ORIGINAL ARTICLE

COMPARATIVE PHARMACODYNAMIC BIO-EQUIVALENCE OF TWO ORALLY ADMINISTERED FORMULATIONS OF ISOSORBIDE DINITRATE IN HEALTHY HUMAN SUBJECTS UNDER FASTING CONDITIONS

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ABSTRACT

BACKGROUND: ***Medical profession has realized the problem of wide variations in the therapeutic effectiveness of various brands of oral formulations containing the same active ingredient in equal amounts. A large number of preparations of isosorbide dinitrate are commercially available on the Indian Market. Manufacturers claim that their products are bioequivalent to the innovator's formulation but it may or may not be. So, the only way to verify these claims is to do a comparative bioequivalence study with the innovator drug formulation using confidence intervals. Hence the present study was undertaken to compare the pharmacodynamic bioequivalence of the only two marketed brands of 10 mg isosorbide dinitrate tablets in India in healthy, adult, male, human subjects under fasting conditions.***

METHODS: ***The study was carried out as single dose, two treatment, two period, two-sequence crossover randomized trial on 8 healthy human subjects under fasting conditions.***

RESULTS: ***All the 8 subjects successfully completed the study. There were no significant protocol deviations. The two drugs were well tolerated by the volunteers. Administration of the reference formulation (Isordil: Wyeth Lederle, Mumbai) showed a maximum reduction in systolic blood pressure of 32 $\pm$ 3.96 mm of Hg (E_{max}) at 49 $\pm$ 10.6 minutes. The area under the effect time curve at time 't' was 2491.875 $\pm$ 555.826 mm. hr. Administration of the test formulation (Sorbitrate: Nicholas Piramal, Madhya Pradesh) showed a maximum reduction in systolic blood pressure of 30 $\pm$ 4 mm of Hg at 45 $\pm$ 17.92 minutes. The area under the effect time curve at time 't' was found out to be 2295.625 $\pm$ 456.829 mm. hr. The test to reference ratios for log transformed data of the test formulation for maximum reduction in the systolic blood pressure and area under the effect time curve were 91.93% and 93.77% respectively. The 90% confidence interval for log transformed data for maximum reduction in the systolic blood pressure***

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and area under the effect time curve were 83.91–100.7 and 81.12–111.27 respectively. There was no period, sequence and formulation effect observed as indicated by the p values.

CONCLUSION: ***The two products were bioequivalent and can be safely substituted for the prophylaxis of angina pectoris and for the management of unstable angina pectoris.***

KEY WORDS: Pharmacodynamic, Bioequivalence, isosorbide dinitrate

INTRODUCTION

Systemic drugs administered orally or parentally must reach the general circulation in their physiologically active form to be distributed throughout the body to exert therapeutic effects at the site of action (1). Variations in the completeness of absorption of the drug are always therapeutically important. Changes in the bioavailability are invariably reflected in the concentration of the drug in the circulation and at the site of action. The intensity of the therapeutic actions of many drugs correlates well with the concentration of the drug in the biological fluid - serum or plasma (2). The rate of absorption is therapeutically important with single doses. When absorption of the drug is slow the minimum effective concentration of the drug at its site of action may never be reached. Conversely rapid absorption of the drug may cause concentrations to rise above the maximum effective concentration and may produce toxic effects. This is especially important in case of narrow therapeutic index drugs (3) where relatively small changes in the concentration can lead to marked changes in pharmacodynamic response.

Medical profession has realized the problem of wide variations in the therapeutic effectiveness of various brands of oral formulations containing the same active ingredient in equal amounts. Lack of bioequivalence among different brands has been well documented with digoxin (4), phenobarbital (5), prednisolone (6), tolbutamide (7), diclofenac sodium (8) and aspirin (9).

Isosorbide dinitrate is an organic nitrate that is used in the prophylaxis of angina pectoris. The oral absorption of conventionally formulated Isosorbide dinitrate is considered to be complete, while mean systemic bioavailability ranges from 19-25%. Peak Isosorbide dinitrate concentration occurs between 30 and 60 min after an oral dose (10). There is however, no published report on the pharmacodynamic bioequivalence of isosorbide dinitrate formulations. Several different formulations of isosorbide dinitrate are available, which differs in their bioavailability and the extent of presystemic metabolism, which is most pronounced following peroral route (11). In one study, hemodynamics of isosorbide dinitrate following different routes and dosage forms were studied. (12). The pharmacokinetics and hemodynamic effects of isosorbide dinitrate have been investigated following administration of single doses as a sublingual spray (2.5 mg), sublingual tablet (5 mg), and per-oral tablet (10 mg) in a randomized, placebo controlled, double-blind crossover trial in 16 healthy volunteers. The haemodynamic effects were quantified using a /b ratio of the finger pulse wave and the systolic blood pressure and heart rate under orthostatic conditions. After the sublingual spray C_{max} was higher (39 ng /ml) and T_{max} was shorter (3.9 min) than after the sublingual (22.8 ng / ml and 13.8 min) and peroral tablet (16.9 ng / ml and 25.6 min).

The AUC did not differ following any of the three formulations (1031, 879, 997 ng/ml. min) for the spray, sublingual tablet and per-oral tablet)

There is an utmost need to document bioequivalence of these formulations. Various manufacturers claim that their products are bioequivalent to the innovator's formulation but it may not be so. Most of the bioequivalence studies on which these claims are based did not use confidence intervals, a current DCGI (Drug Controller General of India) requirement to document bioequivalence with the comparator drug formulation. So, the only way to verify these claims is to do a comparative bioequivalence study with the innovator drug formulation using confidence intervals.

Hence the present study was undertaken to compare the pharmacodynamic bioequivalence of the only two marketed brands of 10-mg isosorbide dinitrate tablets in India in healthy, adult, male, human subjects under fasting conditions.

MATERIALS AND METHODS

Study design: The study was conducted as an open label, balanced, randomized, two period, two treatment, two sequence, crossover single dose study.

Products to be evaluated

Reference (R): Isosorbide dinitrate 10 mg tablet (Isordil: Wyeth Lederle, Mumbai)

Test (T): Isosorbide dinitrate 10 mg tablet (Sorbitrate: Nicholas Piramal, Madhyapradesh).

Study subjects

The study was carried out on 8 healthy human subjects. The average (±SD) age and weight of the subjects were 23.25 (3.99) years (range 18-31 yrs) and 63 (7.4) (range 51-77) respectively. All were in good health as evidenced by the medical histories, complete physical examination and laboratory tests performed with in 28 days prior to the commencement of the study. None had history of any allergy to isosorbide and related compounds. All the volunteers included in the study were normotensives and none had any evidence of cardiac disease at the beginning of the study. Subjects did not receive any medication during the 2 weeks period prior to dosing. They were instructed during screening not to take any prescription and over the counter medications. All the subjects were instructed to abstain from any xanthine containing food or beverages or alcoholic products for 48 hours prior to dosing and throughout the sampling schedule during each period. All subjects were fasted overnight for 11 hours before the morning dose and for 4-hrs post-dose. Drinking water was not allowed 1 hour pre dose to 2-hrs post dose. There after it was allowed at all times. There was a washout period of 3 days between the administration of test and reference products. A single oral dose of 10 mg isosorbide dinitrate was administered during each period according to SAS generated randomization schedule.

Clinical procedures and measurements

Subjects were admitted and housed in the Clinical Pharmacology Unit from 12 hrs before dosing and were discharged 5 hrs after dose administration. Arterial blood pressure both systolic and diastolic (KorotKoff phase 5) was recorded by using a standard calibrated mercury sphygmomanometer. Measurements were made one minute after rising from the supine position (orthostatic challenge). Measurement of blood pressure both systolic and diastolic was carried out at 5, 10, 15, 30, 45, 60, 75, 90, 120 and 150 minutes after drug administration. Systolic blood pressure was used for determining pharmacodynamic bioequivalence.

Ethical considerations

This research was carried out as per ICH Guidelines for Good Clinical Practices (ICH: GCP) and the principles enunciated in the Declaration of Helsinki. Jamia Hamdard Institutional Review Board reviewed the protocol and the corresponding informed consent form (ICF) used to obtain informed consent of study subjects. The study subjects were not dosed until the Board had approved the protocol and the ICF. Subjects were required to understand and sign a consent form summarizing the discussion prior to check-in for the Period I of the study.

Pharmacodynamic analysis

The following pharmacodynamic parameters, i.e. E_{max} (maximum reduction in the systolic blood pressure), T_{max} (time of maximum reduction in the systolic blood pressure) and AUEC $_{(0-t)}$ (Area under the effect time curve as percentage of baseline), were calculated by using WINNONLIN software version 1.5 (SCI, USA).

Statistical analysis: Statistical analysis was performed on data obtained from all the 8 subjects. The analysis was conducted on least square means (LSM) of each pharmacodynamic component of the test product for all treatments using SAS software version 6.12 (SAS Institute Inc. Cary NC, USA). The geometric means, standard deviation and coefficient of variation were calculated using log transformed data. Ninety percent (90%) confidence intervals were calculated for the pharmacodynamic parameters Emax and AUEC $_{(0-t)}$ for untransformed and log transformed data (13). The untransformed and log transformed pharmacodynamic parameters i.e. E_{max} and AUEC $_{(0-t)}$ were analyzed using Analysis of variance (ANOVA) model. A p value < 0.05 was considered statistically significant at 95% level of significance. The intra subject variability for the test product was also calculated for the pharmacodynamic parameters for both untransformed and log transformed data.

RESULTS

There were no significant protocol deviations. All the 8 subjects successfully completed the study. The two drugs were well tolerated by the volunteers. During the study, 3 subjects reported mild headache within five to ten minutes after the administration of the drug. This could be attributed to the drug itself, and it required no treatment.

The various pharmacodynamic parameters namely E_{max}, T_{max} and AUEC $_{(0-t)}$ after the administration of isosorbide dinitrate are shown in Table 1.

The linear mean plot of percentage decrease in systolic blood pressure versus time for the two treatments is shown in Figure 1.

The summary statistics for log transformed pharmacodynamic parameters is shown in Table 2.

Table 1. Mean ± SD pharmacodynamic parameters of two formulations of isosorbide dinitrate in 8 human subjects

Formulation type	E_{max} (mm of Hg)	T_{max} (hr)	AUEC $_{(0-t)}$ (mm.hr)
Reference (Isordil)	32 ± 3.96	49± 10.60	2491.87± 555.82
Test (Sorbitrate)	30± 4.00	45± 17.92	2295.62± 456.82

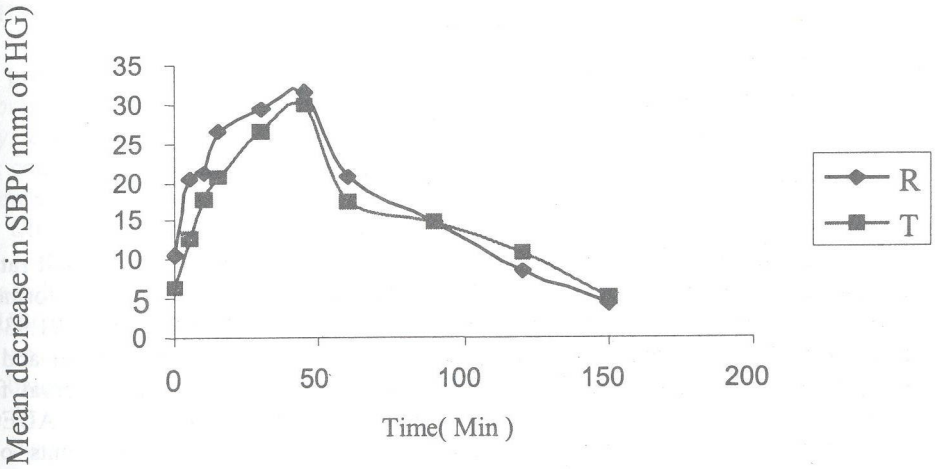

Fig 1. Mean Plot of Percentage decrease in systolic blood pressure versus time for the two treatments

Table 2. Summary statistics of log transformed pharmacodynamic parameters of isosorbide dinitrate

Product	E_{max} (mm of Hg)	$AUEC_{(0-t)}$ (mm.hr)
Reference (Isordil)		
Geometric mean	31.28	2439.67
SD	3.96	555.82
% CV	12.67	22.78
N	8.00	8.00
Test (Sorbitrate)		
Geometric mean	28.76	2287.81
SD	4.00	199.90
% CV	13.90	8.73
N	8.00	8.00
Sorbitrate/Isordil (%)	91.93	93.77
90% CI	83.91-100.70	81.12-111.27

Administration of the reference formulation (Isordil 10 mg) showed a maximum reduction in systolic blood pressure of 32 $\pm$ 3.96 mm of Hg (E_{max}) at 49 $\pm$ 10.6 minutes. The $AUEC_{(0-t)}$ at time 't' was 2491.875 $\pm$ 555.826 mm. hr.

Administration of the test formulation (Sorbitrate 10 mg) showed a maximum reduction in systolic blood pressure of 30 $\pm$ 4 mm of Hg at 45 $\pm$ 17.92 minutes. The $AUEC_{(0-t)}$ at time 't'was found out to be 2295.625 $\pm$ 456.829 mm. hr.

The 90% confidence interval for log transformed data for E_{max}, $AUEC_{(0-t)}$ were 83.91–100.7 and 81.12–111.27 respectively. In this study the T/R ratios for log transformed data of the test formulation for E_{max} and $AUEC_{(0-t)}$ were 91.93% and 93.77% respectively. The upper and lower limits of 90% confidence interval for log transformed parameters (E_{max}, $AUEC_{(0-t)}$) fell within the prescribed limits of bio-equivalence i.e. 80-125% for log transformed data. The intra subject variability for the E_{max}, $AUEC_{(0-t)}$ were 9.54% and 19.58% for the untransformed data and for log transformed data it was 15.32% and 7.97%. There was no period, sequence and formulation effect observed as indicated by the p values using 95% significance level, (P> 0.05, Table 3).

Table 3. P-values (95% significance level) and intra-subject variability for sorbitrate (Test) for log transformed data

	E_{max} (mm of Hg)	$AUEC_{(0-t)}$ (mm.hr)
Intra subject Variability (%)	15.32	7.97
P -values		
Sequence	0.16	0.12
Period	0.28	0.21
Formulation	0.06	0.11

DISCUSSION

In the present study the bioequivalence of two brands of isosorbide dinitrate in healthy, male, adult human subjects under fasting conditions was evaluated. The study was carried out in accordance with ICH Good Clinical Practices. Bioequivalence was assessed by measuring the pharmacodynamic parameters namely E_{max} and $AUEC_{(0-t)}$ for isosorbide dinitrate. The study was conducted by using an open label, balanced, randomized, cross over design in healthy, adult, male volunteers under fasting conditions. All the eight volunteers recruited in the trial successfully completed the study. The T /R ratios of the test products were satisfactory i.e. 91.93% and 93.77% for E_{max} and $AUEC_{(0-t)}$. When 90% Confidence interval was calculated for the pharmacodynamic parameters the confidence limits fell with in the prescribed limits of bio-equivalence as per DCGI guidelines i.e. 80-125% for log transformed data. Thus, these two products were bioequivalent.

A through search on Internet located no study reporting , the assessment of bioequivalence of isosorbide dinitrate formulations using pharmacodynamic parameters. Thus, the present study is the first to use pharmacodynamic parameters to assess bioequivalence of two isosorbide dinitrate formulations. A good decrease in the orthostatically induced systolic blood pressure following administration of both the reference and test products was there and the effect was comparable. This may be due to the formation of a large quantity of active metabolites following peroral treatment. The intrasubject variability for the pharmacodynamic parameters was also low for untransformed and log transformed data and p values did not reveal any treatment, period and sequence effect. In this study the bioavailability of the test formulation was 93.77% with respect to reference formulation.

Thus, the two products were bioequivalent (were comparable in terms of the rate and extent of absorption) and can be safely substituted therapeutically. Thus a patient who is currently stabilized on Isordil can be safely switched over to Sorbitrate and vice versa for the management of angina by the physician/pharmacist, without concern for any adverse effect on efficacy and safety.

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