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Research Paper

Prediction of Human Bioavailability from Human Oral Administration Data and Animal Pharmacokinetic Data Without Data from Intravenous Administration of Drugs in Humans

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Purpose. To predict the absolute oral bioavailabilities (BAs) of drugs in humans without using pharmacokinetic data from intravenous administration in humans.
Methods. The distribution volume of the terminal phase (Vd_β) in humans was predicted by three methods using animal pharmacokinetic data. Then, total body clearance (CL_{tot}) was calculated by multiplying the elimination rate constant and Vd_β , and the BA was calculated as a ratio between CL_{tot} and oral clearance. The predicted and observed values were compared for 67 drugs for which pharmacokinetic data after intravenous administration in humans were available.
Results. For Vd_β , predicted values within twice the observed value were obtained for 72.1% of drugs by both methods Ia and Ib, respectively, in which only rat pharmacokinetic data were used. The corresponding percentage was 75.0% for method II, in which pharmacokinetic data from animals other than rats were used. For BA, predicted values within 1.3 times the observed values were obtained for 66.7% and 57.4% of drugs by methods Ia and Ib, respectively, and 75.0% by method II.
Conclusions. Using the present methods, it is possible to predict BA from human oral administration data combined with animal pharmacokinetic data to a certain level without using intravenous injection data.

KEY WORDS: bioavailabilities; distribution volume; interspecies differences; intravenous administration; oral administration.

INTRODUCTION

The methods used to predict pharmacokinetics in humans include allometric scaling and *in vitro*/*in vivo* scaling. Allometric scaling is an animal scale-up method for the prediction of human pharmacokinetics from the *in vivo* pharmacokinetics of experimental animals that expresses the association between animal weight and pharmacokinetic parameters (mainly clearance or distribution volume) using an allometric formula (1,2) based on anatomical, physiological and biological equivalency in a variety of animals. Allometric scaling is an empirical method that has the disadvantage that species differences in drug metabolism are not considered. On the other hand, attempts have been made to predict *in vivo* clearance from *in vitro* metabolism studies using rats (3–5). This approach has also been applied to humans, and the prediction of human *in vivo* clearance from *in vitro* studies using human liver microsomes, hepatocytes and liver slices have been reported (6). Although allometric scaling shows high predictability for distribution volume, it has low predictability for clearance. Thus, allometric scaling has been performed incorporating *in vitro*/*in vivo* scaling for low clearance drugs (7). With this approach, bioavailability (BA) and plasma concentration profiles after oral administration can be predicted in humans (8). These predictions are extremely effective when selecting

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ABBREVIATIONS: β, elimination rate constant in the terminal phase; AUC_{iv}, area under the plasma concentration–time curve after intravenous administration; AUC_{po}, area under the plasma concentration–time curve after oral administration; BA, bioavailability; CL_{po}, oral clearance; CL_{tot}, total body clearance; f_P, free fraction in plasma; f_T, free fraction in tissue; Hc, hematocrit value; R_B, blood-to-plasma concentration ratio; $R_{E/I}$, ratio of amount of the binding protein in the extracellular fluid to that in plasma; V_B, volume of blood; Vd_β, distribution volume of the terminal phase; Vd_{ss}, distribution volume at steady state; V_E, volume of extracellular fluid outside the plasma; V_T, volume of tissue (except blood cells); V_T/f_T, volume of distribution for the unbound drug.

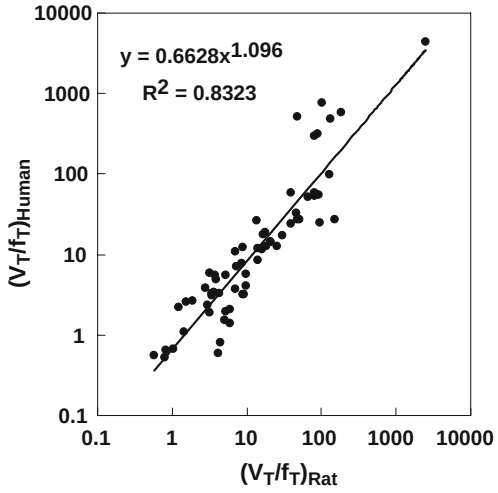

Fig. 1. Correlation between the values of $(V_T/f_T)_{rat}$ and $(V_T/f_T)_{human}$. The *broken line* indicates the regression curve.

compounds with better pharmacokinetic characteristics in the discovery and development stages of new drugs (9). However, most of the BA predictions currently performed address only first-pass metabolism in the liver, ignoring the fraction absorbed and metabolism in the gut after oral administration.

In this study, total body clearance (CL_{tot}) was obtained using the distribution volume of the terminal phase (Vd_β) predicted from animal data and the elimination rate constant (β) after oral administration of drugs in humans, and the absolute BA was calculated by dividing the CL_{tot} by the oral clearance (CL_{po}). In the presented method using human oral administration data, all absorption processes are evaluated, not only first-pass metabolism in the liver. Currently, intravenous administration studies in humans are not always performed during the development of novel oral agents. The purpose of this study was to predict the absolute human BA of drugs without using human intravenous injection data. This is not usually possible; however, we have used a combination of data from human oral administration and animal pharmacokinetic data to do so. The presented prediction methods are not for selecting better compounds during drug development, but for estimating the absolute BA of drugs selected as candidates without using intravenous injection data, even when they are difficult to administer intravenously because of poor solubility or hemolytic properties.

A retrospective analysis of the predictability of human BA was performed based on published data on animal pharmacokinetics and oral administration in humans.

MATERIALS AND METHODS

Data Collection

Drugs for which the absolute BA in human has been reported in Goodman and Gilman's *The Pharmacological Basis of Therapeutics* were selected. After a literature search for animal pharmacokinetic data (the distribution volume of terminal phase; Vd_β, the elimination rate constant; β, the free fraction in plasma; f_P, and the blood-to-plasma concentration ratio; R_B), and human pharmacokinetic data (oral clearance; CL_{po}, Vd_β, β, f_P and R_B), 67 drugs with available data for this study were identified. Supplementary Table in Electronic supplementary material shows the list of studied drugs. The R_B value was assumed to be the same among the animal species if information was available for any one animal species, including humans. For drugs with no available R_B information, R_B was assumed to be 0.945, the average value for 39 drugs for which the R_B value was available.

Prediction of Vd_β

When there are no active transporters in the main distribution organs, the unbound drug concentration at the steady state is the same in all places in the body. Defining Vd as the amount of drug in the body/plasma concentration, the relationship between the distribution volume at steady state (Vd_{ss}) and f_P is expressed as follows (10):

$$Vd_{ss} = V_B \times R_B + f_P \times V_E + (1 - f_P) \times R_{E/I} \times V_B \times (1 - Hc) + V_T \times (f_P/f_T) \quad (1)$$

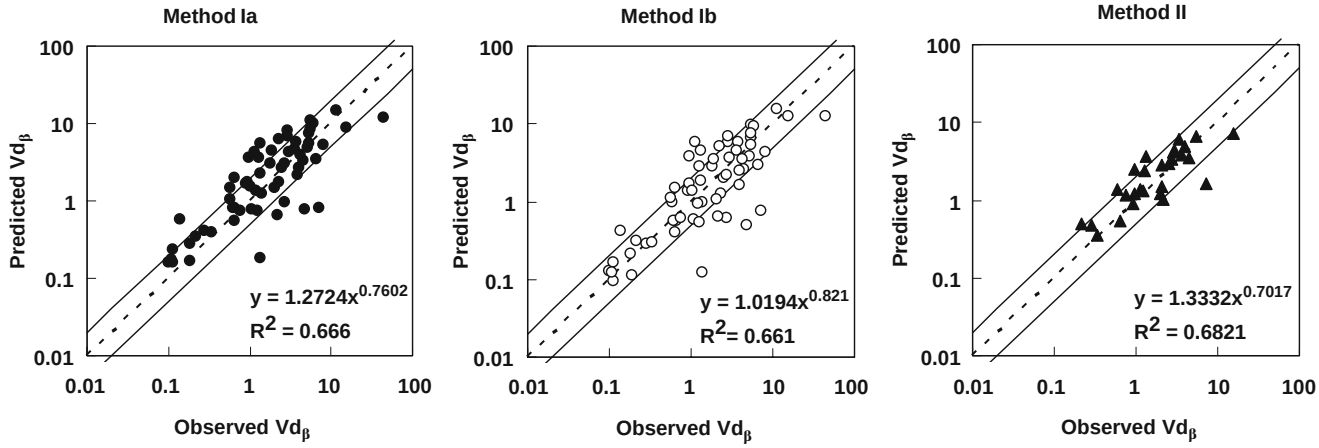

Fig. 2. Correlation between the predicted and observed volumes of distribution of 61 (methods Ia and Ib) and 21 (method II) drugs. The *broken line* indicates 1:1 correspondence. The *area between the solid lines* represents the area within twofold error. The original data for each compound are summarized in Table I.

Table I. Relationship Between Predicted and Observed Values of Volume of Distribution in Human

		Predicted Vd_β (L/kg)			Predicted $Vd_\beta/observed\ Vd_\beta$ [a] (L/kg)		
Drugs	Observed Vd_β (L/kg)	Method Ia	Method Ib	Method II	Method Ia	Method Ib	Method II
Acetaminophen	1.077	0.778	0.601		0.722	0.558	
Acyclovir	1.150			1.385			1.204
Betaxolol	5.370	7.488	6.477	6.477	1.394	1.206	1.206
Bisoprolol	2.897	6.921	5.710	4.063	2.389	1.971	1.403
Carbamazepine	1.400	1.244	0.966		0.889	0.690	
Carvedilol R-	2.697	0.942	0.616		0.349	0.228	
Carvedilol S-	4.805	0.760	0.502		0.158	0.104	
Cefixime	0.282	0.407	0.287	0.471	1.444	1.020	1.671
Chlorpheniramine	3.360			6.083			1.811
Cimetidine	1.200	1.346	0.950	1.337	1.122	0.791	1.114
Ciprofloxacin	2.320	1.728	1.269		0.745	0.547	
Codeine	5.250	4.926	3.843		0.938	0.732	
Cyclosporin	3.810	5.734	5.800		1.505	1.522	
Dexamethasone	0.920	1.650	1.374		1.793	1.493	
Diazepam	0.950	1.774	1.670	1.230	1.867	1.758	1.295
Diflunisal	0.099	0.159	0.130		1.604	1.312	
Digoxin	8.160	5.351	4.289		0.656	0.526	
Diltiazem	4.260	3.907	3.440		0.917	0.808	
Disopyramide	0.910			0.895			0.983
Etodolac S-	2.192	0.661	0.648	1.009	0.302	0.296	0.461
Etodolac R-	0.216	0.350	0.306	0.491	1.618	1.414	2.266
Felodipine	15.700	8.963	12.527	7.307	0.571	0.798	0.465
Flecainide	5.500	10.933	9.883		1.988	1.797	
Fluorouracil	0.659	0.821	0.555		1.245	0.842	
Fluoxetine	43.900	12.006	12.686		0.273	0.289	
Flurbiprofen	0.188	0.167	0.112		0.889	0.597	
Furosemide	0.187	0.273	0.209		1.461	1.117	
Hexobarbital	1.270	0.753	0.530		0.593	0.417	
Ibuprofen	0.112	0.235	0.164		2.098	1.461	
Imipramine	11.500	14.879	15.683		1.294	1.364	
Lidocaine	2.010			1.186			0.590
Meperidine	4.510	3.240	2.641	3.464	0.719	0.586	0.768
Methylprednisolone	1.350	2.283	1.889		1.691	1.399	
Metoprolol	2.900	8.249	6.784		2.844	2.339	
Mexiletine	6.020	10.028	9.463		1.666	1.572	
Midazolam	0.970	3.647	3.732	2.480	3.760	3.847	2.556
Morphine	3.920	2.173	1.618		0.554	0.413	
Nadolol	2.270	6.289	5.141		2.771	2.265	
Nicardipine	4.020	2.691	2.524	4.873	0.669	0.628	1.212
Nifedipine	1.140	4.249	5.817		3.727	5.102	
Nimodipine	0.59	1.034	0.963	1.377	1.753	1.632	2.335
Nitrendipine	7.299	0.818	0.749	1.619	0.112	0.103	0.222
Nizatidine	1.282	3.570	2.808	2.345	2.785	2.191	1.829
Ofloxacin	2.520	2.641	2.005	2.898	1.048	0.795	1.150
Omeprazole	0.340	0.397	0.293	0.352	1.169	0.863	1.035
Pentazocine	5.560	8.370	7.433		1.505	1.337	
Phenobarbital	0.624	0.805	0.570		1.290	0.913	
Phenylpropanolamine	2.087			2.850			1.366
Phenytoin	0.640	0.554	0.408		0.865	0.637	
Prednisolone	0.570	1.440	1.108		2.527	1.944	
Propranolol	3.620	4.621	4.577	3.769	1.276	1.264	1.041
Quinine	1.810	3.068	2.796		1.695	1.545	
Quinidine	3.020	4.259	3.721	4.399	1.410	1.232	1.457
Ranitidine	1.890	4.445	3.471		2.352	1.836	
Salicylic acid	1.380	0.183	0.122		0.133	0.089	
Sotalol	1.350	5.600	4.429	3.655	4.148	3.281	2.707
Sumatriptan	2.700	2.966	2.232	3.320	1.099	0.827	1.230
Tacrine	6.660	3.511	2.976		0.527	0.447	
Tolbutamide	0.112	0.157	0.094		1.402	0.841	
Trazodone	1.040	1.554	1.368		1.494	1.316	
Triazolam	0.630			0.532			0.844
Valproate	0.137	0.586	0.424		4.274	3.094	

Table I. (cotinued)

Drugs	Observed Vd_β (L/kg)	Predicted Vd_β (L/kg)			Predicted Vd_β/*observed* Vd_β [a] (L/kg)		
		Method Ia	Method Ib	Method II	Method Ia	Method Ib	Method II
Verapamil dl	5.490	5.424	5.329		0.988	0.971	
Warfarin	0.108	0.172	0.122		1.596	1.129	
Zalcitabine	0.648	1.990	1.469		3.071	2.267	
Zidovudine	2.060	1.482	1.065	1.486	0.719	0.517	0.721
Zolpidem	0.760	0.754	0.621	1.152	0.992	0.818	1.516

[a] Percent difference between observed and predicted values

where V_B, V_E, and V_T respectively represent the volume of blood, the extracellular fluid and the tissue (except blood cells) into which the drug is distributed; $R_{E/I}$ and Hc are the ratio of the amount of binding protein in the extracellular fluid to that in plasma and the hematocrit value, respectively.

Eq. 2 was derived assuming Vd_β equal to Vd_{ss}.

$$Vd_\beta = V_B \times R_B + f_P \times V_E + (1 - f_P) \times R_{E/I} \times V_B \times (1 - Hc) + V_T \times (f_P/f_T) \quad (2)$$

In this study, V_B, V_E, $R_{E/I}$, and Hc were assumed to be 80 ml/kg (11), 260 ml/kg (12,13), 1.4 (10) and 0.42, respectively. The volume of distribution for the unbound drug, V_T/f_T, can therefore be expressed as follows:

$$V_T/f_T = \left(Vd_\beta - 80R_B - 195f_P - 65\right)\Big/f_P. \quad (3)$$

Although there are some exceptions, it has been reported that plasma protein binding shows some species differences, while tissue binding, and hence the tissue distribution of unbound drug, shows little species differences (10,14). Based on this assumption, Vd_β was predicted by three methods.

Method Ia. Using pharmacokinetics data (Vd_β , R_B and f_P) from rats, which are the most widely used experimental animal, the tissue distribution volume of unbound drug $(V_T/f_T)_{rat}$ was obtained using Eq. 3. Based on the above assumption, this value was assumed to be the same in humans (10), that is:

$$(V_T/f_T)_{human} = (V_T/f_T)_{rat}. \quad (4)$$

The human Vd_β values were calculated from Eq. 3 using $(V_T/f_T)_{human}$ and human values of R_B and f_P.

Method Ib. Vd_β was calculated using the following equation, which incorporates the volume of extracellular fluid in the tissue volume (15).

$$Vd_\beta = V_B \times R_B + V_T \times (f_P/f_T) \quad (5)$$

$$V_T/f_T = \left(Vd_\beta - 80R_B\right)\Big/f_P \quad (6)$$

The values of $(V_T/f_T)_{rat}$ and $(V_T/f_T)_{human}$ were obtained from the data in Sawada's paper (16) and the present data using Eq. 6. Fig. 1 showed the correlation between the values of $(V_T/f_T)_{rat}$ and $(V_T/f_T)_{human}$. The regression formula between humans and rats was obtained as follows:

$$(V_T/f_T)_{human} = 0.6628(V_T/f_T)_{rat}^{1.096} \quad (7)$$

The value of $(V_T/f_T)_{human}$ was obtained from Eq. 7, and human Vd_β was calculated with Eq. 6 using the $(V_T/f_T)_{human}$ and human values for R_B and f_P.

Method II. As in method Ia, the value of V_T/f_T was obtained for each animal using the data not only from rats but also from other animals. Based on the assumption that the value of V_T/f_T is the same for all animals, human Vd_β values were obtained as in method Ia, assuming that the average V_T/f_T value among the animals is equal to the human V_T/f_T (10).

Prediction of BA

The absolute BA can be expressed as in Eq. 8:

$$BA = \frac{AUC_{po}/Dose_{po}}{AUC_{iv}/Dose_{iv}} \quad (8)$$

Table II. Statistical Data Comparing the Accuracy of Predictions

	Vd_β Prediction			BA Prediction		
	Method Ia	Method Ib	Method II	Method Ia	Method Ib	Method II
afe	1.85	1.80	1.56	1.41	1.48	1.30
egrse	2.20	2.25	1.76	1.70	1.86	1.48
r^2	0.666	0.661	0.682	0.664	0.573	0.821
N	61	61	28	61	61	28

afe average fold-error, *egrse* exponential geometric root mean squared prediction error

Table III. Relationship Between Predicted and Observed Values of Bioavailability in Human

Drugs	Observed BA (%)	Predicted BA (%)			Predicted BA/Observed BA[a]		
		Method Ia	Method Ib	Method II	Method Ia	Method Ib	Method II
Acetaminophen	88±15	42.2	32.6		0.480	0.370	
Acyclovir	22.5±7.5			16.3			0.725
Betaxolol	89±5	100.0	100.0	100.0	1.124	1.124	1.124
Bisoprolol	91±10	100.0	100.0	100.0	1.099	1.099	1.099
Carbamazepine	70	73.7	57.2		1.053	0.818	
Carvedilol R-	32±12	9.4	6.1		0.294	0.192	
Carvedilol S-	18±7	2.9	1.9		0.161	0.106	
Cefixime	47±15	60.9	43.1	70.5	1.296	0.916	1.501
Chlorpheniramine	45.5±8.8			48.0			1.056
Cimetidine	59.9±22.6	67.1	47.3	66.6	1.120	0.790	1.112
Ciprofloxacin	69±7	44.1	32.3		0.639	0.469	
Codeine	50±7	68.9	53.8		1.378	1.075	
Cyclosporin	23±7	31.7	32.1		1.380	1.396	
Dexamethasone	78±14	100.0	88.3		1.282	1.132	
Diazepam	100	100.0	100.0	85.8	1.000	1.000	0.858
Diflunisal	90	100.0	100.0		1.111	1.111	
Digoxin	70±13	71.2	57.1		1.018	0.816	
Diltiazem	44±10	39.9	35.1		0.907	0.799	
Disopyramide	81.3±13			79.8			0.981
Etodolac R-	97	100.0	100.0	100.0	1.031	1.031	1.031
Etodolac S-	73	37.0	36.3	56.5	0.507	0.497	0.774
Felodipine	15±8	10.0	14.0	8.2	0.668	0.933	0.544
Flecainide	70±11	100.0	100.0		1.429	1.429	
Fluorouracil	55±12	100.0	100.0		1.818	1.818	
Fluoxetine	60	27.4	28.9		0.456	0.482	
Flurbiprofen	92	94.3	63.3		1.025	0.688	
Furosemide	61±17	42.0	32.1		0.688	0.526	
Hexobarbital	90	62.8	44.2		0.698	0.491	
Ibuprofen	80	100.0	78.9		1.250	0.986	
Imipramine	41.9±9.5	43.8	46.1		1.044	1.101	
Lidocaine	12.8±2.4			21.9			1.707
Meperidine	57.7±4.9	44.0	35.9	47.0	0.763	0.622	0.815
Methylprednisolone	82±13	100.0	100.0		1.220	1.220	
Metoprolol	38±14	100.0	100.0		2.632	2.632	
Mexiletine	87±13	100.0	100.0		1.149	1.149	
Midazolam	44±17	100.0	100.0	100.0	2.273	2.273	2.273
Morphine	24±12	12.3	9.2		0.513	0.382	
Nadolol	34±5	39.3	32.1		1.156	0.945	
Nicardipine	18±11	7.5	7.1	13.7	0.419	0.393	0.759
Nifedipine	50±13	100.0	100.0		2.000	2.000	
Nimodipine	10±4	7.2	6.7	9.6	0.718	0.668	0.956
Nitrendipine	11±5	1.4	1.2	2.7	0.124	0.113	0.245
Nizatidine	90	100.0	100.0	100.0	1.111	1.111	1.111
Ofloxacin	100	100.0	100.0	100.0	1.000	1.000	1.000
Omeprazole	53±29	51.2	37.8	45.3	0.965	0.713	0.854
Pentazocine	18.4±7.8	32.2	28.6		1.748	1.552	
Phenobarbital	100±11	100.0	98.2		1.000	0.982	
Phenylpropanolamine	70			82.0			1.172
Phenytoin	90±3	82.4	60.6		0.915	0.673	
Prednisolone	82±13	100.0	100.0		1.220	1.220	
Propranolol	26±10	20.7	20.4	16.8	0.796	0.785	0.646
Quinine	90	100.0	93.6		1.111	1.040	
Quinidine	87±7	100.0	88.7	100.0	1.149	1.020	1.149
Ranitidine	52±11	100.0	100.0		1.923	1.923	
Salicylic acid	100	100.0	100.0		1.000	1.000	
Sotalol	95	100.0	100.0	100.0	1.053	1.053	1.053
Sumatriptan	14±5	11.5	8.7	12.9	0.822	0.619	0.920
Tacrine	17±3	12.1	10.2		0.711	0.603	
Tolbutamide	93±10	100.0	69.9		1.075	0.752	
Trazodone	81±29	100.0	100.0		1.235	1.235	
Triazolam	44			38.0			0.863
Valproate	94.5±6.4	100.0	100.0		1.058	1.058	

Table III. (continued)

Drugs	Observed BA (%)	Predicted BA (%)			Predicted BA/Observed BA[a]		
		Method Ia	Method Ib	Method II	Method Ia	Method Ib	Method II
Verapamil dl	22±8	19.6	19.2		0.890	0.875	
Warfarin	93±8	85.6	67.3		0.920	0.723	
Zalcitabine	88±17	100.0	100.0		1.136	1.136	
Zidovudine	63±13	49.7	35.7	49.8	0.789	0.567	0.791
Zolpidem	67±20	84.4	69.6	100.0	1.260	1.038	1.493

[a] Percent difference between observed and predicted values

where AUC_{po} and $Dose_{po}$ represent the area under the plasma concentration–time curve (AUC) and the dose (Dose) for oral administration, respectively and AUC_{iv} and $Dose_{iv}$ represent the AUC and Dose for intravenous administration, respectively. Eq. 8 can be converted as follows:

$$BA = \frac{CL_{tot}}{CL_{po}} \tag{9}$$

where CL_{tot} represents the total body clearance. CL_{tot} can also be expressed as $\beta \times Vd_{\beta}$, so BA can be expressed as shown in Eq. 10.

$$BA = {\beta \times Vd_{\beta}}\Big/{CL_{po}} \tag{10}$$

That is, if β and Vd_{β} can be obtained without data after intravenous administration, BA can be obtained without intravenous injection data. In this study, β was obtained from oral administration data assuming the absence of flip-flop phenomena.

Accuracy of Predictions

The observed values of human BA and Vd_{β} (Supplementary Table in Electronic supplementary material) obtained from the literature were compared with the values predicted in this study. In order to compare the accuracy of predictions based on the three models, the exponential geometric root mean squared prediction error (egrse) and the average fold-error (afe) were estimated as measures of precision and bias, respectively, for each set of predictions. This approach prohibited poor overpredictions from being canceled out by equally poor underpredictions. It also kept any single outlier prediction from biasing conclusions concerning a particular prediction method. A method that predicted all observed values perfectly would have a value of 1; one that made predictions that were on average twofold off (100% above or 50% below) would have a value of 2, and so forth.

$$\text{afe} = 10^{\frac{1}{N}\sum\left|\log\frac{\text{Predicted}}{\text{Observed}}\right|}$$

$$\text{egrse} = 10^{\sqrt{\frac{1}{N}\sum\left(\log\frac{\text{Predicted}}{\text{Observed}}\right)^2}}$$

RESULTS

Prediction of Vd_{β}

The Vd_{β} values for 61 of the 67 compounds for which rat pharmacokinetic parameters were available were predicted using methods Ia and Ib. The Vd_{β} values for 28 compounds for which pharmacokinetic parameters of animals other than rats were available were predicted by method II. Fig. 2 and Table I show the relationship between the human Vd_{β} values predicted from the animal data and the values obtained from the literature. The number of compounds (out of 61) for which the predicted value fell within twice as much as the observed value was 44 (72.1%) and 44 (72.1%) by methods Ia and Ib, respectively, both using only rat pharmacokinetic parameters. The corresponding number of compounds was 21 out of 28 (75.0%) for method II, which used pharmacokinetic parameters from animals other than rats. Table II shows the statistical comparison of the three methods. The afe values for methods Ia, Ib and II were 1.85, 1.80 and 1.56, respectively, and the egrse values were 2.20, 2.25 and 1.76, respectively. From the high fraction of compounds whose predicted values fell within twice as much as the observed values and from the statistical comparison of the accuracy, prediction by method II showed the highest accuracy among the three methods.

Prediction of BA

Table III and Fig. 3 show the relationships between the predicted and observed values of BA. Predicted values exceeding 100% are given as 100%. The number of compounds (out of 61) for which the predicted value fell within twice as much as the observed value was 53 (86.9%) and 49 (80.3%) by methods Ia and Ib, respectively, both using only rat pharmacokinetic parameters. The corresponding number of compounds was 26 out of 28 (92.9%) for method II, which used pharmacokinetic parameters from animals other than rats. The number of compounds for which the predicted value fell within 1.3 times as much as the observed value was 40 (66.7%) and 35 (57.4%) out of 61 by methods Ia and Ib, respectively, and 21 (75.0%) out of 28 by method II. Table II shows the statistical comparison of the three methods. The afe values for methods Ia, Ib and II were 1.41, 1.48 and 1.30, respectively, and the egrse values were

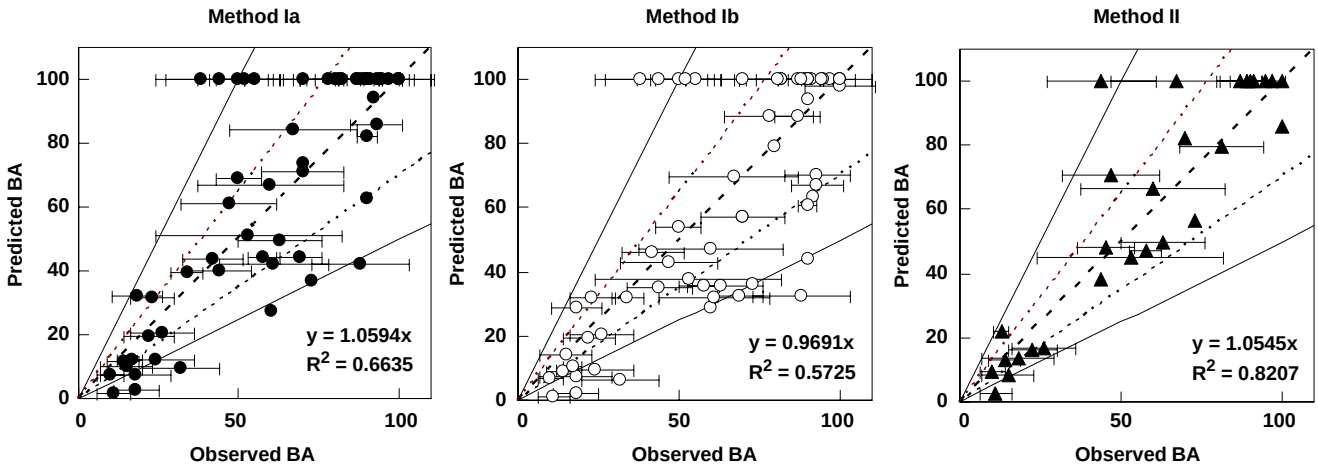

Fig. 3. Correlation between the predicted and observed bioavailability of 61 (methods Ia and Ib) and 21 (method II) drugs. The *broken line* indicates a 1:1 correspondence. The *thin broken lines* indicate the 30% variable limit of the estimation. The *area between the solid lines* represents the area within twofold error. Predicted values over 100% are indicated as 100% in this figure. The original data for each compound are summarized in Table III.

1.70, 1.86 and 1.48, respectively. Judging from the high fraction of compounds whose predicted values fell within twice or 1.3 times as much as the observed values and from the statistical comparison of the accuracy, prediction by method II showed the highest accuracy of the three methods.

To investigate the relationship between the predicted and observed values of BA in relation to the observed values, the compounds were divided into three groups depending on the observed value of BA: less than 30%, from 30% to 70%, and 70% or more. The predicted results by each method are shown in Table IV. Using methods Ia and Ib, of the 12 drugs with an observed BA of less than 30%, the low BA (less than 30%) was successfully predicted in all but two drugs by method Ia (pentazocine and cyclosporine: the observed values were 18.4% and 23%, and the predicted values were 32.2% and 31.7%, respectively), and one drug by method Ib (cyclosporine: the predicted value was 32.1%). Of the 30 drugs with an observed BA of 70% or more, three drugs had predicted values of less than 70% by method Ia (S-etodolac, acetaminophen, and hexobarbital: the observed values were

Table IV. Classification of Predicted Bioavailability in each Prediction Methods

Observed BA	<30%	30~70%	>70%
Method Ia			
Number of drugs	12	19	30
Good estimation	10	11	27
Over estimation	2	6	–
	[31.7(23), 32.2(18.4)]	[100(55), 100(38), 100(44), 100(50), 100(52), 84.4(67)]	
Under estimation	–	2	3
		[9.4(32), 27.4(60)]	[42.2(88), 37.0(73), 62.8(90)]
Method Ib			
Number of drugs	12	19	30
Good estimation	11	12	21
Over estimation	1	5	–
	[32.1(23)]	[100(55), 100(38), 100(44), 100(50), 100(52)]	
Under estimation	–	2	9
		[6.1(32), 28.9(60)]	[32.6(88), 57.2(70), 57.1(70), 36.3(73), 63.3(92), 44.2(90), 60.6(90), 69.9(93), 67.3(93)]
Method II			
Number of drugs	8	9	11
Good estimation	8	6	10
Over estimation	0	3	–
		[70.5(47), 100(44), 100(67)]	
Under estimation	–	0	1
			[56.5(73)]

[] The parenthetic values represent the predicted bioavailability; *()* the parenthetic values represent the observed bioavailability; *Good estimation* the drug which have <30%, 30–70% and >70% of the observed bioavailability predicted <30%, 30–70% and >70% of bioavailability, respectively; *Over estimation* the drug which have <30% and 30–70% of the observed bioavailability predicted >30% and >70% of bioavailability, respectively; *Under estimation* The drug which have 30–70% and >70% of the observed bioavailability predicted <30% and <70% of bioavailability, respectively

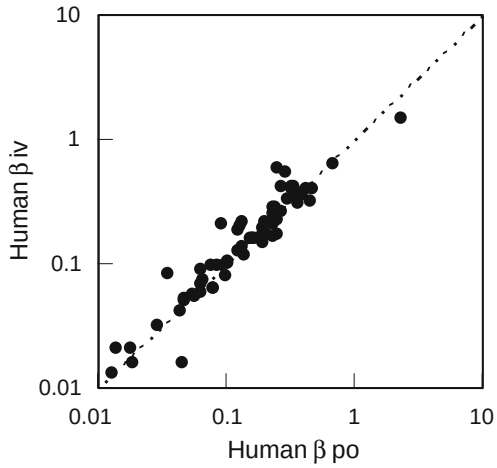

Fig. 4. Correlation of the elimination rate constants in the terminal phase between intravenous administration and oral administration. The *broken line* indicates 1:1 correspondence.

73%, 88%, and 90% and the predicted values were 37.0%, 42.2%, and 62.8%, respectively), and nine drugs had predicted values of less than 70% by method Ib (carbamazepine, digoxin, S-etodolac, acetaminophen, hexobarbital, phenytoin, flurbiprofen, tolbutamide, and warfarin: the observed values were 70%, 70%, 73%, 88%, 90%, 90%, 92%, 93%, and 93%, and the predicted values were 57.2%, 57.1%, 36.3%, 32.6%, 44.2%, 60.6%, 63.3%, 69.9%, and 67.3%, respectively). Among the 19 drugs with observed BA values of 30% to 70%, 11 and 12 had predicted values ranging from 30% to 70% by methods Ia and Ib, respectively, two had predicted values of less than 30% by both method Ia and Ib (R-carvedilol and fluoxetine: the observed values were 32% and 60% and the predicted values by method Ia were 9.4% and 27.4%, respectively, and the predicted values by method Ib were 6.1% and 28.9%, respectively), six had predicted values of more than 70% by method Ia (metoprolol, midazolam, nifedipine, ranitidine, fluorouracil, and zolpidem: the observed values were 38%, 44%, 50%, 52%, 55%, and 67%, and the predicted values were 100%, 100%, 100%, 100%, 100% and 84.4%, respectively), and five had predicted values of more than 70% by method Ib (metoprolol, midazolam, nifedipine, ranitidine, and fluorouracil: the observed values were 38%, 44%, 50%, 52%, and 55%, and the predicted values were all 100%; Table IV).

Using method II, predicted values of less than 30% were obtained for all eight drugs whose observed values were less than 30%. Among the 11 drugs with observed values of more than 70%, predicted values of more than 70% were obtained for all but one drug (S-etodolac: observed value 73%, predicted value 56.5%). As for the drugs with observed values of 30% to 70%, predicted values ranging from 30% to 70% were obtained for six of the nine drugs and predicted values of more than 70% were obtained for the other three drugs (midazolam, cefixime and zolpidem: the observed values were 44%, 47% and 67% and the predicted values were 100%, 70.5% and 100%, respectively; Table IV).

DISCUSSION

In this study, the CL_{tot}, which cannot usually be obtained without intravenous injection data, was estimated from β after oral administration and Vd_β , where Vd_β was predicted from animal data, and the BA was calculated. Several assumptions were necessary to apply this method. The first assumption is the absence of flip-flop phenomena, that is, the elimination phase is the same for both intravenous and oral administration. The validity of this assumption can be confirmed using experimental animals. The elimination process has to be slower than the absorption process so that flip-flop does not occur. In many cases, the absorption process is rate limited by membrane penetration, and species differences are small except for some special cases involving active transport, etc. However, the elimination process consists of metabolism and excretion, where the metabolic clearance is generally known to be larger in experimental animals than in humans. Therefore, considering the small differences in the absorption processes between humans and experimental animals, as well as the slower elimination in humans, it is reasonable to assume that if flip-flop was not observed in the experimental animals, it would not be observed in humans either. The elimination half-lives of the drugs used in the present study after intravenous and oral administration in humans are

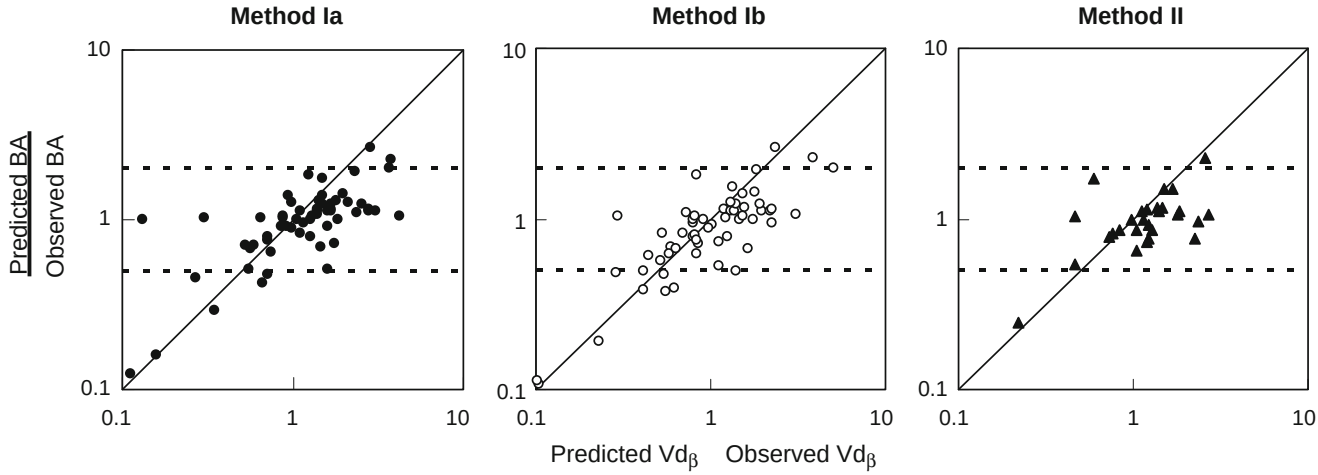

Fig. 5. Correlation of the predicted/observed ratio between the volume of distribution and bioavailability. The *solid line* indicates 1:1 correspondence. The *dotted lines* indicate ratios of the predicted BA to the observed BA of 0.7 and 1.3, respectively.

compared in Fig. 4. As is clearly shown in the figure, the elimination half-lives after oral administration had 1:1 correspondence with those after intravenous administration, indicating the absence of flip-flop phenomena for the tested drugs. Furthermore, although the present prediction assumes $Vd_{ss} = Vd_{\beta}$, there are some exceptions where this assumption does not hold true. Generally, these two values of distribution volume have the relationship of:

$$\mathrm{Vd}_{\beta} > \mathrm{Vd}_{\mathrm{ss}}.$$

While $\mathrm{Vd}_{\mathrm{ss}}$ shows the actual distribution size and is not affected by the clearance magnitude, Vd_{β} overestimates the true distribution volume ($\mathrm{Vd}_{\mathrm{ss}}$) when the clearance is large. However, many drugs are classified as low clearance drugs for humans, so Vd_{β} can be approximated by $\mathrm{Vd}_{\mathrm{ss}}$. Of the 44 of the 67 drugs whose $\mathrm{Vd}_{\mathrm{ss}}$ and Vd_{β} values in humans were available, four drugs (cimetidine, cyclosporine, nicardipine and omeprazole) had Vd_{β} values more than twice the $\mathrm{Vd}_{\mathrm{ss}}$ value, but the prediction of Vd_{β} was not particularly poor for those compounds.

Attempts have been made to discuss the possible reasons for some poor predictions. Fig. 5 shows the relationship between predicted/observed ratio for BA and that for Vd_{β} . The value on the vertical axis is close to 1 for the successful BA predictions. Even for the compounds with poor prediction of BA (a predicted/observed ratio far from 1), the prediction/observed ratio for BA and that for Vd_{β} showed 1 to 1 correspondence, indicating that the poor prediction of BA is largely attributed to the poor prediction of Vd_{β} . One of the reasons for poor prediction of distribution volume contributing to most of the poor predictions of BA may be the invalid assumption of the absence of species difference in the tissue distribution of unbound drugs. Although several parameters were investigated for the possibility of contributing to poor prediction of distribution volume, including lipid solubility (log$P$ value), f_{P} value, Vd_{β} and CL, none of the parameters showed a significant correlation (data not shown).

Alternatively, the Vd_{β} per kilogram body weight can be predicted from the animal Vd_{β} (per kilogram basis) assuming a simple allometry with correction for protein binding, as follows.

$$\mathrm{Vd}_{\beta,\mathrm{human}} = \mathrm{Vd}_{\beta,\mathrm{animal}} \times \left(f_{\mathrm{P,human}} / f_{\mathrm{P,animal}}\right)$$

However, the predicted values using this simplified method differed from those of the presented method for drugs with small Vd_{β} values (data not shown). Compared with the simplified method, the presented method is more faithful to the theoretical equation and resulted in better predictions for Vd_{β} and BA, even when drugs had a small Vd_{β} .

The drugs used in the presented predictions are typically administered orally, so most of them had relatively high BA values. Because BA can be problematic in clinical practice, mainly for drugs with low BA values, it is important to investigate whether a low BA can be predicted by the presented methods. The purpose of the predictions in this study was not to arrive at the same value as the observed value, but to predict a low BA for drugs with low BA and a high BA for those with high BA. When the drugs were divided into groups according to the observed BA values, a low BA (less than 30%) was successfully predicted by method Ia and Ib in all but two and one of 12 drugs, respectively. For the drugs with a BA of 70% or more, the numbers of drugs whose predicted values fell below 70% were three of 30 by method Ia, nine of 30 by method Ib and one of 11 by method II. The goal of predicting high values for high BA drugs and low values for low BA drugs has thus been predominantly attained by the presented methods.

In conclusion, this study showed the possibility of predicting human BA values from human oral administration data and animal pharmacokinetics, without needing data from intravenous injections. A low BA could lead to a large interindividual variation in blood concentration and AUC, affecting the apparent drug effects and adverse events that could cause clinical problems, especially for drugs with a narrow therapeutic range. Successful prediction of a low BA by the presented methods would be useful for vigilance in drug administration and for designing dosage regimens.

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