Results: Following a single intravenous or oral administration of pantoprazole, omeprazole, or lansoprazole at a dose of 5 mg/kg, systemic exposure (i.e., AUC) to total radioactivity (i.e., parent drug + metabolites) and parent drug were the greatest for pantoprazole. Further, oral bioavailability of total radioactivity or parent drug was the greatest for pantoprazole. Following a single intravenous administration of pantoprazole, omeprazole, or lansoprazole at 5 mg/kg, greater than 75% of the parent compound was metabolized. Following a single oral administration of pantoprazole, omeprazole, or lansoprazole at 5 mg/kg, greater than 88% of the parent compound was metabolized. Following a single intravenous or oral dose, half-life of the parent was greatest for pantoprazole; although, half-life of total radioactivity was shortest for pantoprazole. With repeated dosing of pantoprazole at 50 or 200 mg/kg/day, omeprazole at 50 or 141 mg/kg/day, or lansoprazole at 50 mg/kg/day, systemic exposure to total radioactivity (parent compound + metabolites) on days 1 and 7 was relatively constant. With repeated dosing of pantoprazole at 50 and 200 mg/kg/day, systemic exposure to the parent drug on day 7 was 74.6 and 74.5% of systemic exposure on day 1. This would suggest that bioavailability of the parent compound remained relatively constant over a dose range of 50 to 200 mg/kg/day for the 7-day treatment period. With repeated dosing of omeprazole at 50 mg/kg/day, systemic exposure to the parent compound on day 7 was 75% of systemic exposure on day 1; however, at a dose of 141 mg/kg/day, systemic exposure to the parent compound on day 7 had declined to 35.8% of systemic exposure on day 1. The half-life for the omeprazole parent compound on day 7 was 3 times that observed on day 1, which does not appear to correlate with decreased systemic exposure and bioavailability of the parent compound. For lansoprazole at 50 mg/kg/day, systemic exposure to parent drug on day 7 was 45% of systemic exposure on day 1. Pharmacokinetic parameters determined in this study have no value in the interpretation of the carcinogenicity study with pantoprazole in Sprague Dawley rats. Interpretation of the carcinogenicity study was based upon toxicity endpoints and not exposure. Further, the treatment period was only 7 days in the present study and the validity of extrapolating these results to a 2-year study is highly questionable. The observed oral bioavailabilities for the unchanged drugs following single doses are unusually high as compared to other studies in the present submission with pantoprazole or published values for omeprazole and lansoprazole. Further, it should be emphasized that total systemic exposure, consisting of parent drug + metabolites, on days 1 and 7 was relatively similar for each of the three compounds.

Plasma pharmacokinetic parameters for total radioactivity following a single intravenous or oral administration of pantoprazole, omeprazole, or lansoprazole at dose of 5 mg/kg.

Drug	C_{max}, mg-eqv/L		T_{max}, hr		T½, hr		$AUC_{0-\infty}$, mgeqv* hr/L		Oral Bioavailability,%
	IV[A]	Oral	IV	Oral	IV	Oral	IV	Oral	
Pantoprazole	16.658	7.386	-	0.25	3.90	4.40	42.15	27.84	66.05
Omeprazole	11.003	2.237	-	0.25	7.2	7.0	6.45	3.57	55.35
Lansoprazole	6.705	1.339	-	0.29	10.7	9.6	8.58	3.80	44.29

A. Extrapolated value.

Plasma pharmacokinetic parameters for parent compound following a single intravenous or oral administration of pantoprazole, omeprazole, or lansoprazole at dose of 5 mg/kg.

Drug	C_{max}, mg/L		T_{max}, hr		T½ , hr		$AUC_{0-\infty}$, mg*hr/L		Oral Bioavailability,%
	IV[A]	Oral	IV	Oral	IV	Oral	IV	Oral	
Pantoprazole	12.308	3.855	-	0.25	0.47	0.59	10.72	3.35	31.25
Omeprazole	8.522	0.858	-	0.25	0.14	0.13	1.69	0.43	25.44
Lansoprazole	5.863	0.283	-	0.25	0.20	0.19	1.54	0.17	11.04

A. Extrapolated value.

Plasma pharmacokinetic parameters for total radioactivity on days 1 or 7 following oral administration of pantoprazole (50 or 200 mg/kg/day), omeprazole (50 or 141 mg/kg/day), or lansoprazole (50 mg/kg/day). For repeat oral dose studies with pantoprazole, omeprazole, and lansoprazole, ^{14}C-radiolabeled drug was administered on days 1 and 7. Unlabeled drug was administered on days 2 through 6.

Drug	Dose, mg/kg/day	C_{max}, mg-eqv/L		T_{max}, hr		T½, hr		$AUC_{0-\infty}$, mg-eqv*hr/L	$AUC_{144-168hr}$, mg-eqv*hr/L
		Day 1	Day 7	Day 1	Day 7	Day 1	Day 7	Day 1	Day 7
Pantoprazole	50	83.501	67.903	0.38	144.46	4.2	4.5	377.38	327.96
	200	155.03	155.496	0.92	145.17	5.1	5.9	1206.1	1098.9
Omeprazole	50	26.660	17.867	0.25	144.29	6.3	8.0	60.51	50.48
	141	46.039	27.56	0.29	144.29	7.3	7.3	158.03	138.82
Lansoprazole	50	9.814	5.101	0.33	144.33	10.6	10.6	40.23	35.66

Plasma pharmacokinetic parameters for parent compound on days 1 or 7 following oral administration of pantoprazole (50 or 200 mg/kg/day), omeprazole (50 or 141 mg/kg/day), or lansoprazole (50 mg/kg/day). For repeat oral dose studies with pantoprazole, omeprazole, and lansoprazole, ^{14}C-radiolabeled drug was administered on days 1 and 7. Unlabeled drug was administered on days 2 through 6.

Drug	Dose, mg/kg/day	C_{max}, mg/L		T_{max}, hr		T½, hr		$AUC_{0-\infty}$, mg*hr/L	$AUC_{144-168hr}$, mg*hr/L
		Day 1	Day 7	Day 1	Day 7	Day 1	Day 7	Day 1	Day 7
Pantoprazole	50	47.634	35.139	0.25	144.25	0.80	1.10	63.66	47.49
	200	83.133	70.285	0.38	144.29	1.20	1.51	170.76	126.92
Omeprazole	50	1.862	1.069	0.25	144.54	0.84	1.24	2.00	1.50
	141	6.966	2.328	0.58	144.83	0.91	2.90	18.08	6.48
Lansoprazole	50	1.660	0.805	0.25	144.29	1.00	0.62	2.13	0.96

Stereoselective Chiral Inversion of Pantoprazole Enantiomers in Rats (GTR-32139).

Methods: (±)Pantoprazole [(±)-PAN] is a chiral sulfoxide, which is used clinically as a racemic mixture. The disposition kinetics of (+)-PAN and (-)-PAN and its metabolites, pantoprazole sulfone (PAN-SO2), pantoprazole sulfide (PAN-S), 4'-O-demethylated pantoprazole sulfone (PAN-SO2(OH)), and 4'-O-demethylated pantoprazole sulfide (PAN-S(OH)) were evaluated in male Sprague Dawley rats following oral or intravenous

administration of each enantiomer. There were 4 groups of 18 rats each. (+)-PAN and (-)-PAN were synthesized and the optical purity of each enantiomer was > 99.0%. Each enantiomer was dissolved in saline containing 0.5% 1 N NaOH and 5% ethanol and administered by either the oral or intravenous route as a single dose of 10 mg/kg. The dose volume was 5 mL/kg. Blood samples were collected at 0.083, 0.5, 1, 2, 4, and 8 hr after dosing using 3 animals/time point. Serum was prepared and stored at -20°C until analysis. The enantiomers, (+)-PAN and (-)-PAN), were separated on a cellulose-base chiral stationary phase [redacted] Enantiomers were detected by [redacted] The lower limit of quantitation for enantiomers was [redacted] Metabolites in rat serum were measured using a [redacted] method. The analytical column was a [redacted] and metabolites were eluted using a [redacted] Metabolites were detected by [redacted] The lower limits of quantitation for PAN-SO_2 (OH), PAN-S(OH), PAN-SO_2, PAN, and PAN-S were all determined to be [redacted] µg/mL.

Results: Serum pharmacokinetic parameters of (+)-PAN and (-)-PAN following oral or intravenous administration were relatively similar. The major metabolite following oral or intravenous administration of (+)-PAN or (-)-PAN was PAN-SO_2, followed by smaller levels of PAN-SO_2(OH), and negligible to nonexistent levels of PAN-S and PAN-S(OH). Following oral or intravenous administration of (+)-PAN to male rats, there was significant chiral interconversion to form (-)-PAN. However, following oral or intravenous administration of (-)-PAN to male rats, there was no evidence of chiral interconversion to form (+)-PAN. The mechanism for this stereoselective chiral interconversion is unknown; however, the sponsor has speculated that formation of (-)-PAN from (+)-PAN may be due to a reversible reduction/oxidation reaction between the sulfoxide and sulfide or between the sulfoxide and sulfone.

Pharmacokinetic parameters following intravenous administration of (+)-PAN or (-)-PAN to male rats at a dose of 10 mg/kg.

Parameter	Intravenous Administration	
	(+)-PAN	(-)-PAN
AUC_{0-8hr}, µg*hr/mL	15.2	16.5
T½, hr	0.258	0.345
MRT, hr	0.310	0.482
Cl_{TOTAL}, mL/min/kg	11.0	10.1

Pharmacokinetic parameters following oral administration of (+)-PAN or (-)-PAN to male rats at a dose of 10 mg/kg.

Parameter	Oral Administration	
	(+)-PAN	(-)-PAN
T_{max}, hr	0.50	0.08
C_{max}, hr	9.56	9.50
AUC_{0-8hr} µg*hr/mL	9.38	9.87
MRT, hr	0.748	0.935
Cl_{TOTAL}/F, mL/min/kg	17.8	16.1

$AUC_{0\text{-}8hr}$ (μg*hr/mL) of (+)-PAN and (-)-PAN after oral or intravenous administration of (+)-PAN or (-)-PAN to male rats at a dose of 10 mg/kg.

Route	Enantiomer	$AUC_{0\text{-}8hr}$ (μg*hr/mL)		Apparent inversion ratio (%)
		(+)-PAN	(-)-PAN	
Oral	(+)-PAN	9.38	3.66	28.1
	(-)-PAN	0.00	9.87	0
Intravenous	(+)-PAN	15.2	8.66	36.3
	(-)-PAN	0.00	16.5	0

$AUC_{0\text{-}8hr}$ (μg*hr/mL) of PAN-SO_2, PAN-S, PAN-SO_2(OH), and PAN-S(OH) after oral or intravenous administration of (+)-PAN or (-)-PAN to male rats at a dose of 10 mg/kg.

Route	Enantiomer	$AUC_{0\text{-}8hr}$ (μg*hr/mL)			
		PAN-SO_2	PAN-S	PAN-SO_2 (OH)	PAN-S (OH)
Oral	(+)-PAN	15.6	0	1.20	0
	(-)-PAN	17.6	0.09	1.02	0
Intravenous	(+)-PAN	13.4	0	0.676	0
	(-)-PAN	20.3	0	1.45	0

Dog

Kinetics and Bioavailability of Pantoprazole in Dog After Single Oral Doses of the Solution, and Uncoated and Enteric Coated Tablets (GTR-31190).

Methods: Toxicokinetics and bioavailability of pantoprazole in dogs were examined following single oral administration of the drug in either a 1% aqueous solution at pH 10.5, uncoated tablets, or enteric-coated tablets. The theoretical dose of pantoprazole in each formulation was 10 mg/kg. The dose volume for the solution was 1 mL/kg followed by 30 mL of water. Uncoated and enteric-coated tablets were administered in gelatin capsules followed by 50 mL of water. Six beagle dogs received the three formulations of pantoprazole in a randomized schedule. There was a washout period of 2 weeks between administration of each formulation. Blood for determination of serum levels of pantoprazole and its sulfone metabolite were collected at 0 (predose), 0.25, 0.50, 0.75, 1, 1.5, 2, 2.5, 3, 3.5, 4, 4.5, 5, 6, 7, 8, 10, and 24 hr after dosing. Pantoprazole and its corresponding sulfone (B8610-14) were measured by [redacted]. Urine for determination of levels of pantoprazole and metabolite levels was collected prior to treatment and from 0 to 24 hr after dosing (results will be the subject of a separate report).

Results: Systemic exposure to pantoprazole was in the following order: uncoated tablets > enteric-coated tablets > solution. Variability in systemic exposure to pantoprazole was smallest for the uncoated tablets. The uncoated tablets contained sodium carbonate, which may have protected pantoprazole from acid degradation in the stomach. Absorption of pantoprazole from solution and uncoated tablets were relatively rapid. Absorption of pantoprazole was delayed for the enteric-coated tablets since these tablets must pass from the stomach to the duodenum before disintegration. Higher variability in systemic exposure to pantoprazole observed with enteric-coated tablets may have been related to slow dissolution of the coating and release of drug in deeper parts of the intestine, where absorption was slower. Elimination half-lives of pantoprazole with the three formulations were similar at 0.7-0.8 hr. Systemic exposure to the sulfone metabolite was in the following

order: solution > uncoated tablets > enteric-coated tablets. The T_{max} for the sulfone with the enteric-coated tablets was longer than the other two formulations due to slow absorption of the parent drug. Elimination half-lives of the sulfone metabolite with the three formulations were similar at 2.8-3.2 hr.

Toxicokinetic parameters of serum pantoprazole and its sulfone metabolite in dogs following single oral administration of the drug at a dose of 10 mg/kg in either a 1% aqueous solution at pH 10.5, uncoated tablets, or enteric-coated tablets.

Parameter	Solution		Uncoated Tablets		Enteric-Coated Tablets	
	Pantoprazole	Sulfone	Pantoprazole	Sulfone	Pantoprazole	Sulfone
C_{max}, µg/mL	20.4	4.24	20.9	5.17	15.2	3.75
T_{max}, hr						
$AUC_{0-\infty}$, µg*hr/mL	29.7±7.36	36.96	35.1±2.53	36.72	32.2±5.7	23.48
T½, hr	0.7	2.8	0.7	3.2	0.7	3.1
Pantoprazole Bioavailability, % (solution as reference)			140		120	
Pantoprazole Bioavailability, % (uncoated as reference)					86	

Pharmacokinetics of Pantoprazole and Its Sulfone Metabolite in the Dog Following Single and Repeated Oral and Intravenous Doses (GTR-31546).

Methods: The serum pharmacokinetic of pantoprazole and its major metabolite, a sulfone — 97167) were characterized following single and repeated oral or intravenous administration of pantoprazole to beagle dogs. Pantoprazole was administered by the oral route at doses of 40 and 160 mg/animal/day and the intravenous route at 60 mg/animal/day for periods ranging from 15 to 18 days. There were 2 dogs/sex/group. For the oral route, pantoprazole was administered as subcoated 40-mg tablets. For the intravenous route, pantoprazole was administered as a bolus with a dose volume of 15 mL/animal. Serum levels of pantoprazole and its sulfone metabolite were measured on days 1 and 10. Blood for determination of serum levels of pantoprazole and its sulfone metabolite were collected at 0, 0.25, 0.50, 1, 1.5, 2, 3, 4, 6, and 24 hr after dosing. On days 15 to 18, liquor cerebrospinalis and vitreous fluid were collected 3 hr after the last dose for determination of levels of pantoprazole and its sulfone metabolite. Measurement of serum concentrations of pantoprazole and its sulfone metabolite was performed using a — method with — Liquor cerebrospinalis and vitreous fluid samples were —

Results: Following oral administration of doses at 40 or 160 mg/animals/day, serum AUC and C_{max} values for pantoprazole and its sulfone metabolite on days 1 and 10 were not proportional to dose. Serum AUC and C_{max} values for pantoprazole and its sulfone metabolite on day 1 were greater than those observed on day 10. However, clearance and volume of distribution values measured following intravenous administration of 60 mg/animal/day on day 10 were almost twice values observed on day 1. On day 10, elimination of the drug was twice that observed on day 1. Clearance values of 0.54 and 0.99 L/hr/kg on days 1 and 10, respectively, were slightly less than hepatic plasma flow at 1.06 L/hr/kg.

Volume of distribution values exceeded blood volume (0.09 L/kg) suggesting distribution into tissues. Bioavailability of pantoprazole with oral doses at 40 and 160 mg/kg/day based upon the AUC value obtained with an intravenous dose of 60 mg/animal/day were 44 and 146.5%, respectively. The bioavailability of pantoprazole exceeding 100% with an oral dose of 160 mg/animal/day was attributed to the small number of animals used in each group as well as different individual animals in each group. Pantoprazole concentrations in liquor cerebrospinalis and vitrous fluid at 3 hr after an oral dose of 160 mg/animal/day were 0.117 and 0.151 mg/L, respectively. Pantoprazole was not detected in liquor cerebrospinalis or vitrous fluid following administration of an oral dose at 40 mg/animal/day or an intravenous dose at 60 mg/animal/day.

Serum pharmacokinetics of pantoprazole (Panto) and its sulfone metabolite (Sulfo) on days 1 and 10 following administration by the oral route at 40 or 160 mg/animal/day or by the intravenous route at 60 mg/animal/day.

Day	Dose, mg/kg/day and Route	AUC mg*hr/L		C_{max}, mg/L		T_{max}, hr		T½, hr		CL, L/hr/kg	Vd, L/kg
		Panto	Sulfo	Panto	Sulfo	Panto	Sulfo	Panto	Sulfo	Panto	Panto
1	40, Oral	3.22	3.26	2.12	0.74	1.0	2.00	0.78	2.18	-	-
	160, Oral	41.30	36.46	22.54	8.03	0.50	2.50	0.64	-	-	-
	60, IV	10.63	6.69	-	1.62	-	1.50	0.43	1.95	0.554	0.322
10	40, Oral	1.17	1.17	0.69	0.26	0.50	2.63	1.06	-	-	-
	160, Oral	20.67	15.68	9.03	3.54	0.75	2.75	0.83	2.71	-	-
	60, IV	6.43	5.28	-	1.24	-	1.60	0.45	2.95	0.998	0.558

Monkey

Pantoprazole : (^{14}C)-B8610-023: A Study of Absorption, Distribution, and Excretion Following Oral and Intravenous Administration to the Cynomolgus Monkey (GTR-31549).

Methods: The absorption, distribution, and excretion of pantoprazole were examined in cynomolgus monkeys. Four (2 male and 2 female) captive-bred cynomolgus monkeys (Macaca fascicularis) were used for these studies. For oral and intravenous administration studies, ^{14}C-pantoprazole was administered at dose of 5 mg/kg. For oral administration studies, 5 mL of a 0.1 M sodium bicarbonate was administered by oral gavage at 2 min prior to dosing and at 10, 20, and 30 min after dosing in order to maintain a high gastric pH to protect acid labile pantoprazole from premature degradation. The treatment schedule is shown in the table below. A minimum period of 14 days separated each experiment. Blood for determination of total radioactivity, pantoprazole, and metabolite levels was collected at 0, 0.083 (Group D only), 0.25, 0.5, 1, 2, 4, 6, 9, 12, 24, 48, 72, 96, and 144 hr after dosing. Plasma was used for determination of total radioactivity levels and pantoprazole concentrations. Determination of plasma concentrations of pantoprazole and its sulfone metabolite were performed with [redacted] This method also allowed for simultaneous semiquantitative determination of metabolite M2 (sulfate conjugate of pantoprazole demethylated at the 4-position of the pyridine ring) and an unknown metabolite designated MY. Blood cells collected at 0.5, 24, 72, 96, and 144 hr after dosing were extensively washed and radioactivity levels were quantified.

Treatment schedule for pharmacokinetic (PK), excretion, and tissue distribution studies.

Session	Dose Route	Dose, mg/kg	Type of Study
A	Oral	5 mg/kg	PK
B	IV	5 mg/kg	Excretion
C	Oral	5 mg/kg	Excretion
D	IV	5 mg/kg	PK
E	IV	5 mg/kg	Tissue distribution

Results: Following oral administration, the C_{max} of pantoprazole was significantly lower than the C_{max} for total radioactivity. This difference can be attribute to the presence of pantoprazole metabolites (i.e., sulfone metabolite, and metabolites M2 and MY). This data suggested a pronounced first pass effect resulting in extensive metabolism following oral administration. Concentrations of pantoprazole and total radioactivity at 5 min after intravenous dosing were similar suggesting that little metabolic conversion had occurred at that time. However, pantoprazole levels declined more rapidly than total radioactivity levels. Comparison of AUC values for total radioactivity following oral and intravenous administration revealed that systemic bioavailability following oral administration was 51.77%. However, comparison of AUC for unchanged pantoprazole following oral and intravenous administration suggested that bioavailability following oral administration was only 3.5%. This result further indicated that a pronounced first-pass elimination of pantoprazole occurred following oral administration. Low levels of radioactivity were found to be associated with erythrocytes and these levels were much lower than corresponding plasma concentrations.

Pharmacokinetic parameters of total radioactivity in cynomolgus monkeys following oral administration of ^{14}C-pantoprazole at a dose of 5 mg/kg.

Sex	C_{max}, µg equiv/g	T_{max}, hr	T½, hr	AUC_{0-t} µg equiv*hr/g	AUC µg equiv*hr/g
Male	2.640	1.0	11.9	16.074	16.933
Female	4.524	———	17.5	17.521	18.942

Pharmacokinetic parameters of total radioactivity in cynomolgus monkeys following intravenous administration of ^{14}C-pantoprazole at a dose of 5 mg/kg.

Sex	$C_{5\ min}$, µg equiv/g	T½, hr	AUC_{0-t} µg equiv*hr/g	AUC µg equiv*hr/g
Male	27.637	16.7	31.509	32.222
Female	35.875	16.5	37.157	37.775

Pharmacokinetic characteristics of total radioactivity, pantoprazole, and its metabolites in the cynomolgus monkey following a single intravenous or oral dose of pantoprazole at 5 mg/kg.

Route	Compound	$AUC_{0-\infty}$ mg*hr/L	C_{max}, mg/L	T_{max}, hr	T½, hr	CL, L/hr/kg	Vd, L/kg
Oral	Pantoprazole	0.46	0.56	0.63	0.48	-	-
	Sulfone	0.41	0.16	0.75	0.90	-	-
	MY	2.52	0.76	0.75	1.62	-	-
	M2	0.92	0.71	0.63	0.57	-	-
IV	Pantoprazole	6.88			0.30	0.737	0.316
	Sulfone	0.76	0.68	0.12	0.88	-	-
	MY	1.11	1.08	0.25	0.67	-	-
	M2	2.01	3.62	0.12	0.33	-	-

Note: mg*hr/L should be equivalent to μg equiv*hr/g and mg/L should be equivalent to μg equiv/g.

Washed blood cells: Concentrations of radioactivity in cynomolgus monkeys following a single oral or intravenous administration of ^{14}C-pantoprazole at a dose level of 5 mg/kg.

Time, hr	μg equivalents/g of ^{14}C-Pantoprazole			
	Oral Administration		Intravenous Administration	
	Male	Female	Male	Female
0.5	0.032	0.075	0.305	0.324
4	0.031	0.011	0.052	0.046
24	0.011	ND	0.014	0.038
96	ND	ND	ND	ND

N.D. = Not Detected

Distribution:

Protein Binding-In Vitro

^{14}C-Pantoprazole Binding to Rat, Dog, and Human Serum Proteins (GTR-31194).

Methods: Binding of racemic ^{14}C-pantoprazole at concentrations of 0.9 - 230 μmol/L to rat (Wistar, male) and dog (beagle, male and female) serum protein or at concentrations of 0.9-90 μmol/L to human (male) serum proteins was measured by ———————

Results: Pantoprazole binding to rat and human serum protein was high and nearly constant over the concentration ranges examined. Binding ranged from 94.6-96.9% in rat serum and 97.8-98.5% in human serum. Protein binding to male dog serum protein was lower at 78.8-89.2%. Protein binding to female dog serum protein using a drug concentration range of 1-220 μmol/L was similar to male dogs with values of 81.7-90.3%.

In Vitro Binding of Pantoprazole, Omeprazole, and Lansoprazole in Human, Rat, and Mouse Plasma (Report # GTR-27796).

Methods: Comparative in vitro binding of pantoprazole, omeprazole, and lansoprazole with human, rat, and mouse plasma was assessed by ———— Binding was evaluated at concentrations of 0.5 and 3.0 μg/mL with human, rat, and mouse plasma. An additional concentration of 30 μg/mL was used with rat and mouse plasma.

Results: With human, rat, and mouse plasma, binding was concentration independent. In vitro, >90% of pantoprazole, omeprazole, or lansoprazole were bound to human, rat, or mouse plasma.

Plasma protein binding was as follows:

Drug	Percent Bound to Plasma		
	Man	Rat	Mouse
Pantoprazole	98.05	93.99	91.81
Omeprazole	95.76	84.77	90.15
Lansoprazole	97.32	91.71	87.97

Pantoprazole: Estimation of the Plasma: Whole Blood Concentration Ratios In Vitro Using Rat, Dog, and Human Blood (GTR-31199).

Methods: The plasma: whole blood concentration of ^{14}C-pantoprazole was measured using rat (Wistar, male), dog (Beagle, male), and human (male) blood. Radiolabel was located at the C-2 position on the benzimidazole ring. Drug concentration ranges were 0.435-234 μmol/L for rat, 0.885 to 189 μmole/L for dog, and 0.887-99.8 μmole/L for man. Radioactivity in whole blood and plasma were measured ———— If there was no association of pantoprazole with red blood cells from rats, dogs, or humans, the theoretical plasma to whole blood ratios would be 1.85, 1.94, and 1.87, respectively.

Results: The proportion of ^{14}C-pantoprazole associated with red blood cells was 12-26% for rat blood and 19-31% for dog blood over the concentration ranges used. The proportion associated with red blood cells increased with ascending concentration. For rat blood, plasma to whole blood ratios ranged from 1.63 at the lowest drug concentration to 1.36 for the highest drug concentration. Similarly for dog blood, plasma to whole blood ratios ranged from 1.57 to 1.33. The proportion of ^{14}C-pantoprazole associated with red blood cells was 1-5% for human blood. For human blood, plasma to whole blood ratios ranged from 1.83 to 1.78.

Rat

Pharmacokinetic Investigation of ^{14}C-Pantoprazole in Rat: Quantitative Tissue Distribution After a Single Oral Administration (GTR-31197).

Methods: The quantitative distribution of radioactivity was determined in selected organs from male Sprague Dawley following oral administration of ^{14}C-pantoprazole at a dose of 5 mg/kg. The ^{14}C label was located at the C-2 position of the benzimidazole ring. Animals were sacrificed in groups of 5 at 1, 4, 8, 24, 48, and 96 hr after dosing. Blood was collected and separated into plasma and red blood cells. Tissues were collected for measurement of radioactivity content as follows: adrenal glands, brain, fat, gastrointestinal tract (divided into stomach, small intestine, large intestine, and gut contents), heart, kidneys, lacrimal glands, liver, lungs, muscle (thigh), pancreas, salivary glands (i.e., submandibular, sublingual, and parotid), spleen, skin (dorsal surface including fur), testes, thymus, and thyroid gland. Plasma, whole blood, tissues, and organs were processed and radioactivity was determined by [redacted]

Results: Radioactivity was widely distributed following oral administration. Peak concentrations of radioactivity were in all tissue at 1 hr after dosing except in the thyroid gland, large intestine, and red blood cells. High concentrations were found in the stomach, small intestine, liver, kidneys, plasma, adrenals, blood, fat, and lungs. Concentrations generally decreased with time except in the thyroid gland, large intestine, and red blood cells where peak concentrations were not obtained until 8, 4, and 6 hr after dosing, respectively. The skin showed a second peak at 24 hr after dosing. Plasma concentrations declined over time; however, radioactivity levels in whole blood remained relatively constant from 24 to 96 hr after dosing due to association of radiolabeled material with red blood cells. Radioactivity levels continued to decline over the first 24 hr except in the gastrointestinal tract, thyroid gland, red blood cells, and skin. By 96 hr after dosing, tissue radioactivity levels had declined further.

[^{14}C-Pyridyl] Pantoprazole in the Rat: Whole Body [redacted] Study After a Single Oral or Intravenous Administration (GTR-31326).

Methods: Male Sprague Dawley rats were given a single oral (gavage) or I.V. dose of [^{14}C-pyridyl] pantoprazole (5 mg/kg). One rat was killed at 5 min. (I.V. only), 1, 2, 4, 8, 24, 48, 96, 168 (p.o. only) hours after drug administration and then [redacted] Results are of a qualitative nature.

Results: At 5 min after drug administration, radioactivity was widely distributed. The highest concentrations of radioactivity were seen in the lungs, liver, kidneys and stomach. At 96 hr, only small amounts of radioactivity were detected in rat tissues. Similar results were seen when drug was given via oral route.

Addendum: High concentrations of radioactivity were observed in the stomach wall within 5 min after dosing. From 1 to 8 hr, a decline of radioactivity occurred in most organs and tissues; however, the stomach wall maintained a substantially higher content of

radioactivity than most other organs (i.e., a distinct retention of radioactivity was observed in the stomach wall). At later time points, high concentrations of radioactivity were observed in the liver and kidney suggesting these organs functioned as excretory routes.

Investigation of Melanin Binding of Radioactivity in Male Pigmented Rats Following Intravenous and Oral Administration of ^{14}C-Pantoprazole at a Target Dose Level of 5 mg/kg (GTR-31198).

Methods: The extent of radioactivity bound to melanin was examined in male pigmented rats (Lister hooded) following intravenous or oral administration of ^{14}C-pantoprazole (radiolabel at the C-2 position of the benzimidazole ring) at a dose of 5 mg/kg. Animals were sacrificed in groups of 2 at approximately 1, 6, and 24 hr after dosing and in groups of 3 at 72 hr, 7, 14, and 28 days after dosing. The blood, plasma, skin, and eyes were collected, processed, and analyzed for radioactivity content using ———

Results: Following I.V. and oral administration of ^{14}C-pantoprazole, peak levels of radioactivity in the eyes, non-pigmented skin, whole blood, and plasma were observed at 1.1 hr after dosing [in nmole-eqv./g wet tissue: Eyes, ——— Non-pigmented skin, ——— Pigmented skin, ——— Whole blood, ———; and Plasma, ———. For both I.V. and oral administration, radioactivity in the plasma and non-pigmented skin declined rapidly over the first 24 hr, and plasma levels of radioactivity were undetectable by 7 days after dosing. With I.V. administration, levels in non-pigmented skin were negligible by 14 days; however, with oral administration, radioactivity remained at 10% of peak levels from 3-28 days. With I.V. or oral administration, radioactivity in whole blood declined to 5.5-8% of peak levels by 70 hr after dosing; however, levels remained relatively constant through 14 days after dosing. By 28 days, radioactivity was approximately 2-3.7% of peak levels. As noted in earlier studies, there is a strong association between drug-related radioactivity and red blood cells. With I.V. administration, levels in pigmented skin dropped to 23.05% of peak levels at 24 hr after dosing and remained relatively constant through 7 days after dosing. At 14 to 28 days after dosing, levels in pigmented skin were relatively constant at 6 to 10% of peak levels. For oral administration, levels dropped to 17.8% of peak concentrations by 72 hr after dosing and remained relatively constant through 28 days. With I.V. and oral administration, radioactivity in eyes declined to 50% of peak levels between 1 and 3 days and remained at 15-25% of peak levels from 7-28 days. Following I.V. or oral administration of ^{14}C-pantoprazole, there was strong association of compound-related material with melanin.

Pharmacokinetic Investigation of Pantoprazole in the Rat: Whole-Body ——— Study on the Distribution of Radioactivity (GTR-31223).

Methods: Qualitative distribution of radioactivity in male Sprague Dawley rats was determined using ———, following a single intravenous or oral administration of ^{14}C-pantoprazole at 5 mg/kg. Radiolabel was located at the C-2 position of the benzimidazole ring. Animals were sacrificed at 0.083 (I.V. only), 1, 6, 24, 96 and 168 (oral only) hr after dosing. Animals were processed and the distribution of radioactivity was visualized using———

Results: At 5 min after intravenous administration of drug, radioactivity was widely distributed. Highest concentrations were observed in the stomach contents, liver, kidneys, small intestinal contents, blood vessels, and vestibule of the ear. At 1 hr, the highest concentrations of radioactivity were observed in the gastric mucosa, gut contents, liver, and kidneys. At 6 and 24 hr, high concentrations of radioactivity were observed in the gastric mucosa, skin, and gut contents. At 98 hr, high concentrations of radioactivity were observed in the skin and lower concentrations were observed in the liver and kidneys. Distribution of radioactivity showed a similar pattern following oral administration of drug.

Transplacental Transport and Mammoglandular Passage of ^{14}C-Pantoprazole in the Rat (GTR-31203).

Methods: In study 1, radiolabeled pantoprazole (^{14}C at the 2-position of the benzimidazole ring; 5 mg/kg = 4.5 mg free acid/kg) was given orally (gavage) or I.V. to pregnant rats on day 14 or 18 of gestation. Three dams/time point/route of administration were sacrificed at 1, 4, 8, and 24 hr after drug administration. Blood and various tissues (brain, heart, lungs, liver, spleen, kidneys, placenta, fetus and amniotic fluid) were collected. Radioactivity in each sample was measured by [redacted]

Results: After I.V. dose, concentrations of radioactivity in plasma, kidneys and liver were comparable. Significant amount of radioactivity was also seen in fetus, placenta and amniotic fluid. On day 14 of gestation about 0.26% of the administered radioactivity was seen in fetus, while on day 18 of gestation, 0.58% of the administered radioactivity was seen in fetus and the level was slightly higher than that seen in plasma. Data indicate that drug (and its metabolites) crosses placenta and fetal retention of radioactivity increases as the pregnancy progressed. As a function of time, radioactivity declined rapidly from every tissue and reached a negligible amount by the end of 24 hours after drug administration. Similar results were seen after oral administration of the drug.

Percent of Dose/g Tissue at 1 hr. After I.V. Drug Administration		
Tissues	Gestation Day 14	Gestation Day 18
Brain	0.017	0.025
Lungs	0.177	0.221
Heart	0.196	0.200
Liver	0.544	0.641
Spleen	0.095	0.105
Kidneys	0.486	0.543
Fetus	0.236	0.583
Placenta	0.216	0.226
Amniotic fluid	0.131	0.159
Plasma	0.436	0.415
Blood	0.381	0.341

Secretion in the Milk (Study 2):

Methods: Radiolabeled pantoprazole (^{14}C at the 2-position of the benzimidazole ring; 5 mg/kg = 4.5 mg free acid/kg) was given orally (gavage) or I.V. to 5 lactating rats on day 4 of parturition. Milk samples were collected from pups allowed to suckle for 15 min at 1, 4, 8 and 24 hours after drug administration and radioactivity was measured by ——

Results: A milk transfer was observed following oral or intravenous administration of drug. Peak concentrations of radioactivity in the milk occurred at 8 hr (1.832 μg equiv./g) after oral dosing or 24 hr (2.036 μg equiv./g) after intravenous dosing. A maximum of 0.023% of the administered dose was excreted into the milk.

Pharmacokinetic Investigation of ^{14}C-pantoprazole in the Rat: Quantitative Distribution After Single Intravenous Administration (GTR-31224).

Methods: The distribution of radioactivity into selected tissues was measured following a single intravenous administration of ^{14}C-pantoprazole to Sprague Dawley rats at a dose of 5 mg/kg. The radiolabel was at the C-2 position of the benzimidazole ring. Five rats were sacrificed at each time point (0.083, 1, 4, 8, 24, and 96 hr after dosing). Urine and feces were collected up to 96 hr for 2 animals. Two animals were maintained up to a 168 hr after dosing for analysis of radioactive tissue distribution. Tissues were collected for measurement of radioactive content as follows: brain, heart, lungs, liver, kidneys, thymus, testes, adrenal glands, spleen, thyroid gland, pancreas, muscle, fat, gastrointestinal tract (separated into stomach, small and large intestine after removal of contents), skin, red blood cells, parotid gland, mandibular gland, lingual gland, and lacrimal gland. Contents of the gastrointestinal tract, blood, and plasma were retained separated. Samples were processed and radioactivity was determined ——

Results: Peak concentrations of radioactivity were found in all tissues at 5 min after dosing except for the small intestine (see figure below). Highest concentrations of radioactivity were found in the liver, adrenal glands, plasma, and kidneys. At 1 hr after dosing, the highest concentrations were found in the small intestine, liver, kidney, and plasma followed by the adrenal gland, lung, and red blood cells. By 24 hr after dosing, tissue concentrations of radioactivity had declined significantly except for the stomach, intestines, and erythrocytes. By 96 hr after dosing, significant levels of radioactivity were confined to the erythrocytes, lungs, and spleen. Within the first 24 hr, 96% of the radioactivity was excreted in the urine and feces. Radioactivity was retained in red blood cells and skin/hair shafts.

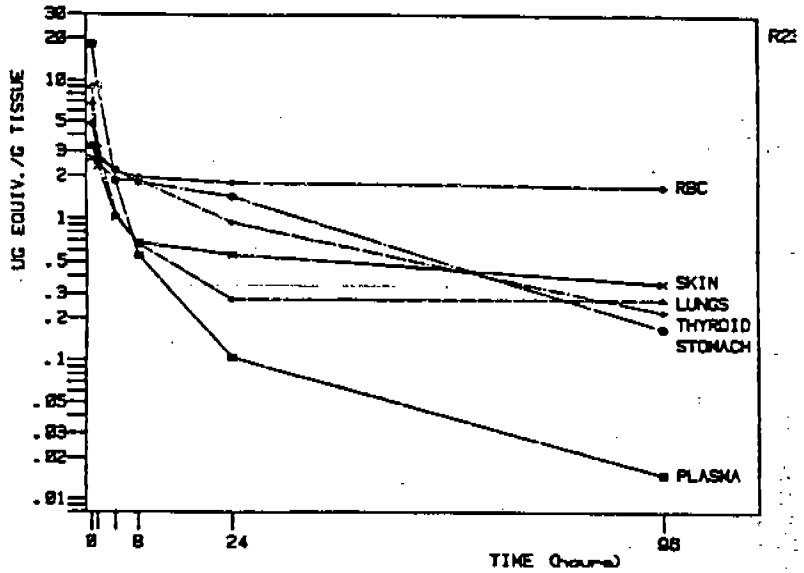

FIG. 1: TOTAL RADIOACTIVITY IN TISSUES I.V. 5 MG/KG (n=5/timepoint)

Distribution of [^{14}C-Pyridyl] Pantoprazole Organs and Tissue in Male Rats After a Single Intravenous Administration (GTR-31314).

Methods: Male rats were given a single I.V. dose of [^{14}C-Pyridyl]-pantoprazole (5 mg/kg). Five rats/time points were sacrificed. The time points were 5, 30 min, 1, 2, 4, 8, 24 and 48 hr after the drug administration. At the end of 4 hr after drug administration, blood samples were collected by cardiac puncture and then various tissues were also collected. In all samples, total radioactivity was determined by ———

Results: The radioactivity was widely distributed in the body. The highest levels of radioactivity were seen in adrenals, plasma, liver and kidneys. As a function of time, radioactivity declined in all tissues except G.I. tract where radioactivity persisted for a longer time. However, by the end of 48 hours most of the organs (including G.I. tract) contained less than 0.03% of administered dose. It should be noted here that plasma and blood levels of radioactivity declined as a function of time. This finding was in contrast to the results of the previous study submitted by the sponsor (GTR-31224) in which ^{14}C-pantoprazole labeled in the benzimidazole moiety was used and the results indicated sequestering of radioactivity in red blood cells. The above experiment clearly indicates that the benzimidazole moiety and not the pyridyl ring of pantoprazole is responsible for binding to cellular component of red blood cells.

Table 2: Tissue distribution of radioactivity following i.v. administration of 5 mg/kg ^{14}C-pantoprazole to male rats.

All the results given are the mean of 5 animals.

Time: 5 min.

Tissue	ug equiv. /g tissue	/tissue	% of dose /g tissue	min.	max.
Brain	1.220	0.255	0.122		
Thymus	3.897	0.171	0.391		
Lungs	8.380	0.886	0.840		
Heart	10.14	0.774	1.016		
Liver	27.59	17.92	2.767		
Pancreas	7.200	0.248	0.722		
Spleen	4.408	0.234	0.442		
Adrenals	22.47	0.081	2.254		
Kidneys	14.04	2.548	1.408		
Testes	1.090	0.234	0.109		
Gl.parotis	7.740	0.168	0.776		
Gl.lingualis	7.285	0.040	0.730		
Gl.submandib.	8.930	0.278	0.895		
Thyroid	8.110	0.009	0.811		
Gl.lacrimalis	7.585	0.159	0.761		
Skin	4.181	11.10	0.419		
Erythrocytes	5.125	1.323	0.514		
Muscle	4.495	-	0.451		
Fat	5.170	-	0.518		
St. contents	n.d.	0.118	n.d.		
SI contents	n.d.	0.654	n.d.		
LI contents	n.d.	0.061	n.d.		
Stomach	4.776	0.772	0.478		
Small intest.	5.630	3.791	0.565		
Large intest.	3.554	0.962	0.356		
Plasma*	16.72	-	1.688		
Blood*	5.260	-	0.531		

* Units used are ug equiv./ml and % of dose/ml
- Total weight of tissue not known
n.d.= not determined, range refer to % of dose
St.= stomach
SI= small intestine
LI= large intestine

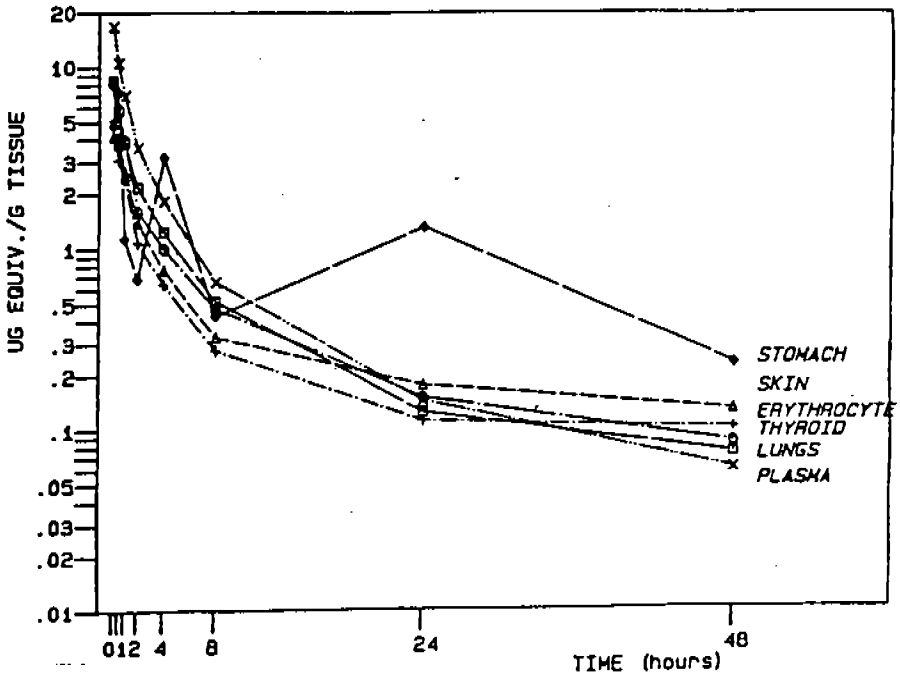

Fig.1: Time course of radioactivity in selected tissues from male rats following a single i.v. administration of 5 mg/kg ^{14}C-pantoprazole (n=5, mean)

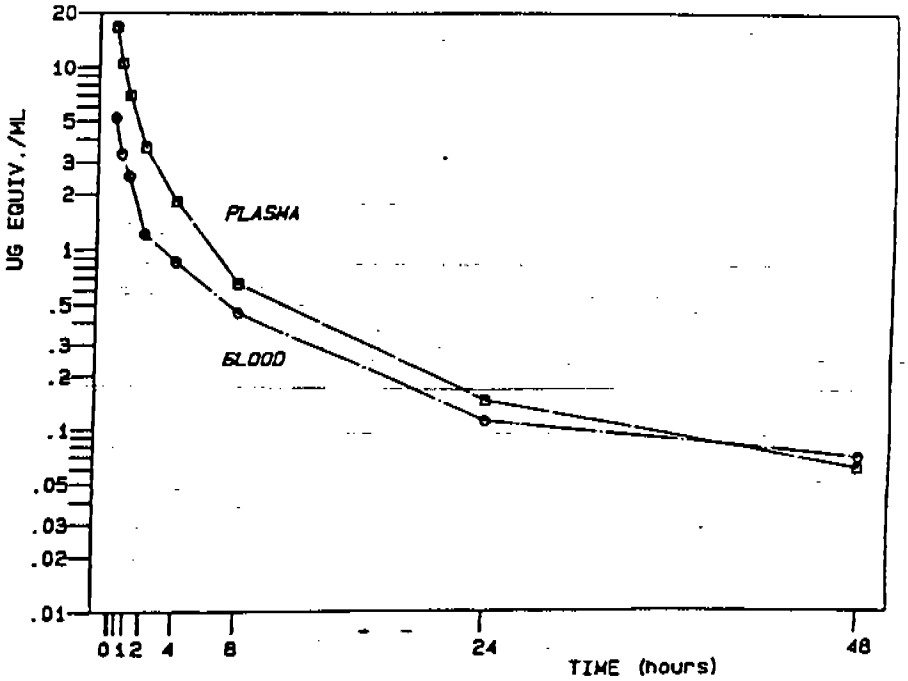

Fig.2: Time course of radioactivity in plasma and blood from male rats following a single i.v. administration of 5 mg/kg ^{14}C-pantoprazole (n=5, mean)

Monkey

Pantoprazole : (^{14}C)-B8610-023: A Study of Absorption, Distribution, and Excretion Following Oral and Intravenous Administration to the Cynomolgus Monkey (GTR-31549).

Methods: The absorption, distribution, and excretion of pantoprazole were examined in cynomolgus monkeys. Four (2 male and 2 female) captive-bred cynomolgus monkeys were used for these studies. For oral and intravenous administration studies, ^{14}C-pantoprazole was administered at dose of 5 mg/kg. For oral administration studies, 5 mL of a 0.1 M sodium bicarbonate was administered by oral gavage at 2 min prior to dosing and at 10, 20, and 30 min after dosing in order to maintain a high gastric pH to protect acid labile pantoprazole from premature degradation. At 1 and 24 hr after intravenous dosing, 1 male and 1 female were sacrificed and the radioactivity levels in the blood, plasma, medulla oblongata, cerebrum, cerebellum, and eye (retina, optic nerve, aqueous humour, and vitreous humour). Radioactivity levels were determined in intact tissues or in tissues following homogenization in deionized water or maceration with scissors. The remaining eyes from sacrificed animals were processed, sectioned, and placed in contact with [redacted] After 10, 35, and 146 days of exposure at –20°C, [redacted] and analyzed.

Results: Following intravenous administration of ^{14}C-pantoprazole, radioactivity distributed to all tissues and fluids with the exception of the aqueous humor. At 1 and 24 hr after dosing, the highest tissue levels of radioactivity were observed in the retina. By 24 hr, radioactivity levels in the retina were significantly higher than levels observed in the blood and plasma. [redacted] studies with the eye found that radioactivity at 1 hr after dosing was primarily associated with the uveal tract (i.e., choroid, ciliary body, and iris), which are melanin rich tissues. Radioactivity was also detected in the sclera; however, none was found in the lens, cornea, and optic nerve. At 24 hr after dosing, radioactivity levels in the uveal tract had declined. No radioactivity was found in the sclera, lens, cornea, and optic nerve.

Tissues : Concentration of radioactivity in cynomolgus monkeys following a single intravenous administration of (^{14}C)-B8610-023 at a nominal dose level of 5 mg free acid/kg body weight - Dose group E

Tissue / Sacrifice time	µg equivalents/g of (^{14}C)-B8610-023			
	1M 1 h	3F 1 h	2M 24 h	4F 24 h
Plasma	4.185	3.450	0.270	0.398
Blood	2.845	2.007	0.178	0.270
Retina	2.226	2.836	1.204	1.984
Optic Nerve	0.696	0.677	ND	0.192
Cerebellum	0.194	0.159	0.097	0.140
Cerebrum	0.166	0.156	0.099	0.142
Medulla Oblongata	0.302	0.175	0.116	0.152
Aqueous Humour	ND	0.386	ND	ND
Vitreous Humour	0.060	0.047	0.023	0.029

ND - Not detected

Tissue concentrations of radioactivity within the eye (determined by _____ following a single intravenous administration of (^{14}C)-B8610-023 to male and female cynomolgus monkeys at a nominal dose level of 5 mg free acid/kg body weight

Animal no and sex / Sacrifice time	µg equivalents of B8610-023 / g of tissue			
	1M 1 h	2M 24 h	3F 1 h	4F 24 h
Choroid	3.585	1.704	4.888	2.298
Ciliary body	2.608	0.777	2.770	1.318
Cornea	NQ	NQ	NQ	NQ
Iris	1.539	0.632	2.188	0.803
Lens	NQ	NQ	NQ	NQ
Optic nerve	NQ	NQ	NQ	NQ
Sclera	3.185	NQ	1.067	NQ

NQ - Tissue concentration not quantifiable

Metabolism:

Enzyme Inhibition and Induction-In Vitro

Inhibition of the Hydroxylation of Lanzolac (Cytochrome P450 Dependent Reaction) With Pantoprazole (GTR-31210).

Methods: In this experiment liver microsomes obtained from phenobarbital pretreated rats were used.

Results: Pantoprazole inhibited in vitro cytochrome-P450 dependent lonazolac hydroxylase activity with a K_i value of 113 μmoles/L. Cytochrome-P450 dependent hydroxylase activity is associated with IIB1 isoform of P450. Hence the drug interacts with cytochrome P450 IIB1.

Pantoprazole: Interaction of Pantoprazole With Ethylmorphine Demethylase Activity (Cytochrome P450) in Rat Liver Microsomes (GTR-31211).

Methods: The inhibitory effect of pantoprazole on ethyl N-morphine N-demethylase was evaluated using rat liver microsomes in vitro. Female rats were given drinking water containing 0.1% phenobarbital for 7 days in order to induce the activity of the cytochrome P450 2B isozyme. Microsomes were prepared from rat livers after the 7-day treatment period. Microsomes were incubated with 300 and 1000 μM ethylmorphine in the presence of pantoprazole at concentrations ranging from 0 to 100 μM. Formaldehyde released from the reaction was reacted with acetylacetone (4%) in 4 M ammonium acetate to form a complex, which was measured spectrophotometrically

Results: The K_i value of pantoprazole for inhibition of ethylmorphine demethylation in rat liver microsomes was 104 μM. Published K_i values for omeprazole and lansoprazole were 68 and 34 μM, respectively (Biochemical Pharmacology 42: 347-355, 1991). The sponsor claimed that pantoprazole had the lowest inhibitory potential toward this cytochrome P450-dependent reaction; however, they did not directly assess the inhibitory effects of omeprazole and lansoprazole in their system.

The Influence of Free Pantoprazole (B8510-29) on 7-Ethoxycoumarin Dealkylase Activity (Cytochrome P450) in Rat Liver Microsomes (GTR-33324).

Methods: The effect of B8510-29 on microsomal conversion of 7-ethoxycoumarin to 7-hydroxy coumarin in an O-dealkylation reaction was examined. The K_m value for 7-ethoxycoumarin is 33 μmol/L. Microsomes were prepared from pentobarbital-treated rats.

Results: B8510-29 possesses a low affinity for cytochrome P450. Interaction with 7-ethoxycoumarin dealkylase was minimal and the K_i was estimated to be >130 μmol/L.

Effect of B8610-23 on 7-Ethoxycoumarin Dealkylase Activity-Cytochrome P450) in Rat Liver Microsomes (GTR-31209).

Methods: In this experiment liver microsomes obtained from phenobarbital pretreated rats were used.

Results: B8610-23 (Pantoprazole) inhibited in vitro 7-ethoxycoumarin dealkylase activity in rat liver microsomes (K_i= 141 ± 0.3 μmoles/L). Interaction with 7-ethoxycoumarin dealkylase was minimal.

Interaction of Pantoprazole with Lonazolac in Human Microsomes (GTR-31217).

Methods: Lonazolac is a substrate for CYP2C9-10 and is converted to hydroxy lonazolac. Microsomes were prepared from human liver.

Results: The K_m for lonazolac with microsomes was determined to be 11 μmole/L. Pantoprazole interacts with lonazolac with a K_i of 64.5 μmol/L. CYP2C9-10 does not appear to be involved with the biotransformation of pantoprazole or it has low affinity for pantoprazole as a substrate.

The Interaction of — 95111 (B8229-24 = Omeprazole), — 95488 (B831-56) and — 96022 (B8510-29) with Rat Liver Cytochrome P450 In Vitro (GTR-31206).

Methods: The in vitro interactions of omeprazole, free pantoprazole (B8510-29), and — 95488, another substituted benzimidazole, with cytochrome P450-dependent microsomal enzymes in a 9000 x g supernatant from rat liver were examined at concentrations of 25, 100, and 200 μM. Microsomal activities of ethylmorphine N-demethylation (EM) and 7-ethoxycoumarin O-deethylation (ECOD) were measured in the presence of these compounds at concentrations of 25, 100, and 200 μM.

Results: Omeprazole inhibited ECOD activity at all three concentration with approximately 50% inhibition at 200 μM. Omeprazole also inhibited EM activity by 40% at a concentration of 200 μM. — 95488 inhibited both ECOD and EM activities at all three concentrations with maximal inhibition of 55%. Pantoprazole at concentrations ≤ 200 μM had no effect on ECOD activity. Pantoprazole inhibited EM activity at concentrations of 100 and 200 μM with maximal inhibition of 20%. Pantoprazole at concentrations ≤ 200 μM had minimal inhibitory on cytochrome P450 reactions, ethylmorphine N-demethylation and 7-ethoxycoumarin O-deethylation.

Pantoprazole: An Evaluation of the CYP1A Induction Potential of Pantoprazole in Primary Rat Hepatocytes: A Comparison with Other Proton Pump Inhibitors (GTR-31189).

Methods: The ability of pantoprazole to affect the expression of the cytochrome P450 1A (CYP1A) subfamily was evaluated in primary cultures of rat hepatocytes. Rat hepatocytes were isolated from untreated female rats. Hepatocytes were cultured for two days, followed by treatment with pantoprazole, omeprazole, or lansoprazole at concentrations of 0, 2, 5, or 10 μM for two days. These concentrations are similar to plasma concentrations found in rats in vivo. The positive control, 3-methylcholanthrene, an inducer of CYP1A, was tested at a concentration of 1 μM. Induction potential of CYP1A was assessed by measurement of 7-ethoxyresorufin O-deethylase (EROD) activity. Isozyme protein contents for CYP1A1 and CYP1A2 were quantified by ———————

Results: Treatment of rat hepatocytes with pantoprazole at concentrations of 2, 5, and 10 μM had no statistically significant effects on EROD activity or protein levels of CYP1A1 and CYP1A2. In contrast, omprazole at 10 μM and lansoprazole at 5 and 10 μM significantly increased EROD activity and protein levels of CYP1A1 and CYP1A2.

7-ethoxyresorufin O-deethylase (EROD) activities and ——— analysis of CYP1A1 and CYP1A2 proteins in rat hepatocytes after treatment with pantoprazole, omeprazole, or lansoprazole at concentrations of 2, 5, or 10 μM for 48 hr.

Treatment (μM)		EROD activity (pmole/min/mg protein)	CYP1A1 (% of control)	CYP1A2 (% of control)
Control	0	1.28	100	100
Pantoprazole	2	1.35	203	137
	5	1.70	194	130
	10	2.63	188	124
Omeprazole	2	2.61	240	170
	5	4.48	242	145
	10	8.74*	633*	358*
Lansoprazole	2	3.51	211	134
	5	11.5*	788*	438*
	10	14.6*	807*	462*
3-methylcholanthrene	1	47.2*	2713*	1166*

* $p \leq 0.05$

Pantoprazole: An Evaluation of the Cytochrome P450 Induction Potential of Pantoprazole in Primary Human Hepatocytes: A Comparison with Other Proton Pump Inhibitors (GTR-32138).

Methods: The cytochrome P450 induction potential of pantoprazole was evaluated and compared with omeprazole and lansoprazole using primary cultures of human hepatocytes. Hepatocytes were obtained from 1 female and 2 male human donors, designated as donors #1, #3, and #4, respectively. Hepatocytes were cultured for two days, followed by treatment with pantoprazole, omeprazole, or lansoprazole at 0, 2, 5, or 10 μM for two days. Positive controls were 3-methylcholanthrene (an inducer of CYP1A) at 1 μM, phenobarbital (an inducer of CYP2B) at 1 mM, and rifampin (an inducer of CYP3A) at 50 μM. Activities of 7-Ethoxyresorufin O-dethylase (EROD), testosterone 16ß-hydroxylase (T16ßH), and testosterone 6ß-hyroxylase (T6ßH) were used to assess the activities of CYP1A, CYP2B, and CYP3A, respectively. Isozyme protein contents of CYP1A and CYP3A were evaluated by ———

Results: Activities and isozyme contents of CYP1A, CYP2B, and CYP 3A following treatment with pantoprazole, omeprazole, or lansoprazole were as follows:

CYP1A: EROD activities for donor #1 were increased by pantoprazole (141-315%), omeprazole (426-994%), and lansoprazole (351-650%) at concentrations of 2, 5, and 10 μM. EROD activities for donors #3 and #4 were increased by omeprazole (181-483% and 906-3565%) and lansoprazole (252-498% and 1135-1929%) at concentrations of 2, 5, and 10 μM. ——— analysis revealed that levels of CYP1A for donors #1, #3, and #4 were increased by omeprazole (608-570%, 225-422%, and 1340-2637%) and lansoprazole (372-314%, 337-453%, and 2011-2175%) at concentrations of 2, 5, and 10 μM. Induction potential for CYP1A was as follows: lansoprazole > omeprazole > pantoprazole.

CYP2B: T16ßH activities for donor #1 were increased by pantoprazole (149-127%) and lansoprazole (138-158%) at concentrations of 2, 5, and 10 μM and omeprazole (146-156%) at 5 and 10 μM. T16ßH activities for donors #3 and #4 were unaffected by treatment. ——— analysis of CYP2B was not performed. Induction potential for CYP2B was as follows: lansoprazole > pantoprazole > omeprazole.

CYP3A: T6ßH activities for donor #1 were increased by pantoprazole (176-150%) and lansoprazole (139-178%) at concentrations of 2, 5, and 10 μM and omeprazole at (177-184%) concentrations of 5 and 10 μM. T6ßH activities for donor #3 were increased by pantoprazole (225-275%), omeprazole (170-184%), and lansoprazole (166-266%) at concentrations of 5 and 10 μM. T6ßH activities for donor #4 were increased by omeprazole (127%) at a concentration of 10 μM. ——— analysis revealed that levels of CYP3A4 for donor #1 were increased by pantoprazole (172%) at 10 μM, lansoprazole (189-241%) at 5 and 10 μM, and omeprazole (255-229%) at 2, 5, and 10 μM. Levels of CYP3A4 for donor #3 were increased by lansoprazole (269-358%) at 5 and 10 μM. Levels of CYP3A4 for donor #4 were unaffected by treatment. Induction potential for CYP3A was as follows: omeprazole > lansoprazole > pantoprazole.

Identification of P450 Isozymes Involved in Metabolism of Pantoprazole on Human Liver Microsomes **(Report # Byk 23/96 K1).**

Methods: The object of this study was to identify P450 isozymes in human microsomes which are involved in phase I biotransformation of pantoprazole. Human liver microsomes fractions containing different specific activities of CYP2C19, CYP2D6 and CYP3A4 were incubated with pantoprazole. Decrease in pantoprazole level was measured over 60 min. time period.

Results: Pantoprazole is mainly metabolized by CYP2C19 and CYP3A4 isozymes of P450 but very little metabolism was carried out by CYP2D6 isozyme.

P450 Isozymes	Percent Decrease of Pantoprazole in 60 min.
CYP2C19	59 ± 7.8
CYP3A4	40 ± 3.8
CYP2D6	29 ± 3.1

Pantoprazole: Biotransformation of Pantoprazole in Human Microsomes. Identification of P450 Isozymes by Selective Inhibitors **(GTR-31216).**

Methods: Cytochrome P450 isozymes involved in the biotransformation of pantoprazole were identified with the use of specific inhibitors as follows: quinidine for CYP2D6, sulphenazole for CYP2C9-10, and ketoconazole for CYP3A4. Liver microsomes were prepared from pieces of human livers. Microsomal protein was incubated with 10 μM pantoprazole in the presence and absence of inhibitors for periods up to 120 min at 37°C.

Results: Biotransformation of pantoprazole was inhibited as follows: 30-40% by ketoconazole (CYP3A4), 14-21% by quinidine (CYP2D6), and 11-14% by sulphaphenazole (CYP2C9-10). Cytochrome P450 isozymes 3A4, 2D6, and 2C9-10 appear to play roles in the biotransformation of pantoprazole. Others isozymes were not examined. Due to the limited scope of this study with only three inhibitors as well as the use of an in vitro system, it is difficult to assess the biological significance of these results.

Enzyme Inhibition and Induction-In Vivo

Interactions of Cimetidine, Omeprazole, Lansoprazole or Pantoprazole With Diazepam in Rats (GTR-31212).

Methods: In this study duration of the effects of diazepam (4 mg/kg, s.c.) on muscle coordination of rats were assessed in the presence of various other drugs cimetidine (99-1189 μmole/kg), omeprazole (87-1013 μmole/kg), lansoprazole (108-217 μmole/kg) and pantoprazole (240-960 μmole/kg). Above drugs were given 60 min prior to diazepam administration.

Results: About 288, 170 and 281 μmole/kg of omeprazole, lansoprazole and cimetidine, respectively, were needed to produce 50% prolongation of diazepam effect in rats. The highest tested dose of pantoprazole (960 μmole/kg) only produced 36% prolongation of diazepam effect in rats. Data indicated that pantoprazole interacts significantly less with the diazepam metabolism than the other tested drugs.

Pantoprazole: Induction of Ethylmorphine Demethylase Activity After Oral Administration of Pantoprazole for Three Days (GTR-31215).

Methods: Induction of cytochrome P450 activity (i.e., ethylmorphine demethylase activity) was examined in female rats that received pantoprazole by oral gavage at doses of 200 and 300 mg/kg/day for 3 days. Rats in positive control groups received either 3-methylcholanthrene at 25 mg/kg/day by the intraperitoneal route for 3 days or phenobarbital at a concentration of 0.1% in drinking water for 7 days. Following treatment, animals were sacrificed and liver microsomes were prepared. Microsomal ethylmorphine demethylase activity was measured.

Results: Pantoprazole at 200 and 300 mg/kg/day increased ethylmorphine demethylase activity by 171.9 and 177.8% of the control (22.1 nmole formaldehyde/mg protein/hr), respectively. Phenobarbital increased activity to 515.8% of the control, while 3-methylcholanthrene had no affect. The induction by pantoprazole appeared to be of the phenobarbital type (CYP2B).

Effects of Pantoprazole on the Drug-Metabolizing Enzyme System in Rat Liver (GTR-31544).

Methods: Groups (n=6/group) of SD female rats (7 weeks old, 150-180 g) were given daily oral doses of vehicle (0.5% CMC), pantoprazole (5, 50 or 300 mg/kg/day), omeprazole (5, 50 or 300 mg/kg/day) or lansoprazole (5, 50 or 300 mg/kg/day) for 7 days There were 2 positive control groups, one received 80 mg/kg/day (i.p.) phenobarbital for 7 days and the other group received 3-MC (3-methylcholanthrene in corn oil: 30 mg/kg/day) for 3 days. Three rats/group were killed at 24 hr after the last dose and the remaining rats were sacrificed at 1 week after the last dose. Liver samples were collected, and microsomes were isolated. Cytochrome P450 and cytochrome b5 contents and NADPH cytochrome C reductase activity, drug metabolizing enzyme activities (aminopyrine N-demethylase, 7-ethoxycoumarin O-deethylase, phenacetin deethylase, 7-ethoxyresorufin O-deethylase, 7-pentoxyresorufin O-dependentylase and testosterone 6ß-hydroxylase) and P450 isozymes were measured.

Results: Hepatic microsomal protein content was not affected by treatment. At 300 mg/kg/day, microsomal cytochrome P450 content were increased by 28%, 50% and 60% in pantoprazole, omeprazole and lansoprazole treated rats respectively, when compared to control values (increases in positive controls: 73% and 61% in 3-MC and phenobarbital treated rats, respectively). Dose-related increases in cytochrome b5 and NADPH-cytochrome C reductase were also seen (MC did not increase NADPH-cytochrome C reductase activity). The results of drug-metabolizing enzyme activity and cytochrome P450 isozyme quantification indicates quantitative as well as qualitative differences among pantoprazole, omeprazole and lansoprazole. For example, CYP1A content in pantoprazole treated rats were less than that observed in omeprazole and lansoprazole treated rats, while CYP2B content in pantoprazole treated rats were larger than that seen in omeprazole and lansoprazole rats. There was good correlation between increases in metabolizing enzyme activities with corresponding increases in P450 isozymes. According to sponsor, "pantoprazole exhibited phenobarbital type induction, whereas omeprazole and lansoprazole exhibited both phenobarbital and MC mixed type induction, although lansoprazole was predominately MC in type".