Prediction of Pharmacokinetics Prior to *In Vivo* Studies. 1. Mechanism-Based Prediction of Volume of Distribution

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ABSTRACT: In drug discovery and nonclinical development the volume of distribution at steady state (V_{ss}) of each novel drug candidate is commonly determined under *in vivo* conditions. Therefore, it is of interest to predict V_{ss} without conducting *in vivo* studies. The traditional description of V_{ss} corresponds to the sum of the products of each tissue:plasma partition coefficient ($P_{t:p}$) and the respective tissue volume in addition to the plasma volume. Because data on volumes of tissues and plasma are available in the literature for mammals, the other input parameters needed to estimate V_{ss} are the $P_{t:p}$'s, which can potentially be predicted with established tissue composition-based equations. *In vitro* data on drug lipophilicity and plasma protein binding are the input parameters used in these equations. Such a mechanism-based approach would be particularly useful to provide first-cut estimates of V_{ss} prior to any *in vivo* studies and to explore potential unexpected deviations between sets of predicted and *in vivo* V_{ss} data, when the *in vivo* data become available during the drug development process. The objective of the present study was to use tissue composition-based equations to predict rat and human V_{ss} prior to *in vivo* studies for 123 structurally unrelated compounds (acids, bases, and neutrals). The predicted data were compared with *in vivo* data obtained from the literature or at Roche. Overall, the average ratio of predicted-to-experimental rat and human V_{ss} values was 1.06 (SD = 0.817, $r = 0.78$, $n = 147$). In fact, 80% of all predicted values were within a factor of two of the corresponding experimental values. The drugs can therefore be separated into two groups. The first group contains 98 drugs for which the predicted V_{ss} were within a factor of two of those experimentally determined (average ratio of 1.01, SD = 0.39, $r = 0.93$, $n = 118$), and the second group includes 25 other drugs for which the predicted and experimental V_{ss} differ by a factor larger than two (average ratio of 1.32, SD = 1.74, $r = 0.42$, $n = 29$). Thus, additional relevant distribution processes were neglected in predicting V_{ss} of drugs of the second group. This was true especially in the case of some cationic-amphiphilic bases. The present study is the first attempt to develop and validate a mechanistic distribution model for predicting rat and human V_{ss} of drugs prior to *in vivo* studies. © 2002 Wiley-Liss, Inc. and the American Pharmaceutical Association J Pharm Sci 91:129–156, 2002

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INTRODUCTION

The volume of distribution at steady state (V_{ss}) and clearance processes characterize the disposition of drugs. The estimation of V_{ss} is therefore essential. V_{ss} corresponds to the equivalent plasma volume in which a drug is distributed into the body. In fact, V_{ss} would equal the plasma volume in addition to the sum of each tissue: plasma partition coefficient ($P_{t:p}$) multiplied with its respective tissue volume.[1] The $P_{t:p}$ represents the concentration ratio between a tissue and

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plasma at steady state. The estimation of V_{ss} *in vivo* by using $P_{t:p}$ data is a time consuming process in drug discovery and nonclinical development because determination of $P_{t:p}$ involves intravenous constant infusions to animals followed by an extraction and quantification of drugs from tissue homogenates.[1,2] At present, V_{ss} under *in vivo* conditions is commonly determined by the product of clearance and mean residence time after a single intravenous dose of a drug according to the statistical moment theory.[3] For efficient screening efforts it could be of interest to predict V_{ss} without conducting *in vivo* studies. In other words, it could be useful to predict V_{ss} *in vivo* from *in vitro* parameters routinely determined in drug discovery.

The tools available to date for predicting V_{ss} or V at non–steady state conditions are semiempirical equations solved after their fit to *in vivo* data that have been previously collected.[1,4,5] General or specific rules for the mechanisms underlying V_{ss} *in vivo* cannot easily be developed from such regression analyses. The semiempirical equations may provide predictions of V_{ss}, but only for the species and kinds of drugs for which the fitting exercises have previously been elaborated. The resulting regression analyses, however, show that drug lipophilicity and plasma protein binding are probably two main determinants of V_{ss}.[1,4] Therefore, the prediction of V_{ss} prior to *in vivo* studies may potentially be performed from mechanistic equations that take into account the compound- and species-specific determinants of V_{ss}, and relate the compound-specific determinants to drug lipophilicity and plasma protein binding determined *in vitro*.

It has recently been demonstrated that *in vivo* $P_{t:p}$ of nonadipose and adipose tissues, the main compound-specific determinants of V_{ss}, can reasonably be predicted by using *in vitro* data on drug lipophilicity and plasma protein binding as sole input parameters in mechanistic tissue composition-based equations.[6,7] The tissue composition-based equations have been validated with literature data on more than 310 $P_{t:p}$'s of mammals for about 80 structurally unrelated drugs. The mechanistic basis of these equations is that each tissue and plasma is a mixture of lipids, water, and plasma proteins in which the drug can be homogeneously distributed. In fact, the first term of these equations is based upon the drug lipophilicity in accordance with the lipophilicity-hydrophilicity balance of tissues and plasma due to their lipid and water contents. The second term of the equations considers the binding to common proteins present in plasma and tissue interstitial space. Thus, the main assumption with these equations is that a passive diffusion process mainly governs tissue distribution of a drug to permit its partitioning into tissue lipids and water or into the extracellular water space only, and its reversible binding to common proteins present in plasma and tissue interstitial space. The drug-specific input parameters required in tissue composition-based equations (i.e., lipophilicity and plasma protein binding data) are generally available in drug discovery from *in vitro* studies. Therefore, the hypothesis of the present study was that V_{ss} *in vivo* could potentially be predicted on a mechanistic basis from data on $P_{t:p}$'s calculated with tissue composition-based equations.[6,7] The other determinants of V_{ss}, the data on tissue volumes, can be obtained from the literature.

The objective of the present study was to use established mechanistic tissue composition-based equations to predict rat and human V_{ss} of 123 structurally unrelated drugs (acids, bases, and neutrals) without using *in vivo* data as input information.

METHODS

The overall method was based on a conventional equation that describes V_{ss}. In this equation the input parameters relative to tissue distribution (i.e., $P_{t:p}$'s) were predicted from established tissue composition-based equations, whereas the input parameters relative to the species (tissue volumes) were obtained from the literature. Two kinds of drugs were considered: those for which a homogeneous distribution into a tissue by passive diffusion can be assumed, and those for which a distribution mainly into the extracellular space can be assumed. In fact, the method was applied to 123 drugs covering a broad range of physicochemical (lipophilicity and ionization) and biochemical (plasma protein binding) properties. These drugs were chosen based on the availability of literature or Roche data on V_{ss} determined *in vivo*. The validation of the method consisted of comparing the predicted rat and human V_{ss} with corresponding experimental *in vivo* data obtained from the literature or at Roche.

Prediction of *In Vivo* V_{ss} in Rats and Humans for 123 Structurally Unrelated Drugs

V_{ss} referring to plasma pharmacokinetics was predicted with this conventional equation:[1]

$$V_{ss} = (\Sigma V_t^* P_{t:p}) + (V_e^* E{:}P) + V_p \qquad (1)$$

where V is the fractional body volume (L/kg) of a tissue (t), erythrocyte (e), and plasma (p), $E{:}P$ is the erythrocyte:plasma ratio, and $P_{t:p}$ is the nonadipose and adipose tissue:plasma partition coefficients (PCs). In eq. 1, the species-specific input parameters (V_t, V_e, V_p) can be obtained from the literature, whereas the drug-specific input parameters ($P_{t:p}$, $E{:}P$) can be estimated without using *in vivo* data. The estimation of $P_{t:p}$ and $E{:}P$ is described in the following section.

Prediction of $P_{t:p}$ of Nonadipose and Adipose Tissues With Tissue Composition-Based Equations for Drugs Which Distribute Homogeneously Into Tissues Mainly by Passive Diffusion

In predicting V_{ss}, the main tissues and plasma where a drug can be distributed were chosen to cover at least 80% of the body weight. Therefore, the nonadipose tissues represent the bone + marrows, brain, gut, heart, kidney, liver, lung, skeletal muscle, skin, and spleen, whereas the adipose tissue refers to subcutaneous white fat. The *in vivo* $P_{t:p}$ values of nonadipose and adipose tissues used in eq. 1 were calculated by using *in vitro* data on drug lipophilicity and plasma protein binding as sole input parameters in mechanistic tissue composition-based equations.[6,7] The basis and assumptions of these equations are already presented in the Introduction section. Briefly, the drug would distribute homogeneously into each tissue (and plasma) mainly by passive diffusion. Consequently, the drug would partition between lipids and water as well as bind reversibly to common proteins present in plasma and tissue interstitial space. The tissue composition-based equations used to predict the $P_{t:p}$'s at the organ level are presented as follows:[6,7]

$$P_{t:p\ \text{nonadipose}} = \frac{[P_{o:w} \times (V_{nlt} + 0.3 \times V_{pht})] + [1 \times (V_{wt} + 0.7 \times V_{pht})]}{[P_{o:w} \times (V_{nlp} + 0.3 \times V_{php})] + [1 \times (V_{wp} + 0.7 \times V_{php})]} \times \frac{fu_p}{fu_t} \qquad (2)$$

$$P_{t:p\ \text{adipose}} = \frac{[D^*_{vo:w} \times (V_{nlt} + 0.3 \times V_{pht})] + [1 \times (V_{wt} + 0.7 \times V_{pht})]}{[D^*_{vo:w} \times (V_{nlp} + 0.3 \times V_{php})] + [1 \times (V_{wp} + 0.7 \times V_{php})]} \times \frac{fu_p}{1} \qquad (3)$$

where $P_{o:w}$ is the n-octanol:buffer PC of the nonionized species at pH 7.4, $D^*_{vo:w}$ is the olive oil:buffer PC of both the nonionized and ionized species at pH 7.4, V is the fractional tissue volume content of neutral lipids (nl), phospholipids (ph), and water (w), t is tissue, p is plasma, and fu is unbound fraction. Each tissue is a mixture of lipids, water, and proteins with a pH set equal to 7.4. $P_{o:w}$ and $D^*_{vo:w}$ were used to estimate hydrophobic interactions of drugs with the biotic lipids of nonadipose and adipose tissues, respectively, whereas fu_p and fu_t were used to estimate a specific macromolecular binding to common proteins present in plasma and tissue interstitial space, respectively. A detailed derivation of eqs. 2 and 3 was recently provided by Poulin et al.[6,7] Each input parameter of eqs. 2 and 3 can be estimated prior to *in vivo* studies. In fact, the physiological data were obtained from a review of the literature, whereas data on drug lipophilicity and protein binding were determined from *in vitro* studies or were calculated (see the section concerning the estimation of these input parameters).

Prediction of $P_{t:p}$ of Nonadipose and Adipose Tissues With Tissue Composition-Based Equations for Drugs Which Distribute Mainly Into Extracellular Space

For drugs distributing mainly into the extracellular space of tissues, their $P_{t:p}$'s of both nonadipose and adipose tissues cannot be calculated with eqs. 2 and 3. Alternatively, other tissue composition-based equations obtained from the literature[6] were used to estimate their values of $P_{t:p}$ used in the general eq. 1:

$$P_{t:p\ \text{nonadipose}} = \frac{V_{eist}}{V_{eisp}} \times \frac{fu_p}{fu_t} \qquad (4)$$

$$P_{t:p\ \text{adipose}} = \frac{V_{eist}}{V_{eisp}} \times \frac{fu_p}{1} \qquad (5)$$

where V_{eis} is the fractional volume content of extracellular (interstitial) space in tissue (t) and plasma (p). This kind of physiological data can be obtained from the literature (see the section

concerning the estimation of these input parameters).

Estimation of the Input Parameters of Eqs. 1–5

Values of the Physiological Input Parameters

Table 1 summarizes the mean rat and human data on tissue volumes used in eq. 1 (V_t, V_e, V_p), tissue composition used in eqs. 2 and 3 (V_{nl}, V_{ph}, V_w), and extracellular space used in eqs. 4 and 5 (V_{eis}). The estimation of these data from the literature is presented in the Appendix.

Values of the Biochemical Input Parameters

The biochemical parameters refer to *E:P* for erythrocyte:plasma ratio of eq. 1 as well as fu_p and fu_t of eqs. 2–5. The *E:P in vivo* was estimated from data on blood:plasma ratio (*B:P*) determined *in vitro* and the hematocrit content in blood (*Ht*) (assumed to be 45%) according to this conventional equation:

$$E{:}P = [B{:}P - (1 - Ht)]/Ht \qquad (6)$$

For drugs investigated in the present study, the mean data on *B:P* determined *in vitro* are presented in Tables 2–4 with their corresponding literature sources. However, the *in vitro* data on *B:P* were not available for all drugs of this study. Alternatively, *B:P* used in eq. 6 was set equal to 1 for the drugs that distribute homogeneously into tissues, whereas it was set equal to 0.55 (1 − hematocrit value) for the drugs that distribute only in the extracellular space. This has a minor impact on the calculated V_{ss} considering that a change of *B:P* from 0.5 to 2.0 changes V_{ss} only by a factor of 15% in the worse cases (not shown).

Values of fu_p were obtained from experimental *in vitro* studies published in the literature (Tables 2–4). The mean experimental value of fu_p of each drug is presented in Tables 2–4 with the corresponding literature sources. For racemic drugs, the average for the two enantiomers is presented. For some drugs, fu_p is concentration dependent. Where a significant concentration dependency of fu_p was reported in the literature cited, fu_p values were chosen according to similar plasma concentrations in both the *in vitro* binding studies and in the *in vivo* distribution studies. The values of fu_t used for tissues were estimated from data on fu_p of plasma and tissue interstitial fluid-to-plasma concentration ratios of albumin, globulins, and lipoproteins; these ratios for mammals were estimated as equal to an intermediate value of approximately 0.5 based on literature

Table 1. Physiological Parameters for Volumes and Composition of Tissues of Adult Male Rats and Humans Used for Predicting V_{ss}[a]

	Tissue Volume		Tissue Composition (Volume Fraction of Wet Tissue Weight)						
	(Fraction of Body Weight; L/kg)		Water (V_w)		Neutral Lipids (V_{nl})		Phospholipids (V_{ph})		Extracellular (Interstitial) Space (V_{eis})
Tissues	Rat	Human	Rat	Human	Rat	Human	Rat	Human	Rat
Adipose	0.0761	0.11957	0.12	0.18	0.853	0.79	0.002	0.002	0.175
Bone	0.041476	0.085629	0.446	0.439	0.0273	0.074	0.0027	0.0011	0.42
Brain	0.0057	0.02	0.788	0.77	0.0392	0.051	0.0533	0.0565	0.162
Gut	0.027	0.0171	0.749	0.718	0.0292	0.0487	0.0138	0.0163	0.39
Heart	0.0033	0.0047	0.779	0.758	0.014	0.0115	0.0118	0.0166	0.156
Kidney	0.0073	0.0044	0.771	0.783	0.0123	0.0207	0.0284	0.0162	0.346
Liver	0.0366	0.026	0.705	0.751	0.0138	0.0348	0.0303	0.0252	0.159
Lung	0.005	0.0076	0.79	0.811	0.0219	0.003	0.014	0.009	0.484
Muscle	0.404	0.40	0.756	0.76	0.01	0.0238	0.009	0.0072	0.115
Skin	0.190	0.0371	0.651	0.718	0.0239	0.0284	0.018	0.0111	0.462
Spleen	0.002	0.0026	0.771	0.788	0.0077	0.0201	0.0136	0.0198	0.264
Plasma	0.0449	0.0424	0.96	0.945	0.00147	0.0035	0.00083	0.00225	1.00
Erythrocytes	0.0367	0.0347	—	—	—	—	—	—	—

[a]Mean values obtained from a review of the literature. See Appendix for the estimation and references concerning V_t, V_p, V_e, V_w, V_{nl}, V_{ph}, and V_{eis}. Data represent a rat of 250 g and a human of 70 kg.
—, Not used in the present study.

Table 2. Values of log $P_{o:w}$, log $D_{vo:w}$, pK_a[c], fu_p, and *B:P* as well as Predicted and Experimental in vivo Values of V_{ss} in Adult Male Rats and Humans

Drugs	Log $P_{o:w}$[a]	Log $D^*_{vo:w}$[b]	pK_a[c]	fu_p[d] Rat	fu_p[d] Human	*B:P*[e] Rat	*B:P*[e] Human	Volume of Distribution at Steady State (V_{ss}) (L/kg)[f] Rat Predicted	Rat Experimental	Human Predicted	Human Experimetnal	References
Acids												
Acetaminophen	0.46	(−0.842)	9.38	0.88	1.0	(1.0)	(1.0)	0.64	0.54–0.797	0.63	0.85–1.42	[8,20–23]
Acetazolamide	−0.26	(−1.941)	7.4	—	0.044	—	(1.0)	—	—	0.32	0.38 ± 0.05	[8,24,25]
Acitretin	6.40	(3.484)	(5.10)	0.01	—	(1.0)	—	4.60	3.57	—	—	[8,26,27]
Ascorbic acid	−1.85	−2.34	4.70	—	0.76	—	0.82	—	—	0.50	0.72	[4,8,28,29]
Mehyl-ethyl barbiturate	0.08	(−1.338)	8.11	1.0	—	1.0	—	0.65	0.78	—	—	[8,30]
Butyl-ethylbar biturate	1.73	0.41	7.81	0.60	—	1.42	—	1.20	1.20	—	—	[8,10,30]
Pentyl-ethyl barbiturate	2.24	0.46	8.00	0.486	—	1.52	—	1.99	1.64	—	—	[8,10,30]
Hexyl-ethyl-bar biturate	2.46	(1.229)	7.74	0.18	—	1.0	—	2.16	1.80	—	—	[8,30]
Heptyl-ethyl barbiturate	2.91	(1.743)	7.78	0.07	—	1.0	—	3.07	1.67	—	—	[8,30]
Nonyl-ethylbar biturate	4.00	(2.970)	7.82	0.01	—	1.0	—	4.31	3.03	—	—	[8,30]
Captopril	0.34	(−4.671)	3.70	—	0.73	—	(1.0)	—	—	0.53	0.70–0.84	[8,31]
Dideoxyinosine	−1.24	(−2.743)	9.0	0.965	0.965	(1.0)	(1.0)	0.62	0.48–0.53	0.55	0.73–0.77	[8,32]
Isoniazid	−0.70	(−7.711)	1.82	—	1	—	(1.0)	—	—	0.56	0.59	[4,8]
Mephenytoin	1.69	(0.502)	8.51	0.63	—	(1.0)	—	1.20	1.29	—	—	[8,33–35]
Metronidazole	−0.02	(−6.152)	2.62	—	0.80	—	(1.0)	—	—	0.53	0.51–1.1	[8,36,37]
Pentobarbital	2.10	0.65	8.11	0.574	0.53	(1.0)	(1.0)	1.81	1.30 ± 0.10	1.96	0.99	[4,8,38,39]
Phenobarbital	1.47	−0.244	7.30	—	0.543	—	0.861	—	—	0.88	0.54–0.62	[8,28,40,41]
Prazosin	1.28	(−0.373)	7.14	—	0.053	—	0.70	—	—	0.49	0.46–0.77	[8,42,43]
Tetrahydrocan nabinol	6.97	(6.421)	10.6	—	0.115	—	0.55	—	—	5.53	8.94 ± 4.23	[8,44–46]
Thiopental	2.85	1.80	7.45	—	0.112	—	1.0	—	—	2.87	1.81–2.20	[8,38,47,48]
Bases												
Acebutolol	1.71	(−1.716)	9.67	—	0.743	—	(1.0)	—	—	1.25	1.17 ± 0.26)	[8,49,50]
Acyclovir	−1.56	(−3.084)	2.4; 3.5	—	0.85	—	(1.0)	—	—	0.52	0.69 ± 0.19	[8,51]
Alfentanil	2.16	(1.007)	6.5	0.158	—	(1.0)	—	1.43	0.41–1.0	—	—	[8,52,53]
Alprenolol	3.34	(0.072)	9.70	—	0.24	—	(1.0)	—	—	3.04	3.37 ± 1.24	[8,49,54]
Amsacrine	2.85	(1.511)	7.43	—	0.0298	—	(1.0)	—	—	2.24	1.56	[8,55,56]
Atropine	1.83	(−1.482)	9.57	—	0.89	—	(1.0)	—	—	1.56	2.90	[8,57,58]
Caffeine	−0.07	−1.08	10.4	—	0.80	—	1.0	—	—	0.53	0.73 ± 0.02	[8,28,59]

Table 2. (*Continued*)

Drugs	Log $P_{o:w}$[a]	Log $D^{*}_{vo:w}$[b]	pK_a[c]	fu_p[d] Rat	fu_p[d] Human	$B{:}P$[e] Rat	$B{:}P$[e] Human	Volume of Distribution at Steady State (V_{ss}) (L/kg)[f] Rat Predicted	Rat Experimental	Human Predicted	Human Experimetnal	References
Carvedilol	4.19	(2.28)	8.40	—	0.0054	—	0.71	—	—	2.68	1.54 ± 0.11	[8,60,61]
Chlorpheniramine	3.38	(0.689)	9.13	—	0.27	—	1.34	—	—	3.26	3.17	[1,8]
Cimetidine	0.40	(−1.001)	6.80	—	0.80	—	(1.0)	—	—	0.56	1.0	[8,62]
Clonazepam	2.41	(1.337)	2.59	—	0.146	—	(1.0)	—	—	2.07	2.88	[4,8]
Cocaine	2.30	(−0.021)	8.61	0.63	—	1.0	—	2.23	2.8–3.3	—	—	[8,63,64]
Diazepam	2.99	2.07	3.5	0.137	0.022	0.38	0.58	4.10	4.50 ± 0.6	2.40	0.90–1.20	[65–68]
Diltiazem	2.84	(1.34)	7.7	0.25	0.22	(1.0)	1.0	3.46	3.60	3.09	2.91	[69–71]
Diphenhydramine	3.27	(0.685)	(9.0)	—	0.016	—	(1.0)	—	—	2.45	2.98	[4,8]
Epiroprim	2.89	1.70	6.96	0.097	0.11	1.0	1.0	3.16	4.63 ± 0.50	2.81	2.62 ± 0.36	[8]
Etomidate	3.05	(2.050)	4.5	0.191	0.235	(1.0)	(1.0)	4.70	4.03 ± 0.24	4.55	2.5 ± 1.1	[8,72–75]
Fentanyl	4.05	(1.565)	8.99	—	0.156	—	(1.0)	—	—	3.51	2.27–4.72	[8,76–79]
Gentamicin	−1.88	(−4.310)	8.20	—	0.73	—	(1.0)	—	—	0.49	0.45 ± 0.13	[8,80–82]
Ketanserin	3.29	(1.986)	7.46	—	0.05	—	(1.0)	—	—	2.88	3.7 ± 0.9	[8,83,84]
Lidocaine	2.44	(0.665)	8.01	0.38	0.296	1.27	0.90	2.37	2.13–3.11	2.18	1.08–1.51	[8,85–87]
Lorazepam	2.39	(−2.285)	11.0	—	0.107	—	(1.0)	—	—	1.69	1.60 ± 0.55	[8,88,89]
Lorcainide	4.85	(1.954)	(9.5)	—	0.15	—	0.77	—	—	4.0	6.4 ± 2.4	[8,90]
Mefloquine (S^+)	4.49	2.56	9.1	0.024	—	(1.0)	—	4.62	7.50	—	—	[8]
Meperidine	2.72	(0.428)	8.63	—	0.357	—	(1.0)	—	—	2.73	4.17	[4,8,91]
Mepivacaine	1.95	(0.307)	7.76	—	0.225	—	0.9	—	—	1.22	1.10	[8,92,93]
Meptazinol	2.70	(−1.310)	(10.37)	—	0.729	—	(1.0)	—	—	3.30	2.36 ± 0.81	[94,95]
Methadone	3.93	1.15	8.94	0.254	0.127	(1.0)	(1.0)	5.23	6.55	3.10	4.20 ± 0.78	[8,28,96–98]
Mibefradil	3.86	(2.953)	4.80	—	0.005	—	(1.0)	—	—	2.70	3.04	[99,100]
Midazolam	2.95	(1.928)	5.82	0.027	0.019	0.55	0.55	2.91	2.54 ± 1.54	2.31	1.41 ± 0.39	[101]
Morphine	0.89	(−1.230)	8.21	—	0.76	—	(1.0)	—	—	0.64	0.92–2.12	[4,8,102]
Nicotine	1.17	−0.41	3.04; 7.8	0.84	—	0.80	—	0.80	0.98	—	—	[4,28,103–105]
Nitrazepam	2.23	1.43	3.4	—	0.123	—	1.0	—	—	1.77	1.90–2.14	[65,106]
Oxazepam	2.24	(−2.453)	2.35	0.15	0.035	(1.0)	(1.0)	1.59	1.32–4.59	1.42	0.59–1.31	[8,107–110]
Pentazocine	3.31	0.828	8.5	0.54	0.389	1.55	1.06	5.50	7.66	3.61	3.48–5.56	[1,65]
Phencyclidine	4.96	(2.176)	9.4	0.47	—	1.0	—	9.91	9.6–15.5	—	—	[8,111–114]
Prilocaine	2.11	(0.383)	7.9	—	0.715	—	(1.0)	—	—	2.12	3.70 ± 1.21	[8,115,116]
Propafenone	(3.37)	(−0.054)	(9.86)	0.01	—	(1.0)	—	3.52	4.90	—	—	[8,117,118]
Propranolol	3.20	(0.164)	9.45	—	0.099	—	(1.0)	—	—	2.61	2.79 ± 0.10	[8,49,119]
Quinidine	3.44	(1.297)	8.56	0.30	0.228	1.85	(1.0)	5.09	4.58 ± 0.80	3.45	3.50	[8,120,121]

Table 2. (*Continued*)

Drugs	Log $P_{o:w}$[a]	Log $D^{*}_{vo:w}$[b]	pK_a[c]	fu_p[d]		$B:P$[e]		Volume of Distribution at Steady State (V_{ss}) (L/kg)[f]				References
								Rat		Human		
				Rat	Human	Rat	Human	Predicted	Experimental	Predicted	Experimetnal	
Sematilide	(1.36)	(−2.127)	9.5; 7.6	—	0.96	—	1.09	—	—	0.98	0.74–0.77	[8,122,123]
Sildenafil	2.75	(1.665)	6.5	0.05	0.04	(1.0)	(1.0)	2.56	1.1–2.0	2.22	1.20	[124]
Sotalol	0.24	(−4.378)	8.3; 9.8	—	1.0	—	(1.0)	—	—	0.60	0.85	[4,8,49]
Sulpiride	0.57	(−2.443)	9.12	—	1.0	—	1.0	—	—	0.64	0.849 ± 0.116	[8,125]
Theophylline	−0.02	(−2.799)	8.81	0.40	0.58	(1.0)	(1.0)	0.47	0.38	0.47	0.54	[8,126]
Verapamil	3.79	(1.752)	(8.49)	0.05	—	0.85	—	4.27	2.99–5.81	—	—	[8,127]
Neutrals												
2-Butanol	0.61	(−0.67)	—	1.0	—	(1.0)	—	0.70	0.73	—	—	[8,128,129]
t-Butyl alcohol	0.35	0.27	—	1.0	—	(1.0)	—	0.79	0.71–1.29	—	—	[8,128,130]
Cloprednol	1.68	(0.523)	—	—	0.16	—	(1.0)	—	—	0.88	0.81 ± 0.14	[8,131]
Coumarin	1.39	(0.20)	—	—	0.853	—	(1.0)	—	—	1.08	1.29	[4,8]
Cyclophosphamide	0.63	(−0.648)	—	—	0.90	—	(1.0)	—	—	0.64	0.78	[4,8]
Cyclosporin	2.92	(1.906)	—	0.0602	0.0727	1.283	1.36	3.14	3.43 ± 0.56	2.77	1.70–2.08	[8,132–135]
Cytosine arabinoside	−2.51	−4.0	—	—	0.98	—	(1.0)	—	—	0.55	0.44–0.86	[8,28,136,137]
Digitoxin	1.85	(0.713)	—	—	0.041	—	(1.0)	—	—	0.92	0.78 ± 0.02	[8,138,139]
Digoxin	1.26	(0.055)	—	0.613	—	(1.0)	—	0.77	1.27	—	—	[8,140]
Enprofylline	0.33	(−0.922)	—	0.226	0.489	(1.0)	(1.0)	0.43	0.34	0.47	0.51	[8,141]
Ethanol	−0.31	−1.34	—	1.0	1.0	(1.0)	(1.0)	0.64	0.78–0.785	0.57	0.54 ± 0.05	[8,28,128,142]
Ethoxybenzamide	0.77	(−0.491)	—	0.50	—	(1.0)	—	0.56	0.56	—	—	[8,143]
Ethylene glycol	−1.36	(−2.866)	—	1.0	—	(1.0)	—	0.63	0.3–0.4	—	—	[8,128,144]
Etretinate	(7.97)	(7.537)	—	0.01	—	(1.0)	—	4.65	3.07	—	—	[8,26,27]
Griseofulvin	2.18	(1.081)	—	—	0.20	—	(1.0)	—	—	1.72	1.40	[4,8]
Hydrocortisone	1.61	(0.445)	—	—	0.075	—	(1.0)	—	—	0.73	0.91	[4,8]
Methylprednisolone	1.82	(0.679)	—	—	0.232	—	(1.0)	—	—	1.13	1.38 ± 0.25	[8,145]
Prednisolone	1.62	(0.456)	—	0.60	0.444	(1.0)	(1.0)	1.07	1.37 ± 0.21	1.07	0.84–0.95	[8,145,146]
Propofol	3.79	(2.876)	—	—	0.022	—	1.25	—	—	3.04	5.82 ± 2.94	[8,147]
Quercetin	(1.48)	(0.30)	—	—	0.02	—	(1.0)	—	—	0.60	1.13	[4,8]
Remoxipride	2.10	(0.992)	—	0.74	0.20	1.20	0.70	2.30	4.5 ± 0.60	1.53	0.7 ± 0.1	[8,148]
Thiopeta	0.53	(−0.759)	—	—	0.90	—	(1.0)	—	—	0.62	0.70 ± 0.11	[8,149,150]
Toluene	2.65	2.78	—	—	1.0	—	(1.0)	—	—	20.1	15.4 ± 4.4	[8,128,151,152]

Table 2. (*Continued*)

Values of log $P_{o:w}$ (octanol:buffer PCs of nonionized species at pH 7.4), log $D^{*}_{vo:w}$ (olive oil:buffer PCs of both nonionized and ionized species at pH 7.4 or 7.56), pK_a (dissociation constant), fu_p (unbound fraction in plasma), and *B:P* (blood:plasma ratio). This table contains 89 drugs for which the predicted V_{ss}'s are within a factor of two of the corresponding experimental V_{ss}'s. A homogeneous distribution was assumed in predicting V_{ss}.

[a]Mean experimental data on log $P_{o:w}$ obtained from the cited literature except for diltiazem, mibefradil, midazolam, and mefloquine (Hoffmann-La Roche). However, for etretinate, propafenone, quecertin, and sematilide, data on log $P_{o:w}$ were calculated using the fragment constant method of Meylan and Howard[8] in the absence of experimental data (values in brackets).

[b]Mean experimental data on log $D^{*}_{vo:w}$ obtained from the cited literature except for diazepam, epiroprim, mefloquine, nitrazepam, and pentazocine (Hoffmann-La Roche). For toluene, its log $D^{*}_{vo:w}$ was obtained by dividing the oil:air PC with saline:air PC. However, for drugs for which experimental data on log $D^{*}_{vo:w}$ were not available in the literature or at Roche, they were calculated from data on log $P_{o:w}$ and pK_a according to eqs. 7–12 as described in the Methods section (values in brackets).

[c]Mean experimental data on pK_a obtained from the cited literature except for diltiazem, epiroprim, lorazepam, mefloquine, mibefradil, midazolam, and oxazepam (Hoffmann-La Roche). However, for acitretin, diphenhydramine, lorcainide, meptazinol, propafenone, and verapamil the data were calculated using the method of ISIS Base™, version 2.2 in the absence of experimental data (values in brackets).

[d]Mean experimental data on fu_p *in vitro* obtained from the cited literature except for epiroprim (rat and human data), mefloquine (rat data), and midazolam (rat data) (Hoffmann-La Roche).

[e]Mean experimental data on *B:P in vitro* obtained from the cited literature except for epiroprim (rat and human data) and midazolam (rat and human data) (Hoffmann-La Roche). However, for drugs for which experimental data on *B:P* were not available in the literature or at Roche, a default value of 1 was presented in the table as explained in the Methods section (values in brackets).

[f]Data predicted from eqs. 1–3. Experimental data on V_{ss} *in vivo* obtained from the cited literature except for epiroprim (rat and human data), mefloquine (rat data), and midazolam (rat data) (Hoffmann-La Roche). The experimental V_{ss} data represent either the means alone, or the means ± SD (single study), or the reported ranges (several studies). Experimental *in vivo* V_{ss} data were obtained for rat and human of approximately 0.250 kg and 70 kg, respectively.

—, Not used or calculated.

information.[6] In fact, fu_t was set equal to $1/(1+\{[(1-fu_p)/fu_p]\times 0.5\})$. The rationale of the mechanism-based estimation of fu_t from fu_p was provided in Poulin and Theil.[6]

Values of the Physicochemical Input Parameters

$P_{o:w}$ (*n*-octanol:buffer PC of nonionized species) used in eq. 2 was experimentally determined *in vitro* with conventional shake-flask methods or was calculated with a fragment constant method. The experimental or calculated $P_{o:w}$ data of several drugs are now available in a database named KowWin.[8] This database was used in the present study to get data on $P_{o:w}$ of most drugs. For some drugs, however, data on $P_{o:w}$ are not available in the database. Therefore, $P_{o:w}$ was obtained from other literature sources including Roche data. The values of log $P_{o:w}$ of all drugs and their references are presented in Tables 2–4.

$P_{o:w}$ is a parameter commonly determined for drugs. The availability of experimental data on $P_{o:w}$ explains why it was used in eq. 2 to estimate drug partitioning between neutral lipids and water of nonadipose tissues. Furthermore, the neutral nonpolar lipids of nonadipose tissues are mainly mixtures of triglycerides and cholesterol. The *n*-octanol may mimic the lipophilicity of such mixtures. The adipose tissue lipids, however, are mostly composed of triglycerides. In this case, it was demonstrated that *n*-octanol is not a good surrogate of triglycerides.[7] Alternatively, olive oil was suggested as an adequate surrogate of triglycerides.[7] Therefore, $D^{*}_{vo:w}$ (olive oil:buffer PCs of nonionized and ionized species) was used in eq. 3 to estimate drug partitioning between lipids and water of adipose tissue. The values of log $D^{*}_{vo:w}$ determined *in vitro* with conventional shake-flask methods were obtained from the literature for some drugs (Tables 2–3). For several drugs, however, an experimental value of log $D^{*}_{vo:w}$ is not available in the literature. Alternatively, the value of log $D^{*}_{vo:w}$ was estimated from data on log $P_{o:w}$ as follows. It is conventionally considered that the ionized species is soluble only in the buffer phase, but the nonionized species is soluble in both the aqueous and the lipophilic phases.[9] As reported previously, $D^{*}_{vo:w}$ takes into account the nonionized and ionized species in the aqueous phase compared to $P_{o:w}$ that considers only the nonionized species. In reporting $D^{*}_{vo:w}$ on a basis of nonionized species only in the aqueous phase to get $D_{vo:w}$, a linear regression analysis can be obtained between

Table 3. Values of Log $P_{o:w}$, Log $D^{*}_{vo:w}$, pK_a, fu_p, and *B:P*, As Well As Predicted and Experimental *In Vivo* Values of V_{ss} in Adult Male rats and Humans

								Volume of Distribution at Steady State (V_{ss}) (L/kg)[c]				
				fu_p[a]		*B:P*[b]		Rat		Human		
Drugs	Log $P_{o:w}$[a]	Log $D^{*}_{vo:w}$[a]	pK_a[a]	Rat	Human	Rat	Human	Predicted	Experimental	Predicted	Experimental	Reference
Acids												
Barbital	0.65	−0.59	7.75	0.964	—	1.0	—	0.70	2.31	—	—	[8,38,30]
Propylethyl-barbiturate	0.66	−0.14	7.77	0.86	—	1.0	—	0.69	1.58	—	—	[8,10,30]
Octylethyl-barbiturate	3.78	(2.713)	7.78	0.03	—	1.0	—	4.57	2.20	—	—	[8,30]
Ketoprofen	3.12	(−0.822)	4.45	0.029	—	(1.0)	—	3.17	1.19	—	—	[8,153]
Thiopental	2.85	1.80	7.45	0.131	—	0.922	—	3.34	0.69–1.35	—	—	[8,38,154,155]
Bases												
Alfentanil	2.16	(1.007)	6.5	—	0.074	—	(1.0)	—	—	1.38	0.226 ± 0.048	[8,52,156]
Amlodipine	3.00	(0.768)	8.6	0.06	0.02	(1.0)	(1.0)	3.03	32.0	2.28	21.0	[8,157,158]
Amiodarone	6.66	(4.348)	9.12	0.05	—	1.1	—	6.29	16.35 ± 0.71	—	—	[159,160]
Biperiden	4.25	(1.972)	8.8	0.135	—	1.16	—	5.41	14.0 ± 1.4	—	—	[8,65,161]
Bupivacaine	3.41	(1.623)	8.16	—	0.044	—	0.60	—	—	2.70	0.63–0.76	[8,92,93]
Chlorpromazine	5.19	(2.531)	9.3	0.106	0.043	(1.0)	1.56	6.18	29.1	3.4	8.8–11.2	[1,65,162,163]
Doxorubicin	1.27	(−0.798)	8.2	—	0.277	—	(1.0)	—	—	0.61	15.4 ± 3.8	[8,164,165]
Disopyramide	2.58	(−1.474)	10.4	0.235	0.325	(1.0)	(1.0)	2.36	0.63	2.35	0.78	[1,8,166]
Etidocaine	3.69	(2.288)	7.7	—	0.06	—	0.6	—	—	3.31	1.15	[8,92,93]
Exaprolol	3.68	(0.877)	9.27	0.54	—	1.25	—	6.15	59.4	—	—	[167]
Haloperidol	4.30	(2.161)	8.66	—	0.125	—	(1.0)	—	—	4.06	17.8 ± 6.5	[8,168,169]
Imipramine	4.80	1.93	9.5	0.24	0.145	1.67	(1.0)	6.45	19.9	3.93	14.2 ± 1.4	[1,8,28,170]
Procainamide	0.88	(−2.176)	9.2	0.92	—	(1.0)	—	0.71	1.77	—	—	[8,171]
Methylphenidate	0.20	−0.26	8.77	0.77	—	(1.0)	—	0.61	1.47	—	—	[8,172,173]
Ropivacaine	2.90	(1.129)	8.07	—	0.06	—	0.69	—	—	2.32	0.55 ± 0.07	[8,92,93,174]
Trimethoprim	0.91	(−0.799)	7.12	—	0.304	—	(1.0)	—	—	0.52	1.96	[4,8]
Neutrals												
Digoxin	1.26	(0.055)	—	—	0.78	—	(1.0)	—	—	0.90	6.9–7.3	[8,175]

Table 3. (*Continued*)

Drugs	Log $P_{o:w}$ [a]	Log $D^*_{vo:w}$ [a]	pK_a [a]	fu_p [a]		$B{:}P$ [b]	Volume of Distribution at Steady State (V_{ss}) (L/kg) [c]					Reference
							Rat		Human			
				Rat	Human	Human	Predicted	Experimental	Predicted	Experimental		
Felodipine	3.86	(2.954)	—	—	0.004	(1.0)	—	—	2.68	10.3 ± 3.0		[8,176]
Zwitterionics												
Ciprofloxacin	0.28	(0.986)	6.2; 8.6	—	0.52	(1.0)	—	—	0.47	1.74–1.90		[8,177–179]
Enoxacin	−0.20	(−1.654)	6.3; 8.3	—	0.485	(1.0)	—	—	0.44	1.96 ± 0.43		[8,177,180,181]

This table contains 25 drugs for which the predicted V_{ss}'s are greater by a factor of two than the corresponding experimental V_{ss}'s. A homogeneous distribution was assumed in predicting V_{ss}.

[a] Mean experimental data on log $P_{o:w}$, log $D^*_{vo:w}$, pK_a, and fu_p *in vitro* obtained from the cited literature. However, for drugs for which experimental data on log $D^*_{vo:w}$ were not available in the literature they were calculated from log $P_{o:w}$ and pK_a according to eqs. 7–12 as described in the Methods section (values in brackets).

[b] Mean experimental data on *B:P in vitro* obtained from the cited literature. However, for drugs for which experimental data on *B:P* were not available in the literature, a default value of 1 was presented as explained in the Methods section (values in brackets).

[c] Data predicted from eqs. 1–3. Experimental data on V_{ss} *in vivo* obtained from the cited literature. The experimental V_{ss} data represent either the means alone, the means ± SD (single study), or the ranges (several studies). Experimental *in vivo* V_{ss} data were obtained for rat and human of approximately 0.250 kg and 70 kg, respectively.

—, Not used or calculated.

experimental data on log $D_{vo:w}$ and log $P_{o:w}$ of several organic chemicals (acids, bases, neutrals):[10]

$$\log D_{vo:w} = 1.115 \times \log P_{o:w} - 1.35, \quad n = 104,\ r = 0.99 \tag{7}$$

For the prediction of $P_{t:p}$ of adipose tissue with eq. 3, data on $D^*_{vo:w}$ are required instead of $D_{vo:w}$.[7] Therefore, data on log $D^*_{vo:w}$ were obtained as follows from data on log $D_{vo:w}$ calculated with eq. 7. For this, it was assumed that $D^*_{vo:w}$ and $D_{vo:w}$ differ by a factor corresponding to the ionized species in the aqueous phase based on classical Henderson-Hasselbalch equations:

1. Monoprotic acid: $\log D^*_{vo:w} = \log D_{vo:w} - \mathrm{Log}\,(1 + 10^{\mathrm{pH} - \mathrm{p}K_a})$ (8)
2. Monoprotic base: $\log D^*_{vo:w} = \log D_{vo:w} - \mathrm{Log}\,(1 + 10^{\mathrm{p}K_a - \mathrm{pH}})$ (9)
3. Diprotic acid: $\log D^*_{vo:w} = \log D_{vo:w} - \mathrm{Log}\,(1 + 10^{\mathrm{pH} - \mathrm{p}K_a1 + \mathrm{pH} - \mathrm{p}K_a2})$ (10)
4. Diprotic base: $\log D^*_{vo:w} = \log D_{vo:w} - \mathrm{Log}\,(1 + 10^{\mathrm{p}K_a1 - \mathrm{pH} + \mathrm{p}K_a2 - \mathrm{pH}})$ (11)
5. Zwitterionic: $\log D^*_{vo:w} = \log D_{vo:w} - \mathrm{Log}\,(1 + 10^{-\mathrm{p}K_a2 + \mathrm{pH} + \mathrm{p}K_a1 - \mathrm{pH}})$, $\mathrm{p}K_a1 = \text{acid}$, $\mathrm{p}K_a2 = \text{base}$ (12)
6. Neutral compound (see eq. 7).

To estimate $D^*_{vo:w}$ with the above equations, additional data on pK_a are required. The experimental or calculated value of pK_a of each drug is presented in Tables 2–3, which includes also the corresponding literature sources. The calculated values of log $D^*_{vo:w}$ are presented in Tables 2–3. Note that data on $P_{o:w}$ and $D^*_{vo:w}$ expressed in a logarithmic form need to be transformed in the inverse log form before their use in eqs. 2 and 3 to predict $P_{t:p}$ of nonadipose and adipose tissues, respectively.

In the literature cited in the tables, data on log $P_{o:w}$ and log $D^*_{vo:w}$ were determined at a temperature of about 25 or 37°C in most cases. However, the known data on temperature dependence of log $P_{o:w}$ and log $D^*_{vo:w}$ as well as other solvent:buffer PCs indicate that the temperature dependence is relatively minor for several organic compounds (Δ log solvent:buffer PCs = 0.009/°C).[10–12] A temperature dependence of log $P_{o:w}$ and log $D^*_{vo:w}$, however, may be a potential source of error in

Table 4. Values of Log $P_{o:w}$, pK_a, and fu_p As Well As Predicted and Experimental *In Vivo* Values of V_{ss} (L/kg) in Adult Male Rats and Humans

			Rat Data[c]				
Drugs	Log $P_{o:w}$[a]	pK_a[b]	fu_p	*B:P*	Predicted V_{ss}	Experimental V_{ss}	References
Cefazolin	−0.58	2.33	0.272	(0.55)	0.16	0.208	[8,182]
Ceftazidime	−1.36	—	0.10	(0.55)	0.14	0.24	[183]
Ceftriaxone	−1.47	2.33	0.725	(0.55)	0.21	0.33 ± 0.19	[184]
Glycyrrhetinic acid	6.90	5.26	0.17	(0.55)	0.15	0.178 ± 0.068	[8,185]
Glycyrrhizin	2.80	2.6,2.6,5.3	0.05	(0.55)	0.14	0.184 ± 0.015	[8,186]
Interferon	—	—	1.0	(0.55)	0.23	0.26	[187]
Lamifiban	—	—	0.92	(0.55)	0.23	0.24	[188]
Nicotinic acid	0.36	4.75	0.55	(0.55)	0.19	0.289	[8,189]
Tolcapone	3.13	4.93	0.001	(0.55)	0.13	0.21	—

This table contains nine drugs for which tissue distribution is assumed to be restricted to the extracellular space based on the literature and for which the predicted V_{ss}'s are within a factor of two of the corresponding experimental V_{ss}'s.

[a]Mean experimental data on Log $P_{o:w}$ obtained from the cited literature except for ceftriaxone and tolcapone for which log $P_{o:w}$ was estimated using the fragment constant method of Meylan and Howard.[8] Data on log $P_{o:w}$ were not used for predicting V_{ss} of these types of drugs, which do not penetrate into cells (see the Methods section).

[b]Data on pK_a estimated with the method of ISIS Base™ except for nicotinic acid for which the experimentally determined pK_a was obtained from the literature.[8] Data on pK_a were not used for predicting V_{ss} of this type of drug, which does not penetrate into cells (see the Methods section).

[c]All data of tolcapone were obtained at Hoffmann-La Roche. The experimental data on *B:P in vitro* were not available in the literature. Therefore, a default value of 0.55 (1 – hematocrit value) was used for each drug that does not penetrate into cells (values in brackets). Data on fu_p represent only the means (single study), whereas data on V_{ss} represent the means alone or the means ± SD (single study). Data on V_{ss} were predicted from eqs. 1, 4, and 5 or were experimentally determined under *in vivo* conditions for rat and human of approximately 0.250 kg and 70 kg, respectively.

—, Data not available.

calculating $P_{t:p}$'s at 37°C. Nevertheless, PC data at 25 or 37°C were used in absence of these data at 37°C for all drugs. It is of interest to remember that the main purpose of the present study was only to verify if reasonable rough first estimates of V_{ss} *in vivo* could be obtained as required in early drug discovery. For the same reason, log $P_{o:w}$ and log $D^*_{vo:w}$ were obtained only at pH 7.4 or 7.56 considering that: these data are not available at other pHs based on the literature, the physiological pH of plasma and extracellular space of tissues is close to 7.4, the overall pH value of an intact tissue is currently unknown for *in vivo* conditions, and the change of *in vivo* $P_{t:p}$ with pH is also unknown.

Validation of the Mechanism-Based Method Used to Predict V_{ss}

The values of V_{ss} of 123 structurally unrelated drugs predicted based on eq. 1 were compared with corresponding *in vivo* values available in the literature (most cases) or obtained at Hoffmann-La Roche. The *in vivo* values of V_{ss} for all drugs and their corresponding literature sources are presented in Tables 2–4. Only V_{ss}'s (i.e., not an apparent V, $V\beta$, or Vz) estimated from plasma concentration-time profiles determined after intravenous administrations to rats and humans were considered for the purpose of the present study. For a few drugs, however, the values of V_{ss} are expressed on a basis of blood concentration. In this case, V_{ss} was multiplied with the blood: plasma ratio to get V_{ss} for plasma. As much as possible, values of V_{ss}'s were obtained for healthy adult male rats (≅ 250 g) and humans (≅ 70 kg). It was observed that V_{ss} is concentration-dependent for some drugs. In this context, see the Methods section concerning the estimation of fu_p.

The experimental or calculated values of V_{ss}, fu_p, *B:P*, $P_{o:w}$, or $D^*_{vo:w}$ were taken from several literature sources of the last 30 years. It is important to note that in collecting these values, no effort or judgment was made to differentiate experimental and analytical errors from true variability, or to classify the values according to the techniques used for their determination/estimation. Sensitivity and variability analyses are beyond the scope of the present study considering that the main objective was to verify

if the current methodology could provide rough first-cut estimates of V_{ss} prior to any *in vivo* experiments as required in drug discovery.

RESULTS

It is of interest to note that fitting exercises were omitted to improve the predictions. In fact, the current study presents only first predictions of V_{ss} by using one set of specified data as the input in eq. 1. The experimental values of V_{ss} *in vivo* obtained from the literature for 123 drugs were compared with the corresponding predicted values. For the 123 drugs investigated in the present study the overall average ratio of predicted-to-mean experimental rat and human V_{ss} values was 1.06 (SD = 0.817, $r = 0.78$, $n = 147$; Tables 2–4, Figure 1A). In fact, 80% of all predicted values were within a factor of two of the corresponding experimental values obtained from the literature. The drugs can therefore be separated into two groups.

First Group

The first group contains 98 drugs for which the predicted V_{ss}'s were within a factor of two of those experimentally determined (average ratio of 1.01, SD = 0.39, $r = 0.93$, $n = 118$; Tables 2 and 4, Figure 1B). These 98 drugs include 89 drugs for which a homogeneous tissue distribution by passive diffusion can be assumed (Table 2), and nine other drugs for which a distribution limited to the extracellular space can be assumed (Table 4). The current assumptions on tissue distribution considered for predicting V_{ss} are probably not violated for these 98 drugs, which include acids, bases, and neutral compounds.

Second Group

The second group contains 25 drugs (acids, bases, zwitterions) for which the predicted and experimental V_{ss}'s differ by a factor larger than two (average ratio of 1.32, SD = 1.74, $r = 0.42$, $n = 29$; Table 3, Figure 1C). It was assumed that these 25 drugs distribute homogeneously into tissues by passive diffusion. Thus, additional relevant distribution processes were probably neglected in predicting V_{ss} of these drugs. This was true especially in the case of some cationic-amphiphilic bases for which their V_{ss} was largely under predicted (Table 3).

DISCUSSION

Distribution and clearances are input parameters in empirical PK compartment models and physiologically based PK models (PBPK).[3,30] For efficient screening efforts with these models it is essential to obtain rough predictions of V_{ss} and clearances from compound-specific determinants routinely determined in early drug discovery. In the present study, a mechanism-based method for predicting rat and human V_{ss} under *in vivo* conditions has been developed and validated for 123 structurally unrelated drugs. The compound-specific determinants used for predicting V_{ss} with this method could be generated in drug discovery without conducting *in vivo* studies. During the drug development process it is evident that the quantity and quality of experimentally determined data on V_{ss} *in vivo* increase with time, which should permit comparison to the predicted V_{ss}. The present prediction method of V_{ss} built on a mechanism-based framework would permit the understanding of unexpected large deviations that may be observed between sets of predicted and experimental V_{ss} data. In general, a significant under-prediction of V_{ss} *in vivo* by the present method may suggest a drug accumulation into tissues due to additional specific binding and/or uptake processes, whereas an over-prediction of V_{ss} may suggest a limited permeation into cells of the peripheral compartment. Figure 1C presents large under- and over-predictions of V_{ss} especially for 25 drugs.

The drugs investigated in the present study have been classified into two groups. The first group represents 98 drugs for which the predicted and experimental V_{ss}'s differ by a factor of less than two (Tables 2 and 4), whereas the second group represents 25 drugs for which the predicted and experimental V_{ss}'s differ by a factor larger than two (Table 3). However, it appears that the current assumptions on tissue distribution used to predict V_{ss} are not violated especially for the first group of drugs. In other words, relevant additional distribution processes have probably been neglected in predicting V_{ss} of the second group of drugs. For the early drug discovery, V_{ss} was judged as accurately predicted solely for drugs of the first group. An obvious matter of concern with the present study is the uncertainty associated with each input parameter of the series of equations used to predict V_{ss}. The mean data of these parameters on tissue composition as well as drug lipophilicity and protein binding were

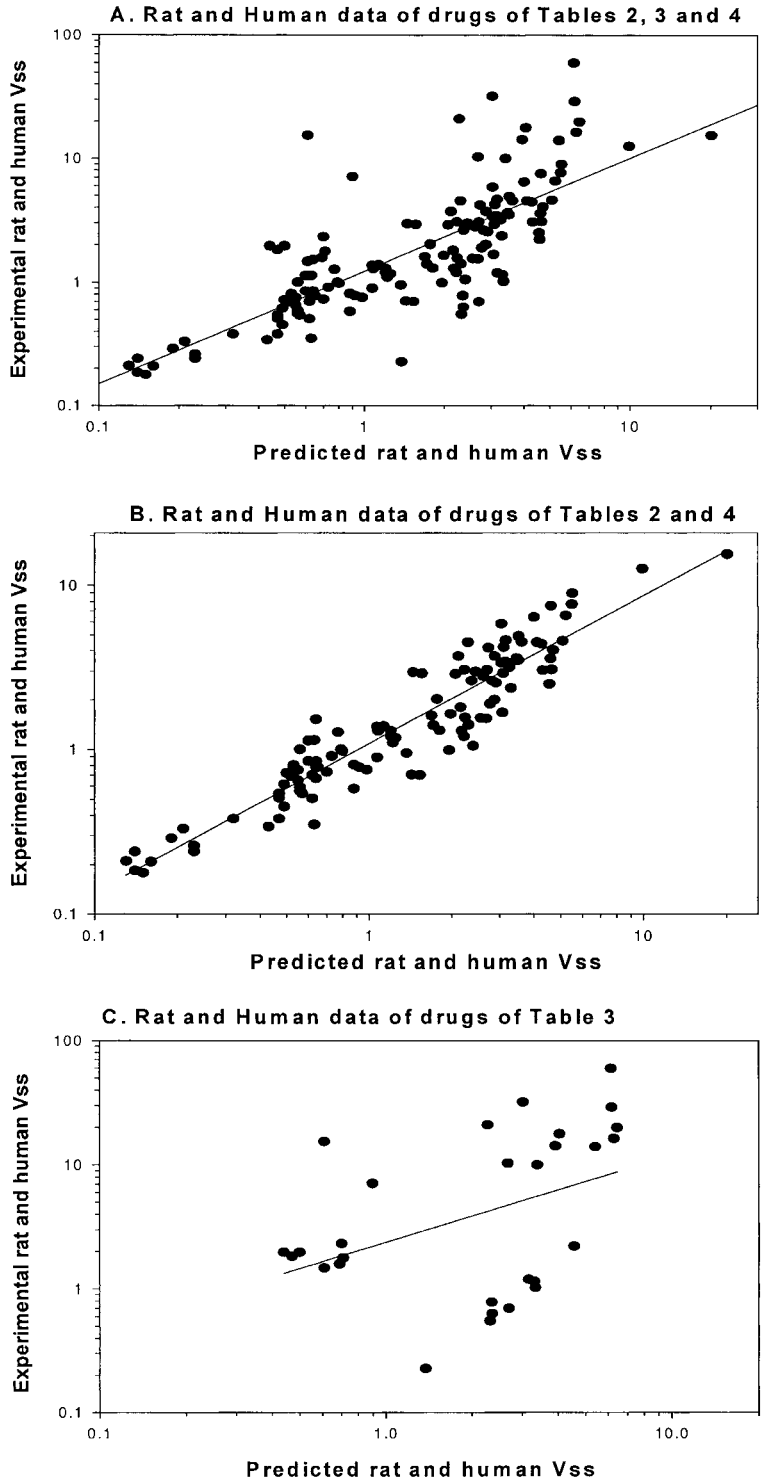

Figure 1. Relationship between predicted and experimentally determined V_{ss} of 123 drugs of Tables 2–4 (A), of 98 drugs of Tables 2 and 4 (B), and of 25 drugs of Table 3 (C). The line indicates the best fit for (A) [$\log y = 0.91383 \cdot \log x + 0.0892$, $r = 0.78$, $n = 147$, average ratio predicted-to-experimental $V_{ss} = 1.06 \pm 0.82$], (B) [$\log y = 0.90180 \cdot \log x + 0.03561$, $r = 0.93$, $n = 118$, average ratio predicted-to-experimental $V_{ss} = 1.01 \pm 0.39$], and (C) [$\log y = 0.70273 \cdot \log x + 0.37492$, $r = 0.42$, $n = 29$, average ratio predicted-to-experimental $V_{ss} = 1.32 \pm 1.74$]. Sigmaplot® version 4.0 was used for plotting.

obtained from various literature sources. Elaborated statistical and sensitivity exercises need to be done, but this was not necessary at this step because the main objective of the present study was only to develop a method to get first rough estimates of V_{ss} *in vivo* as required in drug discovery.

Predictions of V_{ss} *in vivo* within the given accuracy criteria were particularly obtained for the 89 drugs of Table 2, for which a homogeneous distribution into tissues mainly by passive diffusion can be assumed, and for the nine other drugs of Table 4, for which a tissue distribution limited to extracellular space can be assumed. An explanation for such accurate predictions for most drugs may be that muscle and fat are probably two main determinants of V_{ss} (based on their volume and/or lipid content) for which the $P_{t:p}$ of several drugs can reasonably be predicted with tissue composition-based equations.[6,7] Nevertheless, the results for these 98 drugs (Tables 2 and 4) suggest that V_{ss} is mainly governed by partitioning into total lipids and water (or partitioning into extracellular space only) and a reversible binding to common proteins present in plasma and interstitial space (i.e., albumin, globulins, lipoproteins as HDL and LDL). However, some drugs of this study (e.g., disopyramide, imipramine, methadone, mibefradil, propranolol, quinidine) can bind predominantly to other plasma proteins (e.g., alpha-glycoprotein).[13] The rat and human V_{ss}'s of these drugs are also accurately predicted except for disopyramide and imipramine (Tables 2 and 3; for imipramine, another explanation for the under-prediction of its V_{ss} is presented later in the Discussion section). Therefore, a binding to alpha-glycoprotein can potentially be accounted for in predicting V_{ss}. The rationale for that can be the ratio fu_p/fu_t of eq. 2 used to calculate the $P_{t:p}$'s. This ratio was estimated from data on tissue interstitial fluid-to-plasma concentration ratios of albumin, globulins, and lipoproteins[6]; similar ratios may also be found for alpha-glycoprotein. This cannot be confirmed in the absence of experimental data for alpha-glycoprotein.

Specific binding to cell macromolecules other than lipids has been neglected in predicting V_{ss}. Regarding the results of Table 2, it may be suggested that specific binding of drugs to cell macromolecules, if present, does not influence on a quantitative basis the prediction of V_{ss} for 89 drugs. This is also supported by the accurate predictions of rat and human V_{ss} of cyclosporin, quinidine, and methotrexate (Table 2) by neglecting a saturable specific binding to particular cell components reported for these drugs.[14,132] An explanation for this may be the much higher content of lipids and plasma proteins in tissues compared to cell macromolecules. Consequently, this may be a factor explaining that nonspecific binding to lipids and specific binding to plasma proteins (this study) might be the main "apparent" determinants of tissue distribution of most drugs. This needs to be verified, but the results in Table 2 suggest that specific binding to cell macromolecules can be neglected in predicting V_{ss} *in vivo* of several drugs. However, there are exceptions. For instance, for a drug binding extensively to DNA (e.g., doxorubicin),[15] V_{ss} was largely under-predicted (Table 3). In general, a significant specific binding to cell macromolecules may potentially explain unexpected under-predictions of V_{ss} *in vivo* observed for other drugs of Table 3. In this context, additional *in vitro* data on macromolecular binding are needed to predict V_{ss} accurately.

Ionic interactions between drugs and charged lipids of cell membranes as well as a pH gradient between intracellular and extracellular space have also been neglected in predicting V_{ss}. In fact, each tissue has been considered as a mixture of lipids, water, and plasma proteins having a global pH of 7.4. Only the hydrophobic interactions between drugs and tissue lipids have currently been considered for predicting V_{ss}. Consequently, the potential ionic interactions with charged lipids have been neglected. Regarding the predicted and experimental V_{ss}'s provided in Table 2, the present description of tissues is acceptable for several kinds of drugs. However, this appears not to be the case for drugs in Table 3, for which inaccurate predictions of their V_{ss}'s are observed. In fact, an under-prediction of V_{ss} by a factor of at least three is especially observed for some cationic-amphiphilic compounds (amiodarone, amlodipine, biperiden, chlorpromazine, exaprolol, haloperidol, imipramine; $\log P_{o:w} > 3$, $pK_a > 8.6$). This may be explained by the fact that ionic interactions between drugs and charged tissue lipids were neglected in predicting their V_{ss} as compared to hydrophobic interactions. As an example, amiodarone has an *in vivo* V_{ss} value three times larger than the one predicted (Table 3). This drug is extremely lipophilic, amphiphilic, and cationic, properties for which anomalous pharmacokinetic behaviors are reported.[16] Its *B:P* ratio is independent of drug

concentration, which is not the case for V_{ss}, suggesting that distribution into peripheral tissues accounts for the concentration-dependence of V_{ss}.[16] Therefore, it may be speculated that some lipid-rich inclusions of tissues (e.g., cells) act as deep traps considering the high membrane:buffer ratio of amiodarone probably caused by the presence of hydrophobic and ionic interactions.

For other cationic-amphiphilic bases [e.g., alprenolol, carvedilol, chlorpheniramine, fentanyl, lorcainide, mefloquine (S^+), methadone, pentazocine, phencyclidine, propafenone, propranolol, quinidine, verapamil; log $P_{o:w} > 3$, p$K_a > 8.4$], V_{ss} can be more reasonably predicted (Table 2). This is in clear contrast with other bases of Table 3, for which V_{ss}'s are largely under-predicted. It is then suspected that the ionic interactions with charged lipids of cell membranes (a nonspecific binding) are less relevant for bases of Table 2 compared to those of Table 3. This has been observed from preliminary *in vitro* studies (not shown). Therefore, this is now under investigation. If significant saturable nonspecific ionic binding is present in cell membranes of tissues for specific bases, V_{ss} can therefore be dependent on drug concentration, which can be an obvious matter of concern in predicting V_{ss} as a function of dose.

A presence of lysosomal ionic trapping in certain tissues (e.g., muscle, liver, lung)[6,17] may also be a potential source of explanation for the large under-prediction of V_{ss} in the case of cationic-amphiphilic bases of Table 3. The most pronounced effect of the lysosomal ionic trapping may represent 20 to 40% of the total tissue accumulation of a base such as thioridazine under *in vitro* conditions.[206] However, the lysosomes (where the pH is 4.7) represent only 1% of the total tissue volume under *in vivo* conditions.[65] Nevertheless, a significant lysosomal ionic trapping may potentially affect the first passage of a base through the lung, a tissue rich in lysosomes, particularly after intravenous bolus administrations. This may result in a slow release into the plasma leaving this tissue. In other words, the estimation of V_{ss} *in vivo* from data on plasma concentration-time profiles might be biased. Furthermore, a problem of drug solubility in the lung following the intravenous administration of an inadequate formulation may also result in a slow release into the plasma leaving this tissue. At present, however, mechanistic studies are still needed to explain why V_{ss} of cationic-amphiphilic bases can or cannot be accurately predicted with the present method (i.e., Table 2 vs. Table 3).

Enterohepatic recirculation and/or active uptake processes may lead to a contribution of the intestinal lumen to the distribution space. This would be significant particularly if the active uptake processes become important compared to the passive diffusion process. In this case, the *in vivo* V_{ss} would be under-predicted by the present approach, which does not consider any accumulation into the intestinal lumen. It is observed that V_{ss} is under-predicted for ciprofloxacin, enoxacin, and trimethoprim (Table 3), but is more reasonably predicted for cyclosporin, propranolol, and verapamil (Table 2). It is known that these drugs may accumulate into the intestinal lumen to different degrees. In another context, the prediction of V_{ss} is species variant especially for three drugs. For thiopental, its predicted and experimental V_{ss}'s differ by a factor of 3.27 and 1.44 in rat and human, respectively. For digoxin and alfentanil the predicted and experimental V_{ss} differ by a factor of 0.61 and 2 in the rat, respectively. In the human, the factor is 0.13 and 6.1, respectively (Tables 2 and 3). Definitive explanations cannot be provided. However, the disposition of digoxin is governed either by active transport, biliary clearance, and/or Na/K ATPase activity,[18,19,207] which may affect rat and human V_{ss} to different degrees.

In Figures 2 and 3 tridimensional mesh and contour plots are presented for the relationships between the calculated V_{ss} and the compound-specific determinants fu_p (ranging from 0 to 1) and log $P_{o:w}$ (ranging from -2 to 8). These theoretical exercises are mechanism-based relationships between potential determinants of rat and human V_{ss}. For a hypothetical monoprotic base of pK_a equal to 3 or 9, sigmoid relationships between V_{ss}, log $P_{o:w}$, and fu_p are observed. Similar types of relationships were also observed for monoprotic acids and neutral compounds (not shown). The resulting relationships demonstrate that V_{ss} of a drug is enhanced by increasing its lipophilicity, but is reduced by increasing its plasma protein binding. The relevance of ionization on V_{ss} becomes apparent when comparing a base with pK_a of 3 or 9. In other words, the upper profile of the tridimensional plots observed in Figures 2 and 3 is not exactly similar between a base of pK_a equal to 3 or 9 (Figures 2A1 and 3A1 vs. Figures 2A2 and 3A2). The rationale for that is the contribution of $P_{t:p}$ of adipose tissue in V_{ss}. In fact, $P_{t:p}$ of adipose tissue calculated with eq. 3 is

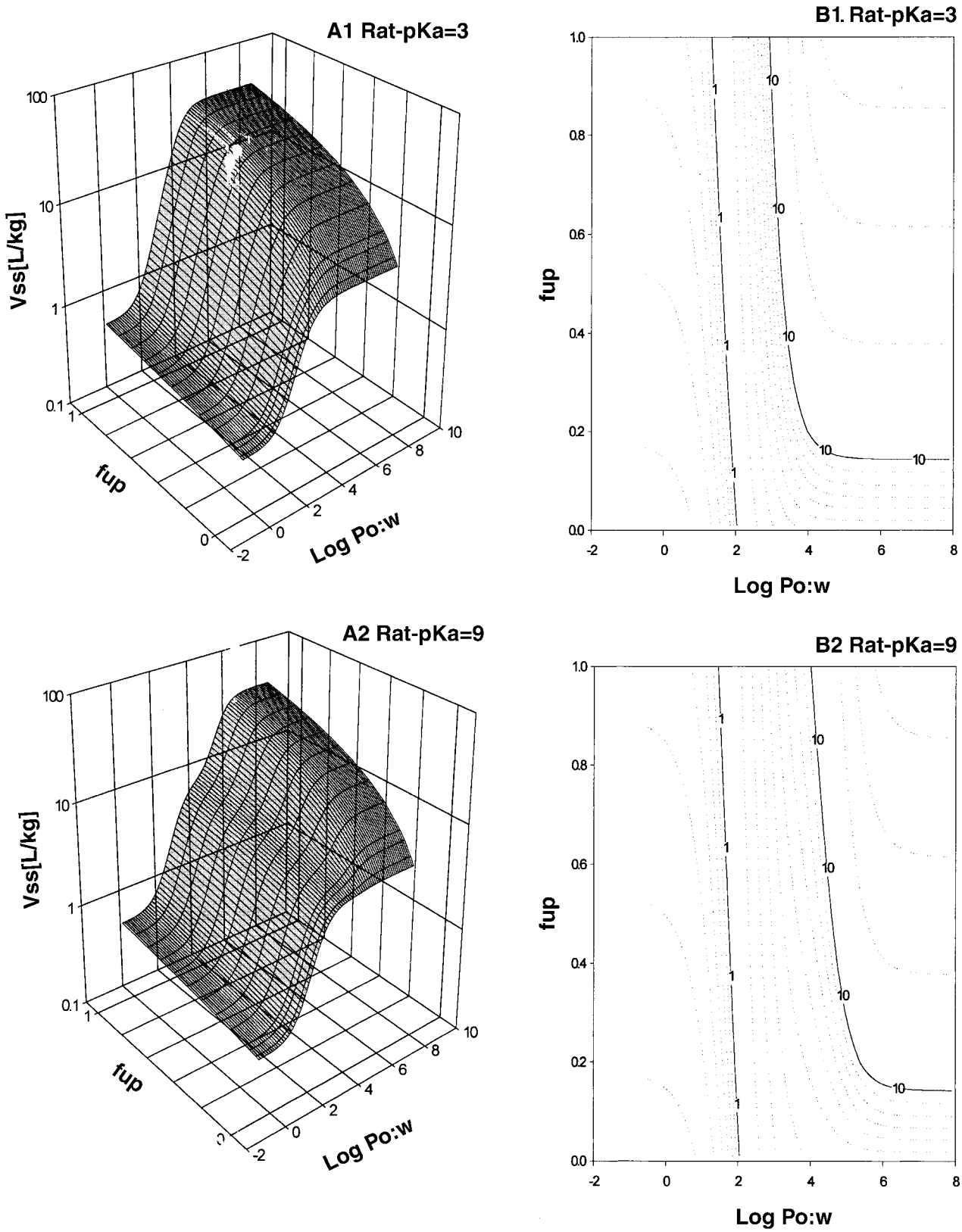

Figure 2. Tridimensional mesh (A) and contour (B) plots for the relationships between fu_p, log $P_{o:w}$, and rat V_{ss} calculated. The plots were performed for a hypothetical monoprotic base of pK_a equal to 3 (A1, B1) or 9 (A2, B2). The rat V_{ss} was calculated with eqs. 1–3 as demonstrated in the Methods section by using theoretical data on fu_p and log $P_{o:w}$ ranging from 0 to 1 and − 2 to 8, respectively. A value of blood:plasma ratio of 1 was used. The pK_a was used to calculate $D^*_{vo:w}$ of eq. 3 from data on $P_{o:w}$. A homogeneous tissue distribution was assumed. Sigmaplot® version 4.0 was used for plotting.

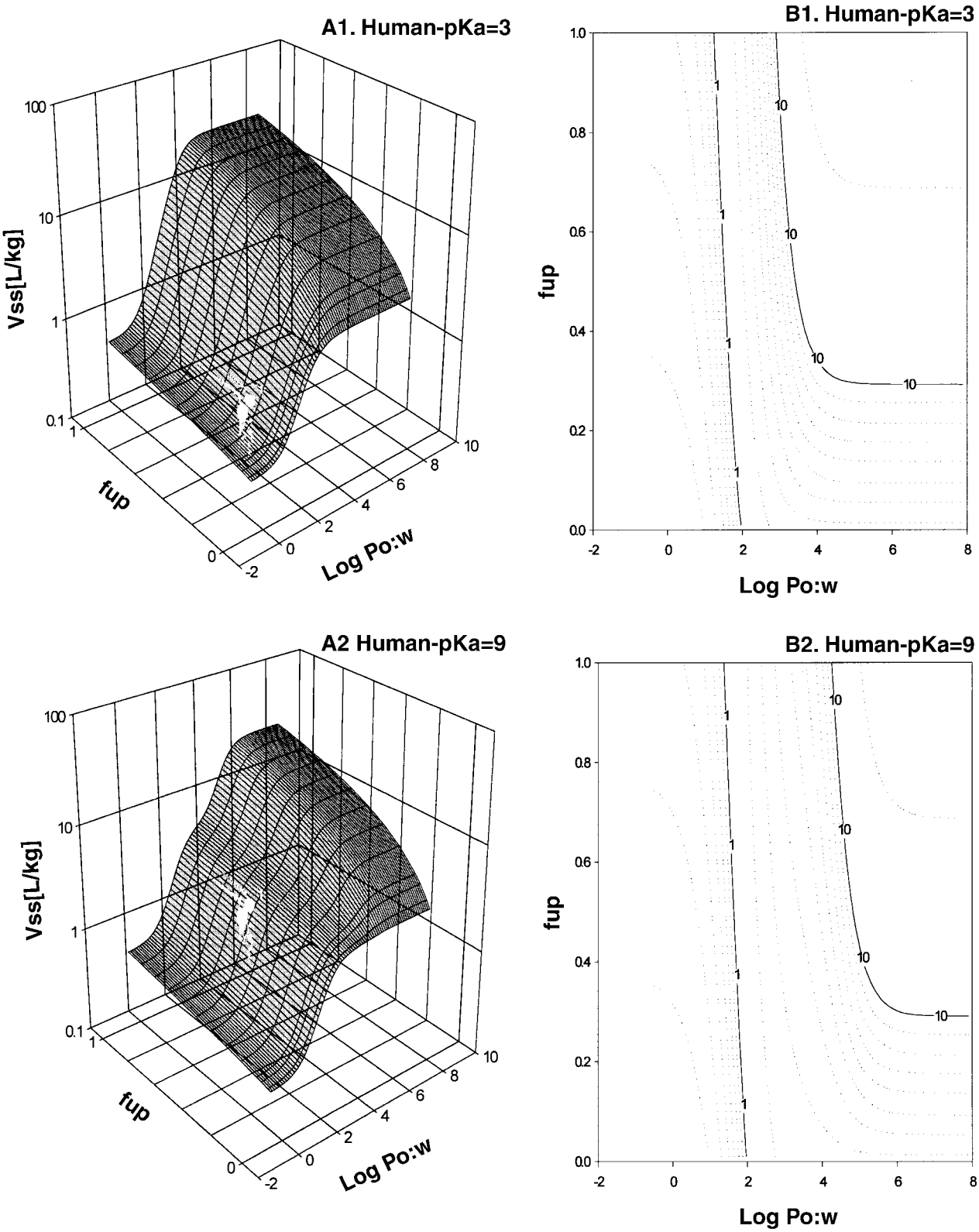

Figure 3. Tridimensional mesh (A) and contour (B) plots for the relationships between fu_p, log $P_{o:w}$, and human V_{ss} calculated. The plots were performed for a hypothetical monoprotic base of pK_a equal to 3 (A1, B1) or 9 (A2, B2). The human V_{ss} was calculated with eqs. 1–3 as demonstrated in the Methods section by using theoretical data on fu_p and log $P_{o:w}$ ranging from 0 to 1 and − 2 to 8, respectively. A value of blood:plasma ratio of 1 was used. The pK_a was used to calculate $D^*_{vo:w}$ of eq. 3 from data on $P_{o:w}$. A homogeneous tissue distribution was assumed. Sigmaplot® version 4.0 was used for plotting.

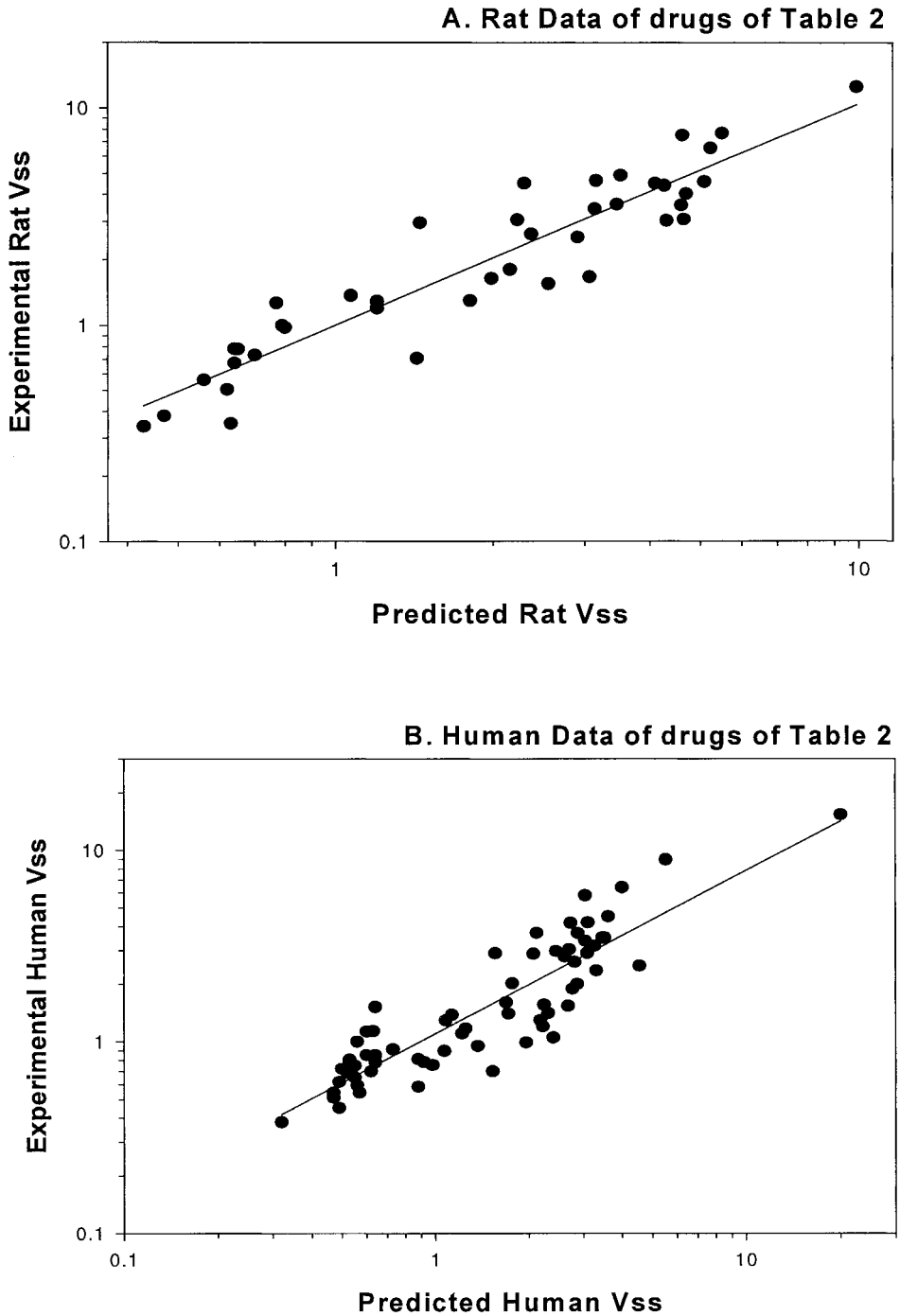

Figure 4. Relationships between the predicted and experimentally determined V_{ss} of rats (A) and humans (B) for the 89 drugs listed in Table 2. The line indicates the best fit for (A) [$\log y = 1.01895 \cdot \log x + 0.00101$, $r = 0.93$, $n = 41$, average ratio predicted-to-experimental $V_{ss} = 1.03 \pm 0.37$] and (B) [$\log y = 0.85347 \cdot \log x + 0.04118$, $r = 0.89$, $n = 68$, average ratio predicted-to-experimental $V_{ss} = 1.03 \pm 0.42$]. Sigmaplot® version 4.0 was used for plotting.

reduced when the input parameter $D^{*}_{vo:w}$ decreases by increasing pK_a.

Potential interspecies differences in V_{ss} are demonstrated in Figures 2 and 3. On a theoretical basis, the interspecies differences in V_{ss} between rat and human are differences in tissue lipid composition and fu_p for similar drug lipophilicity. Therefore, the minimal theoretical value of V_{ss} that can be calculated from eq. 1 is similar for rat and human ($\approx$0.31), which it is not the case for the maximal value that is higher in rat than in human ($\approx$46 vs. 28; Figures 2 and 3). For drugs listed in Table 2, the regression analyses comparing the calculated and experimental V_{ss} are similar for both species (Figure 4).

Eq. 1 can predict V_{ss}'s of drugs distributed homogeneously or nonhomogeneously (i.e., extracellular space only) into tissues. However, approaches are still needed to identify by which of these two scenarios a novel drug candidate will be distributed into tissues. An *in vitro* technique using tissue slices can potentially be used.[208] The drugs residing in the extracellular space are mainly acids that are highly hydrophilic or lipophilic as well as highly bound compounds (Table 4), properties that may reduce cell permeability. Eq. 1 calculates a maximal value of 0.31 L/kg for rat V_{ss} of drugs residing mainly in extracellular space, which corresponds to the minimal calculated value of V_{ss} of drugs distributing homogeneously into tissues. This calculated value of 0.31 is similar to the extracellular water space in the rat body.[205]

In conclusion, the present work suggests that V_{ss} *in vivo* of rat and human can be reasonably predicted prior to *in vivo* studies for most of the drugs investigated. However, the predicted and experimental V_{ss}'s may deviate by a factor larger than two (under- or over-predictions) for other drugs such as some cationic-amphiphilic bases, for which the V_{ss}'s are largely under-predicted. Additional relevant processes may have been neglected in the tissue composition-based equations explaining the less accurate predictions. These processes may refer to ionic interactions with charged lipids of cell membranes, specific macromolecular binding to cell components, active transport processes, or limitation for membrane permeation. These processes need to be described in tissue composition-based equations for a broader applicability. It is essential to adapt eq. 1 to predict V_{ss} according to age, sex, and disease state. Changing data on tissue volume and composition, as well as on fu_p, may do this. The present study is the first attempt to develop and validate a mechanistic distribution model for predicting rat and human V_{ss} *in vivo* of structurally unrelated drugs. Predictions of V_{ss} were made solely from compound-specific data determined *in vitro* ($P_{o:w}$, pK_a, fu_p), as well as tissue composition data obtained from the literature. Thus, the present study may open opportunities for a reduction of animal experiments.

ACKNOWLEDGMENTS

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APPENDIX

Estimation of Tissue Volume and Composition of Standard Adult Male Rats (250 g) and Humans (70 kg)

Tissue Volume Data (V_t) Reported in Table 1 for Adult Male Rats and Humans

The compilation of Brown et al.[190] reports the mean values of tissue volume (V) as a fraction of total body weight (L/kg). These authors provide data for adipose tissue (subcutaneous), skeleton + bone marrows, brain, gut, heart, kidney, liver, lung, muscle, skin, and spleen. In converting the values of tissue volume in kilograms to liters, the density was considered for adipose tissue (0.92 kg/L for rat and human) and skeleton + bone marrows (1.76 kg/L for rat and 1.67 kg/L for human).[190] For all the other tissues a density close to 1 was reported[190] or assumed. The overall density for skeleton + bone marrows was recalculated by multiplying the density of the main bone components (cortical, trabular, yellow, and red marrows) with their respective fractional volume of total bone.[190]

The arterial and venous blood volumes expressed as a fraction of total body weight (L/kg) were obtained from Igari et al.[191] in absence of these data in the compilation of Brown et al.[190] In calculating the volume of plasma and red blood cells, the rat and human blood were assumed to be constituted of 55% of plasma and 45% of erythrocytes (hematocrit) as classical values.

Tissue Composition Data (V_w, V_{nl}, V_{ph}) Reported in Table 1 for Adult Male Rats and Humans

The neutral lipids (nl) refer to the sum of triglycerides, diglycerides, monoglycerides, cholesterol,

and other types of nonpolar lipids. Phospholipids (ph) represent lipids that contain phosphoric acid esterified at one position of the glycerol molecule (e.g., phosphatidylcholine, phosphatidyl ethanolamine, phosphatidylserine, sphingomyelin).

The fractional volume content of total neutral lipids (V_{nl}) in each tissue was obtained from the difference between fractional volume content of total lipids and fractional volume content of total phospholipids (V_{ph}). V_{nl} and V_{ph} are reported on the basis of wet tissue weight. However, V_{nl} and V_{ph} reported on the basis of dry tissue weight were converted on the basis of wet tissue weight as follows: V_{nl} (V_{ph}) for wet tissue weight = V_{nl} (V_{ph}) for dry tissue weight $\times (1 - V_w)$, where V_w is the fractional volume content of water. The mean data on V_w in rat and human tissues were obtained from the compilation of Reinoso et al.,[192] whereas data on V_{nl} and V_{ph} in each rat and human tissue were obtained in the references listed as follows.

Here are the references for V_{nl} and V_{ph} of each tissue: adipose tissue (subcutaneous white fat) of rat and human[193]; bone + marrows of rat[194] and human[195]; whole brain of rat and human[196]; gut/small intestine of rat[197] and human[198]; heart of rat and human[199]; whole kidney of rat and human[200]; liver of rat[200] and human[198]; lung of rat and human[201]; skeletal muscle of rat[199] and human[202]; skin epidermis of rat[197] and human[203];spleen of rat and human[200]; plasma of rat[198] and human.[204]

In the literature cited above, the estimation of V_{nl} and V_{ph} was performed as detailed below. It was assumed that cortical and trabular components of bone contain only traces of lipids.[198] Therefore, V_{nl} and V_{ph} for the overall bone (skeleton) were recalculated by multiplying V_{nl} and V_{ph} of the marrows (yellow and red)[194,195] with their respective fractional volume content in total bone.[190] For the whole brain, V_{nl} and V_{ph} were recalculated from data on V_{nl} and V_{ph} determined for the gray and white matters[196] by assuming that each of these two components constitutes 50% of the brain. In calculating V_{nl} and V_{ph} for gut, heart, kidney, liver, lung, muscle, and spleen, a conventional factor of 25 was used to convert the level of phospholipid expressed in milligrams phosphorus to milligrams phospholipids.[198–201] Data on V_{nl} and V_{ph} for gut (small intestine) and skin (epidermis) represent those for a neonatal mouse[197] in the absence of data for adult rats. These data, however, were available in human adults.[203,204] The composition of human skin epidermis taken from reference 203 corresponds to the average values of V_{nl} and V_{ph} that have been determined with pieces of summer and winter skin of subjects of 25 to 35 years.

In references 199 and 200 data on fractional volume content of total lipid in muscle and liver were reported in both rat and human. However, these data for muscle (10%) and liver (14%) reported for an individual human appear elevated compared to other literature sources. This may be explained by the fact that the sample of human skeletal muscle used for the determination of total lipids may contain subcutaneous fat, whereas the sample of human liver may correspond to a sick and/or an old individual (but no information is provided). Therefore, values of V_{nl} and V_{ph} in the human skeletal muscle (psoas) and liver were calculated from other references, 198 and 202, respectively.

The values concerning the fractional volume content of extracellular (interstitial) space (V_{eis}) in all rat blood-free tissues were also obtained from the literature.[205]

REFERENCES

1. Sawada Y, Hanano M, Sugiyama Y, Harashima H, Iga T. 1984. Prediction of the volume of distribution of basic drugs in human based on data from animals. J Pharmacokinet Biopharm 12:587–596.
2. Lin JH, Sugiyama Y, Awazu S, Hanano M. 1982. *In vitro* and *in vivo* evaluation of the tissue-to-blood partition coefficients for physiological pharmacokinetic models. J Pharmacokinet Biopharm 10:637–646.
3. Gibaldi M, Perrier D. 1982. Pharmacokinetics. Second edition. Drugs and the pharmaceutical sciences, volume 15. New York: Marcel Dekker, 414 p.
4. Ritschel WA, Hammer GV. 1980. Prediction of the volume of distribution from *in vitro* data and use for estimating the absolute extent of absorption. Int J Clin Pharmacol Toxicol 18:298–316.
5. Ritschel WA, Kaul S. 1986. Prediction of apparent volume of distribution in obesity. Meth Find Exp Clin Pharmacol 8:239–247.
6. Poulin P, Theil FP. 2000. *A priori* prediction of tissue:plasma partition coefficients of drugs to facilitate the use of physiologically based pharmacokinetic models in drug discovery. J Pharm Sci 89:16–35.
7. Poulin P, Schoenlein K, Theil FP. 2001. Prediction of adipose tissue:plasma partition coefficients for structurally unrelated drugs. J Pharm Sci 90: 436–447.

8. Meylan WM, Howard PH. 2001. Log *n*-octanol:-water partition coefficients. KOWWIN database. Syracuse Research Corporation, Environmental Science Center, Syracuse, NY 13210.
9. Ueda O, Katsuji O, Kasumi A. 1982. True oil/water partition coefficients of procaine and lidocaine and estimation of their dissociation constants in organic solvents. Anesthesia Analgesia 61:56–61.
10. Leo A, Hansch C, Elkins D. 1971. Partition coefficients and their uses. Chem Rev 71:525–615.
11. Kaufman JJ, Koski WS, Benson DW, Semo NM. 1975/76. Narcotic and narcotic antagonist pKa's and partition coefficients and their significance in clinical practice. Drug Alcohol Dependence 1:103–114.
12. Christensen JH, Andreasen F, Jensen EB. 1983. The binding of thiopental to human serum albumin at variable pH and temperature. Acta Pharmacol Toxicol 52:364–370.
13. Tillement JP, Houin G, Zini R, Uruen S, Albengres E, Barré J, Lecomte M, Dathis P, Sebille B. 1984. The binding of drugs to blood plasma macromolecules: recent advances and therapeutic significance. Adv Drug Res 13:60–94.
14. Harashima H, Sawada Y, Sugiyama Y, Iga T, Hanano M. 1985. Analysis of nonlinear tissue distribution of quinidine in rats by physiologically based pharmacokinetics. J Pharmacokinet Biopharm 13:425–440.
15. Terasaki T, Iga T, Sugiyama Y, Hanano H. 1984. Pharmacokinetic study on the mechanism of tissue distribution of doxorubicin: interorgan and interspecies variation of tissue-to-plasma coefficients in rats, rabbits, and guinea pigs. J Pharm Sci 73:1359–1363.
16. Weiss M. 1999. The anomalous pharmacokinetics of amiodarone explained by nonexponential tissue trapping. J Pharmacokinet Biopharm 27:383–396.
17. Ishizaki J, Yokogawa K, Nakashima E, Ohkuma S, Ichimura F. 1998. Uptake of basis drugs into rat lung granule fraction *in vitro*. Biol Pharm Bull 21:858–861.
18. Okudara K, Sawada Y, Sugiyama Y, Iga T, Hanano M. 1988. Effects of basic drugs on the hepatic transport of cardiac glycosides in rats. Biochem Pharmacol 37:2949–2955.
19. Kitani K, Ohta M, Kanai S, Satp Y. 1985. Sex differences in the biliary excretion of digoxin and its metabolism in aging Wistar rats. Arch Gerontol Geriatr 4:1–12.
20. Nakayama T, Sawamoto T, Karino T, Matsumura M, Sasaki K, Kurosaki Y, Kimura T. 1997. Biopharmaceutical studies on drug/conjugated-metabolite interactions. III. Effect of acetaminophen sulfate and its positional isomers on the pharmacokinetics of acetaminophen in rats. Biol Pharm Bull 20:522–529.
21. Jang SH, Lee MH, Lee MG. 1994. Pharmacokinetics of acetaminophen after intravenous and oral administration to spontaneously hypertensive rats and normotensive Wistar rats. J Pharm Sci 83:810–814.
22. Gazzard BG, Ford-Hutchinson AW, Smith MJH, Williams R. 1973. The binding of paracetamol to plasma proteins of man and pig. J Pharm Pharmac 25:964–967.
23. Abernethy DR, Divoll M, Greenblatt DJ, Ameer B. 1982. Obesity, sex, and acetaminophen disposition. Clin Pharmacol Ther 31:783–790.
24. Sweeney KR, Chapron DJ, Antal EJ, Kramer PA. 1989. Differential effects of fluorbiprofen and aspirin on acetazolamide disposition in humans. Br J Clin Pharmacol 27:866–869.
25. Katritzky AR, Caster KC, Maren TH, Conroy CW, Bar-Ilan A. 1987. Synthesis and physicochemical properties of thiadiazol as candidates for topically effective carbonic anhydrase inhibitors. J Med Chem 30:2058–2062.
26. Urien S, Claudepierre P, Meyer J, Brandt R, Tillement JP. 1992. Comparative binding of etretinate and acitretin to plasma proteins and erythrocytes. Biochem Pharmacol 44:1891–1893.
27. Liu SS, Sandri R, Tang-liu DS. 1990. Systemic pharmacokinetics of acethylenic retinoids in rats. Drug Metab Dispos 18:1071–1077.
28. Oldendorf WH. 1974. Lipid solubility and drug penetration of the blood brain barrier. Proc Soc Exp Biol Med 147:813–816.
29. Zetler G, Seidel G, Siegers CP, Iven H. 1976. Pharmacokinetics of ascorbic acid in man. Eur J Clin Pharmacol 10:273–282.
30. Blakey GE, Nestorov IA, Arundel PA, Aarons LJ, Rowland M. 1997. Quantitative structure-pharmacokinetics relationships: I. Development of a whole-body physiologically based model to characterise changes in pharmacokinetics across a homologous series of barbiturates in the rat. J Pharmacokinet Biopharm 25:277–312.
31. Duchin KL, McKinstry DN, Cohen AI, Migdalof BH. 1988. Pharmacokinetics of captopril in healthy subjects and in patients with cardiovascular diseases. Clin Pharmacokin 14:241–259.
32. Hyo-Jeong KK, Wientjes MG, Au JLS. 1997. Physiologically based pharmacokinetic model of 2,3-dideoxyinosine. Pharm Res 14:337–344.
33. Akrawi SH, Wedlund PJ. 1989. Mephenytoin stereoselective elimination in the rat. I. Enantiomeric disposition following intravenous administration. Eur J Metab Pharmacokin 14:195–200.
34. Akrawi SH, Wedlund PJ. 1989. Mephenytoin stereoselective elimination in the rat. II. Comparison of mephenytoin stereoselective clearance during chronic intravenous and hepatic portal vein administration. Eur J Metab Pharmacokin 14:269–278.

35. Mellett LB. 1977. Physicochemical considerations and pharmacokinetic behaviour in delivery of drugs to the central nervous system. Cancer Treat Rep 61:527–531.
36. Cho MJ, Kurtz RR, Lewis C, Machkovech SM, Houser DJ. 1982. Metronidazole phosphate a water soluble prodrug for parental solutions of metronidazole. J Pharm Sci 71:410–414.
37. Lamp KC, Freeman CD, Klutman NE, Lacy MK. 1999. Pharmacokinetics and pharmacodynamics of the nitroimidazole antimicrobials. Clin Pharmacokinet 36:353–373.
38. Lerster CM, Burns JJ, Brand L, Campomanes CI, Trousof N, Papper EM, Brodie BB. 1958. The passage of thiobarbiturates and their oxygen analogs into brain. J Pharmacol Exp Ther 123: 70–73.
39. Jung D, Prasad PP. 1989. Influence of nutritional status on the pharmacokinetics and pharmacodynamics of pentobarbital. Drug Metab Dispos 17: 365–368.
40. Wilensky AJ, Friel PN, Levy RH, Comfort CP, Kaluzny SP. 1982. Kinetics of phenobarbital in normal subjects and epileptic patients. Eur J Clin Pharmacol 23:87–92.
41. Browne TR, Evans JE, Szabo GK, Evans BA, Greenblatt DJ. 1985. Studies with stable isotopes II: Phenobarbital pharmacokinetics during monotherapy. J Clin Pharmacol 25:51–58.
42. Arranz A, Betono SF, Echevarria C, Moreda JM, Cid A, Valentin JF. 1999. Voltammetric and spectophotometric techniques for the determination of the antihypertensive drug prazosin in urine and formulations. J Pharm Biomed Anal 21:797–807.
43. Jaillon P. 1980. Clinical Pharmacokinetics of prozacin. Clin Pharmacokinet 5:365–376.
44. Wahlqvist M, Nilson M, Sandberg F, Agurell S. 1970. Binding of Δ1-tetrahydrocannabinol to human plasma proteins. Biochem Pharmacol 19: 2579–2584.
45. Garrett ER, Hunt CA. 1974. Physicochemical properties, solubility, and protein binding of Δ^9-tetrahydrocannabinol. J Pharm Sci 63:1056–1064.
46. Hunt CA, Jones RT. 1980. Tolerance and disposition of tetrahydrocannabinol in man. J Pharmacol Exp Ther 215:35–44.
47. Wada DR, Bjorkman S, Ebling WF, Harashima H, Harapat SR, Stanski DR. 1997. Computer simulation of the effects of alterations in blood flows and body composition on thiopental pharmacokinetics in humans. Anesthesiol 87:884–899.
48. Nguyen KT, Stephens DP, McLeish MJ, Crankshaw DP, Morgan DJ. 1996. Pharmacokinetics of thiopental and pentobarbital enantiomeres after intravenous administration of racemic thiopental. Anaesth Anal 83:552–558.
49. Hinderling PH, Schmidlin O, Seydel JK. 1984. Quantitative relationships between structure and pharmacokinetics of beta-adrenoreceptor blocking agents in man. J Pharmacokinet Biopharm 12: 263–287.
50. Meffin PJ, Winkle RA, Peters FA, Harrison DC. 1977. Acebutolol disposition after intravenous administration. Clin Pharmacol Ther 22:557–567.
51. Blum MR, Liao STH, Miranda P. 1982. Overview of acyclovir pharmacokinetic disposition in adults and children. J Am Med 73(1A):186–192.
52. Björkman S, Stanski DR, Verotta D, Harashima H. 1990. Comparative tissue concentration profiles of fentanyl and alfentanil in humans predicted from tissue/blood partition data obtained in rats. Anesthesiology 72:865–873.
53. Cox EH, Kuipers JA, Danhof M. 1998. Pharmacokinetic-pharmacodynamic modeling of the EEG effect of alfentanil in rats: assessment of rapid functional adaptation. Br J Clin Pharmacol 124: 1534–1540.
54. Alvan G, Lind M, Mellström B, von Bahr C. 1977. Importance of first-pass elimination for interindividual differences in steady state concentrations of the adrenergic β-receptor antagonist alprenolol. J Pharmacokinet Biopharm 5:193–204.
55. Paxton JW, Jurlina JL. 1986. Comparison of the pharmacokinetics and protein binding of the anticancer drug, amsacrine and a new analogue, N-5-dimethyl-9-((3-methoxy-4-methylsulfonylamino)-phenyl-amino)-4-acridinecarboxamide in rabbits. Cancer Chemother Pharmacol 16:253–256.
56. Paxton JW, Kim SN, Whitfield R. 1990. Pharmacokinetic and toxicity scaling of the antitumor agents amsacrine and CI-921, a new analogue, in mice, rats, rabbits, dogs and humans. Cancer Res 50:2692–2697.
57. Hinderling PH, Guendert-Remy U, Schmidlin O. 1985. Integrated pharmacokinetics and pharmacodynamics of atropine in healthy humans. I. Pharmacokinetics. J Pharm Sci 74:703–709.
58. Eckert M, Hinderling PH. 1981. Atropine: a sensitive gas chromatography-mass spectrometry assay and prepharmacokinetics studies. Agents Actions 11:520–531.
59. Bonati M, Latini R, Tognoni G, Young JF, Garattini S. 1984–5. Interspecies comparison of *in vivo* caffeine pharmacokinetics in man, monkey, rabbit, rat, and mouse. Drug Metab Rev 15:1355–1383.
60. Fujimaki M, Murakoshi Y, Hakusui H. 1990. Assay and disposition of carvedilol enantiomers in humans and monkeys: evidence of stereoselective presystemic metabolism. J Pharm Sci 79: 568–572.
61. Cubeddu LX, Fuenmayor N, Varin F, Villagra VG, Colindres RE, Powell JR. 1987. Clinical pharma-

cology of carvedilol in normal volunteers. Clin Pharmacol Ther 41:31–43.
62. Somogyi A, Gugler R. 1983. Clinical pharmacokinetics of cimetidine. Clin Pharmacokinet 8: 463–495.
63. Sukbuntherng J, Martin DK, Pak Y, Mayersohn M. 1996. Characterization of the properties of cocaine in blood: blood clearance, blood to plasma ratio, and plasma protein binding. J Pharm Sci 85:567–571.
64. Booze RM, Lehner AF, Wallace DR, Welch MA, Mactutus CF. 1997. Dose-response cocaine pharmacokinetics and metabolite profile following intravenous administration and arterial sampling in unanesthetized, freely moving male rats. Neurotoxicol Teratol 19:7–15.
65. Yokogawa K, Nakashima E, Ishizaki J, Maeda H, Nagano T, Ichimura F. 1990. Relationships in the structure-tissue distribution of basic drugs in the rabbit. Pharm Res 7:691–696.
66. Klotz U, Antonin KH, Bieck PR. 1976. Pharmacokinetics and plasma binding of diazepam in man, dog, rabbit, guinea pig and rat. J Pharmacol Exp Ther 199:67–73.
67. Klotz U, Avant GR, Hoyumpa A, Schenker S, Wilkinson GR. 1975. The effects of age and liver disease on the disposition and elimination of diazepam in adult man. J Clin Invest 55:347– 359.
68. Klotz U, Reichman IW. 1984. Pharmacokinetics and pharmacodynamic interaction study of diazepam and metoprolol. Eur J Clin Pharmacol 26:223–226.
69. Höglung P, Nilsson LG. 1988. Physiological disposition of intravenously administrated C-labeled diltiazem in healthy volunteers. Ther Drug Monitoring 10:401–409.
70. Echizen H, Eichelbaum M. 1986. Clinical pharmacokinetics of verapamil, nifedipine and diltiazem. Clin Pharmacokinet 11:425–449.
71. Lee YH, Lee MH, Shim CK. 1992. Decreased systemic clearance of diltiazem with increased hepatic metabolism in rats with uranyl nitrate-induced acute renal failure. Pharm Res 9:1599–1606.
72. Paepe P, Van Hoey G, Belpaire FM, nRosseel MT, Boon PA, Buylaert WA. 1999. Relationship between etomidate plasma concentration and EEG effect in the rat. Pharm Res 16:924–929.
73. Meuldermans WE, HeyKants JJ. 1976. The plasma protein binding and distribution of etomidate in dog, rat and human blood. Arch Int Pharmacodyn Ther 221:150–162.
74. Sfez M, Le-Mapihan Y, Levron JC, Gaillard JL, Rosemblatt JM, Le Moing JP. 1990. Comparison of the pharmacokinetics of etomidate in children and in adults. Ann Fr Anesth Reanim 9:127–131.
75. Levron JC, Assoune P. 1990. Pharmacokinetics of etomidate. Ann Fr Anesth Reanim 9:123–126.
76. Roy SD, Flynn GL. 1989. Solubility behaviour of narcotic analgesics in aqueous media: solubilities and dissociation constants of morphine, fentanyl, and sufetanil. Pharm Res 6:147–151.
77. Scott JC, Stanski DR. 1987. Decreased fentanyl and alfentanil dose requirements with age. A simultaneous pharmacokinetic and pharmacodynamic evaluation. J Pharmacol Exp Ther 240:159–166.
78. Singleton MA, Rosen JI, Fisher DM. 1988. Pharmacokinetics of fentanyl in the elderly. Br J Anaesth 60:619–622.
79. Meuldermans WE, Hurkmans RM, Heykants JJ. 1982. Plasma protein binding and distribution of fentanyl, sufentanil, alfentanil and lofentanil in blood. Arch Int Pharmacodyn Ther 257:4–19.
80. Lui EC, Bendayan R. 1998. Gentamicin uptake by LLCPK1 cells: effect of intracellular and extracellular pH changes. Can J Physiol Pharmacol 76:155–160.
81. Myers DR, DeFehr J, Bennet WM, Porter GA, Olsen GD. 1978. Gentamicin binding to serum and plasma proteins. Clin Pharmacol Ther 23:356–360.
82. Leskin OL, Longstreth JA, Smith CR, Lietman PS. 1983. Netilmicin and gentamicin multidose kinetics in normal subjects. Clin Pharmacol Ther 34:644–650.
83. Lebrec D, Hadengue A, Gaudin C, Levron JC, Fraitag B, Berthelot P, Benhamou JP. 1990. Pharmacokinetics of Ketanserin in patients with cirrhosis. Clin Pharmacokinet 19:161–166.
84. Barendreg JN, Van-Peer A, Van-der-Hoeven JG, Van-Oene JC, Tjandra YI. 1990. Ketanserin pharmacokinetics in patients with renal failure. Br J Clin Pharmacol 29:715–723.
85. Wing LMH, Miners JO, Birkett DJ, Phil D, Foenander T, Lillywhite K, Wanwimolruk S. 1984. Lidocaine disposition—sex differences and effects of cimetidine. Clin Pharmacol Ther 35: 695–701.
86. Bennett PN, Aarons LJ, Bending MR, Stiner JA, Rowland M. 1982. Pharmacokinetics of lidocaine and its deethylated metabolite: dose and time dependency studies in man. J Pharmacol Biopharm 10:265–281.
87. Shibasaki S, Kawamata Y, Ueno F, Koyama C, Itho H, Nishigaki R, Umemura K. 1988. Effects of cimetidine on lidocaine distribution in rats. J Pharmacobio-Dyn 11:785–793.
88. Camu F, Maes V, Van de Velde A, Sevens C. 1988. Lorazepam fast-dissolving drug formulation (FDDF) and intravenous administration as anaesthetic premedicants: a pharmacokinetic analysis. Eur J Anaesthesiol 5:261–268.
89. Divoll M, Greenblatt DJ. 1982. Effect of age and sex on lorazepam protein binding. J Pharm Phamacol 34:122–123.

90. Klotz U, Muller-Seydlitz P, Heimburg P. 1978. Pharmacokinetics of lorcainide in man: a new antiarrhythmic agent. Clin Pharmacokinet 3: 407–418.
91. Dunkerley R, Johnson R, Schenker S, Wilkinson GR. 1976. Gastric and biliary excretion of miperidine in man. Clin Pharmacol Ther 20:546–551.
92. Lee A, Fagan D, Lamont M, Tucker GT, Halldin M, Scott DB. 1989. Disposition kinetics of ropivacaine in humans. Anesth Analg 69:736–738.
93. Burm AGL. 1989. Clinical pharmacokinetics of epidural and spinal anaesthesia. Clin Pharmacokinet 16:283–311.
94. Franklin RA. 1988. The clinical pharmacokinetics and metabolism of the analgesic meptazinol. Xenobiotica 18:105–112.
95. Gepts E, Sonck WA, Camu F, Vercruysse A. 1987. Pharmacokinetics of intravenously meptazinol during general anesthesia in man. Eur J Anaesthesiol 4:35–43.
96. Nilsson MI, Grönblach L, Winderlöv E, Anggard E. 1983. Pharmacokinetics of methadone in methadone maintenance treatment: characterization of therapeutics failures. Eur J Clin Pharmacol 25:497–501.
97. Eap CB, Cuendet C, Baumann P. 1990. Binding of d-methadone, l-methadone, and dl-methadone to proteins in plasma of healthy volunteers: role of alpha-1-acid glycoprotein. Clin Pharmacol Ther 47:338–346.
98. Garrido MJ, Valle M, Calvo M, Troconiz IF. 1999. Altered plasma and brain disposition and pharmacodynamics of methadone in abstinent rats. J Pharmacol Exp Ther 288:179–187.
99. Welker HA, Wiltshire H, Bullingham R. 1998. Clinical pharmacokinetics of mibefradil. Clin Pharmacokin 35:405–423.
100. Welker HA. 1998. Single and multiple dose mibefradil pharmacokinetics in normal and hypertensive subjects. J Pharm Pharmacol 50:983–987.
101. Trouvin JH, Farinotti R, Haberer JP, Servin F, Chauvin M, Duvaldestin P. 1988. Pharmacokinetics of midazolam in anaesthetised cirrhotic patients. Br J Anaesth 60:762–767.
102. Owen JA, Sitar DS, Berger L, Brownell L, Duke PC, Mitenko PA. 1983. Age-related morphine kinetics. Clin Pharmacol Ther 34:364–368.
103. Nair MK, Chetty DJ, Ho H, Chien YW. 1997. Biomembrane permeation of nicotine: mechanistic studies with porcine mucosae and skin. J Pharm Sci 86:257–262.
104. Gabrielsson J, Bondesson U. 1988. Constant-rate infusion of nicotine and cotinine. I. A physiological pharmacokinetic analysis of the cotinine disposition, and effects on clearance and distribution in the rat. J Pharmcokinet Biopharm 15:583–599.
105. Hieda Y, Keyler DE, Vandevoort JT, Kane JT, Ross CA, Raphael DE, Niedbalas RS, Pentel PR. 1997. Active immunization alters the plasma nicotine concentration in rats. J Pharmacol Exp Ther 283:1076–1081.
106. Jochemsen R, Hogendoorn JJH, Dingemanse J, Hermans J, Boeijinga JK, Breimer DD. 1982. Pharmacokinetics and bioavailability of intravenous, oral, and rectal nitrazepam in humans. J Pharmacokinet Biopharm 10:231–245.
107. Dingemanse J, Sollie FAE, Breimer D, Danhof M. 1988. Pharmacokinetic modeling of the anticonvulsant response of oxazepam in rats using the pentylenetetrazol threshold concentration as pharmacodynamic measure. J Pharmacokin Biopharm 16:203–227.
108. Srivbastava K, Hatanaka T, Aiba T, Katayama K, Koizumi T. 1999. Single dose pharmacokinetics and pharmacodynamics of oxazepam in normal and renal dysfunction rats. Biol Pharm Bull 22:627–632.
109. Sonne J, Loft S, Dossing M, Vollmer-Larsen A, Olesen KL, Victor M, Andreasen F, Andreasen PB. 1988. Bioavailability and pharmacokinetics of oxazepam. Eur J Clin Pharmacol 35:385–389.
110. Grennblatt DJ. 1981. Clinical pharmacokinetics of oxazapam and lorazepam. Clin Pharmacokinet 6:89–105.
111. Valentine JL, Owens M. 1996. Antiphencyclidine monoclonal antibody therapy significantly changes phencyclidine concentrations in brain and other tissues in rats. J Pharmacol Exp Ther 278: 717–724.
112. Shelnutt SR, Gunnell M, Owens M. 1999. Sexual dimorphism in phencyclidine *in vitro* metabolism and pharmacokinetics in rats. J Pharmacol Exp Ther 290:1292–1298.
113. Valentine JL, Arnold LW, Owens M. 1994. Antiphencyclidine monoclonal fab fragments markedly decrease after phencyclidine pharmacokinetics in rats. J Pharmacol Exp Ther 269:1079–1085.
114. Amico D, Kline RP, Maayani S, Weinstein H, Kupersmith J. 1983. Effects of phencyclidine on cardiac action potential: pH dependence and structure-activity relationships. Eur J Pharmacol 88:283–290.
115. Van der Meer AD, Burm AG, Stienstra R, van Kleef JW, Vletter AA, Olieman W. 1999. Pharmacokinetics of prilocaine after intravenous administration in volunteers: enantioselectivity. Anesthesiol 90:988–992.
116. Lai PM, Roberts MS. 1998. Epidermal iontophoresis: II. Application of the ionic mobility-pore model to the transport of local anesthetics. Pharm Res 15:1579–1588.
117. Fernandez J, lligona L, Puigdemont A, Guitart R, Riu JL, Arbox M. 1991. Tissue distribution of

propafenone in the rat after intravenous administration. Eur J Drug Metab Pharmacokin 16:23–27.
118. Puigdemont A, Arboix M, Gaspari F, Bortolotti A, Bonati M. 1989. *In vitro* plasma protein binding of propafenone and protein profile in eight mammalian species. Res Commun Chem Path Pharmacol 64:435–440.
119. Bowman SL, Hudson SA, Simpson G, Munro JF, Clements JA. 1986. A comparison of the pharmacokinetics of propranolol in obese and normal volunteers. Br J Clin Pharmacol 21:528–532.
120. Watari N, Wakamatsu A, Kaneniwa N. 1989. Comparison of disposition parameters of quinidine and quinine in the rats. J Pharmacobio-Dyn 12:608–615.
121. Karbwang J, Davis TME, Looareesuwan S, Molunto P, Bunnag D, White NJ. 1993. A comparison of the pharmacokinetic and pharmacodynamic properties of quinine and quinidine in healthy Tahi males. Br J Clin Pharmacol 35:265–271.
122. Hinderling PH, Dilea C, Koziol T, Millington G. 1993. Comparative kinetics of sematilide in four species. Drug Metab Dispos 21:662–669.
123. Shi J, Ripley E, Gehr TWB, Sica DA, Dandekar KA, Hinderling PH. 1996. Pharmacokinetics of sematilide in renal failure. J Clin Pharmacol 36:131–143.
124. Walker DK, Ackland MJ, James GC, Muirhead GJ, Rance DJ, Wastall P, Wright PA. 1999. Pharmacokinetics and metabolism of sildenafil in mouse, rat, rabbit, dog and man. Xenobiotica 29: 297–310.
125. Brés J, Bressolle F. 1991. Pharmacokinetics of sulpiride in humans after intravenous and intramuscular administrations. J Pharm Sci 80:1119–1124.
126. Gascon AR, Calvo B, Hernandez RM, Dominguez-Gil A, Pedraz JL. 1994. Interspecies scaling of cimetidine-theophilline pharmacokinetic interaction: interspecies scaling in pharmacokinetic interactions. Pharm Res 11:945–950.
127. Manitpisitkul P, Chiou WL. 1993. Intravenous verapamil kinetics in rats: marked arteriovenous concentration difference and comparison with humans. Biopharm Drug Dispos 14:555–566.
128. Poulin P, Béliveau M, Krishnan K. 1999. Mechanistic animal-replacement approaches for predicting pharmacokinetics of organic chemicals. In: Salem H, Katz SA, editors. Toxicity assessments alternatives: methods, issues, opportunities. Totowa, NJ: Humana Press, Inc., p 115–139.
129. Dietz FK, Rodriguez-Giaxola M, Traiger GJ, Stella VJ, Himmelstein KJ. 1981. Pharmacokinetics of 2-butanol and its metabolites in the rat. J Pharmacokinet Biopharm 9:553–571.
130. Poet TS, Valentine JL, Borghoff SJ. 1997. Pharmacokinetics of tertiary butyl alcohol in male and female Fischer 344 rats. Toxicol Lett 92:179–186.
131. Legler UE. 1988. Pharmacokinetics of cloprednol in healthy volunteers. Int J Clin Pharmacol Ther Toxicol 26:173–175.
132. Bernareggi A, Rowland M. 1991. Physiologic modeling of cyclosporin kinetics in rat and man. J Phamacokinet Biopharm 19:21–50.
133. Tayar N, Mark AE, Vallat P, Brunne RM, Testa B, van-Gunsteren WF. 1993. Solvent-dependent conformation and hydrogen-bonding capacity of cyclosporin A: evidence from partition coefficients and molecular dynamics simulations. J Med Chem 36:3757–3764.
134. Lemaire M, Tillement JP. 1982. Role of lipoproteins and erythrocytes in the *in vitro* binding and distribution of cyclosporin A in the blood. J Pharm Pharmacol 34:715–718.
135. Ptachcinski RJ, Venkataramanan R, Burckart GJ, Gray JA, van Thiel DH, Sanghvi A, Rosenthal JT. 1987. Cyclosporin kinetics in healthy volunteers. J Clin Pharmacol 27:243–248.
136. Breithaupt H, Pralle H, Eckhardt T, von-Hattingberg M, Schick M, Loffler H. 1982. Clinical results and pharmacokinetics of high-dose cytosine arabinoside. Cancer 50:1248–1257.
137. Slevin ML, Johnston A, Wollard RC, Piall EM, Lister TA, Turner P. 1983. Relationship between protein binding and extravascular drug concentration of a water-soluble drug, cytosine arabinoside. J R Soc Med 76:365–368.
138. Viani A, Rizzo G, Carrai M, Pacifici GM. 1992. The effect of aging on plasma albumin and plasma protein binding of diazepam, salicylic acid and digitoxin on healthy subjects and patients with renal impairment. Br J Clin Pharmacol 33:299–304.
139. Ochs HR, Verburg-Ochs B, Holler M, Greenblatt DJ. 1985. Effect of ketanserin on the kinetics of digoxin and digitoxin. J Cardiovasc Pharmacol 7:205–207.
140. Evans RL, Owens SM, Ruch S, Kennedy RH, Seifen E. 1990. The effect of age on digoxin pharmacokinetics in Fisher-344 rats. Toxicol Appl Pharmacol 102:61–67.
141. Tsunekawa Y, Hasegawa T, Nadai M, Takagi K, Nabeshima T. 1992. Interspecies differences and scaling for the pharmacokinetics of xanthine derivatives. J Pharm Pharmacol 44:594–599.
142. Matsumoto H, Minowa Y, Nishitani Y, Fukui Y. 1999. An allometric model for predicting blood ethanol elimination in mammals. Biochem Pharmacol 57:219–223.
143. Lin JH, Hayashi M, Awazu S, Hanano M. 1978. Correlation between *in vitro* and *in vivo* drug metabolism rate: oxidation of ethoxybenzamide in rat. J Pharmacokinet Biopharm 6:327–337.

144. Frantz SW, Beskitt JL, Tallant MJ, Zourelias LA, Ballantyne B. 1996. Pharmacokinetics of ethylene glycol. III. Plasma disposition and metabolic fate after single increasing intravenous, peroral, or percutaneous doses in the male Sprague-Dawley rat. Xenobiotica 5:515–539.
145. Szefler SJ, Ebling WF, Georgitis JW, Jusko WJ. 1986. Methylprednisolone versus prednisolone pharmacokinetics in relation to dose in adults. Eur J Clin Pharmacol 30:323–329.
146. Gard V, Jusko WJ. 1994. Effects of indomethacin on the pharmacokinetics and pharmacodynamics of prednisolone in rats. J Pharm Sci 83:747–750.
147. Servin F, Desmonts JM, Haberer JP, Cockshott ID, Plummer GF, Farinotti R. 1988. Pharmacokinetics and protein binding of propofol in patients with cirrhosis. Anesthesiology 69:887–891.
148. Widman M, Nilsson LB, Bryske B, Lundström J. 1993. Disposition of remoxipride in different species. Arzneim-Forsch (Drug Res) 43:287–297.
149. Cohen BE, Egorin MJ, Kohlhepp EA, Aisner J, Gutierrez PL. 1987. Human plasma pharmacokinetics and urinary excretion of thiopeta and its metabolites. Cancer Treat Rep 70:859–864.
150. Hagen B, Nilsen OG. 1987. The binding of thiopeta in human serum and to isolated serum protein fractions. Cancer Chemother Pharmacol 20:319–323.
151. Pierce CH, Dills RL, Morgan MS, Nothstein GL, Shen DD, Kalman DA. 1996. Interindividual differences in $^{2}H_{8}$-Toluene toxicokinetics assessed by a semiempirical physiologically based model. Toxicol Appl Pharmacol 139:49–61.
152. Gargas ML, Burgess RJ, Voisard DE, Cason GH, Andersen ME. 1989. Partition coefficients of low molecular weight volatile chemicals in various tissues and liquids. Toxicol Appl Pharmacol 98: 87–99.
153. Satterwhite JH, Boudinot FD. 1992. Pharmacokinetics of ketoprofen in rats: effect of age and dose. Biopharm Drug Dispos 13:197–212.
154. Srivastava K, Hatanaka T, Katayama K, Koizumi T. 1998. Pharmacokinetics and pharmacodynamic consequences of thiopental in renal dysfunction rats: evaluation with electroencephalograph. Biol Pharm Bull 21:1327–1333.
155. Yu HY, Sawada Y, Sugiyama Y, Iga T, Hanano M. 1981. Effect of sulfadimethoxine on thiopental disposition and elimination in rats. J Pharm Sci 70:323–326.
156. Meuldermans W, Van Peter A, Hendrickx J, Woestenborghs R, Lauwers W, Heykants J, Bussche G, Van Craeyvelt H, Van Der Aa P. 1988. Alfentanil pharmacokinetics and metabolism in humans. Anesthesiology 69:527–534.
157. Stopher DA, Beresford AP, Macrae PV, Humphrey MJ. 1988. The metabolism and pharmacokinetics of amlodipine in humans and animals. J Cardiovas Pharmacol 12:S55–S59.
158. Kass RS, Arena JP. 1989. Influence of pH on calcium channel block by amlodipine, a charged dihydropyridine compound. Implications for location of the dihydropiridine receptor. J Gen Physiol 93:1109–1127.
159. Chatelin P, Laruel R. 1985. Amiodarone partitioning with phospholipid bilayers and erythrocyte membranes. J Pharm Sci 74:783–784.
160. Weir SJ, Ueda CT. 1986. Amiodarone pharmacokinetics. I. Acute dose-dependent disposition studies in rats. J Pharmacokinet Biopharm 14:601–613.
161. Yokogawa K, Nakashima E, Ichimura F. 1990. Effect of fat tissue volume on the distribution kinetics of biperiden as a function of age in rats. Drug Metab Dispos 18:258–263.
162. Yeung F, Hubbard JW, Korchinski ED, Midha KK. 1993. Pharmacokinetics of chlorpromazine and key metabolites. Eur J Clin Pharmacol 45: 563–569.
163. Curry SH. 1972. Relation between binding to plasma protein, apparent volume of distribution, and rate constants of disposition and elimination for chlorpromazine in three species. J Pharm Pharmacol 24:818–819.
164. Mross K, Mayer U, Hamm K, Burk K, Hossfeld DK. 1990. Pharmacokinetics and metabolism of iodo-doxorubicin and doxorubicin in humans. Eur J Clin Pharmacol 30:507–513.
165. Eksborg S, Ehrsson H, Ekqvist B. 1982. Protein binding of anthraquinone glycosides, with special reference to adriamycin. Cancer Chemother Pharmacol 10:7–10.
166. Lima JJ, Boudoulas H, Shields BJ. 1985. Stereoselective pharmacokinetics of disopyramide enantiomers in man. Drug Metab Dispos 13:572–577.
167. Trnovec T, Zemanek M, Faberova V, Bezek S, Durisova M, Ujhazy E, Tomcikova O. 1982. Disposition of exaprolol, a new beta-blocker, in rats. Drug Metab Dispos 10:547–550.
168. Holley FO, Magliozzi JR, Stanski DR, Lombrozo L, Hollister LE. 1983. Haloperidol kinetics after oral and intravenous doses. Clin Pharmacol Ther 33:477–484.
169. Rowell FJ, Hui SM. 1981. Total and free serum haloperidol levels in schizophrenic patients and the effect of age thioridazine and fatty acid on haloperidol-serum protein binding *in vitro*. Br J Clin Pharmacol 11:377–382.
170. Abernethy DR, Greenblatt DJ, Shader RI. 1985. Imipramine disposition in the elderly. J Pharmacol Exp Ther 232:183–188.
171. Gole DJ, Nagwekar JB. 1991. Effect of chronic ethanol ingestion on pharmacokinetics of procainamide in rats. J Pharm Sci 80:232–238.

172. Thai DL, Yurasits LN, Rudolph GR, Perel JM. 1999. Comparative pharmacokinetics and tissue distribution of the d-enantiomeres of *para*-substituted methylphenidate analogues. Drug Metab Dispos 27:645–650.
173. Patrick KS, Ellington KR, Breese GR. 1984. Distribution of methylphenidate and p-hydroxymethylphenidate in rats. J Pharmacol Exp Ther 231:61–65.
174. Niesel HC, Kaiser H, Eilingsfeld T. 1990. Ropivacaine—a new local anesthetic with specific properties. Reg Anaesth 13:54–56.
175. Hinderling PH, Hartmann D. 1991. Pharmacokinetics of digoxin and main metabolites/derivatives in healthy humans. Ther Drug Monit 13: 381–401.
176. Dunselman PH, Edgar B. 1991. Felodipine clinical pharmacokinetics. Clin Pharmacokinet 21:418–430.
177. Escribano E, Calpena AC, Garrigues TM, Freixas J, Domenech J, Moreno J. 1997. Structure-absorption relationships of a series of 6-fluoroquinolones. Antimicrobial Agents Chemother 41: 1196–2000.
178. Zhai S, Wei X, Parker BM, Kunze KL, Vestal RE. 1996. Relation between plasma and saliva concentrations of enoxacin, ciprofloxacin, and theophylline. Ther Drug Monit 18:666–671.
179. Drusano GL, Plaisance KI, Forrest A, Standiford HC. 1986. Dose ranging study and constant infusion evaluation of ciprofloxacin. Antimicrobial Agents Chemother 30:440–443.
180. Nix DE, Lebsack ME, Chapelsky M, Sedman AJ, Norman A. 1993. Effect of oral antacids on disposition of intravenous enoxacin. Antimicrobial Agents Chemother 37:775–777.
181. Barbosa J, Berges R, Sanz-Nebot V. 1998. Retention behaviour of quinolone derivatives in high-performance liquid chromatography. Effect of pH and evaluation of ionisation constants. J Chromatogr A 823:411–422.
182. Tsuji A, Terasaki T, Imaeda N, Nishide K, Tamai I. 1989. Age-related change in tissue-to-plasma partition coefficient of cefazolin for non-eliminating organs in the rat. J Pharm Sci 78:535–540.
183. Granero L, Jimenez J, Molina F, Peris J. 1998. Distribution of ceftazidime in rat tissues. Biopharm Drug Dispos 19:473–478.
184. Kovar A, Costa TD, Derendorf H. 1987. Comparison of plasma and free tissue levels of ceftriaxone in rats by microdyalisis. J Pharm Sci 86:52–56.
185. Kawakami J, Yamamura Y, Santa T, Kotaki H, Uchino K, Sawada Y, Iga T. 1993. Kinetic analysis of glycyrrhetic acid, an active metabolite of glycyrrhizin, in rats: role of enterohepatic circulation. J Pharm Sci 82:301–305.
186. Wang Z, Okamoto M, Kurosaki Y, Nakayama T, Kimura T. 1996. Pharmacokinetics of glycyrrhizin in rats with d-galactosamine-induced hepatic disease. Biol Pharm Bull 19:901–904.
187. Lavé T, Levet B, Schmitt-Hoffmann A, Morgenroth B, Richter W, Chou R. 1995. Interspecies scaling of interferon disposition and comparison of allometric scaling with concentration-time transformations. J Pharm Sci 84:1285–1290.
188. Lavé T, Saner A, Coassolo P, Brandt R, Schmitt-Hoffmann A, Chou R. 1996. Animal pharmacokinetics and interspecies scaling from animals to man of lamifiban, a new platelet aggregation inhibitor. J Pharm Pharmacol 48: 573–577.
189. Iwaki M, Ogiso T, Hayashi H, Tanino T, Benet LZ. 1996. Acute dose dependent disposition studies of nicotinic acid in rats. Drug Metab Dispos 24: 773–779.
190. Brown RP, Delp MD, Lindstedt SL, Rhomberg LR, Beliles RP. 1997. Physiological parameter values for physiologically based pharmacokinetic models. Tox Ind Health 13:407–484.
191. Igari Y, Sugiyama Y, Sawada Y, Iga T, Hanano M. 1983. Prediction of diazepam disposition in the rat and man by a physiologically based pharmacokinetic model. J Pharmacokinet Biopharm 11:577–593.
192. Reinoso RF, Telder BA, Rowland M. 1997. Tissue water content in rats measured by desiccation. J Pharmacol Toxicol Meth 38:87–92.
193. Behnke AR. 1964. Role of fat in gross body composition and configuration. In: Rodahl R, Issekutz B, editors. Fat as a tissue. New York: McGraw Hill Book Company, pp. 285–313.
194. Das SK, Scott MT, Adhikary PK. 1975. Effect of the nature and amount of dietary energy on lipid composition of rat bone marrow. Lipids 10:584–590.
195. Miller SPF, Zirzow GC, Doppelt SH, Brady RO, Barton NW. 1976. Analysis of the lipids of normal and gaucher bone marrow. J Lab Clin Med 127: 353–358.
196. Norton WT. 1981. Formation, structure, and biochemistry of myelin. In: Siegel GL, Albers W, Agranoff BW, Katzman R, editors. Basic Neurochemistry, 3rd ed. Boston: Little, Brown and Company, pp. 63–92 and 355–369.
197. Schoephoerster RT, Wertz PW, Madison KC, Downing DT. 1985. A survey of polar and nonpolar lipids of mouse organs. Comp Biochem Physiol 82b:229–232.
198. Long C. 1961. Biochemist's Handbook. London: E & FN Spon Ltd. 1192 pp.
199. Simon G, Rouser G. 1969. Species variations in phospholipids class distribution of organs: II. Heart and skeletal muscle. Lipids 4:607–614.

200. Simon G, Rouser G. 1969. Species variations in phospholipids class distribution of organs: I. Kidney, liver and spleen. Lipids 4:599–606.
201. Baxter CF, Simon G, Rouser G. 1969. Variations among vertebrates of lung phospholipids class composition. Lipids 4:243–244.
202. Fletcher R. 1973. Lipids of myocardium. Lipids 7:728–732.
203. Nieminen E, Koljonen LM, Kiistala U, Mustakallio KK. 1967. Quantitative analysis of epidermal lipids by thin-layer chromatography with special reference to seasonal and age variation. Acta Derm-Venereol 47:327–338.
204. Diem K, Lentner C. 1972. Ciba-Geigy Scientific Tables, 7th ed. Basel, Switzerland: Ciba-Geigy, Ltd. 203 pp.
205. Pierson RN, Price DC, Wang J, Jain RK. 1978. Extracellular water measurements: organ tracer kinetics of bromide and sucrose in rats and man. Am J Physiol 235:F254–F264.
206. Daniel WA, Wojcikowski J. 1999. The role of lysosome in the cellular distribution of thioridazine and potential drug interactions. Toxicol Appl Pharmacol 158:115–124.
207. Lyon X, Schutz Y, Buclin T, Jequier E, Deriaz O. 1995. Inhibition of Na/K ATPase by digoxin and its relation with energy expenditure and nutrient oxidation rate. Am J Physiol 268:E1051– 1056.
208. Ballard P, Leahy DE, Rowland M. 2000. Prediction of *in vitro* tissue distribution from *in vitro* data. 1. Experiments with markers of aqueous spaces. Pharm Res 17:660–663.