AstraZeneca

Amended Clinical Pharmacology Study Protocol	
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A Phase I, Open-Label, Pharmacokinetic Study Measuring Plasma Exposure of a 20 mg Single Dose Administration of Rosuvastatin Calcium to Healthy Asian Subjects Living in the United States Related to Pharmacokinetic Data Obtained from a Caucasian Control Group

AstraZeneca Clinical Development Team representative

AstraZeneca Research and Development site representative

AstraZeneca Research and Development site representative

The following Amendment(s) and Administrative Changes have been made to this protocol since the date of preparation:

Amendment No. 1	Date of Amendment	Local Amendment No.	Date of local Amendment
Administrative change No.	**Date of Administrative Change**	**Local Administrative change No.**	**Date of local Administrative Change**

ASTRAZENECA PROCEDURES IN CASE OF EMERGENCY, OVERDOSE, OR PREGNANCY

In the case of a medical emergency you may contact the Clinical Study Team Leader. If the Clinical Study Team Leader is not available, contact the Clinical Study Team Physician or the Clinical Study Team Drug Safety Physician at the AstraZeneca Research and Development site shown below.

Role in the study	Name	Address and Telephone number
Clinical Study Team Leader		
Clinical Study Team Physician		
Clinical Study Team Safety Physician		

For further clarifications regarding:

- Procedures in case of medical emergency see Section 8.2
- Procedures in case of overdose see Section 8.3
- Procedures in case of pregnancy see Section 8.4

PROTOCOL SYNOPSIS

A Phase I, Open-Label, Pharmacokinetic Study Measuring Plasma Exposure of a 20 mg Single Dose Administration of Rosuvastatin Calcium to Healthy Asian Subjects Living in the United States Related to Pharmacokinetic Data Obtained from a Caucasian Control Group

Investigator

Study center(s), type and number of subjects planned

This will be a single-center study conducted in the United States. Additional sites may be added if required. Approximately 156 healthy Asian subjects of Chinese, Filipino, Asian-Indian, Korean, Vietnamese, and Japanese heritage will be recruited with a minimum of 22 subjects and a maximum of 26 subjects from each group to obtain at least 132 evaluable subjects. Approximately 26 healthy Caucasian subjects will be recruited to obtain at least 22 evaluable subjects. These subjects will serve as a control group for the study. An evaluable subject is defined as a subject completing all study procedures from the screening period to the final blood sampling for plasma levels of rosuvastatin.

Study period	**Phase of development**
Estimated date of first subject enrolled	I
Estimated date of last subject completed	

Objectives

The primary objective of this study is to summarize rosuvastatin plasma exposure in healthy subjects of Asian heritage who are living in the United States following a single 20 milligram (mg) dose by measuring maximum concentration (C_{max}) of rosuvastatin and area under the concentration curve from zero to infinity (AUC) and to relate those results to rosuvastatin pharmacokinetic data obtained from a Caucasian control group.

The secondary objective of this study is to summarize the pharmacokinetics of 20 mg rosuvastatin by measuring area under the curve of plasma concentration against time from

zero to time of last quantifiable concentration ($AUC_{(0-t)}$), terminal elimination half-life ($t_{½\lambda z}$), time of maximum concentration (t_{max}), and apparent oral clearance (CL/F).

The pharmacokinetics, (C_{max}, AUC, $AUC_{(0-t)}$, $t_{½\lambda z}$, and t_{max},) of rosuvastatin metabolites (N-desmethyl rosuvastatin, and rosuvastatin lactone) will also be assessed and summarized.

Safety and tolerability will be assessed by physical examination, clinical laboratory tests, vital signs, and collection of adverse events.

An additional objective of the present study is to obtain DNA samples from subjects who consent separately to pharmacogenetic research. It is emphasized that subjects who decline to provide such consent may still participate in the main pharmacokinetic study.

Study design

This is a single-center, open label, single dose, pharmacokinetic study in healthy Asian and Caucasian subjects residing in the United States.

Investigational product, dosage and mode of administration

A 20 mg tablet of rosuvastatin calcium will be given as a single oral dose.

Duration of treatment

Subjects meeting inclusion/exclusion criteria and satisfactorily completing screening evaluations during the screening period will participate in a 4 day in-patient period during which they will receive a single dose of 20 mg rosuvastatin calcium with subsequent blood sampling to determine defined pharmacokinetic parameters.

Outcome variables

C_{max} and AUC of rosuvastatin will be the primary endpoints for this study to summarize exposure in the selected population. If AUC data cannot be determined in all subjects completing the study, $AUC_{(0-t)}$ will replace AUC as a primary endpoint. A detailed description of the summaries will be described in the Statistical Analyses Plan (SAP).

- **Pharmacokinetic**

Along with C_{max} and AUC, $AUC_{(0-t)}$, $t_{½\lambda z}$, t_{max}, and CL/F of rosuvastatin will be determined to summarize the pharmacokinetics of rosuvastatin in the selected population**.**

C_{max}, AUC, $AUC_{(0-t)}$, $t_{½\lambda z}$, and t_{max}, of rosuvastatin metabolites, N-desmethyl rosuvastatin and rosuvastatin lactone, will also be determined and summarized in the selected population.

- **Safety**

Safety and tolerability will be assessed by physical examination, clinical laboratory tests, vital signs, and collection of adverse events

- **Pharmacogenetics**

Blood samples will be obtained from consenting subjects and may be used to study genetic polymorphisms that could affect the disposition of rosuvastatin.

Statistical Methods

C_{max} and t_{max} for each subject will be determined by inspection of the rosuvastatin plasma concentration-time profile. The terminal elimination rate constant (λ_z) will be calculated by log-linear regression of the terminal portion of the concentration-time profiles where there are sufficient data (a minimum of 3 plasma concentration values in the terminal log-linear phase, spanning an interval of at least 2 half-lives.) Terminal elimination half-life ($t_{½\lambda z}$) will be calculated as $0.693/\lambda_z$. The $AUC_{(0\text{-}t)}$ will be calculated by the linear trapezoidal rule ($AUC = AUC_{(0\text{-}t)} + C_{last}/\lambda_z$). $AUC_{(0\text{-}t)}$ will be extrapolated to infinity using λ_z to obtain AUC where there are sufficient data. Following the single dose administration of rosuvastatin, the CL/F will be calculated as Dose/AUC.

Pharmacokinetic data, C_{max}, $AUC_{(0\text{-}t)}$, and AUC, will be summarized for each ethnic group using descriptive statistics, including geometric means and coefficient of variation, and listed. In addition, 95% confidence intervals (CI) for the mean pharmacokinetic parameters will also be presented.

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LIST OF ABBREVIATIONS AND DEFINITION OF TERMS

The following abbreviations and special terms are used in this study protocol.

Abbreviation or special term	Explanation
λ_z	Terminal elimination rate constant
ad lib	As desired
ADME	Absorption, distribution, metabolism, excretion
AE	Adverse event
Assessment	An observation made on a variable involving a subjective judgment
ALT	Alanine aminotransferase
AST	Aspartate aminotransferase
AUC	Area under the concentration curve from zero to infinity
$AUC_{(0-t)}$	Area under the curve of plasma concentration against time from zero to time of last quantifiable concentration
BMI	Body mass index
°C	Degrees Celsius
CI	Confidence interval
CK	Creatine kinase
CL/F	Apparent oral clearance
C_{last}	Last quantifiable plasma concentration after single dose or last administration
C_{max}	Maximum concentration
CRC	Clinical research center
CRF	Case report form
DCF	Data clarification form
DMPK	Drug metabolism and pharmacokinetics
DNA	Deoxyribonucleic acid
ECG	Electrocardiogram
EDTA	Ethylenediaminetetraacetic acid
G	Relative centrifugal force
GCP	Good Clinical Practice
HBsAG	Hepatitis B surface antigen
HCG	Human chorionic gonadotrophin

Abbreviation or special term	Explanation
HDL-C	High density lipoprotein cholesterol
HIV	Human immunodeficiency virus
HMG-CoA	3-hydroxy-3methlglutaryl coenzyme A
ICH	International Conference on Harmonization
IEC	Independent Ethics Committee
IRB	Institutional Review Board
LDH	Lactic dehydrogenase
LDL-C	Low density lipoprotein cholesterol
LC/MS/MS	Liquid chromatography/mass spectrometry/mass spectrometry
m	Meter
MeDRA	Medical Dictionary for Drug Regulatory Affairs
mg	Milligram
mL	Milliliter
OAE	Other Significant Adverse Event (ie, adverse events of particular clinical importance, other than SAEs and those AEs leading to discontinuation of the subject from study treatment; see definition in section 4.7)
OTC	Over-the-counter
Outcome variable	A, usually derived, variable specifically defined to be used in the analysis of a study objective
PDR	Physicians' Desk Reference
PP	Per protocol
Parameter	A quantity (usually unknown) that characterizes the distribution of a variable in a population of subjects
Principal investigator	A person responsible for the conduct of a clinical study at a study site. Every study center has a principal investigator.
SAE	Serious adverse event
SAP	Statistical analysis plan
TC	Total cholesterol
TG	Triglycerides
$t_{½\hat{\lambda}z}$	Terminal elimination half life
t_{max}	Time of maximum concentration
ULN	Upper limit of normal
Variable	A characteristic or a property of a subject that may vary eg, from time to time or between subject

1. INTRODUCTION

1.1 Background

The clinical development program for rosuvastatin was comprised of 33 Phase I trials and 27 Phase II/III trials conducted worldwide. The Phase III clinical development program now includes efficacy and safety data from more that 12,500 subjects, with 14,231 subject-years of exposure in the 5 to 80 mg dose range tested. This includes 7,819 subjects at rosuvastatin 10 mg and 4,007 at 40 mg. There are no subjects currently taking rosuvastatin 80 mg in clinical trials.

1.2 Clinical pharmacology

In clinical pharmacology studies in man, peak plasma concentrations of rosuvastatin are reached 3 to 5 hours following oral dosing. Both C_{max} and AUC increase in approximate proportion to rosuvastatin dose. The absolute bioavailability of rosuvastatin in Caucasian subjects is approximately 20%.

Administration of rosuvastatin with food decreased the rate of drug absorption by 20% as assessed by C_{max} but there was no effect on the extent of absorption as assessed by AUC. Plasma concentrations of rosuvastatin do not differ following evening or morning drug administration. Significant LDL-C reductions are seen when rosuvastatin is given with or without food, and regardless of the time of day of drug administration.

Mean volume of distribution at steady state of rosuvastatin is approximately 134 liters. Rosuvastatin is 88% bound to plasma proteins, mostly albumin. This binding is reversible and independent of plasma concentrations.

Rosuvastatin is not extensively metabolized; approximately 10% of a radiolabeled dose is recovered as metabolite. The major metabolite is N-desmethyl rosuvastatin, which is formed principally by cytochrome P450 2C9, and *in vitro* studies have demonstrated that N-desmethyl rosuvastatin has approximately one-sixth to one-half the 3-hydroxy-3-methlglutaryl coenzyme A (HMG-CoA) reductase inhibitory activity of rosuvastatin. Overall, greater than 90% of plasma HMG-CoA reductase inhibitory activity is accounted for by rosuvastatin.

Following oral administration, rosuvastatin and its metabolites are primarily excreted in the feces (90%). The $t_{½\lambda z}$ of rosuvastatin is approximately 19 hours.

After an intravenous dose, approximately 28% of total body clearance was via the renal route, and 72% by the hepatic route.

1.3 Rationale

Phase I studies conducted in healthy Japanese subjects living in Japan indicated an approximate 2-fold increase in rosuvastatin C_{max} and $AUC_{(0-24)}$ compared to similar studies conducted in western Caucasian subjects and that the absolute bioavailability in Japanese subjects in Japan was 29% compared to 20% in Caucasians. The population pharmacokinetic

analysis that incorporated race as a categorical variable also demonstrated an approximate 2-fold increase in rosuvastatin plasma concentrations in Asian subjects, most of whom were Japanese subjects residing in Japan.

Preliminary results of an ongoing trial in Singapore suggest that systemic exposure in Chinese, Malay, and Indian subjects is higher than in a group of Caucasian subjects. The magnitude of difference between Chinese and Caucasian subjects is similar to that observed between Japanese and Caucasian subjects.

The mechanism for the apparent difference in the pharmacokinetic profile of rosuvastatin between these ethnic groups is unknown. Extrinsic (diet, environment, etc.) and/or intrinsic (genetic) factors could be responsible for these differences.

Plasma exposure of rosuvastatin has not been determined in Asian populations residing in the United States. This study will assess and summarize the pharmacokinetics of a single dose of 20 mg rosuvastatin in Chinese, Filipino, Asian-Indian, Korean, Vietnamese, and Japanese subjects residing in the United States. The study population represents the most populous Asian groups residing in the United States as described in the 2000 United States census report (Barnes, Jessica; Bennett, Claudette; U.S. Census Bureau; Census 2000 Brief; "The Asian Population;" issued February 2002, <http://www.census.gov/prod/2002pubs/c2kbr01-16.pdf>). Results will be related to pharmacokinetic data obtained from a Caucasian control group to determine if similar differences in exposure occur between these groups.

In relation to the pharmacogenetic objective of this study, extensive evidence indicates that, for many drugs, variation within genes controlling absorption, distribution, metabolism, and excretion (ADME) can strongly influence pharmacokinetics. Emerging evidence suggests that such genetic variations may indeed be important for the disposition of statins. In a recent study, a functional polymorphism within OATP-C, a hepatic uptake transport protein, was reported to affect the clearance of pravastatin, (Nishizato Y, Jeiri I, Suski H et al., 2003). It is plausible that variations in OATP-C or in other genes relevant to rosuvastatin disposition could account for intra-or inter-racial variation in its pharmacokinetics

1.4 Efficacy

CRESTOR® (rosuvastatin calcium, ZD4522, hereafter referred to as rosuvastatin) is a novel member of the statin class of lipid-lowering agents, and is a potent and competitive synthetic HMG-CoA reductase inhibitor, both in vitro and in vivo.

In the rosuvastatin clinical trial program, the efficacy and safety of rosuvastatin up to 80 mg were evaluated in a broad range of subjects with dyslipidemia representing those requiring lipid-lowering therapy in the general population. The clinical trial program included trials in subjects with Fredrickson Type IIa, IIb, and IV dyslipidemia, as well as specific trials in subjects with heterozygous or homozygous familial hypercholesterolemia (FH).

Rosuvastatin, at doses up to 40 mg, produces substantial improvements in the lipid profile, lowering low density lipoprotein cholesterol (LDL-C) and triglycerides (TG), and raising high

density lipoprotein cholesterol (HDL-C), in patients with dyslipidemia while exhibiting a favorable safety profile.

1.5 Safety

Review of adverse event data show that rosuvastatin is well tolerated. The overall frequency of alanine aminotransferase (ALT) elevations is low. Clinically significant ALT elevations (>3 x the upper limit of normal (ULN) on 2 or more occasions) are uncommon and for the most part resolve with either continued therapy with or without dose reduction or after an interruption of treatment. No cases of irreversible liver injury secondary to rosuvastatin therapy have been identified.

Muscle-related findings (ie, creatine kinase (CK) increases, myalgia, and myopathy) are a well-recognized complication of statin therapy. The frequency of both myalgia and CK elevations is low and similar in the 10 mg to 40 mg dose range. Only 1 case, (at 20 mg dose), of myopathy – defined as CK elevations >10 x ULN associated with muscle symptoms – was considered possibly related to rosuvastatin therapy in the 10 mg to 40 mg dose range. The frequency of these complications in subjects given rosuvastatin is consistent with the data reported for other marketed statins.

The analysis of renal data indicates that proteinuria, predominantly tubular in nature, occurs at a low frequency among subjects receiving rosuvastatin in doses up to and including 40 mg. A low frequency of proteinuria was also observed in subjects treated with other statins in the rosuvastatin clinical program. The proteinuria seen with rosuvastatin is not associated with clinically significant increases in serum creatinine or worsening renal function. The 40 mg rosuvastatin dose is well tolerated in terms of renal effect with both shorter-term and longer-term treatment.

Rosuvastatin is well tolerated in a broad spectrum of subjects independent of gender, age, race, menopausal status, or the presence of comorbid conditions such as hypertension, diabetes, coronary heart disease, or altered hepatic or renal function.

Rosuvastatin can be administered with a range of medications used to treat either comorbid medical conditions or underlying lipid profile abnormalities.

2. STUDY OBJECTIVES

2.1 Primary objective

The primary objective of this study is to summarize rosuvastatin plasma exposure in healthy subjects of Asian heritage who are living in the United States following a single 20 mg dose by measuring C_{max} and AUC of rosuvastatin and to relate those results to pharmacokinetic data obtained from a Caucasian control group.

2.2 Secondary objective(s)

The secondary objectives of the study are:

1. To summarize the pharmacokinetics of rosuvastatin after a 20 mg dose by measuring $AUC_{(0\text{-}t)}$, $t_{½\lambda z}$, t_{max}, and CL/F

2. To summarize the pharmacokinetics of rosuvastatin metabolites by measuring C_{max}, AUC, $AUC_{(0\text{-}t)}$, $t_{½\lambda z}$, and t_{max} of N-desmethyl rosuvastatin and rosuvastatin lactone

3. To assess safety and tolerability by physical examination, clinical laboratory tests, vital signs, and collection of adverse events

2.3 Pharmacogenetic objectives

An additional objective of the present study is to obtain DNA samples from subjects who consent separately to pharmacogenetic research. It is emphasized that subjects who decline to provide such consent may still participate in the main pharmacokinetic study.

3. STUDY PLAN AND PROCEDURES

3.1 Overall study design

This is a single-center, open label, single dose, pharmacokinetic study of subjects having Chinese, Filipino, Asian-Indian, Korean, Vietnamese, and Japanese heritage and are residing in the United States. Approximately156 subjects will be recruited with a minimum of 22 subjects and a maximum of 26 subjects from each ethnic group to obtain at least 132 evaluable subjects. Approximately 26 healthy Caucasian subjects will be recruited to obtain at least 22 evaluable subjects. These subjects will serve as a control group for the study.

Rosuvastatin exposure following a single oral dose of 20 mg will be determined by measuring C_{max} and AUC. In addition, $AUC_{(0\text{-}t)}$, $t_{½\lambda z}$, t_{max}, and CL/F of rousuvastatin and C_{max}, AUC, $AUC_{(0\text{-}t)}$, $t_{½\lambda z}$, and t_{max}, of N-desmethyl rosuvastatin and rosuvastatin lactone will be measured.

Subjects satisfying the inclusion and exclusion criteria and screening exams will enter the clinical research center (CRC) on Day –1. On Day 1 subjects will receive a single dose of rosuvastatin 20 mg under fasting conditions. Blood samples will be taken pre-dose (within 30 minutes) and 0.5, 1, 2, 3, 4, 5, 6, 8, 10, 12, 18, 24, 30, 36, 48, 54, 60, and 72 hours post-dose. After completion of the final blood sample and all safety assessments on Day 4, the subjects will be discharged.

3.1.1 Stopping criteria for dose escalation

Not applicable

- **Figure 1 Study flow chart**

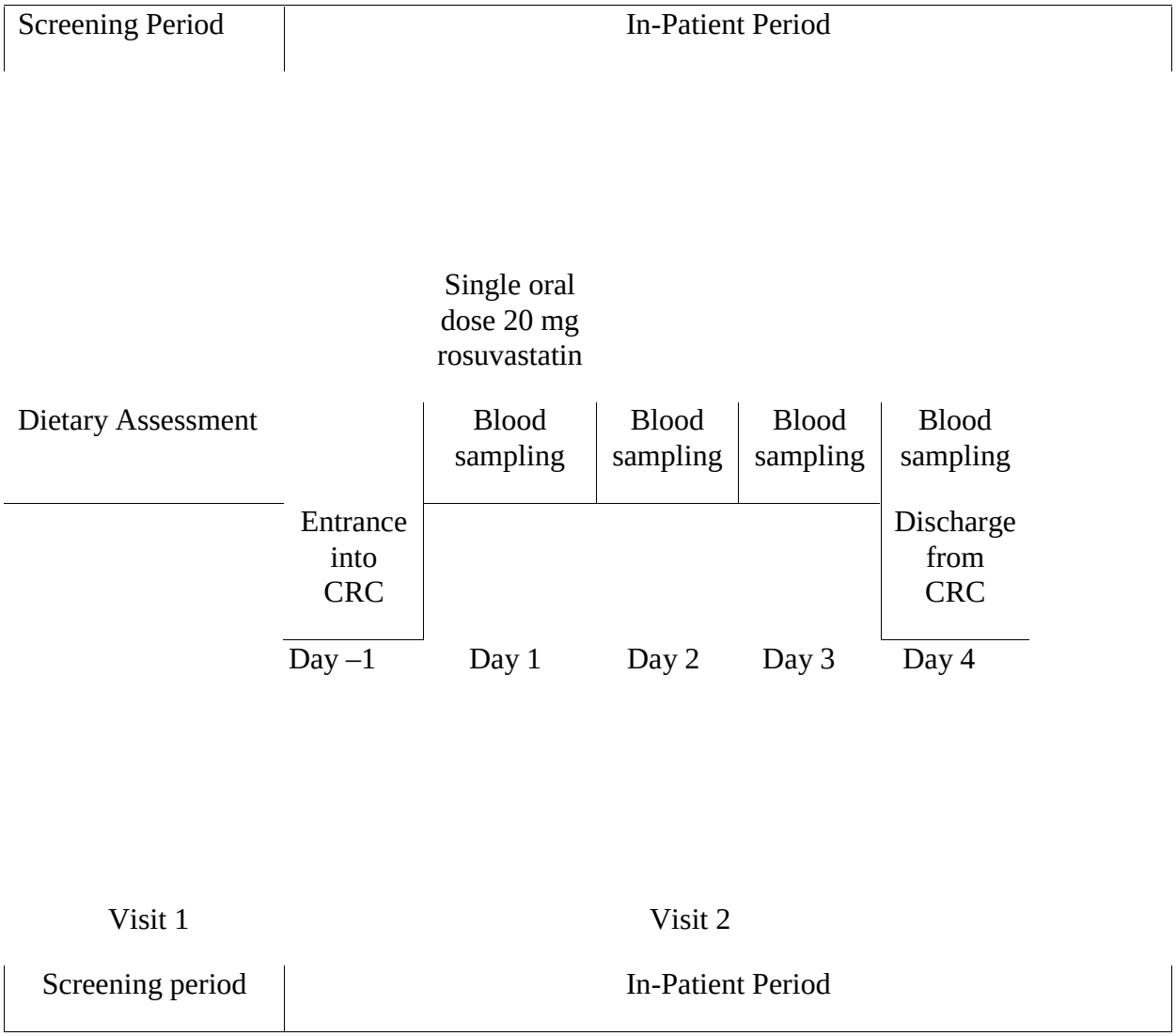

Table 1 Study plan

	Screening Period	In-Patient Period				
	Within 35 Days of Day -1	Day -1	Day 1	Day 2	Day 3	Day 4
Informed Consent	√					
Inclusion/Exclusion Criteria	√	√				
Demographics	√					
Medical History	√					
Drug History	√					
Urine Drug Screen	√	√				
Complete Physical Exam	√					√
Brief Physical Exam		√				
Dietary Assessment[a]	√					
ECG	√					
Clinical Chemistry, Hematology	√	√				√
Fasting Lipid Profile[b]			√			
HIV antibody, HBsAG, Hepatitis C Antibody	√					
Urinalysis	√	√				√
Serum Pregnancy Test	√	√				√
Vital Signs[c]	√	√	√	√	√	√
Sample for Genetic Testing		√				
Administration of 20 mg rosuvastatin			√			
Blood Sampling for rosuvastatin[d,e]			√	√	√	√
AEs/Concomitant Medications		√	√	√	√	√

a. A 3-day diet diary will be completed and returned to the CRC for analysis during the screening period
b. Fasting lipid profile to measure TC, TG, HDL-C, and LDL-C
c. Vital signs will be taken at screening, Day –1, before dosing on Day 1 and in the morning on Days 2 through 4
d. Subjects will remain at the CRC through final blood sampling and exit assessments on the morning of Day 4
e. Serial blood sampling for rosuvastatin will be done at the following time points: Day 1 pre-dose (within 30 minutes), and 0.5, 1, 2, 3, 4, 5, 6, 8, 10, 12, 18, 24, 30, 36, 48, 54, 60, and 72 hours post-dose

3.2 Rationale for study design, doses and control groups

The design of this study is standard for determining the pharmacokinetics of a single dose of 20 mg rosuvastatin.

A 20 mg dose is sufficient to allow complete characterization of the rosuvastatin plasma concentration-time profile.

A Caucasian control group has been included in order to collect pharmacokinetic data that can be related to the pharmacokinetic data collected from the selected Asian population.

3.3 Selection of study population

3.3.1 Study selection record

The study population represents the most populous Asian groups residing in the United States as described in the 2000 United States census report.

3.3.2 Inclusion criteria

For inclusion in the study, subjects must fulfill all of the following criteria:

1. Provision of signed written informed consent, ability to communicate with the investigator, and to understand and comply with the requirements of the study

2. Males and females aged 18-65, inclusive

3. Body Mass Index (BMI) between 18-29, inclusive, BMI will be calculated as weight in kilogram (kg)/height in meters^2 [m^2])

4. Women who are surgically sterilized, post-menopausal for at least one year, or not pregnant and/or lactating. Women of childbearing potential must be willing to abstain from sexual activity or use an effective double barrier method of contraception during the study period (eg, condom and diaphragm, condom and foam, condom and sponge, etc.), or intrauterine devices

5. With the exception of those subjects entered into the Caucasian control group, subjects must identify themselves as a member of one of the following groups: Chinese, Filipino, Asian-Indian, Korean, Vietnamese, or Japanese. Subjects' parents must also be of the same reported race. (See Section 4.1.1)

6. Residence in the United States for a least 12 continuous months

3.3.3 Exclusion criteria

Any of the following is regarded as a criterion for exclusion from the study:

1. Use of prescription medication for a chronic medical condition

2. Acute illness or use of prescription medication for an acute medical condition within two weeks of Day –1

3. History within the last 3 months of extreme or therapeutic diet programs including but not limited to: weight reduction, weight augmentation, high protein, high carbohydrate, low carbohydrate, or low fat

4. Any contraindication determined by review of a detailed medical and drug history, complete physical examination, vital signs, blood chemistry, hematology, and electrocardiogram (ECG).

5. Medical history or psychological conditions which, in the opinion of the investigator, would compromise the subject's safety or successful participation in the study

6. History of adverse drug reaction or hypersensitivity to statins or drugs with a similar chemical structure to rosuvastatin.

7. History or presence of gastrointestinal, hepatic, or renal disease or other conditions known to interfere with ADME of drugs

8. History of alcohol or substance abuse within the past year

9. Positive test results for human immunodeficiency virus (HIV) antibody, hepatitis B surface antigen (HBsAG), or hepatitis C antibody

10. Positive urine drug screen

11. Participation in another study within 30 days of Day –1, apart from non-invasive methodology studies in which no drugs were given.

3.3.4 Restrictions

Subjects will be required to:

1. Refrain from alcohol, grapefruit containing products, and apple juice from CRC admission through the final study evaluation

2. Refrain from using over-the-counter (OTC) medications, and herbal supplements 2 weeks prior to CRC admission through the final study assessments

3. Refrain from eating or drinking starting 8 hours before study drug administration and continuing through 4 hours post study drug administration (with the exception of water which will be allowed ad lib until 2 hours prior to study drug administration and 2 hours post study drug administration). Lunch will be served approximately 4 hours after the dose of rosuvastatin. At all other times during the in-patient period, subjects will be served standard meals on a regular basis.

3.3.5 Discontinuation of subjects from treatment or assessment

Subjects may be discontinued from study treatment and assessments at any time, at the discretion of the investigator(s).

3.3.5.1 Criteria for discontinuation

Specific reasons for discontinuing a subject from this study are:

1. Voluntary discontinuation by the subject, who is at any time free to discontinue their participation in the study without prejudice to further treatment
2. Safety reasons as judged by the investigator or AstraZeneca
3. Eligibility criteria not fulfilled
4. At investigator's discretion

3.3.5.2 Procedures for discontinuation

Subjects who discontinue from the study should always be asked about the reason(s) for their discontinuation and about the presence of any adverse events. If possible, they should be seen and assessed by an investigator(s). Adverse events should be followed up and the subject should return any diary cards, questionnaires and investigational products.

Discontinuation of any subject should be communicated to AstraZeneca. All withdrawals due to a serious adverse event (SAE) must be reported to AstraZeneca within 1 day. Withdrawals due to the occurrence of a non-serious AE must be reported to AstraZeneca within 15 days.

Where possible, subjects must be followed up for 30 days after completion of the study.

There must be at least 22 evaluable subjects per ethnic group; therefore, if a subject is discontinued before completing the study he/she may be replaced as needed at the discretion of the sponsor.

3.4 Treatment(s)

3.4.1 Investigational product(s)

Rosuvastatin calcium 20 mg will be supplied as tablets for oral use.

3.4.1.1 Identity of investigational product

Table 2 Identity of investigational product

Investigational product	Dosage form and strength	Manufacturer	Formulation number
Rosuvastatin calcium	20 mg	AstraZeneca Pharmaceuticals, LP	F12673

3.4.1.2 Labeling

Rosuvastatin calcium 20 mg will be provided in bottles containing 100 tablets per bottle. Each subject will be assigned a bottle of rosuvastatin. Each bottle will be affixed with a two-panel label with tear-off. At least the study number, storage conditions, dosing instructions, and bottle contents will appear on both portions of the label.

Before dispensing the first dose, the date dispensed and the subject's initials and subject number must be written on both the permanent and tear off labels. The tear off portion will be removed from the bottle assigned to the subject and affixed to the appropriate CRF.

3.4.1.3 Storage

Study medication supplies must be stored in a secure place (eg locked cabinet) and will be accessible only to the persons authorized to dispense them to the subjects. Rosuvastatin tablets will be stored at 20-25°C (68-77 °F) and protected from light and moisture.

3.4.1.4 Accountability

The investigational product provided for this study is for use only as directed in the protocol. The AstraZeneca site monitor will return all unused investigational product to . The investigational site personnel will account for all drugs dispensed and returned. Certificates of delivery and return must be signed.

3.4.2 Doses and treatment regimens

On the morning of Day 1, all subjects will receive a single 20 mg dose of rosuvastatin calcium under fasting conditions (See restrictions Section 3.3.4). Tablets will be taken orally with 240 milliliters (mL) of distilled, room temperature water. Tablets are not to be crushed or chewed. Subjects must remain in an upright position (sitting or standing) for 4 hours after dosing.

3.4.3 Method of assigning subjects to treatment groups

Written informed consent will be obtained before enrollment. Each subject will be assigned a unique enrollment number that will identify the site and the ethnic group of the subject enrolled. (See Table 3) This table is representative and the number of sites participating may be more or less than those listed.

Subjects fulfilling the eligibility criteria and continuing in the study will be assigned unique subject numbers that will identify the site and ethnic group of the subject. (See Table 4)

Subjects will be assigned enrollment/subject numbers consecutively as they enroll/enter into the study. If a subject discontinues from the study, the subject number will not be re-used and the subject will not be allowed to re-enter the study.

Table 3 Assignment of enrollment numbers

Site #	Chinese	Filipino	Asian-Indian	Korean	Vietnamese	Japanese	Caucasian
1	E0001101*	E0001201	E0001301	E0001401	E0001501	E0001601	E0001701
2	E0002101	E0002201	E0002301	E0002401	E0002501	E0002601	E0002701

*This table represents the first enrollment number for each site/ethnic group. Numbers will be assigned consecutively as prospective subjects are enrolled.

Table 4 Assignment of subject numbers

Site #	Chinese	Filipino	Asian-Indian	Korean	Vietnamese	Japanese	Caucasian
1	1101	1201	1301	1401	1501	1601	1701
2	2101	2201	2301	2401	2501	2601	2701

*This table represents the first subject number for each site/ethnic group. Subject numbers will be assigned consecutively to subjects as they are entered into the study. NB: assigned enrollment numbers will not necessarily match assigned subject numbers.

3.4.4 Blinding and procedures for unblinding the study

Not applicable

3.4.5 Concomitant medication

During the screening period, subjects will be instructed to consult with the investigative staff prior to taking OTC and prescription medications, dietary supplements, and herbal remedies. All medications, (OTC products, dietary supplements, and herbal remedies) will be discontinued two weeks prior to entry into the CRC. Women with a history of previous oral contraceptive use must have discontinued the regimen at least 3 months prior to Day –1.

Subjects will not be permitted to take any medication, dietary supplements, or herbal remedies during the in-patient period. Acetaminophen preparations will be allowed for analgesia, if absolutely necessary, for the well being of the subject. If acetaminophen is necessary, administration is not to exceed 2 grams in any 12-hour period.

Any medication, which is considered necessary for the subject's safety and well being, may be given at the discretion of the investigator(s). The administration of all medication (including investigational products) must be recorded in the appropriate sections of the CRF.

3.4.6 Treatment compliance

Compliance will be assured by the supervised administration of the study drug by site personnel.

4. MEASUREMENT OF STUDY VARIABLES

4.1 Screening and demographic measurements

Screening of subjects will be done within 35 days of Day –1.

4.1.1 Demographics

At the screening visit, demographic data will be collected including the following: date of birth, sex, and race.

Subjects being entered into the study as part of the Asian study group will self-report race by checking a tick box offered during the screening process. The following tick boxes will be offered: Caucasian, Black, Oriental, American Indian, and Other. Additional tick boxes will be provided to indicate which Asian sub-group the subject belongs. These tick boxes will include Chinese, Filipino, Asian-Indian, Korean, Vietnamese, Japanese, and Other. Subjects reporting mixed ethnicity by selecting more than one Asian sub-group or boxes indicating "Caucasian", "Black", "American Indian" or "Other" will not be included in the study.

In addition, subjects being entered into the study as part of the Asian study group will be asked to identify the race of each parent. Only those subjects with both parents matching the self-reported race (of the subject) will be eligible for the study.

Subjects being entered into the study as part of the Caucasian control group will also self-report race by checking a tick box offered during the screening process. The same tick boxes will be offered to the control group as offered to the Asian groups. Only those choosing "Caucasian" will be considered eligible for the study. Please refer to the "Instructions for the Investigator" located in the CRF for additional direction.

4.1.2 Medical history/Drug history

A complete medical and drug history will be recorded for each subject at the initial screening visit and reviewed for any additions or omissions on admission to the CRC (Day –1). Significant medical conditions that have occurred within the past 2 years, or conditions that are ongoing (ie, headache, backache, indigestion) are to be recorded in the CRF. The drug history must identify any known drug allergies, presence or history of drug abuse, and use of chronic medications.

4.1.3 Urine Drug Screen

A urine screen for drugs of abuse will be conducted at the times specified in Table 1.

If a test result is positive for drugs of abuse, the subject will not participate in the study. The following drugs of abuse will be screened: cannabinoids, cocaine, opiates, amphetamines, benzodiazepines, barbiturates, methaqualone, propoxyphene, and methadone.

4.1.4 Complete physical examination

Each subject will undergo a complete physical examination, which will be conducted at the times specified in Table 1.

This complete physical examination will include an assessment of the following:

- General appearance,
- Skin, head, neck, and lymph nodes
- Musculoskeletal/extremities (including spine)
- Cardiovascular
- Lungs
- Abdomen
- Neurological (reflexes)

Physical examination data to be recorded on the CRF will include: 1) normal/abnormal and 2) a description of any abnormalities.

Height in centimeters (cm) and weight in kg will be measured at screening only and will be recorded on the CRF.

4.1.5 Brief physical examination

The brief physical examination will be conducted at the times specified in Table 1 and will include an assessment of the following:

- General appearance
- Cardiovascular
- Lungs
- Abdomen

Brief physical examination data to be recorded on the CRF will include: 1) normal/abnormal and 2) a description of any abnormalities

4.1.6 Dietary assessment

During the screening period each prospective subject will participate in a 3-day diet diary assessment and analysis.

A trained investigative staff member will instruct subjects on how to complete a 3-day food diary. In addition, subjects will be taught how to estimate food quantities and how to record information regarding food brands and varieties in the diary. Subjects will be instructed not to change their personal eating habits during the 3-day diet assessment, but to eat normally and record all foods and quantities in the diary.

Each subject will record his or her food intake in a diary over 3 days during the screening period (2 days during the week and 1 day during the weekend). Subjects will return to the CRC with their record for analysis prior to Day –1.

A commercially available nutrition analysis software program will be used to analyze the dietary records. The results of the analysis will include the following:

- The contents of each meal and all snacks including all liquids consumed
- The total daily caloric intake for each day
- Total (in grams) carbohydrates, protein, fiber, and fat, (monounsaturated, polyunsaturated and saturated) for each day
- Percent of total caloric intake for each component (carbohydrates, protein, fiber, and fat)
- Dietary cholesterol (mg)
- Minerals (mg)
    - Sodium
    - Potassium
    - Calcium
    - Magnesium
    - Phosphorus
    - Iron
    - Zinc
- Vitamins (mg)
    - Total retinol
    - Thiamine

- Riboflavin
- Niacin
- Vitamin B6
- Vitamin B12
- Vitamin C
- Vitamin D
- Vitamin E

Additionally, the analysis will provide an average of the same components for the 3 days.

These components, with the exception of the contents of each meal, will be recorded on the CRF.

4.2 Pharmacokinetic measurements

For timing of individual samples, refer to the study plan (Table 1) and/or Appendix C.

4.2.1 Blood samples

Blood samples (4mL) for determination of rosuvastatin in plasma will be collected, processed, labeled and shipped as detailed in Appendices C and D. The date and time of collection will be recorded on the appropriate CRF.

4.2.2 Collection and processing of biological samples

Plasma samples for measurement of rosuvastatin concentration will be analyzed by liquid chromatography/mass spectrometry/mass spectrometry (LC/MS/MS). Full details of the methodology will be included in the clinical trial report.

4.3 Pharmacodynamic measurements

Not applicable

4.4 Safety measurements

4.4.1 Clinical laboratory assessments

Fasting blood samples (10 hours) for determination of clinical chemistry, hematology, and lipid profile parameters will be taken at the times given in the study plan (Table 1). The date and time of collection will be recorded on the appropriate CRF.

4.4.2 Urinalysis

A 10 mL midstream urine sample will be collected for urinalysis at the times given in the study plan (Table 1). If a sample is positive for protein or blood, a microscopic examination will be performed. Further investigations may be undertaken at the discretion of the investigator.

4.4.3 Serum pregnancy test

Female subjects of childbearing potential will have a serum pregnancy test conducted at the times specified in Table 1 for human chorionic gonadotrophin (HCG). The serum pregnancy test can be collected with the same sample as the blood chemistry. If the result is positive, the subject will not be allowed to proceed in the trial.

4.4.4 HIV and hepatitis testing

Testing for HIV antibody, HBsAG, and hepatitis C antibody is to be performed on all subjects at screening. If a test result is positive, the subject will not be allowed to proceed in the trial. Although the results of the HIV and hepatitis screens have to be documented in the subject's file, they will not be collected on the CRFs and will therefore not be recorded in the study database.

4.4.5 Sample collection

Samples will be collected in the following volumes specified:

Clinical chemistry/serum pregnancy	- 10mL
Hematology	- 10 mL
Lipid profile	- 10 mL
HIV/HBsAG/HepC	- 10 mL
Urinalysis	- 10 mL

The following laboratory variables will be measured:

Clinical chemistry	**Hematology**
Calcium	Hemoglobin
Phosphate	Hematocrit
Blood glucose	Platelet count
Total bilirubin	Red blood cell count
Alkaline phosphatase	White blood cell count with differential, including neutrophils, lymphocytes, monocytes, eosinophils, and basophils
Urea nitrogen (BUN)	
Uric acid	**Lipid Profile**
Chloride	Total cholesterol

Clinical chemistry	**Hematology**	
Carbon dioxide	TG	
Creatinine	LDL-C	
Total protein	HDL-C	
Albumin		
Aspartate aminotransferase (AST)	**Urinalysis**	Microscopic analysis of an formed element (if applicable)
Alanine aminotransferase (ALT)		
	pH	WBC
Sodium	Glucose	RBC
Potassium	Blood	Casts
Creatine kinase (CK)	Ketones	Crystals
Lactic dehydrogenase (LDH)	Protein	Epithlial cells
	Bilirubin	Bacteria
	Specific gravity	Mucous

4.4.6 Electrocardiographic measurements

A standard resting 12-lead ECG will be obtained at screening only and will be recorded after the subject has been lying down for 10 minutes.

4.4.7 Vital signs

For timing of individual measurements refer to study plan (Table 1).

Vital signs consist of a sitting heart rate, sitting blood pressure, and oral temperature.

Oral temperature will be measured in degrees Celsius (°C) at screening and Day –1 only.

AstraZeneca will provide the investigator reference range values for each vital sign measurement required by the protocol

4.4.7.1 Blood pressure and heart rate

Heart rate and blood pressure will be measured after the subject has been seated for at least 5 minutes. Heart rate will be determined by palpation of the radial pulse for a period of 30 seconds and then multiplied by two. Blood pressure will be measured using a blood pressure device with an appropriate cuff size. The same arm will be used for each measurement.

4.5 Genetic sampling and storage

Blood samples will be obtained from each subject and used to prepare deoxyribonucleic acid (DNA) samples. Consent for genetic research will be obtained on a separate consent form and

is completely optional – ie, refusal to provide such consent does not preclude participation in the main pharmacokinetic study. The blood samples obtained under this consent will be exclusively used to study the effects of genetic polymorphisms on the disposition of rosuvastatin. Currently, genes of special interest include OATP-C (which encodes a hepatic uptake transport protein) and MRP2 (which encodes a transport protein implicated in the biliary excretion of statins). However, other genes implicated in the ADME of statins may also be studied. DNA samples will not be used in other types of genetic or non-genetic research either as test or control specimens.

Blood samples may be stored for up to a year prior to DNA extraction, and the extracted DNA may be stored for up to 15 years. DNA samples, and any remaining blood samples will be destroyed 15 years after study completion (defined here as database lock).

4.5.1 Collection, labeling and shipment of genetic samples

Approximately 9 mL of blood will be collected for the genetic sample. Genetic samples will be collected, labeled and shipped as detailed in Appendix E.

4.6 Volume of blood sampling

The total volume of blood that will be drawn from each subject in this study is as follows:

Table 5 Volume of blood to be drawn from each subject

Assessment		**Sample volume (mL)**	**n of samples**	**Total volume (mL)**
Blood sampling for rosuvastatin		4 mL	19	76 mL
Genetic sample		9 mL	1	9 mL
Safety	Clinical chemistry	10 mL	3	30 mL
	Hematology	10 mL	3	30 mL
	Lipid profile	10 mL	1	10 mL
	HIV/HBsAG/Hepatitis C	10 mL	1	10 mL
Total				**165 mL**

4.7 Adverse events

The definitions of adverse events (AEs), serious adverse events (SAEs) and other significant adverse events (OAEs) are given in Appendix B. Additional guidance on defining a SAE and determining causality are also included in Appendix B. It is of the utmost importance that all staff involved in the study is familiar with the content of these sections. The principal investigator is responsible for ensuring this.

4.7.1 Recording of adverse events

Adverse events will be collected beginning Day –1 through final assessment on Day 4. SAEs will be collected from the time the informed consent is signed through final assessment on Day 4.

The following variables will be recorded for each AE:

Start date, stop date, maximum intensity, action taken, outcome, causality (yes or no) and whether it constitutes an SAE or not.

The intensity rating is defined as:

1 = mild (awareness of sign or symptom, but easily tolerated)

2 = moderate (discomfort sufficient to cause interference with normal activities)

3 = severe (incapacitating, with inability to perform normal activities)

It is important to distinguish between serious and severe AEs. Severity is a measure of intensity whereas seriousness is defined by the criteria in Appendix B. An AE of severe intensity need not necessarily be considered serious. For example, nausea that persists for several hours may be considered severe nausea, but not a SAE. On the other hand, a stroke that results in only a limited degree of disability may be considered a mild stroke but would be a SAE.

Should a pregnancy occur, it must be reported in accordance with the procedures described in Section 8.4, Procedures in case of pregnancy. Pregnancy in itself is not regarded as an AE unless there is a suspicion that an investigational product may have interfered with the effectiveness of a contraceptive medication.

4.7.2 Reporting of serious adverse events

When the investigator becomes aware of an SAE during the course of the study, the SAE must be reported to the local monitor or other AstraZeneca representative within one (1) day.

All SAEs have to be reported, whether or not considered causally related to the investigational product. All SAEs will be recorded in the case report form. The investigator is responsible for informing the Ethics Committee and/or the Regulatory Authority of the SAE as per local requirements.

The AstraZeneca representative will work with the investigator to compile all the necessary information to ensure that the appropriate AstraZeneca Drug Safety Department receives a report by day one for all fatal and life-threatening cases and by day five for all other SAEs.

If a non-serious AE becomes serious, this and other relevant follow-up information must also be provided to AstraZeneca within 1 day as described above.

5. STUDY MANAGEMENT

5.1 Monitoring

The monitoring of this study will be performed in accordance with the principles of Good Clinical Practice (GCP) as laid out in the International Conference on Harmonization (ICH) document "Good Clinical Practice: Consolidated Guideline".

5.1.1 Data verification

It is a prerequisite of this study that the study monitor has direct access to source data for data verification. This will be done by comparing data from the CRFs with those in the subject's medical notes (permission from the subject will be sought as part of the consent process). Such verification is an essential element of quality control, as it allows the rectification of transcription errors and omissions.

Monitoring will routinely be performed prior to the transfer of data to Data Management.

5.1.2 Direct access to source data

Not applicable

5.2 Archiving of study documentation

AstraZeneca will retain all documentation pertaining to this study in the AstraZeneca central file for as long as rosuvastatin is available for human consumption.

The investigator will retain all documentation pertaining to this study for at least 15 years.

5.3 Audits and inspections

Authorized representatives of AstraZeneca, a regulatory authority, an Independent Ethics Committee (IEC) or an Institutional Review Board (IRB) may visit the center to perform audits or inspections, including source data verification. The purpose of an AstraZeneca audit or inspection is to systematically and independently examine all study related activities and documents to determine whether these activities were conducted, and data were recorded, analyzed, and accurately reported according to the protocol, GCP guidelines of the ICH and any applicable regulatory requirements. The investigator should contact AstraZeneca immediately if contacted by a regulatory agency about an inspection at his or her center.

5.4 Training of staff

The principal investigator will maintain a record of all individuals involved in the study (medical, nursing and other staff). He or she will ensure that appropriate training relevant to the study is given to all of these staff, and that any new information of relevance to the performance of this study is forwarded to the staff involved.

5.5 Changes to the protocol

Study procedures will not be changed without the mutual agreement of the principal investigator and AstraZeneca.

If it is necessary for the study protocol to be amended, the amendment and/or a new version of the study protocol must be notified to or approved by each IEC or IRB, and in many countries also the local regulatory authority, before implementation. Local requirements must be followed.

If a protocol amendment requires a change to a particular center's Master Written Informed Consent Form, then AstraZeneca and the center's IEC or IRB must be notified. Approval of the revised Master Written Informed Consent Form by AstraZeneca and by the IEC or IRB is required before the revised form is used.

AstraZeneca will distribute amendments and new versions of the protocol to each principal investigator who in turn is responsible for the distribution of these documents to his or her IEC or IRB, and to the staff at his or her center. The distribution of these documents to the regulatory authority will be handled according to local practice.

5.6 Study agreements

The principal investigator at each center must comply with all the terms, conditions, and obligations of the study agreement for this study. In the event of any inconsistency between this protocol and the study agreement, this protocol shall prevail.

5.7 Study timetable and termination

The study is expected to start on and to be completed by .

5.8 Data management

5.8.1 Case report forms

CRFs will be used to record all data not captured electronically. The forms will be in triplicate on carbonless paper. Data should be recorded directly and legibly from the source documents onto the CRFs in black ballpoint pen. Corrections to the CRFs should be made legibly, initialed, and dated. Correction fluid or covering labels must not be used. The top and middle sheets will be collected and forwarded to data management personnel. The bottom sheet will be retained in the Investigator Study File.

The AstraZeneca Monitor will check data at the monitoring visits to the investigational site. The Investigator, together with the AstraZeneca Monitor, will ensure that the data in the CRFs are accurate, complete and legible.

AstraZeneca Data Management will enter the CRF data on an ongoing basis into their standard commercial database. The data will be verified and cleaned with electronic data checks comprised of validated computer programs and manual data review. Any missing,

impossible or inconsistent recordings in the CRFs will be referred back to the Investigator using a data clarification form (DCF), and be documented for each individual subject before Clean File Data status is declared. Responses should be received by Data Management and updated within an agreed number of days upon generating the data queries. These timelines will be reduced nearing Clean File. Clean File will be declared when all of the following have been completed: all data discrepancies are resolved or accepted; all SAEs have been reconciled with the clinical database; all coding is complete and has been medically reviewed and approved; and quality control of the database against the CRF and relevant data sources has been completed.

5.8.2 Genetic data

Genetic data will not be collected on the CRFs. The results of genetic research will be confidential. The results of any genetic research, any genetic sequences, cell lines, patents, diagnostic tests, drugs and biological products developed directly or indirectly from those samples, are the sole property of the study sponsor (and its successors, licensees, and assignees). Genetic data may be reviewed with research collaborators and published. Otherwise, no genetic data will be provided to the subject, the subject's family, the investigator, or any other physician who is treating the subject or who may treat the subject in the future. Neither the subject's insurance company nor the subject's employer will have any access to these research results. There is no direct benefit to the subject in having genetic research performed.

6. STATISTICAL METHODS AND DETERMINATION OF SAMPLE SIZE

6.1 Statistical evaluation

A comprehensive SAP will be prepared and finalized before database lock.

6.2 Description of outcome variables in relation to hypotheses

Although there are no hypotheses to be addressed in this protocol, this section will list the outcome variables related to the objectives.

Rosuvastatin exposure (primary objective): After a single oral dose of rosuvastatin, C_{max} and AUC will be determined for each subject. If AUC data cannot be determined in all subjects completing the study, $AUC_{(0-t)}$ will replace AUC as a primary endpoint.

Rosuvastatin exposure (secondary objective): The following pharmacokinetic parameters will be estimated and summarized: $AUC_{(0-t)}$, $t_{½\lambda z}$, t_{max}, and CL/F. With exception to CL/F, these pharmacokinetic parameters, along with C_{max} and AUC, will also be estimated and summarized for N-desmethyl rosuvastatin and rosuvastatin lactone.

Safety and tolerability (secondary objective): Incidence and severity of adverse events, blood pressure/heart rate (sitting), laboratory parameters (hematology, clinical chemistry, urinalysis), and physical examination.

Pharmacogenetic objective: Genes of interest include OATP-C and MRP-2, and may include any other gene that may be involved with the ADME of statins. Genetic information will not be included in the SAP or study report.

6.3 Description of analysis sets

There are two populations in this study. All subjects receiving the dose of study drug will be included in the safety population. The per-protocol (PP) population, also known as the evaluable population, is defined as a subset of the safety population, excluding those subjects with a protocol deviation felt to significantly influence the pharmacokinetics of the drug. In order to be evaluable, subjects must have completed all study procedures from the screening period to the final blood sampling for plasma levels of rosuvastatin. Definitions of all such protocol deviations will be made in the SAP. Examples include, but are not limited to, major changes in the administration of study drug or co-administration of medications expected to affect the pharmacokinetics.

All safety data will be summarized using the safety population (defined as any subject who received the dose of study drug). Pharmacokinetic data will be summarized using the PP population. In the case of the PP and safety populations being identical, only the safety population will be used throughout; this decision will be made at the time of data review and documented in the SAP.

A test for homogeneity of variance in C_{max} and AUC will be performed for the subjects in the Asian groups. If the test shows no difference and the means appear similar, then the data will be pooled. This data will be related to pharmacokinetic data generated from the subjects in the Caucasian control group.

The distribution of C_{max}, AUC and $AUC_{(0-t)}$ values for the Caucasian control group and for the Asian subjects residing in the United States will be summarized using descriptive statistics and presented graphically (e.g. box and whisker plots). There will be no statistical comparisons between groups.

6.4 Methods of statistical analyses

Statistical summaries will be carried out by the biostatistical group at AstraZeneca using the SAS system Version 8. Pharmacokinetic analysis will be carried out by the Clinical Pharmacokinetic Section, Experimental Medicine, at AstraZeneca.

Where standard summary statistics are referenced below, this will include the mean, standard deviation, median, minimum and maximum for continuous variables, and counts and percentages for discrete variables.

6.4.1 Demographic and baseline data

All demographic and baseline data, including medications, will be listed and summarized using standard summary statistics. No hypothesis test comparing the Asian subjects with the Caucasian control group will be made. Demographic data from the control group will be summarized as well.

6.4.2 Pharmacokinetics

C_{max} and t_{max} for each subject will be determined by inspection of the rosuvastatin plasma concentration-time profile. The terminal elimination rate constant (λ_z) will be calculated by log-linear regression of the terminal portion of the concentration-time profiles where there are sufficient data (a minimum of 3 plasma concentration values in the terminal log-linear phase, spanning an interval of at least 2 half-lives.) Terminal elimination half-life ($t_{½\lambda z}$) will be calculated as $0.693/\lambda_z$. The $AUC_{(0\text{-}t)}$ will be calculated by the linear trapezoidal rule ($AUC = AUC_{(0\text{-}t)} + C_{last}/\lambda_z$). $AUC_{(0\text{-}t)}$ will be extrapolated to infinity using λ_z to obtain AUC where there are sufficient data. Following the single dose administration of rosuvastatin, the CL/F will be calculated as Dose/AUC.

Pharmacokinetic data, C_{max}, $AUC_{(0\text{-}t)}$ and AUC, will be summarized for each Asian ethnic group and pooled, if appropriate, using descriptive statistics, including geometric means and coefficient of variation, and listed. In addition, 95% CI for the mean pharmacokinetic parameters will also be presented.

6.4.3 Adverse events

Adverse events will be summarized by System Organ Class and High Level Term, using MeDRA. All adverse event data will be listed for all subjects. Separate listings of all serious adverse events, deaths or other significant adverse events will be presented.

6.4.4 Laboratory data

All laboratory safety data, incorporating hematology, clinical chemistry, lipid profiles, and urinalysis data will be listed, with deviations from the normal range explicitly noted on the listings.

Continuous laboratory data will be summarized using standard summary statistics. Both absolute values and change from pre-dose baseline will be summarized.

Discrete laboratory data will be summarized using standard summary statistics.

6.4.5 12-lead ECG data

Twelve-lead ECG data will be summarized and listed using standard summary statistics.

6.4.6 Vital signs

Vital signs data will summarized and listed using standard summary statistics. Both absolute values and change from pre-dose baseline will be summarized. In addition, mean plots showing change within treatment group across time will be produced for absolute values.

6.4.7 Physical examination

Physical examination abnormalities will be listed.

6.5 Determination of sample size

No formal statistical comparisons will be made between the ethnic groups compared to the Caucasians. However, 22 subjects will be sufficient to characterize the pharmacokinetics of rosuvastatin in these populations subsequent to a single 20 mg dose.

6.6 Interim analyses

No interim analysis is planned

6.7 Data Presentation

The details of data presentation will be provided in the SAP.

6.8 Data or safety monitoring committee

Not applicable

7. ETHICS

7.1 Ethics review

The final study protocol and the final version of the Master Written Informed Consent Form must be approved or given a favorable opinion in writing by the IEC or IRB as appropriate. The investigator must submit written approval to AstraZeneca before he or she can enroll any subject into the study.

The final study protocol, including the final version of the Informed Consent Form, must be approved or given a favorable opinion in writing by an IRB or IEC as appropriate. The investigator must submit written approval to AstraZeneca before he or she can enroll any subject into the study.

The Principal Investigator is responsible for informing the IRB or IEC of any amendment to the protocol in accordance with local requirements. In addition, the IRB or IEC must approve all advertising used to recruit subjects for the study. The protocol must be reapproved by the IEC or IRB annually, as local regulations require.

The Principal Investigator(s) is also responsible for providing the IRB with reports of any SAEs from any other study conducted with the investigational product. AstraZeneca will provide this information to the principal investigator.

Progress reports and notifications of SAEs will be provided to the IRB or IEC according to local regulations and guidelines.

7.2 Ethical conduct of the study

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

7.3 Subject information and consent

The principal investigator at each center will ensure that the subject is given full and adequate oral and written information about the nature, purpose, and the possible risk and benefit of the study. Subjects must also be notified that they are free to discontinue from the study at any time. The subject should be given the opportunity to ask questions and allowed time to consider the information provided.

The subject's signed and dated informed consent must be obtained before conducting any procedure specifically for the study.

The principal investigator must store the original, signed Master Written Informed Consent Form. A copy of the Master Written Informed Consent Form must be given to the subject. If modifications are made according to local requirements, the new version has to be approved by AstraZeneca.

7.4 Subject data protection

For study sites within the US or in studies where non-US subjects' protected health information (subject data) will come into the US through a covered entity (eg, Central Lab/Reader), the Master Informed Consent Form will incorporate, or be accompanied by, a separate document incorporating HIPAA-compliant wording by which subjects authorize the use and disclosure of the Protected Health Information by the Investigator and by those persons who need that information for the purposes of the study.

The Master Informed Consent Form will explain that study data will be stored in a computer database, maintaining confidentiality in accordance with national data legislation. All data computer processed by AstraZeneca will be identified by subject number/study code/initials.

The Master Informed Consent Form will also explain that for data verification purposes, authorized representatives of AstraZeneca, a regulatory authority, an IEC or IRB may require direct access to parts of the hospital or practice records relevant to the study, including subjects' medical history.

8. PROCEDURES IN CASE OF EMERGENCY, OVERDOSE, OR PREGNANCY

8.1 Medical emergency contact procedure

In the case of a medical emergency, contact AstraZeneca personnel shown below.

AstraZeneca Contact	Telephone Number & Fax

For Serious Adverse event reporting

- Drug Safety Fax

8.2 Procedures in case of medical emergency

The principal investigator(s) is responsible for ensuring that procedures and expertise are available to cope with medical emergencies during the study. **A medical emergency usually constitutes an SAE and should be reported as such, see Section 4.7.2**

8.3 Procedures in case of overdose

There is no specific antidote available for overdose of rosuvastatin. In the event of overdose, treatment should be symptomatic and supportive.

As with the management of any overdose, the possibility of multiple drug ingestion should be considered. For current information on treatment of any drug overdose, a certified Regional Poison Control Center should be contacted. Telephone numbers are listed in the Physicians' Desk Reference (PDR) or local telephone book.

8.4 Procedures in case of pregnancy

Pregnancy itself is not regarded as an adverse event unless there is a suspicion that the investigational product under study may have interfered with the effectiveness of a contraceptive medication. However, the outcome of all pregnancies (spontaneous

miscarriage, elective termination, normal birth or congenital abnormality) must be followed up and documented even if the subject was discontinued from the study.

All reports of congenital abnormalities/birth defects are SAEs. Spontaneous miscarriages should also be reported and handled as SAEs. Elective abortions without complications should not be handled as AEs. All outcomes of pregnancy must be reported to AstraZeneca on the pregnancy outcomes report form.

9. REFERENCES

1. Nishizato Y, Jeiri I, Suski H et al. Polymorphisms of OATP-C (SLC21A6) and OAT3 (SLC22A8) genes: Consequences for pravastatin pharmacokinetics. Clin Pharmcol Ther 2003;73:554-65

2. Barnes, Jessica; Bennett, Claudette; U.S. Census Bureau; Census 2000 Brief; "The Asian Population;" issued February 2002, <http://www.census.gov/prod/2002pubs/c2kbr01-16.pdf>

AstraZeneca

Clinical Pharmacology Study Protocol: Appendix B	
Drug substance:	Rosuvastatin
Study Code:	D3560C00001
Appendix Edition No:	Final Version 1
Appendix Date:	

Appendix B
Additional Safety Information

1. DEFINITION OF AN ADVERSE EVENT

An adverse event is the development of an undesirable medical condition or the deterioration of a pre-existing medical condition following or during exposure to a pharmaceutical product, whether or not considered causally related to the product. An undesirable medical condition can be symptoms (eg, nausea, chest pain), signs (eg, tachycardia, enlarged liver) or the abnormal results of an investigation (eg, laboratory findings, electrocardiogram). In clinical studies, an AE can include an undesirable medical condition occurring at any time, including run-in or washout periods, even if no study treatment has been administered.

2. DEFINITION OF OTHER SIGNIFICANT ADVERSE EVENTS

An AstraZeneca expert will identify other adverse events (OAEs) during the evaluation of safety data for the Clinical Study Report. Significant adverse events of particular clinical importance, other than SAEs and those AEs leading to discontinuation of the patient from study treatment, will be classified as OAEs. Examples of these are marked hematological and other laboratory abnormalities, and certain events that lead to intervention (other than those already classified as serious), dose reduction or significant additional treatment. For each OAE, a narrative will be written and included in the Clinical Study Report.

3. DEFINITION OF A SERIOUS ADVERSE EVENT

A serious adverse event is an AE occurring during any study phase (ie, run-in, treatment, washout, follow-up), and at any dose of the investigational product, comparator or placebo, that fulfills one or more of the following criteria:

Results in death

Is immediately life-threatening

Requires in-patient hospitalization or prolongation of existing hospitalization

Results in persistent or significant disability or incapacity

Is a congenital abnormality or birth defect

Is an important medical event that may jeopardize the patient or may require medical intervention to prevent one of the outcomes listed above

4. FURTHER GUIDANCE ON THE DEFINITION OF A SERIOUS ADVERSE EVENT (SAE)

Life threatening

'Life-threatening' means that the subject was at immediate risk of death from the AE as it occurred or it is suspected that use or continued use of the product would result in the subject's death. 'Life-threatening' does not mean that had an AE occurred in a more severe form it might have caused death (eg, hepatitis that resolved without hepatic failure).

Hospitalization

Outpatient treatment in an emergency room is not in itself a serious AE, although the reasons for it may be (eg, bronchospasm, laryngeal edema). Hospital admissions and/or surgical operations planned before or during a study are not considered AEs if the illness or disease existed before the subject was enrolled in the study, provided that it did not deteriorate in an unexpected way during the study.

Important medical event or medical intervention

Medical and scientific judgment should be exercised in deciding whether a case is serious in situations where important medical events may not be immediately life threatening or result in death, hospitalization, disability or incapacity but may jeopardize the subject or may require medical intervention to prevent one or more outcomes listed in the definition of serious. These should usually be considered as serious.

Examples of such events are:

- Angioedema not severe enough to require intubation but requiring IV. hydrocortisone treatment
- Hepatotoxicity caused by acetaminophen overdose requiring treatment with N-acetylcysteine
- Intensive treatment in an emergency room or at home for allergic bronchospasm
- Blood dyscrasias (eg, neutropenia or anemia requiring blood transfusion, etc.) or convulsions that do not result in hospitalization
- Development of drug dependency or drug abuse

5. A GUIDE TO INTERPRETING THE CAUSALITY QUESTION

The following factors should be considered when deciding if there is a "reasonable possibility" that an AE may have been caused by the drug.

- Time Course. Exposure to suspect drug. Has the subject actually received the suspect drug? Did the AE occur in a reasonable temporal relationship to the administration of the suspect drug?

- Consistency with known drug profile. Was the AE consistent with the previous knowledge of the suspect drug (pharmacology and toxicology) or drugs of the same pharmacological class? OR could the AE be anticipated from its pharmacological properties?

- Dechallenge experience. Did the AE resolve or improve on stopping or reducing the dose of the suspect drug?

- No alternative cause. The AE cannot be reasonably explained by another etiology such as the underlying disease, other drugs, other host or environmental factors.

- Rechallenge experience. Did the AE reoccur if the suspected drug was reintroduced after having been stopped? AstraZeneca would not normally recommend or support a rechallenge.

- Laboratory tests. A specific laboratory investigation (if performed) has confirmed the relationship?

A “reasonable possibility” could be considered to exist for an AE where one or more of these factors exist.

In contrast, there would not be a “reasonable possibility” of causality if none of the above criteria apply or where there is evidence of exposure and a reasonable time course but any dechallenge (if performed) is negative or ambiguous or there is another more likely cause of the AE.

In difficult cases, other factors could be considered such as:

- Is this a recognized feature of overdose of the drug?

- Is there a known mechanism

Ambiguous cases should be considered as being a “reasonable possibility” of a causal relationship unless further evidence becomes available to refute this. Causal relationship in cases where the disease under study has deteriorated due to lack of effect should be classified as no reasonable possibility.

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Clinical Pharmacology Study Protocol: Appendix C	
Drug substance:	Rosuvastatin
Study Code:	D3560C0001
Appendix Edition No:	Final Version 1
Appendix Date:	

Appendix C
Collection, processing, labeling, and shipping of blood samples

1. COLLECTION AND PROCESSING OF BLOOD SAMPLES

Venous blood samples (4mL) will be collected into tubes containing lithium heparin anticoagulant at the times indicated in the table below. The date and time of collection and tube reference will be recorded on the appropriate CRF.

Blood samples must be protected from light, cooled to 4 °C, and centrifuged at 1500 G (relative centrifugal force) for 10 minutes at 4 °C within 30 minutes of blood sampling. Following centrifugation, a 1.5 mL aliquot of plasma will be transferred to a clean polypropylene tube and an equal volume (1.5 mL) of 0.1 M acetate buffer, pH 4.0, also at 4 °C, will be added and mixed thoroughly with a vortex mixer to provide plasma for analysis of rosuvastatin. See Appendix D for preparation of the buffer solution for plasma samples.

After ensuring that the 3 mL buffered plasma sample (1.5 mL plasma, 1.5 mL buffer solution) has been thoroughly mixed, a fresh pipette will be used to transfer 1.5 mL of the buffered plasma to a clean 2 mL polypropylene tube and the remainder of the buffered plasma will be transferred to a second 2 mL clean polypropylene tube. The two samples will be protected from light, frozen, and stored at -70°C until analysis.

One sample will be used for analysis of rosuvastatin at AstraZeneca Drug Metabolism and Pharmacokinetics (DMPK) and one sample will be kept at the CRC as a spare in the event of loss or damage of the other sample. AstraZeneca DMPK, Wilmington, Delaware, will analyze plasma samples for measurement of drug concentration.

Table 1 Schedule of blood sampling and tube numbers

Day	Scheduled time	Tube number
1	0.5 hour pre-dose	1
1	0.5 hour post-dose	2
1	1 hour post-dose	3
1	2 hours post-dose	4
1	3 hours post-dose	5
1	4 hours post-dose	6
1	5 hours post-dose	7
1	6 hours post-dose	8
1	8 hours post-dose	9
1	10 hours post-dose	10
1	12 hours post-dose	11
2	18 hours post-dose	12
2	24 hours post-dose	13
2	30 hours post-dose	14
2	36 hours post-dose	15
3	48 hours post-dose	16
3	54 hours post-dose	17
3	60 hours post-dose	18
4	72 hours post-dose	19

2. LABELING OF PLASMA SAMPLES

The labels supplied by AstraZeneca must be applied to the plasma sample tubes. The labels will include the following information:

- Study number: D3560C00001
- Subject number
- Tube number
- Scheduled time
- Matrix: (Plasma-rosuva or Plasma-spare)

3. SHIPMENT OF PLASMA SAMPLES

All plasma samples for rosuvastatin assays will be shipped to AstraZeneca Pharmaceuticals, LP. The samples must be shipped frozen in dry ice via expedited overnight delivery. The samples must be packed securely to avoid damage during transit, should be double bagged to contain leaks, and should be packed with a sufficient quantity of dry ice to ensure they remain frozen for at minimum of 72 hours. Allowance should be made for possible delay of shipment. All applicable shipping regulations must be followed. Documentation sufficient to identify each sample must be included in the shipment. The AstraZeneca contact (see below) must be notified by phone or fax when the samples are shipped and all shipping details must be provided.

Ship samples on Mondays-Wednesdays. Do not ship on or the day before a legal holiday.

Samples should be shipped to:

AstraZeneca

Clinical Pharmacology Study Protocol: Appendix D	
Drug substance:	Rosuvastatin
Study Code:	D3560C00001
Appendix Edition No:	Final Version 1
Appendix Date:	

Appendix D
Preparation of buffer solution for rosuvastatin plasma samples

1. STEP 1 – PREPARE 0.1 M ACETIC ACID

To a 1 liter volumetric flask containing approximately 500 mLs of HPLC water, add 5.75 mLs of concentrated glacial acetic acid. Mix thoroughly and fill to volume with HPLC water.

2. STEP 2 – PREPARE 0.1 M SODIUM ACETATE

To a 500 mL volumetric flask containing approximately 200 mLs of HPLC water, add 6.8 grams of sodium acetate trihydrate. Mix to dissolve and fill to volume with HPLC water

3. STEP 3 – PREPARE BUFFER

In a large beaker, combine 820 mLs of 0.1 M acetic acid and 180 mLs of 0.1 M sodium acetate. Adjust to pH 4.0 with 0.1 M sodium acetate.

Store final buffers in a 1-liter plastic screw cap container. Label with a 1-month expiration, store at 4°C.

AstraZeneca

Clinical Pharmacology Study Protocol: Appendix E	
Drug substance:	Rosuvastatin
Study Code:	D3560C00001
Appendix Edition No:	Final Version 1
Appendix Date:	

Appendix E
Collection, labeling, storage, and shipment of blood samples for genetic analysis

1. COLLECTION OF GENETIC SAMPLES

Approximately 9 mL of blood will be collected from consenting subjects into EDTA-coated polypropylene tubes at the time indicated in the protocol study plan (Table 1). The blood should be mixed by gentle inversion of the tube.

Glass tubes must not be used as they may break during transport and freeze-thaw cycles.

Heparin must not be used as an anticoagulant as it may interfere with downstream genotyping methodology.

2. LABELLING OF GENETIC SAMPLES

Collection tubes will be labeled with the following information:

- Genotyping sample
- Date of sample
- Study number
- Subject number

3. STORAGE OF GENETIC SAMPLES AT THE STUDY SITE

After collection, blood samples must be stored appropriately at the site of collection and transported to the AstraZeneca Clinical Genotyping Laboratory as soon as possible.

The samples will be frozen immediately after collection and may be stored at –20°C or –70°C. The samples should be kept frozen until analyzed.

If blood samples are to be stored at –20°C, non-frost free freezers must be used to prevent repeated freeze-thaw of blood, which may reduce yield and quality of the DNA, obtained.

Samples must not be thawed and then re-frozen at any point.

4. SHIPMENT OF GENETIC SAMPLES

For safety reasons, all blood samples must be contained. Standard procedures for transporting biological samples as defined by the courier and in compliance with local regulations will be followed.

Blood samples for genetic analysis collected from each consenting subject will be shipped to AstraZeneca Clinical Genotyping Laboratory. The samples must be shipped frozen in dry ice via expedited overnight delivery. The samples must be packed securely to avoid damage during transit, must be double bagged to contain leaks, and will be packed with a sufficient quantity of dry ice to ensure they remain frozen for a minimum of 72 hours. Allowance should be made for possible delay of shipment.

All applicable shipping regulations must be followed. Documentation sufficient to identify each sample (ie, list of subject identification numbers) must be included in the shipment. The AstraZeneca contact ( ) must be notified by telephone or fax when the samples are shipped. All shipping details, including study ID, number of samples, list of sample ID's, courier name, airway bill number, and date of shipment must be provided.

Samples must be shipped on Mondays or Tuesdays only to ensure delivery before the weekend. Do not ship on or the day before a legal holiday.

Samples will be sent o the following address: