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Original article

BIOEQUIVALENCE STUDY OF GENERIC FINASTERIDE IN HEALTHY MALE VOLUNTEERS

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Abstract The bioequivalence of 5-mg of the generic finasteride tablet, as a test, and the original finesteride tablet, as a reference products, were evaluated. The two products were administered to 12 healthy Thai male volunteers as a single oral dose according to a randomized two-way crossover design. The washout period was 1 week. After drug administration, serial blood samples were collected over a period of 30 hours. Plasma finasteride concentrations were measured by HPLC coupled with UV detection. The pharmacokinetic parameters were analyzed by noncompartmental analysis. The maximum finasteride concentrations (C_{max}, ng/mL) and the median time to reach the C_{max} ($T_{max,}$ hr) for the test and reference were 34.05 (range 26.5-47.49) and 34.39 (23.79-45.96), and 2.25 (0.5-4.0) and 2.50 (1.0-2.5), respectively. Analysis of variance for bioequivalence revealed the mean (90% CI) of the $AUC_{0-\infty}$ and C_{max} ratios [for Test /Reference] of 0.98 (0.81-1.17) and 0.99 (0.89-1.10), respectively. These values were within the bioequivalence range of 0.80-1.25, thus, our study demonstrated the bioequivalence of the two products. **Chiang Mai Med Bull 2003;42(4):131-137.**

Keywords: finasteride, bioequivalence

Finasteride (Proscar®) is a synthetic 4-azasteroid prescribed for the treatment of benign prostatic hypertrophy (BPH).[1] The mechanism of action involves inhibi tion of 5 alpha-reductase, which metabolizes testosterone to the more potent androgen, dihydrotestosterone (DHT).[2-4] Deprivation of DHT in the prostate results in a marked regression of the prostate volume and decrease symptoms associated with urinary tract obstruction.[3,5] Since circulating levels of testosterone are not affected, the desired androgen mediated effects on muscle strength, bone density and sexual function are thus preserved.[6] Treatment with finasteride for four years among men with symptoms of urinary obstruction

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and prostatic enlargement can maintain the control of BPH, while decreasing disease progression and significantly reducing the probability of surgery and acute urinary retention.[7] Finasteride may also be used to prevent hair loss in younger man.[8]

Finasteride is well absorbed and widely distributed after oral administration. It undergoes extensive hepatic metabolism to inactive metabolites, which are eliminated through the bile and urine.[4] Its mean bioavailability is 63%, (range from 33 to 108%).[9] Maximum plasma concentration averages 37 ng/mL (range 27 to 49 ng/mL) and is reached at 1 to 2 hours postdose.[9]

The aim of this study was to determine the pharmacokinetics of finasteride in 12 healthy volunteers after single oral doses of 5 mg Proscar® and the generic finasteride in order to obtain the bioequivalence approval.

Subject, materials and methods

Drugs used: Proscar® at 5 mg (Merck Sharp & Dohme, Australia) Lot No. A 6454 was used as a reference product and Harifin® at 5 mg (the T.O. Chemical 1979 Ltd., Bangkok, Thailand) Lot No. HAR5-01 was used as a test product.

Subjects

Twelve healthy Thai male volunteers aged between 32-47 years old, with a body mass index between 18-24 participated in this study. The subjects were free from medical illness judging from a physical examination and routine blood test. Cigarette smokers, alcohol consumers as well as subjects currently taking any drug known to induce or inhibit hepatic metabolizing enzyme were excluded from the study. All subjects signed a written informed consent before participating in the study.

Method of drug administration

This was a randomized, double-blind, 2-period crossover study. Each subject was randomly assigned to receive a single 5 mg dose of finasteride orally in the morning after an overnight fast. Subjects continued fasting for at least 2 hours after drug administration. Water and lunch were served at 2 hours and 4 hours after dosing. Blood samples were collected immediately before dose administration and thereafter at 0.5, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 15, 24 and 30 hr. The washout period was 1 week and the subjects were crossed - over to receive the other preparation in the same manner.

Determination of the plasma finasteride concentrations[10-11]

Finasteride in plasma was quantified by high performance liquid chromatography (HPLC) with UV detection (220 nm) after C8 solid phase extraction (Sep-Pak®,1 mL, 100 mg, Waters Corporation, MA, USA) and separation on C18 (Inersil®, 150 x 4.6 mm, 5 μm, GL Sciences Inc., Tokyo Japan) at 25 ^{o}C. The mobile phase was a mixture of 15 mM Phosphate buffer (pH 3.5)/Acetonitrile/Tetrahydofuran (50/39/1, v/v/v). The retention times for finasteride and internal standard were approximately 10.27 and 13.26 minutes, respectively. Solutions of finasteride that ranged from 2-50 ng/mL were prepared in plasma to

establish the calibration curve for the validation assay. Linear regression analysis of the peak-height ratios of finasteride/internal standard (IS) versus finasteride concentrations consistency gave determinant (R^2) coefficients of 0.999 or better. Finasteride concentration were quantified from the calibration standard lines with the use of linear regression. The method was validated using 4 sets of 5 control samples (12 samples) from each of 3 different concentrations (7.5, 15, 30 ng/mL) of quality control (QC) samples, and a single calibration curve ran concurrently for within-day accuracy and precision. For inter-day assay precision, 5 sets of three concentrations of QC samples were studied on 4 independent days with 4 concurrent standard calibration curves. The average %CV for within-day and inter-day assays was 6.8% and 10.02%, respectively. The lower limit of quantitative analysis (LLQ) was 2 ng/mL (%CV = 14.7) and the mean recovery of finasteride determined from 5 aliquots of each QC sample was 85.3%.

Pharmacokinetic analysis

Maximal plasma concentration ($C_{max,}$ ng/mL) and time to reach the peak concentration ($T_{max,}$ hr) were obtained directly by visual inspection of each subject's plasma concentration-time profile. The area under the plasma concentration-time curve (AUC) from time 0-infinity ($AUC_{0-\infty}$, ng*hr/mL) to half-life ($t_{1/2}$, hr) was determined by non-compartmental analysis. The slope of the terminal log-linear portion of the concentration-time profile was determined by least-squares regression analysis and used as the elimination rate constant (K_e). The elimination half-life was calculated as $0.693/K_e$. The AUC_{0-t} from time zero to the last quantifiable point (Ct) was calculated using the trapezoidal rule, and extrapolated AUC from Ct to infinity ($AUC_{t-\infty}$) was determined as Ct/Ke. Total $AUC_{0-\infty}$ was the sum of AUC_{0-t} + $AUC_{t-\infty}$. The calculation was performed by using the TopFit, pharmacokinetic data analysis program for PC.

Statistical analysis[12-13]

An analysis of variance (ANOVA) was performed to determine the statistical differences of pharmacokinetic parameters ($AUC_{0-\infty,}$ $C_{max,}$ and T_{max}), which represented the extent and rate of drug absorption. Statistical analysis of AUC and C_{max} was performed on logarithmically (ln) transformed data. The 90% confidence intervals for the ratio of AUC as well as C_{max} values of the test preparation over those of the reference product were estimated using the following equation:

$$90\%\ CI\ (\mu_T - \mu_R) = (\bar{X}_T - \bar{X}_R) \pm t^{v}_{0.1} \sqrt{\frac{2S^2}{n}}$$

- $\bar{X}_T$ and $\bar{X}_R$ are the observed mean of the (ln) transformed parameters (either C_{max} or AUC) for the test (T) and reference products (R).
- S^2 is obtained from the analysis of variance.
- n is the number of subjects.
- $t^{v}_{0.1}$ is the tabulated two-tail t value for 90 % CI. v is the number of degrees of freedom of the error mean square.

The antilogarithm of the confidence interval ($\mu_T - \mu_R$) expressed the bioequivalence as a ratio of the test and reference product [μ_T/μ_R].

The bioequivalence intervals of 0.8-1.25 for the ratio [$\frac{\text{Test}}{\text{Reference}}$] of the average $AUC_{0-\infty}$ and C_{max} were accepted by the Thai FDA. Regarding analysis of T_{max}, the limits for the bioequivalence range were expressed as untransformed data (absolute differences) and the accepted stipulated bioequivalence range of T_{max} difference [Test-Reference] was± 20% of the T_{max} of the reference formulation.

Result and discussion

All subjects completed the study with out any adverse effects. Fig. 1 shows that the mean plasma concentration-time curves of the reference and test were comparable, although the peak finasteride concentration of the reference was slightly higher than that of the test.

Table 1 compared the mean values of pharmacokinetic parameters (C_{max}, T_{max}, $AUC_{0-\infty}$ and $t_{1/2}$) of Proscar® and the test product. Following a single oral dose, the median time to reach the maximum concentration (T_{max}) for the test (2.25 hr, range 0.5-4.0 hr) was faster than that for the reference (2.5 hr, range 1.0-2.5 hr). The 90% CI for the T_{max} difference (μ_T-μ_R) ranged from -0.58 to 0.5 hour, which was outside the stipulated bioequivalence range of ± 0.42 hour, hence, equivalence with respect to the T_{max} could not be concluded. In spite of this, the average (±SD) C_{max} and $AUC_{0-\infty}$ for the test were not significantly different from those for the reference (34.05±6.43 vs 34.39±6.46 ng/mL, and 299.09±79.11 vs 299.94±

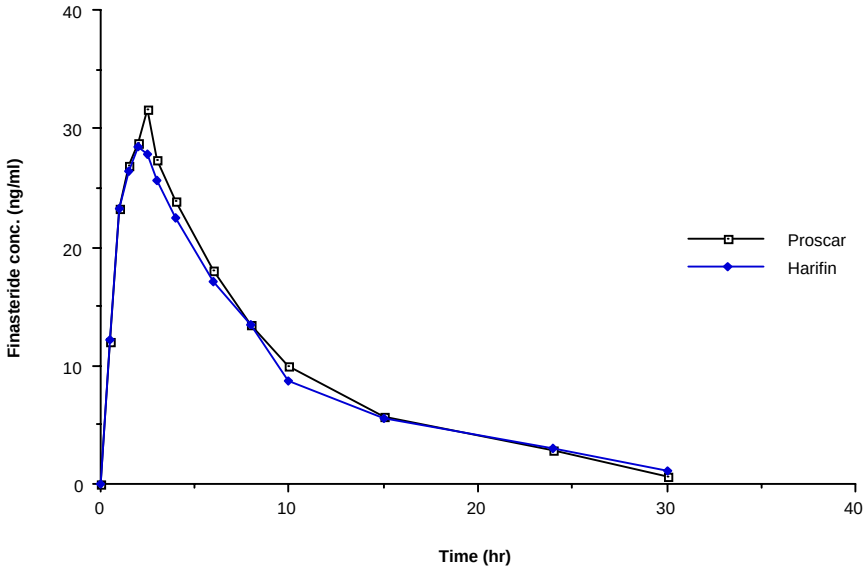

Figure 1. Mean plasma concentration-time profiles after single oral administration of 5 mg finasteride [Reference (- -), Test (-♦-)].

Table 1. Comparison of finasteride pharmacokinetic parameters after oral administration of 5 mg for the test (T) and reference (R).

Pharmacokinetic parameters	T	R
T_{max} (hr) (mean±SD)	2.04±0.92	2.08±0.56
Range	0.50-4.00	1.00-2.50
Median	2.25	2.50
C_{max} (ng/mL) (mean±SD)	34.05±6.43	34.39±6.46
Range	26.50-47.49	23.79-45.96
AUC (ng.hr/mL) (mean±SD)	299.09±79.11	299.94±54.18
Range	158.40-476.40	230.62-391.33
$T_{1/2}$(hr) (mean±SD)	8.01±3.42	7.12±1.64
Range	2.71-16.60	4.02-9.79

Table 2. Parametric 90% CI of the mean pharmacokinetic parameters ($AUC_{0-\infty}$, C_{max} and T_{max}) of the test/ reference.

PK parameters	Mean	90% CI	Acceptable range
$AUC_{0-\infty}$ $(\frac{\text{Test}}{\text{Reference}})$	0.98	0.81 – 1.17	0.80 – 1.25
C_{max} $(\frac{\text{Test}}{\text{Reference}})$	0.99	0.89 – 1.10	0.80 – 1.25
T_{max} (Test – Reference)	-0.04	(-0.58) – 0.50	$\pm$ 0.42

54.18 ng.hr/ mL). The C_{max} of finasteride obtained from this study was comparable to those values reported in the literature [average C_{max} 37 (range 27-49 ng/mL)].[9] Furthermore, the mean elimination half-lives ($t_{1/2}$, hr) of the test [8.01±3.42 (range 2.71–16.6)] and reference [7.12± 1.64 (range 4.02–9.79)] were similar to the values reported in the literature(3-16 hr),[9] with no statistical difference between the two preparations. The relative bioavailability (F_{rel}) calculated from C_{max} and $AUC_{o-\infty}$ of the Test/ Reference was 101.58 % and 103.31 %, respectively. Bioequivalence analysis (Table 2) showed that the mean (90% CI) of the C_{max} and $AUC_{o-\infty}$ ratios for the Test/ Reference were 0.99 (0.89-1.10) and 0.98 (0.81–1.17), respectively. As these were well within the bioequivalence range of 0.8-1.25, our study demonstrated the bioequivalence of the test and reference.

Conclusion

We conducted a bioequivalence study in 12 healthy Thai male volunteers with 5 mg of oral formulations of the generic finasteride. The results showed that both formulations were well tolerated. The pharmacokinetic parameters (C_{max} and $T_{1/2}$) of finasteride obtained from the study were comparable to those values reported in the literature. We also demonstrated the bioequivalence of the two

products concerning the rate (C_{max},) and extent ($AUC_{0-\infty}$) of absorption based on the 90% CI, which was well within the acceptable range of standard Thai FDA guidelines.

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การทดสอบชีวสมมูลของยาฟีเนสเทอไรด์เปรียบเทียบกับยาต้นแบบในอาสาสมัครสุขภาพดี

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บทคัดย่อ การศึกษานี้มีวัตถุประสงค์เพื่อทดสอบชีวสมมูลของยาเม็ดฟีเนสเทอไรด์ขนาด 5 มิลลิกรัม โดยให้ยาทดสอบฟีเนสเทอไรด์ที่ผลิตในประเทศไทย และยาต้นแบบซึ่งศึกษาแบบสุ่มไขว้ ครั้งเดียว ระยะเวลาการศึกษาห่างกัน 1 สัปดาห์ในอาสาสมัครชายไทยสุขภาพดีจำนวน 12 ราย หลังจากให้ยานำตัวอย่างเลือดภายใน 30 ชั่วโมงไปตรวจวัดหาความเข้มข้นของยาฟีเนสเทอไรด์โดยวิธีโครมาโตกราฟฟีชนิดของเหลวสมรรถนะสูงและประเมินค่าทางเภสัชจลนศาสตร์โดยวิเคราะห์แบบ non compartment ผลการศึกษาพบว่าระดับยาสูงสุดในเลือดและค่ามีเดียนของเวลาที่ระดับยาในเลือดสูงสุดของยาทดสอบและยาต้นแบบเท่ากับ 34.05 (26.5-47.49) และ 34.39 (23.79-45.96) นาโนกรัม/มิลลิลิตร, 2.25 (0.5-4.0) และ 2.50 (1.0-2.5) ชั่วโมง ตามลำดับ การวิเคราะห์ชีวสมมูลโดยใช้อะโนวา พบว่าค่าเฉลี่ย (ช่วงความเชื่อมั่นร้อยละ 90) ของอัตราส่วน [ยาทดสอบ / ยาต้นแบบ] ของพื้นที่ใต้กราฟที่เวลา 0 ถึงอสงไขยและค่าความเข้มข้นสูงสุดของยาในเลือด มีค่าเท่ากับ 0.98 (0.81-1.17) และ 0.99 (0.89-1.10) ตามลำดับ ซึ่งอยู่ในช่วงที่ยอมรับคือ 0.80-1.25 ดังนั้นยาทั้งสองตำรับจึงมีชีวสมมูลซึ่งกันและกัน **เชียงใหม่เวชสาร 2546;42(4):131-137.**

คำสำคัญ : ฟีเนสเทอไรด์ ชีวสมมูล
