Research Article

Theme: Challenges and Opportunities in Pediatric Drug Development

Guest Editors: Bernd Meibohm, Jeffrey S. Barrett, and Gregory Knipp

A Workflow Example of PBPK Modeling to Support Pediatric Research and Development: Case Study with Lorazepam

A. R. Maharaj,¹ J. S. Barrett,² and A. N. Edginton^{1,3}

Received 28 September 2012; accepted 29 December 2012; published online 24 January 2013

Abstract. The use of physiologically based pharmacokinetic (PBPK) models in the field of pediatric drug development has garnered much interest of late due to a recent Food and Drug Administration recommendation. The purpose of this study is to illustrate the developmental processes involved in creation of a pediatric PBPK model incorporating existing adult drug data. Lorazepam, a benzodiazepine utilized in both adults and children, was used as an example. A population-PBPK model was developed in PK-Sim v4.2® and scaled to account for age-related changes in size and composition of tissue compartments, protein binding, and growth/maturation of elimination processes. Dose (milligrams per kilogram) requirements for children aged 0–18 years were calculated based on simulations that achieved targeted exposures based on adult references. Predictive accuracy of the PBPK model for producing comparable plasma concentrations among 63 pediatric subjects was assessed using average-fold error (AFE). Estimates of clearance (CL) and volume of distribution (V_{ss}) were compared with observed values for a subset of 15 children using fold error (FE). Pediatric dose requirements in young children (1–3 years) exceeded adult levels on a linear weight-adjusted (milligrams per kilogram) basis. AFE values for model-derived concentration estimates were within 1.5- and 2-fold deviation from observed values for 73% and 92% of patients, respectively. For CL, 60% and 80% of predictions were within 1.5 and 2 FE, respectively. Comparatively, predictions of V_{ss} were more accurate with 80% and 100% of estimates within 1.5 and 2 FE, respectively. Using the presented workflow, the developed pediatric model estimated lorazepam pharmacokinetics in children as a function of age.

KEY WORDS: lorazepam; PBPK; pediatrics.

INTRODUCTION

The Food and Drug Administration (FDA) enacted the Pediatric Research Equity Act in 2003, requiring pharmaceutical companies to assess pharmacokinetics (PK), safety, and efficacy of new drug products in pediatric subjects. Recently, several FDA pediatric submissions have incorporated physiologically based pharmacokinetic (PBPK) models, stimulating an interest in their utility among regulatory authorities (1). In a March 2012 meeting, the majority of the FDA's Pharmaceutical Science and Clinical Pharmacology Advisory Committee voted to support the use of PBPK modeling for pediatric drug development; a decision with potential implications toward the manner in which pediatric drug information is derived.

PBPK modelling is characterized by the use of mathematical algorithms to predict the interplay between drug specific

characteristics and organism anatomy and physiology. Similar to empirically derived compartmental models, the structure of PBPK models includes compartments in order to describe the processes of absorption, distribution, metabolism, and excretion (ADME). In a PBPK model, however, compartments are based on actual organs with inherent volumes and blood flows linked through the vasculature. The mechanistic nature of PBPK models permit rational scaling between organisms (*i.e.*, rat to human) as well as developmental stages (*i.e.*, adult to child). This is the result of defining ADME as a function of anatomy, physiology, and biochemistry: components not accounted for in traditional compartmental models.

Use of pediatric PBPK models offer researchers an *a priori* approach to predict a compound's PK behavior in children, with or without prior PK data in humans, though knowledge of the drug substance's physicochemical characteristics is essential. The developmental processes involved in the creation of pediatric PBPK models has been documented by several researchers and typically include defining physiology and anatomy, protein binding, and clearance, all as a function of age (2–4). Amongst the literature, pediatric PBPK models have been utilized in several different capacities: suggesting starting doses for children of different age groups, predictions of environmental contaminant exposure, optimization of clinical

¹ School of Pharmacy, University of Waterloo, 200 University Ave W, Waterloo, Ontario N2L 3G1, Canada.

² Laboratory for Applied Pharmacokinetics and Pharmacodynamics, Division of Clinical Pharmacology and Therapeutics, The Children's Hospital of Philadelphia, Philadelphia, Pennsylvania, USA.

³ To whom correspondence should be addressed. (e-mail: aedginto@uwaterloo.ca)

drug trial design (sampling schedule, number of patients, etc.), and assessment of potential drug–drug interactions (1,4–6).

Lorazepam, an intermediate acting benzodiazepine, is administered for a variety of “off-label” indications in children including status epilepticus, chemotherapy-induced nausea and vomiting, anxiety, sedation, and procedural amnesia (7). Despite extensive use among pediatric patients (8,9), further information is still needed to adequately define its safety, efficacy, and PK, as demonstrated by lorazepam’s inclusion on the National Institutes of Health (NIH) priority list of medications requiring urgent pediatric studies in 2003 (10). Most published literature regarding lorazepam PK parameters (Table 1) is primarily focused on an adult patient population. In humans, lorazepam exhibits an affinity for albumin with a bound fraction in plasma of approximately 89% (11–13). Hepatic metabolism and renal filtration represent the major and minor pathways of clearance, respectively (14–16).

This study will use a systematic workflow to demonstrate how adult drug data are leveraged in the development of pediatric PBPK models. Using lorazepam as an example, a rational prediction of lorazepam PK will be completed as a function of age and further compared with PK data from a pediatric clinical study.

METHODS

Model Building Workflow

Following the proposed workflow (Fig. 1), the development of a pediatric population PBPK model is presented using lorazepam as an example. The derived model will be used to assess PK differences between children (age 0–18 years) and adults.

Development of the Adult PBPK Model

Model Structure and Parameterization

All simulations (adult and pediatric) were completed using PK-Sim® v 4.2 (Bayer Technology Services, Leverkusen, Germany), which implements a whole-body PBPK model consisting

of 15 organs. Due to the physicochemical nature of lorazepam (low molecular weight, <500 g/mol; moderate lipophilicity; and neutral at physiologic pHs), all organs were considered kinetically equivalent to well-stirred compartments with the exception of the brain. PK-Sim® utilizes a permeation barrier between the plasma and interstitial fluid of the brain to simulate the physiologic equivalent of the blood–brain barrier. Specific physicochemical, ADME, and anatomic/physiologic data used for initial parameterization of the adult lorazepam model are presented in Table 1. Tissue/plasma partition coefficients (K_p) were predicted using the *in silico* tissue composition approach proposed by Rodgers and Rowland (17–19). Using the above procedures, an initial adult model was generated.

Optimization of the Adult Model

Concentration-time data from four separate adult PK studies (15,20–22) were dose-normalized and compared with the output of the initial adult PBPK model. Outputs were generated using the average observed clearance in conjunction with the mean age and weight of participants in the studies to define anatomical and physiological values (23). A visual check was used to evaluate predictive accuracy between simulated (PBPK model) *versus* observed concentration-time data and appropriateness of line shape. Subsequently, the model parameters of intrinsic hepatic clearance ($CL_{int(hep-UGT2B7)}$) as well as lipophilicity ($\log P$), a direct predictor of K_p (s), were optimized using the MoBi® Toolbox for MATLAB® (Bayer Technology Services GmbH, Leverkusen, Germany/The Mathworks Inc., Natick, MA), an integrative modeling tool that permits parameter analysis. Parameters were iteratively optimized to achieve values which minimized the error between observed and model-derived concentration-time points.

Adult Population Model

A virtual population of 100 adult individuals was created using PK-Sim®’s Population Module in order to simulate the effect of anthropometric and intrinsic clearance variability on the PK behavior of lorazepam. The module incorporates the work conducted by Willmann *et al.* (23) to create a population

Table 1. Physicochemical, ADME, and Anatomic/Physiologic Data for Initial Parameterization of the Adult Lorazepam PBPK Model

Data for initial parameterization		
Physicochemical	LogP	2.39 (34)
	pK_a	1.3 (base), 11.5 (acid) (35)
ADME	f_{up}	0.11 (11–13)
	$CL_{int(hep-UGT2B7)}$	0.439 ml/min/g _{liver} (13,15,20–22)
	CL_{GFR}	0.01 ml/min/kg (14–16)
	B/P	0.642 (36)
Anatomic/physiologic	Organ size	Generated using simple demographic information
	Organ blood flow	(sex, male; age, 26.6 years; weight, 68 kg) by the
	Tissue composition	methods described in Willman <i>et al.</i> (23)

Tissue/plasma (K_p) were estimated using the methods described by Rodgers and Rowland (17–19); hepatic (plasma) clearance was estimated using a well-stirred liver model (Eq. 3)

$\log P$ logarithm of the octanol–water partition coefficient (lipophilicity), pK_a negative logarithm of the acid dissociation constant, f_{up} plasma fraction unbound, $CL_{int(hep-UGT2B7)}$ intrinsic clearance of hepatic isozyme UGT2B7, CL_{GFR} renal (plasma) clearance due to glomerular filtration, B/P blood/plasma partition coefficient, ADME absorption, distribution, metabolism, and excretion

Fig. 1. Proposed workflow for scaling adult PBPK models toward children

of individuals with associated variability consistent with real life observations. Demographic constraints (sex, age, weight) of the simulated population were reflective of the adult subjects included among the lorazepam PK studies (15,20–22). All subjects received a 2 mg intravenous dose, representing a commonly prescribed adult dosage. Inter-patient variability associated with UGT2B7 intrinsic activity, the hepatic enzyme responsible for lorazepam metabolism (24), was estimated from

an *ex vivo* metabolism study for the probe substrate zidovudine using human liver microsomes (25). As a result, UGT2B7 intrinsic clearance was varied based on a log-normal distribution with a geometric standard deviation of 1.34. The average area under the plasma concentration-time curve to infinity ($AUC_{0 \rightarrow \infty}$), a measure of systemic exposure, was tabulated to comparatively assess dosage equivalency between adults and children. Extrapolation to infinity occurred by dividing the final

concentration (C_{last}) by the elimination rate constant (λ_z), where λ_z was calculated based on a linear regression of the final 10% of predicted time points transformed on the natural log scale. To evaluate the range of PK variability among the virtual population, 5% and 95% prediction intervals for concentration-time values were generated and compared with dose-normalized observed values (assuming linear PK) from each of the four lorazepam PK studies (15,20–22).

Age-Dependent Scaling of PBPK Model Parameters

Protein Binding. Prediction of pediatric protein binding was estimated using equations presented by McNamara and Alcorn (26). The authors successfully predicted the fraction of protein binding in infants for several compounds using fraction unbound in adults and compound-specific plasma protein affinity. For lorazepam, literature data indicate an affinity for albumin and an average fraction unbound (plasma) of 0.11 in adults (11–13).

Clearance. Total clearance of a compound is calculated as the sum of its individual clearance pathways. For lorazepam, the clearance in children was calculated as the sum of scaled hepatic and renal clearances using a physiologically based approach. The process of physiologic hepatic clearance scaling relies on the following underlying assumptions (24):

1. Pathways of clearance in children are the same as those observed in adults.
2. Well-stirred model conditions hold (hepatic uptake of the compound is a function of blood flow—not permeability across cell membranes).
3. Enzyme metabolism follows first-order kinetics (concentrations are within linear range—no enzyme saturation).

Hepatic UGT2B7 enzyme activity is on average 5% of the adult value at term, increases to 30% by the age of 3 months, and reaches adult activity by 1 year of age. To quantify the relationship between age and enzyme activity, a cubic spline function, as derived by Edginton *et al.* (24), was utilized. Scaled intrinsic clearance in pediatric subjects can be calculated from adult values using the following formula (27),

$$CL_{int\ UGT2B7\ (child)g\ liver} = OSF_{UGT2B7} \times CL_{int\ UGT2B7\ (adult)g\ liver} \quad (1)$$

where $CL_{int\ UGT2B7\ (child)g\ liver}$ is the scaled intrinsic clearance due to UGT2B7 per gram of liver, OSF_{UGT2B7} is the ontogeny scaling factor for UGT2B7 specific to the age of the child, and $CL_{int\ UGT2B7\ (adult)g\ liver}$ is the intrinsic clearance due to UGT2B7 per gram of liver in adults. Total hepatic plasma clearance can be derived from intrinsic clearance using the well-stirred model,

$$CL_{int\ (child)total\ liver} = CL_{int\ UGT2B7\ (child)g\ liver} \times LW_{child} \quad (2)$$

$$CL_{hepatic\ (child)} = \frac{Q_h\ (child) \times f_{u,p\ (child)} \times CL_{int\ (child)total\ liver}}{Q_h\ (child) + \left(f_{u,p\ (child)} \times CL_{int\ (child)total\ liver} \right) / B/P\ (child)} \quad (3)$$

where $CL_{int\ (child)total\ liver}$ is the whole liver intrinsic clearance in the child, LW_{child} is the liver weight of the child, $CL_{hepatic\ (child)}$ is total hepatic clearance in the child, $Q_h\ (child)$ is liver blood flow in the child, $f_{u,p\ (child)}$ is the scaled unbound fraction (plasma) in the child, and $B/P\ (child)$ is the blood/plasma (B/P) ratio in the child. The B/P ratio is affected by both changes in protein binding and hematocrit and can be tabulated using equations derived by Rodgers and Rowland (17),

$$Kp_{rbc,u} = \frac{B/P_{(adult)} - 1 + H_{(adult)}}{H_{(adult)} \times f_{u,p\ (adult)}} \quad (4)$$

$$B/P\ (child) = 1 + [H_{(child)} \times (f_{u,p\ (child)} \times Kp_{rbc,u} - 1)] \quad (5)$$

where $Kp_{rbc,u}$ is the unbound partition coefficient of red blood cells (assumed to be constant between adults and children), $B/P_{(adult)}$ is the blood/plasma ratio in adults, $H_{(adult)}$ is the hematocrit in adults, $f_{u,p\ (adult)}$ is the fraction unbound (plasma) in adults, and $H_{(child)}$ is the hematocrit of the child (28).

The effects of maturation and growth on renal function were examined in a seminal paper by Hayton (29). The study proposed a series of equations to estimate renal function parameters, such as glomerular filtration rate (GFR) and active secretion, in children as a function of age and weight. To scale adult renal clearance values toward pediatric patients, the estimated GFR of the child, as determined using Hayton's algorithm, was used in conjunction with the following equation proposed by Edginton *et al.* (24),

$$CL_{GFR\ (child)} = \frac{GFR_{(child)}}{GFR_{(adult)}} \times \frac{f_{u,p\ (child)}}{f_{u,p\ (adult)}} \times CL_{GFR\ (adult)} \quad (6)$$

where $CL_{GFR\ (child)}$ is the child's clearance due to glomerular filtration, $GFR_{(child)}$ is the estimated GFR of the child, $GFR_{(adult)}$ is the GFR in adults (assumed to be 110 ml/min (24)), and $CL_{GFR\ (adult)}$ the clearance due to glomerular filtration in adults.

Anatomy/Physiology. The age dependence of body weight, height, organ weights, and blood flows were obtained from Edginton *et al.* (3) and represent those values currently used as default values in PK-Sim®. The above parameters were derived based on the mean value for European children ranging from neonates to 18 years old.

Age-Dependent PK Simulations

Pharmacokinetic variability among pediatric populations of various age-groups was assessed by creating virtual populations of 100 individuals aged 0, 3, 7, and 14 days; 1, 2, 3, 6, and 9 months; and 1, 1.5, 2, 3, 4, up to 18 years. All patients received a single 0.05-mg/kg dose of intravenous lorazepam, representing a common pediatric dosage. Inter-patient variability associated with hepatic UGT2B7 intrinsic clearance was assumed to be identical to that of adults. Dosage equivalency between adults and children, the dose (milligrams per kilogram) required to achieve an $AUC_{0 \rightarrow \infty}$ similar to the average value obtained in the adult population model, was calculated for each simulated

population. In addition, to demonstrate the potential utility of this modelling technique, mean tissue concentration-time profiles for targeted organs (adipose, muscle, brain) were simulated for a population of 2-year-old patients ($n=100$) following a 0.05-mg/kg intravenous dose.

Assessment of Model Accuracy

The predictive accuracy of the derived pediatric lorazepam model at estimating concentration-time values and PK parameters was evaluated using an observed data set provided by Chamberlain *et al.* (30). Concentration-time data from 63 pediatric patients, ranging from 5 months to 17 years of age, who received intravenous lorazepam either electively or part of routine treatment of status epilepticus, were included in the sample (30). Forty of the 63 pediatric subjects received a single intravenous dose of lorazepam. Of these subjects, 15 received lorazepam electively. Pediatric PBPK model concentration estimates were based on simulations incorporating the age, weight, and height for each child, in addition to the dose administered. The predictive accuracy of the pediatric PBPK model for estimating observed concentration-time values for each individual patient was assessed by tabulating the average-fold error (AFE).

$$AFE = 10^{\frac{1}{n} \sum \log \left(\frac{\text{predicted}}{\text{observed}} \right)} \quad (7)$$

Observed PK parameters derived from non-compartmental analysis (NCA) were determined for elective patients ($n=15$), for whom intense serial sampling was performed following intravenous lorazepam administration (30). Lorazepam total body clearance (CL) was calculated as $\text{Dose}/\text{AUC}_{0 \rightarrow \infty}$, where $\text{AUC}_{0 \rightarrow \infty}$ was determined using the linear log trapezoidal rule up to the final sample time with extrapolation to infinity based on $C_{\text{last}}/\lambda_z$. Non-compartmental extrapolation of $\text{AUC}_{0 \rightarrow \infty}$ was analogous to the approach used for PBPK model assessment but differed in the algorithm used to estimate λ_z , which was computed based on regression of the natural logarithm of concentration values during the post-distributive phase. The volume of distribution (V_{ss}) was calculated as $V_{ss} = \text{MRT} \times \text{CL}$, in which MRT is the mean residence time. MRT was calculated as $\text{AUMC}_{0 \rightarrow \infty} / \text{AUC}_{0 \rightarrow \infty}$, where $\text{AUMC}_{0 \rightarrow \infty}$ is the area under the first moment curve from zero to infinity. These parameters were similarly calculated from PBPK model simulations. The relative accuracy of predicted CL and V_{ss} values, as determined by physiologic scaling, were assessed against observed values (NCA) for each patient using fold error.

$$\text{fold error} = \frac{\text{predicted}}{\text{observed}} \quad (8)$$

As a final evaluation, the PK variability associated with ontogeny and anthropometrics among a virtual pediatric patient population after receiving a single 0.05 mg/kg intravenous (bolus) dose of lorazepam was compared with individual observed data. Five percent and 95% concentration-time prediction intervals were generated for the entire age range (0–18 years) of the virtual pediatric population with equal representation from each age. The simulated profile was evaluated using individual observed concentration-time data (normalized to 0.05 mg/kg) for 40 of the 63 pediatric patients who received a single intravenous dose of lorazepam (30).

RESULTS

Mean adult hepatic intrinsic clearance and logP values were optimized to 0.416 ml/min/g_{liver} and 2.43, respectively. Figure 2a compares the concentration-time profile derived from the optimized adult population model to dose-normalized observed data from four PK studies (15,20–22). Observed data in the initial phase of distribution were over-predicted by the model; however, subsequent data were well described. The range of PK variability for lorazepam associated with a simulated pediatric population (0–18 years) is displayed in Fig. 2b. Approximately 72% of dose-normalized observed lorazepam concentration-time values from a subset of 40 pediatric patients fell within the 90% prediction interval of the simulated population.

The dosage (milligrams per kilogram) required to achieve similar $\text{AUC}_{0 \rightarrow \infty}$ as those observed in adults after a 2 mg intravenous dose of lorazepam varied as a function of age (Fig. 3a, b). Newborns (0 days old) required approximately one tenth of the weight normalized dose given to adults, whereas infants between the ages 1 and 3 years required dosages (milligrams per kilogram) that exceeded those of adults. After 3 years of age, the weight normalized dose slowly decreased towards adult requirements.

Predicted tissue specific concentrations from the pediatric PBPK model were calculated for a virtual population of 2-year-old patients following a 0.05-mg/kg dose of lorazepam (Fig. 4). Estimates indicate that muscle and adipose tissues equilibrate with plasma concentrations at a much faster rate compared with the brain.

The predictive accuracy of concentration-time estimates derived by our pediatric PBPK model was assessed using individual patient AFE values for all 63 sample patients (Fig. 5a). Forty-four percent, 73%, and 92% of patients had AFE values within 1.25-, 1.5-, and 2-fold deviation from observed values, respectively.

PBPK estimates of CL and V_{ss} were compared with observed values obtained by NCA from the 15 elective patients (Fig. 5b, c). For CL, 40%, 60%, and 80% of predictions were within 1.25-, 1.5-, and 2-fold deviation from observed values, respectively. Comparatively, predictions of V_{ss} were relatively more accurate with 53%, 80%, and 100% of estimates within 1.25-, 1.5-, and 2-fold error from observed values, respectively.

DISCUSSION

Integration of PBPK modelling with pediatric drug research has been illustrated by five recent FDA submissions (31). Common applications of modelling in the aforementioned submissions include optimizing study design, recommending starting doses for different age groups, and facilitating covariate analysis. The workflow presented in Fig. 1 ensures pediatric PBPK models are developed using physiologically rationale approach and incorporates elements from two recently proposed workflows. Similar to Fig. 1, Leong *et al.* (1) and Edginton (32) both presented workflows which included an evaluation of appropriateness of the adult model prior to scaling. Refinement (optimization) of the adult model was also included by Leong *et al.* Edginton's use of virtual populations to assess PK variability among simulated adult and pediatric populations was similar to the approach adopted in this study.

Fig. 2. **a** Predicted (solid line corresponds to geometric mean; dashed lines corresponds to 5th and 95th percentiles; virtual population $n=100$) versus observed (symbols – (15, 20–22)) plasma concentration versus time data following a 2-mg IV lorazepam bolus in adults. Log (concentration) versus Log (time) plot is displayed in insert. **b** Predicted (solid line corresponds to geometric mean; dashed lines corresponds to 5th and 95th percentiles; virtual population $n=1140$) versus observed (symbols – (30)) plasma concentration versus time data following a 0.05 mg/kg IV lorazepam bolus in children aged 0 to 18 years. Log (concentration) versus Log (time) plot is displayed in insert

The workflow's evaluation process ensures that the derived adult model adequately reflects *in vivo* PK data. A major underlying assumption involved in physiologic scaling necessitates pathways of clearance in children are the same as those observed in adults. This ensures that physiologic processes accounted for in the adult model will be mirrored in the pediatric model. As such, models displaying poor predictive accuracy in adults should be re-evaluated for missing drug-specific information that may explain the discrepancy (*i.e.*, additional transporter affinities, alternative clearance pathways, etc.). Scaling to pediatrics should only be considered for adult models found to be in general agreement with observed data. In order to better represent *in vivo* PK, the modeller may opt to refine the adult model based on observed data prior to scaling towards pediatrics. Input parameters, such as lipophilicity and intrinsic clearance, are generally chosen as targets for optimization as a result of uncertainty surrounding literature values of lipophilicity and variability of clearance values between PK studies.

Physiologic scaling also relies on another key assumption in order to extrapolate adult models towards pediatrics: Enzyme metabolism follows first-order kinetics. Validation of this assumption would normally require *in vivo* dose escalation studies in pediatric patients; data which are simply unavailable

for most compounds. Consequently, we are reliant on adult PK data to estimate the likelihood of enzyme saturation occurring among children. For lorazepam, exposure ($AUC_{0 \rightarrow \infty}$) in adults has previously been shown to increase proportionately with intravenous dosage increases from 2 to 4 mg, indicating a lack of saturable metabolism (15).

The derived adult population PK model (Fig. 2a) exhibited an acceptable fit for the majority of observed data with the exception of concentration-time points during the initial phase, post-administration. This lack of fit may be attributed to an unrealistic assumption made by most PK models: Initial mixing of blood in the central compartment is instantaneous. As a result, initial simulated concentration-time points may appear erroneously high as they fail to account for the lag time associated with the introduction of the compound to the systemic circulation and its presence at a peripheral sample site (33). Potential issues related to drug-IV catheter binding may present an alternative explanation for the lack of initial data fit. Reversible binding of drug to the IV catheter would result in delayed administration of the entire dose, resulting in a PK profile similar to a short-term IV infusion as opposed to a bolus dose. Slight over-prediction of initial concentration-time points were also observed in the pediatric population PK model

Fig. 3. Pediatric dose (milligrams per kilogram) required to achieve an equivalent $AUC_{0 \rightarrow \infty}$ of a 2-mg dose in adults. **a** Entire pediatric age-range. **b** Children between 0 and 1 years old

Fig. 4. Mean tissue concentration-time profiles for selected organs among a virtual population ($n=100$) of 2-year-old subjects

(Fig. 2b), a finding which exemplifies the congruent relationship between the adult and pediatric models.

During the first year of life, increasing UGT2B7 enzyme activity corresponds with increases in weight normalized

lorazepam clearance. With clearance being directly proportional to dosage rate, it is not surprising that the dosage (milligrams per kilogram) required to maintain a similar $AUC_{0 \rightarrow \infty}$ as adults after a 2-mg intravenous dose increases during the first year (Fig. 3b). The average dosage requirement in term neonates, where enzyme activity is estimated to be only 5% of adult values, was 0.0037 mg/kg whereas adults received approximately 0.03 mg/kg. For pediatric subjects between the ages of 1 and 3 years, the average weight-normalized clearances exceeded that of adults and, as a result, required increased relative dosages to maintain similar exposure rates. These findings have been corroborated in the literature by studies examining the age-dependence of lorazepam clearance (13,24). Using the process of physiologic scaling as outlined in this study, the increased weight-normalized lorazepam clearance simulated in young children aged 1–3 years was a function of complete hepatic UGT2B7 maturation and a higher liver weight: body weight ratio compared with adults.

Current intravenous dosing recommendations for preoperative administration of lorazepam in adults, 0.044 mg/kg (max 2 mg/dose) 15–20 min before surgery (7), and in infants and children, 0.05 mg/kg (range, 0.02–0.09 mg/kg) (7), were compared with dosage requirements derived from our simulated pediatric populations. While the suggested pediatric dosing range relates to the majority of the simulated children, neonatal dosing (0 days–1 month) does not appear to follow the same regimen. Furthermore, the recommendations provided do not incorporate an age-dependence to dosing. Results from our

Fig. 5. Predictive accuracy plots: Individual AFE values for PBPK model concentration-time predictions for the 63 pediatric patients (plot A), fold error associated PBPK model clearance predictions for the 15 elective patients (plot B), and fold error associated PBPK model volume of distribution predictions for the 15 elective patients (plot C) (dotted line represents 1.5-fold error. Dashed line represents twofold error)

simulations indicate that neonates require substantially lower doses than suggested for infants and children (*i.e.*, 0.05 mg/kg). For example, newborns (0 days old) require approximately one tenth (0.0037 mg/kg) of adult doses whereas 1-month-old neonates require approximately one fourth (0.0077 mg/kg) of adult doses. Accordingly, dosing recommendations derived from population PBPK models circumvent the use of inexact dosing ranges by assigning a unique dosage to particular age groups.

The utility of PBPK models at estimating concentrations values within specific organs is demonstrated in Fig. 4. These estimates highlight the potential future of PBPK models to create targeted dosing regimens based on *a priori* estimates of drug distribution within specific compartments of interest (*i.e.*, site of action). The plasma concentration simulated by the model is reflective of the overall mass-transfer between all organs. As a result, larger organs to which the drug permeates into (*i.e.*, muscle and adipose) will play a larger role in influencing the overall plasma profile compared with smaller organs (brain). By simply evaluating the PBPK model estimates using plasma concentration-time data, we are unable to confirm the reliability of individual organ estimates, especially smaller organs which only influence plasma levels minutely. Consequently, in order to increase reliability in specific compartment estimates, experimental data assessing tissue concentration or effect biomarkers that can link to target concentrations are inherently needed.

Using the presented workflow, retrospective assessment of the derived pediatric model may only take place once sufficient observed data are available. To evaluate our pediatric lorazepam model, concentration-time data from a cohort of 63 pediatric patients was obtained (30). For the majority of sample patients (approximately 73%), PBPK model concentration estimates were within a 1.5-fold prediction error on average. Additionally, the predictive performance of the pediatric PBPK model at estimating CL and V_{ss} was determined among elective patients ($n=15$) included in Chamberlain *et al.*'s study (30). For CL, 60% of model estimates were within 1.5-fold deviation from observed values. Comparatively, model estimates of V_{ss} were associated with a greater predictive accuracy with 80% of estimates lying within 1.5-fold error.

When attempting to estimate individual subject parameters (*i.e.*, CL and V_{ss}), PBPK models normally incorporate superficial biometric data such as age, weight, and height, to derive a complete physiologic/anatomic depiction of the subject. All physiologic/anatomic values (*i.e.*, organ weights, percent enzyme activity) denote values that would be expected from a mean individual of a particular age, weight, and height. What this approach does not account for is the intrinsic (intra-patient) variability that underpins biological research (*i.e.*, not all 8-year-olds, weighing 35 kg, and measuring 120 cm will have the same organ weights). Consequently, the accuracy of individualized PBPK model-derived parameter estimates will be affected by the magnitude of the inherent intra-subject variability that exists. Data-driven population PK estimation techniques represent an alternative approach to estimating individualized PK parameters. Chamberlain *et al.* (30) found no covariates which were statistically significant to necessitate inclusion into their final PK model. Subsequently, estimates of CL and V_{ss} were predicted using 0.14 L/h/kg^{0.75} and 1.37 L/kg, respectively.

When used to estimate individual PK parameters among the elective group ($n=15$), this method predicted 47%, 67%, and 80% of CL values within a 1.25-, 1.5-, and 2-fold deviation from observed values, respectively. For V_{ss} , 33%, 87%, and 100% of estimates fell within 1.25-, 1.5-, and 2-fold error, respectively. These values are quite similar to ones obtained *via* PBPK modelling. One major difference being that PBPK model estimates were generated independent of PK data from children whereas Chamberlain *et al.*'s study derived estimates based on data obtained through *in vivo* experimentation with pediatric subjects. As such, PBPK modelling provided a relatively accurate approach for estimating individualized PK parameters compared with the estimates generated by Chamberlain *et al.* Despite the previous finding, a large proportion of subjects had CL and V_{ss} estimates associated with greater than 1.25-fold error which indicates, in an absolute sense, individualized predictions generated through PBPK modelling still requires further development.

With predictions directed toward a mean individual, it not surprising that PBPK modelling can be used to estimate average parameter values among specific populations. For 14 of the 15 pediatric subjects among the elective group greater than the age of 5 years, the PBPK model produced mean estimates of CL and V_{ss} that exhibited a 14% and -6% relative error, respectively, from the mean observed values calculated by NCA. One patient who was approximately 1 year old was excluded from this analysis as the mean clearance was adjudged to be significantly different than that of the rest of the group, which was comprised of older children. This assertion was based on the increased dosing requirements for children between the ages of 1 and 3 years due to increased weight normalized clearances, as depicted in Fig. 3a, b.

PBPK models also display a unique ability to estimate the magnitude of variability that surrounds PK parameters. Many PBPK modelling applications are capable of generating virtual populations in order to estimate PK variability associated with anthropometric differences between individuals. These populations can also be manipulated to encompass the range of intra-individual variability that exists around each physiological/anatomical value. As such, PBPK population-based modelling techniques may be used to provide *a priori* estimates of variability associated with PK parameters such as CL and V_{ss} for a given population. For example, the coefficient of variation associated with observed clearance values, determined by NCA, for 14 of the 15 elective group patients greater than 5 years of age was 53%. To generate an *a priori* estimate of variability using PBPK modelling, a uniform virtual population of 1,300 children between the ages of 5 and 17 years (100 children per year) was created with associated variability around organ sizes, tissue blood flows, and metabolizing enzyme activity. The coefficient of variation of associated with PBPK model estimates of clearance among this population was 43%. Although this estimate under-predicts the observed variability, it does provide a preliminary estimate of the magnitude of variation that may be observed within the population. Estimates of high degrees of CL and V_{ss} variability among a population for drugs with narrow therapeutic indexes may indicate the need for individualized dosing regimens whereas drugs with wider therapeutic indexes may be administered using fixed dosing levels (*i.e.*, 60 mg for 1–2-year-olds, 80 mg for 2–5-year-olds, etc.).

Although evaluation of the derived PBPK model represents a crucial step in the workflow process, a clear metric has yet to be proposed to aid researchers in classifying a particular model as “good” versus “bad”. As such, acceptable tolerances of predictive performance of the pediatric model should be established on a drug-by-drug basis based on current information available in adults: PK–PD relationship, safety profile, and therapeutic index.

The derived pediatric population PBPK model was able to account for approximately 72% of the variability associated with concentration–time points observed in a subset of 40 pediatric patients. The inability of the model to fully account for the variability observed is likely a result of the uniform nature of age groups included in the simulated population. One thousand one hundred forty pediatric patients, between the ages of 0 days and 18 years, were simulated with 60 patients being selected per year. As a result, PK variability associated with the simulated population was skewed toward the large majority of patients whose clearance values were similar to that of adults (*i.e.*, patients greater than 3 years of age). Patients with relatively lower and higher clearance values, neonates, and children aged 1–3 years, respectively, comprised a small portion of the simulated population and, therefore, only minimally contributed to the PK variability amongst the total population. Consequently, the total range of simulated variability may have been understated with use of a uniformly aged population.

CONCLUSION

Since lorazepam’s inclusion on NIH’s priority list in 2003, two clinical pediatric studies have been completed under the sponsorship of National Institute of Child Health and Human Development with one additional study in the recruitment phase. With future advancements in model development, there is the potential for PBPK models to shift from being used as a compliment to clinical PK studies to a partial replacement. For example, PBPK models may be used to decrease the amount of clinical trials required in children by functioning as the primary exploratory investigation of drug PK. This could potentially eliminate the need for exploratory PK studies in children and permit clinical investigations to function on a confirmatory basis. With the recent FDA recommendation, interest in the use and development of pediatric PBPK models will inevitably increase. The current study demonstrates the fundamental processes required for development of a pediatric PBPK model incorporating existing adult drug data. Using this approach, the model was able to predict lorazepam PK in children as a function of age. With the rise in use of PBPK modelling software in the field of pediatric drug development, the use of procedural workflows, similar to the one presented, will ensure consistency in model outputs and potentially permit for expedited reviews of such research by regulatory bodies.

ACKNOWLEDGMENTS

This work was funded by the Natural Sciences and Engineering Research Council (NSERC) of Canada.

REFERENCES

1. Leong R, Vieira ML, Zhao P, Mulugeta Y, Lee CS, Huang SM, *et al.* Regulatory experience with physiologically based pharmacokinetic modeling for pediatric drug trials. *Clin Pharmacol Ther.* 2012;91(5):926–31.
2. Bjorkman S. Prediction of drug disposition in infants and children by means of physiologically based pharmacokinetic (PBPK) modelling: theophylline and midazolam as model drugs. *Br J Clin Pharmacol.* 2005;59(6):691–704.
3. Edginton AN, Schmitt W, Willmann S. Development and evaluation of a generic physiologically based pharmacokinetic model for children. *Clin Pharmacokinet.* 2006;45(10):1013–34.
4. Johnson TN, Rostami-Hodjegan A. Resurgence in the use of physiologically based pharmacokinetic models in pediatric clinical pharmacology: parallel shift in incorporating the knowledge of biological elements and increased applicability to drug development and clinical practice. *Paediatr Anaesth.* 2011;21(3):291–301.
5. Edginton AN, Ritter L. Predicting plasma concentrations of bisphenol A in children younger than 2 years of age after typical feeding schedules, using a physiologically based toxicokinetic model. *Environ Health Perspect.* 2009;117(4):645–52.
6. Barrett JS, Della Casa AO, Laer S, Meibohm B. Physiologically based pharmacokinetic (PBPK) modeling in children. *Clin Pharmacol Ther.* 2012;92(1):40–9.
7. Lexi-Comp Online (2012) Pediatric & neonatal Lexi-drugs online. Hudson, Ohio, Lexi-Comp, Inc. 18-6-2012. Ref Type: Online Source.
8. Kumar P, Walker JK, Hurt KM, Bennett KM, Grosshans N, Fotis MA. Medication use in the neonatal intensive care unit: current patterns and off-label use of parenteral medications. *J Pediatr.* 2008;152(3):412–5.
9. Twite MD, Rashid A, Zuk J, Friesen RH. Sedation, analgesia, and neuromuscular blockade in the pediatric intensive care unit: survey of fellowship training programs. *Pediatr Crit Care Med.* 2004;5(6):521–32.
10. Federal Register (2003) List of drugs for which pediatric studies are needed. *Federal Register* 2003 Jan 9;68(13):2789–90.
11. Chin PK, Jensen BP, Larsen HS, Begg EJ. Adult age and *ex vivo* protein binding of lorazepam, oxazepam and temazepam in healthy subjects. *Br J Clin Pharmacol.* 2011;72(6):985–9.
12. Divoll M, Greenblatt DJ. Effect of age and sex on lorazepam protein binding. *J Pharm Pharmacol.* 1982;34(2):122–3.
13. Crom WR, Relling MV, Christensen ML, Rivera GK, Evans WE. Age-related differences in hepatic drug clearance in children: studies with lorazepam and antipyrine. *Clin Pharmacol Ther.* 1991;50(2):132–40.
14. Greenblatt DJ, Schillings RT, Kyriakopoulos AA, Shader RI, Sisenwine SF, Knowles JA, *et al.* Clinical pharmacokinetics of lorazepam. I. Absorption and disposition of oral 14C-lorazepam. *Clin Pharmacol Ther.* 1976;20(3):329–41.
15. Greenblatt DJ, Shader RI, Franke K, MacLaughlin DS, Harmatz JS, Allen MD, *et al.* Pharmacokinetics and bioavailability of intravenous, intramuscular, and oral lorazepam in humans. *J Pharm Sci.* 1979;68(1):57–63.
16. Verbeeck R, Tjandramaga TB, Verberckmoes R, De Schepper PJ. Biotransformation and excretion of lorazepam in patients with chronic renal failure. *Br J Clin Pharmacol.* 1976;3(6):1033–9.
17. Rodgers T, Rowland M. Physiologically based pharmacokinetic modelling 2: predicting the tissue distribution of acids, very weak bases, neutrals and zwitterions. *J Pharm Sci.* 2006;95(6):1238–57.
18. Rodgers T, Leahy D, Rowland M. Physiologically based pharmacokinetic modeling 1: predicting the tissue distribution of moderate-to-strong bases. *J Pharm Sci.* 2005;94(6):1259–76.
19. Rodgers T, Leahy D, Rowland M. Tissue distribution of basic drugs: accounting for enantiomeric, compound and regional differences amongst beta-blocking drugs in rat. *J Pharm Sci.* 2005;94(6):1237–48.
20. Greenblatt DJ, Comer WH, Elliott HW, Shader RI, Knowles JA, Ruelius HW. Clinical pharmacokinetics of lorazepam. III. Intravenous injection. Preliminary results. *J Clin Pharmacol.* 1977;17(8–9):490–4.

21. Greenblatt DJ, Divoll M, Harmatz JS, Shader RI. Pharmacokinetic comparison of sublingual lorazepam with intravenous, intramuscular, and oral lorazepam. *J Pharm Sci.* 1982;71(2):248–52.
22. Wermeling DP, Miller JL, Archer SM, Manaligod JM, Rudy AC. Bioavailability and pharmacokinetics of lorazepam after intranasal, intravenous, and intramuscular administration. *J Clin Pharmacol.* 2001;41(11):1225–31.
23. Willmann S, Hohn K, Edginton A, Sevestre M, Solodenko J, Weiss W, *et al.* Development of a physiology-based whole-body population model for assessing the influence of individual variability on the pharmacokinetics of drugs. *J Pharmacokinet Pharmacodyn.* 2007;34(3):401–31.
24. Edginton AN, Schmitt W, Voith B, Willmann S. A mechanistic approach for the scaling of clearance in children. *Clin Pharmacokinet.* 2006;45(7):683–704.
25. Sim SM, Back DJ, Breckenridge AM. The effect of various drugs on the glucuronidation of zidovudine (azidothymidine; AZT) by human liver microsomes. *Br J Clin Pharmacol.* 1991;32(1):17–21.
26. McNamara PJ, Alcorn J. Protein binding predictions in infants. *AAPS PharmSci.* 2002;4(1):E4.
27. Alcorn J, McNamara PJ. Ontogeny of hepatic and renal systemic clearance pathways in infants: part II. *Clin Pharmacokinet.* 2002;41(13):1077–94.
28. Irwin JJ, Kirchner JT. Anemia in children. *Am Fam Physician.* 2001;64(8):1379–87.
29. Hayton WL. Maturation and growth of renal function: dosing renally cleared drugs in children. *AAPS PharmSci.* 2000;2(1):E3.
30. Chamberlain JM, Capparelli EV, Brown KM, Vance CW, Lillis K, Mahajan P, *et al.* Pharmacokinetics of intravenous lorazepam in pediatric patients with and without status epilepticus. *J Pediatr* 2011;160:667–672; Nov 1.
31. Zhao P. Regulatory experience in physiologically-based pharmacokinetic (PBPK) models in pediatric submissions. FDA—Advisory Committee for Pharmaceutical Science and Clinical Pharmacology. 14-3-2012. Conference Presentation.
32. Edginton AN. Knowledge-driven approaches for the guidance of first-in-children dosing. *Paediatr Anaesth.* 2011;21(3):206–13.
33. Chiou WL. Potential pitfalls in the conventional pharmacokinetic studies: effects of the initial mixing of drug in blood and the pulmonary first-pass elimination. *J Pharmacokinet Biopharm.* 1979;7(5):527–36.
34. Pyka A, Babuska M, Zachariasz M. A comparison of theoretical methods of calculation of partition coefficients for selected drugs. *Acta Pol Pharm.* 2006;63(3):159–67.
35. Popovic GV, Sladic DM, Stefanovic VM, Pfendt LB. Study on protolytic equilibria of lorazepam and oxazepam by UV and NMR spectroscopy. *J Pharm Biomed Anal.* 2003;31(4):693–9.
36. Boxenbaum H. Comparative pharmacokinetics of benzodiazepines in dog and man. *J Pharmacokinet Biopharm.* 1982;10(4):411–26.