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Research Paper

Development and Validation of a Physiology-based Model for the Prediction of Oral Absorption in Monkeys

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Purpose. The development and validation of a physiology-based absorption model for orally administered drugs in monkeys is described.
Materials and Methods. Physiological parameters affecting intestinal transit and absorption of an orally administered drug in monkeys have been collected from the literature and implemented in a physiological model for passive absorption previously developed for rats and humans. Predicted fractions of dose absorbed have been compared to experimentally observed values for a set of $N = 37$ chemically diverse drugs. A sensitivity analysis was performed to assess the influence of various physiological model parameters on the predicted fraction dose absorbed.
Results. A Pearson's correlation coefficient of 0.94 (95% confidence interval: [0.88, 0.97]; $p < 0.0001$) between the predicted and observed fraction dose absorbed in monkeys was obtained for compounds undergoing non-solubility limited passive absorption ($N = 29$). The sensitivity analysis revealed that the predictions of fractions dose absorbed in monkeys are very sensitive with respect to inter-individual variations of the small intestinal transit time.
Conclusions. The model is well suited to predict the fraction dose absorbed of passively absorbed compounds after oral administration and to assess the influence of inter-individual physiological variability on oral absorption in monkeys.

KEY WORDS: absorption; modeling; monkey; PBPK; simulation.

INTRODUCTION

The usefulness of monkeys as animal models for drug research and development studies has been discussed in the literature (1–5). Monkeys are closer to humans in terms of evolutionary development then all other commonly used laboratory animals. On the other hand, ethical concerns and the fact that they are difficult to handle and to house in a laboratory environment limit more extensive use in drug research (4).

As a model for oral drug absorption, mainly macaques such as rhesus (*Macaca mulatta*) or cynomolgus (*Macaca fascicularis*) monkeys are used. Based on relatively few studies, there appears to be a high correlation between the fraction dose absorbed in monkeys and in humans (1,2,4).

The aim of this study was to extend a physiological gastro-intestinal flow and absorption model previously developed for rats (6) and humans (7) for use in monkeys. The physiological parameters affecting GI transit and absorption have been collected from the literature and then included into an existing software tool [PK-Sim®, Bayer Technology Services GmbH, Leverkusen, Germany (8,9)], which enables dynamic simulations of the absorption process. Secondly, the model has been validated based on a set of marketed drugs with known fraction dose absorbed (F_{abs}) in monkeys. A sensitivity analysis was then performed to quantify the impact of inter-individual variability in physiological parameters on the fraction dose absorbed. Further, the model results obtained for passively absorbed compounds were compared to the results of the previously developed models for rats (6) and humans (7).

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MATERIALS AND METHODS

Model Structure

The details of the mechanistic GI transit and absorption model used herein have been presented elsewhere (6,7). In short, the gastro-intestinal tract is described as a single tube with spatially varying properties such as pH-value, length, radius, and effective surface area. Transport of an orally administered compound through this tube is modelled by means of a transit function, which defines the temporal and spatial distribution of the drug in the GI tract using a Gaussian profile as a distribution function. After release from the stomach, the center of mass of the drug package moves across the small intestine, while its width oscillates, reflecting the peristaltic movements of the bowel. The

parameters of the transit function (center of mass and width as a function of time) have been determined from published recovery data of a non-absorbable marker administered to rats (10), and have been subsequently scaled to human dimensions by taking the inter-species differences in the length of the various segments of the GI tract as well as differences in the gastric emptying and intestinal transit times into account (7). Two compound specific model parameters are important to determine the rate and extent of oral absorption. These are the compound's intestinal permeability (P_{int}) and solubility (S_{int}). P_{int} is a measure of the compound's ability to cross the membranes of the enterocytes and to be absorbed into the portal vein, whereas S_{int} can limit the rate and extent of absorption, especially in the case of a low solubility and/or high doses (7). If the mass-solubility ratio exceeds a certain value, then the administered mass cannot be fully absorbed during the time of intestinal transit. This value is the critical mass-solubility ratio (7) and it defines the threshold for solubility-limited absorption. Under non-solubility limited conditions, the rate and extent of absorption are controlled by the intestinal permeability. In this case, the fraction dose absorbed is a sigmoidal function of the logarithm of the intestinal permeability coefficient (7). Both parameters are, in general, a function of pH in the case of ionisable compounds. A mechanistic equation (Eq. 1) has been previously derived on the basis of a data-set of 126 chemically diverse marketed drugs with known fraction dose absorbed in humans to calculate the passive intestinal permeability from the compound's lipophilicity (L) and size expressed by an effective molecular weight (MW_{eff}) (7,11),

$$P_{int}(MW_{eff}, L) = A\frac{MW_{eff}^{-\alpha-\beta}L}{MW_{eff}^{-\alpha} + B\,MW_{eff}^{-\beta}L} + C\frac{MW_{eff}^{-\gamma}}{D^{-\gamma} + MW_{eff}^{-\gamma}}\,[cm/s] \quad (1)$$

MW_{eff} includes an empirical correction for compounds which contain halogen atoms in order to obtain a better measure for the volume, which is, according to the Stokes–Einstein law, the parameter that drives a diffusion process. For every halogen atom, a certain amount is deduced from the real molecular weight (CL: −22, F: −17, Br: −62, I: −98) (7). As a lipophilicity measure, the membrane affinity MA, i.e. a partition coefficient between aqueous buffer and an immobilized lipid bilayer (12,13)] is preferred, but other more commonly used lipophilicity measures such as octanol/water partition coefficients (P) might also work, since there is a reasonable correlation between LogMA and LogP in the range of approximately $1 < LogP < 4$ (14). Equation (1) accounts for passive transcellular uptake by diffusion through the membrane (left term of the right hand side) including the influence of the unstirred water layer as well as paracellular transport (right term of the right hand side) via a sigmoidal relation (7,11). The parameter C can be interpreted as the permeability coefficient of pure paracellular transport, and D reflects a cut-off value for the effective molecular weight to allow for paracellular transport. The exponents α and β describe the mass-dependence of the diffusion coefficients in water and in the membrane of the epithelial cells, respectively. The parameters of the equation have been determined previously using an independent data set for humans (7). Their values are shown in Table I.

Table I. Parameters of Model Eq. 1 Taken from (7)

A	B	C	D	α	β	γ
7440	1.0×10^7	2.5×10^{-7}	202	0.60	4.395	16

Model Parameterization

To parameterize the absorption model for use in monkeys, a literature review of the monkey-specific physiological parameters that affect oral absorption such as dimensions of the GI tract, pH profile, and time scales for gastric emptying and intestinal transit has been performed.

Dimensions of the GI Tract

The gastric volume of a rhesus monkey with a BW of approximately 9 kg is around 100 ml (4). The length and diameter of the duodenum are reported to be 5 cm and 1.5 to 2 cm, respectively, and the length of the colon in the rhesus monkey is about 40 to 50 cm (4). In a study by Makita *et al.* (15), the intestinal dimensions of eight Japanese monkeys (*Macaca fuscata*), weighing between 4.5 and 10 kg, were determined. The average length of the small intestine in this cohort was 255 cm (autopsy data) and independent of the body weight. The average lengths of the caecum and colon plus rectum were 4 and 75 cm, respectively. This data suggests that the dimensions of the GI tract in monkeys are similar to the intestinal dimensions in humans and, thus, much larger in proportion to the size of the animal (4). With respect to the rate and extent of absorption, the morphology and density of the villi plays an important role, because they increase the effective surface area (A_{eff}) that is available for absorption (7). In monkeys, villi are present along the entire small intestine. Their shape has been described as cylindrical and finger-shaped (similar to the morphology in humans) in the jejunum and ileum, while they appear to be broader in the duodenum (16). Other sources report that the villi in monkeys are of a leaf-like shape (4,17). According to Grass *et al.* (18), the density of villi in cynomolgus jejunum is approximately 17.8 ± 1.8 mm^{-2} (mean ± s.d.), but it is unknown for other segments of the GI tract. This value, however, is in the range of the villi density reported for human small intestine [between 15 and 40 mm^{-2} (7,19)]. It was therefore assumed in the model that the surface enhancement due to the villi plus the microvilli is similar in monkeys and humans. To calculate A_{eff} in the monkey's small intestine, the same ratio A_{eff}/A_{cyl} was assumed in the duodenum, jejunum, and ileum as in humans (7). Here, $A_{cyl} = 2\pi r\ L$ is the cylindrical surface area of the intestine calculated from the length (L) and radius (r) of each segment.

Gastric and Intestinal pH

In the study of Ikegami *et al.* (1), the pH of gastric fluids ranged between 1.3 and 4.3 with a mean around 2–3 in fasted cynomolgus monkeys. Kondo *et al.* (20) observed pH vs. time profiles in fasted cynomolgus monkeys and reported median profiles between pH 1 and 3. A considerable inter- and intra-individual variability was noted in this study. Some of the

monkeys exhibited fasted gastric pH values up to 9 (20). In the fed state, the pH profiles were elevated depending on the type and amount of food. After consumption of 108 g of a biscuit-type meal, elevated gastric pH values with a median around 5–7 were observed, which lasted for about 9 h, while intake of sweet potatoes or bananas increased the gastric pH to 5 to 7 for approximately 4 h (21). In comparison, the variability of gastric pH values in humans in the fasted state is also very high (between 1.2 and 7.4), whereas the variability is low after consumption of a meal (22).

Little information is available regarding pH values in the small and large intestine. It is suggested that monkeys have comparatively high intestinal pH, between 7 and 9 (4).

Gastric Emptying (GET) and Small Intestinal Transit Time (SITT)

In monkeys, gastric emptying of liquids occurs rapidly in the fasted state and is delayed under fed conditions (1,21). In both cases, gastric emptying follows a mono-exponential kinetic pattern (21). The reported mean half-lives for gastric emptying are 58 min (fasted) and 148 min (fed) (21). In the study by Ikegami *et al.* (1) SITT ranged from 2.2 to 4.2 h in the fasted state and from 2.2 to 3.2 in the fed state. Kondo *et al.* (21) also reported oro-caecal transit times of liquids administered in fasted and fed cynomolgus monkeys. Although the measurement of the oro-caecal transit time includes the gastric emptying process, this time is a reasonable estimate for SITT, because the start of gastric emptying was confirmed at the first sampling point 6 min after administration of the liquid (21). The median values were 2.5 h (fasted) and 1.8 h (fed) (20,21). Again, there was a high inter-individual variability with reported ranges of 1.5–6 h (fasted) and 1–6 h (fed). Both studies concluded that small intestinal transit of liquids is not significantly different in the fasted and the fed state (1,21).

Based on the values obtained in the literature, the model parameters summarized in Table II have been used as reasonable estimates and implemented in the existing GI transit and absorption model (6,7). The transit function derived from rat data (10) was scaled to the intestinal dimensions in the monkey in the same way as described for humans in (7).

Model Validation

With respect to model validation, it is important to point out that this physiological absorption model for monkeys is *generic* in nature. The model only makes use of an empiric equation for the intestinal permeability derived previously on a different data set in humans in combination with physiological knowledge about the parameters in monkeys that affect gastro-intestinal transit and absorption to predict F_{abs} a priori. Contrary to statistical prediction models, this model is not trained based on experimental data. Its validity is therefore assessed on the basis of a comparison of a priori predictions for the fraction dose absorbed with data experimentally observed in monkeys. Chiou and Buehler (2) recently presented a comparison of F_{abs} and bioavailability between monkeys and humans for 43 (in case of F_{abs}) and 35 (in case of bioavailability) drugs. 36 compounds were selected from this study based on the availability of the lipophilicity input required for the calculation of the intestinal permeability coefficient (see Table III). Ciprofloxacin was also added to the list based on availability of monkey absorption data in (23) and *in house* physico-chemical data. For 15 compounds, the membrane affinity was either known from previous in house studies (7) or they were provided by Nimbus Biotechnology, Leipzig, Germany. For the remaining 21 compounds, either experimentally determined or, in the case of bromocriptine, lisuride, menogaril and valacylovir, calculated LogP values were collected from public databases (see references in Table III). The list also includes compounds with reported poor aqueous solubility. Such compounds can be expected to show incomplete absorption due to a solubility limitation. Since the doses at which the fractions absorbed have been measured in monkeys were not reported in (2), compounds with a solubility that could potentially limit the extent of oral absorption were considered separately. The threshold for a potential solubility limitation was set to 100 mg/l based on preliminary simulations results that revealed the onset of solubility limitation around this value for oral doses in the range of 500–1,000 mg in monkeys.

In addition, carrier mediated influx or efflux, as mentioned above, as well as presystemic elimination of the drug in the gut wall are not accounted for in Eq. 1. Therefore, it has to be expected that substrates for active transporters or

Table II. Physiological Parameters of the GI Tract of a Monkey Implemented as Default Values in the Model (for Details and References, See Text)

Parameter	Stomach	Duodenum	Jejunum	Ileum	Caecum	Total Small Intestine
Length [cm]	–	5	100	150	4,5	255
Radius [cm]	–	0,75	1	1	1	0.75–1
V_{cyl} [ml]	100	9	314	471	14	794
A_{cyl} [cm^2]	–	24	628	942	28	1,594
A_{eff} [cm^2]	–	7,400	249,000	175,500	28	431,900
pH (fasted)	2,5	8	8	8	8	8
pH (fed)	6	8	8	8	8	8
GET (fasted) [min]	60	–	–	–	–	–
GET (fed) [min]	150	–	–	–	–	–
SITT (fasted) [h]	–	–	–	–	–	2,5
SITT (fed) [h]	–	–	–	–	–	1,8

Table III. List of Compounds Used for Validation of the Monkey Absorption Model (for Details, See Text)

COMPOUND	Lipophilicity (Log units)	Eff. Molweight	Solubility [mg/l]	P_{int} [10^{-6} cm/s][n]	Dosage Form	F_{abs} (observed)	F_{abs} (predicted)
Atenolol*	1,00 (LogMA)[a]	266	685[a]	1,54	Sol.	45	55
Benazepril	3,22 (LogP)[b]	425	2,23[b]	12,6	Caps.	32	100
Bepiridil	5,15 (LogP)[b]	366	Slightly soluble[a]	21,5	Susp.	83	100
Bisoprolol	3,19 (LogP)[b]	325	2,240[b]	19,0	Sol.	100	100
Bromocriptine	4,51 (cLogP)[c]	593 (1 Br)	800	14,6	Sol.	35	100
Caffeine*	0,60 (LogMA)[a]	194	2,632[a]	2,57	Sol.	100	74
Captopril*	0,48 (LogMA)[a]	217	6,857[a]	1,22	Sol.	79	47
Carbamazepine*	2,52 (LogMA)[a]	236	17,7[a]	21,5	Sol.	100	100
Ceftriaxone*	0,90 (LogMA)[a]	512	958[a]	0,073	Sol.	0	5
Ciprofloxacin*	0,95 (LogMA)[a]	314 (1 Cl)	11,480[a]	0,68	Sol.	31	30
Coumarin*	1,51 (LogMA)[a]	146	1,900[a]	25,1	Sol.	87	100
Etidronate	−2,73 (LogP)[b]	206	High	0,11	Sol.	6	5
Flunisolide	2,90 (LogP)[b]	417 (1 F)	120[d]	9,47	Sol.	100	99
Fluvastatin*	3,10 (LogMA)[a]	395 (1 F)	0,47[a]	13,1	Caps.	100	100
Furosemide*	1,34 (LogMA)[a]	315 (1 Cl)	149[a]	1,59	Sol.	60	56
Guanabenz*	3,50 (LogMA)[a]	209 (2 Cl)	1,055[a]	29,7	Sol.	88	100
Irbesartan	5,38 (LogP)[b]	429	100 in water, higher in intestine[h]	19,5	Susp.	92	100
Latanoprost	4,03 (LogP)[b]	433	8,000[b]	17,8	Sol.	100	100
Lisuride	2,31 (cLogP)[e]	338	Highly soluble[f]	7,70	Sol.	100	98
Lovastatin	4,00 (LogP)[b]	405	0.4[b]	18,8	Sol.	31	100
Menogaril	3,18 (cLogP)[c]	542	90[g]	6,63	Sol.	63	97
Metoprolol*	1,59 (LogMA)[a]	267	4,777[a]	5,05	Sol.	92	93
Moxifloxacin	2,00 (LogMA)	316 (1 F)	High	5,79	Tabl.	82	95
Nadolol*	0,60 (LogMA)[a]	309	2,2400[a]	0,33	Sol.	23	16
Naltrexone	1,92 (LogP)[b]	341	10,0000[b]	3,80	Sol.	100	86
Nisoldipine*	4,90 (LogMA)[a]	388	25[a]	20,6	Susp.	97	100
Pindolol*	2,40 (LogMA)[a]	248	7,883[a]	18,3	Tabl.	100	100
Pirmenol	4,27 (LogP)[e]	375	Unknown	20,6	Sol.	98	100
Propranolol*	3,15 (LogMA)[a]	259	609[a]	24,1	Tabl.	100	100
Rifapentine	5,29 (LogP)[b]	877	Low	11,9	Susp.	100	100
Rolipram	1,09 (LogP)[b]	275	200[i]	1,63	Susp.	100	57
Ropinirole	2,72 (LogP)[b]	260	133,000[b]	20,7	Sol.	100	100
Tilodrunate	0,55 (LogP)[b]	296 (1 Cl)	Unknown	0,36	Sol.	15	17
Valacyclovir	−0,84 (cLogP)[c]	324	174,000[j]	0,10	Sol.	100	5
Viloxazine	1,45 (LogP)[k]	237	Unknown	6,03	Caps.	100	96
Zolpidem	3,32 (LogP)[b]	307	23,000[b]	21,2	Susp.	100	100
Zomepirac	1,81 (LogP)[m]	270 (1 Cl)	Low	7,16	Sol.	94	98

Compound that are marked with an asterisk have been used for the validation of the related model in humans (7).
[a] (7); [b] (28); [c] (36); [d] (37); [e] (38); [f] (39); [g] (40); [h] (41); [i] (42); [j] (43); [k] (44); [m] (45), [n] calculated from Eq. 1

compounds that undergo a significant metabolization in the gut wall are not correctly predicted by the model. The data set contains three compounds that are known substrates for intestinal transporters: Captopril (24) and valacyclovir (25,26) are actively taken up by a peptide carrier, and bromocriptine is a known substrate for p-glycoprotein (p-gp) (27). Several compounds are substrates for cytochrome P450 (CYP) 3A4 and therefore candidates for first pass elimination in the intestine after oral administration. Among them are benazepril (28), bromocriptine, fluvastatine, lovastatin, and nisoldipine (29).

Sensitivity Analysis

A number of the physiological parameters that affect oral absorption in monkeys show large inter-individual variability. Therefore, a local parameter sensitivity analysis was performed to quantify the changes in the fraction dose absorbed associated with variations in these parameters. The gastric emptying and small intestinal transit time in the fasted state as well as the effective surface area available for absorption have been logarithmically varied in independent simulations within a factor of two around their default values (see Table II) for three virtual compounds assumed to have low, medium, and high intestinal permeability. The intestinal permeability parameter was set according to resulting fraction dose absorbed values of 10, 50, and 90%, respectively, under non-solubility limited conditions. The parameters were varied independently, i.e., all other simulation parameters were kept constant at their default values. All simulations were carried out assuming fasted conditions.

Inter-species Comparison

To compare the F_{abs}-model predictions between different species, the fraction dose absorbed was calculated as a

function of the intestinal permeability coefficient using equation (1) in rats (6), humans (7), and monkeys. In all species, the species-specific default parameters for the physiological parameters of the gastro-intestinal tract were used. Therefore, the resulting predicted F_{abs} values represent average values for each species. Further, non solubility-limited, fasted state conditions were assumed and gut wall metabolism was neglected.

RESULTS

Figure 1 shows the correlation between the predicted and the observed F_{abs} values in monkeys. The Pearson's correlation coefficient for the total data set (N = 37) is 0.63 (95% confidence interval: [0.39, 0.79]; $p < 0.0001$). When only passively absorbed compounds with high solubility (closed circles in Fig. 1, N = 29) are considered, 27 out of the 29 are found within the 5 – 20% interval around the line of identity (dotted lines in Fig. 1). For those compounds, the Pearson's correlation coefficient between the predicted and observed fraction dose absorbed in monkeys is 0.94 (95% confidence interval: [0.88, 0.97]; $p < 0.0001$). Most compounds that are predicted outside of this range are either known to be actively transported either by an influx transporter (symbol X) or by an efflux transporter (symbol +), or have a low solubility that potentially limits the fraction dose absorbed *in vivo* (but not in the simulations, because the influence of solubility on absorption could not be accounted for due to a lack of knowledge of the administered dose, indicated by the open circles).

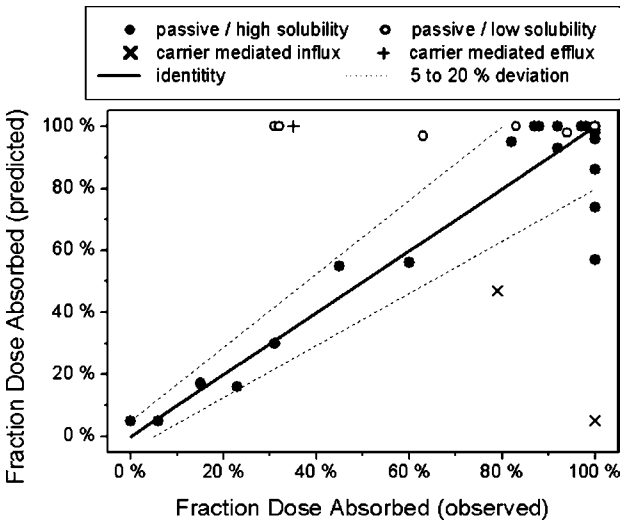

Fig. 1. Correlation of the predicted and the observed fraction dose absorbed in monkeys. *Closed circles* represent passively absorbed compounds with adequate solubility, *open circles* passively absorbed compounds with low aqueous solubility that potentially (depending on the administered dose) limits the extent of absorption. Compounds that are substrates for active transporters are marked as X (active influx) and + (active efflux). The *solid line* represents the line of identity, *dotted lines* indicate a deviation from the identity ranging from ±5% for poorly absorbed compounds to ±20% for completely absorbed compounds (7).

The results of the local sensitivity analysis are presented in Fig. 2. Variations of the small intestinal transit time have the largest effect on the predicted fractions dose absorbed, followed by variations of the effective surface area available for absorption. Changes of the gastric emptying time had only a moderate effect on the predicted fraction dose absorbed. The sensitivities are always largest for compounds with medium permeability and smallest for compounds with low permeability.

The inter-species comparison is shown in Fig. 3. According to the physiological model, monkeys and rats tend, on average, to underestimate the fraction dose absorbed in humans for passively absorbed compounds without solubility limitation and with negligible gut wall metabolism, but rat to a greater extent than monkey.

DISCUSSION

The physiological GI transit and absorption model previously developed for rats (6) and humans (7) has been extended for use in monkeys. The monkey model was set-up in the same way as the model for rats (6) and humans (7), but with monkey-specific values for the physiological input parameters. The same empirical equation for the intestinal permeability as a function of compound-specific input data [lipophilicity and molweight, Eq. 1] with parameters derived based on human data [Table I, (7)] was used. The application of this equation among different species assumes a comparable chemical composition of the membrane of the intestinal mucosa in terms of lipids, and a comparable size, density, and porosity of the tight junctions, which facilitate paracellular absorption.

Therefore, the physiological parameters that affect oral absorption were reviewed and implemented in the existing model structure. The literature review revealed, overall, a high inter-individual variability in the gastro-intestinal physiology in monkeys.

The correlation coefficient for the total data set was 0.63, indicating a reasonable correlation, besides a number of obvious outliers. A potential reason for underprediction of F_{abs} is carrier mediated uptake across the intestinal membrane. Overprediction of F_{abs} is likely to occur for compounds that exhibit solubility limited absorption at the given dose and/or for substrates of active efflux transporters (P-gp) or metabolizing enzymes in the intestinal epithelium (most likely CYP3A4). If only the compounds are considered that have been ranked as highly soluble (>100 mg/l) and predominately passively absorbed, the predictivity of the monkey model for F_{abs} was very good. Ninety-three percent (27/29 = 93%) of the passively absorbed compounds with a solubility >100 mg/l were predicted within a pre-defined linear range between ±5% (for non-absorbable compounds) to ±20% (for completely absorbed compounds) around the line of identity, which is considered to be an acceptable range for use in drug research and development (7). It should be noted, however, that 19 of the 29 compounds (66%) exhibited a fraction dose absorbed of greater than 90% *in vivo*. Thus, the validation data-set is biased towards well absorbed compounds. The two compounds outside of this range were caffeine and rolipram. Caffeine is among the compounds of this data-set that fulfils the criteria for para-

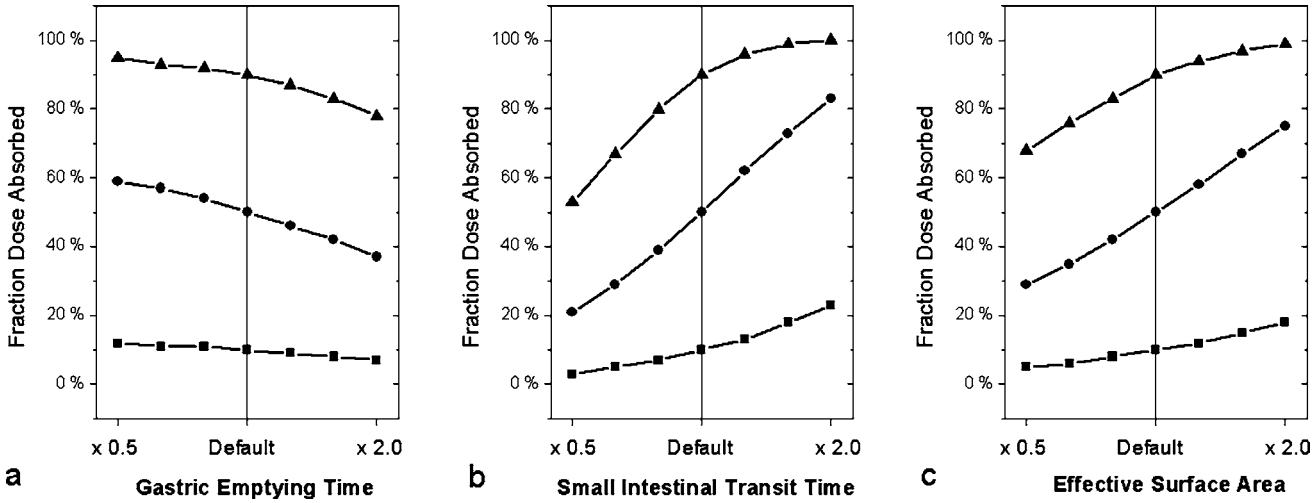

Fig. 2. Parameter sensitivity analysis: The fraction dose absorbed is shown as a function of the gastric emptying time (**a**), small intestinal transit time (**b**) and effective surface area (**c**) for three different compounds with low (*filled square*), medium (*filled circle*), and high (*filled triangle*) intestinal permeability. Parameters were varied within two-fold of their default value (for details see text).

cellular transport, which are high hydrophilicity and low molecular weight. The cut-off values for molecular weight enabling diffusion through the aqueous pores vary in the literature between approximately 150 and 400 (7,30–32). Moreover, paracellular transport rates appear to be species dependent. For example, a two-fold difference between jejunal permeabilities of paracellular markers has been reported between monkeys and rabbits (18). It is therefore possible, that the underestimation of F_{abs} of caffeine is caused by an underestimation of the paracellular contribution to the total intestinal permeability in monkeys, when the parameters for paracellular permeability obtained in humans (7) were applied in this monkey model (Eq. 1 and Table II). On the other hand, such an underestimation is not evident for the bisphosphonates, etidronate and tilodrunate, which are also predominately absorbed via the paracellular route (32). The second outlier, rolipram, is an effective cyclic nucleotide phosphodiesterase enzymes (PDE) inhibitor (33). The PDE enzymes hydrolyze cyclic nucleotides in the gut wall, which mediate the relaxation of the gastrointestinal smooth muscle. Since inhibition of PDE leads to reduced contractile activity in the gut, it has been postulated that rolipram could be used in treatment of hyperactivity of the intestines (33). It can therefore be hypothesized that a rolipram-induced reduction in motility and hence increased time for absorption could lead to more complete absorption. If this is the case, the F_{abs} in monkeys estimated by the model would be underestimated. The hypothesis is supported by the observation that the F_{abs} of rolipram generated in the simulations is very sensitive to changes in the small intestinal transit time.

All other compounds that fall outside of the above defined acceptable region either have a low solubility (<100 mg/l) or are known substrates for active transporters and/or for CYP3A4. The F_{abs} values for the majority of the low solubility compounds have been over predicted by the model, as would be expected. The threshold for a low solubility was set to 100 mg/l in this study because preliminary simulations with the model demonstrated that this solubility value marks the borderline for the onset of solubility limitation at doses of 500–1,000 mg [dependent on the intestinal permeability (7)]. This is considered to be an upper limit for use in monkey experiments. Since the actual doses for which F_{abs} was determined in monkeys was not reported in (2) it is not unexpected that 50% (3/6) of the compounds from the low solubility class still fell into the region of acceptable agreement in Fig. 1.

Captopril (24) and valacylcovir (25,26) are both actively taken up by the intestinal peptide carrier system. Their F_{abs} values were consequently underpredicted on the basis of pure passive absorption as expressed in Eq. 1. Bromocriptine is reported to be a substrate of the p-glycoprotein efflux transporter (27) as well as for CYP3A4 (29), which potentially caused a reduction in the observed F_{abs} relative to the predicted F_{abs}. It is noticeable that the other two compounds that were drastically overpredicted by the model (benazepril and lovastatin) are also biotransformed by CYP3A4 (28,29).

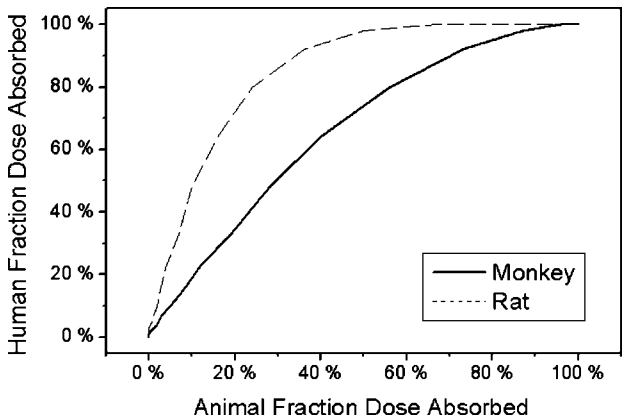

Fig. 3. Inter-species comparison: The predicted fraction dose absorbed in humans is shown as a function of the predicted fraction dose absorbed in monkeys and rats for passively absorbed compounds under non-solubility limited conditions. Default parameters for the fasted state were assumed in these simulations.

CYP3A4 is the most abundant cytochrome in the human gut wall and is also expressed in the intestinal epithelium of monkeys (34,35). Furthermore, only a calculated LogP of 4.51 value was available for bromocriptine. Because the correlation between LogP and LogMA, the preferred lipophilicity measure (7), is not linear at such high LogP values (14), the overestimation of F_{abs} in case of bromocriptine might at least in part also be attributed to an overestimation of the lipophilicity parameter that was used as input. Similarly, the LogP value reported for valacyclovir is well below the region of reasonable LogMA-LogP correlation (14), and could therefore also contribute to the underestimation of F_{abs} of this compound.

Due to the large reported inter-individual variability of many of the physiological values that affect gastro-intestinal absorption in monkeys, such as for example GET and SITT, it is reasonable to perform a sensitivity analysis with respect to these parameters. A_{eff} was included in the sensitivity analysis, because it was (a) estimated assuming the same relative surface enhancement factors (A_{eff}/A_{geom}) in monkeys as in humans (7) and (b) this parameter is known to exhibit substantial inter-individual variability in humans (7). The sensitivity analysis showed that, as one would expect, the sensitivities were always largest for the compound with medium permeability, because the fraction dose absorbed vs. permeability relation has the steepest slope in this region (7). If absorption is poor, the influence of changes in the gastric emptying and intestinal transit time or effective surface area can be expected to be small.

The comparison of predicted F_{abs}-values between species for passively absorbed compounds with no solubility-limitation and negligible gut wall metabolism shows that on average (i. e., assuming the default parameters for gastro-intestinal physiology), rat as well as monkey tend to underestimate the fraction dose absorbed in humans, but rat to a greater extent than monkey (Fig. 3). This is not unexpected since the differences in the predicted F_{abs} reflect the differences in the physiological parameters and dimensions of the gastro-intestinal tract. In this respect, the monkey is much closer to a human than a rat. Although the average small intestinal transit time in rats [6 h (6)] is reported to be longer than in humans [4 h (7)] and monkeys (2.5 h, Table II), this cannot compensate for the much smaller intestinal dimensions, especially the much smaller effective surface area that is available for absorption (approximately 550 cm^2 (6) versus 70 m^2 in humans (7) and 43 m^2 in monkeys).

So far, model predictions of F_{abs} in monkeys only accounted for the lipophilicity and molecular weight of the compound. Solubility- or dissolution-limitation as well as the influence of active transporters or gut wall metabolism are not considered in this model. Therefore, application of the described model can only be suggested for compounds predominately undergoing passive absorption under non-solubility limited conditions with negligible gut wall metabolism. Further validation work is also necessary before the application of this model to controlled release formulations can be suggested. Consumption of a meal has a strong impact on the pH profile in monkeys, which also depends on the type of food. In comparison to humans, the intestinal pH is more basic in monkeys, which can limit their use as animal model for ionized compounds and pH dependent formulations. To overcome the limitations of the current model is the subject of ongoing work.

CONCLUSIONS

A mechanistic absorption model in monkeys based on physiological information available in the literature has been developed and validated. The model is well suited to predict the fraction dose absorbed of passively absorbed compounds after oral administration and to assess the influence of inter-individual physiological variability on oral absorption in monkeys. It can, thus, be useful to interpret pharmacokinetic data obtained after oral administration in monkeys in the drug development process. Future experimental studies to refine the physiological parameter database in monkeys and a demonstration of the validity in case of solubility- or dissolution limited absorption as well as for compounds undergoing active transport processes or metabolism in the intestine are desired.

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