PROTOCOL 202016
IND Number: 132145
Amendment II

A PHASE 1 STUDY TO ASSESS THE PHARMACOKINETICS AND SAFETY OF ASCENDING DOSES OF JOTROL ORAL GELCAPS IN HEALTHY SUBJECTS, AND TO DETERMINE THE INFLUENCE OF FOOD

Contract Research Organization:
inVentiv Health Clinical Research Services LLC, a Syneos Health company (« Syneos Health »)

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Sponsor:
Jupiter Orphan Therapeutics

601 Heritage Drive
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Protocol Historical File

Version number	Brief description/summary of changes	Date
Final	Version submitted to the IEC.	20-AUG-2020
Amendment I	Peripheral blood mononuclear cells will be isolated from additional blood samples in all periods of study Part 1, leading to a total blood volume exceeding 500 mL in Period 3. Hence, a hematology test is included before dosing in Period 3. A clarification to subjects to be included in Part 2 is also provided.	12-NOV-2020
Amendment II	See the description of changes below.	25-NOV-2020

Changes included in Amendment II:

Based on the anticipated bioavailability profile of the JOTROL formulation, the U.S. National Institute of Aging (NIA) recommended to establish a formal Data and Safety Monitoring Board (DSMB) constituted by 3 qualified pharmacovigilance persons, totally independent of Syneos Health, JOT, and the associated investigators on the study. This independent group of experts will periodically review and evaluate the accumulated study data for participant safety, study conduct and progress, and make recommendations concerning the continuation, modification, or termination of the trial. Therefore, there are changes under:

- Section 2. Synopsis of Protocol – Study Design / Subjects / Drug Administration
- Section 6. Study Design
- Section 7.5 Dose Escalation Criteria
- Section 9.5 Drug Administration
- Section 9.11 Subject Withdrawal and Replacement

Two additional guidances are cited for the establishment of DSMB and the Investigator's Brochure has been updated. Therefore, reference numbering is updated throughout the protocol and the section 15. References is also updated.

Signature Page

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Sponsor's representative:

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11/25/2020
Date

Signature Page

Contract Research Organization (CRO)

Investigator:

I have carefully read this study protocol and agree that it contains all necessary information required to conduct this study. I agree to conduct the study according to this protocol (including any amendments) and in accordance with the clinical site's Standard Operating Procedures (SOPs), ICH Good Clinical Practices (GCP), all other applicable regulations, and the recommendations laid down in the most recent version of the Declaration of Helsinki.

PPD

Electronically signed by: PPD
PPD
Reason: I am the approver
Date: Nov 25, 2020 12:40 EST

25-Nov-2020

PPD, M.D.
PPD

Date

1. Facilities and Responsible Staff

1.1 Clinical Research Facilities

This study will be conducted by Syneos Health at the following facility:

1951 NW 7th Avenue, Suites 180 and 450
Miami, FL 33136, USA
Tel.: 1-305-547-5800

1.2 Biomedical Laboratory Facilities

Biomedical laboratory testing will be performed by the following laboratories:

Eccolab Group Co.
8370 W. Flagler Street, Suite 216
Miami, FL 33144, USA
Tel.: 1-800-616-1770

Syneos Health
1951 NW 7th Avenue, Suites 180 and 450
Miami, FL 33136, USA
Tel.: 1-305-547-5800

If another biomedical laboratory is used, this will be documented and annexed to the protocol.

1.3 Clinical Pharmacology and Regulatory Affairs

Syneos Health
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1.4 Bioanalytical Facility

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Table of Contents

2. Synopsis of Protocol

Project No.:	202016
Title:	Phase 1 Study, to Assess the Pharmacokinetics and Safety of Ascending Doses of JOTROL Oral Gelcaps in Healthy Subjects, and to Determine the Influence of Food.
Study Phase and Type:	Phase 1 - Single Ascending Doses (SAD) study
Objectives:	The objectives of this study are: • To characterize the pharmacokinetic (PK) profile of JOTROL (resveratrol) following oral administration of SAD ranging from 200 mg up to a dose currently estimated at 1000 mg, in healthy adult subjects. • To evaluate the effect of food on the PK profile of JOTROL.
Study Design:	Single center, Phase 1, open-label, sequential SAD study, with a food-effect arm. The study will be divided into two parts: • Study Part 1 consists of 3 periods with SAD of JOTROL under fasting conditions. Periods 2 and 3 will be initiated with updated doses after safety, tolerability, and PK data are evaluated by the Safety Committee **and the Data and Safety Monitoring Board (DSMB)** and deemed acceptable for single doses for subsequent doses. • Study Part 2 consists of a single JOTROL dose under fed conditions. JOTROL dose will derive from study Part 1 safety, tolerability, and PK data.
Subjects:	24 healthy adult males or females, ≥ 18 and ≤ 75 years of age, non-smokers will be included in the study Part 1. Only 16 subjects who completed study Part 1 will be included in study Part 2. In order to minimize potential PK variability, elderly (more than 65 years old subjects) will be not be included in study Part 2.These 16 subjects will be selected according to their order of enrolment in the study, provided their consent to continue the study, they follow the study restrictions and they still meet study criteria. Subjects who withdraw or are withdrawn from study Part 1 after dosing, for reasons other than safety and tolerability, may be replaced after consultation between the Safety Committee members **and the DSMB members** in order to assure initiating study Part 2 with 16 subjects. For study Part 1 only, an effort will be made to include to the extent possible, subjects of the following age groups: • ≥ 65 and ≤ 70 years of age; • ≥ 70 and ≤ 75 years of age.
Inclusion Criteria:	1) Normal healthy male or female volunteers, non-smokers (no use of tobacco products within 3 months prior to screening), ≥ 18 and ≤ 75 years of age, with BMI > 18.5 and < 30.0 kg/m^2 and body weight ≥ 50.0 kg for males and ≥ 45.0 kg for females 2) Healthy as defined by: a) the absence of clinically significant illness and surgery within 4 weeks prior to dosing. Subjects vomiting within 24 hours predose will be carefully evaluated for upcoming illness/disease. Inclusion pre-dosing is at the discretion of the Investigator.

	b) the absence of clinically significant history of neurological, endocrine, cardiovascular, respiratory, hematological, immunological, psychiatric, gastrointestinal, renal, hepatic, and metabolic disease as determined by the Investigator. 3) Females of childbearing potential who are sexually active with a male partner must be willing to use one of the following acceptable contraceptive methods throughout the study and for 30 days after the last study drug administration: a) intra-uterine contraceptive device without hormone release system placed at least 4 weeks prior to study drug administration; b) male condom with intravaginally applied spermicide starting at least 21 days prior to study drug administration; c) sterile male partner (vasectomized since at least 6 months). 4) Capable of consent
Exclusion Criteria:	1) Any clinically significant abnormality at physical examination, clinically significant abnormal laboratory test results or positive test for hepatitis B, hepatitis C, or HIV found during medical screening. 2) Positive urine drug screen or urine cotinine test at screening. 3) History of allergic reactions to resveratrol, polyphenols, other related drugs, or to any excipient in the formulation. 4) Positive pregnancy test at screening. 5) Breast-feeding subject. 6) Clinically significant ECG abnormalities or vital sign abnormalities (systolic blood pressure lower than 90 or over 140 mmHg, diastolic blood pressure lower than 50 or over 90 mmHg, or heart rate less than 50 or over 100 bpm) at screening. 7) History of significant alcohol abuse within 1 year prior to screening or regular use of alcohol within 6 months prior to the screening visit (more than 14 units of alcohol per week [1 unit = 150 mL of wine, 360 mL of beer, or 45 mL of 40% alcohol]). 8) History of significant drug abuse within 1 year prior to screening or use of soft drugs (such as marijuana) within 3 months prior to the screening visit or hard drugs (such as cocaine, phencyclidine [PCP], crack, opioid derivatives including heroin, and amphetamine derivatives) within 1 year prior to screening. 9) Use of resveratrol for a medical condition or in the context of another clinical trial within a period of 30 days prior to the first dosing. 10) Participation in a clinical research study involving the administration of an investigational or marketed drug or device within 30 days prior to the first dosing, administration of a biological product in the context of a clinical research study within 90 days prior to the first dosing, or concomitant participation in an investigational study involving no drug or device administration. 11) Use of medications for the timeframes specified below, with the exception of medications exempted by the Investigator on a case-by-case basis because they are judged unlikely to affect the PK profile of the study drug or subject safety (e.g., topical drug products without significant systemic absorption): a) prescription medication within 14 days prior to the first dosing;

	b) over-the-counter products and natural health products (including herbal remedies, homeopathic and traditional medicines, probiotics, food supplements such as vitamins, minerals, amino acids, essential fatty acids, and protein supplements used in sports) within 14 days prior to the first dosing, with the exception of the occasional use of acetaminophen (up to 2 g daily); c) use of any drugs known to induce or inhibit hepatic drug metabolism, including St. John's wort, within 30 days prior to the first study drug administration; d) depot injection or an implant of any drug within 3 months prior to the first dosing. 12) Donation of plasma within 7 days prior to dosing. Donation or loss of blood (excluding volume drawn at screening) of 50 mL to 499 mL of blood within 30 days, or more than 499 mL within 56 days prior to the first dosing. 13) Any reason which, in the opinion of the Investigator, would prevent the subject from participating in the study.
Screening Procedures:	Demographic data, medical and medication histories, physical examination, body measurements, electrocardiogram (ECG), vital signs (blood pressure, heart rate, respiratory rate, and oral temperature), hematology, biochemistry, coagulation, serology (human immunodeficiency virus [HIV], hepatitis B and C tests), urinalysis, urine cotinine test, urine pregnancy test, and urine drug screen.
Confinements, Visits, and Washout:	For each period, subjects will be confined from Day -1 until after the 32-hour post-dose blood draw. There will be a washout period of 14 days or more between each study drug administration.
Drug Administration:	Single oral dose: <u>Study Part 1:</u> Subject will be sequentially dosed in an ascending manner with at least 14 days between the last dosing and the subsequent nominal dose level below, which will be calculated based on observed PK results: • Period 1: 2 x 100 mg JOTROL oral gelcaps, total resveratrol dose 200 mg • Period 2: 5 x 100 mg JOTROL oral gelcaps, total resveratrol dose 500 mg • Period 3: 10 x 100 mg JOTROL oral gelcaps, total resveratrol dose 1000 mg Targeted doses will be achieved with multiple JOTROL' gelcaps of 100 mg resveratrol strength. For Periods 2 and 3, JOTROL's doses to be administered will be reassessed when safety, tolerability, and PK data analysis are available. Adjustment to the currently outlined doses and/or dosing regimen may be necessary, but the dose to be administered in Period 2 or 3 will not exceed the one currently outlined in the protocol. No food will be allowed from at least 10 hours before dosing until at least 4 hours after dosing in each period. <u>Study Part 2:</u>

	In order to minimize potential PK variability, elderly (more than 65 years old subjects) will be not included in study Part 2. There will be at least 14 days between dosing of Periods 3 and 4. • Period 4: one single dose of JOTROL oral gelcaps. Dose to be administered will be derived from study Part 1 PK and safety data. The same dose will be administered in Periods 3 and 4. However, if the exposure is higher than expected in Period 3, the dose to be administered in Period 4 could be lower, in order to keep AUC exposures lower than 2.1 µg•hr/mL, as specified in section 7.2. After a supervised fast of at least 11 hours, subjects will be served a critical, high-fat, high-caloric meal. Drug administration will occur approximately 30±1 minutes after the meal has been started. Except for water given with study medication and fluids provided with the critical breakfast (study Part 2 only), no fluids will be allowed from 1 hour before dosing until 1 hour post-dose. Following completion of each dose level, PK, safety, tolerability data will be evaluated by a Safety Committee **and a DSMB** before proceeding to the next dose. Depending on safety and tolerability as well as on available PK data, the escalation scheme may be modified such that intermediate dose levels are administered. The Safety Committee will be composed by at least the Investigator and one medically qualified Sponsor representative. **A formal DSMB will be established and constituted by an independent group of experts (totally independent of Syneos Health, JOT, and the associated investigators on the study) that advises the NIA Director and the study investigator. The members of the DSMB serve in an individual capacity and provide their expertise and recommendations. The DSMB will be constituted and will act according to the FDA Guidance for Clinical Trial Sponsors - Establishment and Operation of Clinical Trial Data Monitoring Committees[1] and the NIA Guidance on Clinical Trials.[2]** Some adjustments to the currently outlined doses and/or dosing regimen may be implemented by the Safety Committee **or the DSMB** as described in Section 7.
Study Restrictions:	Subjects will be asked to refrain from using products that may potentially affect their safety and/or the PK profile of the study drug. Main study restrictions include: • prescription medication from 14 days prior to the first dosing until after the last PK blood sample collection of the study ; • over-the-counter products from 14 days prior to the first dosing until after the last PK blood sample collection of the study; • any drugs known to be strong inducer or inhibitor of hepatic drug metabolism, including St. John's wort, within 30 days prior to the first dosing until after the last PK blood sample collection of the study; • natural health products from 14 days pre-dose until after the last pharmacokinetic blood sample collection of each period; • food containing poppy seeds within 24 hours prior to admission; • food or beverages containing xanthine derivatives or xanthine-related compounds or energy drinks from 48 hours pre-dose until after the last PK blood sample collection of each period;

	• food or beverages containing grapefruit, starfruit, pomegranate, pineapple, or pomelo from 7 days pre-dose until after the last PK blood sample collection of each period; • foods rich in resveratrol (e.g. grapes, peanuts and their products including juices and wines) from 7 days pre-dose until after the last PK blood sample collection of each period; • alcohol-based products from 24 hours prior to admission until after the last PK blood sample collection of each period. Subjects will be allowed to engage in normal activity but will avoid lying down or sleeping, unless medically necessary or procedurally required, for 2 hours (study Part 1) and 4 hours (study Part 2) after study drug administration.
Blood Sample Collection for PK/PD Biomarkers Analysis:	A total of 17 blood samples will be collected in each period for PK analyses: pre-dose and 0.133, 0.25, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8, 10, 12, 16, 24, and 32 hours post-dose. A total of 5 additional blood samples will be collected in each period for potential RNA-Seq analysis: pre-dose, and 3, 6, 12, and 24 hours post-dose. For study Part 1 only, peripheral blood mononuclear cells (PBMC) will be isolated, from at most (maximum) 10 subjects at the following timepoints, in: • Period 1 at pre-dose, 3, and 24 hours post-dose; • Period 2 at 3, 6, and 24 hours post-dose; and in • Period 3 at 3, 6, and 24 hours post-dose. Other investigations may be performed using remaining samples already collected.
Urine Collection for PK Analysis:	For study Part 1, urinary levels will be quantified for PK analysis. Urine samples will be collected at 6 times or time intervals: spot pre-dose (within 2 hours before dosing), 0-4 hours, 4-8 hours, 8-12 hours, 12-24 hours, and 24-32 hours post-dose.
Subject Safety:	Urine drug screen, alcohol breath test, urine cotinine test, and serum pregnancy test: at check-in of each period. Hematology: at check-in of Period 3 (study Part 1) and study Part 2 (Period 4). Medical surveillance: Subjects will be monitored throughout the study by the clinical staff for adverse events (AEs). In each period, the Investigator or designee will be on site for drug administration and until 2 hours (study Part 1) and 4 hours (study Part 2) post-dose, and available on call for the remainder of the study.
Study Exit Procedures:	Hematology, biochemistry, urinalysis, urine pregnancy test, vital signs (blood pressure, heart rate, respiratory rate, and oral temperature), ECG, and AEs monitoring.
Analytical Method:	Syneos Health Clinical Bioanalytical Facility will analyze resveratrol, resveratrol-3-glucuronide, resveratrol-4'-glucuronide, and resveratrol-3-sulfate in plasma and urine samples using a validated method.
RNA-Seq Analysis, Iduronidase, mRNA Frataxin, and Other Possible Tests	Potential RNA-Seq analysis and other investigational tests such as iduronidase activity measurement and mRNA frataxin will be performed by an external laboratory. Details of this additional laboratory work will be

	available in a separate protocol. Details of the laboratory will be outlined when available in an additional information form.
Pharmacokinetics:	Plasma PK Parameters: AUC_{0-t}, AUC_{0-inf}, C_{max}, Residual area, T_{max}, $T_{½\ el}$, and K_{el}. Urine PK parameters: Ae_{0-t}, R_{max}, and T_{max}.
Statistical Analyses:	A Statistical Analysis Plan (SAP) will be prepared after completion of the final protocol. <u>Safety and tolerability</u>: Treatment-emergent AEs (TEAEs) will be tabulated by treatment. Changes from baseline values in vital signs, ECG, and clinical laboratory parameters will be evaluated. Safety and tolerability data will be reported using descriptive statistics. A listing of all TEAEs will be provided. Results of vital signs, ECG, and clinical laboratory parameters will be listed. <u>PK:</u> • Plasma PK: For all analytes, summary statistics will be used to describe the PK profile for each dose level under fasting conditions (Periods 1, 2, and 3) and for administration under fed conditions (Period 4). Dose proportionality analysis for AUC_{0-t}, AUC_{0-inf} and C_{max} will be performed (using the power model with mixed procedure from SAS®) considering data under fasting conditions (Periods 1, 2, and 3). For evaluation of the food-effect, PK data (ln-transformed $AUC_{0\ t}$, AUC_{0-inf}, C_{max} and untransformed T_{max}) reported under fed conditions (Period 4) and under fasting conditions (for the same dose level) will be compared using ANOVA from SAS®. The ratio (fed/fasting) and 90% geometric confidence interval will also be calculated for AUC_{0-t}, AUC_{0-inf}, and C_{max}. All inferential statistical analyses will be interpreted in an exploratory sense only at an alpha level of 5% for statistical significance. Interim PK analyses will be performed between each dose level and will inform selection of the subsequent dose. • Urine PK: Summary statistics will be used to describe urinary excretion.

3. Schedule of Events

PROCEDURE	Screening	Part 1			Part 2[1]			Study Exit[2]
		Periods 1, 2 & 3			Period 4			
		D-1	D1	D2	D-1	D1	D2	
Demographic Data	X							
Medical and Medication Histories	X							
Review and Monitoring of AEs and Concomitant Medications		X	X	X	X		X	X
Physical Exam. & Body Measurements	X							
Vital Signs	X							X
ECG	X							X
Biochemistry	X							X
Hematology	X	X[3]			X			X
Coagulation	X							
HIV and Hepatitis	X							
Urinalysis	X							X
Urine Drug Screen	X	X			X			
Urine Cotinine Test	X	X			X			
Alcohol breath test		X			X			
Serum Pregnancy Test		X			X			
Urine Pregnancy Test	X							X
Confinement		X	X	X	X	X	X	
Drug Administration			X			X		
Blood PK Samples[4]			X	X		X	X	
Urine PK Samples[5]			X	X				
PBMC Isolation[6]			X	X				
Blood Samples for RNA-Seq Analysis, Iduronidase, mRNA Frataxin, and Other Investigational Tests[7]			X	X		X	X	

1. For 16 subjects who completed study Part 1 only.
2. Study exit procedures for subjects who are not included in study Part 2 will be completed after the last blood draw of Period 3 (after 32 hours post-dose on Day 2) or within 14 days after the last participation of the subject in the study. Study exit procedures for subjects who are included in study Part 2 will be completed after the last blood draw of Period 4 (after 32 hours post-dose on Day 2) or within 14 days after the last participation of the subject in the study.
3. For Period 3 only.
4. Blood samples for PK analysis: pre-dose and 0.133, 0.25, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8, 10, 12, 16, 24, and 32 hours post-dose.
5. Urine samples for PK analysis: Urine samples will be collected at 6 times or time intervals: spot pre-dose (within 2 hours before dosing), 0-4 hours, 4-8 hours, 8-12 hours, 12-24 hours, and 24-32 hours post-dose.

6. PBMC will be isolated in: Period 1 at pre-dose, 3, and 24 hours post-dose; Period 2 at 3, 6, and 24 hours post-dose; and in Period 3 at 3, 6, and 24 hours post-dose.
7. Blood samples for potential RNA-seq analysis, iduronidase activity, and mRNA frataxin /other investigational tests: pre-dose and 3, 6, 12, and 24 hours post-dose.

4. Introduction

4.1 Background Information

Resveratrol is a chemical compound (trans-3,4',5-trihydroxystilbene[3]) that is a member of a unique family of polyphenols called viniferins.[4] It is a polyphenolic compound similar to diethylstilbestrol, a synthetic estrogen. Resveratrol presents itself in both *trans*- and *cis*- isomeric forms. It is found in an abundant amount in red wine, grape berry skins and seeds and, particularly in dried roots of the plant *Polygonum cuspidatum*. Content of resveratrol in grapes varies from 0.16 to 3.54 mg/g; dry grape skin contains about 24 mg/g of resveratrol. Resveratrol is also present in other berries and nuts. For example, cranberry raw juice contains about 0.2 mg/L. In other natural foods, the concentration of resveratrol varies in the range of mg/g (peanuts, pistachios) to ng/g (bilberries, blue berries). It has been documented that red wine contains a much greater amount of polyphenolic compounds than white wine. The concentration of resveratrol ranges from 0.1 to 14.3 mg/L in various types of red wine, while white wines contain only about 0.1–2.1 mg/L of resveratrol.[5]

Resveratrol is also a phytoalexin, a antimicrobial substance synthesized by plants and accumulating rapidly at areas of pathogen infection.[6] Resveratrol is produced in plants usually in response to stress, injury or fungal infection but also in response to mechanical injury, UV irradiation and changes in climate.[4,6,7] In plants, resveratrol exerts antioxidant function by protecting against sun damage. Food products contain both cis- and trans-isoforms of resveratrol, mostly in the glycosylated form. Glycosylation prevents enzymatic oxidation, thereby increasing stability and bioavailability of resveratrol.[5]

Research has recognised the beneficial effects of resveratrol as an important component of the overall injury that occurs in various disorders such as oxidative stress, myocardial injury, anticancer activity, anti-diabetic activity, and antihypercholesterolemic effects.[8] Resveratrol is both a free radical scavenger and a potent antioxidant that promotes the activities of a variety of antioxidant enzymes. Resveratrol is believed to exert its protective actions through the regulation of nitric oxide production and has been shown to have anti-inflammatory, anti-carcinogenic and anti-aging effects. The anti-inflammatory and anti-oxidant properties of resveratrol have been linked with the prevention of ocular diseases such as age-related macular degeneration (AMD), diabetic retinopathy, and glaucoma.[4]

Multiplicity of resveratrol biological effects is mainly caused by the abundance and diversity of molecular targets of this compound like cyclooxygenases/lipooxygenases, a wide range of various kinases, sirtuins, transcription factors, cytokines, DNA polymerase, adenylyl cyclase, ribonucleotide reductase, aromatase and others. It is hypothesized that resveratrol provides a complex physiological action because of its capability to modulate different pathways in a micromolar range. Many studies have shown that resveratrol possesses cardiovascular protective, antiplatelet, antioxidant, anti-inflammatory, blood glucose-lowering, and anti-cancer activities. By increasing the production of nitric oxide, resveratrol inhibits platelet aggregation and stimulates vasodilation. Recently published data have shown that resveratrol protects against some neurodegenerative diseases, such as Alzheimer's disease and obesity as well as is effective in the management of osteoporosis in post-menopausal women without an increased risk of breast cancer.[5]

The trans-isoform is more common in plants. It has been reported that this compound has low toxicity as it was well tolerated in the short-term experiments performed in humans. Recent clinical trials proved that resveratrol is well-tolerated and pharmacologically safe at doses up to 5 g/day. However, the data on toxicity of resveratrol in long term experiments are scarce.[5] However, it is reported that resveratrol treatment at a dose of 8 mg/day for one year significantly reduced a number of cardiac risk factors.[9]

Thousands of basic science experiments *in vitro* and in animal models suggest low toxicity and many positive effects of resveratrol.

4.2 Toxicology

There is ample published nonclinical oral toxicity data regarding the use of resveratrol in acute, subchronic, and chronic settings (Table 7 in the Investigator Brochure).[10] In a 28-day study conducted in male and female CD rats (20/sex/group) where the animals were dosed once daily with 0, 300, 1000 or 3000 mg/kg body weight (bw)/day resveratrol by gavage, no observed adverse effects were observed in animals treated with 300 mg/kg bw/day resveratrol. Changes observed in animals treated with 1000 mg/kg bw/day resveratrol were mild reduced body weight in females, slight increase in white blood count and lymphocytes in males, and decreased adrenal gland weights in females. No histological changes were observed. Male and female rats treated with 3000 mg/kg bw/day of resveratrol showed reduced body weight and food consumption, elevated kidney weight, and increased incidences of kidney lesions.[11]

The incidence and severity of nephropathy was similar to the animals who received placebo. It was concluded that the nephropathy seen in resveratrol groups was not treatment related; however, the kidney was identified as the target organ of toxicity in animals treated with 3000 mg/kg bw/day resveratrol. The 300 mg/kg bw/day resveratrol body weight was identified as the no-observed-adverse-effect-level (NOAEL).[11]

Three 28-day studies were conducted in rat, dog, and rabbit with daily oral doses ranging from 250 mg/kg to 3 g/kg. The resveratrol used in these trials was a micronized formulation. In the rat study, the only finding was a slight hemolytic anemia in male rats at the 1,000 mg/kg body weight per day dose that was not associated with microscopic changes in bone marrow or spleen. Recovery animals were not anemic. The NOAEL for both male and female rats was considered to be 300 mg/kg/day. For the dog study, dosing up to 300 mg/kg/day did not show any cardiovascular effects and there were no changes in weight gain or food consumption. As in the rat study, the NOAEL for both male and female dogs was considered to be 300 mg/kg/day. In rabbits, the highest daily dose given was 750 mg/kg. At this dose, 5 of 8 males and 1 of 8 females were euthanized because of body weight loss. Other clinical findings included discolored urine and stool abnormalities; and in females, increases in total bilirubin and urea nitrogen were seen. These clinical chemistries were not seen after recovery. In this study, the kidneys were identified as the target organs of toxicity in males treated with 750 mg/kg/day and in females treated with 500 mg/kg/day and 750 mg/kg/day. As with the clinical chemistries, all resveratrol-related histopathological findings in the kidneys were reversed following the 4-week recovery period. For male rabbits, the NOAEL was considered to be 500 mg/kg/day and 250 mg/kg/day for females.[12]

4.2.1 Subchronic Toxicity

In two studies conducted in CD rats and beagle dogs, rats received daily gavage doses of 0, 200, 400, or 1000 mg/kg bw of resveratrol for 90 days while dogs received daily orally doses of 0, 200, 600, or 1200 mg/kg bw of resveratrol for 91 days. No mortality was seen in either study nor were there any clinical observations of treatment-related gross toxicities. Female rats in all dose groups experienced a reduction in body weight during weeks 10-13; however, females at the highest dose experienced statistically significant decrease in body weight. For both male and female dogs, there was a statistically significant decrease in body weight at the highest dose that started at week 5 in male dogs and at week 6 in female dogs. Other clinical findings for rats included a small, but statistically significant increase in mean total bilirubin levels in both sexes of rats in the high dose group at study termination. Additionally, a modest but statistically significant and dose-related hepatomegaly was seen in both male and female rats. There was no evidence of any neurotoxicity in either sex. For dogs, only minimal toxicity was seen. Treatment-related changes were limited to inflammatory infiltrates in the urinary bladder and kidneys of which the severity ranged from minimal to mild with most dogs demonstrating only changes of minimal severity. No other changes were noted in other organs or tissues. The NOAEL for rats was determined to be 200 mg/kg/day and 600 mg/kg/day for dogs. The NOAEL for both species was determined on the decreases in body weight.[13]

4.2.2 Chronic Toxicity

A 6-month rat study was conducted using oral doses of resveratrol at 300, 1000, and 2,000 mg/kg/day or placebo. Weight loss occurred in the 2,000 mg/kg group. Total bilirubin values were increased in the 1000 mg/kg group with the values returning to normal after the recovery period. The NOAEL was considered to be 300 mg/kg/day.[12] Another 6-month study using oral daily doses of 100, 300, and 500 mg/kg or placebo was conducted in rabbits. No drug-related mortality was observed during the study and there were no apparent drug-related clinical abnormalities. For animals in the 500 mg/kg dose group, modest increases were seen in clinical chemistry parameters of total protein, albumin, GGT, globulin, glucose, urea nitrogen, indirect bilirubin and calcium along with decreases in potassium, but these findings were not present in the recovery animals. The NOAEL was considered to be 500 mg/kg/day.[12]

4.2.3 Reproductive Toxicity

A study was conducted in rats using oral daily doses of 300, 1000, and 3000 mg/kg or placebo administered on days 7 through 17 of presumed gestation. Five of the 25 animals in the 3000 mg/kg dose group were sacrificed due to adverse clinical conditions. No effects on the fetus were noted. The maternal NOAEL was considered to be 300 mg/kg/day and the developmental NOAEL was considered to be 1000 mg/kg/day.[12]

4.3 Clinical

4.3.1 Safety

In a 12-week clinical study in Friedreich's ataxia patients including 27 participants who received the study drug, 24 completed the study. Thirteen patients received 1 g/day of resveratrol while the other 14 patients received 5 g/day of resveratrol. No serious adverse events were noted.

Gastrointestinal (GI) side effects were the most frequent occurring adverse effects and were deemed to be dose-related.

The following side effects were seen in 2% to 15% (except if otherwise stated) of the 13 patients with Friedreich's ataxia taking resveratrol 500 mg twice daily (total daily dose: 1000 mg) for 12 weeks:

Infections

- Urinary tract infection (23%)
- Tonsilitis (inflammation of tonsil)
- Inflammation of the sinus (sinusitis)

Nervous system disorders

- Headache (31%)
- Fatigue (23%)

Gastrointestinal disorders

- Loose stools
- Abdominal pain/cramps
- Bloating
- Diarrhea
- Nausea
- Indigestion/upset stomach

Respiratory disorders

- Upper respiratory tract infection (31%)

Cardiac disorders

- Rapid and irregular heartbeat
- Increase in the level of substances (called enzymes) in blood that may indicate heart problems

Kidney and urinary disorders

- Proteins in urine

Liver, gallbladder or pancreas disorders

- Increase in the level of substances (called enzymes) in blood that may indicate liver problems

Overall, 1,000 mg/day resveratrol was well tolerated.[14]

Another randomized, placebo-controlled, double-blind, multicenter 52-week trial of resveratrol in subjects with mild to moderate Alzheimer's disease examined the drug's safety and tolerability. A total of 119 individuals were randomized and 104 completed through to week 52. Fifty-five subjects received placebo. The starting dose was 500 mg/day of resveratrol or matching placebo. The dose was escalated at 500 mg increments every 13 weeks as follows: 500 mg/day am; 500 mg twice a day; 1000 mg am and 500 mg pm; and 1000 mg twice a day. No differences between the resveratrol and placebo groups were found on vital signs, physical examinations, or neurologic examinations.

The side effects that were seen in 2% to 15% (except if otherwise stated) of the 64 patients with mild to moderate Alzheimer's disease who received resveratrol and in equal or greater frequency than in the placebo group are listed below. These patients were taking resveratrol 500 mg/day ending with 1000 mg twice daily (total daily dose of resveratrol: 2000 mg).[15]

Infections and infestations (42%)

Nervous system disorders (39%)

- Headache

Gastrointestinal disorders (42%)

- Diarrhea (41%)
- Nausea

Psychiatric disorders (36%)

General disorders

- Weight loss (17%)
- Fall (34%)

Respiratory problem (20%)

Vascular disorders

4.3.2 Pharmacokinetics

There are many published human studies dealing with the pharmacokinetics, metabolism, and disposition of resveratrol. Absorption, bioavailability, and metabolism of ^{14}C-resveratrol after oral and intravenous (IV) doses was investigated in six human volunteers. The absorption of a dietary relevant 25-mg oral dose was at least 70%, but poorly bioavailable (≤1%).[6,16] Peak plasma concentrations of trans-resveratrol were reached at 0.8-1.5 h following oral trans-resveratrol administration at doses 25, 50, 100 or 150 mg, six times/day.[17] A second peak of plasma concentration occurring at 6 hours after the oral dose (but not after the IV dose) may be due to enterohepatic recirculation of conjugated metabolites by reabsorption after intestinal hydrolysis.[16] Absorption is reduced when resveratrol is given with a high fat meal with a delayed time of peak plasma concentrations.[18] There was an increase in systemic exposure to trans-resveratrol with increasing doses of 25-150 mg. Over this dose range, there was a more than dose-proportional increase in AUC_{0-t} and C_{max}. The dose proportionality factor was 2.8 for C_{max} and 6.6 for AUC_{0-t} following a single dose. With repeated doses, steady-state was reached at least by dose 7.[17]

Resveratrol has poor water solubility and thus has to be bound to plasma proteins to assure its body distribution and bioavailability. The plasma protein binding of resveratrol in human plasma was 98%.[19]

Following oral trans-resveratrol, three metabolic pathways were identified, i.e., sulfate and glucuronic acid conjugation of the phenolic groups and hydrogenation of the aliphatic double bond, the latter likely produced by the intestinal microflora. There was no evidence of enzymatic

oxidation of resveratrol. The rapid appearance of a sulfate conjugate pair in plasma at 2 hours after either IV or oral doses suggests that sulfation by the intestine/liver may indicate a rate-limiting step in resveratrol's bioavailability. Although presystemic metabolism of resveratrol may be important for the oral dose, iv doses also demonstrate highly efficient systemic metabolism. In humans, the major metabolites were the glucuronide- and sulfate-conjugates of resveratrol and of dihydro-resveratrol, with most of the dose (64 to 70%) recovered in the urine. The total recovery of glucuronic and sulfate conjugations in human urine and feces was about 83 % after oral doses and 74 % after intravenous doses. Resveratrol was shown to inhibit the phenotypical index of CYP3A4, CYP2D6 and CYP2C9 and was able to induce the phenotypical index of CYP1A2. On the other hand, GST and UGT1A1 activity was minimally affected.[20] Most of the oral dose was recovered in urine. Plasma half-life is 9.2 ± 0.6 hours.[16] An almost complete elimination of resveratrol and its metabolites from tissues is observed by 72 h after a single dose.[21]

Additional PK parameters are shown in Table 1 and concentration versus time profiles in Figure 1.[22] Further detailed PK information can be found in the Investigator Brochure.[10]

4.4 Rationale for Study Conduct

Absorption of micronized resveratrol is good (~70%) but the compound is almost totally eliminated by first pass metabolism, primarily to glucuronides and sulfates.[10] One of the biggest challenges in translating the therapeutic effects of resveratrol is the poor bioavailability of the drug when it is administered orally. Resveratrol is eliminated from the body extremely quickly and maintaining a therapeutic level in the bloodstream is difficult.[16,23] In an attempt to improve the oral bioavailability of resveratrol, various formulations and drug delivery systems have been tested but none have been forthcoming, with the exception of multiparticulate formulations, which have shown increased bioavailability.[24] Results have shown that high doses of resveratrol are needed to affect disease state; however this causes serious side effects that limit treatment utilization. To bypass the first pass effect, a micellar formulation could allow access to lymphatic distribution. The structure of the product micelle, with a size of appr. 30 nm, is similar to the structure of the naturally formed physiological mixed micelles containing water-insoluble compounds in its core which is enclosed by ambiphilic molecules. The higher bioavailability, which is obtained by the product micellation, is based on the independence of this transport system from the limiting parameters in the formation of the physiological mixed micelle. It is important to appreciate the product micelle's character as an "encapsulation and transport medium". This must not be confused with "nanoparticles" which are often spoken about in nanotechnology. Jupiter Orphan Therapeutics (JOT) is developing a new formulation (called JOTROL) of trans-resveratrol (3,4',5-trihydroxy-trans stilbene) for the treatment of Mucopolysaccharidosis type I (MPS I) and Friedreich ataxia (FRDA). JOTROL is a micellar 10% resveratrol solubilization formulation that is thought to increase the amount of resveratrol and metabolite concentrations in the blood via the lymphatic system.[10]

4.5 Rationale for the Study Population

As above mentioned, published data have shown that resveratrol protects against some neurodegenerative diseases, such as Alzheimer's disease and obesity as well as is effective in the management of osteoporosis in post-menopausal women. In order to assess the potential clinical

use of resveratrol in usual populations with these medical conditions, the upper age limit of study participants will be 75 years.

Since there have been no studies with pregnant women, it is uncertain whether there is human fetal risk associated with the use of resveratrol. Therefore, non-pregnant, non-lactating females will be included in the study. In addition, females of childbearing potential will be included if they use appropriate methods of contraception.

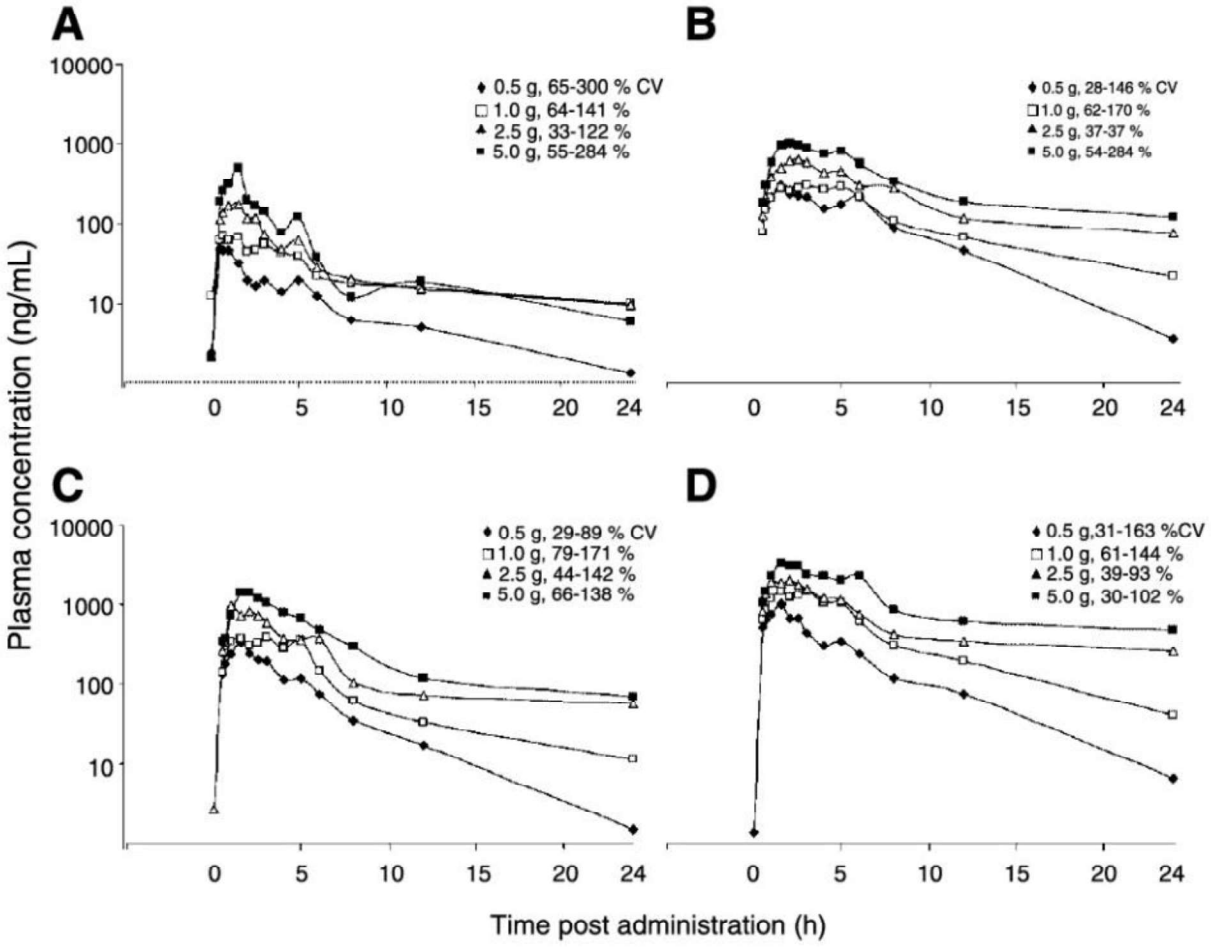
Given the fact that smoking may alter the metabolism of resveratrol, smokers will not be included in this study.

Table 1 Plasma PK of Resveratrol, Two Resveratrol Monoglucuronides and Resveratrol 3-sulfate After Single Oral Doses of Drug

Variable	Dose level (g)			
	0.5	1.0	2.5	5.0
Resveratrol				
AUC_{inf} (ng h/mL)	223.7*	544.8 (57.2)	786.5 (36.2)	1,319 (59.1)
C_{max} (ng/mL)	72.6 (48.9)	117.0 (73.1)	268.0 (55.3)	538.8 (72.5)
T_{max} (h)	0.833 (0.5-1.5)	0.759 (0.5-4.0)	1.375 (0.5-4.0)	1.500 (0.67-5.0)
C_{av} (ng/mL)	8.36 (57.8)	18.04 (51.6)	32.25 (43.0)	51.90 (80.7)
Half-life (h)	2.85*	8.87 (91.1)	4.22 (51.6)	8.52 (95.8)
CL/F (L/h)	2,235*	2,593 (65.1)	3,471 (29.9)	4,930 (50.0)
CL_R (L/h)	1.177 (102.5)	0.696 (71.5)	0.656 (53.1)	1.443 (139.2)
V/F (liters)	9,198*	19,298 (54.3)	22,226 (67.3)	66,991 (112)
Glucuronide 1				
AUC_{inf} (ng h/mL)	1,919 (33.6)	3,059 (60.9)	5,664 (27.7)	9,923 (40.9)
C_{max} (ng/mL)	404.6 (35.3)	473.6 (76.8)	874.4 (37.5)	1,285 (55.4)
T_{max} (h)	2.00 (1.0-6.0)	2.250 (1.0-6.0)	2.375 (1.0-8.0)	2.00 (1.5-5.0)
C_{av} (ng/mL)	76.9 (37.2)	110.3 (56.1)	215.5 (43.5)	344.1 (51.5)
Half-life (h)	2.85 (48.6)	7.27 (93.9)	10.6 (92.9)	7.90 (39.1)
CL/F (L/h)	282.7 (27.3)	493.5 (74.7)	469.5 (25.7)	590.6 (45.2)
Glucuronide 2				
AUC_{inf} (ng h/mL)	1,287 (21.7)	2,589 (66.4)	4,320 (32.9)	8,546 (62.3)
C_{max} (ng/mL)	369.5 (39.6)	672.6 (81.1)	1,626 (71.5)	1,735 (66.4)
T_{max} (h)	1.500 (1.0-5.0)	1.750 (1.0-5.1)	2.000 (1.0-6.0)	2.520 (1.5-8.0)
C_{av} (ng/mL)	51.0 (27.6)	99.9 (66.2)	193.8 (39.3)	317.8 (65.6)
Half-life (h)	3.09 (69.8)	6.64 (92.1)	8.42 (88.9)	5.83 (51.2)
CL/F (L/h)	408.8 (26.7)	642.5 (83.0)	636.9 (32.6)	1,017 (94.6)
3-Sulfate				
AUC_{inf} (ng h/mL)	4,049 (26.6)	10,053 (73.2)	16,984 (41.7)	30,898 (46.1)
C_{max} (ng/mL)	1,135 (25.7)	2,102 (81.3)	2,786 (27.2)	4,294 (48.0)
T_{max} (h)	1.500 (1.0-5.0)	2.000 (1.0-5.0)	2.000 (1.0-5.2)	2.050 (1.0-6.0)
C_{av} (ng/mL)	172.0 (23.2)	402.6 (70.5)	597.0 (27.0)	1,089 (49.4)
Half-life (h)	3.21 (56.6)	4.51 (82.8)	11.5 (95.5)	7.71 (42.3)
CL/F (L/h)	131.2 (25.8)	151.8 (62.7)	171.2 (40.0)	207.8 (63.9)

NOTE: Values are the mean of n = 10 with coefficient of variation (in percent) or range in brackets.
Abbreviations: AUC_{inf}, area under the concentration versus time curve to time infinity; C_{max}, maximal plasma concentration; T_{max}, median time of maximal plasma concentration; C_{av}, average plasma concentration; CL/F, apparent total body clearance (calculated as dose/AUC_{inf}); CL_R, apparent renal clearance approximated by amount excreted with urine within 24 h over AUC_{0-24}; V/F, apparent volume of distribution.
*n = 1, value for AUC_{inf} at the lowest dose could be established in only one participant.

Figure 1 Mean Plasma Concentrations of Resveratrol (A), Two Resveratrol Monoglucuronides (B and C), and Resveratrol-3-sulfate (D) Versus Time in Healthy Volunteers Who Received a Single Dose of Resveratrol at 0.5, 1.0, 2.5, or 5.0 g Doses.



5. Objectives

The objectives of this study are:

- To characterize the PK profile of JOTROL (resveratrol) following oral administration of SAD ranging from 200 mg up to a dose currently estimated at 1000 mg, in healthy adult subjects;
- To evaluate the effect of food on the PK profile of JOTROL.

6. Study Design

This will be a single center, Phase 1, open-label, sequential SAD study, with a food-effect arm. The study will be divided into two parts:

- Study Part 1 consists of 3 periods with SAD of JOTROL under fasting conditions. Periods 2 and 3 will be initiated with updated doses after safety, tolerability, and PK data are evaluated by the Safety Committee **and the DSMB** and deemed acceptable for single doses for subsequent doses.
- Study Part 2 consists of a single oral JOTROL dose under fed conditions. JOTROL dose for this study part will derive from study Part 1 safety, tolerability, and PK data.

This study is intended for filing under FDA regulations.

7. Rationale for Dose Selection

7.1 Starting Dose

Diarrhea or other gastrointestinal symptoms were reported in clinical studies in healthy volunteers at doses of 1,000 mg/day or higher of unmodified resveratrol. Studies do not indicate significant adverse effects below 1,000 mg.[20,25] In addition, results of metabolic interactions with cytochrome P-450 complex enzymes at doses higher than 1,000 mg/day suggested that this dose of unmodified resveratrol is the upper limit for clinical studies.[20] Considering different NOAEL values obtained with different animal species in subchronic and chronic toxicity studies (sections 4.2.1, 4.2.2, and 4.2.3) that are within 200 mg/kg and 700 mg/kg[25] it may be assumed that resveratrol shows a good safety profile within a certain range dose. In addition, a dose of 50 mg/kg of study product JOTROL administered to rats produced an AUC (2.5 µg•hr/mL) roughly equivalent to that seen after a 2,500 mg dose in humans.[10] Applying a security factor of 10, the entry dose might be 250 mg. Consequently, a starting dose of resveratrol 200 mg can be considered safe.

7.2 Maximum Dose in this Study

The FDA preliminary review of the published data on resveratrol shows that the nonclinical data can only support daily oral doses of unmodified resveratrol (i.e. non-micronized drug with no absorption enhancers) of up to 3,000 mg/day for no more than 13 weeks. Published data from a 26-week study in mice demonstrated renal toxicity at 1,000 mg/kg/day, for which the Human Equivalent Dose is approximately 5,000 mg. Based on published data in healthy volunteers, AUC exposures at a dose of 3,000 mg/day of unmodified resveratrol is estimated to be

2.1 µg•hr/mL. Because JOTROL formulation has been modified to enhance bioavailability, JOTROL doses to be used in this study should produce AUC exposures less than 2.1 µg•hr/mL.[26]

7.3 Dose Escalation Scheme (Study Part I)

The dose escalation scheme for this SAD study (Part 1) was established considering the following assumptions:

- PK animal data (rat, with a dose of 50 mg/kg) suggest that resveratrol administered as JOTROL produce an AUC (2.5 µg•hr/mL) roughly equivalent to that seen after a 2,500 mg dose in humans;
- AUC exposures at a dose of 3,000 mg/day of unmodified resveratrol is estimated to be 2.1 µg•hr/mL. AUC exposures during this study must not be higher than 2.1 µg•hr/mL;
- The bioavailability of resveratrol from JOTROL increases linearly with the dose.

The nominal planned dose escalation scheme for Study Part I corresponds to a 2.5-fold increment between the two first doses, and 2 fold increment between the second and third dose levels. Both current second and third dose levels are nominal as JOTROL's doses to be administered in Periods 2 and 3 will be reassessed when safety, tolerability, and PK data analysis are available. These dose levels will be adjusted, lowered or increased, but in any case corresponding AUC exposures must be lower than 2.1 µg•hr/mL.

7.4 Dose Level in Study Part II

The same dose is planned to be administered in Periods 3 (Part I) and 4 (Part 2). However, if the exposure is higher than expected in Period 3, the dose to be administered in Period 4 could be lower, in order to keep AUC exposures lower than 2.1 µg•hr/mL, as specified in section 7.2.

7.5 Dose Escalation Criteria

Each dose level will be dosed sequentially in an ascending fashion, with at least 14 days between dosing of consecutive dose levels. Following completion of each level, safety, tolerability, and PK data will be evaluated by a Safety Committee **and a DSMB** before proceeding to the next dose. The Safety Committee **and the DSMB** will review the whole safety and tolerability data, as well as PK data up to 32 hours post-dose, in order to make decisions regarding progression to the next prescribed dose level, decreasing the next dose level, repeating a dose level or not evaluating any additional dose level, based on consideration of the clinical significance of several safety and tolerability parameters, as well as PK data. The Safety Committee will be composed by at least the Investigator, one medically qualified Sponsor representative, and an independent third party physician. **A formal DSMB will also be established and constituted by an independent group of experts (totally independent of Syneos Health, JOT, and the associated investigators on the study) that advises the NIA Director and the study investigator. The members of the DSMB serve in an individual capacity and provide their expertise and recommendations. The DSMB will be constituted and will act according to the FDA Guidance for Clinical Trial Sponsors - Establishment and Operation of Clinical Trial Data Monitoring Committees[1] and the NIA Guidance on Clinical Trials.[2]** Dose

escalation may occur only following mutual agreement between the Safety Committee **and the DSMB** members.

Minutes of the safety review committee meeting **and the DSMB** will be prepared and signed by all voting participants. Minutes will include the decision to escalate the dose, confirmation of the next dose level, confirmation of the safety monitoring and washout period, rationale for the decisions and supportive data. The minutes will be shared with the Independent ethics committee (IEC)/Institutional review board (IRB).

Some adjustments to the currently outlined doses may be implemented by the Safety Committee, **or the DSMB** but the dose to be administered at a given dose level will not exceed the dose corresponding to AUC exposures of 2.1 μg•hr/mL.

Stopping Rules for Dose Escalation

Dose escalation will be halted if any of the events described below occur within one dose level:

- Individual stopping rules met for ≥ 2 subjects in the same study period suggesting that subjects receiving higher dose levels would be at risk for similar adverse drug reactions.
- The occurrence of one serious adverse event (SAE) considered at least possibly related to the study drug.
- The occurrence of severe AEs considered at least possibly related to the study drug in ≥ 2 subjects and independent of within or not within the same system organ class (SOC).
- Clinically significant abnormalities of the same character, at least of moderate severity, and considered at least possibly related to the study drug in ≥ 2 subjects

Cumulative Study Events

In addition to the events listed above, any of the events described below occurring at different dose levels administered throughout the study (i.e., cumulative events) will also result in a temporary halt of dose escalation:

- The occurrence of severe AEs of the same character considered at least possibly related to the study drug in ≥ 2 subjects.
- Clinically significant abnormalities of the same character, at least of moderate severity, and considered at least possibly related to the study drug in ≥ 3 subjects
- Determination by the SRC that a pattern of AEs precludes further dose escalation, even if no other stopping rules have been met.

Study Stopping Rules

The study will be terminated if any of the following criteria are met:

- The occurrence SAEs considered at least possibly related to the study drug in ≥ 2 subjects and independent of within or not within the same SOC.

- Determination by the SRC that a pattern of AEs precludes any further dosing.

8. Study Population

8.1 Sample Size

A total of 24 healthy adult male or female volunteers will be included in study Part 1. Only 16 subjects who completed study Part 1 will be included in study Part 2. In order to minimize potential PK variability, elderly (more than 65 years old subjects) will be not included in study Part 2. These 16 subjects will be selected according to their order of enrolment in the study, provided their consent to continue the study, they follow the study restrictions and they still meet study criteria. Subjects numbers are judged adequate to achieve the study objectives.

For study Part 1 only, an effort will be made to include to the extent possible, subjects of the following age groups:

- ≥ 65 and ≤ 70 years of age;
- ≥ 70 and ≤ 75 years of age.

8.2 Inclusion Criteria

Subjects enrolled in this study will be members of the community at large. The recruitment advertisements may use various media types (e.g., radio, newspaper, the clinical site Web site and volunteer database). Subjects must meet all of the following criteria to be included in the study:

1) Normal healthy male or female volunteers, non-smokers (no use of tobacco products within 3 months prior to screening), ≥ 18 and ≤ 75 years of age, with BMI > 18.5 and < 30.0 kg/m^2 and body weight ≥ 50.0 kg for males and ≥ 45.0 kg for females.
2) Healthy as defined by:
   a) the absence of clinically significant illness and surgery within 4 weeks prior to dosing. Subjects vomiting within 24 hours predose will be carefully evaluated for upcoming illness/disease. Inclusion pre-dosing is at the discretion of the Investigator.
   b) the absence of clinically significant history of neurological, endocrine, cardiovascular, respiratory, hematological, immunological, psychiatric, gastrointestinal, renal, hepatic, and metabolic disease as determined by the Investigator.
3) Females of childbearing potential who are sexually active with a male partner must be willing to use one of the following acceptable contraceptive methods throughout the study and for 30 days after the last study drug administration:
   a) intra-uterine contraceptive device without hormone release system placed at least 4 weeks prior to study drug administration;
   b) male condom with intravaginally applied spermicide starting at least 21 days prior to study drug administration;
   c) sterile male partner (vasectomized since at least 6 months).
4) Capable of consent

8.3 Exclusion Criteria

Subjects to whom any of the following applies will be excluded from the study:

1) Any clinically significant abnormality at physical examination, clinically significant abnormal laboratory test results or positive test for hepatitis B, hepatitis C, or HIV found during medical screening.

2) Positive urine drug screen or urine cotinine test at screening.

3) History of allergic reactions to resveratrol, polyphenols, other related drugs, or to any excipient in the formulation.

4) Positive pregnancy test at screening.

5) Breast-feeding subject.

6) Clinically significant ECG abnormalities or vital sign abnormalities (systolic blood pressure lower than 90 or over 140 mmHg, diastolic blood pressure lower than 50 or over 90 mmHg, or heart rate less than 50 or over 100 bpm) at screening.

7) History of significant alcohol abuse within 1 year prior to screening or regular use of alcohol within 6 months prior to the screening visit (more than 14 units of alcohol per week [1 unit = 150 mL of wine, 360 mL of beer, or 45 mL of 40% alcohol]).

8) History of significant drug abuse within 1 year prior to screening or use of soft drugs (such as marijuana) within 3 months prior to the screening visit or hard drugs (such as cocaine, phencyclidine [PCP], crack, opioid derivatives including heroin, and amphetamine derivatives) within 1 year prior to screening.

9) Use of resveratrol for a medical condition or in the context of another clinical trial within a period of 30 days prior to the first dosing.

10) Participation in a clinical research study involving the administration of an investigational or marketed drug or device within 30 days prior to the first dosing, administration of a biological product in the context of a clinical research study within 90 days prior to the first dosing, or concomitant participation in an investigational study involving no drug or device administration.

11) Use of medications for the timeframes specified below, with the exception of medications exempted by the Investigator on a case-by-case basis because they are judged unlikely to affect the PK profile of the study drug or subject safety (e.g., topical drug products without significant systemic absorption:

    a) prescription medication within 14 days prior to the first dosing;

    b) over-the-counter products and natural health products (including herbal remedies, homeopathic and traditional medicines, probiotics, food supplements such as vitamins, minerals, amino acids, essential fatty acids, and protein supplements used in sports) within 14 days prior to the first dosing, with the exception of the occasional use of acetaminophen (up to 2 g daily);

c) use of any drugs known to induce or inhibit hepatic drug metabolism, including St. John's wort, within 30 days prior to the first study drug administration;

d) depot injection or an implant of any drug within 3 months prior to the first dosing.

12) Donation of plasma within 7 days prior to dosing. Donation or loss of blood (excluding volume drawn at screening) of 50 mL to 499 mL of blood within 30 days, or more than 499 mL within 56 days prior to the first dosing.

13) Any reason which, in the opinion of the Investigator, would prevent the subject from participating in the study.

9. Clinical Procedures

Unless otherwise specified, procedures, data collection and evaluation will be conducted as per the clinical site SOPs.

9.1 Screening Procedures

Subject screening procedures will be performed within 28 days preceding administration of study medication. Subjects must provide written informed consent prior to initiation of any screening procedures. The consent to perform some general screening procedures may be obtained on a consent document other than the Informed Consent Form (ICF) specific to this study, and therefore, some screening test results could be obtained before signature of the ICF specific to this study. The study-specific ICF must be signed and dated by the subject before participation to study-specific procedures.

Screening procedures will include: demographic data, medical and medication histories, physical examination, body measurements, ECG (12-lead), vital signs (blood pressure, heart rate, respiratory rate, and oral temperature), hematology, biochemistry, coagulation, serology (HIV, hepatitis B and C tests), urinalysis, urine cotinine test, urine pregnancy test, and urine drug screen.

For eligibility purposes, abnormal laboratory or vital signs results may be repeated once if abnormal result is observed at the initial reading. Moreover, abnormalities found in the ECG may need to be confirmed by repeated measurements. In the event that the participation of a subject in the study is delayed and some screening procedures had been performed outside the prescribed screening window, outdated screening procedures can be repeated.

9.2 Confinements, Visits, and Washout

For each period, subjects will be confined from Day -1 until after the 32-hour post-dose blood draw.

There will be a washout of 14 days or more between doses. The washout period may be increased for logistical considerations. Participation of each subject in this study should last approximately 1 month (for subjects participating in study Part 1 only) and 1.5 months (for subjects participating in both study parts).

9.3 Study Medication

Treatment A (Period 1): JOTROL (resveratrol) 100 mg oral gelcap (Jupiter Oprhan Therapeutics, USA) given as 2 x 100 mg gelcaps under fasting conditions, total resveratrol dose: 200 mg

Treatment B (Period 2): JOTROL (resveratrol) 100 mg oral gelcap (Jupiter Oprhan Therapeutics, USA) given as 5 x 100 mg gelcaps under fasting conditions, total resveratrol dose: 500 mg

Treatment C (Period 3): JOTROL (resveratrol) 100 mg oral gelcap (Jupiter Oprhan Therapeutics, USA) given as 10 x 100 mg gelcaps under fasting conditions, total resveratrol dose: 1000 mg

Treatment D (Period 4): JOTROL (resveratrol) 100 mg oral gelcap (Jupiter Oprhan Therapeutics, USA) given as 10 x 100 mg gelcaps under fed conditions, total resveratrol dose: 1000 mg

Targeted doses will be achieved with multiple JOTROL' gelcaps containing 100 mg of resveratrol. For Periods 2 and 3, JOTROL's doses to be administered will be reassessed when safety, tolerability, and PK data analysis are available. The same dose is planned to be administered in Periods 3 and 4. However, if the exposure is higher than expected in Period 3, the dose administered could be lower in Period 4, in order to keep AUC exposures lower than 2.1 μg•hr/mL, as specified in section 7.2.

9.4 Drug Supplies and Accountability

It is the responsibility of the Sponsor to ensure that study medication provided for this study are manufactured under Good Manufacturing Practices (GMP) and are suitable for human use. The Sponsor is responsible to ship a sufficient amount of dosage units to allow the clinical site to maintain an appropriate sampling for the study.

Study medication will be stored at the clinical site as per applicable requirements. The medications will be stored in a locked, environmentally-controlled medication room with restricted access. Container(s) will bear a label containing at least the name of the study drug, lot and/or batch number, and manufacturing and/or expiry/retest date.

Individual doses for each subject will be dispensed at the clinical site, as per appropriate SOP. Individual doses will be dispensed in appropriate envelopes/containers indicated with at least the project number and the subject number/spare number.

All study drug received at the site will be inventoried and accounted for throughout the study and the result recorded in the drug accountability/retention record according to the clinical site appropriate SOP.

9.5 Drug Administration

Study medication will be administered to each subject with approximately 240 mL of water and a hand and mouth check will be performed to ensure consumption of the medication. Additional water may be provided if needed.

Study Part 1 (Periods 1, 2, and 3): All subjects will be sequentially dosed in an ascending manner with at least 14 days between the last dosing and the subsequent nominal dose level which will be calculated based on observed safety, tolerability, and PK results. Subjects will receive a single oral dose of JOTROL gelcaps under fasting conditions using the following nominal dosage levels:

Study period	Planned dose levels of JOTROL
1	200 mg (given as 2 x 100 mg gelcaps), total dose of resveratrol: 200 mg
2	500 mg (given as 5 x 100 mg gelcaps), total dose of resveratrol: 500 mg
3	1000 mg (as 10 x 100 mg gelcaps), total dose of resveratrol: 1000 mg

For Periods 2 and 3, JOTROL's doses to be administered will be reassessed when safety, tolerability, and PK data analysis are available. Adjustment to the currently outlined doses and/or dosing regimen may be necessary, but the dose to be administered in Period 2 or 3 will not exceed the one currently outlined in the protocol.

Time of dosing will be set equal to the time when the first gelcap is administered to the subject. The complete dosing procedure must be completed within 3 minutes.

Following completion of each dose level, PK, safety, tolerability data will be evaluated by a Safety Committee **and a DSMB** before proceeding to the next dose.

The same JOTROL dose will be administered in Periods 3 and 4.

Study Part 2 (Period 4): Only 16 subjects who completed study Part 1 will receive the highest dose of JOTROL tested during study Part 1, as a single oral dose of JOTROL gelcaps under fed conditions. In order to minimize potential PK variability, elderly (more than 65 years old subjects) will be not included in study Part 2. However, depending on safety and tolerability as well as on available PK data from Part 1 (in order to keep AUC exposures lower than 2.1 µg•hr/mL, as specified in section 7.2.), the dose to be used during study Part 2 may be reduced.

9.6 Study Restrictions

9.6.1 Food and Fluids

Study Part 1 (Periods 1, 2, and 3): No food will be allowed from at least 10 hours before dosing until at least 4 hours after dosing.

Study Part 2 (Period 4): After a supervised fast of at least 11 hours, subjects will be served a critical, high-fat, high-calorie meal of approximately 800 to 1000 calories (approximately 50% of total caloric content of the meal derived from fat). This test meal should derive approximately 150, 250, and 500-600 calories from protein, carbohydrate, and fat, respectively. The meal will consist of two eggs fried in butter, two slices of toast with butter, two strips of bacon, 120 g of hash brown potatoes, and 240 mL of whole milk. Subjects will be informed to start their meal as soon as it is served and to complete it in 30 minutes or less. Subjects will be informed that they should eat the entire meal. Drug administration will occur 30 ± 1 minutes after the meal has been started. Subjects will fast for not less than 4 hours after drug administration.

Meals will be standardized and similar in composition between periods.

Except for fluids provided with the critical breakfast (study Part 2 only) and water given with study medication, no fluids will be allowed from 1 hour before dosing until 1 hour post-dose. Water will be provided *ad libitum* at all other times.

In addition, subjects will be required to abstain from:

- food containing poppy seeds within 24 hours prior to admission;
- food or beverages containing xanthine derivatives or xanthine-related compounds or energy drinks from 48 hours prior to dosing until after the last PK blood sample collection of each period;
- food or beverages containing grapefruit, starfruit, pomegranate, pineapple, or pomelo from 7 days prior to dosing until after the last PK blood sample collection of each period;
- foods rich in resveratrol (e.g. grapes, peanuts and their derived-products including wines and juices) from 7 days pre-dose until after the last PK blood sample collection of each period.

9.6.2 Tobacco, Alcohol, and Illicit Drugs

Subjects will be required to abstain from using soft or hard drugs or any tobacco products from screening and throughout the study.

Consumption of alcohol-based products will be prohibited from 24 hours prior to admission until after the last PK blood sample collection of each period.

9.6.3 Concomitant Medications

Subjects will be required to avoid using prescription and over-the-counter medications for the period of time specified in exclusion criterion 11) and throughout the study. Subjects will be required to avoid using natural health products (including herbal remedies, homeopathic and traditional medicines, probiotics, food supplements such as vitamins, minerals, amino acids, essential fatty acids, and protein supplements used in sports) from 14 days prior to dosing until after the last PK blood sample collection of each period.

No concomitant medications are allowed during the study, with the exception of one(s) required for the medical management of an adverse event, medications exempted by the Investigator on a case-by-case basis that are judged unlikely to affect the pharmacokinetic profile of the study drug or subject safety (e.g., topical drug products without significant systemic absorption) and occasional use of acetaminophen.

All medications taken by subjects after screening until the last study day will be documented as concomitant medications. Any concomitant medication use, other than the allowed medications stated above, will be reviewed and evaluated on a case-by-case basis by the Investigator to determine if they affect a subject's eligibility or continued participation in the study, or for potential impact on the study results.

9.6.4 Posture and Physical Activity

Subjects will be allowed to engage in normal activity but will avoid lying down or sleeping, unless medically necessary or procedurally required, for 2 hours (study Part 1) and 4 hours (study Part 2) after drug administration. Vigorous activity will be prohibited at all times during the confinement.

9.7 Sample Collection and Processing

9.7.1 Blood Sample Collection

In each period :

- a total of 17 blood samples will be drawn from each subject for PK analyses. Blood samples will be collected prior to drug administration and 0.133, 0.25, 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8, 10, 12, 16, 24, and 32 hours post-dose (3 mL for each sampling time). The time tolerance window for blood sample collection during the confinement period will be ±1 minute for all samples collected before 8 hours post-dose and ±3 minutes for subsequent samples. Sample collections done outside the pre-defined time windows will not be considered as protocol deviations since actual post-dose sampling times will be used for PK and statistical analyses. Unless otherwise specified or for subject safety, when blood draws and other procedures coincide, blood draws will have precedence. A saline intravenous catheter will be used for blood collection to avoid multiple skin punctures, when appropriate. Otherwise, blood samples will be collected by direct venipuncture.

- a total of 5 additional blood samples (for plasma and buffycoats) will be collected for potential RNA Seq analysis, iduronidase activity, and mRNA frataxin: pre-dose, and 3, 6, 12, and 24 hours post-dose (10 mL for each sampling time).

For study Part 1 only, PBMC will be isolated, from at most (maximum) 10 subjects at the following timepoints (16 mL for each sampling time):

- Period 1: pre-dose, 3, and 24 hours post dose;

- Period 2: 3, 6, and 24 hours post dose;

- Period 3: 3, 6, and 24 hours post dose.

PBMC will be isolated as per the Project Specific Procedure (PSP).

The total volume of blood including that collected for eligibility and safety purposes should not exceed 733 mL for the whole study (605 mL for subjects participating in study Part I only).

9.7.2 Urine Sample Collection for PK analysis

In study Part 1, urine will be collected for quantitation for PK analysis. Urine samples will be collected at 6 times or time intervals: spot pre-dose (within 2 hours before dosing), 0-4 hours, 4-8 hours, 8-12 hours, 12-24 hours, and 24-32 hours post-dose.

If a subject cannot void his or her bladder within 30 minutes before dosing, a sample from earlier that morning may be used as the pre-dose sample. Voids that occur within the time interval will

be pooled, and subjects will be asked to void their bladder within 10 minutes before the end of each collection interval, so that each new interval will begin with an empty bladder. Any urine voided by subjects at the intersection (within 10* minutes) of two intervals will be included in the earlier sample. Any urine voided by subjects but not collected will be documented.

**For Day 2 (24 hours post-dose) urine collection, subjects will be asked to void their bladder within 15 minutes before the end of the collection interval (12-24 hours).

Plasma and urine samples will be collected and processed as per the Analytical Methodology Information Sheet.

Since resveratrol is sensitive to light, blood and urine collection tubes will be protected from light, sample processing will be performed under sodium lamp, and urine and plasma samples will be transferred into amber vials.

Other investigations may be performed using remaining samples already collected.

9.8 Subject Monitoring

Subjects will be monitored throughout the study by the clinical staff for AEs. In each period, the Investigator or designee will be on site for drug administration and until 2 hours (study Part 1) and until 4 hours (study Part 2) post-dose, and available on call for the remainder of the study. If necessary, the Investigator or designee at the clinical site or a healthcare professional in a nearby hospital will administer treatment for any AE(s). A crash cart or emergency bag containing the necessary rescue material and appropriate medications will be available in the clinic to allow rapid intervention in case of emergency.

Safety parameters, including laboratory results and ECG, will be assessed by the Investigator or designee, using the clinical site's criteria for biomedical laboratory and ECG acceptance ranges as suggested guidelines in making the medical assessment.

Scheduled safety measurements will be repeated according to the clinical site SOPs or upon request from the Investigator or designee. Any abnormal repeated measurement will be evaluated by the Investigator or designee and repeated if judged necessary. Further action may be taken upon the Investigator or designee's request.

Subjects will be advised to notify their health care professional(s) (e.g. physician, dentist, and/or pharmacist) that they are participating in a clinical research study on a drug called resveratrol before taking any medicines or undergoing any medical procedure.

9.8.1 Vital Signs

Blood pressure, respiratory rate, oral temperature, and heart rate will be measured in a sitting position (except for safety reasons) at screening and study exit.

9.8.2 ECG

Supine ECG will be performed at screening and study exit.

9.8.3 Physical Examination

A physical examination will be performed at screening.

9.8.4 Cotinine, Drug, and Alcohol Screen

A urine drug screen (amphetamines, methamphetamines, barbiturates, benzodiazepines, tetrahydrocannabinol, cocaine, opiates, PCP, MDMA, methadone) and a urine cotinine test will be performed at screening. A urine drug screen, a urine cotinine test, and an alcohol breath test will be performed before dosing at check-in of each period.

9.8.5 Pregnancy Test

A urine pregnancy test will be performed at screening and at study exit, and a serum pregnancy test will be performed at check-in of each period.

9.8.6 Laboratory Assessments

9.8.6.1 RNA-Seq Analysis, Iduronidase, mRNA Frataxin, and Other Possible Tests

Potential RNA-Seq analysis will be performed before dosing and 3, 6, 12, and 24 hours post-dose in each period. Details for this analysis will be included in a separate protocol. Other investigational tests such as iduronidase activity measurement and mRNA frataxin may be performed by an external laboratory. Details of this additional laboratory work will be available in a separate protocol.

9.8.6.2 PBMC Isolation

PBMC will be isolated, from at most (maximum) 10 subjects on Days 1-2 in study Part 1.

9.8.6.3 Biochemistry

Biochemistry will be performed at screening and study exit. The following will be assessed: albumin, alkaline phosphatase, AST, ALT, urea, calcium, chloride, glucose, phosphorus, potassium, creatinine, sodium, total bilirubin, and total protein.

9.8.6.4 Serology

Serology will be performed at screening. The following will be assessed: HIV antigen and antibody, hepatitis B surface antigen (HBsAg), and hepatitis C virus (HCV) antibody.

9.8.6.5 Hematology

Hematology will be performed at screening, at check-in of **Period 3 (study Part 1) and of** study Part 2 (Period 4), and at study exit. The following will be assessed: complete blood count with differential, hemoglobin, and hematocrit.

9.8.6.6 Coagulation

Coagulation tests will be performed at screening. The following will be assessed: prothrombin time/international normalized ratio (PT/INR) and activated partial thromboplastin time (aPTT).

9.8.6.7 Urinalysis

Urinalysis will be performed at screening and at study exit. The following will be assessed: macroscopic examination, pH, specific gravity, protein, glucose, ketones, bilirubin, occult blood, nitrite, urobilinogen, and leukocytes. Unless otherwise specified, microscopic examination will be performed according to internal procedures.

9.9 Study Exit Procedures

Hematology, biochemistry, urinalysis, urine pregnancy test, vital signs (blood pressure, respiratory rate, heart rate, and oral temperature), ECG, and AE monitoring will be performed on the last study day. For subjects who are not included in study Part 2, study exit procedures will be completed after the last blood draw of Period 3 (after 32 hours post-dose on Day 2). For subjects who are included in both study parts, study exit procedures will be completed after the last blood draw of Period 4 (after 32 hours post-dose on Day 2). If not possible, all efforts will be made to complete study exit procedures within 14 days after the last participation of the subject in the study.

9.10 Data Collection and Evaluation

Subjects' personal information will be stored in an electronic data capture (EDC) system (Initiator™ or Alphadas®). All clinical raw data will be recorded promptly, accurately, and legibly; either directly into the EDC system as e-source data or indelibly on paper (e.g., ECG readings or raw data sheets when electronic data capture is not possible). A detailed list of the type (electronic or paper) and location for all source data will be included in the Trial Master File. When recorded electronically, Case Report Forms will be electronically generated afterwards. All raw data will be conserved in order to maintain data integrity. The Investigator and/or the clinical staff have the responsibility of ensuring the completeness and accuracy of the clinical data. Initiator™ and Alphadas® are validated and are Code of Federal Regulations (CFR) part 11 compliant applications.

9.11 Subject Withdrawal and Replacement

Subjects will be advised that they are free to withdraw from the study at any time. Over the course of the study, the Sponsor and the Investigator or designee may withdraw any subject from the study for one of the reasons described below; subject withdrawal will be done in accordance with the clinical site's SOP:

- safety reason;
- non-compliance with protocol requirements;
- significant protocol deviation;
- positive cotinine, alcohol, drug, or pregnancy test;
- vomiting within 3 hours (study Part 1) and 7 hours (study Part 2) after dosing.

Subjects excluded from dosing in one period as per criteria listed above, may be invited to participate in subsequent periods of the study if deemed appropriate by the Investigator and appropriate from a statistical standpoint (i.e. would permit adequate statistical comparison). However, subjects with positive cotinine, alcohol, drug, or pregnancy test will be definitively withdrawn from the study. Hematology results will be reviewed by the Investigator or designee prior to dosing in Period 3 (study Part 1) and in Period 4 (study Part 2). Subjects will be

withdrawn from the study if it is deemed that the subject's safety may be at risk on the basis of these test results. Subjects who withdraw or are withdrawn from study Part 1 after dosing, for reasons other than safety and tolerability, may be replaced after consultation between the Safety Committee members **and the DSMB members** in order to assure initiating study Part 2 with 16 subjects. Such replacement resulting in dosing more subjects than planned in this protocol would be documented in a protocol amendment.

Subjects who withdraw or are withdrawn will be asked to remain at the clinic until the Investigator or a designee agrees that the subject is fine and can be discharged. As soon as subject withdrawal is confirmed, blood sampling will be stopped. A PK blood draw may be collected at the time of withdrawal if deemed required by the Investigator. Study exit procedures will be performed at the time of withdrawal from the study or as soon as possible thereafter.

9.12 Adverse Events

9.12.1 Recording of Adverse Events

AEs will be recorded and evaluated for their seriousness, severity, and relationship to the study medication. AEs will be collected and documented during the course of the study and until 4 days following the last study drug administration, if reported. AEs will be followed-up until complete resolution, or until the Investigator judges it to be safe to discontinue follow-up. The relationship to the study medication will be classified according to the clinical site SOPs.

9.12.2 Serious Adverse Events

9.12.2.1 Serious Adverse Event Reporting to the Sponsor

Any SAE will be reported to the Sponsor via telephone, fax, e-mail or in person, within 24 hours of knowledge by the Investigator, and then in writing as soon as possible, but no later than 7 calendar days after first knowledge of the SAE. The notification must be directed to:

PPD , Ph.D.
PPD

Jupiter Orphan Therapeutics
601 Heritage Drive
Jupiter, Florida 33458
Tel.: PPD

9.12.2.2 Serious Adverse Event Reporting to Regulatory Agency(ies)

The Sponsor is responsible for notifying the FDA of suspected, unexpected, serious adverse drug reactions observed during conduct of studies in which the investigational drug is administered.

FDA notification of fatal or life-threatening suspected, unexpected, serious adverse drug reaction must be made as soon as possible, but no later than 7 calendar days after becoming aware of the information. FDA notification of all other suspected, unexpected, serious adverse drug reactions that are neither fatal nor life-threatening must be made as soon as possible, but no later than 15 calendar days after becoming aware of the information.

The Sponsor is responsible to comply with any other applicable regulatory requirement(s) related to the reporting of SAE to other regulatory authority(ies).

9.12.2.3 Serious Adverse Event Reporting to the Independent Ethics Committee

It is the responsibility of Syneos to report as soon as possible, but no later than 7 calendar days after first knowledge by the Investigator, fatal or life-threatening suspected, unexpected, serious adverse drug reactions to the IEC responsible for the study.

It is the responsibility of Syneos to report to the IEC all other suspected, unexpected, serious adverse drug reactions that are neither fatal nor life-threatening, as soon as possible, but no later than 14 calendar days after first knowledge by the Investigator.

9.13 Pregnancy

In the event a dosed female subject becomes pregnant during or shortly after participation in the study, this pregnancy will be reported to the Sponsor within 24 hours of first knowledge of the event. Any subject who becomes pregnant during the study will be immediately withdrawn. Follow-up information regarding the course and outcome of the pregnancy will be documented as per the clinical site's SOP. If the outcome of the pregnancy meets the criteria for classification as an SAE, reporting of the event to the IEC and regulatory agency(ies) will be performed as per site's SOP.

9.14 Reportable Disease

In the case a subject has or manifested any clinical signs characteristic of a reportable disease or condition (e.g., HIV, tuberculosis, SARS, COVID-19), it is the responsibility of the Medical Director to notify the public health department of the State of Florida within 72 hours after becoming aware of the information.

10. Premature Termination of the Study

The study may be prematurely terminated by the Investigator following consultation with the Sponsor, by the Sponsor or by the regulatory authorities. Following a decision to discontinue the trial, the Investigator will promptly inform the active study subjects and the IEC responsible for this trial, stating the reasons for discontinuation of the study and, furthermore, advise them in writing of any potential risks to the health of study subjects or other persons. It is the Sponsor's responsibility to report the premature termination of the study to the regulatory authority(ies), when required by the applicable regulatory requirement(s).

11. Analytical Methodology

When applicable, samples will be transported to the bioanalytical facility in at least two separate shipments, with each set of aliquots in separate shipments. Once the bioanalytical laboratory confirms receipt of the first shipment, the second set of aliquots may be sent. The samples should be packed on sufficient dry ice to keep them frozen for at least 72 hours.

Syneos Health Clinical Bioanalytical Facility will analyze resveratrol, resveratrol-3-glucuronide, resveratrol-4'-glucuronide, and resveratrol-3-sulfate in plasma and urine samples using a validated method.

Samples for potential RNA-Seq analysis and for the other possible investigational tests (iduronidase activity measurement and mRNA frataxin) will be transported to the external laboratory, when identified. Details of this additional laboratory work will be available in a separate protocol.

Analyst and Watson LIMS (Laboratory Information Management System) will be used at different steps of the analysis.

The bioanalytical work in support of the study will be conducted in compliance with the GCP, using the SOPs in place by Syneos Health Clinical Bioanalytical Facility. These SOPs are in accordance with applicable regulations in the industry: Guidelines on Bioanalytical Method Validation, Good Laboratory Practice (GLP), and Guideline for GCP ICH E6 (R2).

Samples from subjects included in the pharmacokinetic population (see section 12.2.2) and from subjects who were withdrawn from the study due to AEs or vomiting episodes will be analyzed.

12. Pharmacokinetic and Statistical Analyses

PK analysis will be performed using Phoenix® WinNonlin®. Inferential statistical analyses will be performed using SAS® according to FDA guidelines.

12.1 Pharmacokinetics

12.1.1 PK Parameters Calculated With Plasma Concentrations

The following PK parameters will be calculated by standard non-compartmental methods for resveratrol, resveratrol-3-glucuronide, resveratrol-4'-glucuronide, and resveratrol-3-sulfate:

1) AUC_{0-t}: area under the concentration-time curve from time zero to the last non-zero concentration
2) AUC_{0-inf}: area under the concentration-time curve from time zero to infinity (extrapolated)
3) C_{max}: maximum observed concentration
4) Residual area: calculated as $100*(1- AUC_{0-t} / AUC_{0-inf})$
5) T_{max}: time of observed C_{max}
6) $T_{½\ el}$: elimination half-life
7) K_{el}: elimination rate constant

12.1.2 PK Parameters Calculated With Urine Concentrations

Urine samples will be used to calculate the following parameters for resveratrol, resveratrol-3-glucuronide, resveratrol-4'-glucuronide, and resveratrol-3-sulfate:

1) Ae_{0-t}: cumulative urinary excretion from time zero to time t, calculated as the sum of the amounts excreted over each collection interval. The amount excreted in urine for each time interval is calculated as the urine concentration multiplied by the urine volume.
2) R_{max}: maximum rate of urinary excretion, calculated by dividing the amount of drug excreted in each collection interval by the time over which it was collected.
3) T_{max}: time of R_{max}, calculated as the midpoint of the collection interval during which R_{max} occurred.

Additional PK analysis may be performed. Upon the Sponsor's request, PK repeats might be performed according to Syneos Health's SOP. If re-assays are requested for PK reasons, final results will include re-assay values, while results with original values will be presented in an appendix of the clinical study report as supportive data.

12.2 Analysis Populations

12.2.1 Safety Population

The safety population is defined as all subjects who received at least one dose of the study medication.

12.2.2 Pharmacokinetic Population

The PK population will include all subjects completing at least one period and for whom the PK profile can be adequately characterized.

Any subject with pre-dose concentrations will be excluded from the PK and statistical analysis for the respective analyte for the concerned period if the pre-dose concentration is greater than 5% of the C_{max} value of that period for that subject.

Data from subjects who experienced emesis during the sampling interval and who were not withdrawn as per criterion established under section 9.11 may be evaluated after completion of the PK analysis. Any subject who experienced emesis within 2 times median T_{max} of the current study (based on the reference product) will be excluded from the statistical analysis. Data (concentrations and PK parameters) from subjects excluded due to a pre-dose concentration greater than 5% of their C_{max} or from subjects withdrawn due to AEs or vomiting episodes will be presented but excluded from descriptive statistics for the concerned period.

12.3 Statistical Analyses

Demographic parameters will be summarized descriptively. TEAEs will be summarized descriptively by treatment for all subjects who were dosed (safety population). No inferential statistical analysis of safety data is planned.

Individual and mean plasma concentration versus time curves will be presented for both linear and semi-log scales. Descriptive statistics (arithmetic and geometric means, standard deviation [SD], coefficient of variation [CV%], minimum [Min], maximum [Max], and median) of the plasma concentrations versus time will be presented as well for the pharmacokinetic parameters.

A Statistical Analysis Plan (SAP) will be prepared after completion of the final protocol.

Safety and tolerability:

TEAEs will be tabulated by treatment. Changes from screening to study exit values in vital signs, ECG, and clinical laboratory parameters will be evaluated. Safety and tolerability data will be reported using descriptive statistics. Results of vital signs, ECG, and clinical laboratory parameters, urine drug screens, clinical laboratory tests, pregnancy tests, alcohol breath test, and urine cotinine test will be listed.

PK

- Plasma PK:

For all analytes, summary statistics will be used to describe the PK profile for each dose level under fasting conditions (Periods 1, 2, and 3) and under fed conditions (Period 4).

Dose proportionality analysis for AUC_{0-t}, AUC_{0-inf} and C_{max} will be performed (using the power model with mixed procedure from SAS®) considering data under fasting conditions (Periods 1, 2, and 3).

For evaluation of the food-effect, PK data (ln-transformed AUC_{0-t}, AUC_{0-inf}, C_{max} and untransformed T_{max}) reported under fed conditions (Period 4) and under fasting conditions (for the same dose level) will be compared using ANOVA from SAS®. The ratio (fed/fasting) and 90% geometric confidence interval will also be calculated for AUC_{0-t}, AUC_{0-inf} and C_{max}.

All inferential statistical analyses will be interpreted in an exploratory sense only at an alpha level of 5% for statistical significance.

Additional statistical analysis may be performed.

- Urine PK:

Summary statistics will be used to describe urinary excretion of resveratrol, resveratrol-3-glucuronide, resveratrol-4'-glucuronide, and resveratrol-3-sulfate.

Additional statistical analysis may be performed.

13. Regulatory Considerations and Quality Assurance

13.1 Independent Ethics Committee Approval of Protocol and Other Study Documents

The Investigator(s) agree to provide the IEC with all appropriate documents, including a copy of the protocol/amendments, ICFs, advertising text (if any), Investigator's Brochure (if any) and any other written information provided to study subjects. The trial will not begin until the Investigators have obtained the IEC favourable written approvals for the above-mentioned study documents. A properly executed written ICF shall be read, signed, and dated by each subject prior to entering the trial or prior to performing any study procedure. The original signed and dated ICF will be kept at Syneos Health and a copy will be given to the subject.

In the event that the protocol is amended, the revised protocol must be approved by the IEC prior to its implementation, unless the changes involve only logistical or administrative aspects of the trial. If a revised ICF is introduced during the study, each subject's further consent must be obtained. The new version of the ICF must be approved by the IEC, prior to subsequently obtaining each subject's consent.

The Investigator and the Sponsor's representative must sign the protocol and its amendments (if any) before initiating the study.

It is the Sponsor's responsibility to submit the protocol and its amendments (if any), and the ICFs to regulatory authorities when necessary.

13.2 Compliance

This study will be conducted in compliance with the protocol, GCP, and all applicable regulations, including the Federal Food, Drug and Cosmetic Act, U.S. applicable Code of Federal Regulations (title 21), and any IEC requirements relative to clinical studies. The study will also be conducted in compliance with the recommendations laid down in the most recent version of the Declaration of Helsinki, with the exception that registration of such Phase 1 trials in a publicly accessible database is not mandatory.

13.3 Quality Assurance Program

Syneos Health has established Quality Control (QC) and Quality Assurance (QA) systems with written SOPs to ensure that the study will be conducted and data will be generated, recorded, and reported in compliance with the protocol, GCP, and applicable regulatory requirements. A rigorous QC program is applied to ensure accuracy of all data and reports. QA oversees a complementary risk-based program of audits to assure compliance with applicable regulations and Syneos Health's prescriptive documentation.

13.4 Audits, Inspections and Monitoring

In accordance with the principles of GCP and GLP, the study may be inspected by regulatory authorities, the Sponsor and Syneos Health. The Sponsor is entitled to access information about the status of the study and to review the original documents of the study.

14. Confidentiality

This document contains trade secrets and commercial information that is confidential and may not be disclosed to third parties. Persons to whom this study protocol is disclosed must be informed that all the information herein is confidential and may not be further divulged. These restrictions will apply as well to all future communications if deemed privileged or confidential. Publication of the study results may only be allowed with written permission from the Sponsor.

All information on a subject obtained during the conduct of the study will be kept confidential. Subjects will be identified by an anonymized identifier on all samples and study records provided to the Sponsor or designee. In compliance with ICH GCP, the Sponsor's authorized representatives, monitor(s), auditor(s), IEC, and regulatory authority(ies) will be granted direct access to the subject's original trial-related records for verification of clinical trial procedures and/or data, without violating the confidentiality of the subject, to the extent permitted by the applicable laws and regulations. Consent from the subject for disclosure of such information will be obtained in writing in the ICF. In addition, should a subject require medical care or hospitalization during the course of the study, the clinical site may contact the treating physician with the subject's consent, except that consent may not be requested if there is an emergency situation. If the results of the study are published, the subject's identity will remain confidential.

The clinical site will maintain adequate study records for 2 years after the marketing application is approved for the drug for the indication for which it is being investigated; or, if no application

is to be filed or if the application is not approved for the drug, until 2 years after the study is discontinued. The Sponsor will be notified prior to the destruction of study records.

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