2a study (MS1819/16/01) was conducted to investigate the safety of escalating doses of MS1819 in 11 patients with EPI caused by chronic pancreatitis or pancreatectomy.

The primary objective of this study was to investigate the safety of escalating doses of MS1819 (280 mg/day, 560 mg/day, 1120 mg/day, and 2240 mg/day). Each dose was given over a two-

week period. A CFA was obtained at baseline after washout, and after each escalating dose period. Safety assessments included immunoallergic side effects, digestive symptoms, and clinical laboratory tests. The secondary objective of this study was to investigate the efficacy of MS1819 by assessment of CFA.

Eleven patients were enrolled into the study, and ten completed all doses. One patient withdrew during the 1120 mg/day dose level due to severe diarrhea. This was not assessed as related to study drug.

In the Intent-to-Treat population, mean CFA rose from a baseline of 46.1% to 62.4% at the highest dose ($p = 0.001$). A number of symptoms improved comparing the highest dose to baseline, including stool consistency ($p < 0.001$), number of bowel movements ($p = 0.006$), steatorrhea ($p = 0.008$), and abdominal pain ($p = 0.148$). The treatment was well tolerated, with only two SAEs reported. Only one SAE was considered by the investigator to be possibly related to study drug, which was a patient with mild, transient elevations of liver enzymes. The elevations were between 2 and 3 X upper limit of normal (ULN), and lasted for about two weeks, a period of time that the patient was no longer receiving study drug. The patient had no symptoms of liver disease.

6.6.3 AZ-CF2001, A Phase 2, Open-Label, Multicentre, 2x2 Crossover Trial to assess the Safety and Efficacy of MS1819-SD in Patients with Exocrine Pancreatic Insufficiency due to Cystic Fibrosis

This study, referred to as the OPTION study, was the first experience with MS1819 in patients with cystic fibrosis. The study began in February of 2019, and all patients completed the trial by July 2019. The study was conducted in 14 sites, with 11 in the U.S. and 3 in Poland. All sites participated in enrollment. The primary objectives of the study were to ascertain safety of 2240 mg/day dose of MS1819 in patients with cystic fibrosis and to compare the efficacy of MS1819 to commercial PERT in digestion of fat. The study was not powered for inferential statistics. A total of at least 30 patients were planned to complete the study.

Inclusion criteria were cystic fibrosis patients aged 18 or older, who had been clinically well-controlled while on a stable dose of PERT for at least 30 days. The dose of MS1819 employed was 2240 mg/day. This dose, used previously in a Phase 2 study of chronic pancreatitis, was the highest dose used in man prior to the OPTION study. In discussions with FDA, it was agreed that the safety profile in the prior study in chronic pancreatitis patients was sufficient to allow this as the dose for a first-time study in cystic fibrosis.

Patients were randomized to begin the study in a treatment arm of MS1819, or a treatment arm using the same PERT, and same dose, they had been receiving prior to enrollment. After three weeks on treatment, a CFA was measured, and the patient was crossed over to the opposite treatment. After three more weeks of treatment, a CFA was measured, and the patient was returned to their pre-study PERT. An end of study visit was done two weeks after completing the treatments or upon early withdrawal of patients.

A total of 46 patients were screened, 41 were enrolled and 32 completed the study. Of the 9 withdrawals, one patient discontinued after completing both study treatment periods, three

patients discontinued during treatment with PERT and five patients discontinued during treatment with MS1819. The withdrawals of patients who discontinued during treatment with MS1819 were primarily attributed to increased symptoms of exocrine pancreatic insufficiency.

The mean CFA for the MS1819 group was 56% (range 7% to 92%), and the mean CFA for the PERT group was 86% (range 57% to 97%). This mean CFA of 56% on a dose of 2240 mg/day MS1819 was similar to the mean CFA of 62% in the chronic pancreatitis study at the same dose. The mean coefficient of nitrogen absorption (CNA) for the MS1819 group was 93% and the mean CNA for the PERT group was 97%. Symptoms of exocrine pancreatic insufficiency were more prominent in the MS1819 group.

MS1819 was well-tolerated, with no remarkable adverse events. Three SAEs occurred during the screening period, but none during the study itself. Each of these SAEs was due to pulmonary exacerbations of cystic fibrosis. Laboratory values were unremarkable during the study.

6.6.4 AZ-CF2002: A Phase 2, Open-Label, Multicentre, 2x2 Crossover Study to assess the Safety and Efficacy of MS1819 in Enteric Capsules in Patients with Exocrine Pancreatic Insufficiency due to Cystic Fibrosis; with an Extension Phase Evaluation of Immediate-Release MS1819 Capsules

This study, referred to as the OPTION 2 study, was the second experience with MS1819 in patients with cystic fibrosis. The study began in August of 2020, and all patients completed the study by April 2021. The study was conducted in 9 sites, with 5 in the U.S. and 4 in Poland. All sites participated in enrollment.

The primary objectives of the study were to assess the safety and efficacy of MS1819 in enteric capsules vs porcine pancreatic enzyme replacement therapy (PERT) in patients with exocrine pancreatic insufficiency (EPI) due to cystic fibrosis (CF). The safety and tolerability of MS1819 treatment (2240 and 4480 mg/day; enteric capsules) were assessed during the crossover phase, as was efficacy by comparing coefficient of fat absorption (CFA) percentages across the MS1819 (2240 and 4480 mg/day) versus porcine PERT treatment groups. The exploratory objective during the dose extension phase was to identify immediate-release MS1819 (4.4 or 6.7 g/day) doses that were safe and resulted in CFA percentages residing within the current PERT therapeutic range. The study was not powered for inferential statistics. A total of at least 30 patients were planned to complete the study.

A total of 59 patients were screened, 44 enrolled and randomized and 40 completed the study. A total of four patients withdrew from the study. Three patients withdrew from the crossover phase of the study, 2 in the MS1819 DR treatment group and 1 in the PERT treatment group. One additional patient in the extension MS1819 IR 4480 mg/day treatment group discontinued the study due to mild TEAEs, which resolved.

Mean CFA percentages were lower in the crossover groups (MS1819 DR 2240 and 4480 mg/day) and in dose extensions (MS1819 IR 4.4 g/day, 6.7 g/day), ranging from 50.6 to 66.0% versus 83.3 to 89.3% for the corresponding PERT groups.

MS1819 was well-tolerated, with no remarkable adverse events and laboratory values were unremarkable during the study.

6.6.5 MS1819/18/02: A Multicentre, Open-Label Phase 2 Study with Escalating Doses of MS1819-SD On Top of a Stable Dose of PPEs, to Investigate the Efficacy and Safety of This Combination for the Compensation of Severe Exocrine Pancreatic Insufficiency in CF Patients Not Fully Compensated with Only PPEs

This study was the first experience with MS1819 in CF patients not fully compensated with commercially available porcine PERTs. The study began in June 2019 and all patients completed the study by June 2021. This was a multicenter study with 4 study centers located in Hungary and 6 study centers located in Turkey that screened at least one subject.

The primary objectives of the study were to assess the safety, tolerability and efficacy of escalating doses of MS1819 on top of a stable dose of PERTs in patients with severe EPI caused by CF. A total of at least 20 patients were planned to complete the study.

A total of 62 patients were screened, 21 enrolled and 20 completed the study. There was one withdrawal due to COVID restrictions. One subject was excluded from the efficacy analysis due to a major protocol deviation (stool samples not collected properly).

For the primary endpoint, in the FAS population, mean CFA significantly increased by 5.67 (p-value = 0.0323) from baseline to Visit 4 (end of the 2-week cycle on MS1819- SD 700 mg/day); increased by 4.85 (p-value = 0.122) from baseline to Visit 5 (end of the 2-week cycle on MS1819-SD 1120 mg/day); and increased by 3.57 (p-value = 0.3653) from baseline to Visit 6 (end of the 2-week cycle on MS1819-SD 2240 mg/day). In the PP population, mean CFA significantly increased by 5.99 (p-value = 0.0316) from baseline to Visit 4; increased by 5.89 (p-value = 0.0513) from baseline to Visit 5; and increased by 5.66 (p-value = 0.1147) from baseline to Visit 6

In the full analysis set (N=20) the mean %CFA at baseline was 67.3 and mean increase in %CFA at Visit 4, visit 5 and visit 6 were 5.7, 6.6 and 5.0 respectively. The average improvement in CFA over baseline was 5.7 percentage points. These results were supported by secondary endpoints including improvements in number of stools and stool weight per day, body weight and BMI. A mean increase in body weight of 1.7kg (~4 lbs) over 45 days was observed with the addition of adrulipase. Only descriptive statistical testing was performed for these parameters.

MS1819 was well-tolerated, with no remarkable adverse events and laboratory values were unremarkable during the study.

6.7 Rationale and Dose Justification

Past *in vitro* studies have shown adrulipase to be relatively acid stable when compared to porcine derived lipases (see Investigator Brochure). However, it is also evident that below pH 4.0, there is increasing inactivation of adrulipase as the pH drops. Work by Gelfond, et al (2013), has shown that in CF patients, the pH in the upper duodenum may be as low as 3.5 for a short period, and the pH in the stomach could be even lower. The gastric emptying time of liquid or solid food may range from 1h-4h or longer and without any gastroprotection adrulipase is likely to have been exposed to low pH in patients with or without gastric acid suppression. Given that in the OPTION study, mean CFA with a dose of 2240 mg/day was 56%, a level below our target of 80%, it was deemed important to provide more protection of adrulipase against acid inactivation in the stomach and upper duodenum to provide adequate adrulipase dose and optimal lipolytic activity in the duodenum. AZ-CF2002 evaluated adrulipase in an enteric capsule, but the results were still below our target of 80%. This was likely due to the inconsistent gastric emptying of the size 0 enteric capsules resulting in adrulipase not being available to mix with and digest food.

Accordingly, for this study, adrulipase as an enteric microgranule formulation in size 00 IR capsules will be employed. Intrinsically, the enteric microgranule formulation is designed to not only provide adrulipase protection against degradation in the gastric environment but also allows for mixing with the food in the stomach and emptying along with the food in the duodenum. The dissolution characteristics of the adrulipase enteric microgranules show lipase availability at pH 5.5 and above, and <15% lipase release below pH 3 i.e., adequate gastroprotection. This matches nicely with our peak enzyme activity at pH 6 and allows rapid release in the duodenum where digestion is taking place.

The use of mass (g) for dose expression, rather than USP lipase units is the result of the biochemical difference between porcine pancreatic-derived lipase and the microbial-derived lipase from the yeast Yarrowia lipolytica (Aloulou et al, 2015). Porcine lipase is active at pH >7, while peak yeast-derived lipase activity occurs at pH 6 and has no activity at pH 9. The USP assay for porcine lipase units is conducted using an olive oil substrate at pH 9, while the enzymatic activity for adrulipase is assayed using tributyrin as substrate at pH 6. At pH 6 it

is difficult to assess the true lipolytic activity of adrulipase using olive oil as a substrate given the high pKa of oleic acid (the main fatty acid component of olive oil), it is not possible to monitor the lipolysis directly as fatty acid will remain in a protonated form and the assay requires the acid form. No validated conversion factor is available for conversion of USP lipase units to tributyrin units. It has been suggested by the FDA for microbial-derived lipase products that mass rather than units be used for dose expression (Heubi et al, 2016).

In all previous adrulipase studies the mass referred to the mass of the lipase:maltodextrin spray dried complex. Moving forward we will be referring to the actual mass of lipase in the capsule to directly communicate the mass of adrulipase (g) in the capsule. For reference to past studies, the low dose in Part 1 of the present study (1.2 g adrulipase/day) corresponds to the 2 g lipase:maltodextrin /day dose. The medium dose (1.95 g adrulipase/day) corresponds to the 6 g lipase:maltodextrin /day. The high dose (2.4 g adrulipase/day) corresponds to an 8 g lipase:maltodextrin /day. There were no safety concerns with the 1.2 g or 1.95 g adrulipase/day doses used in prior studies. While the 2.4 g adrulipase/day dose was not used in prior studies, it is expected to be well tolerated.

The highest dose possible in Part 1 of this study is 2.4 g adrulipase/day (equivalent to 8 g lipase:maltodextrin complex/day). As of the authoring of protocol version 2.0, six patients are enrolled in FWB-CF-2.03 under the Part 1 study design. Five of the 6 patients have been dosed at 2.4 g adrulipase/day, and there have been no AEs reported. Therefore, it is anticipated that the 2.4 g/day dosing will continue to be well-tolerated.

In Part 2, in keeping with past incremental increases in dose, a dose of 3.3 g adrulipase/day (equivalent to 10 g lipase:maltodextrin/day) is used. While the 3.3 g adrulipase/day dose has not been used in prior studies, it is expected to be well tolerated based on adrulipase's safety profile in the current study (FWB-CF-2.03) and prior clinical studies to date (discussed in Section 6.6). This higher dose requires two more capsules per meal, but the same number of capsules per snack.

7 STUDY OBJECTIVES

7.1 Primary Objectives

The Primary objective of this pilot study is to establish safety and efficacy of a new enteric microgranule formulation of adrulipase in patients with EPI due to CF.

The primary safety objective of this study is to assess the safety and tolerability of the highest dose received of adrulipase (for subjects under Part 1, 1.2 g/day, 1.95 g/day or 2.4 g/day; for subjects under Part 2, 3.3 g/day) provided in size 00 capsules containing ~150 mg adrulipase. Efficacy will be evaluated by comparing CFA values during treatment with the highest dose of adrulipase versus baseline CFA obtained during screening of the patient's pre-study porcine PERT.

8 INVESTIGATIONAL PLAN

8.1 Overall Study Design and Plan: Description

This is a Phase 2, single arm pilot study assessing the safety and efficacy of adrulipase in an enteric microgranule formulation. Patients with a confirmed diagnosis of cystic fibrosis who are 18 years of age or greater will be screened for eligibility if they have been clinically controlled, including minimal signs and symptoms of EPI, on a stable dose of commercial pancreatic enzyme replacement therapy (PERT) for at least one month.

Patients on cystic fibrosis transmembrane conductance regulator (CFTR) modulator therapies must have been on a stable dose for at least 3 months prior to study entry, and no dose changes will be made during the study. Patients receiving gastric acid suppressants must have been on a stable dose for at least one month prior to study entry and no dose changes will be made during the study.

Upon obtaining an informed consent, potentially eligible patients will receive dietary counseling during the week prior to confinement for collecting stool samples for calculation of baseline coefficient of fat absorption (CFA). This counseling will emphasize the importance of dietary stability during the study. Patients found to have a CFA of 80% or greater while receiving their commercial PERT and meeting the other eligibility criteria will be enrolled into the study.

The study will enroll patients into either Part 1 (under protocol version 1.0) or Part 2 (under protocol version 2.0). Upon study enrollment, the patient will be switched from their commercial PERT to receive adrulipase. The patient will remain on study for approximately three weeks for Part 1 patients or two weeks for Part 2 patients, after which a repeat CFA will be obtained under supervised confinement. In special circumstances, upon discussion with First Wave BioPharma, the end of study supervised confinement may be done prior to the two or three weeks on adrulipase.

For Part 1, a dose titration scheme will be used for determining whether a low, medium, or high dose of adrulipase can succeed in controlling signs and symptoms of exocrine pancreatic insufficiency (EPI) and provide a CFA of 80% or greater. Patients will initially receive a low dose of adrulipase. Upon the appearance of EPI symptoms for three or more days, and upon discussion with the investigator, the patient will be switched to the medium dose of adrulipase. If signs and symptoms of EPI persist for three or more days, the patient will be switched to the high dose of adrulipase. After patients reach three weeks of study and complete their end of study CFA, they will be returned to their pre-study commercial PERT preparation therapy.

For Part 2, a dose of 3.3 g/day adrulipase will be used for all patients. After patients reach two weeks of study treatment and complete their end of study CFA, they will be returned to their pre-study commercial PERT. An end of study safety visit will be scheduled to occur one week after finishing adrulipase therapy.

An end of study safety visit will be scheduled for approximately one week after finishing adrulipase therapy.

Safety assessments will be made by collecting adverse events, safety lab assessments, and immunologic assays to assess drug induced immune responses.

For Part 1, dosages will ideally be fractionated as follows: 1/4 of the daily dose at each of 3 main meals, and 1/8 at each of the 2 snacks. The patients will be instructed to consume ~25 g of fat with every main meal and 12.5 g of fat with each of the 2 snacks to approximate a high fat meal of 100 g of fat/day. The 1.2 g/day dose will be fractionated as follows: 2 capsules of 150 mg with the morning, noontime, and evening meals, plus 1 capsule of 150 mg with the morning and evening snacks. The 1.95 g/day dose will be fractionated as follows: 3 capsules with each of the three main meals, and 2 capsules with each of the two snacks. The 2.4 g/day dose will be fractionated as follows: 4 capsules with each of the three main meals and 2 capsules with each of the two snacks.

For Part 2, The 3.3 g/day dose will be fractionated as follows: 6 capsules with each of the three main meals, and 2 capsules with each of the two snacks.

Individual variations may occur if patients have discussed the variation with their PI or care team and total daily dose is achieved.

This study is divided into 3 main periods.

- Screening Period including supervised confinement (up to 14 days)
- Adrulipase Treatment Period including supervised confinement (approximately 21 days for Part 1 patients or 14 days for Part 2 patients)
- End-of-Study/Early Withdrawal (approximately 7 days)

Refer to Section 3 for the Schedule of Assessments.

8.1.1 Amended Study Design

For analysis purposes, this amended portion of the study will be referred to as Part 2. Refer to Section 3 for the amended SoA for Part 2.

Patients who begin on Part 2 of the protocol will receive a higher dose (3.3 g/day) for treatment. Patient safety data for adrulipase at lower doses (ranging from 1.2 g/day to 1.95 g/day) supports it as safe and well-tolerated (Section 6.7). Therefore, patients will not undergo titration in Part 2, thus treatment is two weeks rather than three weeks.

8.2 Study Endpoints

8.2.1 Primary Efficacy Endpoints

Efficacy will be assessed by comparing CFA values during treatment with adrulipase versus baseline CFA obtained during screening of the patients pre-study porcine PERT, using descriptive methods.

8.2.2 Secondary Efficacy Endpoints

The secondary efficacy endpoints of the study include:

- A comparison of 72-hour stool weights, adrulipase vs porcine PERT
- Signs and symptoms of malabsorption
    - Stool frequency (number of bowel movements per day)
    - Stool consistency (graded as 0=hard, 1=formed/normal, 2=soft, 3=watery, or 4=overt diarrhea)
    - Bloating (graded as 0=none, 1=mild, 2=moderate, 3=severe)
    - Abdominal pain (graded as 0=none, 1=mild, 2=moderate, or 3=severe)
    - Flatulence (graded as 0=none, 1=mild, 2=moderate, or 3=severe)
    - Incidence of visible oil/grease in stool (Yes/No)
    - Increased stool quantity (graded as 0=none, 1=mild, 2=moderate, or 3=severe)
    - Worsening of overall bowel habit (graded as 0=none, 1=mild, 2=moderate, or 3=severe)
- CNA
- Body weight
- Body mass index

8.2.3 Safety Endpoints

AEs, SAEs, safety lab values, and discontinuations due to adverse events.

In addition, laboratory test results will be summarized (Section 16.2):

- Hematology
- Biochemistry
- Fasting lipid profile
- Urinalysis
- Serum adrulipase concentrations
- Antibodies against adrulipase

8.3 Discussion of Study Design

8.3.1 Number of Planned Patients

Approximately 12 patients are planned to complete the study.

The statistical considerations on which the planned number of patients is based are described in Section 11.

8.3.2 Inclusion Criteria

To be eligible for study entry, patients must satisfy all of the following inclusion criteria:

1. Signed and dated informed consent form by patient as required by First Wave BioPharma or designee and approved by an Institutional Review Board/Independent Ethics Committee (IRB/IEC)
2. Age ≥18 years at the time of screening
3. Male or female
4. Confirmed diagnosis of cystic fibrosis, based on 2 clinical features consistent with CF, plus either a new/historic sweat chloride ≥60 mmol/L by quantitative pilocarpine iontophoresis (measured while not on a CFTR modulator) or genotype
5. On stable dose of porcine PERT ≥1 month (30 days) prior to screening; stable dose is defined as dose of medication not changed during this time period, and the medication must be commercially available and be administered in the recommended dose range.
6. CFA = or > 80% at screening while on stable PERT
7. Normal nutritional status as defined by BMI ≥ 18.5 kg/M2 for patients ≥ 18 years of age.
8. Fecal elastase <100 µg/g of stool at screening
9. Clinically stable with no documented evidence of acute respiratory symptoms that would require administration of new oral or intravenous antibiotics, oxygen supplementation, or hospitalization within 30 days of screening or during the screening period.
10. Male and Female patients, if of childbearing potential, must use a reliable method of contraception during the study. A reliable method of birth control is defined as one of the following: oral or injectable contraceptives, intrauterine device, contraceptive implants, tubal ligation, hysterectomy, or a double-barrier method (diaphragm with spermicidal foam or jelly, or a condom), abstinence or vasectomy. Periodic abstinence (calendar, symptothermal, or post-ovulation methods) is not an acceptable method of contraception. The preferred and usual lifestyle of the patient must also be evaluated in determining if sexual abstinence is a reliable method of birth control.
11. Be considered reliable and capable of adhering to the protocol, according to the judgment of the Investigator

8.3.3 Exclusion Criteria

Patients will be excluded from the study if one or more of the following criteria are applicable:

1. History or diagnosis of fibrosing colonopathy
2. Total or partial gastrectomy
3. A history of solid organ transplant or significant surgical resection of the bowel; significant resection of the bowel is defined as any resection of the terminal ileum or ileocecal valve. Patients who have had qualitative, long-term changes in nutritional status after any other bowel resection (e.g., increased, or new need for pancreatic enzyme supplementation compared with preoperative status to maintain the same nutritional status) should also be excluded.
4. Any chronic diarrheal illness unrelated to pancreatic insufficiency (e.g., infectious gastroenteritis, sprue, inflammatory bowel disease)
5. Known hypersensitivity or other severe reaction to any ingredient of the investigational medicinal product (IMP)
6. Alanine aminotransferase (ALT) or aspartate aminotransferase (AST) level ≥3 ×upper limit of normal (ULN), or total bilirubin level ≥2.2 mg/dl at the Screening visit, unless in the opinion of the investigator this does not present a safety issue based on the patient's clinical status. Cases of suspected or confirmed Gilbert's syndrome should be discussed with the Medical Monitor.
7. Signs and/or symptoms of liver cirrhosis or portal hypertension (e.g., splenomegaly, ascites, esophageal varices), or documented liver disease unrelated to CF
8. Patients with a known allergy to the stool marker (FD&C Blue #2)
9. Feeding via an enteral tube during 6 months before screening
10. Routine use of anti-diarrheals, anti-spasmodics, or cathartic laxatives, or a change in chronic osmotic laxatives (e.g., polyethylene glycol) regimen in the previous 3 months prior to screening
11. History of severe constipation with <1 evacuation/week under appropriate laxative therapy within the last 12 months before screening
12. Documentation of distal intestinal pseudo-obstruction syndrome within the last 12 months before screening
13. Forced expiratory volume ≤30% at the screening visit
14. Lactation or known pregnancy or positive pregnancy test at screening for women of childbearing potential
15. Participation in another clinical study involving an investigational product within 30 days prior to screening for this study.
16. Patient's with poorly controlled diabetes according to the Investigator's judgment

17. Changes in CFTR modulator therapy during the 3 months prior to screening, for patients already on CFTR modulator therapy.
18. Changes in gastric acid suppressant therapy during the one month prior to screening for patients already on suppressant therapy.
19. Any condition that the Investigator believes would interfere with the intent of this study or would make participation not in the best interests of the patient.

8.3.4 Removal of Patients from the Study

A patient may be withdrawn from the study at any time for any of the following reasons:

- Lost to follow-up
- Voluntary withdrawal of consent by the patient, for any reason, at any time
- Major protocol deviations that could compromise the interpretation of the study results
- Occurrence of an immunoallergic reaction
- Occurrence of an adverse reaction (i.e., an SAE related to IMP) that justifies the discontinuation of the IMP
- Pregnancy

The date of withdrawal and the reason for withdrawal must be fully documented in the case report form (CRF) and a short narrative description should be added in the patient's medical records.

If at the time of withdrawal, the patient has received the investigational study product (partially or totally), study staff should encourage the patient to complete the Early Termination Visit (Visit 7) for follow-up safety investigations. These patients will be included in the mITT efficacy and safety analyses.

If the reason for discontinuation is due to an AE, then the Investigator will seek to obtain follow-up information and to document the event until its resolution or stabilization. In any case, the Investigator will take all necessary measures to ensure the patient's safety and ensure the patient is treated in accordance with local standard of care.

If a patient is lost to follow-up, then the Investigator should make every effort to obtain maximum information on the reasons for the nonattendance to the visit and on the patient's state of health. All attempts will be documented in the patient's medical records.

Pregnancy

Patients will be instructed that known or suspected pregnancy occurring during the study, in patients or female partners of male patients, should be confirmed and immediately reported to

the Investigator. Any patient who becomes pregnant during the study must be promptly withdrawn from the study. Upon discontinuation from the study, only those procedures that would not expose the patient to undue risk will be performed. The Investigator should also be notified of pregnancy occurring during the study but confirmed after completion of the study. If a patient is subsequently found to be pregnant after inclusion in the study, any pregnancy will be followed to term, and the status of mother and child will be reported to First Wave BioPharma after delivery.

Full details will be recorded in the CRF, or an SAE report will be completed if the patient has completed the study.

8.4 Investigational Products Administered

8.4.1 Adrulipase Administration

At the time of prescribing, the Investigator or delegated site staff will fill in a source document with the appropriate dosing instructions.

For Part 1, dosages will typically be fractionated as follows: 1/4 of the daily dose at each of 3 main meals, and 1/8 at each of the 2 snacks. The patients will be instructed to consume ~25 g of fat with every main meal and 12.5 g of fat with each of the 2 snacks to approximate a high fat meal of 100 g of fat/day. The 1.2 g/day dose will be fractionated as follows: 2 capsules of 150 mg with the morning, noontime, and evening meals, plus 1 capsule of 150 mg with the morning and evening snacks. The 1.95 g/day dose will be fractionated as follows: 3 capsules with each of the three main meals, and 2 capsules with each of the two snacks. The 2.4 g/day dose will be fractionated as follows: 4 capsules with each of the three main meals, and 2 capsules with each of the two snacks.

For Part 2, The 3.3 g/day dose will be fractionated as follows: 6 capsules with each of the three main meals, and 2 capsules with each of the two snacks.

Individual variations may occur if patient has discussed the variation with their PI or care team and total daily dose is achieved.

Please refer to Table 2 for dosing guidance.

Table 3: Adrulipase Dosing Schedule

	Part 1 Dosing Schedules			**Part 2 Dosing Schedule**
	Dose: 1.2 g/day	**Dose: 1.95 g/day**	**Dose: 2.4 g/day**	**Dose: 3.3 g/day**
Breakfast	2 x 150 mg capsules	3 x 150 mg capsules	4 x 150 mg capsules	6 x 150 mg capsules
Snack 1	1 x 150 mg capsule	2 x 150mg capsule	2 x 150mg capsule	2 x 150mg capsule

	Part 1 Dosing Schedules			Part 2 Dosing Schedule
	Dose: 1.2 g/day	**Dose: 1.95 g/day**	**Dose: 2.4 g/day**	**Dose: 3.3 g/day**
Lunch	2 x 150 mg capsules	3 x 150 mg capsules	4 x 150 mg capsules	6 x 150 mg capsules
Snack 2	1 x 150 mg capsule	2 x 150mg capsule	2 x 150mg capsule	2 x 150mg capsule
Dinner	2 x 150 mg capsules	3 x 150 mg capsules	4 x 150 mg capsules	6 x 150 mg capsules
Total	**8 capsules/day**	**13 capsules/day**	**16 capsules/day**	**22 capsules/day**

Note: Individual variations may occur if patient has discussed the variation with their PI or care team and correct total daily dose is achieved.

8.4.2 Method of Assigning Patients to Adrulipase Treatment

For Part 1, all eligible patients will first start on the low dose (1.2 g/day) and optimize to medium or high dose if required.

For Part 2, all eligible patients will be assigned to adrulipase 3.3 g/day dose.

8.5 Previous and Concomitant Therapy

Standard-of-care medications are allowed (e.g., antibiotics, mucolytic agents, aerosols, CFTR modulators). Patients taking CFTR modulators should be on stable doses of the same modulator(s) for at least 3 months prior to screening. Patients should not start taking, or modify dose of, CFTR modulators for the duration of the study.

Gastric acid suppressants are allowed but must be on stable dosage of the same suppressant for 30 days before screening and must not be altered in dose or stopped during the study.

Prohibited medications during the entire clinical study will be as follows:

- Orlistat lipase inhibitor (e.g., Alli®, Xenical®)
- Laxatives consisting of mineral oil and castor oil; chronic use of osmotic laxatives is permitted
- Symptomatic treatments of diarrhea: loperamide (e.g., loperamide generic, Imodium®, Imodium A-D®, Diamode®, Imotil®, Kao-Paverin®); atropine/diphenoxylate (Lonox®); and atropine/diphenoxylate (Lomocot®).

8.6 Study Drug Materials and Management

8.6.1 Adrulipase

Study drug Adrulipase will be supplied by First Wave BioPharma for use in the protocol and is limited to investigational use only. Please refer to current Investigator's Brochure for additional information.

Adrulipase is a ~70% pure preparation of LIP2 protein. The concentrated ultra-filtration product obtained from fermentation is mixed with maltodextrin and a generally regarded as safe enteric polymer then subjected to spray drying to produce the drug substance. The drug product is formulated with inactive excipients and formulated in non-animal origin HPMC capsules. Adrulipase will be supplied as 150 mg, size 00 (double zero) capsules.

Adrulipase will be provided in appropriately labeled HDPE bottles.

8.6.1.1 Adrulipase Shipment, Receipt, and Storage

Adrulipase will be supplied in bottles to the sites by First Wave BioPharma, in accordance with local requirements. The IMP will be shipped under controlled room temperature condition. The site is responsible for the appropriate storage of the IMP. The IMP must be stored in a secured limited-access area and maintained at controlled room temperature. Controlled room temperature is the temperature maintained thermostatically that encompasses the usual and customary working environment of 20°-25° (68°-77 °F). Excursions between 15° and 30° (59° and 86 °F) that are experienced in pharmacies, hospitals, and warehouses, and during shipping are allowed.

8.6.1.2 Adrulipase Misuse/Overdose

Any IMP misuse or overdose associated or not associated with any AE should be reported to First Wave BioPharma, or designee, as an SAE only if it meets the criteria of an SAE. Overdose is considered as dose taken above the prescribed daily dose for the current dosing phase.

8.6.1.3 Adrulipase and porcine PERT Compliance

The Investigator is responsible for ensuring compliance. Compliance with adrulipase will be cross-checked with study drug accountability and compliance with pre-study porcine PERT will be evaluated by subject self-report.

8.6.1.4 Adrulipase Supply, Resupply, and Accountability

Upon initial receipt of IMP at the investigational site, the pharmacist or delegated site staff will record the date, details of the bottle, and quantity of capsules.

The investigational sites will be re-supplied with IMPs according to their respective recruitment rates.

The pharmacist or delegated site staff will be provided with specific forms for accountability of the IMP (including the returned bottles). Records will be kept up to date throughout the study and must be complete and accurate.

Used and unused IMPs must be made available to the Monitor or First Wave BioPharma designee who will verify the IMP accountability and cross-check pharmacy and Investigator records for compliance to the protocol requirements. Any discrepancy must be fully accounted for and documented.

8.6.1.5 Adrulipase Return, Destruction, and Recall

Return of IMP:

- Unused, partially used bottles will be returned by the patient to the investigational site.
- At the end of the study, the Monitor or First Wave BioPharma will conduct a final reconciliation between delivered, dispensed, and used/unused IMPs.

Destruction of IMP:

- Unused, partially used bottles must *not* be destroyed at the Investigative Sites without written authorization from First Wave BioPharma.
- Unused, partially used, or empty bottles returned to the pharmacy will be destroyed at the Hospital Pharmacy or First Wave BioPharma's Drug Distributor only after IMP accountability forms have been fully and accurately completed and verified by the Monitor.
- If an on-site destruction is requested, then the investigational site must provide procedural documentation and obtain written authorization for destruction from First Wave BioPharma, which will be filed along with the certificate of destruction in the IMP section of the pharmacy site file.

Recall:

- If an IMP batch is suspected to be defective, then First Wave BioPharma will immediately inform the Investigator and the hospital pharmacist.
- The Monitor will coordinate with the investigative site staff for the return of the concerned batches as per the return procedure. Depending on the study status, new batches may be sent to the investigational site.

8.6.2 Nonabsorbable Dye Marker

The dye marker FD&C Blue #2, also named Indigo carmine or Indigotin, will be supplied in accordance with current Good Manufacturing Practices (cGMP).

8.6.2.1 Shipment, Receipt, and Storage and Dispensing

The dye makers must be stored in a secured limited-access area and maintained at controlled room temperature ≤ 25°C and will not require temperature monitoring during shipment.

9 TIMING OF STUDY PROCEDURES

Patients will provide written informed consent before any study-related procedures are performed.

9.1 The study Schedule of Assessments for Part 1 and Part 2 are included in Section 3.Screening Period

Throughout the screening period, patients are to remain on their prestudy porcine PERT.

9.1.1 Visit 1, D-42 to D-14: Screening Visit

The following procedures will be performed at the Screening Visit (which may take place over more than one day):

- Obtain signed informed consent from the patient before any study-related assessments are made
- Collect relevant medical history, including concomitant illnesses/diseases, previous/concomitant medications, and record AEs
- Full physical exam
- Specific CF assessment including a pulmonary function test by spirometry to determine FEV1 >30% of predicted normal for age, sex, and height at screening
- Record body height and weight
- Record vital signs while sitting (blood pressure, pulse rate, breathing rate, and temperature)
- Assess for eligibility (against the inclusion and exclusion criteria)
- Record demographic data such as ethnic origin, date of birth, and sex
- Collect urine sample for urinalysis
- Perform a serum pregnancy test, if applicable
- Collect samples for hematology, clinical chemistry, PT/INR, activated partial thromboplastin time (aPTT)
- Patients will be required to fast for at least 8 hours before the collection of screening laboratory tests (insulin dependent diabetics may be excluded from fasting).
- Collect samples for fasting lipids

- Obtain a stool sample for fecal pancreatic elastase concentration in the selected central laboratory

9.1.2 Visit 2, D-7 to D-21: Stool Collection in Controlled Supervised Facility

In advance of Visit 2, patients will be contacted by telephone and instructed to:

- Refrain from alcohol consumption for 24 hours before the visit
- Bring their prestudy porcine PERT with them to the clinic

Patients will be admitted for a maximum of 7 days to a facility providing supervised confinement for collection of stools for determination of CFA, CNA, and stool weight. The timing of the previous visit (Visit 1) should be made with the aim of accommodating the patients' schedules for the supervised confinement. To accommodate patients' schedules for the supervised confinement, a +7-day visit window is permitted.

- Patients will undergo admission to a controlled supervised facility for stool collection to support the CFA and CNA assessment and weight. A +7-day visit window is allowed to accommodate investigative site staff and patients' schedules for the supervised confinement.
- After appearance of fecal stool dye markers has been measured, patients CFA and CNA will be measured under their prestudy dose of porcine PERT using standardized high-fat meals during the 3-day standardized diet. Additional detailed information provided in the study operations manual should be followed.
- The CFA calculation will be based on the measured fecal fat content in relation to ingested fat quantities during the 3-day stool collection period.

The stool collection and dietary procedures will be conducted as described in Section 9.2.4.1. (Instructions for Supervised Confinement).

The following procedures will be performed at Visit 2:

- Review Inclusion /Exclusion Criteria
- Begin 72-hour controlled diet
- Focused physical exam to evaluate gastrointestinal tract, heart, and lungs
- Record body height and weight
- Record vital signs while sitting (blood pressure, pulse rate, breathing rate, and temperature)
- Record any changes in AEs and concomitant medication
- Instruct/dispense study drug

- Assess and record malabsorption signs and symptoms
- Marker-to-marker stool collection and stool weight (central laboratory will weigh the stool that is collected)
- Collect a serum sample for circulating anti-adrulipase antibodies and adrulipase concentrations
- Record all fat, protein, and dose of study drug taken with every meal and snack during the confinement period
- Verify study drug count on the last day of confinement

At the end of the supervised confinement (once the second blue dye marker sample has been collected), patients will remain on their prestudy porcine pert treatment. Patients will be contacted once the CFA results are available and if the patient is eligible to participate in the study visit 3 will be scheduled.

9.2 Treatment Period (Visits 3, 4 (T), 5 (T), and 6)

9.2.1 Visit 3 (Day 1): Adrulipase Dispensation Visit - Must Occur Within 21 Days of Visit 1

During the treatment period, patients will be randomized to receive adrulipase. Visit 3 will take place if the patient is eligible to continue based on data obtained at the Screening visit. The following procedures will be performed:

- Review Inclusion /Exclusion Criteria
- Perform a urine pregnancy test (urine dip stick), if applicable
- Assess and record malabsorption signs and symptoms
- Record any changes in AEs and concomitant medication
- Begin dose of adrulipase (For Part 1 subjects, 1.2 g/day; for Part 2 subjects, 3.3 g/day) in-clinic (optional if patient plans to eat immediately after the visit)
- Instruct patients on use and dosing (including patients' specific dose-titration for Part 1 patients).
- Instruct/dispense study drug to patient

9.2.2 Visit 4 (Telephone) (Part 1 Day 5 ±2 or Part 2 Day 3 ± 2) and Visit 5 (Telephone) (Part 1 Day 10 ± 2 or Part 2 Day 7 ± 2)

The following procedures will be performed during a telephone visit:

- Record any changes in AEs and concomitant medication review.
- Assess and record malabsorption signs and symptoms
- Confirm patient scheduled date for Visit 6 (second stool collection in a controlled supervised facility) and remind patients to arrive in a fasted status.
- PI and/or site staff may contact patient more frequently to enable patient-specific dose optimization (for Part 1 patients) as required
- Patient will remain on their optimized dose of adrulipase till visit 6
- Patients will be instructed to bring their adrulipase (all bottles dispensed, including full, partial or empty bottles) and prestudy porcine PERT with them to their next in-clinic visit.

9.2.3 Visit 6: Stool Collection in Controlled Supervised Facility (Part 1 Day 16 + 5 or Part 2 Day 10+3)

Patients will be admitted for a maximum of 7 days to a facility providing supervised confinement for collection of stools for determination of CFA, CNA, and stool weight. The timing of the previous visits should be made with the aim of accommodating the patients' schedules for the supervised confinement. To accommodate patients' schedules for the supervised confinement, a ±5 day visit window is permitted.

- On Day 16 + 5 days (Part 1) or Day 10 + 3 (Part 2) (Visit 6), patients will undergo admission to a controlled supervised facility for stool collection to support the CFA and CNA assessment and weight. A +5-day (Part 1) or +3-day (Part 2) visit window is allowed to accommodate investigative site staff and patients' schedules for the supervised confinement.
- After appearance of fecal stool dye markers has been measured, patients CFA and CNA will be measured under their dose of adrulipase using standardized high-fat meals during the 3-day standardized diet. Additional detailed information provided in the study operations manual should be followed.
- The CFA calculation will be based on the measured fecal fat content in relation to ingested fat quantities during the 3-day stool collection period.

After completion of the initial treatment period during the completion of V6, patients will be instructed to switch their study medication to their prestudy porcine PERT. The stool collection and dietary procedures will be conducted as described in Section 9.2.4.1. (Instructions for Supervised Confinement)

The following procedures will be performed at Visit 6:

- Begin 72-hour controlled diet

- Focused physical exam to evaluate gastrointestinal tract, heart, and lungs
- Record body height and weight
- Record vital signs while sitting (blood pressure, pulse rate, breathing rate, and temperature)
- Record any changes in AEs and concomitant medication
- Instruct/dispense study drug for confinement period
- Assess and record malabsorption signs and symptoms
- Marker-to-marker stool collection and stool weight (central laboratory will weigh the stool that is collected)
- Collect urine sample for urinalysis
- Perform a urine pregnancy test (urine dip stick), if applicable
- Collect samples for hematology, clinical chemistry, PT/INR, and aPTT
- Collect samples for fasting lipids after patients have been in a fasting status for at least 8 hours (insulin dependent diabetics may be excluded from fasting).
- Record all fat, protein, and dose of study drug taken with every meal and snack during the confinement period
- Verify study drug count on the last day of confinement
- Return adrulipase on the last day of confinement

At the end of the supervised confinement (once the second blue dye marker sample has been collected), patients will return to their prestudy porcine PERT dose.

9.2.4 Visit 7: End-of-Study /Early Termination Visit (Part 1 Day 28 ± 2 or Part 2 Day 21 ± 2)

Patients will return to the clinic for their End of Study visit approximately 1 week after their last dose of study drug. If patients do not complete Visit 6, an Early Termination visit will be held approximately 1 week after the last dose of study drug. The procedures scheduled for the End-of-Study visit are the same as those that should be conducted for the Early Termination visit. The following procedures will be performed at the End-of-Study/Early Termination Visit:

- Focused physical exam to evaluate gastrointestinal tract, heart, and lungs
- Record any changes in AEs and concomitant medication
- Assess and record malabsorption signs and symptoms
- Collect a serum sample for circulating anti-adrulipase antibodies and adrulipase concentrations

- Perform a urine pregnancy test (urine dip stick), if applicable

9.2.4.1 Instructions for Supervised Confinement for V2 and V6

Dosing must have occurred for at least 16 days (Part 1) or 10 days (Part 2) prior to the supervised confinements. Beginning at breakfast (the first meal) on Day 1 of the supervised confinement, patients will be placed on a 72-hour controlled diet, with ~25 g fat/meal and 12.5 g fat/snack for a total of 100 g fat/day (3 meals and 2 snacks per day) and a minimum of 1.5 to 2 g of protein per kg of body weight. Although it is preferred to have the patient consume exactly 100 g of fat per day, the fat excretion assay remains valid with a fat intake range within 15% of the goal (i.e., 85 to 115 g of fat per day). The nutritional planning and calculation of dietary fat and nitrogen (protein) intake will be conducted by a qualified dietician at each site. The actual fat and nitrogen (protein) intake will be calculated from the recorded type and amount of food consumed. A central laboratory will calculate CFA, CNA based on dietary intake and the fat and nitrogen excreted in stool. Stool weight values will also be recorded.

An indicator marker (500 mg of Food, Drug, and Cosmetics [FD&C] Blue #2) will be provided as two 250-mg capsules. The first marker (consisting of two 250-mg capsules) will be given at the beginning of breakfast or first meal on Day 1 of the supervised confinement to mark the start of the controlled diet. The second marker (two 250-mg capsules) will be given at the beginning of breakfast or first meal on Day 4 to mark the end of the controlled diet. Stool collection for fecal fat and nitrogen assessments must begin after the first marker has passed (the first stool containing the first marker is discarded) and is completed when the second marker has passed (the first stool containing the second marker is collected). Patients must continue the supervised confinement until the second stool marker is passed. The controlled diet will be maintained until the patient takes the second marker. Although it is not part of the controlled diet, the breakfast or first meal consumed on Day 4 of the supervised confinement should mimic (as closely as possible) the breakfast or first meal consumed on Day 1 of the supervised confinement.

If the patient has not passed the first blue dye marker within 4 days after ingesting the first dye marker capsules, or within 2 days after ingesting the second dye marker capsules, then 5-10 mg of oral bisacodyl may be given. Bisacodyl is the only laxative that may be given during the marker-to-marker stool collection period. Note: Patients on a stable chronic osmotic laxative may continue this therapy during the marker-to-marker stool collection.

The supervised confinement may take place within an inpatient facility or within an alternative confinement setting. This will be discussed and approved on an individual case basis by First Wave BioPharma. The alternative confinement setting will require the supervision of trained study site staff to oversee the study procedures.

10 SAFETY ASSESSMENTS

The planned Schedule of Assessments is included in Section 3.

10.1 Definitions

10.1.1 Adverse Event

An AE is any untoward medical occurrence experienced by a patient in a clinical investigation; it does not necessarily have a causal relationship with this study drug. An AE can therefore be any unfavorable and unintended sign, symptom, or disease temporally associated with the use of a medicinal product, whether or not considered related to the medicinal product.

Active or worsening of EPI is not considered an AE for the purposes of this study. Worsening of EPI disease should not be recorded as an AE but should be recorded in the appropriate EPI CRF.

Adverse events may also include post dose complications that occur as a result of protocol-mandated procedures (e.g., invasive procedures such as venipuncture, biopsy, etc.). Preexisting events that increase in severity or change in nature during, or as a consequence of, use of a medicinal product in a human clinical study will also be considered AEs. Any preexisting medical condition or diagnosis associated with a clinically significant laboratory abnormality should be documented on the CRF or eCRF. The Investigator should attempt to establish a diagnosis of the event based on signs, symptoms, and other clinical information. In such cases, the unifying diagnosis should be documented as the AE, rather than the individual signs and symptoms (e.g., runny nose, scratch throat, cough, and low grade fever should be recorded as an upper respiratory infection and not each of the individual symptoms).

An AE does NOT include the following:

- Medical or surgical procedures (e.g., surgery, endoscopy, tooth extraction, transfusion); the condition that necessitates the procedure is an AE
- Any preexisting disease or condition or laboratory abnormality present or detected before the start of administration of study medication that does not worsen
- Laboratory abnormalities without clinical manifestations, which do not require medical intervention, or that do not result in termination or delay of study medication
- Situations where an untoward medical occurrence has not occurred (e.g., hospitalization for elective surgery, social, or convenience admissions)
- Overdose of any study drug or concomitant medication without any signs or symptoms, unless the patient is hospitalized for observation
- Worsening and/or flares of EPI disease activity. This should be recorded in the appropriate EPI CRF. However, if disease worsening meets any of the criteria for an SAE, it must be recorded on the SAE form and reported to First Wave BioPharma or designee within 24 hours of becoming aware of the event.

10.1.2 Serious Adverse Event

An SAE is defined as any adverse experience occurring at any dose of study medication that occurs between the time the patient signs the informed consent form through the end-of-study that results in any of the following outcomes:

- Death
- Life-threatening situation (patient is at immediate risk of death)
- In-patient hospitalization or prolongation of existing hospitalization for a clinically relevant reason (note that this excludes "social" hospitalization for nonmedical causes such as lack of transportation to home)
- Persistent or significant disability/incapacity
- Congenital anomaly/birth defect in the offspring of a patient who received study drug
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient or patient may require medical or surgical intervention to prevent one of the outcomes listed in this definition. Examples may include, but are not limited to:
    - Intensive treatment in an emergency room or at home for allergic bronchospasm
    - Blood dyscrasias that do not result in hospitalization
    - Seizures that do not result in hospitalization

A hospitalization meeting the regulatory definition for "serious" is any in-patient hospital admission that includes a minimum of an overnight stay in a health care facility.

Worsening and/or flares of EPI disease activity should be recorded in the appropriate EPI CRF. However, if disease worsening meets any of the criteria for an SAE, then it must be recorded on the SAE form and reported to First Wave BioPharma or designee within 24 hours of becoming aware of the event.

10.1.3 "Serious" vs "Severe" Adverse Event

To avoid confusion or misunderstanding over the difference between the terms "serious" and "severe," which are not synonymous, the following note of clarification is provided (excerpted from International Council for Harmonisation [ICH] E2A):

The term "severe" is often used to describe the intensity (severity) of a specific event (as in mild, moderate, or severe myocardial infarction); the event itself, however, may be of relatively minor medical significance (such as severe headache). This is not the same as "serious," which is based on patient/event outcome or action criteria usually associated with events that pose a

threat to a patient's life or functioning. Seriousness (not severity) serves as a guide for defining regulatory reporting obligations.

10.1.4 SAE Definition Clarifications

- Death is an outcome of an AE, and not an AE in itself.
- All deaths during study through the end-of-study, regardless of cause or relationship, must be reported.
- "Occurring at any dose" does not imply that the patient is actively receiving study drug at the time of the event.
- "Life-threatening" means that the patient was at immediate risk of death from the event as it occurred. This does not include an event that might have led to death, had it occurred with greater severity.
- Complications that occur during hospitalizations are AEs. If an AE prolongs hospitalization, it is an SAE.
- "In-patient hospitalization" means the patient has been formally admitted to a hospital for medical reasons, for any length of time, excepting situations where an untoward medical occurrence has not occurred (e.g., hospitalization for elective surgery, social, or convenience admissions). Hospitalization may or may not be overnight. It does not include presentation and care within an emergency department (although an emergency department visit may define a medically important event, which is also considered an SAE).
- If any AE worsens during the study and eventually meets the criteria for an SAE the AE should be recorded as a new SAE.

10.2 Reporting Procedures for All Adverse Events

AEs will be recorded in the CRF for all patients from the time the patient signs the informed consent form through the end of the study or patient withdrawal.

The Investigator is responsible for ensuring that all AEs (as defined in Section 10.1 and as further specified below) observed by the Investigator or reported by patients are collected and recorded in the patients' medical records, in the CRF, and as an SAE in the electronic data capture system. These AEs will include the following:

1. All SAEs (as defined in Section 10.1.2) that occur.
1. All nonserious AEs (as defined in Section 10.1.1) that occur.

The following AE attributes must be assigned by the Investigator:

- AE diagnosis or syndrome(s) (if known, signs or symptoms if not known);
- event description (with detail appropriate to the event);

- dates of onset and resolution; severity; assessment of relatedness to study drug; and
- action taken with study medication.

The Investigator may be asked to provide follow-up information, discharge summaries, and extracts from medical records or CRFs.

It will be left to the Investigator's clinical judgment to determine whether the relatedness of an AE, and of sufficient severity to require the patient's removal from treatment or from the study. A patient may also voluntarily withdraw from treatment because of what he or she perceives as an intolerable AE. If either of these situations arises, the patient should be strongly encouraged to complete the End-of-Therapy assessments and be under medical supervision until symptoms cease or the condition becomes stable.

10.3 Grading of Adverse Events

The Investigator will be asked to provide an assessment of the severity of the AE using the categories noted below. This assessment is subjective, and the Investigator should use medical judgment to compare the reported AE to similar events observed in clinical practice. It is important to recognize that severity is not equivalent to event seriousness.

Grade 1 (Mild): usually transient; requires no special treatment and does not generally interfere with the patient's daily activities.

Grade 2 (Moderate): produces a mild to moderate level of inconvenience to the patient and may interfere with daily activities. These events are usually ameliorated by simple therapeutic measures.

Grade 3 (Severe): significantly interrupts daily activity and requires systemic drug therapy or other medical treatment.

10.4 Relationship to Study Drug

For each reported AE, the Investigator must make an assessment of the relationship of the event to the study drug using the following scale:

Unrelated: The event is definitely not or unlikely associated with study drug administration and is judged because of causes other than the study drug.

Related: Events considered to be related are those that follow a reasonable temporal sequence from administration of the study drug, that are not easily explained by another cause such as known characteristics of the patient's clinical state or other treatment or confirmed by improvement on stopping or slowing administration of the study agent (de-challenge), if applicable.

10.5 Serious Adverse Event Reporting Procedures

Serious adverse events will be recorded from the time the patient signs the informed consent form through the end of study. AEs will be followed until the event resolves or stabilizes, or until the end of study for that patient.

If the AE is an SAE and is assessed as "related to study drug", it must be followed until either the event is considered stable or resolved.

Any SAE assessed as "unrelated to study drug" will be followed as clinically indicated until its resolution or, if not resolving, until considered stable or until the final study visit for that patient whichever comes first.

All SAEs that occur must be reported within 24 hours of discovery or notification of the event. Initial SAE information and all amendments or additions must be recorded as an SAE in the electronic data capture system and sent to First Wave BioPharma or designee.

If a patient is permanently withdrawn from the study because of an SAE, then this information must be captured as an SAE in the initial or follow-up electronic data capture record as well as the End-of-Therapy CRF.

The Investigator should notify the appropriate IEC/IRB of SAEs occurring at the site and other SAE reports received from First Wave BioPharma, in accordance with local procedures and statutes.

First Wave BioPharma will ensure that applicable regulatory authorities receive all relevant information on an SAE, in accordance with regulatory requirements. Results of First Wave BioPharma's investigation of other safety information shall be submitted, as required.

10.6 Pregnancy Reporting Procedures

Patients must be instructed to inform the Investigator immediately if they or their partners become pregnant after the patient has received their first dose of investigational product during the study. The following actions should be taken in the event of a confirmed pregnancy:

1. For female patients, study drug should be discontinued immediately.
2. The pregnancy should be reported to the Safety Group within 24 hours of notification using the applicable Pregnancy Report Form.
3. The Investigator should counsel the patient regarding the possible effects of previous study drug exposure on the fetus and the need to inform the study site of the outcome of the pregnancy.
4. The patient or the patient's pregnant partner must be monitored until the immediate postnatal period or until termination of the pregnancy. The outcome should be reported to the Medical Monitor using the Pregnancy Outcome Form.

Pregnancy is not an AE in and of itself. However, any pregnancy complications or elective terminations of a pregnancy for medical reasons will be recorded as an AE or SAE. A spontaneous abortion is always considered an SAE and will be reported as described in the AE and SAE sections (Section 10.2 and Section 10.5). Furthermore, any SAE occurring as an adverse pregnancy outcome poststudy must be reported to the Medical Monitor.

10.7 Hepatic Monitoring

The following guidelines are provided for the management of serum transaminase elevations in patients with new, clinically meaningful increases in liver function tests occurring during the study. All cases of new (i.e., since screening) elevations in ALT or AST ε 3 times ULN or any questions concerning the management of a patient with elevated serum transaminases should be discussed with the study Medical Monitor.

Patients who experience a new (i.e., since screening) elevation in ALT or AST level to ≥3 × ULN OR total bilirubin ≥1.5 × ULN will have hepatic monitoring (ALT, AST, Bilirubin) until one of the following occurs:

- Withdrawal from study if elevation in ALT or AST level to ≥3 × ULN is associated with a rise in total serum bilirubin to ≥2 × ULN (without laboratory findings of cholestasis [elevated serum alkaline phosphatase ≥2 × ULN])
- Withdrawal from study if elevation in ALT or AST level to ≥5 × ULN is associated with new or exacerbated gastrointestinal (GI) symptoms (i.e., nausea, vomiting, right upper quadrant pain, and/or jaundice) and other causes are not evident
- Withdrawal from the study if elevation in ALT or AST is ≥10 × ULN regardless of serum bilirubin level

Causes of acute elevation of transaminases should be considered and ruled out (e.g., viral hepatitis, concomitant medications).

Patients discontinued from study drug administration should resume porcine PERT therapy according to physician prescription.

10.8 Appropriateness of Measurements

The efficacy and safety assessments planned for this study are widely used and generally recognized as reliable, accurate, and relevant to the assessment of biological products administered to patients with CF.

11 STATISTICAL METHODS

11.1 General Considerations

The statistical methods will be further detailed in the Statistical Analysis Plan (SAP).

Any change to the data analysis methods described in the protocol will require an amendment only if it changes a principal feature of the protocol. Any other change to the data analysis methods described in the protocol, and the description for making the change, will be described in the SAP and in the clinical study report. Additional exploratory analyses of the data will be conducted as deemed appropriate.

Statistical methodology and analyses will be in accordance with the principles outlined by the International Council for Harmonisation (ICH) E9 guidelines.

Tables, listings, and figures will be produced in accordance with the principles outlined by the ICH E3 guidelines.

Summaries will consist of descriptive statistics including the nonmissing counts, mean, standard deviation, median, Interquartiles (IQ), minimum and maximum values for continuous variables, and number and percentage of patients in each defined category for categorical variables.

The primary efficacy endpoint analysis aims to compare the mean CFA after receiving adrulipase vs porcine PERT. With the sample size to be employed, primary efficacy analysis will be descriptive rather than inferential.

As a general rule, missing data will not be replaced.

11.2 Sample Size Determination

Based upon CFA data obtained in previous studies with adrulipase, 12 patients per adrulipase dose should provide sufficient point estimates of CFA in each group. Given the acceptable safety profile obtained in the previous study, a sample of 12 patients should be adequate for observation of safety per dose of adrulipase. This study will use descriptive analyses for both safety and efficacy analyses.

11.3 Analysis Sets

- Safety Set: patients receiving at least one dose of treatment. Patients will be analyzed according to the treatment actually received. The Safety Set will be used for all analyses of safety endpoints unless specified otherwise and for the summaries of patients in listings related to dosing of study drug.
- Efficacy sets:
    - mITT Set: All randomized patients receiving at least one dose of treatment and have at least one valid stool collection and CFA post baseline while receiving their assigned study drug. The mITT set is considered as the primary set for the efficacy analysis. The mITT set will be used for all analyses of efficacy endpoints.
    - Per Protocol Set: Subset of the mITT set comprising all patients who do not violate the terms of the protocol in a way that would affect the study outcome significantly, as determined by the study clinician. Protocol

deviations will be captured throughout the study and classified as minor or major and reviewed at a data review meeting before database lock. Attendees will include appropriate individuals from First Wave BioPharma and contract research organization. Each major deviation will be categorized as either important or not important with respect to the effect on the primary endpoint analysis. A sensitivity analysis will be performed in the Per Protocol Set.

11.4 Handling Missing Data

Missing data will not be replaced.

11.5 Demographic, Other Baseline Characteristics and Medication

Demographics, other baseline characteristics, and medications will be summarized for the Safety Set unless specified otherwise.

11.5.1 Patient Disposition and Withdrawals

The following will be summarized:

- Number of screen failures
- Number of patients included in the ITT, and PP sets
- Number of patients who completed the study
- Number of patients who discontinued the study early with reasons

The patient disposition, and protocol deviations will be listed.

11.5.2 Demographic and Other Baseline Characteristics

Demographic and other baseline characteristics will be summarized in the Safety Set:

- For each dose (i.e., 1.2 g/day, 1.95 g/day, 2.4 g/day, or 3.3 g/day)
- for all patients

11.6 Safety Analysis

Safety (AEs, SAEs, discontinuations due to adverse events, and safety laboratory values) will be assessed by descriptive methods.

11.7 Efficacy Analysis

11.7.1 Primary Efficacy Endpoint

The primary efficacy endpoint is defined as the CFA assessed at the end of the approximately 21-day treatment period (Part 1) or 14-day treatment period (Part 2). CFA will be calculated as follows:

$$\text{CFA (\%)} = 100 ((\text{fat intake (g)} - \text{fat excretion (g)})/ \text{fat intake (g)})$$

Primary Analysis:

The primary analysis of the primary endpoint will be a comparison of the mean CFAs from the adrulipase treatment vs the PERT treatment (screening), using descriptive statistics. Data will be analyzed according to the protocol version under which patients enroll (ie, Part 1 [protocol version 1.0] or Part 2[protocol version 2.0]) according to the SAP.

11.7.2 Secondary Efficacy Endpoints

Coefficient of nitrogen absorption (CNA) will be calculated and analyzed in the same manner as CFA.

Stool weights, signs and symptoms for malabsorption, body weight, and will be analyzed using descriptive methods.

12 QUALITY ASSURANCE AND QUALITY CONTROL

12.1 Audit and Inspection

An audit/inspection may be carried out by qualified First Wave BioPharma staff member, by subcontracted auditors or by representatives of national or foreign Health Authorities to ensure that the study is conducted as per protocol and in accordance with regulatory requirements, and to ensure the validity of the data. Participation in this study implies acceptance to cooperate in any potential audit/inspection.

The audit/inspection may consist of an inspection of the premises and equipment together with verification of the study documents and data. The investigational team must be available for inspection or audit. When First Wave BioPharma or the Investigator is informed that an inspection is to be performed, the other party must be informed immediately.

Audits/inspection may take place after the end of the study.

12.2 Monitoring

Data for each patient will be recorded on a CRF by the investigational site. Data collection must be completed for each patient who signs an informed consent form (ICF).

In accordance with cGCP and ICH guidelines, the study monitor will carry out source document verification at regular intervals to ensure that the data collected in the CRF are accurate and reliable.

The Investigator must permit the monitor, the IEC/IRB, the First Wave BioPharma's internal auditors, and representatives from regulatory authorities, direct access to all study-related documents and pertinent hospital or medical records for confirmation of data contained within the CRFs.

12.2.1 Responsibilities of the Monitor

Instructions for monitoring will be documented in a clinical monitoring plan. The clinical monitoring plan will include options for off-site/remote data verification.

The responsibilities of the study monitor are defined in ICH-E6, Chapter 5. The Monitor, who is mandated by First Wave BioPharma, must ensure that the study is conducted in accordance with GCP guidelines and all applicable local laws, and that the rights, the security and the well-being of the patients are respected.

During the conduct of the study, the monitor reports any deviations or persistent poor compliance with the study requirements and First Wave BioPharma will make decisions about appropriate corrective actions.

Compliance:

- The monitor has the responsibility of assessing the progress of the study, checking that the informed consent forms have been signed by the patient ensuring adhesion to and compliance with the study protocol and other study-related documents, and of ensuring the accuracy and completeness of the CRFs. Inconsistencies in the study records are to be resolved.

Source Data Verification:

- The monitor will perform source document verification and validation and request clarification to ensure the accuracy, completeness, and reliability of data.

Investigational Medicinal Product:

- The monitor must ensure that IMP handling is properly carried out and documented.

He/she must ensure that the Investigator site file is up to date with regard to essential documents.

12.3 Data Management and Coding

The assigned contract research organization will be responsible for activities associated with the data management of this study. This will include setting up a relevant database and data transfer mechanisms, along with appropriate validation of data and resolution of queries.

Investigational sites will enter data directly into an electronic data capture system by completing the CRF. Data entered into the eCRF must be verifiable against source documents at the study center. Data to be recorded directly on the eCRF will be identified and the eCRF will be considered the source document. Any changes to the data entered into the electronic data capture system will be recorded in the audit trail and will be FDA CFR 21 Part 11 compliant.

13 ETHICS

13.1 Ethics and Regulatory Considerations

The current study is to be conducted in accordance with globally accepted standards of GCP (ICH-E6), the Clinical Trial Directive 2001/20/EC, the GCP Directive 2005/28/EC, the revised version of the Declaration of Helsinki set out in the European Directive, and with applicable local requirements.

The protocol will be submitted to the Health Authorities and a properly constituted IEC or IRB for formal approval of the study conduct in accordance with local regulations.

The study may not begin until the protocol has received appropriate approval from the Health Authorities in accordance with local requirements.

In accordance with specific local requirements, the Investigator may be responsible for submitting the protocol and any amendments to the local IEC/IRB. A copy of the decision letter, a list and versions of documents submitted, and a list of IEC/IRB members and their affiliation should be provided by the Investigator to First Wave BioPharma.

During the study, First Wave BioPharma should promptly notify the Investigators and Health Authorities of any relevant information that could affect the safety of patients or effect on the conduct of the study. The regulatory authorities will be notified that the study has ended on completion of the study.

13.2 Ethical Conduct of the Study

The Investigator(s) and all parties involved in this study should conduct the study in adherence to the ethical principles based on the Declaration of Helsinki, GCP, ICH guidelines, and applicable national laws, local laws, and regulatory requirements.

13.3 Informed Consent

An IRB/IEC approved telephone informed consent to ensure that subjects come into clinic only if interested in proceeding with screening procedures is encouraged.

Before a patient's participation in this clinical study, the Investigator is responsible for obtaining written informed consent from the patient or legally acceptable representative after adequate explanation of the aims, methods, anticipated benefits, and potential hazards of the study and before any protocol-specific screening procedures or any investigational products

are administered. A legally acceptable representative is an individual or other body authorized under applicable law to consent, on behalf of a prospective patient, to the patient's participation in the clinical study. The Investigator is also responsible for asking the patient if the patient agrees to have his/her primary care physician informed of the patient's participation in the clinical study. If the patient agrees to such notification, the Investigator shall inform the patient's primary care physician of the patient's participation in the clinical study.

The acquisition of informed consent and the patient's agreement or refusal of his/her notification of the primary care physician should be documented in the patient's medical records, and the informed consent form should be signed and personally dated by the patient or a legally acceptable representative and by the person who conducted the informed consent discussion (not necessarily an Investigator). The original signed informed consent form should be retained in accordance with institutional policy, and a copy of the signed consent form should be provided to the patient or legally acceptable representative.

If a potential patient is illiterate or visually impaired and does not have a legally acceptable representative, then the Investigator must provide an impartial witness to read the informed consent form to the patient and must allow for questions. Thereafter, both the patient and the witness must sign the informed consent form to attest that informed consent was freely given and understood.

13.4 Patient Confidentiality

Patient data will be kept strictly confidential. Patient anonymity will be protected by using number codes and /or initials.

14 REPORTING AND PUBLICATION, INCLUDING ARCHIVING

The Investigator shall not divulge unpublished data or information related to the study provided by First Wave BioPharma, including but not limited to the study product characteristics, the Investigator's Brochure, the study protocol, case report forms, assay methods, or scientific data, to any third party without written approval from First Wave BioPharma. In addition, any new information that may become available during the course of the study shall be considered as confidential and shall not be used for any purpose other than the performance of the clinical study.

The study data are the property of First Wave BioPharma. The Investigator and any of the research staff shall obtain written approval from First Wave BioPharma before the publication/communication of the results of any work carried out during or in relation to the study. Publication and/or communication of the results of the clinical study will be of a cooperative nature involving authors representing First Wave BioPharma, the Investigators, and the scientific committee, if any.

First Wave BioPharma reserves the right to request modification of the content and/or timing of any publication or presentation if a patent application, an existing patent, or other proprietary rights may be jeopardized.

Authorship of any publication related to the study and the order of presentation of the authors' names shall be approved by First Wave BioPharma. First Wave BioPharma shall not use an Investigator's name in any publication without his/her written permission and vice versa.

First Wave BioPharma should retain all essential study-related documents, i.e., documents which permit evaluation of the conduct of a study and the quality of the data produced, in accordance with the applicable regulatory requirements of his/her country. These essential documents include, but are not limited to, signed protocol, Investigator's Brochure, printed CRFs, medical records, laboratory reports, informed consent forms, drug disposition records, safety reports, information regarding participants who discontinued, and other relevant documents and data.

Study-related documents should be kept together in the Investigator site file provided to the Investigator by First Wave BioPharma. Sufficient information about the identity of all study patients (e.g., name, medical records number, patient number, and study number) should be retained by the Investigator so that any First Wave BioPharma representatives, auditors, or inspectors may access this information when required. The Investigator must retain all records for 15 years or longer if required by specific local requirements. The Investigator will contact First Wave BioPharma for authorization before the destruction of any study records or in the event of accidental loss or destruction of any of them. All records should be kept in a secure area; however, in the cases of audit or inspection, they should be rapidly made available.

15 REFERENCES

Airinei GC, Gaudichon C, Bos C, Bon N, Kapel B, Bejou JJ, et al. Postprandial protein metabolism but not a faecal test reveals protein malabsorption in patients with pancreatic exocrine insufficiency. Clin Nutr. 2011; 30: 831–7.

Aloulou A, Rodriguez D, Puccinelli N, Mouz J, Leclaire Y, Leblond Y, et al. Purification and biochemical characterization of the LIP2 lipase from Yarrowia lipolytica. Biochim Biophys Acta. 2007;1771: 228–37.

Aloulou A, Schue M, Puccinelli D, Milano S, Delchambre C, Leblond Y, et al. Yarrowia lipolytica lipase 2 is stable and highly active in test meals and increases fat absorption in an animal model of pancreatic exocrine insufficiency. Gastroenterology. 2015; 149: 1910-1919.

Banks PA, Conwell DL, Toskes PP. The management of acute and chronic pancreatitis. Gastroenterol Hepatol (N Y). 2010;6(2 Suppl 3):1–16.

Bodewes FAJA, Verkade HJ, Taminiau JAJM, Borowitz D, Wilschanski M. Cystic fibrosis and the role of gastrointestinal outcome measures in the new era of therapeutic CFTR modulation. J Cyst Fibros. 2015;14:169–77.

Cherney, B. 2008. Viral Safety Issues for the Pancreatic Enzyme Product CREON. F. report, FDA. https://www.fda.gov/ohrms/dockets/ac/08/slides/2008-4402s1-02-FDA-Cherney%20.pdf. (Accessed 25 October 2017.)

Cystic Fibrosis Foundation Patient Registry. Annual Data Report 2016.

Cystic Fibrosis Foundation. What is cystic fibrosis? http://www.cff.org/AboutCF/. Accessed 20 October 2017.

Dhanasekaran R, Toskes PP. Pancrelipase for pancreatic disorders: An update. Drugs Today (Barc). 2010;46: 855–66.

DiMagno EP, Clain JE, Layer P. 1993.Chronic Pancreatitis: In GO VI ed. The Pancreas: biology, pathophysiology and disease. 2nd ed. New York, Raven Press; 665-706.

Dominguez-Munoz JE. Pancreatic enzyme therapy for pancreatic exocrine insufficiency. Current Gastroenterol Rep.2007; 9: 116-122.

Dominguez-Munoz JE, Iglesias-Garcia J, Iglesias-Rey M, Vilarino-Insua M. Optimising the therapy of exocrine pancreatic insufficiency by the association of a proton pump inhibitor to enteric pancreatic extracts. Gut. 2006;55: 1056–7.

Engelen MPKJ, Com G, Deutz NEP. Protein is an important but undervalued macronutrient in the nutritional care of patients with cystic fibrosis. Current Opinion Clin Nutr Metab Care. 2014; 17: 515-520.

Farrell, PM. The prevalence of cystic fibrosis in the European Union. J Cyst Fibros. 2008;7: 450–3.

Ferrone M, Raimondo M, Scolapio JS. Pancreatic enzyme pharmacotherapy. Pharmacotherapy. 2007; 27: 910-920.

Gelfond D, Ma C, Semler J, Borowitz D. 2013. Intestinal pH and gastrointestinal transit profiles in cystic fibrosis patients measured by wireless motility capsule. Dig Dis Sci. 2013 Aug;58(8):2275-81

Greenberger N, Toskes P. 2008. Acute and Chronic Pancreatitis. Harrison's Principles of Internal Medicine. A. Fauci, E. Braunwald, D. Kasper et al. Columbus, OH, McGraw-Hill.

Greenberger N, Toskes P. 2013. Acute and Chronic Pancreatitis. Harrison's Gastroenterology and Hepatology. D. Longo and A. Fauci. Columbus, OH, McGraw-Hill Medical.

Hammer HF. Pancreatic exocrine insufficiency: diagnostic evaluation and replacement therapy with pancreatic enzymes. Dig Dis. 2010; 28: 339-343.

Heubi JE, Schaeffer D, Ahrens RC, Sollo N, Strausbaugh S, Graff G, et al. Safety and efficacy of a novel microbial lipase in patients with exocrine pancreatic insufficiency due to cystic fibrosis: a randomized controlled clinical trial. The Journal of Pediatrics. 2016; 176: 156-161.

Kimura, R.E., et al. Indomethacin and pancreatic enzymes synergistically damage intestine of rats. Dig Dis Sci, 1998. 43: 2322-32.

Kimura, R.E., et al. The effects of high-dose ibuprofen and pancreatic enzymes on the intestine of the rat, J Pediatr Gastroenterol Nutr, 1999; 29:178-83.

King CE, Leibach L, Toskes PP. Clinically significant vitamin B12 deficiency secondary to malabsorption of protein-bound vitamin B12. Dig Dis Sci. 1979;24: 397–402.

Ladas SD, Giorgiotis K, Raptis SA. Complex carbohydrate malabsorption in exocrine pancreatic insufficiency. Gut. 1993; 34: 984-987.

Ledder O, Haller W, Couper RT, Lewindon P, Oliver M. Cystic fibrosis: an update for clinicians. Part 2: hepatobiliary and pancreatic manifestations. J Gastroenterol Hepatol. 2014; 29: 1954–62.

Pignede G, Wang H, Fudalej F, Gaillardin C, Seman M, Nicaud JM. Characterization of an extracellular lipase encoded by LIP2 in Yarrowia lipolytica. J Bacteriol. 2000;182: 2802–10.

Safdi M, Bekal PK, Martin S, Saeed ZA, Burton F, Toskes PP. The effects of oral pancreatic enzymes (Creon 10 capsule) on steatorrhea: a multicenter, placebo-controlled, parallel group trial in subjects with chronic pancreatitis. Pancreas. 2006;33:156–62.

Stallings VA, Stark LJ, Robinson KA, Feranchak AP, Quinton H. Clinical Practice Guidelines on, S. Nutrition and G. Ad Hoc Working. Evidence-based practice recommendations for nutrition-related management of children and adults with cystic fibrosis and pancreatic insufficiency: results of a systematic review. J Am Diet Assoc. 2008;108: 832–9.

Struyvenberg MR, Martin CR, Freedman SD. Practical guide to exocrine pancreatic insufficiency - Breaking the myths. BMC Med. 2017;15:29.

Van Hoozen CM, Peeke PG, Taubeneck M, Frey CF, Halsted CH. Efficacy of enzyme supplementation after surgery for chronic pancreatitis. Pancreas. 1997;14: 174–80.

Waters DL, Dorney SF, Gaskin KJ, Gruca MA, O'Halloran M, Wilcken B. Pancreatic function in infants identified as having cystic fibrosis in a neonatal screening program. N Engl J Med. 1990;322:303–8.

16 APPENDICES

16.1 Investigator Signature Page

Protocol Title	A Phase 2, Open Label, Multicenter, Pilot Study to Assess Safety and Efficacy of an Enteric Microgranule Formulation of Adrulipase in Patients with Exocrine Pancreatic Insufficiency (EPI) due to Cystic Fibrosis (CF)
Study Name	**TBD:** A Phase 2, Open Label, Multicenter, Pilot Study to Assess Safety and Efficacy of an Enteric Microgranule Formulation of Adrulipase in Patients with Exocrine Pancreatic Insufficiency (EPI) due to Cystic Fibrosis (CF)
Protocol Number	FWB-CF-2.03
Protocol Date	May 12, 2023
Protocol Version	2.0

Confidentiality and cGCP Compliance Statement

I have read and understand the protocol (Study No. FWB-CF-2.03) and the Investigator's Brochure and I agree that it contains all the ethical, legal, and scientific information necessary to conduct this study. I will conduct this protocol as outlined herein and will make every effort to complete the study within the time designated.

I will provide copies of the protocol and access to all information furnished by First Wave BioPharma, Inc. to study personnel under my supervision. I will discuss this material with them to ensure that they are fully informed about the drug and the study.

I understand that the study may be terminated, or enrollment suspended at any time by First Wave BioPharma, Inc., with or without cause, or by me if it becomes necessary to protect the best interest of the study patients.

_____

Investigator Name (print)

_____

Signature Date

16.2 Clinical Laboratory Tests

Clinical Chemistry:	**Hematology:**
Serum Concentrations of:	**Hematocrit**
Sodium	**Hemoglobin**
Potassium	**Erythrocyte count (RBC)**
Chloride	**Leukocytes (WBC)**
Bicarbonate	**Absolute counts of:**
Blood urea nitrogen (BUN)	**Neutrophils, segmented**
Total Calcium	**Neutrophils, juvenile (bands)**
Phosphorus	**Lymphocytes**
Magnesium	**Monocytes**
Glucose	**Eosinophils**
Creatinine	**Basophils**
Albumin	**Platelets**
Prealbumin	
Total protein	**Other:**
Alkaline phosphatase	**Serum pregnancy test[a]**
Alanine aminotransferase (ALT)	**Urine pregnancy test[a]**
Aspartate aminotransferase (AST)	**Activated partial thromboplastin time (aPTT)**
Lactate dehydrogenase (LDH)	**Prothrombin time/International normalized ratio (PT/INR)**
Total bilirubin	
Direct bilirubin	
Uric Acid	
Fasting Lipid Profile:	**Stool:**
Total cholesterol	**Marker-to-marker stool collection**
Triglycerides	**Fecal Elastase**
Low Density Lipoprotein (LDL)	
High Density Lipoprotein (HDL)	
Very Low Density Lipoprotein (VLDL)	

Note: All labs will be assayed by First Wave BioPharma designated central laboratory unless otherwise noted.

a. **Only for females of childbearing potential**