

PHARMACOKINETIC INTERACTION STUDY BETWEEN IVABRADINE WITH FLUOXETINE OR METRONIDAZOLE IN HEALTHY VOLUNTEERS

LAURIAN VLASE^{1*}, ADINA POPA², MARIA NEAG², DANA
MUNTEAN¹, SORIN E. LEUCUȚA¹

¹*Faculty of Pharmacy, Department of Pharmaceutical Technology and
Biopharmaceutics,*

²*Faculty of Pharmacy, Department of Clinical Pharmacy, "Iuliu
Hațieganu" University of Medicine and Pharmacy, Cluj-Napoca, 13 Emil
Isac, 400023, Romania*

**corresponding author: vlaselaur@yahoo.com*

Abstract

The pharmacokinetic interaction between ivabradine with fluoxetine and metronidazole in healthy volunteers was evaluated. In two separate experiments, a single dose of either 5 or 10 mg ivabradine was administered alone or in combination with fluoxetine or metronidazole to 18 healthy male volunteers in a two treatment study design, separated by a period in which the fluoxetine or metronidazole alone were administered daily. The pharmacokinetic parameters of ivabradine administered alone or in combination with fluoxetine or metronidazole were calculated using the non-compartmental analysis. Non-statistically significant differences have been observed for the main pharmacokinetic parameters of ivabradine when administered alone or with fluoxetine or metronidazole, demonstrating the lack of pharmacokinetic interaction between these drugs.

Rezumat

A fost evaluată interacțiunea medicamentoasă farmacocinetică dintre ivabradină și fluoxetină sau metronidazol, la voluntari sănătoși. În două studii diferite, la 18 subiecți a fost administrată o doză unică de ivabradină de 5 respectiv 10 mg singură sau în combinație cu fluoxetina sau metronidazolul, urmând un design cu două perioade între care s-a realizat un tratament cu fluoxetină sau metronidazol. Parametrii farmacocinetici ai ivabradinei administrată singură sau în combinație cu fluoxetina sau metronidazolul au fost calculați utilizând analiza farmacocinetica non-compartimentală. Nu au fost găsite diferențe semnificative între parametrii farmacocinetici ai ivabradinei administrată singură sau împreună cu fluoxetina sau metronidazolul, demonstrând lipsa interacțiunii farmacocinetice între aceste substanțe medicamentoase.

Keywords: pharmacokinetic interaction, ivabradine, fluoxetine, metronidazole.

Introduction

Ivabradine is a novel heart rate-lowering agent that selectively and specifically inhibits the depolarizing cardiac pacemaker I_f current in the sinus node. Its activity provides pure heart rate reduction at rest and during

exercise, which improves myocardial oxygen balance and increases coronary perfusion, without any relevant influence on conduction, contractility, ventricular repolarization or blood pressure. The anti-ischemic efficacy and the safety of ivabradine have been demonstrated in patients with stable *angina pectoris* [1-5]. Despite its therapeutical benefit, ivabradine has some important side effects, including bradycardia, atrioventricular (AV) block, ventricular extrasystoles and luminous phenomena [1,3,5]. Due to the high potential of ivabradine to give adverse reactions on overdosing, but also the lack of therapeutic effect on underdosing, it is important to know the way in which some other substances modify the ivabradine pharmacokinetics.

After oral administration, the metabolic clearance of ivabradine accounts for about 80% of its total clearance, with the other 20% corresponding to renal clearance. Mainly the cytochrome P₄₅₀ isoform 3A4 (CYP3A4) is involved in the metabolism of ivabradine, so numerous potential interactions can therefore arise with CYP3A4 inhibitors and inducers [6,7].

Fluoxetine is an antidepressant for oral administration that is effective through selective inhibition of serotonin reuptake. Fluoxetine is metabolized by N-demethylation to an active metabolite, norfluoxetine [8]. Both fluoxetine and norfluoxetine have been shown to be potent inhibitors of the cytochrome P₄₅₀ isoform 2D6 (CYP2D6), whereas the latter is also a moderate inhibitor of CYP3A4 [9].

Metronidazole is used to treat *Giardia* infections of the small intestine, amebic liver abscess and dysentery (amebic colon infection causing bloody diarrhea) and is a moderate inhibitor of CYP3A4 [10].

It is important to determine whether a potentially harmful pharmacokinetic interaction occurs between ivabradine and fluoxetine or metronidazole, this being the aim of our study.

Materials and methods

Subjects

In each study, eighteen non-smoking males, aged 20-26 years were enrolled (Table I). The study was conducted according to the principles of the Declaration of Helsinki (1964) and its amendments (Tokyo 1975, Venice 1983, Hong Kong 1989) and Good Clinical Practice (GCP) rules. The clinical protocol was reviewed and approved by the Ethics Committee of the University of Medicine and Pharmacy "Iuliu Hatieganu", Cluj-Napoca, Romania. All volunteers gave their written informed consent prior to study inclusion. The volunteers were healthy according to history,

physical examination and laboratory tests, had no history of alcohol or drug abuse and did not take any regular medication.

Study design.

Each study consisted of 2 periods: Period 1 (Reference), when each volunteer received a single dose of ivabradine and Period 2 (Test), when each volunteer received a single dose of ivabradine and either fluoxetine or metronidazole. Between the two periods, the subjects were treated with fluoxetine or metronidazole (Table I). All the drugs were administered in fasted state. The pharmaceutical products used were Corlentor[®] (5 mg tablets, producer Les Laboratoires Servier, France), Fluoxin[®] (20 mg capsules, producer Vim Spectrum SRL, Romania) and Flagyl[®] (500 mg tablets, producer Aventis, France). Venous blood (5 mL) was collected into heparinized tubes, in the first and in the last day of the study, before drug administration as well as at 0.5, 1, 1.5, 2, 2.5, 3, 4, 5, 6, 7, 8, 10 and 12 hours after drug administration and the separated plasma was stored frozen (-20°C) until analysis.

Table I

The design of the pharmacokinetic interaction studies between ivabradine with fluoxetine or metronidazole

	Study 1	Study 2
	Pharmacokinetic interaction	
	Ivabradine-fluoxetine	Ivabradine-metronidazole
Study type	Open label, two periods – Reference and Test	Open label, two periods – Reference and Test
Subjects	18 healthy volunteers	18 healthy volunteers
Drugs administered in Reference period:	Ivabradine, 5 mg	Ivabradine, 10 mg
CYP3A4 inhibitor drug	fluoxetine	metronidazole
Dose of inhibitor drug	60 mg p.o.	2x500 mg p.o., b.i.d*
Days of treatment with inhibitor	7	3
Drugs administered in Test period:	Ivabradine, 5 mg + Fluoxetine, 60 mg	Ivabradine, 10 mg + Metronidazole, 500 mg

*b.i.d. – *bis in die* (lat.) – twice a day.

Analysis of plasma samples

Ivabradine plasma concentrations were determined by a validated Liquid Chromatography – Mass Spectrometry (LC/MS) method [11].

Pharmacokinetic analysis

The noncompartmental pharmacokinetic analysis method was employed to determine the pharmacokinetic parameters of ivabradine given alone or in combination with fluoxetine or metronidazole. The maximum

plasma concentration (C_{max} , ng/mL) and the time to reach the peak concentration (t_{max} , hr) were obtained directly by the visual inspection of each subject's plasma concentration-time profile. The area under the concentration-time curve (AUC_{0-t}) has been estimated by integration using the trapezoidal rule. The area was extrapolated to infinity ($AUC_{0-\infty}$) by addition of C_t/k_{el} to AUC_{0-t} where C_t is the last quantifiable drug concentration and k_{el} is the elimination rate constant. The pharmacokinetic analysis was performed using Kinetica 4 (ThermoLabsystems, USA) [12].

Statistical analysis

In order to evaluate a possible statistical or clinical significance of the pharmacokinetic interaction, an analysis of variance (ANOVA) was performed on the main pharmacokinetic parameters calculated, using general linear model procedures, in which the sources of variation were the subject and the treatment. Then the 90% confidence intervals of the test/reference period ratios for C_{max} and $AUC_{0-\infty}$ (log transformed) were determined by the Schuirmann's two one-sided t test [13-18].

Results and discussion

The mean plasma concentrations of ivabradine when administered alone or in combination with either fluoxetine (in Study 1) or metronidazole (in Study 2) after pre-treatment with fluoxetine or metronidazole, are shown in figure 1.

The mean pharmacokinetic parameters of ivabradine administered alone or in combination with fluoxetine (Study 1) or metronidazole (Study 2) are given in table II and table III, respectively, as well as the statistical significance following their comparison.

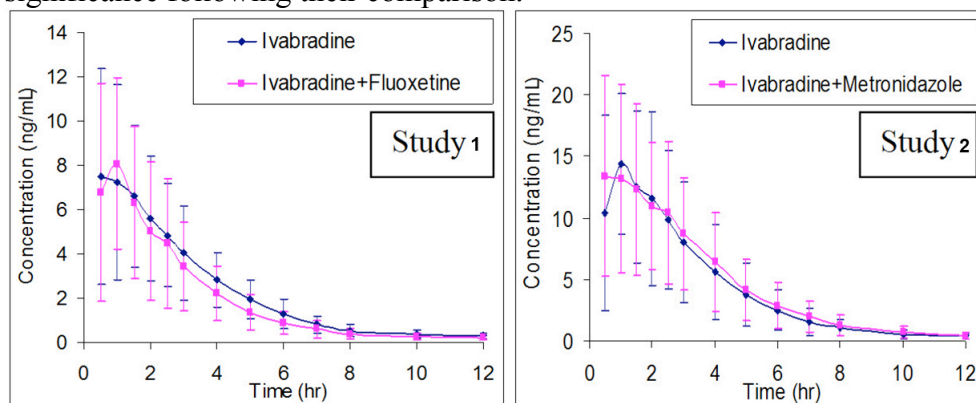


Figure 1

Mean (\pm SD) plasma levels of ivabradine given alone or in combination with fluoxetine (Study 1) or metronidazole (Study 2), after pre-treatment with fluoxetine or metronidazole

Table II
Pharmacokinetic parameters of ivabradine administered alone or after treatment with fluoxetine (Study 1)

Pharmacokinetic parameter (\pm SD)	Ivabradine alone	Ivabradine + fluoxetine	p* value, ANOVA
C_{\max} (ng/mL)	8.52 \pm 4.37	9.28 \pm 4.51	0.332, NS
t_{\max} (hr)	0.86 \pm 0.41	0.86 \pm 0.44	0.782, NS
AUC _{0-∞} (ng.hr/mL)	27.6 \pm 13.6	23.8 \pm 12.1	0.074, NS
$t_{1/2}$ (hr)	1.84 \pm 0.32	1.70 \pm 0.35	0.177, NS
MRT(hr)	3.10 \pm 0.39	2.73 \pm 0.33	0.069, NS

* significance for $p < 0.05$; MRT – mean residence time; NS – not significant

Peak plasma concentrations (C_{\max}) of ivabradine before and after the treatment with multiple doses of fluoxetine (8.52 ng/mL vs. 9.28mg/mL) were not significantly different between the two treatments. The same was found when comparing t_{\max} , AUC_{0- ∞} , $t_{1/2}$ and the mean residence time (MRT) parameters.

Table III
Pharmacokinetic parameters of ivabradine administered alone or after treatment with metronidazole (Study 2)

Pharmacokinetic parameter (\pm SD)	Ivabradine alone	Ivabradine + metronidazole	p* value, ANOVA
C_{\max} (ng/mL)	16.24 \pm 7.30	17.14 \pm 7.78	0.457, NS
t_{\max} (hr)	0.97 \pm 0.46	1.00 \pm 0.91	0.899, NS
AUC _{0-∞} (ng.hr/mL)	52.5 \pm 27.5	56.2 \pm 27.6	0.066, NS
$t_{1/2}$ (hr)	1.91 \pm 0.43	1.90 \pm 0.34	0.941, NS
MRT(hr)	3.15 \pm 0.51	3.32 \pm 0.64	0.198, NS

* significance for $p < 0.05$; MRT – mean residence time; NS – not significant

None of the pharmacokinetic parameters of ivabradine is significantly changed after the pre-treatment and co-administration of metronidazole.

The pharmacokinetic parameters C_{\max} , t_{\max} and AUC_{0- ∞} , calculated for ivabradine administered with fluoxetine or metronidazole were also used for the bioequivalence evaluation of ivabradine administered during the Test and Reference period from each study. The parametric 90% confidence interval for the ratio Test/Reference period of the mean pharmacokinetic

parameters C_{\max} and $AUC_{0-\infty}$ (log transformed) of ivabradine and the significance of the difference of t_{\max} are shown in table IV.

Table IV
Bioequivalence evaluation of pharmacokinetic parameters of ivabradine administered alone or after treatment with fluoxetine or metronidazole.

Pharmacokinetic parameter	90% Confidence intervals	
	Study 1	Study 2
	ivabradine-fluoxetine	ivabradine-metronidazole
$AUC_{0-\infty}$ (ng.h/mL)	0.83-0.98 (ANOVA, NS)	1.01-1.18 (ANOVA, NS)
C_{\max} (ng/mL)	0.93-1.21 (ANOVA, NS)	0.95-1.20 (ANOVA, NS)
t_{\max} (hr)	$\chi^2=3.841$ (Friedman, NS)	$\chi^2=3.841$ (Friedman, NS)

NS – not significant

For both pharmacokinetic studies, the 90% confidence intervals for the geometric mean of ivabradine in Test/Reference individual ratios for C_{\max} and $AUC_{0-\infty}$ were in the acceptable limits of bioequivalence (0.8-1.25) and the difference between mean t_{\max} values were not statistically significant.

Conclusions

The treatment with either fluoxetine or metronidazole until steady-state does not significantly influences the pharmacokinetics of ivabradine. No systemic metabolic drug-drug interaction was observed between ivabradine-fluoxetine or ivabradine-metronidazole, and the half-life is not changing between treatments and the drug exposure (C_{\max} and $AUC_{0-\infty}$) is about the same. Since the drug exposure related pharmacokinetic parameters were in the bioequivalence interval, the pharmacokinetic interaction may not have clinical significance.

Acknowledgments

This work was supported by a PNII-IDEI project, code 462, contract 229/2007 financed by CNCSIS Romania.

References

1. Prasad U.K., Gray D., Purcell H., Review of the I-f selective channel inhibitor ivabradine in the treatment of chronic stable angina, *Advances In Therapy*, 2009, 26(2):127-137.
2. Khawaja M.Z., Walker D.M., Ivabradine - the beauty of a slow heart rate, *International Journal Of Clinical Practice*, 2009, 63(4): 542-546.

3. Tardif J.C., Ponikowski P., Kahan T., Efficacy of the I_f current inhibitor ivabradine in patients with chronic stable angina receiving beta-blocker therapy: a 4-month, randomized, placebo-controlled trial, *European Heart Journal*, 2009, 30(5):540-548.
4. Milliez P., Messaoudi S., Nehme J., Rodriguez C., Samuel J.L., Delcayre C., : Beneficial effects of delayed ivabradine treatment on cardiac anatomical and electrical remodeling in rat severe chronic heart failure, *American Journal Of Physiology-Heart And Circulatory Physiology*, 2009, 296(2): H435-H441.
5. Riccioni G., Vitulano N., D'Orazio N., Ivabradine: Beyond heart rate control, *Advances In Therapy*, 2009, 26(1):12-24.
6. Portoles A., Calvo A., Terleira A., Laredo L., Resplandy G., Gorostiaga C., Moreno A., Lack of pharmacokinetic interaction between omeprazole or lansoprazole and ivabradine in healthy volunteers: An open-label, randomized, crossover, pharmacokinetic interaction clinical trial, *Journal of Clinical Pharmacology*, 2006, 46(10):1195-1203.
7. Portoles A., Terleira A., Calvo A., Effects of *Hypericum perforatum* on ivabradine pharmacokinetics in healthy volunteers: An open-label, pharmacokinetic interaction clinical trial, *Journal Of Clinical Pharmacology*, 2006, 46(10): 1188-1194
8. Altamura A.C., Moro A.R., Percudani M., Clinical pharmacokinetics of fluoxetine. *Clin Pharmacokinet.* 1994; 26(3):201-214.
9. Baumann P., Rochat B., Comparative pharmacokinetics of selective serotonin reuptake inhibitors: a look behind the mirror. *Int Clin Psychopharmacol.* 1995; 10(1):15-21.
10. Roedler R., Neuhauser M.M., Penzak S.R., Does metronidazole interact with CYP3A substrates by inhibiting their metabolism through this metabolic pathway? Or should other mechanisms be considered?, *Ann Pharmacother.* 2007;41(4):653-658.
11. Vlase L., Muntean D., Leucuta S. E., Baldea I., High-throughput determination of ivabradine from human plasma by LC/MS/MS and its application to pharmacokinetic studies, *Studia Universitatis Babes-Bolyai Chemia*, 2009; 54(2):43-52.
12. http://www.thermo.com/eThermo/CMA/PDFs/Product/productPDF_27347.pdf
13. U. S. Department of Health and Human Services, Food and Drug Administration, Bioavailability and Bioequivalence Studies for Orally Administrated Drug Products – General Considerations, Rockville, USA, 2003, <http://www.fda.gov/cder/guidance/index.htm>
14. The European Agency for the Evaluation of Medicinal Products. Note for Guidance on the Investigation of Bioavailability and Bioequivalence, London, UK, 2001 (CPMP/EWP/QWP/1401/98).
15. Sandulovici R., Prasacu I., Mircioiu C., Voicu V., Medvedovici A., Anuța V., Mathematical and phenomenological criteria in selection of pharmacokinetic model for m1 metabolite of pentoxyphylline, *Farmacia*, 2009, 57(2):235-246.
16. Pop A., Vlase L., Leucuța S.E., Pharmacokinetic study of felodipine after single oral dose of slow release formulations in healthy volunteers, *Farmacia*, 2008, 56(5):474-482.
17. Rădulescu F., Miron D., Mircioiu C., Voicu V., Distribution profiles of drugs with active metabolites by compartmental analysis, *Farmacia*, 2007, 55(4):390-395.
18. Anuța V., Aldea A., Neagu O., Mircioiu I., Miron D., Radulescu F., Soare-Rada M., Enache V., Bioequivalence estimation based on peak areas of unknown metabolites, *Farmacia*, 2007, 55(6):680-690.