REPORT ON BIOEQUIVALENCE STUDY

(Protocol No.: BEQ-1346-ATOR-2014, Version No.: 01, Date: 24th November 2014)

Study Title:

Bioequivalence study of single dose Atorvastatin Calcium tablets 80 mg manufactured by Macleods Pharmaceuticals Ltd., India in comparison with Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) Distributed by Pfizer Inc., USA in healthy, adult, human subjects under fasting condition.

Study Design:

An open label, balanced, analyst blind, randomized, two-treatment, four-period, two-sequence, single dose, fully replicated with reference scaled crossover bioequivalence study on 30 healthy, adult, human subjects under fasting condition.

Investigational Product Details		**Dose**
i) Test Formulation (T)	: Atorvastatin Calcium tablets 80 mg Macleods Pharmaceuticals Ltd., India	1 × 80 mg
ii) Reference Formulation (R)	: Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) Distributed by Pfizer Inc., USA	1 × 80 mg

Duration of Clinical Phase	:	12th January 2015 – 23rd February 2015
Duration of Bioanalytical Phase	:	02nd July 2015 – 23rd July 2015
Duration of Statistical Phase	:	29th July 2015 – 30th July 2015
Report Status	:	Final
Version	:	01
Dated	:	01st August 2015
Supersedes Version	:	None
Dated	:	Not Applicable

Sponsor	**Clinical Facility**
MACLEODS PHARMACEUTICALS LTD. G-2, Mahakali Caves Road, Shanti Nagar, Andheri - (East), Mumbai – 400 093. India Telephone No.: 91-22-28306435 / 28314611 Fax No.: 91-22-28304641	MACLEODS PHARMACEUTICALS LTD. Bioequivalence Department, R & D-II, Plot no. 95, Road no. 16, Opp. Suncity Hotel, MIDC, Andheri - (East), Mumbai 400 093, India. Telephone No.: 91-22-67258412 Fax No.: 91-22-28304641 Email: drashish@macleodspharma.com

Statement of Compliance: This study was conducted in compliance with ICH GCP including archiving of essential documents.

Principal Investigator **:** Dr. Rajendra Sonde

Study Director **:** Dr. Ashish Mungantiwar

Sponsor's Representative **:** Ms. Shalmali Ambekar

2.0 SYNOPSIS OF THE REPORT

Name of the Sponsor: Macleods Pharmaceuticals Ltd., India **Name of the Finished Product:** Atorvastatin Calcium tablets 80 mg **Name of Active Ingredient:** Atorvastatin Calcium	Individual Study Table Referring to part of the Dossier **Volume:** N/AP **Page:** N/AP	(For National Authority Use only)

Title of Study: Bioequivalence study of single dose Atorvastatin Calcium tablets 80 mg manufactured by Macleods Pharmaceuticals Ltd., India in comparison with Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) Distributed by Pfizer Inc., USA in healthy, adult, human subjects under fasting condition.

Investigator: Dr. Rajendra Sonde, Principal Investigator

Study Centre: Macleods Pharmaceuticals Ltd., Bioequivalence Department

Publication (reference): Not Applicable

Study period

Date of Screening	:	26th December 2014 – 10th January 2015
Duration of Clinical Phase	:	12th January 2015 – 23rd February 2015
Period 1	:	12th January 2015 – 15th January 2015
Period 2	:	19th January 2015 – 22nd January 2015
Period 3	:	26th January 2015 – 29th January 2015
Period 4	:	02nd February 2015 – 05th February 2015
Duration of Bioanalytical Phase	:	02nd July 2015 – 23rd July 2015
Duration of Statistical Phase	:	29th July 2015 – 30th July 2015

Objectives:

i) Pharmacokinetic: To evaluate the comparative oral bioavailability of single dose of Atorvastatin Calcium tablets 80 mg manufactured by Macleods Pharmaceuticals Ltd., India with Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) Distributed by Pfizer Inc., USA in healthy, adult, human subjects under fasting condition.

ii) Safety: To monitor the safety and tolerability of a single oral dose of Atorvastatin Calcium tablets 80 mg when administered in healthy, adult, human subjects under fasting condition.

Name of the Sponsor: Macleods Pharmaceuticals Ltd., India	Individual Study Table Referring to part of the Dossier **Volume:** N/AP **Page:** N/AP	(For National Authority Use only)
Name of the Finished Product: Atorvastatin Calcium tablets 80 mg		
Name of Active Ingredient: Atorvastatin Calcium		

Methodology:

Serial blood samplings (pre-dose and at 0.17, 0.33, 0.50, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00, 2.33, 2.67, 3.00, 3.50, 4.00, 4.50, 5.00, 5.50, 6.00, 8.00, 12.00, 16.00, 24.00, 34.00 and 48.00 hours post-dose) were done before and up to 48.00 hours post-drug administration.

Each blood sample (1 × 5 mL) were collected into 5 mL blood collection tube containing K_2EDTA as anticoagulant.

Analysis of plasma samples for atorvastatin and for ortho- and parahydroxylated metabolites of atorvastatin were done using an in-house validated LC-MS/MS method.

During the activity from sample collection to storage and during analysis, the samples were maintained in cold condition by placing the sample containers in ice water slurry.

A non-compartmental method was used to calculate pharmacokinetic parameters using drug concentration time profile. Statistical comparison of the pharmacokinetic parameters of both the formulations was done to assess bioequivalence.

Number of Subjects (planned and analyzed):

A total of 30 subjects were planned and enrolled, of these 30, subject number 24 was withdrawn from the study in period 1 (post-dose) on principal investigator advice due to serious adverse events. Subject number 06 and 25 were withdrawn from the study in period 1 (post-dose) on principal investigator advice due to adverse events. Subject number 02, 12, 26 and 29 did not report to the facility due to personal reason for period 2 thus considered dropped-out from the study. Subject number 14 and 18 did not report to the facility due to personal reason for period 3 thus considered dropped-out from the study. Subject number 11 was found to participate in other clinical trial and thus was withdrawn from the study based on exclusion criteria during check-in of period 3 (pre-dose). Subject number 03 was withdrawn from the study in period 3 (post-dose) on principal investigator advice due to adverse event. Thus nineteen subjects completed all the periods of the study.

The plasma samples of nineteen subjects (subject number 01, 04, 05, 07 to 10, 13, 15, 16, 17, 19 to 23, 27, 28 and 30) completing the study was analyzed for atorvastatin and for ortho- and parahydroxylated metabolites of atorvastatin concentration level and the data were taken for pharmacokinetic and statistical evaluations.

The plasma samples of subject number 02, 03, 06, 11, 12, 14, 18, 24 to 26 and 29 who were considered dropped-out from the study were not analyzed.

<table>
<tr><td>Name of the Sponsor:
Macleods Pharmaceuticals Ltd., India</td><td rowspan="3">Individual Study Table Referring to part of the Dossier
Volume: N/AP
Page: N/AP</td><td rowspan="3">(For National Authority Use only)</td></tr>
<tr><td>Name of the Finished Product:
Atorvastatin Calcium tablets 80 mg</td></tr>
<tr><td>Name of Active Ingredient:
Atorvastatin Calcium</td></tr>
<tr><td colspan="3">Diagnosis and main criteria for inclusion: Healthy human subjects within the age range of 18 to 45 years with body-mass index (BMI between 18.50 kg/m^2 and 29.99 kg/m^2 (both inclusive) with body weight not less than 50 kg (for males) and with body weight not less than 45 kg (for females) and having absence of significant disease, clinically significant laboratory values, absence of clinically significant medical history and normal physical examination during the screening and complying with inclusion and exclusion criteria were the criteria for inclusion.</td></tr>
<tr><td colspan="3">i) Test Formulation (T):
Atorvastatin Calcium Tablets 80 mg
Batch No.: BAC9402A
Manufacturing Date: June 2014
Expiry Date: May 2016
Manufactured by: Macleods Pharmaceuticals Ltd. India
Dose: 1 tablet
Mode of administration: Administered orally with 240 mL of drinking water.
Batch size: 1,25,000 Tablets
Assay: 102.1%</td></tr>
<tr><td colspan="3">ii) Reference Formulation (R):
Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg)
Lot No.: V122400
Manufacturing Date: N/AV
Expiry Date: Oct-15
Distributed by: Parke-Davis Division of Pfizer Inc., NY, NY 10017
NDC 0071-0158-23
Dose: 1 tablet
Mode of administration: Administered orally with 240 mL of drinking water.
Batch size: Not Available
Assay: 99.9%</td></tr>
<tr><td colspan="2">Duration of treatment</td><td>Single dose in all the four periods</td></tr>
</table>

Name of the Sponsor: Macleods Pharmaceuticals Ltd., India **Name of the Finished Product:** Atorvastatin Calcium tablets 80 mg **Name of Active Ingredient:** Atorvastatin Calcium	Individual Study Table Referring to part of the Dossier **Volume:** N/AP **Page:** N/AP	(For National Authority Use only)

Criteria for Evaluation:

Efficacy:

If $S_{WR} \geq 0.294$, i.e. intra-subject C.V. for reference formulation ≥ 30% then reference-scaled average bioequivalence approach is used to determine bioequivalence (BE) for the relevant pharmacokinetic parameters (C_{max}, AUC_{0-t} and $AUC_{0-\infty}$).

If $S_{WR} < 0.294$, i.e. intra-subject C.V. for reference formulation < 30% then the two-one sided test (Unscaled average bioequivalence approach) is used to determine BE for the relevant pharmacokinetic parameters (C_{max}, AUC_{0-t} and $AUC_{0-\infty}$).

Where S_{WR} is the within-subject standard deviation (SD) of the reference formulation

Reference-Scaled Average Bioequivalence Approach:

If the 95% Upper Bounds for $(\left(\overline{Y}_T - \overline{Y}_R\right)^2 - \theta * S_{WR}^2)$ are ≤ 0 and the point estimate (Ratio T/R) of atorvastatin are entirely included in the range of 80.00 – 125.00% for C_{max}, AUC_{0-t} and $AUC_{0-\infty}$ then the treatments would be claimed to be bioequivalent.

Unscaled Average Bioequivalence Approach:

If the 90 % confidence interval for C_{max}, AUC_{0-t} and $AUC_{0-\infty}$ of atorvastatin are form the basis for concluding the bioequivalence of atorvastatin in product R and T. If the confidence intervals are entirely included in the range of 80.00 – 125.00% for C_{max}, AUC_{0-t} and $AUC_{0-\infty}$ log-transformed then the treatments would be claimed to be bioequivalent.

O-hydroxy atorvastatin and p-hydroxy atorvastatin the following data submitted: Individual and mean concentrations, individual and mean pharmacokinetic parameters, and geometric means and ratios of means for C_{max}, AUC_{0-t} and $AUC_{0-\infty}$. The measurement of o-hydroxy atorvastatin and p-hydroxy atorvastatin metabolite levels in plasma is not used for comparison of bioequivalence.

Safety: To monitor the safety and tolerability of a single oral dose of Atorvastatin Calcium tablets 80 mg when administered in healthy, adult, human subjects under fasting condition.

Statistical Methods: Method for statistical analysis using the reference-scaled average bioequivalence mixed scaling approach:

Reference-Scaled Average Bioequivalence Approach:

The log-transformed pharmacokinetic parameter C_{max} of atorvastatin analysed using an ANOVA model. Calculated Point estimate (Ratio T/R) and 95% Upper Bound for $(\left(\overline{Y}_T - \overline{Y}_R\right)^2 - \theta * S_{WR}^2)$ of C_{max}. Test factors effects by using the log-transformed pharmacokinetic parameter C_{max} of atorvastatin are analysed using an ANOVA model.

Name of the Sponsor: Macleods Pharmaceuticals Ltd., India **Name of the Finished Product:** Atorvastatin Calcium tablets 80 mg **Name of Active Ingredient:** Atorvastatin Calcium	Individual Study Table Referring to part of the Dossier **Volume:** N/AP **Page:** N/AP	(For National Authority Use only)

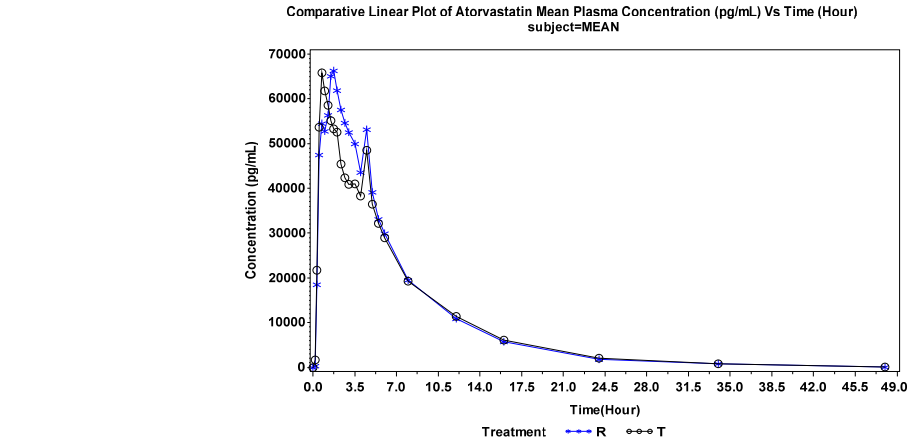
Unscaled Average Bioequivalence Approach: The log-transformed pharmacokinetic parameters (AUC_{0-t} and $AUC_{0-\infty}$) of atorvastatin analysed using an ANOVA model. Calculated 90% confidence interval for the ratio of both the products averages (geometric means) and power of AUC_{0-t} and $AUC_{0-\infty}$.

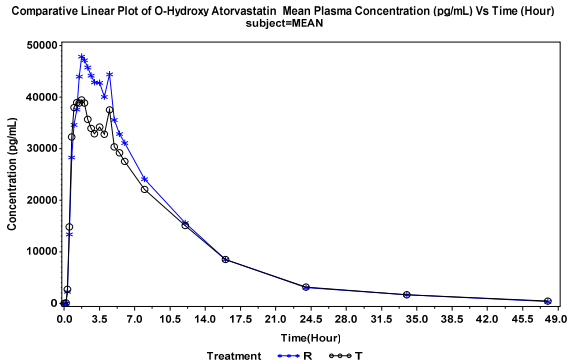
Ratios of mean AUC_{0-t} to mean $AUC_{0-\infty}$ for test and reference are expressed in percentage, Individual Ratio of AUC_{0-t} to $AUC_{0-\infty}$ for test and reference are expressed in percentage, Ratio T/R for each subject at C_{max}, AUC_{0-t} and $AUC_{0-\infty}$ and % Bioavailability of C_{max}, AUC_{0-t} and $AUC_{0-\infty}$ Analysis were performed using SAS® version 9.4.

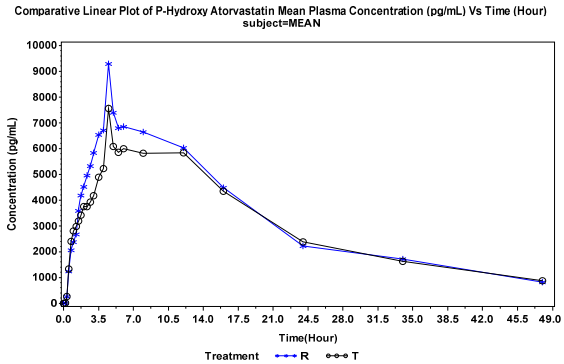
O-hydroxy atorvastatin and p-hydroxy atorvastatin the following data submitted: Individual and mean concentrations, individual and mean pharmacokinetic parameters, and geometric means and ratios of means for C_{max}, AUC_{0-t} and $AUC_{0-\infty}$. The measurement of o-hydroxy atorvastatin and p-hydroxy atorvastatin metabolite levels in plasma were not used for comparison of bioequivalence.

SUMMARY - CONCLUSION

EFFICACY RESULTS:







Name of the Sponsor: Macleods Pharmaceuticals Ltd., India	Individual Study Table Referring to part of the Dossier **Volume:** N/AP **Page:** N/AP	(For National Authority Use only)
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The 95 % Upper Bound of ln-transformed parameters for atorvastatin summarized below:

Pharmacokinetic Parameters	Within subject S.D. for Reference (S_{WR})	Within Subject Variance for Reference (S^2wr)	Intra Subject C.V. for Reference (%)	Point Estimate (Ratio (T/R) (%))	95% Upper Bound
C_{max} (pg/mL)	0.302	0.091	30.90	99.80	-0.0
AUC_{0-t} (pg*hrs/mL)	0.239	0.057	24.25	94.98	-0.0
$AUC_{0-\infty}$ (pg*hrs/mL)	0.237	0.056	24.04	95.37	-0.0

The 90 % confidence intervals of ln-transformed parameters for atorvastatin summarized below:

Geometric Mean, Ratio & Power and 90 % Confidence Interval for Atorvastatin					
Pharmacokinetic Parameters	Geometric Mean		Ratio (T/R) (%)	Power (%)	90 % Confidence Interval (%)
	Test (T)	Reference (R)			
AUC_{0-t} (pg*hrs/mL)	412677.267	434478.335	94.98	99.83	87.82 - 102.72
$AUC_{0-\infty}$(pg*hrs/mL)	421562.572	442031.352	95.37	99.87	88.33 - 102.97

The 90 % confidence intervals of ln-transformed parameters for o-hydroxy atorvastatin summarized below:

Geometric Mean& Ratio for O-Hydroxy Atorvastatin			
Pharmacokinetic Parameters	Geometric Mean		Ratio (T/R) (%)
	Test (T)	Reference (R)	
C_{max} (pg/mL)	46018.088	52747.775	87.24
AUC_{0-t} (pg*hrs/mL)	404899.236	443962.113	91.20
$AUC_{0-\infty}$(pg*hrs/mL)	416361.909	456601.032	91.19

The 90 % confidence intervals of ln-transformed parameters for p-hydroxy atorvastatin summarized below:

Geometric Mean& Ratio for P-Hydroxy Atorvastatin			
Pharmacokinetic Parameters	Geometric Mean		Ratio (T/R) (%)
	Test (T)	Reference (R)	
C_{max} (pg/mL)	6733.313	8460.222	79.59
AUC_{0-t} (pg*hrs/mL)	124743.129	136827.696	91.17
$AUC_{0-\infty}$(pg*hrs/mL)	142592.862	155476.628	91.71

Name of the Sponsor: Macleods Pharmaceuticals Ltd., India	Individual Study Table Referring to part of the Dossier **Volume:** N/AP **Page:** N/AP	(For National Authority Use only)
Name of the Finished Product: Atorvastatin Calcium tablets 80 mg		
Name of Active Ingredient: Atorvastatin Calcium		

SAFETY RESULTS:

Three subjects (subject number 03, 06, and 25) experienced adverse events during study period and one subject (subject number 24) experienced serious adverse events (SAE) during study period. During post-study safety assessment adverse events were reported for one subject (subject number 12).

CONCLUSION:

The 95% Upper Bound for ($\left(\overline{Y}_T - \overline{Y}_R\right)^2 - \theta * S_{WR}^2$) for C_{max} was <=0 and Point estimate (Ratio T/R) of atorvastatin was entirely included in the range of 80.00 – 125.00% for C_{max}.

The 90% confidence interval for the ratio (Test/Reference) of AUC_{0-t} and $AUC_{0-\infty}$ of atorvastatin were within the acceptable limits of bioequivalence 80.00% - 125.00%.

Thus it is concluded that the test product, Atorvastatin Calcium tablets 80 mg manufactured by Macleods Pharmaceuticals Ltd., India is bioequivalent to the reference product; Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) Distributed by Pfizer Inc., USA in healthy, adult, human subjects under fasting condition.

Date of the report: 01st August 2015

2.1 STATEMENT OF INVESTIGATORS

STUDY TITLE:

Bioequivalence study of single dose Atorvastatin Calcium tablets 80 mg manufactured by Macleods Pharmaceuticals Ltd., India in comparison with Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) Distributed by Pfizer Inc., USA in healthy, adult, human subjects under fasting condition.

The study was conducted in accordance with the ethical principles that have their origins in the Declaration of Helsinki; ICH GCP; Schedule-Y and other regulatory provisions under the Drug and Cosmetics Rules; GCP Guidelines issued by Central Drugs Standard Control Organization (CDSCO); "Ethical Guidelines for Biomedical Research on Human Subjects" published by Indian Council of Medical Research (ICMR) and in accordance with USFDA requirement.

We have read this report and confirm that to the best of our knowledge it accurately describes the conduct and results of the study.

Dr. Rajendra Sonde, Principal Investigator — Date: 01/08/15

Dr. Ashish Mungantiwar, Study Director — Date: 1/8/15

Macleods Pharmaceuticals Ltd.,
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India

3.0 TABLE OF CONTENTS FOR THE INDIVIDUAL CLINICAL STUDY REPORT

4.0 LIST OF ABBREVIATIONS AND DEFINITION OF TERMS

µL	:	Microlitre
ALT	:	Alanine transaminase
ANOVA	:	Analysis of Variance
ANA	:	Antinuclear Antibodies
apo B	:	apolipoprotein B
AST	:	Aspartate Aminotransferase
AUC	:	Area under Curve
AUC_{0-t}	:	Area under the concentration versus time curve calculated using the Trapezoidal rule upto the last measurable time point
$AUC_{0-\infty}$	:	Area under the concentration versus time curve from time zero to infinity
BLQ	:	Below Limit of Quantification
BMI	:	Body Mass Index
B.P	:	Blood Pressure
BPM	:	Beats Per Minute
BUN	:	Blood Urea Nitrogen
CBC	:	Complete Blood Count
CK	:	Creatine Kinase
C_{max}	:	Maximum observed concentration
COA	:	Certificate of Analysis
CPK	:	Creatine phosphokinase
CPU	:	Clinical Pharmacology Unit
CV	:	Coefficient of Variance
CYP	:	Cytochrome
^0C	:	Degree Centigrade
D	:	Day
ECG	:	Electro-Cardiogram
ESR	:	Erythrocyte Sedimentation Rate
GCP	:	Good Clinical Practices
GGT	:	Gama Glutamyl Transpeptidase
GLP	:	Good Laboratory Practices
cGMP	:	current Good Manufacturing Practices
HBsAg	:	Hepatitis B Surface Antigen
HC	:	Health Canada
HCV	:	Hepatitis C Virus
HDL-C	:	High Density Lipoprotein-Cholesterol
HIV	:	Human Immunodeficiency Virus
HMG-CoA	:	3-hydroxy-3-methylglutaryl-coenzyme A
HPFB	:	Health Products and Food Branch
hrs	:	Hour(s)

I.D.	:	Identity
ICF	:	Informed Consent Form
ICH	:	International Conference on Harmonization
ICMR	:	Indian Council of Medical Research
IDL	:	Intermediate Density Lipoproteins
IEC	:	Independent Ethics Committee
IP	:	Investigational Product
IU	:	International Unit
IUD	:	Intrauterine device
K_{el}	:	Elimination rate constant
K_e_first	:	First time point of the terminal log-linear phase used to estimate the terminal disposition rate constant (K_e) with bestfit regression method used.
K_e_last	:	Last time point of the terminal log-linear phase used to estimate the terminal disposition rate constant (K_e) with bestfit regression method used.
K_2EDTA	:	Dipotassium Ethylene diamine tetra-acetic acid
LC-MS/MS	:	Liquid Chromatography with Tandem Mass Spectrometry
LDL	:	Low Density Lipoprotein
LDL-C	:	Low Density Lipoprotein-Cholesterol
L/kg	:	Liter / kilogram
L/h/kg	:	Liter / hour / kilogram
LLOQ	:	Lower Limit of Quantification
Lp	:	lipoprotein
LSM	:	Least Square Means
ME	:	Medical Examination
mg	:	milligram
mcg/ml	:	microgram per millilitre
mL	:	millilitre
N/AP	:	Not Applicable
N/AV	:	Not Available
npoints	:	No. of time points of the terminal log-linear phase used to estimate the terminal disposition rate constant (K_e) with bestfit regression method used.
oz	:	Ounce
PA view	:	Posterior Anterior View
PCV	:	Packed Cell Volume
pg/mL	:	picogram per millilitre
PK	:	Pharmacokinetic
QA	:	Quality Assurance
RBC	:	Red Blood Cell
RH	:	Relative Humidity
rpm	:	Revolution Per Minute

SAE(s)	:	Serious Adverse Event(S)
SADR	:	Serious Adverse Drug Reactions
SAS	:	Statistical Analysis System
SD	:	Standard Deviation
SGOT	:	Serum Glutamate Oxaloacetate Transaminase
SGPT	:	Serum Glutamate Pyruvate Transaminase
SOP(s)	:	Standard Operating Procedure(S)
SQ	:	Subject Questionnaire
TIA	:	Transient Ischemic Attack
TG	:	Triglycerides
TPD	:	Therapeutic Products Directorate
$t_{1/2}$	:	Terminal Half-life
t_{max}	:	Time of maximum measured plasma concentration. If maximum value occurs at more than one point, T_{max} is defined as the first point with this value in each period
ULN	:	Upper limit of normal
USFDA	:	United States Food and Drug Administration
VLDL-C	:	Very Low Density Lipoprotein-Cholesterol
w/v	:	Weight by Volume

5.0 ETHICS

5.1 Independent Ethics Committee (IEC)

The protocol, BEQ-1346-ATOR-2014, version No. 01, dated: 24th November 2014 and English ICF, version no. 01, dated: 24th November 2014, Hindi and Marathi ICFs, version No. 01, dated: 24th November 2014, Case Record Form (CRF) version no. 01, dated: 24th November 2014 along with COA were sent to were sent to CBP - Independent Ethics Committee on 24th November 2014. The CBP - Independent Ethics Committee in its meeting held on 29th November 2014 gave approval to the submitted protocol, ICFs and CRF.

Approval was granted after the review of documents and the approved version which was followed for the study conducted bears the following version number and date:

Protocol	: Version No. 01, dated: 24th November 2014
English ICF	: Version No. 01, dated: 24th November 2014
Hindi ICF	: Version No. 01, dated: 24th November 2014
Marathi ICF	: Version No. 01, dated: 24th November 2014
Case Record form	: Version No.01, dated: 24th November 2014

Dr. Mohan Kembhavi (Chairperson) chaired the IEC meeting. The copy of the IEC approved protocol is appended as appendix 16.1.1. The details of IEC consulted along with approval letter and IEC approved ICFs are given in appendix 16.1.3.

5.2 Ethical Conduct of the Study

The study was conducted in accordance with the ethical principles that have their origins in the Declaration of Helsinki; ICH GCP; Schedule-Y and other regulatory provisions under the Drug and Cosmetics Rules; GCP Guidelines issued by Central Drugs Standard Control Organization (CDSCO); “Ethical Guidelines for Biomedical Research on Human Subjects” published by Indian Council of Medical Research (ICMR) and in accordance with USFDA requirement.

5.3 Subject Information and Consent

Subjects from the pool of healthy volunteers who were screened within 21 days prior to the dosing day were considered as potential participants in the study. Before admission of volunteers into the Clinical pharmacology unit (CPU) on the pre-study day, they were given a verbal presentation of the information on the study together with a written document (in the language that they can understand best) describing the purpose, procedures, and risks of the study together with a description of the obligations of the subjects. Volunteers gave their written consent for participating in the study by signing with date the informed consent form. The signed copy of the form is kept in the study specific ICF file at the investigator’s/ institution site and the subjects were also given a copy for their own retention. A copy of the consent form used is appended in appendix 16.1.3. Before the volunteers undergo the pre-screening process for acceptance into healthy volunteer bank, they were provided with written and verbal information about the nature of the tests to be performed following which all willing volunteers gave their written consent.

6.0 INVESTIGATORS AND STUDY ADMINISTRATIVE STRUCTURE

Sponsor's Representative

Name : Ms. Shalmali Ambekar (M. Pharm Pharmacology)
Assistant Manager (Medical Services)

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BIOEQUIVALENCE DEPARTMENT PERSONNEL

Study Director:

Name : Dr. Ashish Mungantiwar (Ph.D. Pharmacology)

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Principal Investigator:

Name : Dr. Rajendra Sonde (M.B.B.S., PGDHHM, D.PHARM)

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A) Clinical Facility : Macleods Pharmaceuticals Ltd.,
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B) Pathology Laboratory : Macleods Pharmaceuticals Ltd.,
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C) X-Ray Facility : Macleods Pharmaceuticals Ltd.,
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D) Bioanalytical Facility : Macleods Pharmaceuticals Ltd.,
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E) Statistical Operations : Macleods Pharmaceuticals Ltd.,
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Ext. No.: 267
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The list of investigators with their affiliations, their role in the study and their curriculum vitae are appended as appendix 16.1.4. The list of other important participants in the study is appended in appendix 16.1.4. The declaration statement by the principal investigator is appended as appendix 16.1.5.

7.0 INTRODUCTION

Pharmacology

LIPITOR is a selective, competitive inhibitor of HMG-CoA reductase, the rate-limiting enzyme that converts 3-hydroxy-3methylglutaryl-coenzyme A to mevalonate, a precursor of sterols, including cholesterol. Cholesterol and triglycerides circulate in the bloodstream as part of lipoprotein complexes. With ultracentrifugation, these complexes separate into HDL (high-density lipoprotein), IDL (intermediate-density lipoprotein), LDL (low-density lipoprotein), and VLDL (very-low-density lipoprotein) fractions. Triglycerides (TG) and cholesterol in the liver are incorporated into VLDL and released into the plasma for delivery to peripheral tissues. LDL is formed from VLDL and is catabolized primarily through the high-affinity LDL receptor. Clinical and pathologic studies show that elevated plasma levels of total cholesterol (total-C), LDL-cholesterol (LDL-C), and apolipoprotein B (apo B) promote human atherosclerosis and are risk factors for developing cardiovascular disease, while increased levels of HDL-C are associated with a decreased cardiovascular risk. In animal models, LIPITOR lowers plasma cholesterol and lipoprotein levels by inhibiting HMG-CoA reductase and cholesterol synthesis in the liver and by increasing the number of hepatic LDL receptors on the cell surface to enhance uptake and catabolism of LDL; LIPITOR also reduces LDL production and the number of LDL particles. LIPITOR reduces LDL-C in some patients with homozygous familial hypercholesterolemia (FH), a population that rarely responds to other lipid-lowering medication(s).

A variety of clinical studies have demonstrated that elevated levels of total-C, LDL-C, and apo B (a membrane complex for LDL-C) promote human atherosclerosis. Similarly, decreased levels of HDL-C (and its transport complex, apo A) are associated with the development of atherosclerosis. Epidemiologic investigations have established that cardiovascular morbidity and mortality vary directly with the level of total-C and LDL-C, and inversely with the level of HDL-C.

LIPITOR reduces total-C, LDL-C, and apo B in patients with homozygous and heterozygous FH, nonfamilial forms of hypercholesterolemia, and mixed dyslipidemia. LIPITOR also reduces VLDL-C and TG and produces variable increases in HDL-C and apolipoprotein A-1. LIPITOR reduces total-C, LDL-C, VLDL-C, apo B, TG, and non-HDL-C, and increases HDL-C in patients with isolated hypertriglyceridemia. LIPITOR reduces intermediate density lipoprotein cholesterol (IDL-C) in patients with dysbetalipoproteinemia.

Like LDL, cholesterol-enriched triglyceride-rich lipoproteins, including VLDL, intermediate density lipoprotein (IDL), and remnants, can also promote atherosclerosis. Elevated plasma triglycerides are frequently found in a triad with low HDL-C levels and small LDL particles, as well as in association with non-lipid metabolic risk factors for coronary heart disease. As such, total plasma TG has not consistently been shown to be an independent risk factor for CHD. Furthermore, the

independent effect of raising HDL or lowering TG on the risk of coronary and cardiovascular morbidity and mortality has not been determined.

LIPITOR, as well as some of its metabolites, are pharmacologically active in humans. The liver is the primary site of action and the principal site of cholesterol synthesis and LDL clearance. Drug dosage, rather than systemic drug concentration, correlates better with LDL-C reduction. Individualization of drug dosage should be based on therapeutic response. [1]

Clinical Pharmacokinetics

Absorption: LIPITOR is rapidly absorbed after oral administration; maximum plasma concentrations occur within 1 to 2 hours. Extent of absorption increases in proportion to LIPITOR dose. The absolute bioavailability of atorvastatin (parent drug) is approximately 14% and the systemic availability of HMG-CoA reductase inhibitory activity is approximately 30%. The low systemic availability is attributed to presystemic clearance in gastrointestinal mucosa and/or hepatic first-pass metabolism. Although food decreases the rate and extent of drug absorption by approximately 25% and 9%, respectively, as assessed by Cmax and AUC, LDL-C reduction is similar whether LIPITOR is given with or without food. Plasma LIPITOR concentrations are lower (approximately 30% for Cmax and AUC) following evening drug administration compared with morning. However, LDL-C reduction is the same regardless of the time of day of drug administration.

Pharmacokinetics Parameters of Atorvastatin and its active metabolites are as follows [2] -

Parameters	Units	Atorvastatin	Ortho- hydroxylated derivatives	Parahydroxylated derivatives
C_{max}	µg/ml	13.4 (9.5)	9.8 (6.1)	1.2 ± 0.6 (0.8–1.6)*
T_{max}	hours	1.00 to 2.00	2 (0.5–4)	9.2 ± 8.8 (3.36–15.2)*
$T_{1/2}$	hours	7 (3.7)	10.8 (3.5)	-
Bioavailability	%	≅14%	-	-
Volume of distribution	liters	≅381	-	-
Plasma proteins binding	%	≥98	-	-
blood/plasma ratio	-	≅0.25	-	-

*in haemodialysis patients

Distribution: Mean volume of distribution of LIPITOR is approximately 381 liters. LIPITOR is ≥98% bound to plasma proteins. A blood/plasma ratio of approximately 0.25 indicates poor drug penetration into red blood cells. Based on observations in rats, LIPITOR is likely to be secreted in human milk.

Metabolism: LIPITOR is extensively metabolized to ortho-and parahydroxylated derivatives and various beta-oxidation products. *In vitro* inhibition of HMG-CoA reductase by ortho-and parahydroxylated metabolites is equivalent to that of LIPITOR. Approximately 70% of circulating inhibitory activity for HMG-CoA reductase is attributed to active metabolites. *In vitro* studies suggest the importance of LIPITOR metabolism by cytochrome P450 3A4, consistent with increased plasma concentrations of LIPITOR in humans following co-administration with erythromycin, a known inhibitor of this isozyme. In animals, the ortho-hydroxy metabolite undergoes further glucuronidation.

Excretion: LIPITOR and its metabolites are eliminated primarily in bile following hepatic and/or extra-hepatic metabolism; however, the drug does not appear to undergo enterohepatic recirculation. Mean plasma elimination half-life of LIPITOR in humans is approximately 14 hours, but the half-life of inhibitory activity for HMG-CoA reductase is 20 to 30 hours due to the contribution of active metabolites. Less than 2% of a dose of LIPITOR is recovered in urine following oral administration.

Gender: Plasma concentrations of LIPITOR in women differ from those in men (approximately 20% higher for C_{max} and 10% lower for AUC); however, there is no clinically significant difference in LDL-C reduction with LIPITOR between men and women. [1]

Indications and Uses

Therapy with lipid-altering agents should be only one component of multiple risk factor intervention in individuals at significantly increased risk for atherosclerotic vascular disease due to hypercholesterolemia. Drug therapy is recommended as an adjunct to diet when the response to a diet restricted in saturated fat and cholesterol and other nonpharmacologic measures alone has been inadequate. In patients with CHD or multiple risk factors for CHD, LIPITOR can be started simultaneously with diet.

LIPITOR has not been studied in conditions where the major lipoprotein abnormality is elevation of chylomicrons. [1]

Adverse Reactions

The following serious adverse reactions are discussed in greater detail as: Rhabdomyolysis and myopathy Liver enzyme abnormalities [1]

Because clinical trials are conducted under widely varying conditions, the adverse reaction rates observed in the clinical studies of a drug cannot be directly compared to rates in the clinical trials of another drug and may not reflect the rates observed in clinical practice.

In the LIPITOR placebo-controlled clinical trial database of 16,066 patients (8755 LIPITOR vs. 7311 placebo; age range 10–93 years, 39% women, 91% Caucasians, 3% Blacks, 2% Asians, 4% other) with a median treatment duration of 53 weeks, 9.7% of patients on LIPITOR and 9.5% of the patients on placebo discontinued due to adverse reactions regardless of causality. The five most common adverse reactions in patients treated with LIPITOR that led to treatment discontinuation and occurred at a rate greater than placebo were: myalgia (0.7%), diarrhea

(0.5%), nausea (0.4%), alanine aminotransferase increase (0.4%), and hepatic enzyme increase (0.4%).

The most commonly reported adverse reactions (incidence ≥ 2% and greater than placebo) regardless of causality, in patients treated with LIPITOR in placebo controlled trials (n=8755) were: nasopharyngitis (8.3%), arthralgia (6.9%), diarrhea (6.8%), pain in extremity (6.0%), and urinary tract infection (5.7%).

The frequency of clinical adverse reactions, regardless of causality, reported in ≥ 2% and at a rate greater than placebo in patients treated with LIPITOR (n=8755), from seventeen placebo-controlled trials.

Table summarizes the frequency of clinical adverse reactions, regardless of causality, reported in ≥ 2% and at a rate greater than placebo in patients treated with LIPITOR (n=8755), from seventeen placebo-controlled trials.

Nasopharyngitis
Arthralgia
Diarrhea
Pain in extremity
Urinary tract infection
Dyspepsia
Nausea
Musculoskeletal
pain
Muscle Spasms
Myalgia
Insomnia
Pharyngolaryngeal pain

Post marketing Experience

The following adverse reactions have been identified during postapproval use of LIPITOR. Because these reactions are reported voluntarily from a population of uncertain size, it is not always possible to reliably estimate their frequency or establish a causal relationship to drug exposure.

Adverse reactions associated with LIPITOR therapy reported since market introduction, that are not listed above, regardless of causality assessment, include the following: anaphylaxis, angioneurotic edema, bullous rashes (including erythema multiforme, Stevens-Johnson syndrome, and toxic epidermal necrolysis), rhabdomyolysis, fatigue, tendon rupture, fatal and non-fatal hepatic failure, dizziness, depression, peripheral neuropathy, and pancreatitis.

There have been rare reports of immune-mediated necrotizing myopathy associated with statin use.

There have been rare postmarketing reports of cognitive impairment (e.g., memory loss, forgetfulness, amnesia, memory impairment, confusion) associated with statin use. These cognitive issues have been reported for all statins. The reports are generally nonserious, and reversible upon statin discontinuation, with variable times to symptom onset (1 day to years) and symptom resolution (median of 3 weeks).

Precautions and Contraindications

Skeletal Muscle

Rare cases of rhabdomyolysis with acute renal failure secondary to myoglobinuria have been reported with LIPITOR and with other drugs in this class. A history of renal impairment may be a risk factor for the development of rhabdomyolysis. Such patients merit closer monitoring for skeletal muscle effects.

Atorvastatin, like other statins, occasionally causes myopathy, defined as muscle aches or muscle weakness in conjunction with increases in creatine phosphokinase (CPK) values >10 times ULN. The concomitant use of higher doses of atorvastatin with certain drugs such as cyclosporine and strong CYP3A4 inhibitors (e.g., clarithromycin, itraconazole, and HIV protease inhibitors) increases the risk of myopathy/rhabdomyolysis.

There have been rare reports of immune-mediated necrotizing myopathy (IMNM), an autoimmune myopathy, associated with statin use. IMNM is characterized by: proximal muscle weakness and elevated serum creatine kinase, which persist despite discontinuation of statin treatment; muscle biopsy showing necrotizing myopathy without significant inflammation; improvement with immunosuppressive agents.

Myopathy should be considered in any patient with diffuse myalgias, muscle tenderness or weakness, and/or marked elevation of CPK. Patients should be advised to report promptly unexplained muscle pain, tenderness, or weakness, particularly if accompanied by malaise or fever or if muscle signs and symptoms persist after discontinuing LIPITOR. LIPITOR therapy should be discontinued if markedly elevated CPK levels occur or myopathy is diagnosed or suspected.

The risk of myopathy during treatment with drugs in this class is increased with concurrent administration of cyclosporine, fibric acid derivatives, erythromycin, clarithromycin, the hepatitis C protease inhibitor telaprevir, combinations of HIV protease inhibitors, including saquinavir plus ritonavir, lopinavir plus ritonavir, tipranavir plus ritonavir, darunavir plus ritonavir, fosamprenavir, and fosamprenavir plus ritonavir, niacin, or azole antifungals. Physicians considering combined therapy with LIPITOR and fibric acid derivatives, erythromycin, clarithromycin, a combination of saquinavir plus ritonavir, lopinavir plus ritonavir, darunavir plus ritonavir, fosamprenavir, or fosamprenavir plus ritonavir, azole antifungals, or lipid-modifying doses of niacin should carefully weigh the potential benefits and risks and should carefully monitor patients for any signs or symptoms of muscle pain, tenderness, or weakness, particularly during the initial months of therapy and during any periods of upward dosage titration of either drug. Lower starting and

maintenance doses of atorvastatin should be considered when taken concomitantly with the aforementioned drugs. Periodic creatine phosphokinase (CPK) determinations may be considered in such situations, but there is no assurance that such monitoring will prevent the occurrence of severe myopathy.

Prescribing recommendations for interacting agents are summarized in table:

Table: Drug Interactions Associated with Increased Risk of Myopathy/Rhabdomyolysis

Interacting Agents	Prescribing Recommendations
Cyclosporine, HIV protease inhibitors (tipranavir plus ritonavir), hepatitis C protease inhibitor (telaprevir)	Avoid atorvastatin
HIV protease inhibitor (lopinavir plus ritonavir)	Use with caution and lowest dose necessary
Clarithromycin, itraconazole, HIV protease inhibitors (saquinavir plus ritonavir*, darunavir plus ritonavir, fosamprenavir, fosamprenavir plus ritonavir)	Do not exceed 20 mg atorvastatin daily
HIV protease inhibitor (nelfinavir) Hepatitis C protease inhibitor (boceprevir)	Do not exceed 40 mg atorvastatin daily

Cases of myopathy, including rhabdomyolysis, have been reported with atorvastatin co-administered with colchicine, and caution should be exercised when prescribing atorvastatin with colchicine.

LIPITOR therapy should be temporarily withheld or discontinued in any patient with an acute, serious condition suggestive of a myopathy or having a risk factor predisposing to the development of renal failure secondary to rhabdomyolysis (e.g., severe acute infection, hypotension, major surgery, trauma, severe metabolic, endocrine and electrolyte disorders, and uncontrolled seizures).

Liver Dysfunction

Statins, like some other lipid-lowering therapies, have been associated with biochemical abnormalities of liver function. Persistent elevations (>3 times the upper limit of normal [ULN] occurring on 2 or more occasions) in serum transaminases occurred in 0.7% of patients who received LIPITOR in clinical trials. The incidence of these abnormalities was 0.2%, 0.2%, 0.6%, and 2.3% for 10, 20, 40, and 80 mg, respectively.

One patient in clinical trials developed jaundice. Increases in liver function tests (LFT) in other patients were not associated with jaundice or other clinical signs or symptoms. Upon dose reduction, drug interruption, or discontinuation, transaminase levels returned to or near pretreatment levels without sequelae. Eighteen of 30 patients with persistent LFT elevations continued treatment with a reduced dose of LIPITOR.

It is recommended that liver enzyme tests be obtained prior to initiating therapy with LIPITOR and repeated as clinically indicated. There have been rare postmarketing reports of fatal and non-fatal hepatic failure in patients taking statins, including atorvastatin. If serious liver injury with clinical

symptoms and/or hyperbilirubinemia or jaundice occurs during treatment with LIPITOR, promptly interrupt therapy. If an alternate etiology is not found, do not restart LIPITOR.

LIPITOR should be used with caution in patients who consume substantial quantities of alcohol and/or have a history of liver disease. Active liver disease or unexplained persistent transaminase elevations are contraindications to the use of LIPITOR.

Endocrine Function

Increases in HbA1c and fasting serum glucose levels have been reported with HMG-CoA reductase inhibitors, including LIPITOR.

Statins interfere with cholesterol synthesis and theoretically might blunt adrenal and/or gonadal steroid production. Clinical studies have shown that LIPITOR does not reduce basal plasma cortisol concentration or impair adrenal reserve. The effects of statins on male fertility have not been studied in adequate numbers of patients. The effects, if any, on the pituitary-gonadal axis in premenopausal women are unknown. Caution should be exercised if a statin is administered concomitantly with drugs that may decrease the levels or activity of endogenous steroid hormones, such as ketoconazole, spironolactone, and cimetidine.

CNS Toxicity

Brain hemorrhage was seen in a female dog treated for 3 months at 120 mg/kg/day. Brain hemorrhage and optic nerve vacuolation were seen in another female dog that was sacrificed in moribund condition after 11 weeks of escalating doses up to 280 mg/kg/day. The 120 mg/kg dose resulted in a systemic exposure approximately 16 times the human plasma area-under-the-curve (AUC, 0-24 hours) based on the maximum human dose of 80 mg/day. A single tonic convulsion was seen in each of 2 male dogs (one treated at 10 mg/kg/day and one at 120 mg/kg/day) in a 2-year study. No CNS lesions have been observed in mice after chronic treatment for up to 2 years at doses up to 400 mg/kg/day or in rats at doses up to 100 mg/kg/day. These doses were 6 to 11 times (mouse) and 8 to 16 times (rat) the human AUC (0-24) based on the maximum recommended human dose of 80 mg/day.

CNS vascular lesions, characterized by perivascular hemorrhages, edema, and mononuclear cell infiltration of perivascular spaces, have been observed in dogs treated with other members of this class. A chemically similar drug in this class produced optic nerve degeneration (Wallerian degeneration of retinogeniculate fibers) in clinically normal dogs in a dose-dependent fashion at a dose that produced plasma drug levels about 30 times higher than the mean drug level in humans taking the highest recommended dose.

Use in Patients with Recent Stroke or TIA

In a post-hoc analysis of the Stroke Prevention by Aggressive Reduction in Cholesterol Levels (SPARCL) study where LIPITOR 80 mg vs. placebo was administered in 4,731 subjects without CHD who had a stroke or TIA within the preceding 6 months, a higher incidence of hemorrhagic stroke was seen in the LIPITOR 80 mg group compared to placebo (55, 2.3% atorvastatin vs. 33,

1.4% placebo; HR: 1.68, 95% CI: 1.09, 2.59; p=0.0168). The incidence of fatal hemorrhagic stroke was similar across treatment groups (17 vs. 18 for the atorvastatin and placebo groups, respectively). The incidence of nonfatal hemorrhagic stroke was significantly higher in the atorvastatin group (38, 1.6%) as compared to the placebo group (16, 0.7%). Some baseline characteristics, including hemorrhagic and lacunar stroke on study entry, were associated with a higher incidence of hemorrhagic stroke in the atorvastatin group.

CONTRAINDICATIONS

Active liver disease, which may include unexplained persistent elevations in hepatic transaminase levels Hypersensitivity to any component of this medication

Pregnancy

Women who are pregnant or may become pregnant. LIPITOR may cause fetal harm when administered to a pregnant woman. Serum cholesterol and triglycerides increase during normal pregnancy, and cholesterol or cholesterol derivatives are essential for fetal development. Atherosclerosis is a chronic process and discontinuation of lipid-lowering drugs during pregnancy should have little impact on the outcome of long-term therapy of primary hypercholesterolemia. There are no adequate and well-controlled studies of LIPITOR use during pregnancy; however in rare reports, congenital anomalies were observed following intrauterine exposure to statins. In rat and rabbit animal reproduction studies, atorvastatin revealed no evidence of teratogenicity.

LIPITOR SHOULD BE ADMINISTERED TO WOMEN OF CHILDBEARING AGE ONLY WHEN SUCH PATIENTS ARE HIGHLY UNLIKELY TO CONCEIVE AND HAVE BEEN INFORMED OF THE POTENTIAL HAZARDS. If the patient becomes pregnant while taking this drug, LIPITOR should be discontinued immediately and the patient apprised of the potential hazard to the fetus. [1]

Nursing mothers

It is not known whether atorvastatin is excreted into human milk; however a small amount of another drug in this class does pass into breast milk. Because statins have the potential for serious adverse reactions in nursing infants, women who require LIPITOR treatment should not breastfeed their infants. [1]

Study Rationale

The Macleods Pharmaceuticals Ltd. has developed generic alternative to the reference drug product of Atorvastatin Calcium Tablets 80 mg. Therefore, its bioequivalence with the reference drug product must be evaluated in fasting and Fed condition. In the present study, the single dose of test product Atorvastatin Calcium tablets 80 mg manufactured by Macleods Pharmaceuticals Ltd., India was compared with Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) distributed by Pfizer Inc., USA under fasting condition.

Justification of Choice of Reference Product

Lipitor® 80 mg (atorvastatin calcium tablets 80mg) is qualified as acceptable reference listed drug product by USFDA.

8.0 STUDY OBJECTIVES

The bioequivalence study presented here was carried out for evaluating the following objectives:

8.1 Pharmacokinetic:

To evaluate the comparative oral bioavailability of single dose of Atorvastatin Calcium tablets 80 mg manufactured by Macleods Pharmaceuticals Ltd., India with Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) Distributed by Pfizer Inc., USA in healthy, adult, human subjects under fasting condition.

8.2 Safety

To monitor the safety and tolerability of a single oral dose of Atorvastatin Calcium tablets 80 mg when administered in healthy, adult, human subjects under fasting condition.

9.0 INVESTIGATIONAL PLAN

9.1 Overall Study Design and Plan – Description

This was an open label, balanced, analyst blind, randomized, two-treatment, four-period, two-sequence, single dose, fully replicated with reference scaled crossover bioequivalence study on 30 healthy, adult, human subjects under fasting condition.

ADMISSION AND STAY

Period 1:

On 12th January 2015, informed consent was presented to all the volunteers. At the time of check-in, eligibility was assessed by carrying out urine test for drugs of abuse, medical examination, confirming compliance to protocol based upon inclusion and exclusion criteria and recording vitals. Breath of the volunteer was analyzed to check the consumption of alcohol using breath alcohol analyzer. The thirty volunteers who were found to be compliant with the requirements of the protocol were checked in Clinical Pharmacology Unit (CPU) between 18:57 hours to 20:54 hours.

During the in-house stay, monitoring of vital signs and subject questionnaire at the time of check-in, pre-dose and at 1.00, 3.00, 9.00 and 24.00 hours post-dose (Time points being relative to the investigational product dosing) was done. Medical examinations were carried out at the time of check-in and check-out of the study.

All the subjects enrolled into the study were housed in the Clinical Pharmacology Unit (CPU) until check-out on 14th January 2015.

The subjects were asked to visit the clinical facility for ambulatory blood sample collection at 34.00 and 48.00 hours post-dose. Breath of the subject was analyzed to check the consumption of alcohol using breath alcohol analyzer during each ambulatory visit. Vital signs and subject questionnaire was done at 34.00 and 48.00 hours post-dose and medical examinations were carried out at 48.00 hours post-dose.

Period 2 :

On 19th January 2015, twenty-three subjects reported to the facility. At the time of check-in, eligibility was assessed by carrying out test for drugs of abuse, medical examination, confirming compliance to protocol based upon exclusion criteria and recording vitals. Breath of the subject was analyzed to check the consumption of alcohol using breath alcohol analyzer. Twenty-three subjects who were found to be compliant with the requirements of the protocol were checked in Clinical Pharmacology Unit (CPU) between 19:15 hours to 20:46 hours.

During the in-house stay, monitoring of vital signs and subject questionnaire at the time of check-in, pre-dose and at 1.00, 3.00, 9.00 and 24.00 hours post-dose (Time points being relative to the investigational product dosing) was done. Medical examinations were carried out at the time of check-in and check-out of the study.

All the subjects checked-in into the study were housed in the Clinical Pharmacology Unit (CPU) until check-out on 21st January 2015.

The subjects were asked to visit the clinical facility for ambulatory blood sample collection at 34.00 and 48.00 hours post-dose. Breath of the subject was analyzed to check the consumption of alcohol using breath alcohol analyzer during each ambulatory visit. Vital signs and subject questionnaire was done at 34.00 and 48.00 hours post-dose and medical examinations were carried out at 48.00 hours post-dose.

Period 3:

On 26th January 2015, twenty-one subjects reported to the facility. At the time of check-in, eligibility was assessed by carrying out test for drugs of abuse, medical examination, confirming compliance to protocol based upon exclusion criteria and recording vitals. Breath of the subject was analyzed to check the consumption of alcohol using breath alcohol analyzer. Subject number 11 was found to participate in other clinical trial and thus was withdrawn from the study based on exclusion criteria. Twenty subjects who were found to be compliant with the requirements of the protocol were checked in Clinical Pharmacology Unit (CPU) between 18:40 hours to 20:22 hours.

During the in-house stay, monitoring of vital signs and subject questionnaire at the time of check-in, pre-dose and at 1.00, 3.00, 9.00 and 24.00 hours post-dose (Time points being relative to the investigational product dosing) was done. Medical examinations were carried out at the time of check-in and check-out of the study.

All the subjects checked-in into the study were housed in the Clinical Pharmacology Unit (CPU) until check-out on 28th January 2015.

The subjects were asked to visit the clinical facility for ambulatory blood sample collection at 34.00 and 48.00 hours post-dose. Breath of the subject was analyzed to check the consumption of alcohol using breath alcohol analyzer during each ambulatory visit. Vital signs and subject questionnaire was done at 34.00 and 48.00 hours post-dose and medical examinations were carried out at 48.00 hours post-dose.

Period 4:

On 02nd February 2015, nineteen subjects reported to the facility. At the time of check-in, eligibility was assessed by carrying out test for drugs of abuse, medical examination, confirming compliance to protocol based upon exclusion criteria and recording vitals. Breath of the subject was analyzed to check the consumption of alcohol using breath alcohol analyzer. Nineteen subjects who were found to be compliant with the requirements of the protocol were checked in Clinical Pharmacology Unit (CPU) between 18:05 hours to 20:00 hours.

During the in-house stay, monitoring of vital signs and subject questionnaire at the time of check-in, pre-dose and at 1.00, 3.00, 9.00 and 24.00 hours post-dose (Time points being relative to the investigational product dosing) was done. Medical examinations were carried out at the time of check-in and check-out of the study.

All the subjects checked-in into the study were housed in the Clinical Pharmacology Unit (CPU) until check-out on 04th February 2015.

The subjects were asked to visit the clinical facility for ambulatory blood sample collection at 34.00 and 48.00 hours post-dose. Breath of the subject was analyzed to check the consumption of alcohol using breath alcohol analyzer during each ambulatory visit. Vital signs and subject questionnaire was done at 34.00 and 48.00 hours post-dose and medical examinations were carried out at 48.00 hours post-dose.

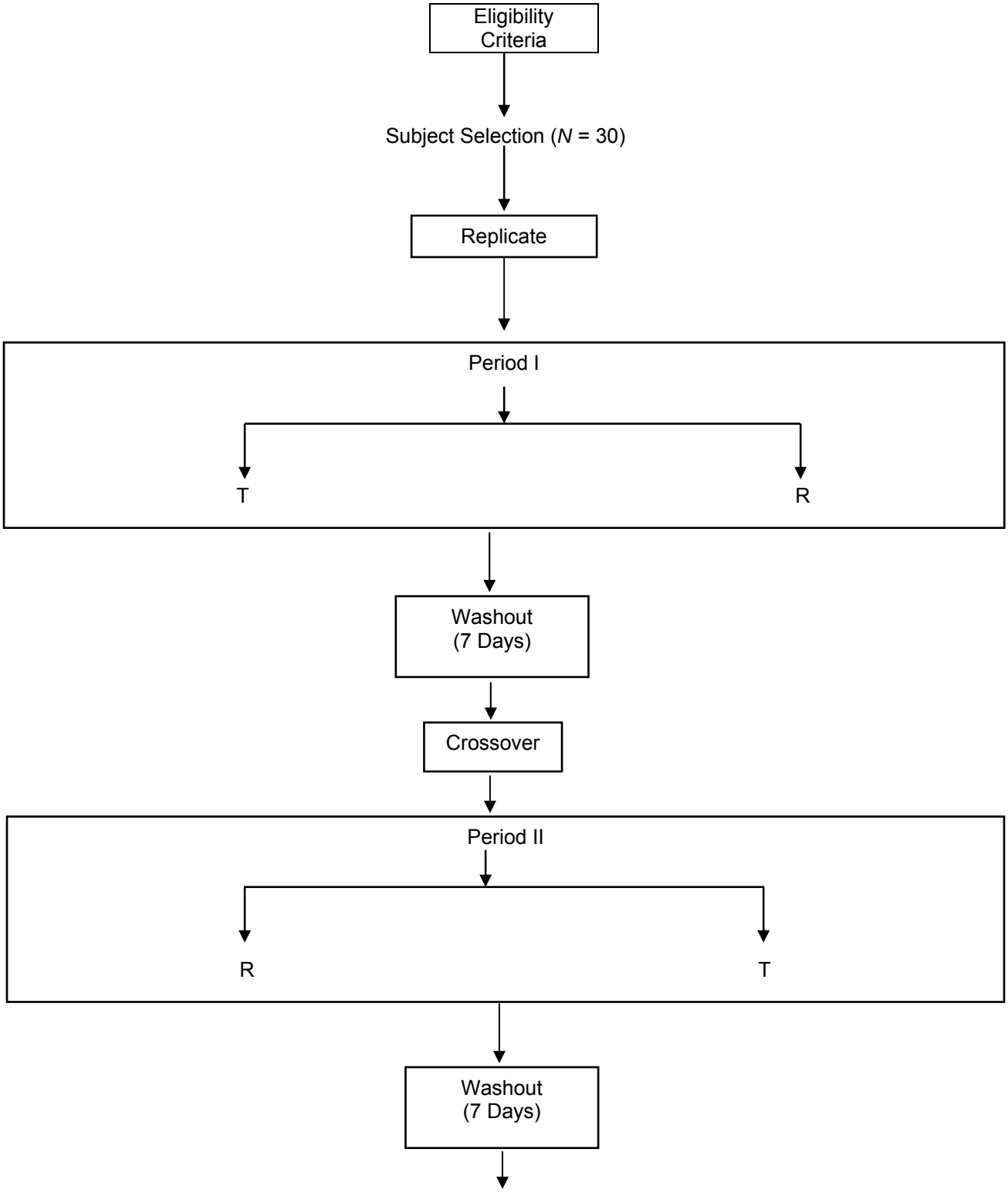
Washout Period:

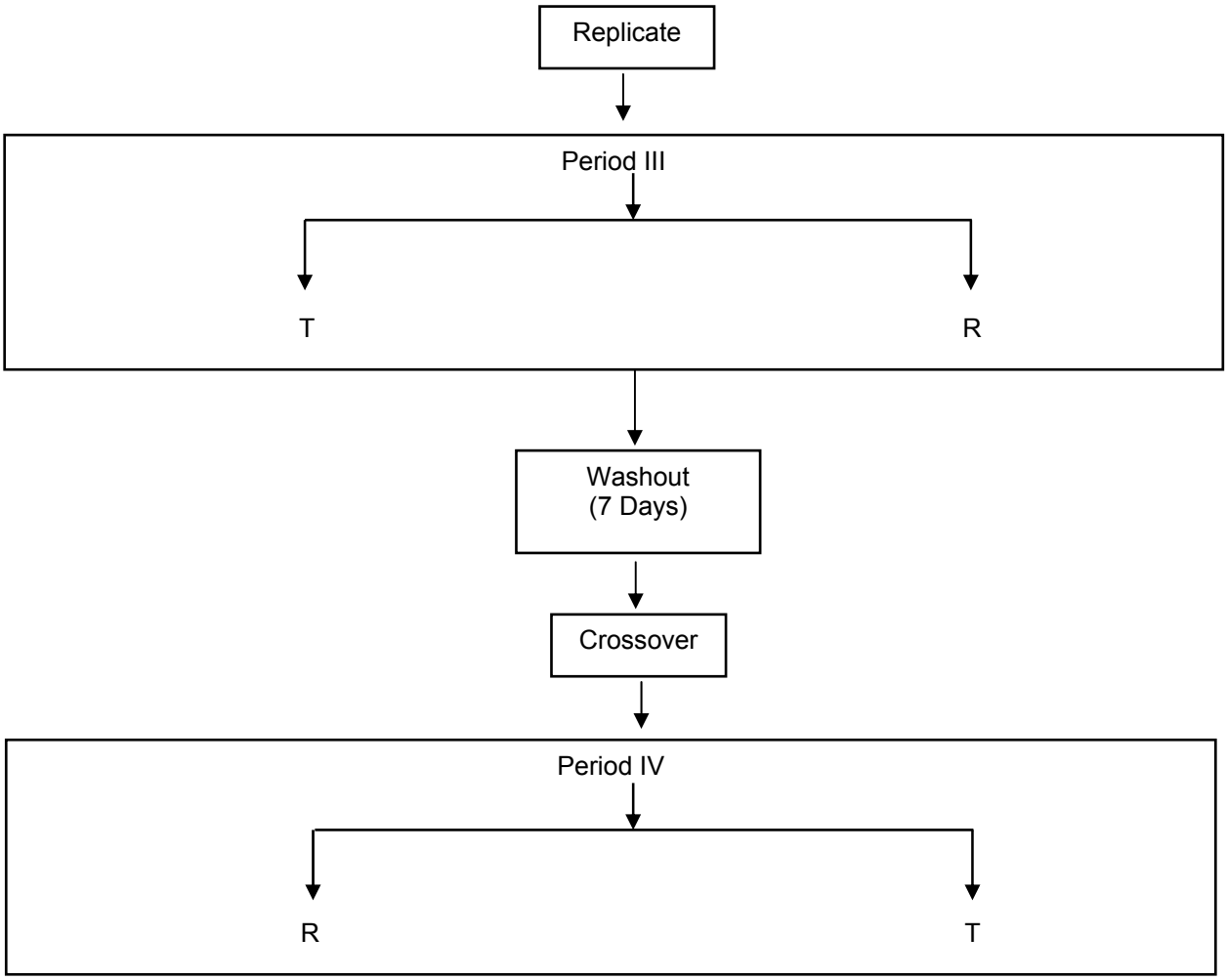
There was a washout period of 7 days from the completion of dosing between two consecutive periods.

The study schematic is given in figure-1 on next page.

FIGURE – 1 STUDY PLAN

STUDY FLOW CHART





N: Number of Subjects

R: Reference Product

T: Test Product.

9.2 Discussion of Study Design, Including the Choice of Control Group

It was an open labeled study because it was not possible to blind the appearance of the products. The analysts concerned, however, were blinded to the sequence of administration of test and reference product to the individual subjects.

The order of receiving treatment was randomized to avoid bias in allocation of sequence to the subjects.

There were two treatments: the sponsor's product was the test product while the innovator product was the reference product.

The subjects served as their own control, the study being crossover.

Since there were two treatments, the trial design was four periods, two sequences. The effect of period and sequence on primary efficacy criteria was analyzed by ANOVA (Analysis of variance).

To reduce variability in the biomedical experimentation and to control factors, which may affect the evaluation and comparison of primary efficacy factors; healthy, adult, human subjects were selected.

The number of subjects to be included in the study was derived based on previous bioequivalence study.

With the mean plasma elimination half-life of atorvastatin in humans is approximately 14 hours, the two dosing days i.e. for period 1, period 2, period 3 and period 4 were separated by a washout period of 7 days.

9.3 Selection of Study Population

The general screening was carried out after obtaining the written consent on IEC approved 'Informed Consent for Screening' from the volunteers. The screening procedure included Demographic data including sex, completed age, height and weight, Body Mass Index (BMI), diet, history of tobacco use, intake of abusive/recreational drugs, alcohol intake, history of blood donation and history of participation in a drug research study. Medical history, including relevant past medical / surgical history, family history, history of allergies (food / drug / any other), past medication history in the last 90 days. Medical examination including recording of vital signs (Blood Pressure (BP), Pulse, Respiration and Temperature), general examination, physical and systemic examination. 12-lead ECG for heart rate, rhythm and specific finding (if any). Chest X-ray (PA view); Laboratory parameter investigation including Complete blood count – Leucocyte count, Erythrocyte count, Haemoglobin, Hematocrit, Platelet count, differential leucocyte count (DLC); and ESR, Biochemistry – blood sugar (random), cholesterol and triglycerides, Alkaline Phosphatase, Hepatic Profile – SGPT, SGOT, GGT and Serum Bilirubin (Total, Direct, Indirect), Renal profile – serum creatinine, BUN, Serum Calcium, Serum Electrolytes (sodium, potassium. Chlorides) and Infectious Diseases – HbsAg, HIV and HCV and routine urine examination.

No clinically significant abnormalities in ECGs, Chest X-ray (PA view) were reported in subjects who were included in the study. Additionally, serological tests (HbsAg, HIV and HCV) were negative. The volunteers with laboratory values within normal limits or with clinically non-

significant values were called one day prior to the study for study informed consent form presentation.

All the baseline clinical and laboratory results are given in appendix 16.2.8 (Table A) and the individual clinical impression of ECG and chest X-ray (PA view) have been appended as appendix 16.2.8 (Table C). All the laboratory results, which were outside the Reference range but within the 'acceptable limit (For Acceptable limits refer Appendix V of protocol) were considered clinically non significant. There were few values as given below, which were outside the acceptable limits, however since the results were considered clinically non-significant based on clinical correlation, these subjects were included in the study.

Subject No.	Laboratory parameter	Results	Reference range
05	Triglycerides	217.5 mg/dL	< 150.0 mg/dL
06	Triglycerides	174.0 mg/dL	< 150.0 mg/dL
09	Bilirubin Total	1.17 mg/dL	0.20 – 1.00 mg/dL
	Bilirubin direct	0.26 mg/dL	0.00 – 0.20 mg/dL
13	Alkaline Phosphatase	160.3U/L	50.0 – 136.0 U/L
14	Bilirubin Total	1.13 mg/dL	0.20 – 1.00 mg/dL
15	SGPT	72.5 U/L	30.0 – 65.0 U/L
18	SGPT	76.8 U/L	30.0 – 65.0 U/L
21	Triglycerides	171.8 mg/dL	< 150.0 mg/dL
22	ESR	26 mm/hr	< 15 mm/hr
23	Triglycerides	166.3 mg/dL	< 150.0 mg/dL
24	Triglycerides	217.4 mg/dL	< 150.0 mg/dL
25	Triglycerides	175.5 mg/dL	< 150.0 mg/dL

Only those volunteers who signed the study informed consent form were checked in for the study on the day of check-in (one day prior to dosing).

All the volunteers who were checked in the study were found negative for breath alcohol test and urine test for drugs of abuse test [Cocaine (COC), Amphetamines (AMP), Marijuana (THC), Morphine (MOP), Barbiturates (BAR), and Benzodiazepine (BZO)].

Volunteers were given the rank orders based on their reporting time to the facility on pre-study day. Based on their rank orders and depending on the compliance to the requirements of the protocol, subject numbers were allotted serially. Thirty fit and consenting subjects fulfilling inclusion/ exclusion criteria and complying with the requirements of the protocol were enrolled in the study.

9.3.1 Inclusion Criteria

Subjects had to fulfill all of the following criteria to be considered for inclusion into this study:

1. Healthy volunteers within the age range of 18 to 45 years.
2. Presently non-tobacco users (smokers and chewers).

3. Willingness to provide written informed consent to participate in the study.
4. Body-mass index (BMI) between 18.50 kg/m^2 and 29.99 kg/m^2 (both inclusive) with body weight not less than 50 kg (for males) and with body weight not less than 45 kg (for females).
5. Absence of significant disease or clinically significant abnormal laboratory values or laboratory evaluation, medical history or physical examination during the screening.
6. Have a normal 12-lead ECG or one with abnormality considered to be clinically insignificant.
7. Have a normal chest X-ray PA view or one with abnormality considered to be clinically insignificant.
8. Comprehension of the nature and purpose of the study and compliance with the requirement of the distributed ICF.
9. Volunteer is regularly menstruating / Volunteer is in postmenopausal phase for at least 1 year / is surgically sterile (for females).
10. Volunteer of child bearing potential practicing an acceptable method of birth control for the duration of the study as judged by the investigator(s) such as condoms, foams, jellies, diaphragm, and intrauterine device (IUD) or abstinence etc. except hormonal contraceptives (for females).

9.3.2 Exclusion Criteria

The subjects were to be excluded based on the following criteria:

1. Personal history of allergy or hypersensitivity to Atorvastatin or allied drugs.
2. Any major illness in the past 90 days or any clinically significant ongoing chronic medical illness e.g. Congestive Cardiac Failure (Heart failure), Hepatitis, Hypotensive episodes, Hyperglycemia etc.
3. Presence of any clinically significant abnormal laboratory values during screening e.g. Significant abnormality of liver function test, renal (kidney) function test etc.
4. Severe cardiac, renal or liver impairment, gastro-intestinal disease or other conditions, any other organ or system impairment.
5. History of seizures, epilepsy or any kind of Neurological disorders.
6. Past history of Anaphylaxis or angioedema.
7. Presence of disease markers of HIV or Hepatitis B or Hepatitis C virus.
8. History of chronic consumption of any kind of alcoholic beverages for more than 2 years or having consumed alcohol within 48 hours prior to dosing.
9. Consumption of products containing xanthine derivatives (chocolates, tea, coffee or cola drinks) or tobacco products within 48 hours prior to dosing.
10. Consumption of grapefruit or grapefruit containing products or any cruciferous vegetables (eg. broccoli, brussels sprouts, etc.) or char-broiled meat prior 7 days of investigational product administration.
11. Use of any recreational drug or a history of drug addiction.
12. Participation in any clinical trial within the past 90 days.

13. History of difficulty with donating blood or difficulty in accessibility of veins in left or right arm.
14. Donation of blood (one unit or 350 mL) within 90 days prior to receiving the first dose of study medication.
15. Consumption of any other prescription drug or over the counter (OTC) drugs (including vitamins and medicinal products from natural origin) within two weeks prior to receiving the first dose of study medication or repeated use of drugs within the last four weeks.
16. An unusual diet for whatever reason e.g. low sodium diet, for two weeks prior to receiving any medication and throughout subject's participation in the study.
17. Recent history of dehydration from diarrhoea, vomiting or any other reason within a period of 48 hours prior to the study.
18. Use of oral contraceptive in last 90 days (for females).
19. Pregnant / lactating volunteers (for females).

9.3.3 Removal of Subjects from Therapy or Assessment

The subjects were free to withdraw from the study at any time without having to give any reasons thereof. The Principal Investigator, at his discretion, could also withdraw the subject from the study for any of the valid reason, which he deems to be appropriate in view of the safety and well being of subject, GCP principles or objectives of the project, in particular for:

1. If the subject suffers from significant illness.
2. If the subject requires concomitant medications which may interfere with pharmacokinetic of the study drug.
3. If the subject has entered the study in violation of the inclusion and the exclusion criteria.
4. If the subject is found to be non co-operative.
5. If the subject decides to voluntarily dropout from the study.

In any of these cases the compensation to the subject would have been made as per the guidelines regarding these i.e. ICH-GCP and National guidelines (ICMR guidelines for Clinical Trials) and as per the compensation structure approved by IEC.

Whatever reason if a subject would not satisfactorily complete the study, would be asked to attend for the post-study examination. Whenever possible, the post-withdrawal follow up would be done immediately after the subject is withdrawn. In case a subject is not willing to undergo such medical examination, it would be documented so.

9.4 Treatments

9.4.1 Treatments Administered

An oral dose of reference product (R) or test product (T) was administered at 0.00 hour during each period with 240 mL (about 8 oz) of drinking water at room temperature as per the randomization schedule under the supervision of the medical officer where end time of the dosing are recorded in investigational product administration forms. Subjects received the alternate 'treatment' in such a way that each subject completing the study received both the treatments test and reference twice at the end of the study.

Subjects were dosed while in sitting posture and were instructed to remain seated or be ambulatory (avoiding any strenuous activity) for first two hours following the investigational product administration (except during recording of vitals). During this interval, under supervision, subjects were permitted to use the washroom facilities. Thereafter the subjects were allowed to engage only in normal activities while avoiding severe physical exertion. However should any adverse event occur at any time during housing the subjects would have been placed in an appropriate posture.

9.4.2 Identity of Investigational Product(s):

i) Test Formulation (T): Atorvastatin Calcium Tablets 80 mg
Batch No.: BAC9402A
Manufacturing Date: June 2014
Expiry Date: May 2016
Manufactured by: Macleods Pharmaceuticals Ltd. India
Dose: 1 tablet
Mode of administration: Administered orally with 240 mL of drinking water.
Batch size: 1,25,000 Tablets
Assay: 102.1%

ii) Reference Formulation (R): Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg)
Lot No.: V122400
Manufacturing Date: N/AV
Expiry Date: Oct-15
Distributed by: Parke-Davis Division of Pfizer Inc., NY, NY 10017
NDC 0071-0158-23
Dose: 1 tablet
Mode of administration: Administered orally with 240 mL of drinking water.
Batch size: Not Available
Assay: 99.9%

9.4.3 Method of Assigning Subjects to Treatment Groups

The subjects were assigned to the sequence either test or reference product, according to the randomization schedule.

The order of receiving test or reference product for each subject during the study was determined according to randomization schedule (generated using SAS®).

Subject number was allocated as per the rank order of the reporting time of the subject to the clinical facility.

9.4.4 Selection of Doses in the Study

The available strengths of Lipitor® atorvastatin calcium tablets are 10 mg, 20 mg, 40 mg and 80 mg. The maximum recommended adult oral single dose of Lipitor® atorvastatin calcium tablets is 80 mg. Therefore single, oral dose of 1 tablet of Lipitor® 80 mg (atorvastatin calcium tablets 80 mg) is chosen, since it is safe (well tolerable) for healthy volunteers and was expected to provide measurable plasma concentrations and single dose study is adequate for this product's bioequivalence.

9.4.5 Selection and Timing of Dose for Each Subject

Dosing was done between 09:00 hrs to 09:42 hrs (0.00 hrs) in the batch of two subjects during all the periods. The dosing interval between successive subjects was 3 minutes.

The test or reference products were orally administered to the subjects while in sitting posture, with 240 mL (about 8 oz) of drinking water at room temperature as per the randomization schedule under the supervision of the Medical Officer followed by examination of the oral cavity.

The subjects fasted for at least 10 hours prior to administration of the study drug. Fasting was continued for 4.00 hours post-dose, and then meals were provided at specified intervals. Drinking water was disallowed for 1 hour pre-dose and 1 hour post-dose administration, except 240 mL of drinking water during administration of the drug dose. Subsequently, drinking water was provided ad libitum.

9.4.6 Blinding, Packaging and Labeling

This study comprised of a randomized, open label design. Study monitors and subjects involved in the study were not blinded. However the analyst concerned, were blinded to the sequence of administration of test or reference products to the individual subject. The plasma sample storage vials were labeled; mentioning study number, period number, subject number, sample number, time point (hrs) and aliquot number, but the identity of product administered was not mentioned. The randomization schedule was in the custody of principal investigator and the investigational product dispensing raw data record was under lock and key until the completion of statistical analysis.

An adequate number of investigational products (IPs) in sealed condition along with certificate of analysis (COA) were received at Bioequivalence department of Macleods by the registered pharmacist.

Dispensing:

As per the randomization schedule, the registered pharmacist prepared the doses under the supervision of trained personnel and in the presence of Quality Assurance personnel in all the periods. Remaining investigational products were stored in their original container as retention samples in stability chamber at 22 ± 2°C and 60 ± 5% RH.

The dispensed investigational products were transferred to the drug-dispensing containers as unit doses. The drug-dispensing containers used for dispensing were properly labeled for the study

number, subject number, treatment ID, period number, Batch/Lot number, dosage form and expiry date, number of unit dispensed, route of administration, storage condition and initial and date of the person dispensing the product. The drug dispensing containers along with duplicate label (similar to that stuck on the dispensing container) were placed in zip lock bag.

Investigational product accountability included the records of the receipts, intake of investigational products during all the periods of the study and remaining quantities of investigational products.

The investigational products were stored in stability chamber at 22 ± 2°C and 60 ± 5% RH and inventory were maintained in the logbook of investigational product. Data for temperature and humidity was monitored and recorded regularly through data logger.

Only authorized personnel has access to the investigational product storage areas.

For certificate of analysis of test and reference products, refer appendix – 16.1.12.

9.4.7 Prior and Concomitant Therapy

Receipt of any other prescription drug or over the counter (OTC) drugs (including vitamins and medicinal products from natural origin) within two weeks prior to receiving the first dose of study medication or repeated use of drugs within the last four weeks was an exclusion criterion. Further, the subjects were not supposed to consume any medication during the conduct of the study. All subjects, who checked-in the study, confirmed that they did not consume any medication within the 2 weeks of the start of first period or during the study.

9.4.8 Treatment Compliance

Subjects were provided with the identity card (I-card). While dosing, the staff on duty confirmed the subjects identity with I-card and the information mentioned on the investigational product administration form.

After administration of the dose of investigational product, examination of the oral cavity was performed under supervision of medical officer to assess the compliance to this procedure and the same was noted in the investigational product administration form of each subject. An additional label was affixed to the investigational product administration form in the appropriate place to confirm correct administration of IP. The time of actual dose administration was recorded in the investigational product administration form.

Investigational product accountability record and evaluation of the plasma drug concentration of the samples confirmed 100% compliance of all the subjects from whom the data was analyzed.

9.5 Efficacy and Safety Variables

9.5.1 Efficacy and Safety Measurements Assessed

Efficacy Measurements Assessed

Efficacy Measurements Assessed

The following pharmacokinetic parameters (variables) of atorvastatin, o-hydroxy atorvastatin and p-hydroxy atorvastatin were estimated after drug administration under fasting conditions:

Primary Efficacy Variables

$C_{max,}$ AUC_{0-t} and $AUC_{0-\infty}$

Secondary Efficacy Variables

$T_{1/2,}$$K_{el}$, $T_{max,}$ npoints, K_e_first & K_e_last

These parameters were derived individually for each subject from their atorvastatin, o-hydroxy atorvastatin and p-hydroxy atorvastatin concentration in plasma. Actual time of blood collection was considered for pharmacokinetic calculations.

For estimation of PK parameters, concentrations that were below level of quantification (BLQ) were assigned a value of zero if they preceded quantifiable samples in the initial portion of the profile. A BLQ that occurred at the end of the profile was set to zero. A BLQ or zero concentration that was embedded between two quantifiable points was assigned a value of missing. If consecutive BLQs in the terminal portion of the profile were followed by quantifiable determinations, these quantified values were excluded from PK analysis by assigning them a value of missing. In the calculations of PK parameters, missing values was ignored. Plasma concentrations used to determine PK parameters would be listed.

The pharmacokinetic parameters were calculated by non-compartmental methods using SAS® version 9.4.

The calculations of the individual pharmacokinetic parameters were carried out as follows:

C_{max} : Maximum measured plasma concentration following each treatment.

AUC_{0-t} : The area under the plasma concentration versus time curve from time zero to the last measurable concentration, as calculated by the linear trapezoidal method.

$AUC_{0-\infty}$: The area under the plasma concentration versus time curve, from zero to infinity.

$AUC_{0-\infty}$ is calculated as the sum of the AUC_{0-t} plus the ratio of the last measurable concentration to the elimination rate constant.

T_{max} : Time of maximum measured plasma concentration. If maximum value occurs at more than one point, T_{max} is defined as the first point with this value in each period.

K_{el} : Apparent first order elimination or terminal rate constant calculated from semi log plot of the plasma concentration versus time curve. The parameters were calculated by linear least square regression analysis using at least the last three non-zero plasma concentration.

$T_{1/2}$: Time required for the plasma drug concentration to decrease to one half.

npoints : No. of time points of the terminal log-linear phase used to estimate the terminal disposition rate constant (K_e) with bestfit regression method used.

K_e_first : First time point of the terminal log-linear phase used to estimate the terminal disposition rate constant (K_e) with bestfit regression method used.

K_e_last : Last time point of the terminal log-linear phase used to estimate the terminal disposition rate constant (K_e) with bestfit regression method used.

Note:

- In case the pre-dose concentration obtained is greater than 5 percent of C_{max}, the subject was to be dropped from bioequivalence study evaluations.
- In case the subject experience emesis during the course of study, the data was to be deleted from statistical analysis, if vomiting occurs within 2 times median $T_{max.}$

Safety Measurements Assessed

Safety was evaluated by monitoring clinical adverse events during study periods. Vital signs (Blood Pressure, Temperature and Pulse Rate) and subject questionnaire was done at the time of check-in, pre-dose and at 1.00, 3.00, 9.00, 24.00, 34.00 and 48.00 hours post-dose (Time points being relative to the investigational product dosing). Medical examinations were carried out at the time of check-in, checkout, and at 48.00 hours post-dose of all the periods.

Subjects were monitored for adverse events, if any, throughout the course of the study by asking them if they were feeling fine or had any discomfort, at the time of clinical examination and recording of vital signs and recording the same in their respective CRF. The following evaluations were done at screening and in the follow-up phase:

- Medical examination (vital signs, 12 lead ECG and general and systemic examination).
- Clinical laboratory tests (hematology, clinical biochemistry) and urine analysis (only during pre study screening).

9.5.2 Appropriateness of Measurements

The plasma samples of subjects were analyzed by a validated LC-MS/MS method. The limit of quantification of 502.4 pg/mL for atorvastatin, 504.0 pg/mL for ortho- hydroxylated and 100.3 pg/mL for parahydroxylated metabolites of atorvastatin was enough to quantify the analyte from the plasma samples collected up to 48.00 hours after drug administration. The linearity range of 502.4 pg /mL to 200013.3 pg/mL for atorvastatin, 504.0 pg/mL to 199862.3 pg/mL for ortho-hydroxylated and 100.3 pg/mL to 30013.8 pg/mL for parahydroxylated metabolites of atorvastatin was enough to quantify the expected concentration range of atorvastatin and ortho- and parahydroxylated metabolites of atorvastatin from subject plasma with the proposed dose of 80 mg of atorvastatin calcium.

9.5.3 Primary Efficacy Variable(s)

The following pharmacokinetic parameters were assessed as primary efficacy variables,

C_{max}: Maximum measured plasma concentration following each treatment.

AUC_{0-t}: The area under the plasma concentration versus time curve from time zero to the last measurable concentration, as calculated by the linear trapezoidal method.

$AUC_{0-\infty}$: The area under the plasma concentration versus time curve, from zero to infinity. $AUC_{0-\infty}$ is calculated as the sum of the AUC_{0-t} plus the ratio of the last measurable concentration to the elimination rate constant.

9.5.4 Drug Concentration Measurements

Concentration of atorvastatin, ortho- and parahydroxylated metabolites of atorvastatin was measured in plasma samples of the subjects. Blood samples (1 × 5 mL) were collected in 5 mL blood collection tube containing K_2EDTA as anticoagulant during all the periods. The venous blood samples were withdrawn pre-dose and at 0.17, 0.33, 0.50, 0.75, 1.00, 1.25, 1.50, 1.75, 2.00, 2.33, 2.67, 3.00, 3.50, 4.00, 4.50, 5.00, 5.50, 6.00, 8.00, 12.00, 16.00, 24.00, 34.00 and 48.00 hours post dose (time points being relative to the investigational product dosing).

Note: 5 mL additional blood sample was collected at pre-dose in all the periods.

Post-dose samples up to 24.00 hrs collected through an indwelling cannula placed in a forearm vein. The pre-dose samples were collected within one hour prior to investigational product dosing. The post-dose samples up to 24.00 hours in house stay were collected within 2 minutes of the scheduled time where the end time of collection to the nearest minute was recorded. Similarly, the post-dose samples during ambulatory visit were collected within one hour of the scheduled time where the end time of collection to the nearest minute was recorded. However there were few deviations in this regard. Refer section 10.2 'Protocol Deviation'.

During ambulatory visit, the blood sample was collected -1.00 hour to + 2.00 hours of specified schedule time of blood collection. No ambulatory blood sample collection was made after 2 hours of specified schedule time of blood collection for any subject.

The breath of the subjects was checked to see whether they have consumed alcohol during ambulatory visit of the study using breath alcohol analyzer. If subjects found breath alcohol test positive, further blood sample was not taken for respective period.

Intravenous indwelling cannula was kept in place as long as required by injecting not more than 0.5 mL of normal saline solution during the collection of multiple samples. In such a case, the blood sample was collected after discarding the first 0.5 mL of normal saline blood from the tubing. Blood would also be withdrawn from vein by using disposable syringe and needle if the cannula was blocked /removed or the cannula is removed for other reasons.

Each blood sample (1 × 5 mL) were collected into 5 mL blood collection tube containing K_2EDTA as anticoagulant. The blood samples collected at each time point were centrifuged between 4 to 8°C and at 4000 rpm for 10 minutes to separate plasma, after receiving the blood samples from all the subjects (For ambulatory samples, the samples collected till the scheduled time of last subject were centrifuged together and the samples collected later were centrifuged separately according to their collection time).

Post blood collection the samples were kept cool by placing the samples in ice water slurry and after centrifugation samples were separated in the ice water slurry and kept in ice water slurry until storage in deep freezer. The separated plasma was aliquoted in duplicate in prelabelled polypropylene tubes during all the periods. 2 mL of plasma sample was transferred in the first aliquot and remaining plasma sample was transferred in the second aliquot. These tubes were labelled with study number, period number, subject number, sample number, time point (hrs) and aliquot number.

These tubes were then transferred to a deep freezer maintained below –50°C or colder for storage and further analysed by bio analytical section.

The investigational products were administered in fasting conditions and no food was served till 4.00 hours post-dose. No fluid, except 240 mL drinking water administered with the investigational products was allowed from 1 hour pre-dose and 1 hour post dose. Subjects were dosed while in sitting posture and were instructed to remain seated or be ambulatory (avoiding any strenuous activity) for first two hours following the investigational product administration (except during recording of vitals). During this interval, under supervision, subjects were permitted to use the washroom facilities. Thereafter the subjects were allowed to engage only in normal activities while avoiding severe physical exertion. However should any adverse event occur at any time during housing the subjects would have been placed in an appropriate posture.

Subjects were instructed to abstain from alcohol and products containing xanthine derivatives (chocolates, tea, coffee or cola drinks) and tobacco products, for at least 48 hours, prior to dosing, for all the periods and during their participation in the study including ambulatory visit.

Subjects were instructed to abstain from grapefruit or grapefruit containing products or any cruciferous vegetables (eg. broccoli, brussels sprouts, etc.) or char-broiled meat prior 7 days of investigational product administration and during their participation in the study including ambulatory visit.

Subjects were instructed to abstain from an unusual diet for whatever reason e.g. low sodium diet, for two weeks prior to receiving the first investigational product and throughout their participation in the study including ambulatory visit.

Validated LC-MS/MS method was employed for the estimation of atorvastatin, ortho- and parahydroxylated metabolites of atorvastatin in plasma. During estimation of atorvastatin, ortho- and parahydroxylated metabolites of atorvastatin in plasma quality control samples were distributed throughout each batch of study samples.

During extraction process and through out the analysis, the samples were maintained in cold condition by placing the sample containers in ice water slurry.

Whenever possible, samples from each subject were analyzed on the same standard curve. Samples with drug concentration greater than upper limit of the validated range of the analysis would be reanalyzed as per the standard test procedure based on method validation report.

The analysts concerned were blinded with respect to the randomization code, and as a result to the order of administration of the study medication.

9.6 Data Quality Assurance

The quality control personnel performed the quality control check of the case report forms and of all source documentation.

The Quality Assurance department of Macleods Pharmaceuticals Bioequivalence Department conducted both in process and retrospective audits of Clinical, Bioanalytical and Pharmacokinetic and Statistical phase of the study. The audits were conducted as per in-house standard operating

procedure as appended in appendix 16.1.10. The findings in brief were reported to the management.

The Quality Assurance statement is appended as appendix 16.1.8.

9.7 Statistical Methods Planned in the Protocol and Determination of Sample Size

9.7.1 Statistical and Analytical Plans

Following were the plans for statistical analysis:

- Use descriptive statistics (number of subjects, mean, SD, CV, minimum and maximum) to summarize the plasma concentrations at each time of measurement.
- For purpose of descriptive linear and semi-logarithmic plots of the mean and individual plasma concentration by scheduled sampling time provided.
- Report missing samples or unreportable concentration values as 'missing/ not reported' and document the reason for the same.
- Actual time of blood collection was considered for pharmacokinetic calculations.
- Use SAS® system version 9.4 for estimation of pharmacokinetic parameters and its statistical analysis for atorvastatin, o-hydroxy atorvastatin and p-hydroxy atorvastatin from their plasma concentration data.
- Report the summary statistics for all pharmacokinetic parameters for both the test and reference products. The reported parameters are the minimum, maximum, arithmetic means, median, standard deviation and the coefficient of variation for untransformed data.
- Calculate ratio of geometric mean of test to geometric mean of reference for each subject for all relevant pharmacokinetic parameters (C_{max}, AUC_{0-t} and $AUC_{0-\infty}$).
- Report ratio of mean AUC_{0-t} to mean $AUC_{0-\infty}$ for test and reference in percentage.
- Report ratio of AUC_{0-t} to $AUC_{0-\infty}$ for each subject for both test and reference are expressed in percentage.
- Report % bioavailability of $C_{max,}$ AUC_{0-t} and $AUC_{0-\infty}$
- Method for Statistical Analysis Using the Reference-Scaled Average Bioequivalence Mixed Scaling Approach
- If $S_{WR} \geq 0.294$, i.e. intra-subject C.V. for reference formulation $\geq$ 30% then reference-scaled average bioequivalence approach is used to determine bioequivalence (BE) for the relevant pharmacokinetic parameters (C_{max}, AUC_{0-t} and $AUC_{0-\infty}$).
- If $S_{WR} < 0.294$, i.e. intra-subject C.V. for reference formulation < 30% then the two-one sided test (Unscaled average bioequivalence approach) is used to determine BE for the relevant pharmacokinetic parameters (C_{max}, AUC_{0-t} and $AUC_{0-\infty}$), Where S_{WR} is the within-subject standard deviation (SD) of the reference formulation.
- Reference-Scaled Average Bioequivalence Approach:

- Analyze intermediate analysis for log-transformed pharmacokinetic parameters ICmax, $IAUC_{0-t}$ and $IAUC_{0-\infty}$ of atorvastatin using an ANOVA model with sequence. “PROC GLM” procedure is used to obtain the ANOVA.
- Analyze intermediate analysis for log-transformed pharmacokinetic parameters of DC_{max}, $DAUC_{0-t}$ and $DAUC_{0-\infty}$ of atorvastatin using an ANOVA model with sequence. “PROC GLM” procedure is used to obtain the ANOVA.
- Use a separate ANOVA model to analyze each of the parameters.
- Use SAS procedure ‘PROC GLM’ to perform analysis of variance.
- Calculate the intra subject coefficient variation for reference using 100 x $\sqrt{\left(e^{S_{WR}^2}-1\right)}$ with help of SAS® version 9.4. Where S_{WR}^2 Within subject variance obtained from Analysis of Variance model.
- Calculate Point estimate (Ratio T/R) and 95% upper bound for ($\left(\overline{Y}_T-\overline{Y}_R\right)^2-\theta * S_{WR}^2$).
- Claim the treatment to be bioequivalent If the 95% Upper Bounds for ($\left(\overline{Y}_T-\overline{Y}_R\right)^2-\theta * S_{WR}^2$) are ≤ 0 and the point estimate (Ratio T/R) of atorvastatin are entirely included in the range of 80.00 – 125.00% for C_{max}, AUC_{0-t} and $AUC_{0-\infty}$.
- Test factors effects by analyze the log-transformed pharmacokinetic parameters (C_{max}, AUC_{0-t} and $AUC_{0-\infty}$) of atorvastatin using an ANOVA model with fixed effect of sequence, period and formulation and random effect of subjects nested within sequence.
- Use a separate ANOVA model to analyze each of the parameters.
- Use a 10% level of significance to test significance of sequence effects.
- Use a 5% level of significance to test significance of formulation & period effects.
- Unscaled Average Bioequivalence Approach:
- Analyze the log-transformed pharmacokinetic parameters (C_{max}, AUC_{0-t} and $AUC_{0-\infty}$) of atorvastatin using an ANOVA model with fixed effect of sequence, period and formulation and random effect of subjects nested within sequence.
- Use a separate ANOVA model to analyze each of the parameters.
- Use a 10% level of significance to test significance of sequence effects.
- Use a 5% level of significance to test significance of formulation & period effects.
- Each analysis of variance including calculation of mean square error, p-value of factors and the associated degrees of freedom.
- Use SAS procedure ‘PROC MIXED’ to perform analysis of variance.
- Calculate and report ratio of geometric means using the LSM for log transformed C_{max}, AUC_{0-t} and $AUC_{0-\infty}$.
- Report the geometric means of the test and reference product
- Calculate the power of the ANOVA model to detect the ratio of the two products averages (geometric means) being equal to 125% (or 80%) at the 5 % significance level for analyses using the log-transformed data. It is calculated using the following formula:

$$\text{Power} = 100 * P\left[X > \left(\frac{\ln(1.25)}{se} - \left| t_{0.05,DF} \right| \right) \right]$$

Where, se is standard error and DF is obtained from Analysis of Variance model.

- Calculate the Intra-subject coefficient of variance for test, reference and global using 100 x $\sqrt{e^{MSE} - 1}$ with help of SAS® version 9.4. Where MSE is mean squared error obtained from Analysis of Variance model.
- Calculate a 90% confidence interval for the ratio of both the products averages (geometric means) by first calculating the 90% confidence interval for the differences in the averages (least square means) of the log-transformed data and then taking the antilogarithms of the obtained confidence limits.
- Claim the treatment to be bioequivalent if the confidence intervals of atorvastatin are entirely included in the range of 80.00% – 125.00% for log-transformed AUC_{0-t}, $AUC_{0-\infty}$ and C_{max}.
- For o-hydroxy atorvastatin and p-hydroxy atorvastatin, the following data should be submitted: Individual and mean concentrations, individual and mean pharmacokinetic parameters, and geometric means and ratios of means for AUC_{0-t}, $AUC_{0-\infty}$ and C_{max}. The measurement of o-hydroxy atorvastatin and p-hydroxy atorvastatin metabolite levels in plasma were not used for comparison of bioequivalence.

9.7.2 Determination of Sample Size

Sample size is calculated using SAS®. As estimated from previous bioequivalence study (BEQ-1048-ATOR-2012) of single dose of Atorvastatin Calcium Tablets 80 mg under fasting condition. The pharmacokinetic parameters and statistical results for Atorvastatin are as follows:

Pharmacokinetic Parameters	Intra subject C.V. (%)			Ratio (T/R) (%)	90 % C.I. (%)	
	ISVR	ISVT	ISVGLB		Lower	Upper
C_{max} (ng/mL)	29.36	30.21	29.79	96.44	78.49	118.75
AUC_{0-t} (ng*hr/mL)	26.89	24.47	25.71	94.04	83.69	107.75
$AUC_{0-\infty}$ (ng*hr/mL)	25.17	22.23	23.74	94.49	84.78	107.31

Sample size is calculated using SAS®. The highest intra subject C.V. for global of Atorvastatin was observed to be 29.79%. Hence for this intra subject C.V. the sample size calculation is as follows:

Two-Sample Equivalence

Multiplicative Model Lower Bound = 0.80 Upper Bound = 1.25

Coefficient of Variation = 0.2979 Alpha = 0.05

Null Ratio	Power	N per Group
0.95	0.800	38
	0.850	43
	0.900	51
	0.950	64
1.00	0.800	30
	0.850	34
	0.900	38
	0.950	46
1.05	0.800	37
	0.850	42
	0.900	49
	0.950	62

The highest intra subject C.V. for Atorvastatin was observed to be 29.79% for C_{max} (ng/mL) in the previous bioequivalence study. So to achieve 80% power a sample size of 19 i.e.38 observation per formulation was concluded sufficient to conclude bioequivalence. Thus accounting for dropout or withdrawal of subjects during conduct of the study, 30 healthy subjects were decided to be recruited in a fully replicated crossover study design to achieve the desired sample size to conclude bioequivalence.

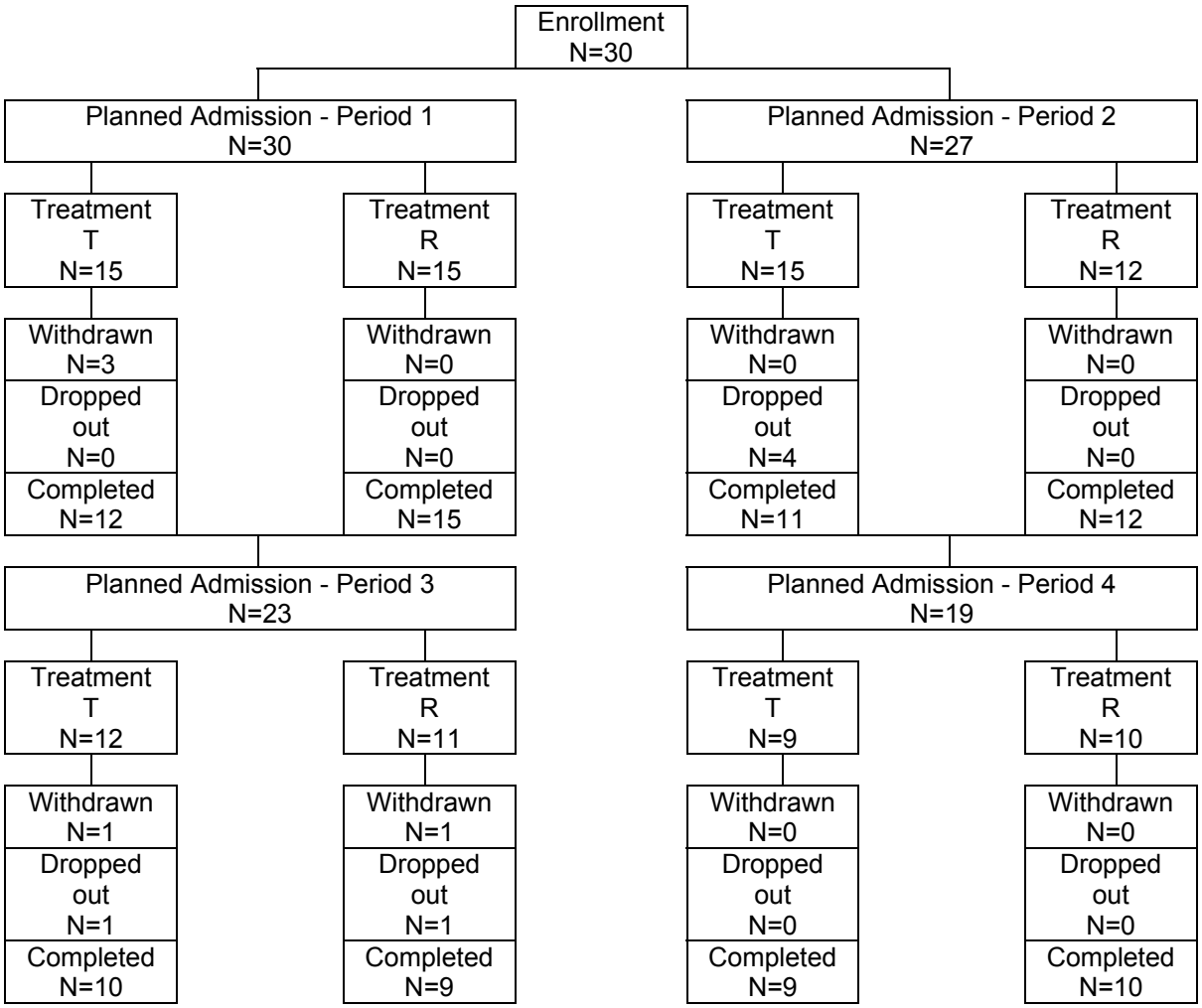
9.8 Changes in the Conduct of the Study or Planned Analyses

There were no changes in the conduct of the study or planned analyses.

10.0 STUDY SUBJECTS

10.1 Disposition of Subjects

A total of 30 subjects were planned and enrolled, of these 30, subject number 24 was withdrawn from the study in period 1 (post-dose) on principal investigator advice due to serious adverse events. Subject number 06 and 25 were withdrawn from the study in period 1 (post-dose) on principal investigator advice due to adverse events. Subject number 02, 12, 26 and 29 did not report to the facility due to personal reason for period 2 thus considered dropped-out from the study. Subject number 14 and 18 did not report to the facility due to personal reason for period 3 thus considered dropped-out from the study. Subject number 11 was found to participate in other clinical trial and thus was withdrawn from the study based on exclusion criteria during check-in of period 3 (pre-dose). Subject number 03 was withdrawn from the study in period 3 (post-dose) on principal investigator advice due to adverse event. Thus nineteen subjects completed all the periods of the study.



10.2 Protocol Deviations

- As per protocol blood samples should be collected within two minutes of scheduled time for blood collection for in-house samples and within one hour for all ambulatory visit samples. All actual times of the sample withdrawal were recorded in the bleed sheet. However; there were 6 deviations in period 1, 4 deviations in period 2 and 6 deviations in period 4 from the schedule time of the collection above the permitted deviation time in the study.

Sr. No.	Subject No.	Sample Time Point(hrs)	Scheduled Time	Actual Time	Deviation in Hour	Reason for Deviation
Period 1						
1.	08	12.00	21:21 hrs	21:25 hrs	0.07	Cannula block
2.	09	4.50	13.:54 hrs	13:57 hrs	0.05	Poor blood flow
3.	22	0.33	09:38 hrs	09:42 hrs	0.07	Cannula block
4.	28	34.00	19:36 hrs	20:50 hrs	1.23	Subject reported late
5.	29	34.00	19:39 hrs	20:53 hrs	1.23	Subject reported late
6.	05	48.00	09:12 hrs	10:40 hrs	1.47	Subject reported late
Period 2						
1.	01	2.33	11:20 hrs	11:24 hrs	0.07	Cannula block
2.		3.00	12:00 hrs	12:03 hrs	0.05	Poor blood flow

Sr. No.	Subject No.	Sample Time Point(hrs)	Scheduled Time	Actual Time	Deviation in Hour	Reason for Deviation
3.		3.50	12:30 hrs	12:34 hrs	0.07	Poor blood flow
4.		5.00	14:00 hrs	14:04 hrs	0.07	Poor blood flow
Period 4						
1	04	0.50	09:39 hrs	09:42 hrs	0.05	Poor blood flow
2	07	0.33	09:38 hrs	09:41 hrs	0.05	Poor blood flow
3	16	1.25	10:15 hrs	10:18 hrs	0.05	Poor blood flow
4	22	2.00	11:18 hrs	11:22 hrs	0.07	Poor blood flow
5		5.50	14:48 hrs	14:52 hrs	0.07	Poor blood flow
6	23	34.00	19:21 hrs	21:09 hrs	1.80	Subject reported late

- As per protocol total 25 samples should be collected per subject in each period. However there was 1 deviation in period 1 and 2 deviations in period 4 in this regard. Samples were not collected since the subjects didn't report to the facility except subject number 02 (period 1, 48.00 hrs), who reported after 2 hrs from the scheduled time of blood collection.

Sr. No.	Time Point (Hrs)	Subject No.			
		Period 1	Period 2	Period 3	Period 4
1.	34.00	-	-	-	17
2.	48.00	02	-	-	17

The above deviations were duly incorporated during pharmacokinetic analysis.

The above deviations were recorded as protocol deviation and are recorded in appendix 16.2.2.

11.0 EFFICACY EVALUATION

11.1 Data Sets Analyzed

The plasma samples of nineteen subjects (subject number 01, 04, 05, 07 to 10, 13, 15, 16, 17, 19 to 23 , 27, 28 and 30) completing the study was analyzed for atorvastatin and for ortho- and parahydroxylated metabolites of atorvastatin concentration level and the data were taken for pharmacokinetic and statistical evaluations.

The plasma samples of subject number 02, 03, 06, 11, 12, 14, 18, 24 to 26 and 29 who were considered dropped-out from the study were not analyzed.

11.2 Demographic and Other Baseline Characteristics

The demographic characteristics of the 30 subjects recruited in the study were as follows:

- Age between 20 and 37 years [28.9 (mean) ± 4.83 (SD) years].
- Height between 1.56 and 1.85 meters [1.676 (mean) ± 0.0577 (SD) meters].
- Weight between 52.2 and 81.0 kg [65.46 (mean) ± 7.978 (SD) kg].
- BMI between 18.96 and 27.14kg/m^2 [23.289 (mean) ± 2.5177 (SD) kg/m^2].

The demographic characteristics of the 19 subjects completed the study were as follows:

- Age between 24 and 37 years [30.4 (mean) ± 4.42 (SD) years].
- Height between 1.56 and 1.74 meters [1.666 (mean) ± 0.0534 (SD) meters].
- Weight between 52.2and 81.0 kg [64.02 (mean) ± 8.761 (SD) kg].
- BMI between 18.96 and 27.06 kg/m^2 [23.037 (mean) ± 2.6020 (SD) kg/m^2].

The demographic data is summarized in section 14.1. The demographic data for individual subjects are appended in appendix 16.2.4.

11.3 Measurements of Treatment Compliance

All the subjects took the medications as administered. Examination of the oral cavity immediately after drug administration was performed under supervision of medical officer to assess the compliance to this procedure. Further, the evaluation of the plasma drug concentration of the samples confirmed 100% compliance of all the subjects from whom the data were analyzed.

Plasma levels of atorvastatin, ortho- and parahydroxylated metabolites of atorvastatin in individual subjects at different time points following reference and test formulations are given in appendix 16.2.5.

11.4 Efficacy Results and Tabulations of Individual Subject Data

11.4.1 Analysis of Efficacy

The various un-transformed mean pharmacokinetic parameters estimated for both the reference and test formulations of atorvastatin under fasting conditions are as follows:

Pharmacokinetic Parameters	Test Product					
	N	Arithmetic Mean	S.D.	C.V. (%)	Median	Range
C_{max} (pg/mL)	38	90797.99	46727.300	51.46	82834.1	27004.3 - 244801.1
AUC_{0-t} (pg*hrs/mL)	38	447481.74842	173455.988200	38.76	446834.2938	152971.7360 - 860786.5575
$AUC_{0-\infty}$ (pg*hrs/mL)	38	455621.34846	173650.129440	38.11	452570.4024	157688.6395 - 872271.5131
T_{max} (hrs)	38	1.664	1.4240	85.58	0.89	0.50 - 4.52
$T_{1/2}$ (hrs)	38	6.02061	1.986616	33.00	5.4464	3.9957 - 14.0398
K_{el} (hr^{-1})	38	0.12360	0.028419	22.99	0.1273	0.0494 - 0.1735

Pharmacokinetic Parameters	Reference Product					
	N	Arithmetic Mean	S.D.	C.V. (%)	Median	Range
C_{max} (pg/mL)	38	89331.23	42076.832	47.10	82982.3	25874.3 - 228846.1
AUC_{0-t} (pg*hrs/mL)	38	467782.88230	167845.522970	35.88	479497.1110	138286.8055 - 817817.5720
$AUC_{0-\infty}$ (pg*hrs/mL)	38	475087.48443	169462.061370	35.67	483270.0687	146199.8011 - 831203.3702
T_{max} (hrs)	38	1.805	1.3012	72.08	1.50	0.50 - 4.52
$T_{1/2}$ (hrs)	38	5.42718	1.654193	30.48	5.1556	3.3627 - 12.0963
K_{el} (hr^{-1})	38	0.13630	0.031547	23.14	0.1344	0.0573 - 0.2061

The various un-transformed mean pharmacokinetic parameters estimated for both the reference and test formulations of o-hydroxy atorvastatin under fasting conditions are as follows:

Pharmacokinetic Parameters	Test Product					
	N	Arithmetic Mean	S.D.	C.V. (%)	Median	Range
C_{max} (pg/mL)	38	51122.06	23399.162	45.77	49915.5	20911.9 - 124837.9
AUC_{0-t} (pg*hrs/mL)	38	438931.58080	176413.463600	40.19	408546.2918	192249.1945 - 844266.2250
$AUC_{0-\infty}$ (pg*hrs/mL)	38	449999.61838	178433.189500	39.65	418207.3212	205468.1162 - 886116.0995
T_{max} (hrs)	38	2.413	1.5532	64.35	1.88	0.75 - 4.52
$T_{1/2}$ (hrs)	38	7.85044	2.558319	32.59	7.1584	5.0195 - 16.0319
K_{el} (hr^{-1})	38	0.09521	0.023135	24.30	0.0968	0.0432 - 0.1381

Pharmacokinetic Parameters	Reference Product					
	N	Arithmetic Mean	S.D.	C.V. (%)	Median	Range
C_{max} (pg/mL)	38	59137.24	32050.126	54.20	53698.6	21841.0 - 177371.9
AUC_{0-t} (pg*hrs/mL)	38	476579.32750	185150.003210	38.85	445629.2118	206503.6020 - 908514.0960
$AUC_{0-\infty}$ (pg*hrs/mL)	38	489182.85665	186627.402680	38.15	464184.4774	217690.8412 - 915379.4350
T_{max} (hrs)	38	2.752	1.3275	48.23	2.50	0.75 - 4.52
$T_{1/2}$ (hrs)	38	7.06886	2.127328	30.09	6.5152	4.1924 - 13.6523
K_{el} (hr^{-1})	38	0.10533	0.026116	24.79	0.1064	0.0508 - 0.1653

The various un-transformed mean pharmacokinetic parameters estimated for both the reference and test formulations of p-hydroxy atorvastatin under fasting conditions are as follows:

Pharmacokinetic Parameters	Test Product					
	N	Arithmetic Mean	S.D.	C.V. (%)	Median	Range
C_{max} (pg/mL)	38	7913.41	4116.076	52.01	7790.8	1561.0 - 18525.9
AUC_{0-t} (pg*hrs/mL)	38	145125.02962	83026.826590	57.21	114133.5578	40031.7230 - 370761.8465
$AUC_{0-\infty}$ (pg*hrs/mL)	38	167183.85653	99190.312420	59.33	124356.7359	43681.6046 - 431256.4303
T_{max} (hrs)	38	6.528	4.5259	69.33	4.50	1.52 - 24.00
$T_{1/2}$ (hrs)	38	15.60647	5.877286	37.66	13.5385	8.6386 - 34.6891
K_{el} (hr^{-1})	38	0.04931	0.014593	29.60	0.0512	0.0200 - 0.0802

Pharmacokinetic Parameters	Reference Product					
	N	Arithmetic Mean	S.D.	C.V. (%)	Median	Range
C_{max} (pg/mL)	38	9855.84	5203.617	52.80	9157.7	2087.9 - 23269.1
AUC_{0-t} (pg*hrs/mL)	38	153520.40588	73236.952473	47.71	124196.9035	41513.8135 - 324262.3170
$AUC_{0-\infty}$ (pg*hrs/mL)	38	173263.01017	78985.005787	45.59	140767.1066	45381.7532 - 355984.7286
T_{max} (hrs)	38	5.597	3.0999	55.39	4.50	0.75 - 12.00
$T_{1/2}$ (hrs)	38	14.80612	4.323992	29.20	14.1787	8.6020 - 27.4190
K_{el} (hr^{-1})	38	0.05041	0.013336	26.45	0.0489	0.0253 - 0.0806

The summary results are tabulated in section 14.2 and the individual subjects and mean pharmacokinetic parameters for both the test and reference formulations have been tabulated in appendix 16.2.6. The statistical output and pharmacokinetic from SAS® version 9.4 is appended in appendix 16.1.9.2.

95% Upper bound obtained from ANOVA, Within subject standard deviation (S.D.) for reference, within subject variance for reference, Intra-subject C.V. for reference and Point estimate (Ratio T/R) for the pharmacokinetic parameters C_{max}, AUC_{0-t} and $AUC_{0-\infty}$ for atorvastatin under fasting conditions are summarized in the following table:

Pharmacokinetic Parameters	Within subject S.D. for Reference (S_{WR})	Within Subject Variance for Reference (S^2wr)	Intra Subject C.V. for Reference (%)	Point Estimate (Ratio (T/R) (%))	95% Upper Bound
C_{max} (pg/mL)	0.302	0.091	30.90	99.80	-0.0
AUC_{0-t} (pg*hrs/mL)	0.239	0.057	24.25	94.98	-0.0
$AUC_{0-\infty}$(pg*hrs/mL)	0.237	0.056	24.04	95.37	-0.0

The ln-transformed least square mean and 90% confidence interval based on least square mean obtained from ANOVA and ratio of geometric means for the pharmacokinetic parameters AUC_{0-t} and $AUC_{0-\infty}$ for atorvastatin under fasting conditions are summarized in the following table:

Geometric Mean, Ratio & Power and 90 % Confidence Interval for Atorvastatin					
Pharmacokinetic Parameters	Geometric Mean		Ratio (T/R) (%)	Power (%)	90 % Confidence Interval (%)
	Test (T)	Reference (R)			
AUC_{0-t} (pg*hrs/mL)	412677.267	434478.335	94.98	99.83	87.82 - 102.72
$AUC_{0-\infty}$(pg*hrs/mL)	421562.572	442031.352	95.37	99.87	88.33 - 102.97

Geometric means & ratio of geometric means for the pharmacokinetic parameters C_{max}, AUC_{0-t} and $AUC_{0-\infty}$ for o-hydroxy atorvastatin and p-hydroxy atorvastatin under fasting conditions are summarized in the following tables:

Geometric Mean & Ratio for O-hydroxy Atorvastatin			
Pharmacokinetic Parameters	Geometric Mean		Ratio (T/R) (%)
	Test (T)	Reference (R)	
C_{max} (pg/mL)	46018.088	52747.775	87.24
AUC_{0-t} (pg*hrs/mL)	404899.236	443962.113	91.20
$AUC_{0-\infty}$ (pg*hrs/mL)	416361.909	456601.032	91.19

Geometric Mean & Ratio for P-hydroxy Atorvastatin			
Pharmacokinetic Parameters	Geometric Mean		Ratio (T/R) (%)
	Test (T)	Reference (R)	
C_{max} (pg/mL)	6733.313	8460.222	79.59
AUC_{0-t} (pg*hrs/mL)	124743.129	136827.696	91.17
$AUC_{0-\infty}$ (pg*hrs/mL)	142592.862	155476.628	91.71

ANOVA RESULTS:

ATORVASTATIN

Formulation Effect

Formulation effect found to be statistically insignificant (p-value >=0.05) for C_{max}, AUC_{0-t} & $AUC_{0-\infty}$.

Period Effect

Period effect found to be statistically significant (p-value < 0.05) for C_{max}, AUC_{0-t} & $AUC_{0-\infty}$.

A significant period effect is caused by the fact that in one of the two periods, the plasma levels C_{max}, AUC_{0-t} & $AUC_{0-\infty}$ are higher / lower than in other. This may be due to different conditions in period 1 and period 2. Some of the causes for significant period effects are volunteers, meal plan, dosing procedure and environmental conditions. However in this study all the conditions in period 1 and period 2 were identical. These are:

- Same volunteers were enrolled in both periods. Moreover, none of the volunteers had taken any medicine between period 1 and period 2.
- Same meal plan was given to volunteers in both periods.
- Dosing procedure was same in both periods.
- Environmental condition during dispensing and sample separation were similar in both periods.

Thus based on the above observations, since the conditions in period 1 and period 2 were identical, the period effect appears to be insignificant in nature and may not have any clinical consequences

Sequence Effect

Sequence effect found to be statistically insignificant (p-value >=0.1) for C_{max}, AUC_{0-t} & $AUC_{0-\infty}$.

Reference-Scaled Average Bioequivalence Approach:

Within Subject Standard Deviation (S_{WR}) AND Intra Subject Variability

Within subject standard deviation (S_{WR}) for reference formulation for ln-transformed pharmacokinetic parameters C_{max}, AUC_{0-t} & $AUC_{0-\infty}$ was found to be 0.302, 0.239 & 0.237 respectively.

Intra subject coefficient of variation for reference formulation for ln-transformed pharmacokinetic parameters C_{max}, AUC_{0-t} & $AUC_{0-\infty}$ was found to be 30.90%, 24.25% & 24.04% respectively.

Here $S_{WR} \geq 0.294$ for relevant pharmacokinetic parameter C_{max} i.e. intra-subject C.V. for reference formulation $\geq$ 30% for relevant pharmacokinetic parameter C_{max} then reference-scaled average bioequivalence approach was used to determine bioequivalence (BE) for the relevant pharmacokinetic parameter for C_{max} and

$S_{WR} < 0.294$ for all relevant pharmacokinetic parameters AUC_{0-t} & $AUC_{0-\infty}$ i.e. intra-subject C.V. for reference formulation < 30% for all relevant pharmacokinetic parameters AUC_{0-t} & $AUC_{0-\infty}$ then un-scaled average bioequivalence approach was used to determine bioequivalence (BE) for the all relevant pharmacokinetic parameters for AUC_{0-t} & $AUC_{0-\infty}$.

Point Estimate (Ratio T/R) AND 95% Upper Bound

The Point estimate (Ratio T/R) and 95% upper bound for the ln-transformed pharmacokinetic parameters C_{max}, AUC_{0-t} & $AUC_{0-\infty}$ were found to be respectively 99.80% -0.0; 94.98% -0.0 and 95.37% & -0.0.

Unscaled Average Bioequivalence Approach:

Ratio and 90% Confidence Interval

The ratio of geometric mean and 90% confidence interval for the ln-transformed pharmacokinetic parameters AUC_{0-t} & $AUC_{0-\infty}$ were found to be respectively 94.98% & 87.82% - 102.72% and 95.37% & 88.33% - 102.97%.

Power AND Intra Subject Variability

The power for ln-transformed pharmacokinetic parameters AUC_{0-t} & $AUC_{0-\infty}$ was found to be 99.83% and 99.87% respectively.

Intra subject coefficient of variation for test as well as reference formulation for ln-transformed pharmacokinetic parameters AUC_{0-t} & $AUC_{0-\infty}$ was found to be 16.11% & 23.24% and 15.54% & 22.91% respectively.

O-Hydroxy Atorvastatin

Ratio

The ratio of geometric mean for the ln-transformed pharmacokinetic parameters C_{max}, AUC_{0-t} & $AUC_{0-\infty}$ were found to be respectively 87.24%; 91.20% and 91.19%.

P-Hydroxy Atorvastatin:

Ratio

The ratio of geometric mean for the ln-transformed pharmacokinetic parameters C_{max}, AUC_{0-t} & $AUC_{0-\infty}$ were found to be respectively 79.59%; 91.17% and 91.71%.

11.4.2 Statistical / Analytical Issues

Statistical Issues

There were no statistical issues.

Analytical Issues

There were no analytical issues.

11.4.3 Tabulation of Individual Response Data

All individual subject concentration data that was used for pharmacokinetic analysis at each time point is appended as appendix 16.2.5. Individual plasma concentration time curves are presented in linear and log-linear scale in appendix 16.2.5.

11.4.4 Drug Dose, Dose Concentration and Relationships to Response

In the present bioequivalence study the pharmacokinetic end points were considered for the bioequivalence conclusion and hence the pharmacodynamic was not measured. Thus the drug dose, dose concentration and relationship to response were not evaluated.

11.4.5 Drug-Drug and Drug-Disease Interactions

Not Applicable

11.4.6 By-Subject Displays

Not applicable

11.4.7 Efficacy Conclusions

The 95% Upper Bound for ($\left(\overline{Y}_T - \overline{Y}_R\right)^2 - \theta * S_{WR}^2$) for C_{max} was <=0 and Point estimate (Ratio T/R) of atorvastatin was entirely included in the range of 80.00 – 125.00% for C_{max}.

The 90% confidence interval for the ratio (Test/Reference) of AUC_{0-t} and $AUC_{0-\infty}$ of atorvastatin were within the acceptable limits of bioequivalence 80.00% - 125.00%.

Thus it is concluded that the test product, Atorvastatin Calcium tablets 80 mg manufactured by Macleods Pharmaceuticals Ltd., India is bioequivalent to the reference product; Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg) Distributed by Pfizer Inc., USA in healthy, adult, human subjects under fasting condition.

12.0 SAFETY EVALUATION

12.1 Extent of Exposure

- Nineteen subjects (subject number 01, 04, 05, 07 to 10, 13, 15, 16, 17, 19 to 23, 27, 28 and 30) who completed the study were exposed to Atorvastatin Calcium 80 mg; four times as per randomization schedule.

- Subject number 03 [who was withdrawn from the study in period 3 (post-dose) on principal investigator advice due to adverse event] was exposed to Atorvastatin Calcium 80 mg; thrice as per randomization schedule.
- Subject number 14 and 18 [who did not report to the facility due to personal reason for period 3 thus considered dropped-out from the study] and subject number 11 [who was found to participate in other clinical trial and thus was withdrawn from the study based on exclusion criteria during check-in of period 3 (pre-dose)] were exposed to Atorvastatin Calcium 80 mg; twice as per randomization schedule.
- Subject number 24 [who was withdrawn from the study in period 1 (post-dose) on principal investigator advice due to serious adverse events]; subject number 06 and 25 [who were withdrawn from the study in period 1 (post-dose) on principal investigator advice due to adverse events] and subject number 02, 12, 26 and 29 [who did not report to the facility due to personal reason for period 2 thus considered dropped-out from the study] were exposed to Atorvastatin Calcium 80 mg; once as per randomization schedule.

12.2 Adverse Events (AEs)

12.2.1 Brief Summary of Adverse Events

Three subjects (subject number 03, 06 and 25) experienced adverse event during study period.

- Subject number 03 had nausea and one episode of vomiting after 4 hours 25 minutes of dosing of period 3.
- Subject number 06 had blackout in front of the eye associated with heaviness in head after 1 hour 7 minutes of dosing of period 1.
- Subject number 25 had severe pain over abdominal region associated with weakness after 5 hours 38 minutes of dosing of period 1.

There were few out of reference range laboratory values obtained at the post-study assessment but these were not clinically significant except for subject number 12. The details for the same are given in section 12.4.2. The clinically significant out of reference range values for the post study examination of the subject's laboratory investigations are tabulated in 14.3.4.

12.2.2 Displays of Adverse Events

The list of adverse events occurred after initiation of the study is displayed in summary table in section 12.2.4 and the post-study clinically significant out of reference range laboratory results are listed in section 14.3.4.

12.2.3 Analysis of Adverse Events

The relationship of the drug to the adverse events experienced by subjects during conduct of the study is given in section 12.2.4.

The out of reference range laboratory values obtained during post-study evaluation except for that listed in section 12.2.4 were considered to be clinically non significant. The relationship of the

drug to the clinically significant out of reference range laboratory values obtained during post-study evaluation is given in section 12.2.4.

12.2.4 Listing of Adverse Events by Subjects

Three subjects (subject number 03, 06 and 25) experienced adverse events during conduct of the study.

The relationships of the drug to the adverse events experienced by the subjects are as mentioned below:

Sub. No.	Adverse Event	Date and Time of Last Dosing	Date and time of Occurrence	Time of AE since Last Dose	Date and time of Resolution	Duration of AE (From Occurrence to Resolution)	Relationship with the Study Drug	Treatment Received (Sequence)
03	Nausea and one episode of vomiting	27th January 2015 09:06 hrs	27th January 2015 13:31 hrs	4 hours 25 minutes	28th January 2015 07:30 hrs	17 hours 59 minutes	Possible	R (RTRT)
06	Blackout in front of the eye associated with heaviness in head	13th January 2015 09:15 hrs	13th January 2015 10:22 hrs	1 hour 07 minutes	14th January 2015 07:50 hrs	21 hours 28 minutes	Possible	T (TRTR)
25	Severe pain over abdominal region associated with weakness	13th January 2015 09:27 hrs	13th January 2015 15:05 hrs	5 hours 38 minutes	14th January 2015 08:00 hrs	16 hours rf55 minutes	Possible	T (TRTR)

The relationship of the drug to the clinically significant out of reference range laboratory values obtained during post-study evaluation is as mentioned below:

Subject Number	Laboratory Parameter	Safety Assessment Results	Reference Range	Remark	Relationship with the Study Drug
12	SGPT	91.0U/L	30.0- 65.0 U/L	Increased	Possible
	SGOT	43.6 U/L	15.0- 37.0 U/L		

12.3 Deaths, Other Serious Adverse Events and Other Significant Adverse Events

One subject (subject number 24) experienced serious adverse event during study period.

- Subject number 24 had severe abdominal cramps associated with headache, restlessness and lethargy after 1 hour 34 minutes of dosing of period 1. Subject was withdrawn from the study on principal investigator advice and was hospitalized for further management.

The relationship of the drug to the serious adverse event experienced by the subject is as mentioned below:

Sub. No.	Adverse Event	Date and Time of Last Dosing	Date and time of Occurrence	Time of AE since Last Dose	Date and time of Resolution	Duration of AE (From Occurrence to Resolution)	Relationship with the Study Drug	Treatment Received (Sequence)
24	Severe abdominal cramps associated with headache, restlessness and lethargy	13th January 2015 09:24 hrs	13th January 2015 10:58 hrs	1 hour 34 minutes	14th January 2015 14:59 hrs (SAE) 19th January 2015 16:31 hrs (AE)	6 days 5 hours and 33 minutes	Possible	T (TRTR)

Note: Subject was hospitalized on 13th January 2015 and was discharged on 14th January 2015, however his treatment was continued.

12.4 Clinical Laboratory Evaluation

12.4.1 Listing of Individual Laboratory Measurements by Subjects and Each Abnormal Laboratory Value

Listed in appendix 16.2.8 are the various biochemical, haematological and urine sample assessment for the subjects, pre and post-clinical phase. The clinically significant out of reference range values for the post-study examination of the subject's laboratory investigations are tabulated in 14.3.4.

12.4.2 Evaluation of Each Laboratory Parameters

There were few out of reference range laboratory values obtained at the post-study assessment, but these were not clinically significant except for subject number 12 who showed clinically significant post-study out of reference range laboratory value. All the post-study safety assessment clinical and laboratory results are given in appendix 16.2.8, 'Listing of Individual Laboratory Parameters by Subject' [B- Laboratory Tests Report (Post-study safety Assessment)]. All the laboratory results, which were outside the reference range but within the 'acceptable limit', (for acceptable limit refer IEC approved Protocol, appendix V) were not considered clinically significant. There were few values as given below, which were outside the acceptable limits, however not considered clinically significant based on the clinical co-relation and have not been included as adverse events. For subject 12 the laboratory result outside acceptable range is considered as clinically significant based on the clinical co-relation and are reported as adverse event.

Sub. No.	Laboratory Parameter	Safety Assessment Results	Baseline Results	Reference Range	Remark
01	SGPT	77.3 U/L	62.4 U/L	30.0 – 65.0 U/L	Clinically not significant
05	Potassium	5.40 mmol/L	5.30 mmol/L	3.50 – 5.10 mmol/L	Clinically not significant
06	Triglycerides	287.4 mg/dL	174.0 mg/dL	< 150.0 mg/dL	Clinically not significant
12	SGPT	91.0U/L	69.8 U/L	30.0- 65.0 U/L	Clinically significant
	SGOT	43.6 U/L	34.4 U/L	15.0- 37.0 U/L	
13	ESR	22 mm/hr	6 mm/hr	< 15 mm/hr	Clinically not significant
	Alkaline Phosphatase	155.5 U/L	160.3 U/L	50.0- 136.0 U/L	
18	SGPT	80.8 U/L	76.8 U/L	30.0- 65.0 U/L	Clinically not significant
22	ESR	21 mm/hr	26 mm/hr	< 15 mm/hr	Clinically not significant
23	Eosinophils	10%	7%	1 – 6%	Clinically not significant
25	SGOT	41.9 U/L	24.4 U/L	15.0- 37.0 U/L	Clinically not significant
27	Potassium	5.40 mmol/L	4.60 mmol/L	3.50 – 5.10 mmol/L	Clinically not significant
29	Triglycerides	244.0 mg/dL	140.0 mg/dL	< 150.0 mg/dL	Clinically not significant

The clinically significant out of reference range values for the post-study examination of the subject's laboratory investigations are tabulated in 14.3.4.

12.5 Vital Signs, Physical Findings and Other Observations Related to Safety

Laboratory assessments, including urine test for drugs of abuse and alcohol breath test were carried out prior to the study.

In the pre-study assessments, all out of range clinical laboratory values were clinically acceptable, for the subjects enrolled in the study.

Vital signs and subject questionnaire was done at the time of check-in, pre-dose and at 1.00, 3.00, 9.00, 24.00, 34.00 and 48.00 hours post-dose (Time points being relative to the investigational product dosing).

Individual recording of vital signs for all the periods has been appended as appendix 16.2.8 (Table D).

Medical examinations were carried out at the time of check-in, check-out and at 48.00 hours post-dose of all periods. All the subjects enrolled and dosed had clinically acceptable vital signs values.

A medical officer was available within the clinical facility whenever the subjects were housed (from check-in to check-out).

At the end of the study period, post-study safety assessments of all the enrolled and dosed subjects (except subject number 02, 14 and 26) were carried out.

Subject number 02, 14 and 26 were considered dropped-out from the study; however subjects were asked to attend for the post-study examination but they did not report to the facility which is documented in respective follow-up log.

Post-study safety assessments included: Medical examination including recording of vital signs [Blood Pressure (BP), Pulse and Temperature], general examination, physical and systemic examination. 12-lead ECG for heart rate, rhythm and specific finding (if any). Laboratory parameter investigation including complete blood count – erythrocyte count, platelet count, haemoglobin, hematocrit, leucocyte count, ESR and differential leucocyte count (DLC); Biochemistry –blood sugar (random), total cholesterol and triglycerides; Hepatic profile – SGOT, SGPT, GGT, alkaline phosphatase and serum bilirubin (total, direct, indirect), Renal profile – serum creatinine, BUN, calcium, electrolytes (sodium, potassium, chlorides).

No clinically significant abnormalities in ECGs were reported in subject's post-study safety assessments. The individual clinical impression of ECG has been appended as appendix 16.2.8 (Table C).

Laboratory values outside the reference range were considered clinically not significant based on clinical co-relation and have not been included as adverse events as given in section 12.4.2. The individual recordings of laboratory parameters are appended in appendix 16.2.8 (Table B).

12.6 Safety Conclusions

Three subjects (subject number 03, 06 and 25) experienced adverse events during study period and one subject (subject number 24) experienced serious adverse event (SAE) during study period. During post-study safety assessment adverse event was reported for one subject (subject number 12).

13.0 DISCUSSION AND OVERALL CONCLUSIONS

A total of 30 subjects were planned and enrolled, of these 30, subject number 24 was withdrawn from the study in period 1 (post-dose) on principal investigator advice due to serious adverse events. Subject number 06 and 25 were withdrawn from the study in period 1 (post-dose) on principal investigator advice due to adverse events. Subject number 02, 12, 26 and 29 did not report to the facility due to personal reason for period 2 thus considered dropped-out from the study. Subject number 14 and 18 did not report to the facility due to personal reason for period 3 thus considered dropped-out from the study. Subject number 11 was found to participate in other clinical trial and thus was withdrawn from the study based on exclusion criteria during check-in of period 3 (pre-dose). Subject number 03 was withdrawn from the study in period 3 (post-dose) on principal investigator advice due to adverse event. Thus nineteen subjects completed all the periods of the study.

Single dose administration of Atorvastatin Calcium 80 mg was well tolerated and no new safety issues were identified during the study. Three subjects discontinued study treatment due to adverse events, which were possibly related to the study drug. One subject experienced serious adverse events withdrawn from the study, was possibly related to the study drug. There were few

clinically relevant changes in laboratory safety variable for one subject; which was possibly related to the study drug.

The plasma samples of nineteen subjects (subject number 01, 04, 05, 07 to 10, 13, 15, 16, 17, 19 to 23, 27, 28 and 30) completing the study was analyzed for atorvastatin and for ortho- and parahydroxylated metabolites of atorvastatin concentration level and the data were taken for pharmacokinetic and statistical evaluations.

The plasma samples of subject number 02, 03, 06, 11, 12, 14, 18, 24 to 26 and 29 who were considered dropped-out from the study were not analyzed.

The mean plasma concentration time profiles of atorvastatin, o-Hydroxy atorvastatin and p-hydroxy atorvastatin for the two treatments are shown in Figure 1 to 6 (Refer section 14.2 'Efficacy Data').

Atorvastatin

After oral administration of the reference product under fasting condition the drug was absorbed with median t_{max} of 1.50 hrs. Where for other PK parameters mean C_{max} 89331.23 pg/mL (range 25874.3 - 228846.1 pg/mL), AUC_{0-t} 467782.88230 pg*hrs/mL (range 138286.8055 - 817817.5720 pg*hrs/mL) and $AUC_{0-\infty}$ 475087.48443 pg*hrs/mL (range 146199.8011 - 831203.3702 pg*hrs/mL).

After oral administration of the test product under fasting condition the drug was absorbed with median t_{max} of 0.89 hrs. Where for other PK parameters mean C_{max} 90797.99 pg/mL (range 27004.3 - 244801.1 pg/mL), AUC_{0-t} 447481.74842 pg*hrs/mL (range 152971.7360 - 860786.5575 pg*hrs/mL) and $AUC_{0-\infty}$ 455621.34846 pg*hrs/mL (range 157688.6395 - 872271.5131 pg*hrs/mL).

O-Hydroxy Atorvastatin

After oral administration of the reference product under fasting condition the drug was absorbed with median t_{max} of 2.50 hrs. Where for other PK parameters mean C_{max} 59137.24 pg/mL (range 21841.0 - 177371.9 pg/mL), AUC_{0-t} 476579.32750 pg*hrs/mL (range 206503.6020 - 908514.0960 pg*hrs/mL) and $AUC_{0-\infty}$ 489182.85665 pg*hrs/mL (range 217690.8412 - 915379.4350 pg*hrs/mL).

After oral administration of the test product under fasting condition the drug was absorbed with median t_{max} of 1.88 hrs. Where for other PK parameters mean C_{max} 51122.06 pg/mL (range 20911.9 - 124837.9 pg/mL), AUC_{0-t} 438931.58080 pg*hrs/mL (range 192249.1945 - 844266.2250 pg*hrs/mL) and $AUC_{0-\infty}$ 449999.61838 pg*hrs/mL (range 205468.1162 - 886116.0995 pg*hrs/mL).

P-Hydroxy Atorvastatin

After oral administration of the reference product under fasting condition the drug was absorbed with median t_{max} of 4.50 hrs. Where for other PK parameters mean C_{max} 9855.84 pg/mL (range 2087.9 - 23269.1 pg/mL), AUC_{0-t} 153520.40588 pg*hrs/mL (range 41513.8135 - 324262.3170 pg*hrs/mL) and $AUC_{0-\infty}$ 173263.01017 pg*hrs/mL (range 45381.7532 - 355984.7286 pg*hrs/mL).

After oral administration of the test product under fasting condition the drug was absorbed with median t_{max} of 4.50 hrs. Where for other PK parameters mean C_{max} 7913.41 pg/mL (range 1561.0 - 18525.9 pg/mL), AUC_{0-t} 145125.02962 pg*hrs/mL (range 40031.7230 - 370761.8465 pg*hrs/mL) and $AUC_{0-\infty}$ 167183.85653 pg*hrs/mL (range 43681.6046 - 431256.4303 pg*hrs/mL).

Bioequivalence was assessed using Reference-Scaled and unscaled Average Bioequivalence Approach:

$S_{WR} \geq 0.294$ for relevant pharmacokinetic parameter C_{max}, i.e. intra-subject C.V. for reference formulation $\geq$ 30% for the relevant pharmacokinetic parameter C_{max} of atorvastatin, then reference-scaled average bioequivalence approach was used to determine bioequivalence (BE) for relevant pharmacokinetic parameter for C_{max} and

$S_{WR} < 0.294$ for all relevant pharmacokinetic parameters AUC_{0-t} and $AUC_{0-\infty}$, i.e. intra-subject C.V. for all reference formulation < 30% for all relevant pharmacokinetic parameters AUC_{0-t} and $AUC_{0-\infty}$ of atorvastatin, then un-scaled average bioequivalence approach was used to determine bioequivalence (BE) for all relevant pharmacokinetic parameters for AUC_{0-t} and $AUC_{0-\infty}$.

The 95% Upper Bound for ($\left(\overline{Y}_T - \overline{Y}_R\right)^2 - \theta * S_{WR}^2$) for C_{max} was <=0 and Point estimate (Ratio T/R) of atorvastatin was entirely included in the range of 80.00 – 125.00% for $C_{max.}$

The 90% confidence intervals for AUC_{0-t} and $AUC_{0-\infty}$ for atorvastatin were within the acceptable limits of bioequivalence 80.00% - 125.00% then the products were claimed to be bioequivalent.

In summary, the test formulation is bioequivalent to the reference in terms of both the rate and extent of absorption.

Both the formulations are well tolerated following a single dose administration of the investigational product.

14.0 TABLES, FIGURES AND GRAPHS REFERRED TO BUT NOT INCLUDED IN THE TEXT

14.1 Demographic Data

Demographic Data of Subjects Recruited in the Study:

	Age (Yrs)	Height (m)	Weight (kg)	BMI (kg/m^2)
Number	30	30	30	30
Median	28.00	1.67	65.45	23.19
Mean	28.9	1.676	65.46	23.289
Standard Deviation	4.83	0.0577	7.978	2.5177
Minimum	20	1.56	52.2	18.96
Maximum	37	1.85	81.0	27.14

Demographic Data of Subjects Completed the Study

	Age (Yrs)	Height (m)	Weight (kg)	BMI (kg/m^2)
Number	19	19	19	19
Median	29.00	1.65	64.00	22.49
Mean	30.4	1.666	64.02	23.037
Standard Deviation	4.42	0.0534	8.761	2.6020
Minimum	24	1.56	52.2	18.96
Maximum	37	1.74	81.0	27.06

14.2 Efficacy Data

TABLE-1 (Reference-Scaled Average Bioequivalence Approach)

Summary statistics of pharmacokinetic parameters for Atorvastatin after administration of single dose of Atorvastatin Calcium tablets 80 mg in 19 healthy, adult human subjects under fasting conditions.

Product/Statistics	C_{max} (pg/mL)	AUC_{0-t} (pg*hrs/mL)	$AUC_{0-\infty}$ (pg*hrs/mL)
Untransformed Reference Product (R)			
Arithmetic Mean	89331.23	467782.88230	475087.48443
S.D.	42076.832	167845.522970	169462.061370
C.V. %	47.10	35.88	35.67
N	38	38	38
Test Product (T)			
Arithmetic Mean	90797.99	447481.74842	455621.34846
S.D.	46727.300	173455.988200	173650.129440
C.V. %	51.46	38.76	38.11
N	38	38	38
Ratio of Arithmetic Mean (% Bioavailability)			
T/R (%)	101.64	95.66	95.90
Ratio (%) for Mean AUC_{0-t} to Mean $AUC_{0-\infty}$			
Reference		98.46	
Test		98.21	
Point Estimate (Ratio T/R) (%)	99.80	94.98	95.37
Within Subject Standard Deviation (S_{WR})			
Reference	0.302	0.239	0.237
Within Subject Variance (S^2_{WR})			
Reference	0.091	0.057	0.056
Intra Subject C.V. (%)			
Reference	30.90	24.25	24.04
95 % Upper Bound	-0.0	-0.0	-0.0
P-value (ANOVA) for ln-transformed Data			
Formulation	0.9756	N/AP	N/AP
Period	0.0200	N/AP	N/AP
Sequence	0.8520	N/AP	N/AP

Note: N/AP: Not Applicable

TABLE-2 (Unscaled Average Bioequivalence Approach)

Summary statistics of pharmacokinetic parameters for Atorvastatin after administration of single dose of Atorvastatin Calcium tablets 80 mg in 19 healthy, adult human subjects under fasting conditions.

Product/Statistics	**AUC_{0-t} (pg*hr/mL)**	**$AUC_{0-\infty}$ (pg*hr/mL)**
Log Transformed (Natural Log) Least Square Mean		
Reference	12.982	12.999
Test	12.930	12.952
Geometric Mean		
Reference	434478.335	442031.352
Test	412677.267	421562.572
Ratio of Geometric Mean		
T/R (%)	94.98	95.37
90 % Confidence Interval (T/R)		
Lower limit (%)	87.82	88.33
Upper limit (%)	102.72	102.97
Power (%)	99.83	99.87
D.F.	40.8	40.8
Intra Subject C.V. (%)		
Reference	23.24	22.91
Test	16.11	15.54
Mean Square Error (MSE)		
Reference	0.0526	0.0511
Test	0.0256	0.0239
P-value (ANOVA) for ln-transformed Data		
Formulation	0.2752	0.3040
Period	0.0002	0.0001
Sequence	0.7021	0.7086

TABLE-3

Summary statistics of pharmacokinetic parameters for O-Hydroxy Atorvastatin after administration of single dose Atorvastatin Calcium tablets 80 mg in 19 healthy, adult human subjects under fasting conditions.

Product/Statistics	C_{max} (pg/mL)	AUC_{0-t} (pg*hrs/mL)	$AUC_{0-\infty}$ (pg*hrs/mL)
Untransformed Reference Product (R)			
Arithmetic Mean	59137.24	476579.32750	489182.85665
S.D.	32050.126	185150.003210	186627.402680
C.V. %	54.20	38.85	38.15
N	38	38	38
Test Product (T)			
Arithmetic Mean	51122.06	438931.58080	449999.61838
S.D.	23399.162	176413.463600	178433.189500
C.V. %	45.77	40.19	39.65
N	38	38	38
Ratio of Arithmetic Mean (% Bioavailability)			
T/R (%)	86.45	92.10	91.99
Ratio (%) for Mean AUC_{0-t} to Mean $AUC_{0-\infty}$			
Reference	97.42		
Test	97.54		
Geometric Mean			
Reference	52747.775	443962.113	456601.032
Test	46018.088	404899.236	416361.909
Ratio of Geometric Mean			
T/R (%)	87.24	91.20	91.19

TABLE-4

Summary statistics of pharmacokinetic parameters for P-Hydroxy Atorvastatin after administration of single dose Atorvastatin Calcium tablets 80 mg in 19 healthy, adult human subjects under fasting conditions.

Product/Statistics	C_{max} (pg/mL)	AUC_{0-t} (pg*hrs/mL)	$AUC_{0-\infty}$ (pg*hrs/mL)
Untransformed Reference Product (R)			
Arithmetic Mean	9855.84	153520.40588	173263.01017
S.D.	5203.617	73236.952473	78985.005787
C.V. %	52.80	47.71	45.59
N	38	38	38
Test Product (T)			
Arithmetic Mean	7913.41	145125.02962	167183.85653
S.D.	4116.076	83026.826590	99190.312420
C.V. %	52.01	57.21	59.33
N	38	38	38
Ratio of Arithmetic Mean (% Bioavailability)			
T/R (%)	80.29	94.53	96.49
Ratio (%) for Mean AUC_{0-t} to Mean $AUC_{0-\infty}$			
Reference	88.61		
Test	86.81		
Geometric Mean			
Reference	8460.222	136827.696	155476.628
Test	6733.313	124743.129	142592.862
Ratio of Geometric Mean			
T/R (%)	79.59	91.17	91.71

Figure 1
Comparative Linear Plot of Atorvastatin Mean Plasma Concentration (pg/mL) Vs Time (Hour)
subject=MEAN

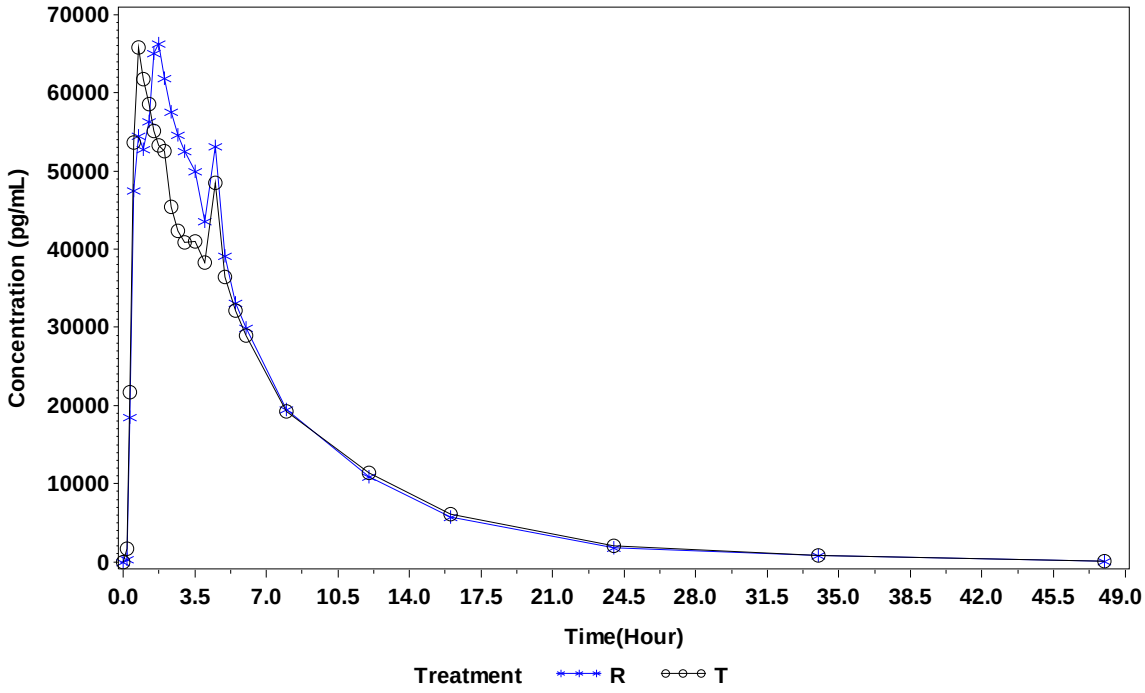


Figure 2
Comparative Semi Log Plot of Atorvastatin Mean Plasma Concentration (pg/mL) Vs Time (Hour)
subject=MEAN

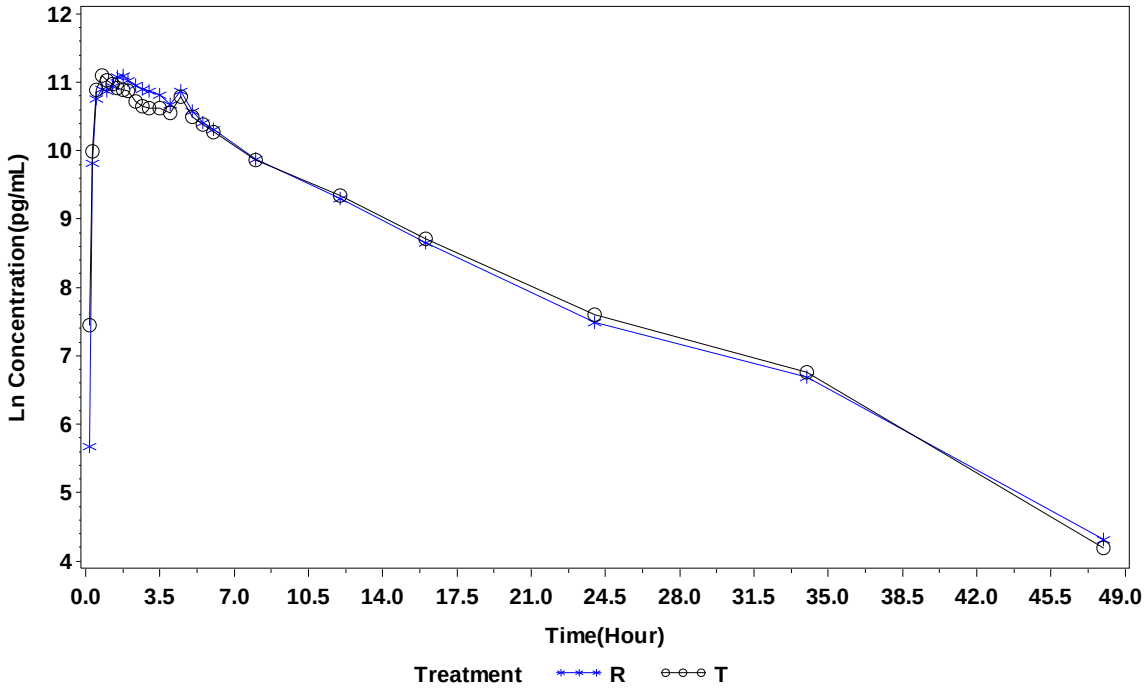


Figure 3
Comparative Linear Plot of O-Hydroxy Atorvastatin Mean Plasma Concentration (pg/mL) Vs Time (Hour)
subject=MEAN

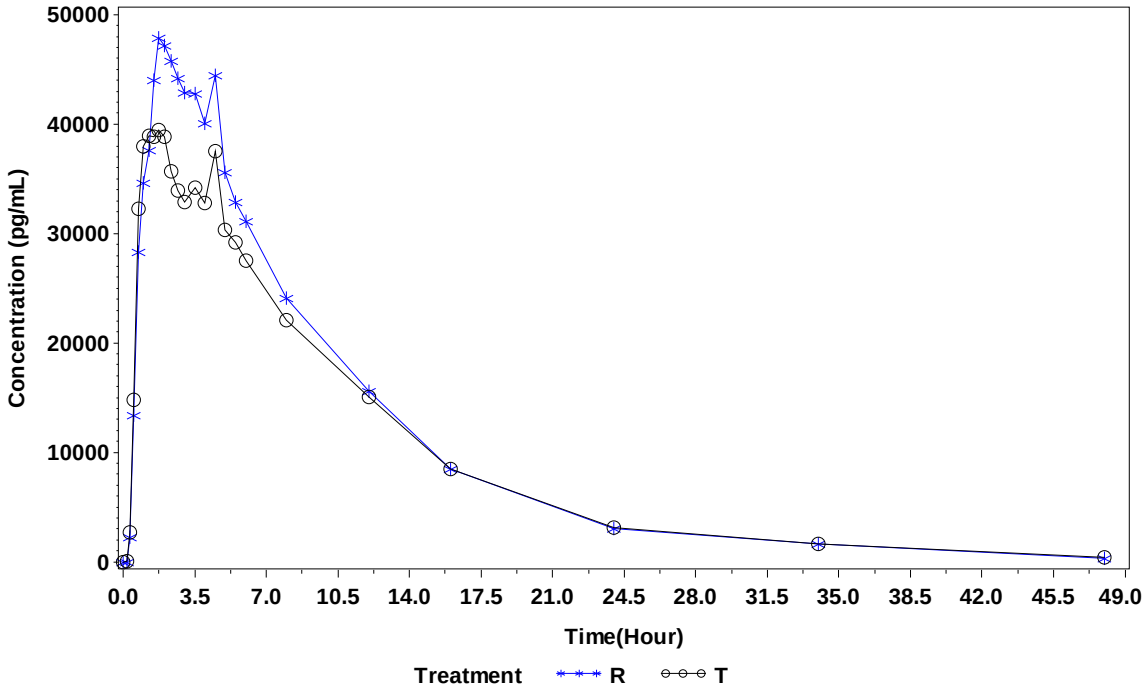


Figure 4
Comparative Semi Log Plot of O-Hydroxy Atorvastatin Mean Plasma Concentration (pg/mL) Vs Time (Hour)
subject=MEAN

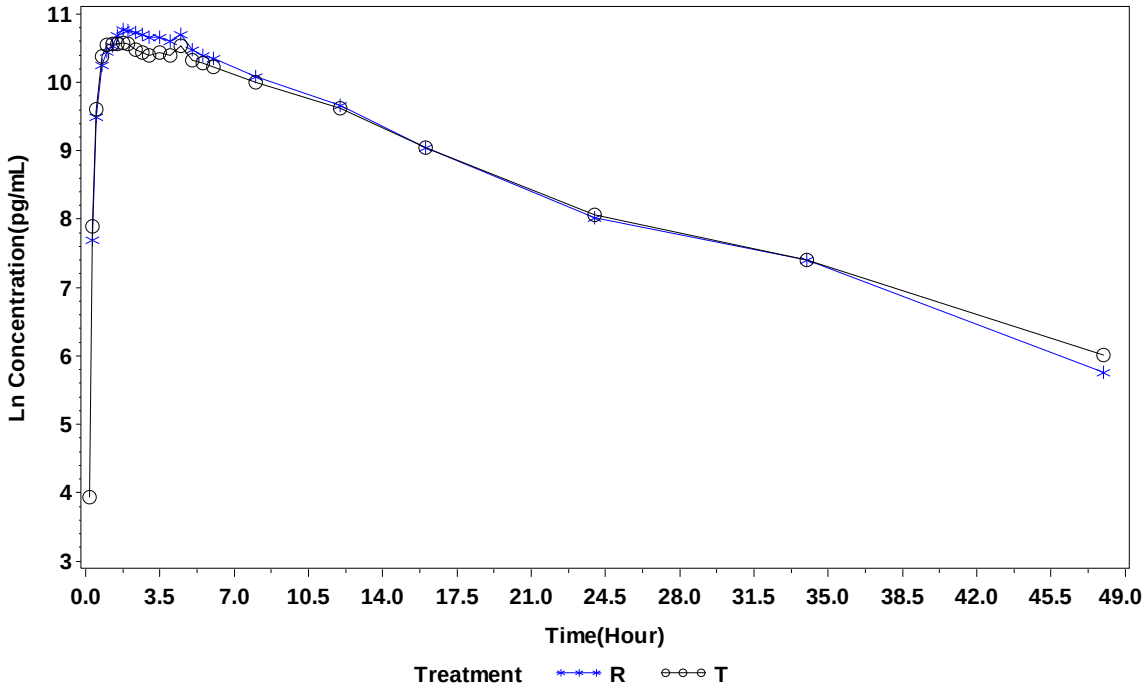


Figure 5
Comparative Linear Plot of P-Hydroxy Atorvastatin Mean Plasma Concentration (pg/mL) Vs Time (Hour) subject=MEAN

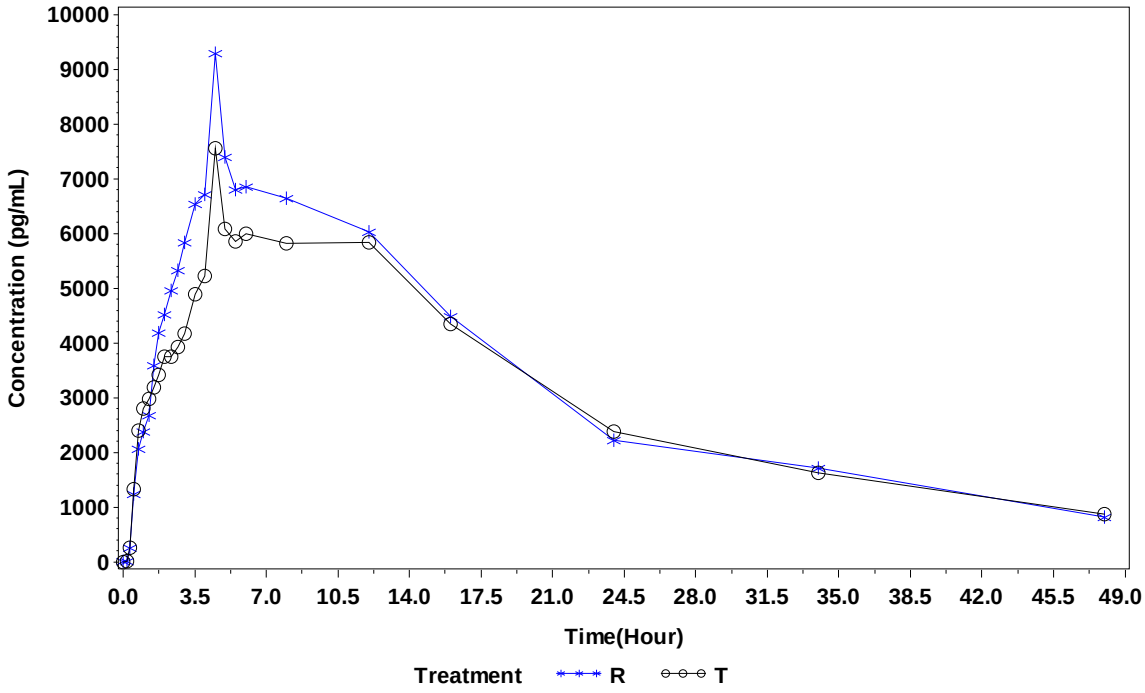
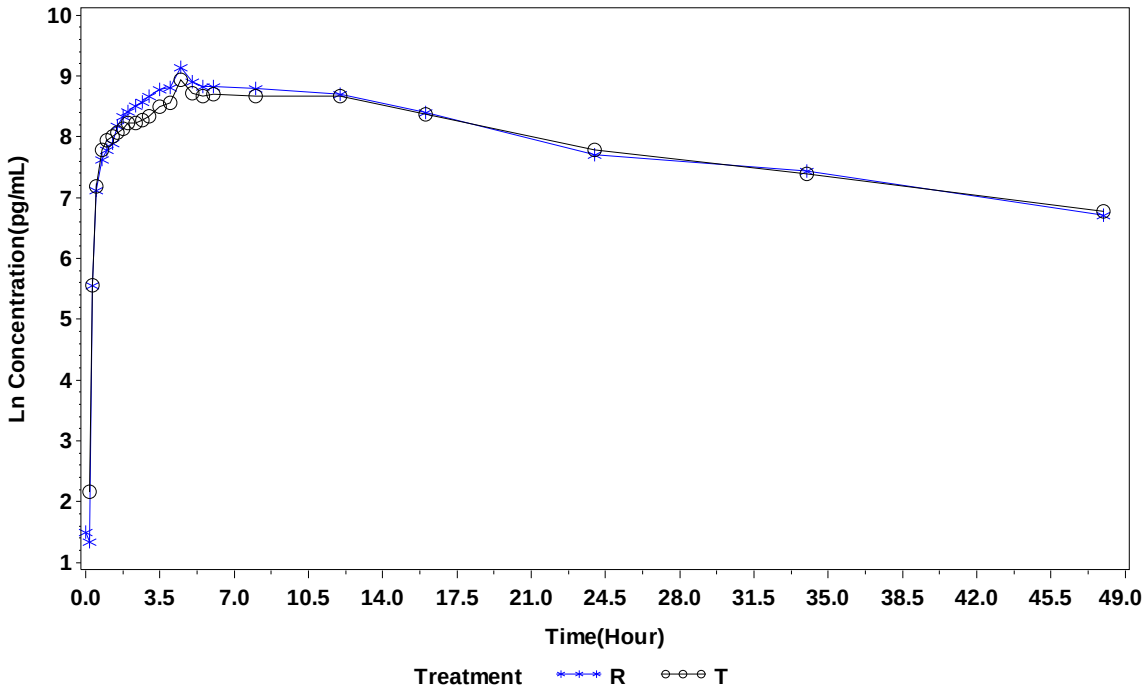


Figure 6
Comparative Semi Log Plot of P-Hydroxy Atorvastatin Mean Plasma Concentration (pg/mL) Vs Time (Hour) subject=MEAN



14.3 Safety Data

14.3.1 Display of Adverse Events

Three subjects (subject number 03, 06, and 25) experienced adverse events during study period and one subject (subject number 24) experienced serious adverse events (SAE) during study period. One subject (subject number 12) enrolled in the study was found to have clinically significant post-study laboratory value. The list of clinically significant post-study out of reference range laboratory value is listed in section 14.3.4.

The detailed description of the adverse events and their handling are given in appendix 16.2.7.

14.3.2 Listings of Deaths, Other Serious and Significant Adverse Events

Subject (subject number 24) experienced serious adverse events (SAE) during study period.

14.3.3 Narrative of Deaths, Other Serious and Certain Other Significant Adverse Events

Not applicable

14.3.4 Abnormal Laboratory Value Listing

Following table gives details of outside acceptable limit findings, which are clinically significant in laboratory test of subject post-study.

Subject No.	Laboratory Parameter	Reference Range	Safety Assessment Results	Follow-up Result	Comments
12	SGPT	30.0 - 65.0 U/L	91.0 U/L	58.1 U/L	Resolved
	SGOT	15.0 - 37.0 U/L	43.6 U/L	31.0 U/L	

15.0 REFERENCE LIST

(Refer appendix 16.1.12; 'Important Publications Referenced in the Report')

1. U.S. Food and Drug Administration-Drug product label of Lipitor® 80 mg (Atorvastatin calcium tablets 80 mg). NDA No. 020702, approved on 21/05/2014 Cited on 14 /11/2014. Available From: http://www.accessdata.fda.gov/drugsatfda_docs/label/2014/020702s064lbl.pdf
2. Pharmacokinetics of atorvastatin and its metabolites after single and multiple dosing in hypercholesterolaemic haemodialysis patients. Nephrology Dialysis Transplantation. Volume 18, Number 5 Pp. 967-976. Cited on 14 /11/2014. Available From: http://ndt.oxfordjournals.org/content/18/5/967/T2.expansion.html