ORIGINAL ARTICLE

Comparative Bioavailability Study of Two Ketoconazole Tablet Preparations

K H Yuen, PhD, J W Wong, Bpharm, N Billa, Bpharm, W P Choy, BSc, T Julianto, Bpharm
School of Pharmaceutical Sciences, University of Science Malaysia, 11800 Penang

Summary

The bioavailability of a generic preparation of ketoconazole (Zorinax^{R} from Xepa-Soul Pattinson, Malaysia) was evaluated in comparison with the innovator product (Nizoral^{R} from Janssen Pharmaceutica, Switzerland). Eighteen healthy male volunteers participated in the study conducted according to a two-way crossover design. The bioavailability was compared using the parameters, total area under the plasma concentration-time curve ($AUC_{0-\alpha}$), peak plasma concentration (C_{max}) and time to reach peak plasma concentration (T_{max}). No statistically significant difference was observed between the values of the two products in all the three parameters. Moreover, the 90% confidence interval for the ratio of the logarithmic transformed $AUC_{0-\alpha}$ and C_{max} values of Zorinax^{R} over Nizoral^{R} was found to lie between 0.82 - 1.04 and 0.83 - 1.02, respectively, being within the acceptable equivalence limit of 0.80 - 1.25. These findings indicate that the two preparations are comparable in the extent and rate of absorption. In addition, the elimination rate constant (k_e) and apparent volume of distribution (V_d) were calculated. For both parameters, there was no statistically significant difference between the values obtained from the data of the two preparations. Moreover, the values are comparable to those reported in the literature.

Key Words: Bioequivalence - Ketoconazole - Generic

Introduction

Generic versions of popularly prescribed drugs usually become widely available, and often from more than one source, upon expiry of their patent. While generic preparations are generally cheaper and can help to lower medication costs, it is also known that excipients, unfavourable physico-chemical properties and other processing variables can markedly affect their bioavailability and hence efficacy. As such, a generic preparation should be proven to be bioequivalent to the innovator product if it is to be used as a substitute for the latter. Currently, there is a paucity of data on the bioequivalence of generic preparations, especially in the South East Asian region where bioequivalence assessment is not a regulatory requirement. Therefore, information obtained from such bioequivalence studies will aid a prescriber in the selection of a suitable product when several generic versions are available.

Ketoconazole is a new effective imidazole derivative which is active against both superficial and systemic fungal infections after oral administration[1]. However, ketoconazole possesses unfavourable physicochemical and pharmacokinetic properties such as low water solubility, erratic absorption from the gastro-intestinal tract as well as undergoing extensive and rapid metabolism in the liver, some possibly through first pass[2]. These properties warrants a bioequivalence study to be conducted on its generic preparations with reference to its original preparation to ensure that the bioavailability is within acceptable range.

In the present study, the bioavailability of a new tablet formulation of ketoconazole (Zorinax[R]) produced locally in Malaysia was investigated in comparison with the innovator product, Nizoral[R].

Materials and Methods

Products Studied

The ketoconazole preparations were:

- Nizoral[R] tablets, 200mg (Janssen Pharmaceutica, UK), batch no: 96104/170, manufacturing date: 09/1996, expiry date: 09/2001.
- Zorinax[R] tablets, 200mg (Xepa-Soul Pattison (M) Sdn Bhd), batch no: 47261, manufacturing date: 05/1998, expiry date: 05/2001.
- Ketoconazole reference standard were obtained from United States Pharmacopeia (MD, USA). All other reagents used were AR or HPLC grade.

In Vivo Study Design

The study was approved by a Joint School of Pharmaceutical Sciences, USM - General Hospital Penang Committee on Bioavailability studies. Eighteen healthy male volunteers of Asian origin between 20 and 47 years old and weighing from 59 to 85kg, participated in a standard 2 period, 2 sequence crossover study after providing written informed consent. All were judged to be healthy and were not receiving any medication during the study period. The volunteers were randomly divided into 2 groups of 9 each.

On the first trial period, each volunteer in group 1 was given one tablet of Nizoral[R], while those of group 2, one tablet of Zorinax[R]. After a washout period of one week, each volunteer then received the alternate product. All products were administered in the morning (10.00 a.m.) after an overnight fast with 150ml of water. Food was withheld for at least 2 hours after dosing. Lunch and dinner comprising chicken with rice, were served at 4 hours and 9 hours after dosing. Blood samples of 5ml volume were collected in vacutainers (containing sodium heparin as anticoagulant) at 0 (before dosing), 20min, 40min, 1, 1.5, 2, 2.5, 3, 4, 6, 8, 10, 14, 18 and 24 hours after dosing via an in-dwelling cannula placed in the forearm. The blood samples were centrifuged for 15min at 2000 G and the plasma transferred to separate glass containers to be kept frozen until analysis.

Analysis for Plasma Levels of Ketoconazole

Plasma level of ketoconazole was analyzed using a reversed-phase high performance liquid chromatography method employing fluorescence detection reported by Yuen and Peh[3].

Data Analysis

The two preparations were compared using the parameters, total area under the plasma concentration-time curve ($AUC_{0-\alpha}$), peak plasma concentration (C_{max}) and time to reach peak plasma concentration (T_{max}). The C_{max} and T_{max} were obtained directly from the plasma concentration data[4], while $AUC_{0-\alpha}$ was obtained by adding the area from time zero to the last sampling time (AUC_{0-t}) and the area from the last sampling to infinity ($AUC_{t-\alpha}$). The former was calculated using the trapezoidal formula, and the latter by dividing the last measurable plasma concentration with the elimination rate constant, k_e. In all cases, the $AUC_{t-\alpha}$ was found to be less than 10% of the $AUC_{0-\alpha}$. The k_e value was estimated from the terminal slope of the plasma concentration versus time plot through logarithmic transformation of the concentation values and application of linear regression[5]. In addition, the apparent volume of distribution (V_d) of the drug was also calculated as $Dose/(AUC_{0-\alpha} \cdot k_e)$.

Statistical Analysis

For each of the parameters, $AUC_{0-\alpha}$, C_{max}, k_e and V_d, the values obtained for the two products were analyzed statistically using an analysis of variance procedure (ANOVA) which distinguishes effects due to group, subjects/group, period, and treatment[6]. The $AUC_{0-\alpha}$ and C_{max} values were logarithmic transformed prior to the analysis. On the other hand, the T_{max} values of the two preparations were compared using the Wilcoxon signed-rank test for paired samples. A statistically significant

difference was considered at $p<0.05$. In addition, the 90% confidence interval of $AUC_{o-\alpha}$ as well as C_{max} for the ratio of test and reference was also determined based on a multiplicative model.

Results and Discussion

Figure 1 shows the mean plasma ketoconazole concentration versus time profiles of Nizoral[R] and Zorinax[R]. Both preparations produced a rapid increase in plasma concentration, reaching a peak at approximately 1.5 hours after dosing and declined rapidly thereafter, being typical for conventional immediate release preparations. Except for a slightly lower mean peak concentration, the plasma concentration profile of Zorinax[R] was essentially superimposable with that of Nizoral[R].

The individual numerical values of $AUC_{o-\alpha}$, C_{max} and Tmax are shown in Table I. The parameter T_{max} and $AUC_{o-\alpha}$ are related to the rate and extent of absorption, while C_{max} is related to both processes[7]. These three are the usual pharmacokinetic parameters used in determining the bioequivalence of generic preparations[8]. When the parameters were analyzed using the ANOVA procedure described previously, there was no statistically significant difference between the logarithmic transformed $AUC_{o-\alpha}$ ($p=0.2678$), as well as the logarithmic transformed C_{max} ($p=0.1775$) values of the two preparations. From the above analyses, it was also observed that the sequence (or group) effect was not statistically significant in both cases, suggesting that there was no significant treatment-by-period interaction. In addition, the 90% confidence interval for the ratio of the logarithmic transformed $AUC_{o-\alpha}$ values of Zorinax[R] over those of Nizoral[R] was estimated to be between 0.82 - 1.04, while that of C_{max} was between 0.83 - 1.25, both being within the acceptable bioequivalence interval of 0.80 - 1.25[9]. In the case of the parameter T_{max}, there was no statistically significant difference ($p>0.2$) between the values of the two preparations when analyzed using the Wilcoxon Signed Rank Test.

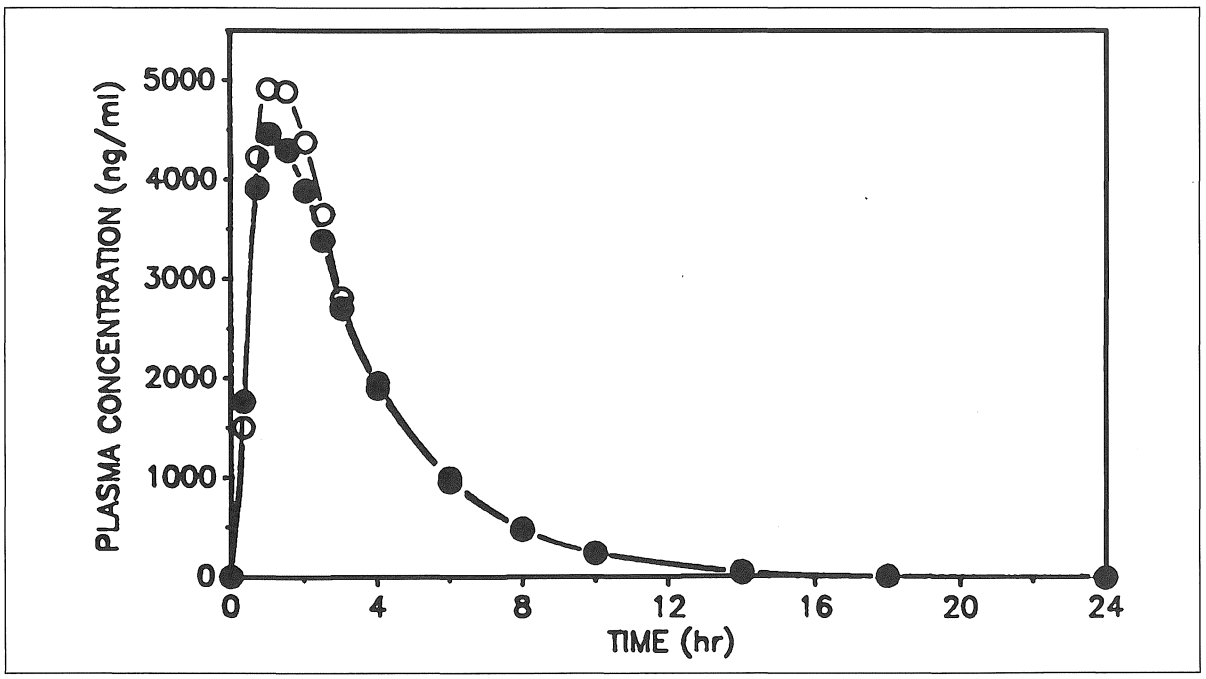

Fig. 1: Mean plasma ketoconazale concentration versus time profiles of Nizoral[R] (O) and Zorinax[R] (•). N=18.

Table I
Individual Numerical Values of C_{max}, T_{max} and $AUC_{0-\alpha}$ of Nizoral[R] and Zorinax[R]

Subject	Nizoral[R]			Zorinax[R]		
	C_{max} (ng/ml)	T_{max} (hr)	$AUC_{0-\alpha}$ (ng.hr/ml)	C_{max} (ng/ml)	T_{max} (hr)	$AUC_{0-\alpha}$ (ng.hr/ml)
1	4946.34	1.5	19248.02	4536.19	2.5	13624.25
2	6183.99	1.0	22322.04	5363.15	0.7	16302.50
3	5285.23	1.0	22168.89	3883.07	2.5	14206.98
4	4580.22	1.5	14627.82	4925.10	1.0	17689.71
5	3638.91	1.0	11154.21	4641.02	1.5	17663.73
6	4400.07	1.0	15494.47	2630.05	1.0	8379.17
7	5237.31	0.7	15614.42	5482.34	1.0	18540.80
8	6748.82	0.7	21533.69	6570.89	0.7	24075.38
9	6209.14	1.0	21766.09	6498.14	1.5	24636.52
10	6066.96	1.0	28531.54	6765.40	0.7	28081.47
11	5122.43	1.0	16958.81	6879.87	1.0	22862.17
12	4978.47	1.5	12579.89	2969.09	1.5	12357.26
13	7824.59	0.7	28392.07	5469.21	0.7	17031.11
14	4974.87	2.0	23903.46	6561.58	1.0	24917.20
15	5544.85	0.7	22212.66	3844.16	1.5	18372.43
16	6599.97	2.0	28010.02	6266.83	1.5	27646.58
17	4748.55	1.5	19169.21	3987.63	1.0	15291.82
18	3767.86	2.0	14205.98	3452.03	0.7	11998.77
Geometric Mean	5282.54	[a]1.0	19179.64	4847.75	[a]1.0	17680.63
Exp(Mean ±SD) ln data	4333.26 6439.79		14489.84 25387.34	3606.65 6515.93		12783.83 24453.12

[a]*Median*

The $AUC_{0-\alpha}$ values showed wide intersubject variation, as evidenced by the large standard deviation values shown in Table I and could be attributed to differences in body weight and drug disposition among the volunteers. In contrast, the intrasubject variation, estimated using the mean square error obtained from the ANOVA analysis[10] appeared to be relatively small, with a coefficient of variation value of 21.5%. Based on this value, the study which employed 18 volunteers has a test power (1-β) of approximately 80% for concluding that the two formulations are equivalent with respect to the $AUC_{0-\alpha}$ at a type 1 error rate (α) of 0.05, if the true difference is equal to or less than 20%[10]. In the case of the parameter C_{max}, the coefficient of variation was estimated to be 18.4%, thus a test power of greater than 80% was also attainable in the analysis of this parameter.

The mean numerical values of k_e and V_d, estimated from the plasma drug concentration profiles of Nizoral^R were 0.3783 hr^{-1} (SD, 0.0776) and 0.4023 liter/kg (SD, 0.1119), respectively, while those of the same parameters estimated from the plasma profiles of Zorinax^R were 0.3508 hr^{-1} (SD, 0.0958) and 0.5169 (SD, 0.3289). There was no statistically significant difference ($p>0.05$) between the values of the two products for both k_e and V_d. Moreover, the values which were obtained from volunteers of Asian origin are comparable to those reported in the literature[11,12]. According to the study by Schafer-Korting *et al.* (1984) and Daneshmend *et al.* (1981), the mean value of ke was 0.4227 hr^{-1} and 0.3466 hr^{-1} respectively, as compared to 0.3783 hr^{-1} obtained in the current study.

Conclusion

In summary, Zorinax^R was found to be comparable to Nizoral^R in both the rate and extent of absorption. The pharmacokinetic values obtained from the subjects of Asian origin are also comparable to those reported in the literature.

References

1. Heel RC, Brogden RN, Carmine A, Morley PA, Speight TM and Avery GS. Ketoconazole: review of its therapeutic efficacy in systemic fungal infection. Drugs 1982; 23: 1-36.
2. Martindale, The Extra Pharmacopoeia. The Pharmaceutical Press, London, 1989; 426-29.
3. Yuen KH and Peh KK. Simple high-performance liquid chromatographic method for determination of ketoconazole in human plasma. Journal of Chromatography B 1998; 715: 436-40.
4. Weiner DL. Design and analysis of bioavailability studies. In: Buncher CR and Tsay JY (eds). Statistics in the Pharmaceutical Industry. New York: Marcel Dekker, 1981: 205-29.
5. Gibaldi M and Perrier D. harmacokinetics. New York: Marcel Dekker, 1982.
6. Wagner JG. Fundamentals of Clinical Pharmacokinetics. Hamilton, IL: Drug Intelligence Publication 1975; 285-305.
7. Grahnen A. Design of bioavailability studies. Pharm Int 1984; 5: 100-3.
8. Chow SC and Liu JP. Recent statistical developments in bioequivalence trials - a review of the FDA guidance. Drug Infor J 1994; 28: 851-64.
9. USP 23 - NF 18, Supplement 5 The United States Pharmacopeial Convention, Inc, USA, 1996; 3490-531.
10. Diletti E, Hauschke D and Steinijans VW. Sample size determination for bioequivalence assessment by means of confidence intervals. Int J Clin Pharmacol Ther Toxicol 1991; 29: 1-8.
11. Daneshmend TK, Warnock DW, Turner A and Roberts CJC. Pharmacokinetics of ketoconazole in normal subjects. J Antimicrob Chemother 1981; 8: 299-304.
12. Schafer-Korting M, Korting HC, Dorn M and Mutschler E. Ketoconazole concentrations in human skin blister fluid and plasma. Int J Clin Pharmacol Ther Toxicol 1984; 22(7): 371-74.