JYP

Microdosing vs. Therapeutic Dosing for Evaluation of Pharmacokinetic Data: A Comparative Study

Mahajan R, Parvez A[1], Gupta K[2]

Department of Pharmacology, Adesh Institute of Medical Sciences & Research, Bathinda-151109 (Punjab), India; [1]Department of Experimental Medicine, Irvine School of Medicine, University of California, Irvine, CA-92697; [2]Department of Biochemistry, Adesh Institute of Medical Sciences & Research, Bathinda-151109 (Punjab), India

Address for correspondence: *Dr. Rajiv Mahajan; E- mail: drrajivmahajan01@yahoo.co.in*

ABSTRACT

Objectives: Human microdosing studies or phase 0 studies have been proposed to supplement pharmacokinetic (PK) studies in animals. In phase 0 studies, extremely low, nonpharmacologically active doses of a drug are given to a few subjects before phase 1, to define the agent's PK profile in humans. This study has been conducted to compare the values of different PK parameters, as determined by microdosing and conventional therapeutic dose studies in healthy volunteers, and target patient population. **Methods:** In the first phase of study, 30 healthy adult male volunteers were divided into three groups of 10 each; receiving 14C-labelled atenolol, enalapril and losartan orally, in single microdose. After a wash-out period of 10 days, the same individual received the same drug in single therapeutic dose. In second phase of study, 30 hypertensive patients were divided into three groups and given same drugs. Parameters studied were t½, AUC, Cmax and tmax. Blood samples collected at intervals were subjected to accelerator mass spectrometry (AMS) and high performance liquid chromatography (HPLC), for microdosing and therapeutic dose studies respectively. **Results:** Microdosing results were comparable with therapeutic dose values for all the drugs studied and showed linearity over therapeutic dose. **Conclusions:** Microdosing PK parameters are comparable to the ones determined by therapeutic dose studies up to a permissible limit. So the idea of phase 0 PK studies supplementing phase 1 PK studies can be furthered.

Key words: Microdosing, Phase I studies, pharmacokinetics, therapeutic dose

DOI: 10.4103/0975-1483.59315

INTRODUCTION

During drug development, preliminary pharmacokinetic (PK) data of a new chemical entity (NCE) is first pooled from pre-clinical (animal) studies and only then that compound is introduced to human beings. During human studies, the pharmacokinetics (PKs) of therapeutic agents is generally studied in young, healthy volunteers during phase I. Safety and efficacy studies are performed on patients during phase II and further, but PK studies are not conducted in patients.

Recently, two new approaches, fully backed by Food and drug Administration (FDA), i.e. phase 0 studies[1] and Population Pharmacokinetics (PopPK)[2] have been introduced. These approaches will minimize dependency on animal testing for preliminary pharmacokinetic data and generate pharmacokinetic data in target patient population.

Microdosing or phase 0 studies are the processes of evaluating PKs of a drug by administrating sub-therapeutic doses of that drug to a small group of healthy volunteers before phase I studies. Dose given is less then 1/100th of the dose calculated to yield a pharmacological effect, to a maximum of less then 100μgs (<30nM for protein products).[1] A microdose study provides early PK data in humans and only requires minimal preclinical toxicology safety testing.[3] Thus, phase 0 trials have been proposed as a means of reducing the long drug development timeline.[4] But microdosing is a relatively recent innovation and there remains a degree of uncertainty as to whether such a small dose will adequately predict the PKs of the therapeutically active dose. Moreover, the non-therapeutic aspect and the fact that a phase 0 trial will not obviate phase I testing have caused reluctance in patients, industry, and academia.

Population pharmacokinetics (PopPKs) is the study of sources and correlates of variability in drug concentrations among individuals who are the target patient population receiving clinically relevant doses of a drug of interest.[5] In simple words, PopPKs seeks to obtain relevant pharmacokinetic information in patients who are representative of the target population to be treated with the drug.

Keeping in view all these arguments, we hypothesized if pharmacokinetic parameters as derived by microdosing studies, therapeutic dose studies in healthy volunteers and therapeutic dose study in target patient population fall within permissible/normal limit of each other, it can be easily interpolated that microdosing studies can replace/ supplement conventional phase I PK studies and can obviate the need for PopPKs studies.

MATERIALS AND METHODS

Our study was a single dose, ethically designed pharmacokinetic study completed at two centers, in two phases. The first phase study (conducted abroad) was a cross-over design including 30 healthy, adult male volunteers; after taking informed written consent. This phase of the study was conducted in a center well equipped with AMS technique. In this phase, these 30 healthy individuals were divided into three groups of 10 individual each. In the first leg of this first phase of the study, 10 volunteers in these groups received ^{14}C labeled (200nanoCuries) atenolol, enalapril and losartan orally in a single dose of 100μgs.[6] This leg of first phase was marked as group I and individual groups were marked as group Ia, group Ib and group Ic respectively. Blood samples were collected at predetermined intervals through heparinized tubes. Plasma samples were separated and stored at -20^{0}C until analysis. These blood samples were subjected to AMS. The amount of radioactivity arising from such a small labeling does not require regulatory approval for human administration.[7]

After a wash-out period of 10 days, these same individuals participated in the second leg of first phase where same individual received same drug in a single therapeutic dose i.e. 50 mg atenolol, 10mg enalapril and 50mg losartan. This leg was marked as group II and individual groups were labeled as group IIa, group IIb and group IIc respectively. Blood samples were again collected by the same time frame as in first leg and were subjected to HPLC.

In the second phase of this study (conducted in India), 30 male hypertensive patients were randomized into three groups of 10 each to receive 50mg atenolol, 10mg enalapril and 50mg of losartan as a single dose respectively, after taking due informed written consent. This phase was labeled as group III and individual groups were labeled as group IIIa, IIIb and IIIc. In these patients, blood samples were collected by same protocol as in first phase and were subjected to HPLC.

Pharmacokinetic parameters studied were maximum concentration achieved (C_{max}), time to achieve maximum concentration (t_{max}), plasma half life of the drugs ($t_{½}$,) and area under the curve (AUC). Area under the curve (AUC) was calculated by integration method.[8] No statistical test was applied, only descriptive statistical analysis was done. For pharmacokinetic linearity of microdose to therapeutic dose, a prediction within a factor of two was considered appropriate.[9] Pharmacokinetic data collected from both phases (groups I, II and III) was also compared with pharmacokinetic data available in literature, which was considered as true value.[10] Based on the currently adopted approach of allometric scaling of animal data to human pharmacokinetics, any prediction within a factor of two of true value was considered acceptable.[11]

RESULTS

Age of adult males who participated in the two phases ranged from 33-45 years for first phase and 38-50 years for second phase respectively. Mean age in two phases was 36.8 years and 44.7 years respectively.

Atenolol

The pharmacokinetic parameters for atenolol, as derived from microdosing (Ia), therapeutic doses in healthy

volunteers (IIa) and therapeutic doses in hypertensive patients (IIIa) and their comparison with literature values are given in Table 1, while graph showing area under curve (AUC) of atenolol in different groups is in Figure 1.

Enalapril

The pharmacokinetic parameters for enalapril, as derived from microdosing (Ib), therapeutic doses in healthy volunteers (IIb) and therapeutic doses in hypertensive patients (IIIb) and their comparison with literature values are given in Table 2, while graph showing area under curve (AUC) of enalapril in different groups is in Figure 2.

Losartan

The pharmacokinetic parameters for losartan, as derived from microdosing (Ic), therapeutic doses in healthy volunteers (IIc) and therapeutic doses in hypertensive patients (IIIc) and their comparison with literature values are given in Table 3, while graph showing area under curve (AUC) of losartan in different groups is in Figure 3.

Comparative results between PK parameters as derived from microdosing studies and from therapeutic studies in healthy volunteers are shown in Table 4 and between microdosing and hypertensive patients are shown in Table 5.

DISCUSSION AND CONCLUSION

As is evident from tables 4 and 5, all pharmacokinetic parameters derived from microdosing fall within a factor of two (0.5-2 times) of therapeutic doses in healthy volunteers

Table 1: PK parameters as calculated for atenolol in different groups

Parameter	Group Ia	Group IIa	Group IIIa	Literature values
C_{max} (ng/ml)	395±21.34*	353.80±13.29	335.90±17.74	280±0.09
t_{max} (hrs)	4.46±0.33	4.14±0.37	4.28±0.30	3.3±1.3
$t_{½}$, (hrs)	7.11±0.81	7.23±0.50	7.39±0.46	6.1±2.0
AUC (hr.ng/ml)	4440±462.61*	3636±395.66	3711±387.87	NA

*: Normalized to therapeutic dose of 50mg (Microdosing Cmax was 0.79±0.04 ng/ml)

Table 2: PK parameters as calculated for enalapril in different groups

Parameter	Group Ib	Group IIb	Group IIIb	Literature values
C_{max} (ng/ml)	91±8.55*	96.60±9.29	95.00±7.32	69±37
t_{max} (hrs)	4.34±0.50	3.70±0.44	3.85±0.23	3.0±1.6
$t_{½}$, (hrs)	12.10±0.79	11.76±0.67	11.71±0.44	11
AUC (hr.ng/ml)	1302±159.98*	1200±160.17	1302±172.12	NA

*: Normalized to therapeutic dose of 10mg (Microdosing Cmax was 0.91±0.09ng/ml)

Table 3: PK parameters as calculated for losartan in different groups

Parameter	Group Ic	Group IIc	Group IIIc	Literature values
C_{max} (ng/ml)	438.50±48.31*	429.70±20.60	414.50±16.81	296±217
t_{max} (hrs)	1.45±0.21	1.42±0.18	1.35±0.16	1.0±0.5
$t_{½}$, (hrs)	3.31±0.48	3.41±0.23	3.17±0.34	2.5±1.0
AUC (hr.ng/ml)	1809±210.76*	1707±211.41	1738±199.57	NA

*: Normalized to therapeutic dose of 50mg (Microdosing Cmax was 0.88±0.10ng/ml)

Table 4: PK values by microdosing vs. therapeutic dosing in healthy volunteers

Drugs	C_{max}	t_{max}	$t_{½}$,	AUC	Linearity
Atenolol	1.12	1.08	0.98	1.22	Linear 0.1-50 mg dose
Enalapril	0.94	1.17	1.03	1.09	Linear 0.1-10 mg dose
Losartan	1.02	1.02	0.97	1.06	Linear 0.1-50 mg dose

Table 5: PK values by microdosing vs. therapeutic dosing in hypertensive patients

Drugs	C_{max}	t_{max}	$t_{½}$,	AUC	Linearity
Atenolol	1.18	1.04	0.96	1.20	Linear 0.1-50 mg dose
Enalapril	0.96	1.13	1.03	1.00	Linear 0.1-10 mg dose
Losartan	1.06	1.07	1.04	1.05	Linear 0.1-50 mg dose

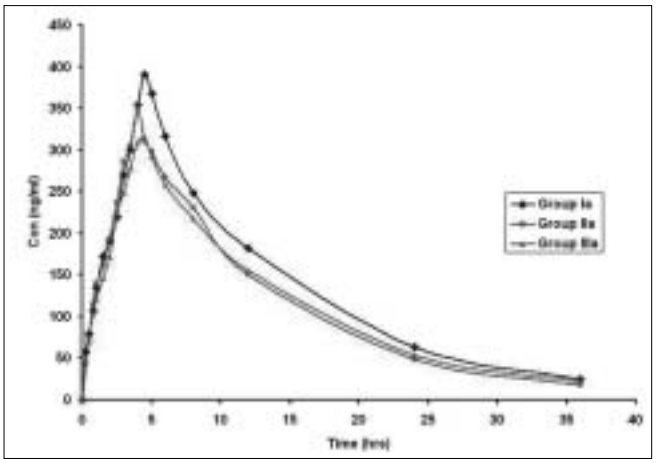

Figure 1: Graph showing AUC of atenolol in different groups

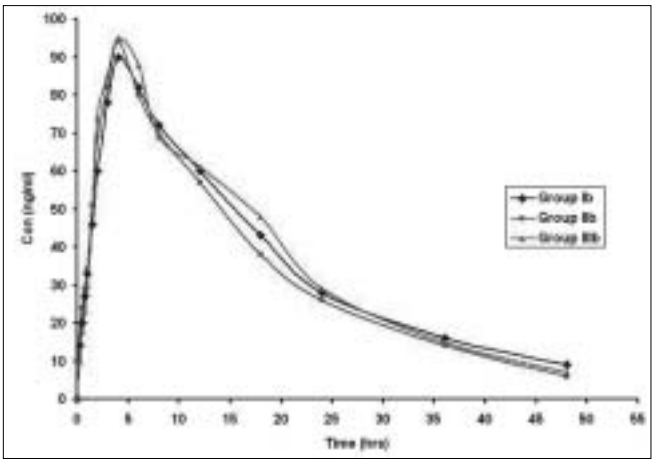

Figure 2: Graph showing AUC of enalapril in different groups

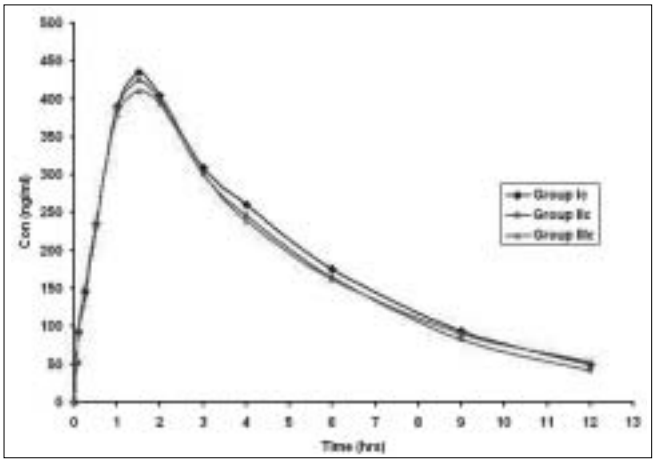

Figure 3: Graph showinh AUC of losartan in different groups

and hypertensive patients, showing a linear relationship for all drugs studied i.e. atenolol, enalapril and losartan over therapeutic range for all pharmacokinetic parameters studied.

As is evident from Tables 1, 2 and 3, C_{max} (ng/ml), t_{max} (hrs) and $t_{½}$, (hrs) for all three drugs used in microdoses, therapeutic doses in healthy volunteers and therapeutic doses in hypertensive patients are within a factor of two of literature (true) values. It can be fairly concluded that microdose studies can supplement pharmacokinetic data obtained from phase I studies and can also obviate the need of PopPks.

Consistent with the FDA's critical path initiative, it is believed that human microdosing offers a faster, more accurate method of developing drugs -- bridging the gap between the laboratory and the clinic. Over the last 10 years, Xceleron has developed a database which compares PK data at microdose; therapeutics dose levels. It shows that for 25 compounds studied, microdose PK scales to therapeutic dose in over 80% of cases.[12]

Although, presently microdose studies are conducted only to make a decision about the viability of a new compound at an early time, future of microdosing studies is bright and these studies, not only can replace the need of preliminary animal pharmacokinetic studies, but can also supplement phase I pharmacokinetic studies and population kinetic studies. An interesting prospectus in the future may be to conduct microdosing studies in target patient population for extracting pharmacokinetic data.

ACKNOWLEDGMENTS

We are thankful to George H Miller, Laboratory Director, Lawrence Livermore National Laboratory, California, for conducting AMS and Nilesh Arora, Incharge-Clinical Laboratory, Ranbaxy Ltd, Mohali for HPLC studies.

REFERENCES

1. Usha Rani P, Naidu MUR. Phase 0- Microdosing strategy in clinical trials. Indian J Pharmacol 2008;40:240-2.
2. Sandeep G, Santhosh M, Srikanth T. Population Pharmacokinetics: Novel Approach for Pharmacokinetic Modeling. The Internet Journal of Pharmacology. 2009 Jul [cited 2009 Jul 31]; 7 (1): [about 7 p.]. Available from: http://www.ispub.com/journal/the_internet_journal_of_pharmacology/volume_7_number_1_27/article/population-pharmacokinetics-novel-approach-for-pharmacokinetic-modeling.html
3. Lappin G, Colin GR. A Review of Human Phase 0 (Microdosing) Clinical Trials Following the US Food and Drug Administration Exploratory Investigational New Drug Studies Guidance. Int J Pharma Med 2006;20:159-65.
4. Bates SE. CCR Focus. Clin Cancer Res 2008;14:3657.
5. Aarons L. Population pharmacokinetics: theory and practice. Br J Clin Pharmacol 1991;32:669-70.
6. Cream trial update. 2005 Jan 01. [cited 2009 Aug 02]. Available from: http://www.xceleron.com/metadot/index.pl?id=2582&isa=DBRow&op=show&dbview_id=2597.

7. European Microdosing AMS Partenrship Programme (EUMAPP). Outcomes from EUMAPP-A study comparing in vitro, in silico, microdose and pharmacological dose pharmacokinetics. [cited 2009 Aug 02]. Available from: http://www.eumapp.com/pdfs/EUMAPP%20SUMMARY.pdf.
8. Area under a curve: Numerical Integration. 2005 Oct. [cited 2009 Aug 04]. Available from: http://people.stfx.ca/bliengme/ExcelTips/AreaUnderCurve.xls
9. Lappin G. Microdosing: Where we are today. 2008 [cited 2009 Aug 05]. Available from: http://www.xceleron.com/metadot/index.pl?id=3088&isa=DBRow&field_name=speaker2_attachment&op=download_file
10. Thummel KE, Shen DD, Isoherranen N, Smith HE. Design and optimization of dosage regimens; Pharmacokinetic data. In: Brunton LL, Lazo JS, Parker KL. editors. Goodman and Gilman's The pharmacological basis of therapeutics. 11 th ed. McGraw-Hill: New York; 2006. p. 1787-888.
11. Lappin G, Kuhnz W, Jochemsen R, Kneer J, Chaudhary A, Oosterhuis B, *et al*. Use of microdosing to predict pharmacokinetics at the therapeutic dose: Experience with 5 drugs. Clin Pharmacol Ther 2006;80:203-15.
12. 12. Microdosing- Current status and future predictions. 2008 Jul 10. Xceleron: Accelerating drug development. [cited 2009 Aug 05]. Available from: http://www.xceleron.com/metadot/index.pl?id=3088&isa=DBRow&op=show&dbview_id=3049.

Source of Support: Nil, **Conflict of Interest:** None declared.