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Research Article

An Open-label, Randomized, Single-dose, Crossover Study in Healthy Adult Volunteers to Evaluate Pharmacokinetic Interactions and Tolerability of a Fixed-dose Combination with Rosuvastatin and Ezetimibe

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ABSTRACT

Recent studies have shown that adding ezetimibe to statins could further reduce levels of Low-Density Lipoprotein Cholesterol (LDL-C), total cholesterol and triglycerides, and likely increase high-density lipoprotein cholesterol levels. We conducted an open-label, crossover, single-dose, 3 periods study in 34 healthy Mexican volunteers under fasting conditions, randomly allocated into 3 treatment groups: Rosuvastatin (20 mg), ezetimibe (10 mg), and FDC of rosuvastatin (20 mg) and ezetimibe (10 mg), to evaluate the pharmacokinetics (PKs) of drug interactions between rosuvastatin and ezetimibe as well as the tolerability of the Fixed-Dose Combination (FDC). All subjects were administered the FDC tablet as well as the same dose of both drugs given separately as monotherapy. The geometric mean ratio (90% CI) for rosuvastatin in FDC over the single dose was 1.032 (0.937-1.138) for C_{max} and 1.09 (0.998-1.190) for $AUC_{0\text{-}inf}$. In the case of ezetimibe C_{max} was 0.897 (0.829-0.971) with an $AUC_{0\text{-}inf}$ of 0.993 (0.916-1.076). A total of 8 Adverse Events (AEs) were reported, the frequency was similar for FDC than in the treatments administered separately. No clinically significant PK interactions between rosuvastatin and ezetimibe were found, studied parameters were within conventionally accepted bioequivalence criteria. Tolerability profiles showed to be similar; therefore, the FDC was well tolerated.

Keywords: Rosuvastatin; Ezetimibe; Pharmacokinetic; Drug-Drug Interaction (DDI); Fixed-Dose Combination (FDC); Bioequivalence

INTRODUCTION

Rosuvastatin is a synthetic 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitor that effectively reduces low density lipoprotein cholesterol levels [1], designed to obtain potent enzyme inhibition at the HMGCoA reductase site. The oral bioavailability of rosuvastatin is approximately 20%, its peak plasma concentration (C_{max}) of 6.1 ng/ml occurs at 5 hours (T_{max}) after a single oral 20 mg dose [2]. Reductions in levels of triglycerides (up to 35%) and increments in levels of high density lipoprotein cholesterol (up to 14%) also have been consistently observed with rosuvastatin, that is why this drug is well tolerated [3].

Ezetimibe is a selective cholesterol absorption inhibitor, approved for use as a monotherapy or combination therapy with 3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors for patients with hypercholesterolemia when statin monotherapy does not achieve acceptable low-density lipoprotein cholesterol levels [4]. Following oral administration, ezetimibe is rapidly absorbed and extensively metabolized (>80%) to the pharmacologically active ezetimibe-glucuronide [5]. Absorption of ezetimibe is rapid and not altered by food content following oral administration. The drug is not metabolized by the cytochrome P450 system but extensive glucuronidation takes place in the intestine. Its combination with statins has obtained a greater effect, specified to the patient to reach the recommended target figures. Recent studies have shown that adding ezetimibe to statins could further reduce levels of Low-Density Lipoprotein Cholesterol (LDL-C), total cholesterol and triglycerides, and likely increase high density lipoprotein cholesterol levels [6].

The main purpose of this study was to examine the pharmacokinetic (PKs) interactions and tolerability of rosuvastatin and ezetimibe in FDC (Trezete®) when administered in healthy Mexican subjects.

MATERIALS AND METHODS

Patient selection

Healthy male and female volunteers aged 18-55 years with body mass index of 18.0 kg/m^2-27.0 kg/m^2 were eligible for the study.

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All subjects were recognized to be in good health based on medical history, physical examinations, vital signs (blood pressure, heart rate, and body temperature), 12-lead Electrocardiogram (ECG), clinical laboratory tests (hematology, blood chemistry, and urinalysis), serology (hepatitis B surface antigen, hepatitis C virus antibodies, and HIV antigen/antibodies), Venereal Disease Research Laboratory (VDRL) test, pregnancy test (when applied) and urine drug screening (amphetamines, methamphetamines, barbiturates, cocaine, opiates, benzodiazepine, cannabinoids) within 12 hours before the first administration of the study drug. Subjects with a known allergy or hypersensitivity to rosuvastatin or ezetimibe, subjects with a history of dyspepsia, gastritis, esophagitis, duodenal or gastric ulcer, subjects with a medical history or diet that might interfere with drug absorption, distribution, metabolism, or excretion or with a history of drug or alcohol abuse, were excluded.

30 subjects were required to detect a 20% difference between test and reference drugs with 80% statistical power at a 5% level of significance, assuming 34% of Intra-subject or within-subject coefficient of variation peak plasma concentration (C_{max}) for ezetimibe, an additional 6 subjects were considered in case of contingency for subjects who might drop out or no compliance to protocol [7]. The study protocol and informed consent form were approved by an Independent Ethics Committee, a Research Committee (protocol number: CEI_024_0417BD_187s) and the Ministry of Health in Mexico (COFEPRIS), and all subjects provided written informed consent before participating.

Study design

The study design was open-label, randomized, single-dose, 3-treatment, 3-period, 6-sequence, crossover, with 10 days washout interval between periods (Figure 1). The test medication was 20 mg rosuvastatin/10 mg ezetimibe FDC tablets (Trezete®; Laboratorios Silanes), and the reference tablets were 20 mg of rosuvastatin (Crestor®; AstraZeneca) and 10 mg of ezetimibe (Ezetrol®; MSD).

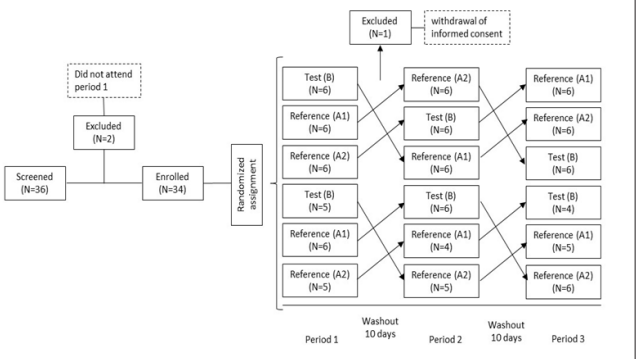

Figure 1: Study design: Open-label, single-dose, three-treatment, three-period, six-sequence, cross over. Test (B): Rosuvastatin+ Ezetimibe (FDC), 20 mg/10 mg, Reference (A1): Crestor® (Rosuvastatin), 20 mg, Reference (A2): Ezetrol® (Ezetimibe), 10 mg

All subjects were administered a FDC tablet containing 20 mg rosuvastatin and 10 mg ezetimibe and were also administered the same dose of both drugs given separately as monotherapy. All treatments were given under fasting conditions with 250 mL of water.

To measure the plasma concentration of rosuvastatin and ezetimibe, blood samples were drawn at 0 (pre-dose), 0.25, 0.5, 0.75, 1, 2, 3, 4, 5, 6, 7, 8, 10, 14, 24, 36, 48, 72 and 96 h after administration in each period. The samples were centrifuged at 4,500 rpm for 5 minutes and stored at -70°C ± 10°C until analysis.

Any Adverse Events (AE) were reviewed by integrating data from vital signs, clinical laboratory tests, physical examinations, and patient interviews. AEs were recorded in terms of symptoms and signs, duration, severity, relationship to the study drug, the action is taken, outcome, and seriousness.

Bioanalysis

Two independent analytical methods were validated to determine the plasma concentration of rosuvastatin and ezetimibe. No relevant cross-talk or matrix effects were recognized.

Rosuvastatin

The plasma concentration of rosuvastatin was analyzed using a liquid chromatographytandem mass spectrometric method (CLAR MS/MS) (AB Sciex, API 4500 QTrap). The analytical method was validated in a range of 0.4 ng/mL-100 ng/mL.

Ezetimibe

The plasma concentration of ezetimibe was analyzed using a liquid chromatographytandem mass spectrometric method (CLAR MS/MS) (Agilent Technologies, G6410B). The analytical method was validated in the range of 0.15 ng/mL-16 ng/mL.

PK analysis

The PK parameters of rosuvastatin and ezetimibe were assessed by non-compartmental analysis using Phoenix®/WinNonlin software. The AUC_{0-t} was de-termined using the trapezoidal rule. The area under the plasma concentration-time curve from time zero to infinity (AUC_{0-inf}) was calculated using the formula: $AUC_{0-inf}=AUC_{0-t}+Ct/k$, where Ct is the last measured plasma concentration and k is the terminal elimination rate constant. The C_{max} and the time to reach C_{max} (T_{max}) were determined from the plasma concentration time curve.

Statistical analysis

Statistical analysis was performed using SAS® software 9.2. Continuous variable data are expressed as means ± Standard Deviation (SD), and categorical data as counts or percentages. To assess PK equivalence, C_{max} and AUC were log transformed, and the Geometric Mean Ratio (GMR) and their 90% CIs were determined.

RESULTS

Population characteristics

34 healthy subjects were enrolled (mean age, 29.58 ± 8.52 years; mean weight, 60.28 kg ± 7.61 kg; mean height 159.5 cm ± 9.6 cm). One subject did not complete the study due to withdraw consent. Tolerability profiles were determined using data from 34 subjects who were administered study drugs. PK analysis was performed using data from 33 subjects who completed the study. The demographic characteristics of the study subjects are shown in Table 1.

Table 1: Demographic characteristics of study subjects (n=34).

Characteristics	Mean ± SD
Age (years)	29.58 ± 8.52 (18–45)
Height (cm)	159.5 ± 9.6 (141–183)
Body weight (kg)	60.28 ± 7.61 (48–76)

Body mass index (Kg/m^2)	23.65 ± 1.77 (20.55–23.65)

Note: Data are expressed as mean ± standard deviation (min-max)

Pharmacokinetic profile

The mean plasma concentration-time profiles for rosuvastatin and ezetimibe ad-ministered separately or in FDC are shown in Figure 2. The PK parameters data are shown in Table 2.

Data are expressed as arithmetic mean ± standard deviation (CV%); one subject was excluded because during period 1 presented diarrhea; one subject was excluded because the subject only had two points after C_{max}. PK, pharmacokinetic; FDC, fixed-dose combination; C_{max}, maximum plasma concentration; T_{max}, time to reach C_{max}; AUC_{0-t}, area under the plasma concentration-time curve from time zero to the last sampling time; AUC_{0-inf}, area under the curve from zero to infinity; $t_{1/2}$, terminal elimination half-life.

(90% CIs) of C_{max} and AUC_{0-inf} were 1.032 (0.937-1.138) and 1.09 (0.998-1.190) for rosuvastatin and 0.897 (0.829-0.971) and 0.993 (0.916-1.076) for ezetimibe, respectively (Table 3).

During the study, a total of 8 Adverse Events (AEs) were reported, of these, 6 (75%) were with rosuvastatin (Crestor®), also was observed that women had higher frequency to AEs. Only the AE nausea was considered to be "related". The most frequently AE reported was a mild-to-moderate headache that was self-limiting. The number of subjects reporting AEs following the rosuvastatin tablet (n=4), ezetimibe tablet (n=1), and FDC (n=1) was similar (Table 4).

Two events required intervention: 1 case of moderate headache, the subject self-administered acetaminophen, and 1 case of acute upper respiratory infection required ceftriaxone and acetaminophen. The other AEs were mild and resolved spontaneously; there were no serious AEs.

No clinically significant abnormalities were found in vital signs, laboratory test results, during the physical examinations, on the electrocardiograms, or in physical examinations.

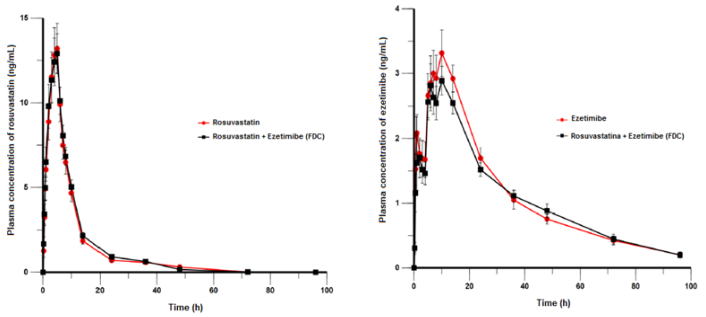

Figure 2: Mean (SE) plasma concentration-time curves for rosuvastatin 20 mg (A) and ezetimibe 10 mg (B)

Table 2: Pk parameters of rosuvastatin, 20 mg, and ezetimibe 10 mg, administered separately or in FDC in healthy subjects.

PK parameters	Rosuvastatin Crestor® (n=32) [a]	Ezetimibe Ezetrol® (n=33)	Rosuvastatin+Ezetimibe	
			Rosuvastatin FDC (n=32)	Ezetimibe FDC (n=33)
C_{max} (ng/mL)	13.96+8.78 (62.95)	4.76+2.54 (53.45)	14.05+7.92 (56.40)	4.22+2.33(55.11)
AUC_{0-t} (h.ng/mL)	120.20+74.31 (61.82)	105.99+52.98 (49.99)	126.42+62.74 (49.63)	101.34+45.95 (45.34)
AUC_{0-inf} (h.ng/mL)	131.72+81.99 (62.25)	120.37+60.18 (49.99)[b]	136.32+64.00 (46.95)	117.83+57.34 (48.66)
$t_{1/2}$ (h)	13.14+14.20 (108.09)	26.92+17.40 (64.63)[b]	11.43+7.33 (64.12)	26.11+16.86 (64.58)
T_{max} (h)	4.21+1.23 (29.21)	8.23+4.27 (51.92)	4.50+1.58 (35.25)	7.14+3.81(53.34)

Note: Data are expressed as arithmetic mean ± standard deviation (CV %); [a] one subject was excluded because during period 1 presented diarrhea; [b] one was excluded because the subject only had two points after C_{max}. PK, pharmacokinetic; FDC, Fixed-Dose Combination; C_{max}, maximum plasma concentration; T_{max}, time to reach C_{max}; AUC_{0-t}, area under the plasma concentration-time curve from time zero to the last sampling time; AUC_{0-inf}, area under the curve from zero to infinity; $t_{1/2}$, terminal elimination half-life.

Table 3: Geometric mean ratio (90% CI) of the PK parameters of rosuvastatin and ezetimibe following administration of the FDC tablet containing 20 mg rosuvastatin and 10 mg ezetimibe or administration of the same dose as monotherapy in healthy Mexican subjects.

API	PK parameter	Geometric mean ratio (90% CI)	Intra-individual CV (%)
Rosuvastatin	C_{max}	1.032 (0.937–1.138)	23.13
	AUC_{0-t}	1.082 (1.005–1.175)	18.45
	$(AUC)_{0-inf}$	1.090 (0.998–1.190)	20.87
Ezetimibe	C_{max}	0.897 (0.829–0.971)	18.96
	AUC_{0-t}	0.976 (0.900–1.057)	19.42
	AUC_{0-inf}	0.993 (0.916–1.076)	19.17

Note: API: Active Pharmaceutical Ingredient; PK: pharmacokinetic; FDC: Fixed-Dose Combination; C_{max}: maximum plasma concentration; AUC_{0-t}: area under the plasma concentration-time curve from time zero to the last sampling time; CI: Confidence Interval; CV: Coefficient of Variation

Table 4: Summary of Adverse Events (AEs).

	Rosuvastatin	Ezetimibe	Rosuvastatin+Ezetimibe, FDC	Total AEs
Headache[a]	1 (2)	1 (1)	0	3
Diarrhea[a]	1 (1)	0	0	1
Upper respiratory infection[a]	1 (1)	0	0	1
Nausea[a]	1 (2)	0	1 (1)	3
Total AEs	6 (75%)	1 (12.5%)	1 (12.5%)	8 (100%)
Number of subjects presented AEs	4 (3 female, 1 male)	1 (female)	1 (female)	

Note: [a] Data are presented as number of subjects (number of adverse events)

DISCUSSION

This study evaluated the PK and tolerability profile of a FDC formulation containing 20 mg rosuvastatin and 10 mg ezetimibe, compared to the same doses of both drugs given separately to determine the potential PKs interactions. The 90% CIs for GMR of the PK parameters C_{max} and AUC_{0-t} were within the acceptable limits of bioequivalence (0.8-1.25), demonstrating that the formulation of the FDC was bioequivalent to monotherapy. Additionally, no significant clinical differences were observed among the AEs that followed the administration of each formulation.

The efficacy and safety of rosuvastatin/ezetimibe in FDC were evaluated in the LANCE study. It was a non-inferiority trial of rosuvastatin/ezetimibe versus simvastatin/ezetimibe in the control of LDL cholesterol levels in Brazilian patients diagnosed with primary hypercholesterolemia or mixed dyslipidemia. The results showed a higher proportion of patients that achieved <100 mg/dL of LDL-c in the group treated with rosuvastatin/ezetimibe (week 4: 84.8% vs. 68.2%; p=0.0257). At the end of the study, the proportions were 81.8% and 73.0%, respectively, without a statistically significant difference between the groups (p=0.2313) [8].

In 2013, the CARMELA Study (Cardiovascular Risk Factors Multiple Evaluation in Latin-America) presented results regarding the prevalence of dyslipidemia in Mexico City and its relationship with other cardiovascular risk factors, finding a cholesterol prevalence of ≥ 240 mg/dl of 16.4%, while 34.1% had values from 200 to 240 mg/dl, in addition to very high triglyceride levels were recorded in 29.9% of the study participants [8]. Combination therapy is recommended if the target LDL (Low-Density Lipoprotein) is not reached after monotherapy [9]. Kim H, et al conducted their study with 24 healthy male volunteers and concluded that according to the results its necessary larger sample size when comparing a fixed-dose combination product with separate tablets [10]. The sample size for our study was calculated using the intra-individual variation information of ezetimibe C_{max} from another study, and in our study was lower (18.96%) with a statistical power >90%. Therefore, the 33 subjects that completed the study were enough to compare the PK profiles of the FDC and separate administration of rosuvastatin and ezetimibe.

Our data are consistent with the results reported by Kosoglou, et al. and Kim, et al. in steady-state pharmacokinetic [5, 10].

CONCLUSION

The administration of Trezete® was bioequivalent to concurrent administration of the corresponding individual tablets as GMR with 90% CI of C_{max}, AUC_{0-t} and AUC_{0-inf} of rosuvastatin and ezetimibe were within conventionally accepted bioequivalence criteria. The corresponding tolerability profiles were also similar; therefore the FDC was well tolerated.

AUTHOR CONTRIBUTIONS

Author contributions to this article are as follows: Study investigator: L.J.G.A; manuscripts preparation: J.G.C, L.L.S. data interpretation, manuscript review and revisions, and final approval of manuscript: All authors. J.G.C, Y.R.A, and L.L.S.

FUNDING

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INSTITUTIONAL REVIEW BOARD STATEMENT

The study protocol and informed consent form were approved by an Independent Ethics Committee, a Research Committee (protocol number: CEI_024_0417BD_187s) and the Ministry of Health in Mexico (COFEPRIS).

OTHER REGISTRATIONS

National Clinical Trials Registry (RNEC by its Spanish acronym): 173300410B0163.

CLINICAL TRIALS PROTOCOL REGISTRATION

NCT04895059.

INFORMED CONSENT STATEMENT

All subjects provided written informed consent before participating in this study.

DATA AVAILABILITY STATEMENT

The authors confirm that the data supporting the findings of this study are available from the corresponding author, upon reasonable request.

CONFLICTS OF INTEREST

J.G.C, Y.R.A and L.L.S are employees of Laboratorios Silanes, S.A. de C.V, and L.J.G.A is employee of Investigación Farmacológica y Biofarmacéutica (IFaB).

REFERENCES

1. Adams SP, Sekhon SS, Wright JM. Rosuvastatin for lowering lipids. Cochrane Database Syst Rev. 2014;11.
2. White CM. A review of the pharmacologic and pharmacokinetic aspects of rosuvastatin. J Clin Pharmacol. 2002; 42 (9): 963-970.
3. Martin PD, Dane AL, Schneck DW, Warwick MJ. An open-label, randomized, three-way crossover trial of the effects of coadministration of rosuvastatin and fenofibrate on the pharmacokinetic properties of rosuvastatin and fenofibric acid in healthy male volunteers. Clin Ther. 2003; 25 (2): 459-471.
4. Toth P, Phan, Dayspring T. Ezetimibe therapy: Mechanism of action and clinical update. Vasc Health Risk Manag. 2012; 8: 415-427.
5. Kosoglou T, Statkevich P, Johnson-Levonas AO, Paolini JF, Bergman AJ, Alton KB. Ezetimibe: A review of its metabolism, pharmacokinetics and drug interactions. Clin Pharmacokinet. 2005; 44 (5): 467-494.
6. Zhan S, Tang M, Liu F, Xia P, Shu M, Wu X. Ezetimibe for the prevention of cardiovascular disease and all-cause mortality events. Int Psychogeriatr. 2018; 11 (11): CD012502.
7. Escobedo-de la Peña J, de Jesús-Pérez R, Schargrodsky H, Champagne B. Prevalencia de dislipidemias en la ciudad de México y su asociación con otros factores de riesgo cardiovascular. Resultados del estudio CARMELA. Am J Geriatr Psychiatr. 2014; 150: 128-136.
8. Aché L. Informe final estadístico estudio. Lancet. 2015.
9. Ballantyne CM, Weiss R, Moccetti T, Vogt A, Eber B, Sosef F, et al. Efficacy and safety of rosuvastatin 40 mg alone or in combination with ezetimibe in patients at high risk of cardiovascular disease (Results from the EXPLORER study). Am J Cardiol. 2007; 99 (5): 673-680.
10. Kim H, Choi HY, Kim Y-H, Bae K-S, Jung J, Son H, et al. Pharmacokinetic interactions and tolerability of rosuvastatin and ezetimibe: An open-label, randomized, multiple-dose, crossover study in healthy male volunteers. Drug Des Devel Ther. 2018; 12: 815-821.