A Systematic Evaluation of the Use of Physiologically Based Pharmacokinetic Modeling for Cross-Species Extrapolation

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ABSTRACT: Transfer of knowledge along the different phases of drug development is a fundamental process in pharmaceutical research. In particular, cross-species extrapolation between different laboratory animals and further on to first-in-human trials is challenging because of the uncertain comparability of physiological processes. Physiologically based pharmacokinetic (PBPK) modeling allows translation of mechanistic knowledge from one species to another by specifically considering physiological and biochemical differences in between. We here evaluated different knowledge-driven approaches for cross-species extrapolation by systematically incorporating specific model parameter domains of a target species into the PBPK model of a reference species. Altogether, 15 knowledge-driven approaches were applied to murine and human PBPK models of 10 exemplary drugs resulting in 300 different extrapolations. Statistical analysis of the quality of the different extrapolations revealed not only species-specific physiology as the key determinant in cross-species extrapolation but also identified a synergistic effect when considering both kinetic rate constants and gene expression profiles of relevant enzymes and transporters. Moreover, we show that considering species-specific physiology, plasma protein binding, enzyme and transport kinetics, as well as tissue-specific gene expression profiles in PBPK modeling increases accuracy of cross-species extrapolations and thus supports first-in-human trials based on prior preclinical knowledge. © 2014 Wiley Periodicals, Inc. and the American Pharmacists Association J Pharm Sci 104:191–206, 2015

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INTRODUCTION

Development of novel drugs is a time-consuming and laborious process. In particular, the translation of preclinical knowledge generated in laboratory animals to first-in-human studies is a critical step with attrition rates above 30%.[1] In this regard, reliable cross-species extrapolations are needed to guarantee safety in human clinical trials. Current approaches for a cross-species extrapolation are often based on empirical allometric scaling techniques.[2] In this context, pharmacokinetic (PK) parameters such as the clearance of administered drugs are correlated to the body weight by using a power law function. This, however, requires observations of that parameter for a series of reference species.[3] In a similar approach, Dedrick plots may be used to predict the plasma drug concentration–time profile based on simple dose normalizations and species-invariant time methods.[4] In addition, allometric scaling laws have been developed by taking into account the brain weight or the maximum life span potential in order to improve the predictive accuracy.[5] Such approaches have been used successfully for single compounds such as dolasetron.[6] However, limitations of allometric scaling techniques have also been shown.[7,8]

Recent approaches incorporate physiologically based pharmacokinetic (PBPK) models to extrapolate between species.[9,10] PBPK models describe physiological processes governing the fate of a drug in the body. In PBPK models, relevant tissues and organs of an organism are represented as compartments which are connected by blood flow. Organs are further subdivided into more detailed subcompartments such as blood cells, plasma, interstitium, and intracellular space. Notably, PBPK models are based on prior information regarding species-specific physiology (SP).[11] Mass transfer is described by using so called distribution models which are parameterized based on the physicochemical properties of a drug.[12–15] Because of the large degree of mechanistic information included in PBPK models, they are particularly well suited for extrapolation to new treatment scenarios or specific subgroups of patients. Human PBPK models have been used before, for instance, for pediatric scaling, dose extrapolation, and prediction of adverse events in

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high-risk subgroups of patients.[16–18] PBPK models can also be used for cross-species extrapolation from one species to another. For cyclosporine, a PBPK modeling framework combined with physiological scaling laws has been used successfully to extrapolate from rats to pigs, monkeys, and humans.[9] Also, scaling of physiological, metabolic, and excretory variables from a mouse PBPK model of docetaxel was used for the prediction of drug plasma levels in men.[10] Although both approaches yielded reasonable predictions for single compounds, a more generalized investigation of the benefit of using PBPK modeling for extrapolation from one species to another is still missing.

We here systematically evaluated different approaches for cross-species extrapolation by using murine and human PBPK models for 10 exemplary drugs. PK of a target species was predicted by stepwise adjusting physiological and biochemical parameters in a reference PBPK model of another species. Thereby, the improvement in model accuracy for different model parameter domains reflecting different degrees of prior information was quantified. We first established a naive model, which neglects any knowledge about interspecies differences and which we henceforth used as a benchmark model. Secondly, we generated various extrapolated models based on different degrees of prior knowledge and quantified the relative improvement in model error (ME) in these models with respect to the naive benchmark model. This systematic workflow allowed us to assess the benefit of different knowledge-driven approaches for cross-species extrapolation, which predict pharmacokinetic profiles in a target species based on mechanistic knowledge obtained in a reference species.

MATERIALS AND METHODS

Software

The software tool PK-Sim® (version 5.1; Bayer Technology Services GmbH, Leverkusen, Germany) was used to build the PBPK models for the considered drugs.[11,15,19,20] The different knowledge-driven approaches for cross-species extrapolation were implemented in MATLAB (version 7.11.0; The MathWorks, Inc., Natick, Massachusetts) by use of the MoBi® Toolbox for MATLAB (version 2.3; Bayer Technology Services GmbH). Statistical computations are performed using R (version 3.0.2, 2013; R Core Team, http://www.R-project.org).

Drug Selection

In total, PBPK models for 10 different drugs (torsemide, talinolol, midazolam, caffeine, pravastatin, morphine, docetaxel, dextromethorphan, cyclosporine, and erythromycin) were developed. Only intravenous administration was considered. The drugs have been selected such that the corresponding route of degradation is mainly governed by a single reaction, that is, either enzyme-catalyzed metabolization or active drug transport. This selection criterion ensures that the PBPK models developed have a comparable structural complexity.

Major physicochemical properties of the considered drugs are listed in Table 1. The octanol/water partition coefficient (log *P*) ranges from −0.07 (caffeine) to 3.64 (cyclosporine). The majority of the molecular weights (MWs) are between 190 and 430 g/mol except for erythromycin (733.92 g/mol), docetaxel (807.88 g/mol), and cyclosporine (1202.60 g/mol). Acid dissociation constants (p*K*a) lie in the range of 4.56 (pravastatin) to 11.83 (cyclosporine).

Table 1. Physicochemical Properties

Drug	Log *P*	MW (g/mol)	p*K*a
Torsemide[21]	0.57[22]	348.40[22]	7.10[22]
Talinolol[23]	2.30[24]	363.50[25]	9.43[25]
Midazolam[26]	2.70[27]	325.77[28]	6.04[28]
Caffeine[29]	−0.07[28]	194.20[28]	10.40[28]
Morphine[30]	0.89[28]	285.30[28]	8.20[28]
Docetaxel[31]	2.92[28]	807.88[28]	10.96[28]
Dextromethorphan[32]	3.60[28]	271.39[28]	9.85[28]
Cyclosporine[33]	3.64[28]	1202.60[28]	11.83[28]
Erythromycin[34]	3.06[28]	733.92[28]	8.88[28]
Pravastatin[35]	1.65[28]	424.53[28]	4.56[28]

Molecular weight (MW), acid dissociation constant (p*K*a), and the octanol/water partition coefficient (log *P*) for the 10 considered drugs.

Experimental Data

The developed PBPK models for mouse and human are validated by comparing the simulated concentration–time profiles with measured data of intravenous studies obtained from the literature. For torsemide, talinolol, and pravastatin, own experimental data were measured quantifying pharmacokinetic profiles in mice. Table 2 provides an overview of the administration route, the dose, the measuring and infusion time, as well as the sampling site from which the blood samples were collected.

Experimental Procedure

Mouse Hepatocyte Isolation and Cultivation

Primary mouse hepatocytes were isolated from male C57BL-6N mice by a two-step perfusion technique as previously described.[50] The viability of cells was checked with trypan blue exclusion; the minimal viability was 97%. In order to prepare confluent sandwich cultures; cells were cultivated in six-well plates coated with collagen-1 (Roche diagnostics GmbH, Manheim, Germany) at a density of 800,000 cells/well in Williams E medium supplemented with 10% fetal calf serum (FCS), 2 mM L-glutamine, 100 units/mL penicillin, 0.1 mg/mL streptomycin, 10 μg/mL gentamycin (PAN Biotech, Aidenbach, Germany) and 100 nM dexamethasone (Sigma–Aldrich, Munich, Germany). The cells were allowed to attach for 3 h at 37°C, 5% CO_2. Subsequently, the cells were washed three times with Williams E medium and a second layer of collagen was added. Cells were further cultivated overnight in cultivation media without FCS at 37°C, 5% CO_2 before starting the experiment.[50]

Determination of Enzyme Kinetics

Enzyme kinetics was estimated by incubating primary mouse hepatocytes for 30 min with midazolam or 60 min with caffeine, codeine, or torsemide at 37°C. Substrate concentrations ranged from 1 to 500 μM for torsemide, caffeine, and codeine, and from 0.5 to 250 μM for midazolam. Culture supernatants (50 μL) were then collected and mixed with 5 μL of 250 mM formic acid and stored at −20°C until analysis by LC–MS/MS. After thawing, samples were centrifuged at 16,000*g* for 5 min and the supernatant spiked with the internal standard mixture, and 10 μL was used for LC–MS/MS analysis (Table S1, Supporting Information).