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ORIGINAL ARTICLES

Propranolol: Pooled Michaelis-Menten parameters and the effect of input rate on bioavailability

Average steady-state propranolol plasma concentration ($\bar{C}_{ss}$) were calculated from published steady-state propranolol clearance data for dose rates (R_0) of 40, 80, 160, 240, and 320 mg/day in divided doses every 6 hours. The $\bar{C}_{ss}$-R_0 data for each of four subjects were fit essentially perfectly by the equation: $\bar{C}_{ss} = K_m R_0/(V_m - R_0)$. Very similar V_m and K_m values were obtained with the V_{mi} and K_{mi} values for four parallel Michaelis-Menten pathways of propranolol metabolism. It is shown by use of the mean V_m and K_m values that the propranolol input rate profoundly affects its bioavailability, which is expected for a first-pass drug that follows Michaelis-Menten elimination kinetics after oral dosing. This most likely explains the poor bioavailability of propranolol after a sustained-release formulation. The decreased bioavailability of propranolol when the number of subdivisions of the daily dose is increased is also explained. (CLIN PHARMACOL THER 37:481-487, 1985.)

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Garg et al.[3] administered 40 mg propranolol as regular tablets four times a day and 160 mg propranolol as a long-acting capsule once a day to 24 healthy men. The plasma $AUC_{0-\infty}$ averaged 584 ng/ml · hr for the long-acting preparation and 1071 ng/ml · hr for the regular tablets, yielding a relative bioavailability of only 54.5% for the long-acting formulation.

Ohashi et al.[4] described another study in which 60 mg propranolol once a day as a long-acting formulation was compared with 20 mg in conventional form given three times a day. Each dose was taken for 8 consecutive days by six subjects. At steady state on day 8, the average AUC for the long-acting formulation was 219 ng/ml · hr, compared with the value of 428 ng/ml · hr for the conventional form. The relative bioavailability of the long-acting product in that report was thus 51.2%.

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Abbreviations

$AUC_{0-\infty}$	AUC extrapolated to infinity
$AUC_{0-\tau}$ bolus	AUC during a dosing interval at steady state when repetitive bolus doses are given every τ hours by mouth
$AUC_{0-\tau}$ zero	AUC during a dosage interval at steady state when there is a constant rate (zero order) of input by mouth; equivalent to $\tau\bar{C}_{ss}$
β	Limiting apparent elimination rate constant; $\beta = V_m/VK_m$
C_0	Initial plasma concentration
$\bar{C}_{ss}$	Average steady-state concentration when R_0 is continued to steady state
C_{ss}^{min}	Minimum steady-state concentration when multiple bolus oral doses are continued to steady state at intervals of τ
Cl_i	Intrinsic clearance; $Cl_i = V_m/K_m$
Cl_{ss}	Steady-state clearance; $Cl_{ss} = R_0/\bar{C}_{ss}$
D_m	Maintenance dose at steady state; $D_m = R_0\tau$
F_{ss}	Steady-state bioavailability (Eq. 6)
K_m	Pooled Michaelis constant, equal to the concentration when the velocity of metabolism is one-half maximal (in concentration units)
Q	A dimensionless parameter that replaces $\beta\tau$ as an exponential exponent when Michaelis-Menten kinetics are operative; $Q = (1 - r)\beta\tau = (V_m\tau - D_m)/VK_m = (V_m - R_0)\tau/VK_m$
Q_L	Liver blood flow
R_0	Zero-order input rate
r	Zero-order input rate as a fraction of V_m; $r = R_0/V_m$
τ	Uniform dosage interval for the bolus doses
V	Volume of distribution
V_m	Pooled maximal velocity of metabolism (mass/time); also, $V'_m = V_m/V$
VK_m	Pooled Michaelis constant in terms of mass

Dvornik et al.[2] reported that, when measured at steady state on the seventh day, 80 mg propranolol as conventional tablets was about 30% more bioavailable when given as 40 mg twice a day than when given as 20 mg four times a day. The 24-hour steady-state AUCs averaged 340.1 ng/ml · hr after dosing four times a day and 446.0 ng/ml · hr after dosing twice a day. The relative bioavailability of the twice-daily regimen was thus 31.4% greater than that after dosing four times a day.

Wood et al.[13] reported that both oral and systemic clearances of propranolol were considerably lower after the seventh dose than after a single dose when 80 mg propranolol in conventional form was taken three times a day. Similar data were reported by Silber et al.[8] at steady state after oral doses of 40, 80, 160, 240, and 320 mg/day. They also measured metabolites of propranolol and reported values for V_m and K_m for propranolol glucuronide, 4-hydroxypropranolol glucuronide, α-naphthoxylactic acid, and unidentified metabolic pathways.

The objective of this article is to show how to estimate pooled V_m and K_m values from the data of Silber et al.[8] These average values are then used to explain the observations of Garg et al.,[3] Ohashi et al.,[4] and Dvornik et al.[2]

METHODS

The propranolol $\bar{C}_{ss}$ was calculated for each of four subjects at each of five R_0 values by dividing the Cl_{ss} value listed in Table II of the article of Silber et al.[8] into the R_0 while correcting for units. These five $\bar{C}_{ss}$ values and their corresponding R_0 values were fit to Eq. 1 by the method of Wilkinson,[12] which has been shown to give essentially the same parameter values as nonlinear least-squares regression:

$$\bar{C}_{ss} = \frac{K_m R_0}{V_m - R_0} \quad (1)$$

Eq. 1 is applicable to both one-[11] and two-compartment[9] models when there is zero-order input by mouth. The equation can also be applied when input is irregular and not zero order, as is the case with these propranolol data.

Silber et al.[8] showed that the scheme represented in Fig. 1 applied to propranolol. The fitting of $\bar{C}_{ss}$-R_0 data to Eq. 1 indicates that the four parallel Michaelis-Menten pathways pool, such that the individual V_{mi} and K_{mi} (i = 1 to 4) values are not observed; only a pooled V_m and K_m value may be obtained from plasma or whole blood data. Sedman and Wagner[7] published both dose-independent and -dependent equations to estimate the pooled V_m and K_m values from individual V_{mi} and K_{mi} values. The dose-dependent equations could not be applied to propranolol, because steady-state plasma or whole blood concentrations of individual propranolol

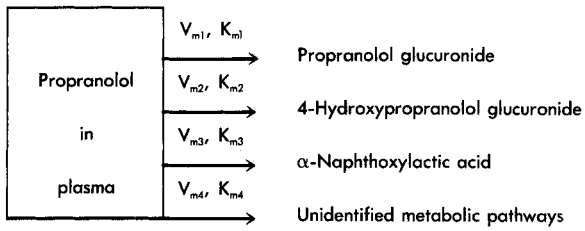

Fig. 1. Schematic representation of propranolol metabolic pathways.

metabolites have not been reported. However, the dose-independent equations[7] were applied to the data of Silber et al.[8] and the equations are as follows:

$$V_m = \sum_{i=1}^{n} V_{mi} \tag{2}$$

$$K_m = \frac{\sum_{i=1}^{n} V_{mi}}{\sum_{i=1}^{n} \frac{V_{mi}}{K_{mi}}} \tag{3}$$

Pooled V_m and K_m values for propranolol were also estimated from the whole-blood propranolol single-dose clearance and Cl_{ss} values reported by Wood et al.[13] by the use of Eqs. 4 and 5, which were derived by Wagner[10] and also reported by Wagner et al.[9] Clearance estimated from first-dose data was used as the estimate of Cl_i.

$$K_m = \frac{R_0}{Cl_i - Cl_{ss}} \tag{4}$$

$$V_m = \frac{R_0}{1 - (Cl_{ss}/Cl_i)} \tag{5}$$

The propranolol F_{ss} varies with the R_0 and is calculated by Eq. 6[9]:

$$F_{ss} = \frac{1}{1 + \frac{V_m - R_0}{Q_L K_m}} \tag{6}$$

Additional equations used in the construction of Fig. 4 are included in the Appendix.

RESULTS

Results of fitting the $\bar{C}_{ss}$-R_0 data of the four subjects to Eq. 1 are shown in Fig. 2, and the estimated values of V_m and K_m are listed in Table I, along with the Cl_i and coefficients of determination for the fittings. The V_{mi} and K_{mi} values shown in Fig. 1[8] were substituted into Eqs. 2 and 3 to provide the estimates of the pooled V_m and K_m values shown in Table II. An item-by-item

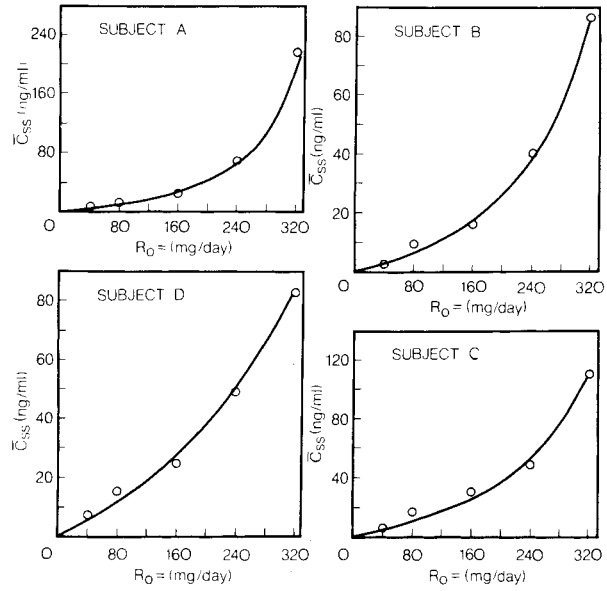

Fig. 2. Results of fitting $\bar{C}_{ss}$-R_0 data of four subjects to Eq. 1. The *circles* are the observed propranolol $\bar{C}_{ss}$ values at various R_0 levels, and the *solid lines* are values based on the estimates of V_m and K_m listed in Table I.

matching of Tables I and II indicates reasonable similarity of data for subjects A, B, and C, while those for subject D should not be compared.

Substitution of the Cl_i and Cl_{ss} values of Wood et al.[13] into Eqs. 4 and 5 gave the pooled V_m and K_m values listed in Table III. It should be noted that the V_m and K_m values listed in Tables I and II are based on propranolol levels in plasma, while those in Table III are based on propranolol levels in whole blood. Substitution of the mean V_m and K_m values in Table I and a Q_L of 1.5 L/min into Eq. 6 gave the *circles* in Fig. 3, while substitution of the mean V_m and K_m values in Table III and the same Q_L value into Eq. 6 gave the *triangles* in Fig. 3. The mean bioavailability as shown in Fig. 3 was 33.8% and ranged from 20.0% to 45.6% in six subjects (as reported by Wood et al.[13]).

Fig. 4 is a plot of the ratio $AUC_{0-\tau}$zero/$AUC_{0-\tau}$bolus as given by Eq. 12 in the Appendix for propranolol when the mean V_m and K_m values in Table I and a V of 300 L with a τ of 6 hours are substituted into the equation. Propranolol V is linearly related to the free fraction of propranolol in blood,[5] and the use of a value for V of 300 L is representative of a centrally located volume. On the same plot, the AUC ratio for verapamil is shown as a fraction of R_0; the mean V_m (469 mg/day) and K_m (48.8 ng/ml) as reported by

Table I. Pooled parameter estimates for propranolol based on fitting average $\bar{C}_{ss}$-R_0 data to Eq. 1 by Wilkinson's method[12]

Subject	V_m *(mg/day)*	K_m *(ng/ml)*	Cl_i *(L/min)*	r^2
A	376	37.8	6.91	0.9992
B	438	32.2	9.45	0.9973
C	457	48.4	6.56	0.9913
D	605	76.7	5.48	0.9894
$\bar{X}$	469	48.8	7.10	
Coefficient of variation (%)	20.7	40.6	23.7	

Table II. Pooled parameter estimates for propranolol based on the dose-independent equation of Sedman and Wagner[7] (namely, Eqs. 2 and 3 with the V_{mi} and K_{mi} values for propranolol glucuronide, 4-hydroxyglucuronide, α-naphthoxylactic acid, and unidentified metabolic pathways of Silber et al.[8])

Subject	V_m *(mg/day)*	K_m *(ng/ml)*	Cl_i *(L/min)*
A	382	34.0	7.80
B	496	33.7	10.2
C	513	50.2	7.10
D	490*	59.7*	5.70
$\bar{X}$	470	44.4	7.70
Coefficient of variation (%)	12.7	28.8	24.4

*Omitting the V_m of 4577 mg/day and K_m of 6297 ng/ml for propranolol glucuronide.

Table III. Pooled parameter estimates for propranolol obtained with Eqs. 4 and 5 from data of Wood et al.[13] based on whole blood assays

Subject	V_m *(mg/day)*	K_m *(ng/ml)*	Cl_i *(L/min)*
1	378	26.8	9.79
2	853	161.8	3.66
3	342	41.8	5.68
4	878	161.8	3.77
5	414	83.3	3.45
6	567	62.9	6.26
$\bar{X}$	572	89.7	5.44
Coefficient of variation (%)	42.0	65.8	44.7

Wagner[10] with V = 300 L and τ = 6 hours were used in construction of the curve.

To approximately simulate the data of Dvornik et al.,[2] numerical integrations were performed on a microcomputer with use of the one-compartment open model with Michaelis-Menten elimination kinetics, the mean V_m and K_m values in Table I, and a V value of 300 L. To simulate 20 mg every 6 hours, the initial C_0 value was $(20 \times 10^3)/300 = 66.67$ ng/ml, while that for the 40 mg every 12 hours regimen was $(40 \times 10^3)/300 = 133.3333$ ng/ml. The C_0 values for subsequent doses were obtained by adding the minimum value from the previous dose to this C_0 value. The $AUC_{0-\infty}$ values were obtained at steady state by application of the logarithmic trapezoidal rule. For the 20 mg four times a day regimen, the simulated data gave an $AUC_{0-\tau}$ of 330.4 ng/hr · ml, compared with the 340.1 ng/ml · hr reported by Dvornik et al.[2] The 40 mg twice a day regimen gave an $AUC_{0-\tau}$ of 466.4 ng/ml · hr, compared with the 446.0 ng/ml · hr reported by Dvornik et al.[2] Thus the simulated data indicated an increased bioavailability of $(466.4 - 330.4)/330.4 \times 100 = 41.4\%$ for the twice-daily dosing regimen, while the observed value[2] was 31.4%.

DISCUSSION

Results in Fig. 2 and Tables I and II indicate that the pooled Michaelis-Menten parameters V_m and K_m adequately describe the propranolol $\bar{C}_{ss}$-R_0 data and that the much more complicated models used by Silber et al.[8] are unnecessary. Parameter values for subjects A, B, and C (Tables I and II) are of the same order, item by item, but those for subject D are not, because the V_m and K_m values for propranolol glucuronide were so large that they had to be omitted in the calculation of the pooled V_m and K_m values from Eqs. 2 and 3.

Although the data are not strictly comparable because values in Table I are based on plasma levels and those in Table III are based on whole blood levels, t tests were applied. The mean V_m of 469 mg/day in Table I does not differ from the mean of 572 mg/day in Table III ($t = 0.8$; $df = 8$). The mean K_m of 48.8 ng/ml in Table I likewise does not differ from the mean of 89.7 ng/ml in Table III ($t = 0.95$; $df = 8$).

The mean V_m and K_m values in Table I substituted

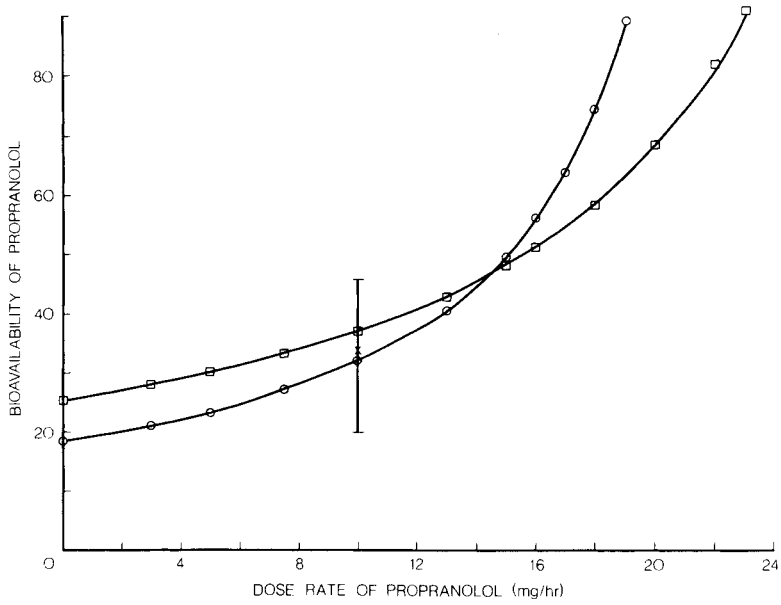

Fig. 3. Plots of propranolol F_{ss} against R_0 from Eq. 6. The symbol *X* is the mean bioavailability and the *bars* represent the range of individual subject bioavailabilities reported by Wood et al.[13] The intercepts of the curves on the y-axis are the intrinsic bioavailabilities (see text). ○ = Calculated with the use of V_m and K_m data from Table I; △ = calculated with the use of V_m and K_m data from Table III.

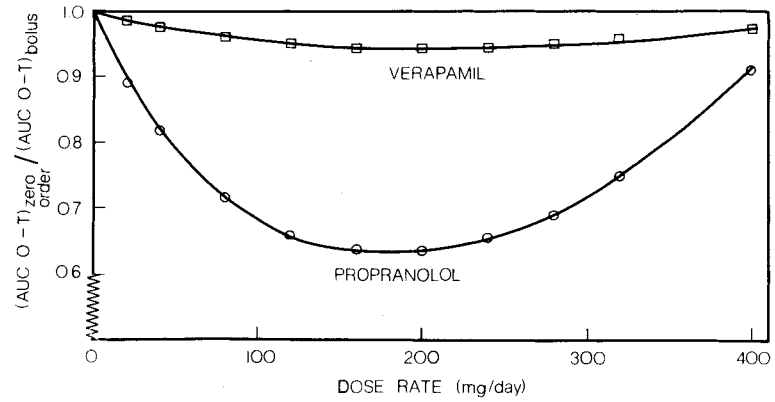

Fig. 4. The ratio $AUC_{0-\tau}$ zero/$AUC_{0-\tau}$ bolus plotted against the R_0 based on the equations in the Appendix, the mean V_m and K_m values listed for propranolol in Table II, and the mean V_m and K_m values for verapamil reported by Wagner.[10]

into appropriate equations offer adequate explanations of reported observations and also lead to other interesting predictions. For example, application of Eq. 6 led to Fig. 3. This indicates that the intrinsic bioavailability (i.e., the bioavailability extrapolated to an R_0 of zero) is predicted to be 18.4% from use of the mean V_m and K_m values in Table I and 25.3% from those values in Table III. Bioavailability increases as R_0 increases, until bioavailability is 100%, when the R_0 is equal to the V_m, namely, 19.5 mg/hr (469 mg/day) from data in Table I and 23.8 mg/hr (572 mg/day) for data in Table III. These predictions exactly bracket the observed bioavailabilities reported by Wood et al.[13] for an R_0 of 10 mg/hr (Fig. 3).

Bolus dosing and constant rate (zero order) are the extremes in input rates. Hence a plot of the ratio $AUC_{0-\tau}$ zero/$AUC_{0-\tau}$bolus as a function of R_0 indicates the sensitivity of a drug to changes in bioavailability in

relation to both rate of absorption and drug intake rate. Fig. 4 shows that, with respect to bioavailability changes, propranolol is very much more sensitive to input rate than is verapamil. Fig. 4 data for propranolol explain the observations of Garg et al.[3] and Ohashi et al.[4] of markedly reduced bioavailabilities of long-acting propranolol formulations relative to conventional ones. The minimum value in the AUC ratio/dose plot occurs approximately at an R_0 of 200 mg/day, when the area ratio is 63.6%. The data of Bottini et al.[1] could be construed as an exception to results in Fig. 4. The latter compared a so-called once-a-day propranolol formulation and conventional tablets in six subjects and reported mean $AUC_{0-\infty}$ values of 838.6 and 860.4 ng/ml · hr after the same total dose of 160 mg propranolol. However, careful inspection of the mean propranolol plasma concentration profiles strongly suggest that the once-a-day regimen provided only delayed absorption, with a time shift in the concentration-time curve rather than a formulation with truly slow absorption.

Simulation of the results of Dvornik et al.[2] was also quite successful and indicated that the slower input of the same dose (20 mg four times a day compared with 40 mg twice a day) resulted in appreciable loss of bioavailability, namely, 76.1% observed[2] compared with 70.8% simulated here.

It does not appear to be recognized generally that the bioavailability of first-pass drugs that obey Michaelis-Menten kinetics will be sensitive to rate of drug input. Before the formulation of long-acting or sustained-release formulations of such drugs is begun, it would be wise to conduct a kinetic study at steady state in which different absorption and input rates are simulated by oral aliquots of an aqueous solution of the drug at different rates. Thus the magnitude of the decrease in bioavailability with change in input rate could be assessed. If Eq. 1 herein fits the data from such a study well, then pooled V_m and K_m values could be estimated and projections such as those reported herein could be made for a different drug.

APPENDIX
Equations used in construction of Fig. 4.

For intermittent bolus dosing to steady state, Wagner et al.[11] derived Eq. 7 for the one-compartment open model with Michaelis-Menten elimination:

$$AUC_{0-\tau}\text{bolus} = \frac{D_m}{V_m}\left(\frac{D_m}{2V} + C_{ss}^{min} + K_m\right) \quad (7)$$

For continuous zero-order input to steady state, both the one-[11] and two-compartment[9] open models with Michaelis-Menten elimination give Eq. 8:

$$AUC_{0-\tau}\text{zero} = \frac{K_m D_m}{V_m - R_0} \quad (8)$$

A ratio of Eq. 8 to Eq. 7, with some simplification, gives Eq. 9:

$$\frac{AUC_{0-\tau}\text{zero}}{AUC_{0-\tau}\text{bolus}} = \left(\frac{V_m}{V_m - R_0}\right)\left(\frac{K_m}{\frac{D_m}{2V} + C_{ss}^{min} + K_m}\right) \quad (9)$$

Sawchuk and Rector[6] showed that C_{ss}^{min} for the one-compartment open model with Michaelis-Menten elimination is given by Eq. 10:

$$C_{ss}^{min} = \frac{D_m}{V}\left(\frac{1}{e^Q - 1}\right) \quad (10)$$

in which Q is given by the first term on the right side of Eq. 11 (I have shown that Q is also given by the second term on the right side of Eq. 11):

$$Q = \frac{V_m\tau - D_m}{VK_m} = (1 - r)\beta\tau \quad (11)$$

Substituting from Eqs. 10 and 11 into Eq. 9, followed by division of the numerator and denominator by K_m, yields Eq. 12:

$$\frac{AUC_{0-\tau}\text{zero}}{AUC_{0-\tau}\text{bolus}} = \left[\frac{1}{1 - r}\right]\left[\frac{1}{1 + \frac{D_m}{VK_m}\left(\frac{1}{2} + \frac{1}{e^Q - 1}\right)}\right] \quad (12)$$

The terms D_m/VK and $1/(e^Q - 1)$ have opposite effects. As VK_m increases, then D_m/VK_m decreases and $1/(e^Q - 1)$ increases. Because $D_m = R_0\tau$, there is a minimum in the area ratio as R_0 increases. The area ratio is equal to unity when $R_0 = 0$, since $r = 0$ and $D_m = 0$; the area ratio again becomes equal to unity when $R_0 = V_m$.

Author's note added in proof

If a sustained-release form of propranolol were to travel through the gastrointestinal tract and reach the colon without releasing all of its drug, then it is feasible that drug released in the colon could escape the first-pass effect and provide a greater AUC than expected from the equations in this article.

References

1. Bottini PB, Caulfield EM, Devane JG, Geoghegan EJ, Panoz DE: Comparative oral bioavailability of conventional propranolol tablets and a new controlled-absorption propranolol capsule. Drug Dev Indust Pharmacy **9:**1475-1493, 1983.
2. Dvornik D, Kraml M, Dubuc J, Fencik M, Weidler D, Mullane JF: Propranolol concentrations in healthy men given 80 mg daily in divided doses: Effect of food and circadian variation. Curr Ther Res **32:**214-224, 1982.

3. Garg DC, Duncan RC, Mishriki A, Jallad MS, Kraml M, Fencik M, Weidler DJ: Comparative pharmacokinetics and pharmacodynamics following single and multiple doses of conventional and long-acting propranolol. CLIN PHARMACOL THER **35:**242, 1984. (Abst. No. A25.)
4. Ohashi K, Ebihara A, Kondo K, Usami M: Clinical pharmacokinetics and pharmacological actions of a long-acting formulation of propranolol. Arzneimittelforsch **34:**507-512, 1984.
5. Branch RA, James J, Read AE: A study of factors influencing drug disposition in chronic liver disease, using the model drug (+)-propranolol. Br J Clin Pharmacol **3:**243-249, 1976.
6. Sawchuk RJ, Rector TS: Steady-state plasma concentrations as a function of the absorption rate and dosing interval for drugs exhibiting concentration-dependent clearance: Consequences for phenytoin therapy. J Pharmacokinet Biopharm **7:**543-555, 1979.
7. Sedman AJ, Wagner JG: Quantitative pooling of Michaelis-Menten equations in models with parallel formation paths. J Pharmacokinet Biopharm **2:**149-160, 1974.
8. Silber BM, Holford NHG, Riegelman S: Dose-dependent elimination of propranolol and its major metabolites in humans. J Pharm Sci **72:**725-732, 1983.
9. Wagner JG, Szpunar GJ, Ferry JJ: Commentary: Exact mathematical equivalence of the venous equilibration ("well-stirred") model, the sinusoidal perfusion ("parallel-tube") model, and a specific two-compartment open model. Drug Metab Dispos **12:**385-388, 1984.
10. Wagner JG: Predictability of verapamil steady-state plasma levels from single-dose data explained. CLIN PHARMACOL THER **36:**1-4, 1984.
11. Wagner JG, Szpunar GJ, Ferry JJ: Michaelis-Menten elimination kinetics: Areas under curves, steady-state concentrations and clearances for compartment models with different types of input. Biopharm Drug Dispos. (In press.)
12. Wilkinson GN: Statistical estimation in enzyme kinetics. Biochem J **80:**324-332, 1961.
13. Wood AJJ, Carr K, Vestal RE, Belcher S, Wilkinson GR, Shand DG: Direct measurement of propranolol bioavailability during accumulation to steady-state. Br J Clin Pharmacol **6:**345-350, 1978.