Comparative Pharmacokinetics and Bioequivalence of Two 50 mg Atenolol Tablet Formulations in Healthy Korean Male Volunteers

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Key words
- atenolol
- bioequivalence study
- volunteers

Abstract

Atenolol is a selective β1 receptor antagonist that is available as a racemic mixture. The objective of this study was to compare the pharmacokinetics and evaluate the bioequivalence of 50 mg atenolol test and reference formulations in 24 healthy Korean male volunteers. This study was a single-dose, randomized, open-label, 2 period crossover study. 24 healthy Korean male volunteers randomly received 50 mg of either test or reference atenolol formulations in a 2 × 2 crossover study. There was a 1 week washout period between doses. The area under the curve (AUC)0–24 h and Cmax of 50 mg atenolol were the primary criteria for evaluation of bioequivalence. The mean ± standard deviation (SD) values of the Cmax, Tmax, AUC0–24 h, AUC0–∞, k-e, and t1/2 of the test and reference formulations were 268.4 (78.96) and 256.9 (79.34), 2,750 (0.9555) and 3,104 (1.053), 1,981 (729.2) and 1,872 (604.8), 2,228 (697.1) and 2,187 (628.5), 0.1332 (0.02748) and 0.1421 (0.04223), 5.419 (1.110) and 5.442 (2.357), respectively. The 90% confidence intervals for AUC0–24 h and Cmax were 0.9037–1.166 and 0.9169–1.1987, respectively. These results were within the accepted bioequivalence range of 0.80–1.25, which satisfied the bioequivalence criteria of the European Committee for Proprietary Medicinal Products and the US Food and Drug Administration guidelines. In conclusion, the findings of this study indicate that the 2 formulations of 50 mg atenolol that were tested are bioequivalent. Therefore, these formulations may be prescribed interchangeably.

Introduction

Atenolol is a selective β1 receptor antagonist without intrinsic partial agonist or membrane-stabilizing activities. Atenolol is commercially available as a racemic mixture. The (−)-S-form is the active isomer, whereas the R(+) isomer has no significant pharmacological activity [1,2]. Importantly, it is widely used to treat hypertension, angina acute myocardial infarction, supraventricular tachycardia, ventricular tachycardia, and congestive heart failure [3]. The onset of atenolol starts 2–4 h after oral administration. The absorption of atenolol has been shown to be rapid and consistent but incomplete, with a bioavailability of 50% [4]. Approximately 6–16% of atenolol is bound to plasma protein [3], and very little atenolol is metabolized by the liver. Instead, approximately 40–50% of non-metabolized atenolol is excreted in urine after administration [3,5]. The objective of this study was to compare the pharmacokinetics and bioequivalence of 50 mg atenolol test drug manufactured by Hana Pharmaceuticals (Seoul, Korea; Hana Atenolol®) with 50 mg atenolol reference drug manufactured by Hyundai Pharmaceuticals (Seoul, Korea; Tenormin®) in 24 healthy Korean male volunteers.

Materials and Methods

Test and reference medications

The test medication (Hana Atenolol, 50 mg, lot No. 6003, Hana Pharmaceuticals) and the reference medication (Hyundai Tenormin, 50 mg, lot No. 66042, Hyundai Pharmaceuticals) were supplied as tablets.

Subjects and methods

This study was a single-dose, randomized, open-label, 2 period crossover study. The study was...
conducted at Bestian Hospital in Seoul, Korea. The protocol of this study was approved by the Research Institute of Pharmaceutical Sciences, College of Pharmacy, Seoul National University (Seoul, Korea) and performed according to the rules of Good Clinical Practice and in accordance with the Declaration of Helsinki. All participants provided a written informed consent after they had been informed of the nature and details of the study in accordance with Korean Guidelines for Bioequivalence Tests [KGBT 2006].

24 healthy male Korean volunteers, ranging in age from 20 to 28 years (median: 24), weight from 58 to 95 kg (71.0 ± 9.15 kg), and height from 168 to 183 cm (176 ± 1.47 cm) completed the study. Volunteers were selected after passing a clinical screening procedure. The screen consisted of a physical examination and laboratory tests, including a blood analysis of hemoglobin, hematocrit, white blood cell (WBC), platelets, differential counting of WBC, blood urea nitrogen, serum creatinine, total protein, albumin, alanine aminotransferase (ALT), aspartate aminotransferase (AST), total bilirubin, cholesterol, glucose fasting, and alkaline phosphatase. In addition, a urine analysis was conducted for specific gravity, color, pH, glucose, protein, bilirubin, red blood cells (RBC), and WBC assessment. Volunteers were excluded if they were possibly sensitive to this type of medication, had a history of any illness of hepatic, renal, or cardiovascular systems, or had taken alcohol or other medications that may affect drug metabolism over a long period of time. These exclusion criteria were used to ensure that any illness or other medication would not induce variation in assessment. All subjects avoided using other drugs for at least 1 month before the study and until study completion. Patients were also required to refrain from consuming alcoholic beverages and xanthine-containing foods and beverages 48 h prior to each dosing and until collection of the last blood sample. The volunteers were randomly assigned to 2 groups and received an oral dose of 50 mg of metoprolol, 500 μg aliquot of 0.5N NaOH, and 5 mL aliquot of ether:dichloromethane (70:30 v/v) were added to a 1 mL aliquot of plasma sample. After 10 min of extraction, the tubes were centrifuged at 5400 × g for 10 min. The organic layer was then collected and dried under a gentle stream of nitrogen gas at 40 °C. A 1.25 μL aliquot of the mobile phase was added to reconstitute the residue and a 50 μL aliquot was injected directly onto a reverse phase HPLC column. The mobile phase consisted of acetonitrile:methanol:10 mM KH2PO4 (pH 3.0) (15:15:70, v/v) and was processed at a flow rate of 1.0 mL/min. The column effluent was monitored using a fluorescence detector with excitation and emission wavelengths set at 222 nm and 300 nm, respectively.

Pharmacokinetic analysis

Standard methods [6] were used to calculate the pharmacokinetic parameters using a non-compartmental analysis (WinNonlin®; Pharsight Corporation, Mountain View, CA). The Cmax and Tmax were compiled from the concentration-time data. The area under the curve (AUC0–24h) was calculated using a linear-log trapezoidal formula up to the last measured time at 24 h in plasma, and the AUC0–∞ was calculated by extrapolation [7]. The terminal elimination rate constant, k_e, was defined as the terminal elimination rate constant estimated by log-linear regression analysis on data visually assessed to be a terminal log-linear phase. The apparent terminal elimination t1/2 was calculated as follows:

\[ t_{1/2} = 0.693/k_e \]

Statistical analysis of data

Analysis of variance (ANOVA) was performed using logarithmically transformed AUC0–24h and Cmax was used to assess group or sequence, period, subjects per group, and drug effects. The Schuermann’s 3-sided t-tests were conducted to test the bioequivalence of the pharmacokinetic characteristics of the 2 formulations [8]. The range of bioequivalence for parametric analysis was set to the commonly accepted 80–125 % of the test/reference ratios of AUC0–24h and Cmax according to Korean Guidelines for Bioequivalence Tests [KGBT 2006]. All statistical comparisons were made using the Equiv test and confirmed with the K–BE test program [9].

Results

Both atenolol formulations were well-tolerated by the subjects of this study. Clinically relevant drug-related side effects were not observed in any of the 24 volunteers, and no volunteer withdrew from the study.

A representative chromatogram of blank plasma spiked with internal standard (metoprolol) and the chromatogram of a plasma sample are shown in Fig. 1. In this HPLC method, no interference from endogenous substances was observed in human plasma. This method, with slight modification, was able to determine the atenolol plasma concentration within 5 min for each plasma sample. The retention times for atenolol and internal standard were approximately 0.7 and 2.8 min, respectively. The detection limit for atenolol in human plasma was 20 ng/mL based on a signal-to-noise ratio of 10. The standard curve was achieved from the least squared regression equation: \( y = 0.0007x + 0.0068 \) (\( r^2 = 0.9983 \)). The intra- and inter-day precision coefficients of variation for human plasma were 3.839–9.274 % and 7.475–10.19 %, respectively, and the intra-and inter-day accuracy coefficients of variation for human plasma were 95.66–105.2 % and 97.91–108.8 %, respectively, from atenolol plasma concentrations in the range of 20–1000 ng/mL.
Table 2 Standard bioequivalence analysis for log-transformed AUC_{0–24 h} (ng/mL × h) and log-transformed C_{max} (ng/mL) based on multiplicative model after a single-dose administration of 2 formulations of atenolol 50 mg tablets in healthy Korean male volunteers. AUC_{0–24 h} (ng/mL × h) and C_{max} (ng/mL) were also analyzed based on additive model for completeness.

<table>
<thead>
<tr>
<th>Parameter</th>
<th>Test drug*</th>
<th>Reference drug*</th>
<th>90% Confidence Interval</th>
</tr>
</thead>
<tbody>
<tr>
<td>AUC_{0–24 h}</td>
<td>1.026</td>
<td>0.9037–1.166</td>
<td></td>
</tr>
<tr>
<td>C_{max}</td>
<td>1.048</td>
<td>0.9169–1.1987</td>
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</tbody>
</table>

Table 1 Pharmacokinetic parameters of atenolol 50 mg after a single oral dose of test and reference formulations of atenolol 50 mg tablets in healthy Korean male volunteers.

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<tr>
<td>C_{max} (ng/mL)</td>
<td>268.4 (78.96)</td>
<td>256.9 (79.34)</td>
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<td>T_{max} (h)</td>
<td>2.750 (0.9555)</td>
<td>3.104 (2.357)</td>
</tr>
<tr>
<td>AUC_{0–24 h}</td>
<td>1981 (729.2)</td>
<td>1872 (604.8)</td>
</tr>
<tr>
<td>AUC_{0–∞} (ng/mL × h)</td>
<td>2228 (697.1)</td>
<td>2187 (628.5)</td>
</tr>
<tr>
<td>k_{e} (h⁻¹)</td>
<td>0.1332 (0.02748)</td>
<td>0.1421 (0.04223)</td>
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<tr>
<td>t_{1/2} (h)</td>
<td>5.419 (1.110)</td>
<td>5.442 (2.357)</td>
</tr>
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Data are expressed as the mean ± standard deviation (SD).

*atenolol 50 mg test drug manufactured by Hana Pharmaceuticals (Seoul, Korea) (Hana Atenolol®); **atenolol 50 mg reference drug manufactured by Hyundai Pharmaceuticals (Seoul, Korea) (Tenormin®) in 24 healthy Korean male volunteers

The mean plasma atenolol concentration-time profiles are shown in Fig. 2. We found that the mean plasma atenolol concentration profiles of the test and reference formulations of atenolol 50 mg tablets in healthy Korean male volunteers are shown in Fig. 2. We found that the mean plasma atenolol concentration profiles of the 2 formulations were almost identical (Fig. 2). The mean extrapolated section of AUC from the last measurable point was 14.78%. The mean pharmacokinetic parameters, such as C_{max}, T_{max}, AUC_{0–24 h}, AUC_{0–∞}, k_{e}, and t_{1/2} after the administration of test and reference atenolol formulations are listed in Table 1. The 90% confidence intervals of AUC_{0–24 h} and C_{max} are presented in Table 2. The pharmacokinetic parameters, including C_{max}, T_{max}, AUC_{0–24 h}, AUC_{0–∞}, k_{e}, and t_{1/2}, were comparable between the 2 atenolol formulations and no statistical difference was found. Importantly, our findings in this study were similar to data previously reported [4, 5, 10].

The ANOVA test showed that the F-test values were lower than the US Food and Drug Administration guidelines. The pharmacokinetic parameters, including C_{max}, T_{max}, AUC_{0–24 h}, AUC_{0–∞}, k_{e}, and t_{1/2} were comparable between the 2 atenolol formulations and no statistical difference was found. Importantly, our findings in this study were similar to data previously reported [4, 5, 10]. The ANOVA test showed that the F-test values were lower than the US Food and Drug Administration guidelines. The pharmacokinetic parameters, including C_{max}, T_{max}, AUC_{0–24 h}, AUC_{0–∞}, k_{e}, and t_{1/2} were comparable between the 2 atenolol formulations and no statistical difference was found. Importantly, our findings in this study were similar to data previously reported [4, 5, 10].

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Acknowledgement

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Conflict of Interest

The authors declare no conflicts of interest with respect to this paper.

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