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Stereoselective Pharmacokinetics of Stable Isotope (+/−)-[^{13}C]-Pantoprazole: Implications for a Rapid Screening Phenotype Test of CYP2C19 Activity

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Abstract

Aims—We have previously shown that the (±)-[^{13}C]-pantoprazole breath test is a promising noninvasive probe of CYP2C19 activity. As part of that trial, plasma, breath test indices and CYP2C19 (*2, *3 and *17) genotype were collected. Here, we examined whether [^{13}C]-pantoprazole exhibits enantioselective pharmacokinetics and whether this enantioselectivity is correlated with indices of breath test.

Methods—Plasma (−)- and (+)-[^{13}C]-pantoprazole that were measured using a chiral HPLC were compared between CYP2C19 genotypes and correlated with breath test indices.

Results—The $AUC_{(0-\infty)}$ of (+)-[^{13}C]-pantoprazole in PM (*2/*2, n=4) was 10.1- and 5.6- fold higher that EM (*1/*1or *17, n=10) and IM (*1/*2or *3, n=10) of CYP2C19 respectively ($p<0.001$). The $AUC_{(0-\infty)}$ of (−)-[^{13}C]-pantoprazole only significantly differed between PMs and EMs (1.98-fold; $p=0.05$). The $AUC_{(0-\infty)}$ ratio of (+)-/(−)-[^{13}C]-pantoprazole was 3.45, 0.77 and 0.67 in PM, IM and EM genotypes respectively. Breath test index, Delta Over Baseline (DOB_{max}) show significant correlation with $AUC_{(0-\infty)}$ of (+)-[^{13}C]-pantoprazole (Pearson's $r = 0.62$; $p<0.001$).

Conclusions—[^{13}C]-pantoprazole exhibits enantioselective elimination. (+)-[^{13}C]-pantoprazole is more dependent on CYP2C19 metabolic status and may serve as a more attractive probe of CYP2C19 activity than (−)-[^{13}C]-pantoprazole or the racemic mixture.

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Competing Interest

Z.D., A.M., and D.A.F. are co-owners of a patent on a clinical test designed to determine CYP2C19 activity using the [^{13}C]-pantoprazole breath test (patent #: US 7,833,744 B2).

D.A.F. has served as a paid consultant for Roche Molecular Diagnostics, Indianapolis, IN.

A.M. is employed by Cambridge Isotope Laboratories which manufactures the ^{13}C substrate used in the study for the breath test. Commercialization of the pantoprazole breath test could be financially beneficial to the company.

The other authors declare no conflict of interest.

Introduction

The human cytochrome P450 (CYP) CYP2C19 metabolizes several clinically important drugs, including those with a narrow therapeutic range.[1–3] This enzyme is also critical in the metabolic activation of some prodrugs such as clopidogrel,[4] cyclophosphamide,[5,6] and thalidomide[7] to pharmacologically active metabolites. The activity of this enzyme varies widely among individuals, leading to substantial differences in the clearance or metabolic activation of its substrates among patients. This variability is mainly controlled by functionally relevant genetic variations in the *CYP2C19* gene coding for the enzyme.[1–3] In addition, nongenetic factors that include exposure to drugs that directly inhibit the CYP2C19 enzyme or enhance its expression contribute to this variability. Clinical studies suggest that differences in CYP2C19 metabolic status have important clinical implications in terms of efficacy or adverse effects for a number of drugs that include proton pump inhibitors,[8] clopidogrel,[9,10], voriconazole,[11] diazepam,[1,12], cyclophosphamide,[6] thalidomide,[7] and citalopram.[13] It follows that knowledge of CYP2C19 metabolic status is important to optimize therapy and avoid adverse effects of drugs metabolized by this enzyme.

Recently, we and others have tested and demonstrated that stable isotope labeled racemic (±)-[^{13}C]-pantoprazole breath test is a promising tool to assess CYP2C19 metabolic status.[14–16] This test has a number of practical advantages (captures genetic and nongenetic causes of CYP2C19 activity, noninvasiveness, easy and rapid to perform) and has the potential to offer greater clinical utility than existing approaches (e.g. genetic tests). Studies have shown that the pharmacokinetics of stable isotope unlabelled pantoprazole is enantioselective and that the (+)-enantiomer is more dependent on CYP2C19 activity than the (−)-enantiomer of pantoprazole.[17] Whether the metabolism and pharmacokinetics of the stable isotope racemic [^{13}C]-labeled pantoprazole exhibits enantioselectivity similar to that of [^{13}C]-unlabeled pantoprazole has not been determined.

In this study, we have performed additional studies to test the hypothesis that [^{13}C]-labeled pantoprazole is metabolized the same as the [^{13}C]-unlabeled pantoprazole such that its pharmacokinetics is stereoselective. If so, the metabolism of the (+)-[^{13}C]-enantiomer, which is more exclusively catalyzed by CYP2C19 than the (−)-[^{13}C]-enantiomer or racemic mixture may be a better candidate for further development as a CYP2C19 phenotyping tool. The secondary purpose of the study is to relate the pharmacokinetics of each enantiomer with breath test indices.

Methods

In a previous study, we investigated the utility of racemic [^{13}C]-pantoprazole breath test as a marker of CYP2C19 activity.[14] As part of that clinical trial, plasma for pharmacokinetic studies as well as breath test indices and CYP2C19 (*2, *3 and *17) were collected. The present report focuses on determination of enantioselective pharmacokinetics of [^{13}C]-pantoprazole in the plasma samples collected from the completed trial. The trial which was detailed in our previous publication[14] is briefly described below.

Study Subjects

A total of 25 healthy male (10) and female (15) volunteers, of Asian origin (18–49 years old, with body weight of at least 110 pounds and body mass index ≤30) and pre-genotyped for *CYP2C19*2, *3*, and **17* alleles were studied at the outpatient clinic of the Indiana University School of the Medicine Indiana Clinical Research Center (ICRC). This study was approved by the Institutional Review Board of the Indiana University. Investigative Device Exemption application G070004 to conduct the study was also approved by the Food and Drug Administration. This trial was registered at http://www.ClinicalTrials.gov (identifier: NCT00668902). All study subjects provided written informed consent before participation. Details of the inclusion criteria of the participants were published previously.[14]

Study Design

This was an open-label, single-dose clinical trial. Eligible subjects were administered a single 100mg oral dose of (±)-[^{13}C]-pantoprazole sodium-sesquihydrate (4-O-[methyl-^{13}C]-pantoprazole, 99%; CLM-7831-SP; lot#. PR-17177; Cambridge Isotope Laboratories, Inc., Andover, MA) with 2.1 g sodium bicarbonate. Breath samples were collected at baseline and at 2.5, 5, 10, 15, 20, 25, 30, 40, 50, 60, 90, and 120 min post dosing. Venous blood samples (10 ml) were collected at baseline and at 0.5, 1, 1.5, 2, 2.5, 3, 3.5, 4, 5, 6, 8, 10, and 12 h after [^{13}C]-pantoprazole administration. Plasma was separated by centrifugation and stored frozen at −80°C until use.

CYP2C19 Genotyping

Extraction of genomic DNA and genotyping for *CYP2C19*2, *3* and **17* were reported previously.[14] In brief, genomic DNA was extracted from human whole blood with the QIAGEN DNA MiniKit (QIAGEN, Valencia, CA) and genotyping for *CYP2C19*2* (rs4244285), *CYP2C19*3* (rs4986893), and *CYP2C19*17* (rs12248560) by TaqMan Assay-Reagents Allelic Discrimination Kits (Applied Biosystems, Foster City, CA) according to the supplier's instructions.

Quantitation of $^{13}CO_2$

The concentrations of $^{13}CO_2$ and $^{12}CO_2$ in exhaled breath samples were determined using the UBiT-IR300 IR spectrometry (Meretek Diagnostics, Rockville, MD) equipped with interference filters that are wavelength-selective for the absorbance of $^{13}CO_2$ and $^{12}CO_2$. Enrichment of $^{13}CO_2$ in expired air was calculated at each sampling point. The delta over baseline (DOB) in the $^{13}CO_2/^{12}CO_2$ ratio after [^{13}C]-pantoprazole relative to predose (baseline) $^{13}CO_2/^{12}CO_2$ ratio was calculated as described elsewhere by our group.[14]

Measurement of [^{13}C]-Pantoprazole

Plasma concentrations of [^{13}C]-pantoprazole enantiomers were measured using a previously described method, with slight modification.[14] Plasma samples were extracted with 6 ml of ethyl acetate, after the addition of 20 µl of internal standard (50 µg/ml phenacetin) and 250 µl of 2% ammonium hydroxide. The samples were then vortexed and spun and the resultant supernatant was transferred to a new tube to be evaporated. The dried residue was reconstituted with 100 µl of acetonitrile and 50 µl were injected onto the HPLC. Separation

was performed using a Chiralcel OJ-RH column (4.6mmx150mm) (Chiral Technologies Inc. West Chester, PA) and a mobile phase consisting of 75% 50mM sodium perchlorate and 25% acetonitrile (flow rate, 0.5 ml/min). Analytes were detected by UV at 290nm. (Figure 1)

Quantification of [^{13}C]-pantoprazole was performed using a standard curve generated in blank plasma. The limit of quantification of the assay was 0.025 μg/ml. The inter-day and intra-day coefficient of variation (% CV) of the assay was less than 20%.

Analysis of Breath Test Indices and Pharmacokinetics

Breath test indices and pharmacokinetic parameters were determined by fitting the DOB data or plasma concentration data to a standard noncompartmental analysis using WinNonlin professional software (version 5.01; Pharsight, Mountain View, CA) as described before.[14]

Statistical Analysis

Continuous variables were summarized by groups using descriptive statistics. Differences in pharmacokinetic parameters and breath test indices among different genotypes of CYP2C19 were analyzed by the ANOVA test with Dunnett's multiple comparison post test assuming the patients were sampled from a Gaussian population. Pearson's correlation analysis was performed to determine relationships between breath indices and pharmacokinetic parameters. All statistical tests were conducted using PASW Statistics 17.0 (SPSS Software Inc., Chicago, IL). A p-value of $p < 0.05$ was considered statistically significant.

Results

In this study, 24 subjects had both pharmacokinetic and breath test indices and were included for analysis. The genotype predicted CYP2C19 phenotypes were: 4 PM (*2/*2), 10 IM (*1/*2, n=9; and *1/*3, n=1) and 10 EM (*1/*1, n=9; and *1/*17, n=1). One poor metabolizer (PM) of CYP2C19 subject that was included in the previous publication[14] was excluded because of unavailability of plasma samples. The demographic characteristics among the different genotypes were provided previously.[14]

The plasma concentration time profile of [^{13}C]-pantoprazole enantiomers in the three groups of genotypes is shown in Figure 2. The corresponding pharmacokinetic parameters are listed in Table 1. Statistically significant differences in the elimination half-life ($p<0.001$), $AUC_{(0-\infty)}$ (<0.05) and weight adjusted oral clearance (<0.05) of (−)-[^{13}C]-pantoprazole were observed among the 3 genotypes [PM (n=4), IM (n=10), EM (n=10)]. Post-hoc analysis showed that PMs had a significantly longer half-life, higher $AUC_{(0-\infty)}$ and lower oral clearance than EM subjects; no difference was observed between IMs versus PMs or IMs versus EMs except for the elimination half-life (Table 1), but these did not reach statistical significance. A more pronounced effect of CYP2C19 genotypes was observed on the elimination parameters of (+)-[^{13}C]-pantoprazole (Table 1). The elimination half-life, $AUC_{(0-\infty)}$, and weight adjusted oral clearance of (+)-[^{13}C]-pantoprazole were markedly and significantly different among the three genotype groups ($p<0.001$; ANOVA). Post-hoc analysis revealed that PM subjects had significantly higher AUC ($p < 0.001$), longer half-life ($p < 0.001$) and lower oral clearance ($p < 0.01$) compared to EM of CYP2C19; higher AUC

($p < 0.001$) and longer half-life ($p < 0.001$) were also observed in PM compared to IM, but the oral clearance was not statistically significant between these groups (Table 1).

The impact of genotype on ratios of (+)−/(−)-[^{13}C]-pantoprazole concentrations is shown (Figure 3). The stereoselectivity is much more pronounced in the PM group than EM and IM groups. Accordingly, the AUC ratios of (+)−/(−)-[^{13}C]-pantoprazole (with 95% CI) were 3.49 (2.26,4.72), 0.77 (0.72,0.84) and 0.62 (0.52,0.73) in PM, IM and EM respectively; similarly the ratios of the elimination half-lives were 2.99 (1.89,4.09), 0.78 (0.62,0.94) and 0.75 (0.54,0.97) respectively. No stereoselectivity was observed with regard to Vd/F. (+)-[^{13}C]-pantoprazole was more dependent on CYP2C19 function than that of (−)-[^{13}C]-pantoprazole was. Thus, the impact of PM on (+)-[^{13}C]-pantoprazole elimination was much greater than IM and EMs compared to the (−)-[^{13}C]-pantoprazole counterpart. As a result, there is a strong gene-dose effect in (+)-[^{13}C]-pantoprazole more so than with (−)-[^{13}C]-pantoprazole.

The results of the (±)-[^{13}C]-pantoprazole breath test reported previously[14] were recalculated and summarized in Table 2. As with the PK parameters, the breath test indices were significantly affected by CYP2C19 genotypes (Table 2). Correlation analyses between pharmacokinetic parameters and breath test indices are listed in Table 3. Breath test indices show no or marginal correlation with (−)-[^{13}C]-pantoprazole elimination parameters, while statistically significant correlations were observed with the elimination of (+)-[^{13}C]-pantoprazole enantiomer, again pointing towards a much bigger role of CYP2C19 in (+)-[^{13}C]-pantoprazole elimination and breath test indices. For example, DOB_{max} and $AUC_{(0\text{-}120)}$ were more significantly correlated with $AUC_{(0\text{-}\infty)}$ of (+)-[^{13}C]-pantoprazole (Pearson correlations: −0.46 & −0.68) than they were with (−)-[^{13}C]-pantoprazole (Pearson correlations: −0.40 & −0.35 for DOB_{max} and $AUC_{(0\text{-}120)}$, respectively) (Tables 3 & Figure 4).

Discussion

In the present study, we have demonstrated that (±)-[^{13}C]-pantoprazole showed stereoselective metabolism, the (+)-[^{13}C]-pantoprazole is preferentially catalyzed by CYP2C19 and that breath test indices correlate best with the elimination of (+)-[^{13}C]-pantoprazole more than that of (−)-[^{13}C]-pantoprazole. These results demonstrate that stable isotope labeled and unlabeled pantoprazole are metabolized by the same mechanism. Incorporation of the [^{13}C]-label at the O-methyl site of pantoprazole does not alter patterns of stereoselective metabolism of pantoprazole. The results further suggest that (+)-[^{13}C]-pantoprazole breath test may be a better selective probe of CYP2C19 activity than the racemic mixture.

That CYP2C19 is the major clearance mechanism (up to 80% of the administered dose) for pantoprazole and other proton pump inhibitors has been well established.[18–20] Pantoprazole is O-demethylated primarily by CYP2C19 to 4-hydroxypantoprazole and a methyl group.[18,20] The hydroxylated metabolite formed undergoes rapid sulfation, while the methyl group passes through the carbon pool before it eventually traverses to the lung and is exhaled.[14,18] A minor pathway of pantoprazole involves CYP3A-mediated sulfoxidation to

pantoprazole sulfone. In a previous report[14] we have shown that the (±)-[^{13}C]-pantoprazole breath test indices and pharmacokinetics were significantly associated with CYP2C19 genotypes, suggesting a major role of CYP2C19 in the metabolism of (±)-[^{13}C]-pantoprazole and isotope unlabeled pantoprazole. Since pantoprazole is a chiral molecule and its pharmacokinetics show enantioselectivity,[21] we tested in this study whether (±)-[^{13}C]-pantoprazole metabolism is also enantioselective. Our data show that, indeed, the metabolism of (±)-[^{13}C]-pantoprazole is enantioselective and that this is mainly dependent on CYP2C19 activity. These data and our previous report suggest that incorporation of the [^{13}C]-label at the O-methyl site of pantoprazole alters neither the metabolic patterns of racemic (±)-[^{13}C]-pantoprazole nor its stereoselective elimination.

The pattern of stereoselectivity of the [^{13}C]-labeled pharmacokinetic data from the present study are broadly consistent with those previously reported for the isotope unlabeled pantoprazole by Tanaka et al.[21] Specifically, the elimination of (+)-[^{13}C]-pantoprazole was more dependent on CYP2C19 activity in both studies compared to that of (−)-[^{13}C]-pantoprazole. These data concur with the fact that (+)-[^{13}C]-pantoprazole is predominantly catalyzed by CYP2C19-mediated O-demethylation in both studies, while CYP3A-mediated sulfoxidation contributes significantly to the metabolism of (−)-[^{13}C]-pantoprazole.[18,19] Consistent with the major role of CYP2C19 in (+)-[^{13}C]-pantoprazole elimination compared to that of (−)-[^{13}C]-pantoprazole, better correlation of (±)-[^{13}C]-pantoprazole breath test indices were observed with the elimination of (+)-[^{13}C]-pantoprazole than with that of (−)-[^{13}C]-pantoprazole when all data were considered or PMs were excluded. It is also interesting to note that both the elimination parameters of (+)-[^{13}C]-pantoprazole and breath test indices are more significantly associated with CYP2C19 genetic variation compared to those of (−)-[^{13}C]-pantoprazole. Despite this, it is interesting to note that the correlations of breath test indices with the exposure of the (+)-enantiomer (Pearson r= −0.46 and −0.68) are not perfect. Clearly, our data show that the breath test is generated from both enantiomers when (±)-[^{13}C]-pantoprazole is administered. In addition, the [^{13}C] may be distributed with time in to the carbon pool of the body and contributing to the relatively lower correlation. It is also possible that sequential metabolism via the sulfone metabolite (O-demethylation) also contribute to this. Nevertheless, these data suggest that (+)-[^{13}C]-pantoprazole is the main source of $^{13}CO_2$ exhaled in the lung. Therefore, O-demethylation is the primary clearance mechanism of (+)-[^{13}C]-pantoprazole, whereas other pathways such as CYP3A-mediated sulfoxidation appear to significantly contribute to the metabolism of (−)-[^{13}C]-pantoprazole.

Some of the pharmacokinetic parameters derived from this study [shorter t_{max}, higher C_{max} and higher $AUC_{(0\text{-}\infty)}$] were different from the study by Tanaka et al.[17] However, it is difficult to directly compare our values with those of Tanaka et al. First, the doses used were different (100 mg in this study versus 40 mg in the other study). Second, the formulations used were different (solution in this study versus enteric coated tablet in the Tanaka study). The t_{max} values in this study (0.5 hr) was consistently lower than in the other study, which agrees well with the difference in formulations and thus rate of absorption. Third, IM and EM were analyzed as EM in the Tanaka study [19] while in our case IM and EM of CYP2C19 were analyzed separately. Additional factors that may contribute to this discrepancy may include the way the phenotypic groups were classified (genotype-predicted phenotype in our study and S-mephenytoin probe activity in the other study.[21])

Concluding Remarks

In summary, we have shown that the metabolism of isotope [^{13}C]-stable labeled pantoprazole and unlabeled pantoprazole is similar qualitatively and quantitatively, providing a rationale for using the O-demethylation of [^{13}C]-labeled pantoprazole as a noninvasive novel biomarker of CYP2C19 activity. Previously, we have performed a feasibility study showing that (±)-[^{13}C]-pantoprazole breath test is a reliable marker of CYP2C19 activity in healthy volunteers. While this test completely discriminated PM from EM and IM of CYP2C19, there was a significant overlap between EM and IM of CYP2C19, which could be due to differences in the quantitative contribution of CYP2C19 and pathways towards the different enantiomers. The data from the present study indicate that (+)-[^{13}C]-pantoprazole is more exclusively metabolized by CYP2C19 than (−)-[^{13}C]-pantoprazole. Studies are ongoing to test whether (+)-[^{13}C]-pantoprazole breath test is superior to (±)-[^{13}C]-pantoprazole breath test to probe CYP2C19 activity.

Acknowledgments

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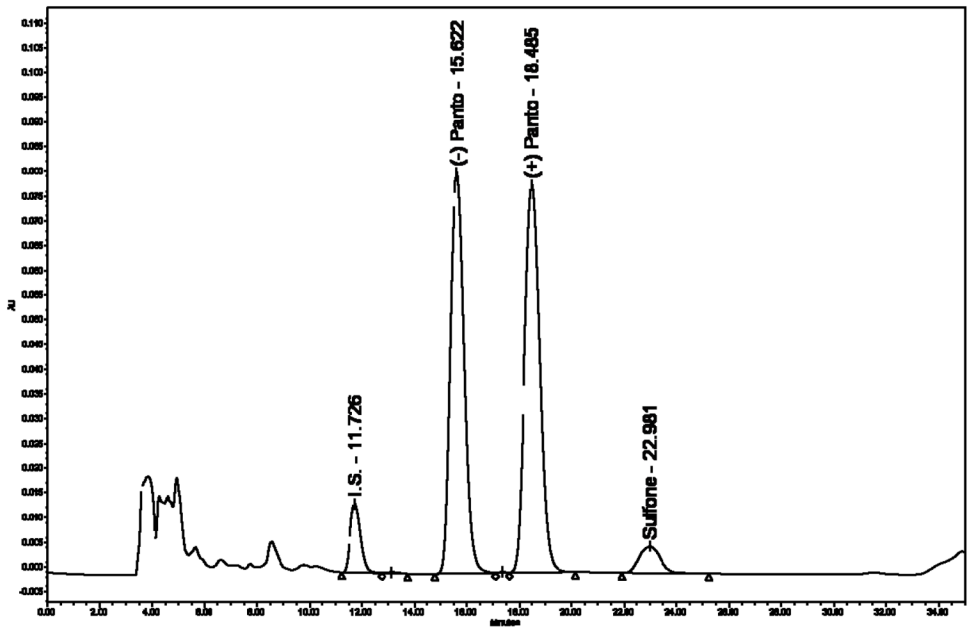
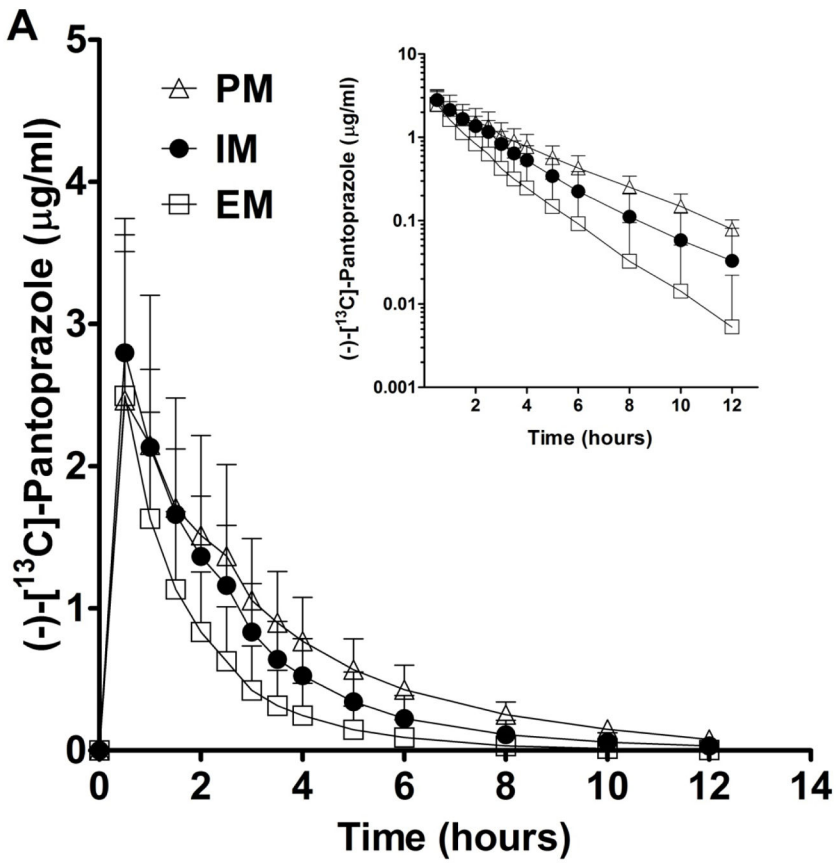


Figure 1.

Representative chromatogram of an EM, showing the separation of both the (−)-[^{13}C]-pantoprazole and the (+)-[^{13}C]-pantoprazole enantiomers.



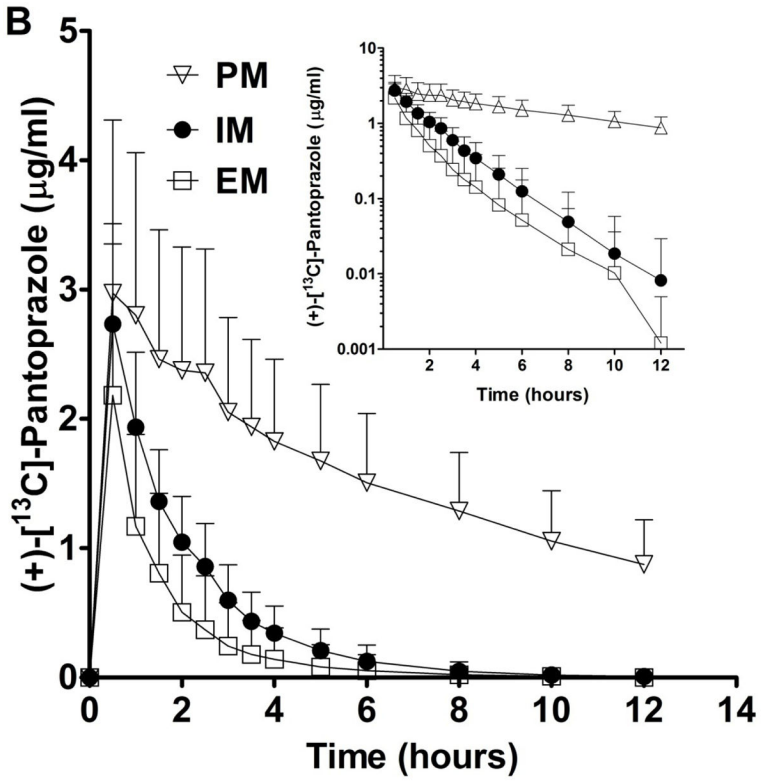


Figure 2.

Mean (±S.D.) of the plasma concentration versus time curves of (−)-[^{13}C]-pantoprazole **(A)** and (+)-[^{13}C]-pantoprazole **(B)** after administration of a single 100mg oral dose of (±)-[^{13}C]-pantoprazole sodium-sesquihydrate to healthy volunteers with PM (n=4), IM (n=10) and EM (n=10) genotypes of CYP2C19.

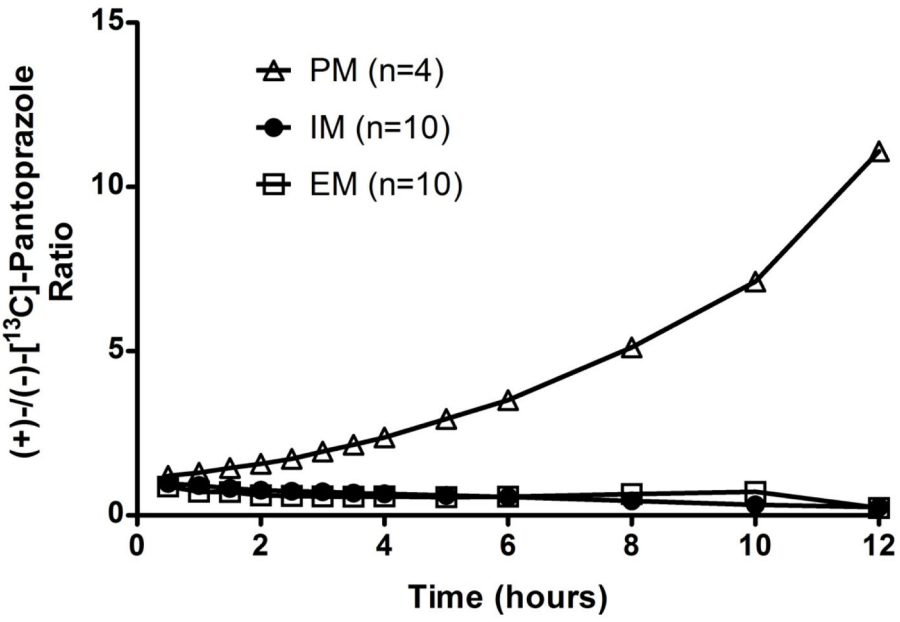


Figure 3.

Mean (±S.D.) ratios of plasma concentrations of (+)-[^{13}C]-pantoprazole/(−)-[^{13}C]-pantoprazole versus time after administration of a single 100mg oral dose of (±)-[^{13}C]-pantoprazole sodium-sesquihydrate to healthy volunteers with PM (n=4), IM (n=10) and EM (n=10) genotypes of CYP2C19.

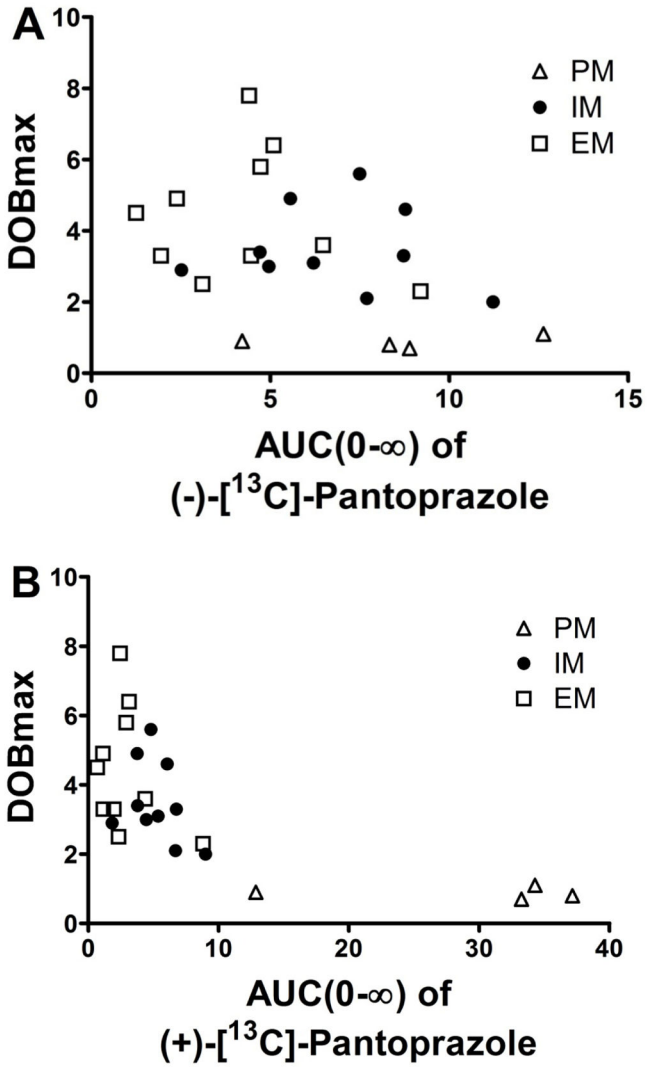


Figure 4.

Correlations of Maximal Delta Over Baseline (DOBmax) with $AUC_{(0-\infty)}$ of (−)-[^{13}C]-pantoprazole **(A)** and of (+)-[^{13}C]-pantoprazole **(B)** after administration of a single 100mg oral dose of (±)-[^{13}C]-pantoprazole sodium-sesquihydrate to healthy volunteers with PM (n=4), IM (n=10) and EM (n=10) genotypes of CYP2C19.

Table 1

Mean (95% C.I.) pharmacokinetic parameters of (−)-[^{13}C]-pantoprazole and (+)-[^{13}C]-pantoprazole after a single 100mg oral dose of (±)-[^{13}C]-pantoprazole in PM, IM and EM genotypes.

	(−)-Pantoprazole				(+)-Pantoprazole			
	PM (n=4)	**IM (n=10)**	**EM (n=10)**	**p Value[a]**	**PM (n=4)**	**IM (n=10)**	**EM (n=10)**	**p Value[a]**
t_{max} (hr)	0.50 (0.50,0.50)	0.50 (0.50,0.50)	0.50 (0.50,1.00)	0.52	0.50 (0.50,2.00)	0.50 (0.50,0.50)	0.50 (0.50,1.00)	0.12
C_{max} (µg/ml)	2.46 (0.61,4.32)	2.80 (2.29,3.31)	2.57 (1.69,3.46)	0.82	2.98 (0.87,5.09)	2.73 (2.18,3.29)	2.06 (1.19,2.92)	0.24
$t_{1/2}$ (hr)	2.6** (2.1,3.1)	1.7$^{@}$ (1.3, 2.1)	1.2$^{\#\#\#}$ (0.9,1.5)	< 0.001	7.7*** (5.3,10.1)	1.3 (1.0,1.6)	0.9$^{\#\#\#}$ (0.6,1.3)	< 0.001
$AUC_{(0-\infty)}$ (µg* hr/ml)	8.52 (3.03,14.00)	6.79 (5.00,8.58)	4.31$^{\#}$ (2.63,5.98)	< 0.05	29.38*** (11.65,47.11)	5.25 (3.82,6.68)	2.90$^{\#\#\#}$ (1.21,4.58)	< 0.001
$AUC_{(0-12)}$ (µg* hr/ml)	8.20 (2.81,13.57)	6.69 (4.98,8.40)	4.28$^{\#}$ (2.63,5.93)	< 0.05	19.32*** (8.00,30.64)	5.20 (3.81,6.60)	2.87$^{\#\#\#}$ (1.21,4.54)	< 0.001
V_d/F (L)	26.6 (0.6,52.6)	18.8 (16.4,21.2)	24.7 (15.8,33.7)	0.33	21.8 (4.3,39.4)	19.0 (14.2,23.8)	34.2 (14.2,54.1)	0.22
Cl/F (L/hr)	6.86 (1.37,12.36)	8.67 (5.50,11.83)	15.87 (8.40,23.34)	0.06	2.05 (0.10,4.00)	11.35$^{@}$ (6.95,15.76)	27.73$^{\#\#}$ (13.45,42.01)	< 0.01
Cl/F* kg (L/hr* kg)	0.11 (0.02,0.20)	0.13 (0.08,0.19)	0.23 (0.15,0.31)	< 0.05	0.03 (0.002,0.07)	0.18$^{@}$ (0.10,0.25)	0.40$^{\#\#}$ (0.23,0.56)	< 0.001

PM vs IM :

* = p<0.05,

** = p<0.01,

*** = p<0.001

IM vs EM :

$^{@}$ = p<0.05,

$^{@@}$ = p<0.01,

$^{@@@}$ = p<0.001

PM vs EM :

$^{\#}$ = p<0.05,

$^{\#\#}$ = p<0.01,

$^{\#\#\#}$ = p<0.001

^{a} = One-Way ANOVA with Dunnett 2-Sided

§ t_{max} [Median (min,max)]

Table 2

Mean (±95% C.I.) breath test parameters after a single 100mg oral dose of (±)-[^{13}C]-pantoprazole in PM, IM and EM genotypes.

	PM (n=4)	IM (n=10)	EM (n=10)	p Value[a]
t_{max} (minutes)[§]	27.5 (10,50)	30.0 (25,60)	30.0 (20,50)	0.50
Maximum Delta Over Baseline (DOB_{max})	0.88** (0.60,1.15)	3.49 (2.64,4.34)	4.44### (3.16,5.72)	< 0.01
$AUC_{(0-120)}$ (minutes* DOB)	56.07** (19.85,92.29)	315.40 (242.7,388.1)	378.22### (282.7,473.8)	< 0.001
DOB_{max}/Kg	0.0143 (0.01,0.02)	0.0555 (0.03,0.08)	0.0695# (0.04,0.10)	< 0.05

PM vs IM :

* = p<0.05,

** = p<0.01,

*** = p<0.001

IM vs EM :

@ = p<0.05,

@@ = p<0.01,

@@@ = p<0.001

PM vs EM :

= p<0.05,

= p<0.01,

= p<0.001

[a] = One-Way ANOVA with Dunnett 2-Sided

[§] t_{max} [Median (min,max)]

Table 3

Correlations between pharmacokinetic parameters of [^{13}C]-pantoprazole enantiomers and breath test indices.

Pearson (r) (p value)	Cl/F*kg (−)(L/hr*kg)	$AUC_{(0-\infty)}$ (+)(μg*hr/ml)	Cl/F*kg (+)(L/hr*kg)	DOBmax Breath	$AUC_{(0-120)}$ Breath (min*DOB)
$AUC_{(0-\infty)}$ (−)(μg*hr/ml)	−0.85 (<0.001)	0.63 (<0.001)	−0.80 (<0.001)	−0.40 (0.053)	−0.35 (0.089)
Cl/F*kg (−)(L/hr*kg)		−0.46 (<0.05)	0.94 (<0.001)	0.30 (0.150)	0.25 (0.238)
$AUC_{(0-\infty)}$ (+)(μg*hr/ml)			−0.58 (<0.05)	−0.62 (<0.001)	−0.68 (<0.001)
Cl/F*kg (+)(L/hr*kg)				0.48 (<0.05)	0.44 (<0.05)
DOBmax					0.98 (<0.001)

Data were analyzed using Pearson's Correlation Test. P<0.05 was considered significant.