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Using ontogeny information to build predictive models for drug elimination

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Our incomplete understanding of the developmental maturation of drug elimination mechanisms poses a serious challenge to paediatric dosage regimen design and toxicological risk assessment. The dynamic and variable nature of maturation also limits our ability to acquire pharmacokinetic data in all relevant paediatric populations. However, recent attempts to use the available human ontogeny data to build predictive models of paediatric drug elimination hold promise to assist dosage regimen design and risk assessment. This review identifies population pharmacokinetic, allometric scaling and physiologically based clearance scaling models as principal approaches to estimate paediatric systemic clearance in the absence of comprehensive age-group-specific data.

Drug dosage regimen design and toxicological assessment in paediatric populations, especially those under the age of 2 years, remain significant challenges in clinical care [1–3] and environmental risk assessment [4–5]. Overcoming such challenges will require a comprehensive understanding of the developmental maturation of drug pharmacokinetic processes, in particular systemic clearance mechanisms. Recent efforts of regulatory agencies, paediatric pharmacologists and researchers have improved the available paediatric age-group-specific pharmacokinetic information [3,6–10]. However, the overwhelming logistics, cost and ethics associated with well-controlled clinical trials or toxicological assessments in paediatric age groups [3,11] will severely limit their use for many drugs. A possible aid to the well-controlled clinical trial has emerged with the latest advancements in predictive modeling approaches. Such approaches hold promise to assist in trial design and help limit the number of samples and costs associated with paediatric drug trials. This article provides a review of available modeling methods for the estimation or prediction of drug elimination in paediatric populations as age-dependent differences in drug dosages and toxicological risk often relate to developmental maturation of drug elimination processes.

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Drug clearance as the principle PK process determining age-dependent differences in drug dosage regimens and toxicological risk assessments

Previously, drug therapy or toxicological assessment in paediatric populations considered only the proximate linear relationship between postnatal growth and body weight. Hence, exposure dose was adjusted on the basis of age-dependent differences in body weight or body surface area [12,13]. At present, we premise such assessments (when possible) upon the relationship between plasma drug concentration and response owing to the knowledge that the dose–response relationship varies with age. In addition to the size of the bioavailable dose, the total exposure to a drug (as measured by the metrics area-under-the-plasma-concentration-versus-time curve, AUC, or steady state plasma concentration, C_{SS}, following single or multiple doses, respectively) is determined by the efficiency of the elimination mechanisms as measured by systemic clearance (Cl_S) (Eq. (1)).

$$AUC_0^\infty = \frac{F \times \text{Dose}}{Cl_S} \quad \text{or} \quad C_{SS} = \frac{(F \times \text{Dose})/\tau}{Cl_S} \tag{1}$$

Age-dependent differences in bioavailability (F) and Cl_S usually account for differences in plasma concentrations and exposure metrics (e.g. AUC_0^∞, C_{SS}) between paediatric age groups. Simple adjustments of exposure dose on per kilogram basis (or surface area) in no way considers the developmental changes as a

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result of both growth and maturation of the physiological and biochemical processes governing F and Cl_S.

Approaches for estimation of systemic clearance in paediatric populations

The availability of methods to predict or estimate paediatric clearance is crucial for appropriate paediatric dosage regimen design [14] and environmental risk assessment [5,12]. Since many drug exposures follow from oral administrations, apparent clearance (the ratio of systemic clearance and bioavailability (Cl_S/F)) becomes important in the estimation of therapeutic doses or for risk assessments. However, modeling approaches for apparent clearance predictions among human paediatric age groups has received little attention [15,16]. Population PK approaches may allow estimates of apparent clearance [15], but most attempts to predict this PK parameter use allometric models and interspecies PK data [17]. The limited *in vivo* bioavailability data in paediatric age groups and our poor understanding of the developmental maturation of physiological factors that determine drug bioavailability (i.e. gastrointestinal blood flow, pH, gastric and intestinal motility, intestinal and hepatic first-pass mechanisms) represent major stumbling blocks. Consequently, the following section focuses on the major approaches for systemic clearance estimation or prediction in paediatric populations.

Population pharmacokinetic approaches

Ethical and technical barriers often preclude the collection of complete plasma drug concentration versus time information from a single paediatric patient [14]. Sparse data sampling techniques and population PK approaches often replace the classical PK study in paediatric drug trials. With population PK approaches only two to three blood samples collected at random intervals following a dosage administration are required from a single paediatric patient. Samples collected from numerous different individuals are then utilized simultaneously to provide an overall composite plasma drug concentration versus time profile [18]. These data are fitted using Bayesian algorithms [13,18] to provide estimates of population or individual PK parameters [14,15]. Population PK approaches can consider variation associated with gender, age and other variables (e.g. renal function status, other medications, among others). In addition, logistic regression can assess the relationship between the calculated exposure metrics (e.g. AUC_0^∞, $C_{SS,ave}$) and pharmacological effect to allow for dosage regimen design [8,19]. A recent variation on the population PK approach allowed for sequential updating of both population and individual PK parameters on the basis of new individual plasma concentration data from the treated individual [20]. According to the author, this approach may show promise as a therapeutic support tool for drugs requiring individualization of dosage regimens [20].

Although population PK modeling is widely used in paediatric PK studies, several issues unique to paediatric populations continue to challenge this modeling approach [21]. Unlike models applied to adult populations, paediatric population PK models must consider a method for body size correction in the analysis of paediatric data, the limited sampling strategy (and its validation) to be used in the paediatric PK study, and how to integrate prior knowledge (both adult and paediatric data) into the population PK model [21]. Furthermore, the population approach is limited to drugs with prior use in the paediatric population as it requires *in vivo* pharmacokinetic data from the appropriate paediatric population to build the population model. This approach has no practical value in clinical drug development.

Scaling approaches

The pharmacokinetic differences observed among paediatric age groups and with adult populations largely relate to the anatomical, physiological and biochemical changes that occur during postnatal development through to adulthood. The scaling approaches, to a variable degree, combine existing knowledge of adult PK with a consideration of the impact of such developmental changes on paediatric PK estimates [16,22–27] to elaborate predictive models for systemic clearance by scaling 'adult parameters' to the paediatric patient.

Allometric scaling

In general terms, allometric scaling is an empirical technique used to relate a physiological function to body size. Allometric scaling as applied to the prediction of paediatric PK parameters (such as systemic clearance) involves an extrapolation of adult data (as a starting point) to paediatric populations on the basis of the relationship of the PK parameter to a body size parameter, usually body weight or body surface area [28,29]. Eq. (2) shows a general allometric scaling model for estimation of paediatric PK parameters on the basis of the known adult PK parameter estimate and some power function (or exponent) of body weight (BW) [13]. The allometric exponent, b, typically assumes a value of 1 (when the parameter is directly proportional to age-dependent changes in body weight), 3/4 (when the PK parameter is related to age-related changes in physiological processes), or 2/3 (when the PK parameter is approximately proportional to age-dependent changes in body surface area) [23,29].

$$\text{PK parameter}_{\text{paediatric}} = \text{PK parameter}_{\text{adult}} \left(\frac{\text{BW}_{\text{paediatric}}}{\text{BW}_{\text{adult}}} \right)^{b} \qquad (2)$$

The allometric exponents have no physiological meaning [28] and, hence, cannot *a priori* account for the influence of growth and developmental maturation on PK processes. Furthermore, this method considers developmental changes in body size owing to normal growth processes but does not accommodate maturation of specific biochemical processes, particularly drug transporter and metabolic enzyme capacity, which in turn influence intrinsic clearance. Since organ or body size alone do not trigger the regulation of these gene products (i.e. transporters and enzymes) then it is unlikely that a ready correlation would exist between size and PK. Several studies that critically examined the ability of simple allometric scaling to provide reasonable estimates for neonatal and infant systemic clearance suggest that the complexity of the maturation process precludes a simple extrapolation of adult parameters to paediatric parameters [23,28,30] and found this approach unreliable until disposition processes (i.e. CYP enzyme activity) begin to approach adult levels [22,31].

Physiologically based clearance scaling modeling

Physiologically based clearance scaling models are mechanism-based approaches for predictions of drug elimination. These mod-

els consider the underlying physiological and biochemical processes that govern drug elimination to predict systemic clearance [7,22,26] in paediatric populations. The more generalized physiologically based pharmacokinetic models provide descriptions of drug absorption and disposition, which lies beyond the scope of this review [12,27,32–35]. Although such models require a tremendous input of physiological data [10,27,33,35,36], their advantage over other modeling approaches is the ability to incorporate more specific information on the ontogeny of the various anatomical, physiological and biochemical processes governing drug PK and elimination to allow predictions in different paediatric age groups [12,27,34,35].

For many drugs, systemic clearance is an additive function of hepatic (Cl_H) and renal (Cl_R) elimination mechanisms. Fundamentally, these models involve a scaling of known adult Cl_H and Cl_R data to predict systemic clearance in different paediatric age groups. Such scaling is achieved by the use of some 'scaling factor' that is based on known ontogeny data relative to adult values (i.e. enzyme activity as a percent of adult activity) available for the different clearance mechanisms. With regards to Cl_H, biotransformation and biliary excretion are the principal mechanisms of hepatic elimination. Almost no information is available on the ontogeny of biliary excretion mechanisms. Hence, most physiologically based clearance scaling models almost always preclude this hepatic elimination mechanism from consideration in their estimation of paediatric systemic clearance.

Hepatic clearance

Most existing data on the developmental maturation of human hepatic biotransformation pathways comes from *in vitro* hepatic microsomal studies of cytochrome P450 (CYP) enzyme-mediated activity [9,25]. Although still limited, recent efforts have improved the available ontogeny data for other Phase I enzymes (such as the flavin-containing monooxygenases, epoxide hydrolase, alcohol dehydrogenase) [37,38], as well as Phase II enzymes, such as the UDP-glucuronosyltransferases, sulfotransferases, glutathione-*S*-transferases and *N*-acetyltransferases [6,26,39–41]. Hence, physiologically based clearance scaling approaches usually apply to CYP data, since most drugs are CYP substrates. However, these approaches can be applied to other Phase I and Phase II enzymes and hepatic transport systems when ontogeny information is available [26,42].

The process involves an *in vitro–in vivo* extrapolation of enzyme activity data determined in hepatic microsomal preparations from different paediatric age groups as well as the adult [7,22,26,32,33]. Alternatively, an estimation of *in vivo* clearance extrapolated from enzyme levels in the liver (rather than enzyme activity) has been reported [27,43]. Extrapolation of microsomal enzyme activity data requires the assumption that uptake into the hepatocyte and rate of drug delivery to the hepatocyte (as defined by hepatic blood flow) are not rate-limiting to overall hepatic elimination of the drug. When hepatocellular uptake is rate-limiting, then the extrapolation process must consider these events.

Hepatic enzyme function is often expressed as intrinsic clearance (Cl_{int}), the ratio of the Michaelis–Menten constants, V_{Max} and K_M, under linear conditions. If CYP enzyme affinity (K_M) remains constant during postnatal development then intrinsic clearance reduces to maximum enzyme activity (V_{Max}) (expressed as activity per milligram microsomal protein) for a particular enzyme pathway. A scaling factor is then constructed from the product of the age-dependent V_{max} values, amount of microsomal protein yielded from a gram of liver, and the ratio of total liver and body weight for a particular paediatric age group. This is subsequently normalized to a similarly constructed scaling factor on the basis of adult parameters. This normalized scaling factor then scales a known adult *in vivo* intrinsic clearance value ($Cl^{adult}_{int(j)}$) to *in vivo* intrinsic clearance in the paediatric patient ($Cl^{paediatric}_{int,t}$). This is represented in Eq. (3), where Cl_{int} is intrinsic clearance and SF is a scaling factor specific for a pathway of elimination (j) and age of the paediatric patient (t). For a particular drug, several CYP enzymes may contribute to its overall elimination. The scaling factor then becomes a sum of all contributing pathways of elimination. Hence, in Eq. (3), $Cl^{paediatric}_{(t)}$ is a measure of the total metabolic enzyme activity towards a drug and represents the sum ($\sum_{j=1}^{n}$) of all individual pathways of metabolism (j) mediating the hepatic elimination of that drug. Eq. (3) can incorporate the contribution of any elimination mechanism (i.e. biliary excretion, Phase II metabolism, hepatocellular uptake or efflux transporters) to Cl_{int} if appropriate ontogeny data is available. Furthermore, adult estimates of intrinsic clearance ($Cl^{adult}_{int(j)}$) are often attainable from the literature as well as the enzymes involved and their relative contribution to total drug elimination [7,16,22,26].

$$Cl^{paediatric}_{int,t} = Cl^{adult}_{int(j)} \times \sum_{j=1}^{n} SF_{(j,t)} \quad (3)$$

The estimated intrinsic clearance value for the paediatric age group ($Cl^{paediatric}_{int,t}$) is then incorporated into a model of hepatic clearance to provide an estimate of hepatic clearance. Often the well-stirred model of hepatic clearance (Eq. (4)) is used as a simple model expressing the inter-relationship between the three physiological determinants of Cl_H, namely hepatic blood flow (Q_H), intrinsic clearance (Cl_{int}) and unbound fraction in the blood binding ($f_{u(b)}$).

$$Cl_H = \frac{Q_H \times f_{u(b)} \times Cl_{int}}{Q_H + f_{u(b)} Cl_{int}} \quad (4)$$

Use of the well-stirred model requires a consideration of the age-related changes in Q_H and $f_{u(b)}$ [44] to attain *in vivo* estimates of hepatic metabolic drug clearance in specific paediatric age groups. Age-dependent changes in $f_{u(b)}$ may be estimated for a particular paediatric age group on the basis of known paediatric plasma protein binding ontogeny data [44]. This approach scales the unbound fraction from adult to paediatric subjects on the basis of a correlation between unbound fraction and the relative levels of the major plasma binding proteins (albumin and α_1-acid glycoprotein) assuming a constant value for binding affinity between adults and paediatric subjects [44].

Developmental changes in Q_H are poorly understood. Age-dependent changes in Q_H may influence hepatic clearance when hepatic elimination is limited by the rate of hepatic drug delivery and not enzyme activity [10]. With blood flow rate limited drugs, developmental changes in metabolic enzymes likely will impact Cl_H only when enzyme activity is exceedingly underdeveloped (such that enzyme activity becomes the rate-limiting factor to overall drug elimination). Despite this concern, many drugs exhi-

bit relatively low hepatic extraction ratios and liver blood flow has limited impact on their hepatic clearance (i.e. the unbound fraction and intrinsic clearance noted in Eq. (4) are the principal determinants of *in vivo* hepatic clearance). Hence, the use of an adult estimate for Q_H or an allometric scaling model for Q_H [22,33] should have minimal impact on *in vivo* Cl_H determinations in physiologically based clearance scaling modeling approaches. However, use of an adult parameter or allometric scaling model for Q_H may not be appropriate for newborns since a patent *ductus venosus* will shunt a significant portion of hepatic portal blood flow away from the functioning hepatocytes in the first week or two of life [45].

The use of physiologically based clearance scaling places a significant burden on the reliability of the reported adult estimates for both intrinsic clearance and the relative contribution of each clearance mechanism to systemic clearance, since the paediatric estimate is scaled directly from the adult estimate. This becomes particularly problematic when important metabolism pathways in paediatric populations have only minor contributions to overall drug elimination in adults [12]. Since the approach depends upon an *in vitro–in vivo* extrapolation of enzyme activity data to predict *in vivo* elimination characteristics, *in vitro* enzyme activity estimates must also have reasonable reliability. Such assessments can depend markedly on the probe substrate. The available data on CYP enzyme ontogeny can show variable results with respect to the rate and pattern of individual CYP enzyme development depending upon probe substrate [22,46,47]. Additionally, several *in vivo* studies examining the maturation of specific enzyme-mediated reactions [48–50] report limited age-dependent changes despite significant changes noted in *in vitro* evaluations. This discrepancy may relate to issues with probe substrate specificity but could also associate with the significant postnatal developmental changes of the liver in general. Quantitative and qualitative changes in cell number, volume, distribution and function occur during hepatic maturation [25] as well as changes in the hepatocellular distribution and expression of drug metabolizing enzymes (i.e. CYP enzymes show homogeneous distribution in fetal and early neonatal life with increasing focal distribution in the perivenous hepatocytes of the hepatic acinus) [46,51]. Hepatic microsomal preparations cannot reflect these developmental changes *in vivo*. Such factors would complicate predictions of Cl_H particularly during the early postnatal period using estimates of *in vitro* intrinsic clearance that are based on hepatic microsomal activity data.

When used to provide predictions of systemic clearance, physiologically based clearance scaling approaches can account for the known interindividual variation in the physiological components that determine *in vivo* hepatic clearance [14]. For example, the Simcyp® software (Simcyp Clearance and Interaction Simulator®) can simulate *in vivo* unbound metabolic clearances from *in vitro* data by using Monte Carlo methods that integrate genetic, anatomical, physiological, demographic and clinical attributes of the population with information on developmental physiology and ontogeny of systemic clearance mechanisms that can influence the *in vitro–in vivo* extrapolation [16]. The paediatric physiological based pharmacokinetic model, Simcyp® Paediatric, simulated 2000 virtual paediatric patients and compared these simulated values to values reported in the literature from *in vivo* clinical trials [14]. The overall predictability of the algorithm resulted in >70% of the predicted unbound clearance values to be within twofold of the actual reported values [14], a clear indication of the software's ability to capture population variability.

Renal clearance

Similar approaches are applied to the scaling of Cl_R from adult values to paediatric populations, since renal elimination depends on the maturation of several physiological parameters such as renal blood flow, glomerular filtration and renal tubular function [7,26]. For most drugs, developmental changes in glomerular filtration (GFR) and tubular secretion (TS) largely determine age-dependent changes in Cl_R [24]. Renal elimination mechanisms in the paediatric population are estimated from adult values by incorporation of scaling factors (SF) that consider the developmental of maturation glomerular filtration and tubular secretion [7,22,24,26], and changes in the unbound fraction [44] relative to the adult value as shown in Eq. (5).

$$Cl_{RM}^{paediatric} = \frac{RM_t^{paediatric}}{RM^{adult}} \times \frac{f_{u(b)t}^{paediatric}}{f_{u(b)}^{adult}} \times Cl_{RM}^{adult} = SF_{(RM,t)} \times Cl_{RM}^{adult} \quad (5)$$

The RM refers to the renal elimination mechanism [glomerular filtration (GFR) or tubular secretion (TS)], $f_{u(b)}$ is the unbound fraction of drug in the blood, t is the particular paediatric age, and Cl_{RM}^{adult} is the renal clearance owing to the particular renal elimination mechanism in the adult. Hayton [24] elaborated an allometric model, which separates the contribution of maturation and of growth on renal function parameters, to describe the ontogeny of glomerular filtration and tubular secretion [24]. This allometric model provides estimates for $RM_t^{paediatric}$ (where RM is either GFR or TS). Normalization of $RM_t^{paediatric}$ to the adult value for the particular renal clearance mechanism (RM^{adult}), with consideration of age-dependent changes in plasma protein binding, results in a scaling factor ($SF_{(RM,t)}$) that allows a prediction of renal clearance (owing to GFR or TS) based upon known adult clearance values (Cl_{RM}^{adult}).

Systemic clearance

Since renal and/or hepatic elimination mechanisms principally determine the systemic clearance of most drugs, systemic clearance ($Cl_{S(t)}^{paediatric}$) then becomes an additive function of all elimination pathways contributing to the irreversible removal of drug from the body as shown in Eq. (6). According to physiologically based clearance scaling approaches, systemic clearance at a paediatric age, t ($Cl_{S(t)}^{paediatric}$), is estimated by scaling the respective known adult estimates for all contributing clearance pathways.

$$Cl_{S(t)}^{paediatric} = SF_{(GFR,t)} \times Cl_{(GFR)}^{adult} + SF_{(TS,t)} \times Cl_{(TS)}^{adult} + \sum_{j=1}^{n} SF_{(j,t)} \times Cl_{int(j)}^{adult} \quad (6)$$

Recently, an integrated systems approach for systemic clearance expanded on the existing scaling approaches for prediction of systemic clearance [26]. The integrated systems approach uses both *in vitro* enzyme ontogeny data and *in vivo* clearance data in paediatric subjects for drugs primarily eliminated via one mechanism to build age-dependent, elimination pathway-specific scaling factors [26]. The approach models the age-dependent

changes in renal clearance (owing to glomerular filtration and tubular secretion), biliary clearance and hepatic clearance owing to Phase I and II enzymes providing the most comprehensive physiologically based clearance scaling modeling approach for estimation of systemic drug clearance in paediatric populations. Model predictions of systemic clearance correlated highly with observed data, with approximately 83% of estimates within 50% of the observed systemic clearance values in all paediatric age groups including premature neonates.

In general, comparisons of predicted clearance estimates from physiologically based clearance scaling model approaches with observed clinical data highly supports their potential use in toxicological risk assessments to determine the impact of age dependent changes in PK processes on the dose received and plasma concentration profiles in paediatric age groups [12,14,22,26,27,33–35,52]. As well, the paediatric physiologically based clearance scaling models provide better estimates of drug clearance in paediatric age groups relative to allometric scaling models owing to incorporation of age-related differences in the physiological variables that determine drug disposition [7,14,16,22,33].

General limitations

Our relatively poor understanding of the developmental maturation of the organs, and the physiological and biochemical processes governing systemic clearance mechanisms poses a serious limitation to current ontogeny models. Most of the available information emerges from a limited number of published studies that often involve disease patients, incorporate a small number of subjects, fail to evaluate discrete age groups, use probe substrates without sufficient specificity to describe enzyme-specific ontogeny leading to tremendous variability in the reported data, and do not generally consider genotypic and environmental variation in the population [9]. Models elaborated from such data will fail to capture population variability adequately, as well as discrete differences across the paediatric age groups, thereby reducing the robustness of possible predictions. In addition to the limited ontogeny data, few pharmacokinetic data is available in young paediatric age groups [27,53]. This poses an important limitation since all modeling approaches require *in vivo* systemic clearance data to draw comparisons between observed and model predicted estimates for model validation purposes.

For many drugs a relationship (whether direct or indirect) exists between plasma concentrations and response. When the relationship between paediatric age groups and adult populations is similar the various modeling approaches for predicting paediatric systemic clearance from known adult data can provide important information towards dosage determination and risk assessment. When the plasma concentration-response profiles are dissimilar effective predictions require additional knowledge of the pharmacodynamics of the drug in paediatric populations relative to the adult. For some drugs, *in vivo* clinical trial data suggests pharmacodynamic differences between the age groups and the process of maturation can lead to 'windows of susceptibility' at certain developmental periods leading to different drug responses [5,12,54–57].

Conclusions

Despite limitations, modeling approaches that incorporate known ontogeny data provide a scientific approach towards predictive systemic clearance estimation in different paediatric age groups. These models hold promise to improve dosage regimen design or selection, facilitate the design of paediatric clinical studies, minimize cost and time for paediatric dose determination, and aid in risk assessment. Although these approaches cannot replace the carefully designed paediatric clinical study, rapid postnatal developmental maturation enormously complicates the conduct of such classical PK studies and few drugs will ever undergo a comprehensive *in vivo* evaluation in all relevant paediatric age groups. Continued research in this area will bridge knowledge gaps that will iteratively improve and refine the available modeling approaches advancing their application in risk assessment and dosage determination in paediatric populations.

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References

1 Cuzzolin, L. *et al.* (2006) Off-label and unlicensed prescribing for newborns and children in different settings: a review of the literature and a consideration about drug safety. *Expert Opin. Drug Saf.* 5, 703–718
2 Choonara, I. (2004) Unlicensed and off-label drug use in children: implications for safety. *Expert Opin. Drug Saf.* 3, 81–83
3 Baber, N. and Pritchard, D. (2003) Dose estimation for children. *Br. J. Clin. Pharmacol.* 56, 489–493
4 Ginsberg, G. *et al.* (2004) Incorporating pharmacokinetic differences between children and adults in assessing children's risks to environmental toxicants. *Toxicol Appl. Pharmacol.* 198, 164–183
5 Barton, H.A. (2005) Computational pharmacokinetics during developmental windows of susceptibility. *J. Toxicol. Environ. Health A* 68, 889–900
6 McCarver, D.G. and Hines, R.N. (2002) The ontogeny of human drug-metabolizing enzymes: phase II conjugation enzymes and regulatory mechanisms. *J. Pharmacol. Exp. Ther.* 300, 361–366
7 Alcorn, J. and McNamara, P.J. (2002) Ontogeny of hepatic and renal systemic clearance pathways in infants: part II. *Clin. Pharmacokinet.* 41, 1077–1094
8 FDA, C. Guidance For Industry: General Considerations for Pediatric Pharmacokinetic Studies (http://www.Fda.Gov/Cder)
9 Kearns, G.L. *et al.* (2003) Developmental pharmacology – drug disposition, action, and therapy in infants and children. *New Engl. J. Med.* 349, 1157–1167
10 Ginsberg, G. *et al.* (2002) Evaluation of child/adult pharmacokinetic differences from a database derived from the therapeutic drug literature. *Toxicol. Sci.* 66, 185–200
11 Kauffman, R.E. and Kearns, G.L. (1992) Pharmacokinetic studies in paediatric patients. Clinical and ethical considerations. *Clin. Pharmacokinet.* 23, 10–29
12 Ginsberg, G. *et al.* (2004) Physiologically based pharmacokinetic (PBPK) modeling of caffeine and theophylline in neonates and adults: implications for assessing children's risks from environmental agents. *J. Toxicol. Environ. Health A* 67, 297–329
13 Anderson, B.J. *et al.* (1997) Size, myths and the clinical pharmacokinetics of analgesia in paediatric patients. *Clin. Pharmacokinet.* 33, 313–327
14 Johnson, T.N. (2005) Modelling approaches to dose estimation in children. *Br. J. Clin. Pharmacol.* 59, 663–669
15 Jullien, V. *et al.* (2006) Population analysis of weight-, age-, and sex-related differences in the pharmacokinetics of lopinavir in children from birth to 18 years. *Antimicrob. Agents Chemother.* 50, 3548–3555
16 Johnson, T.N. *et al.* (2006) Prediction of the clearance of eleven drugs and associated variability in neonates, infants and children. *Clin. Pharmacokinet.* 45, 931–956

17 Mahmood, I. and Balian, J.D. (1996) Interspecies scaling: predicting clearance of drugs in humans. Three different approaches. *Xenobiotica* 26, 887–895
18 Lee, P.I. (2001) Design and power of a population pharmacokinetic study. *Pharm. Res.* 18, 75–82
19 Johnson, T.N. *et al.* (2002) Contribution of midazolam and its 1-hydroxy metabolite to preoperative sedation in children: a pharmacokinetic-pharmacodynamic analysis. *Br. J. Anaesth.* 89, 428–437
20 Micallef, S. *et al.* (2007) Sequential updating of a new dynamic pharmacokinetic model for caffeine in premature neonates. *Clin. Pharmacokinet.* 46, 59–74
21 Meibohm, B. *et al.* (2005) Population pharmacokinetic studies in pediatrics: issues in design and analysis. *AAPS J.* 7, E475–E487
22 Bjorkman, S. (2006) Prediction of cytochrome p450-mediated hepatic drug clearance in neonates, infants and children: how accurate are available scaling methods? *Clin. Pharmacokinet.* 45, 1–11
23 Johnson, T.N. (2007) The problems in scaling adult drug doses to children. *Arch. Dis. Child.* published online 15 Mar 2007, doi:10.1136/adc.2006.114835
24 Hayton, W.L. (2000) Maturation and growth of renal function: dosing renally cleared drugs in children. *AAPS Pharm. Sci.* 2, E3
25 Alcorn, J. and McNamara, P.J. (2002) Ontogeny of hepatic and renal systemic clearance pathways in infants: part I. *Clin. Pharmacokinet.* 41, 959–998
26 Edginton, A.N. *et al.* (2006) A mechanistic approach for the scaling of clearance in children. *Clin. Pharmacokinet.* 45, 683–704
27 Yang, F. *et al.* (2006) Population-based analysis of methadone distribution and metabolism using an age-dependent physiologically based pharmacokinetic model. *J. Pharmacokinet. Pharmacodyn.* 33, 485–518
28 Mahmood, I. (2006) Prediction of drug clearance in children from adults: a comparison of several allometric methods. *Br. J. Clin. Pharmacol.* 61, 545–557
29 Holford, N.H. (1996) A size standard for pharmacokinetics. *Clin. Pharmacokinet.* 30, 329–332
30 Anderson, B.J. and Meakin, G.H. (2002) Scaling for size: some implications for paediatric anaesthesia dosing. *Paediatr. Anaesth.* 12, 205–219
31 Johnson, T.N. (2004) A comparison of methods to predict drug clearance in neonates, infants and children. *Br. J. Clin. Pharmacol.* 57, 677–678
32 Edginton, A.N. *et al.* (2006) Development and evaluation of a generic physiologically based pharmacokinetic model for children. *Clin. Pharmacokinet.* 45, 1013–1034
33 Bjorkman, S. (2004) Prediction of drug disposition in infants and children by means of physiologically based PK modeling: theophylline and midazolam as model drugs. *Br. J. Clin. Pharmacol.* 59, 691–704
34 Price, K. *et al.* (2003) Physiological modeling of age-specific changes in the pharmacokinetics of organic chemicals in children. *J. Toxicol. Environ. Health A* 66, 417–433
35 Clewell, R.A. *et al.* (2007) Perchlorate and radioiodide kinetics across life stages in the human: Using PBPK models to predict dosimetry and thyroid inhibition and sensitive subpopulations based on developmental stage. *J. Toxicol. Environ. Health A* 70, 408–428
36 Nesterov, I. (2003) Whole body pharmacokinetic models. *Clin. Pharmacokinet.* 42, 883–908
37 Hines, R.N. (2006) Developmental and tissue-specific expression of human flavin-containing monooxygenases 1 and 3. *Expert. Opin. Drug Metab. Toxicol.* 2, 41–49
38 Hines, R.N. and McCarver, D.G. (2002) The ontogeny of human drug-metabolizing enzymes: phase I oxidative enzymes. *J. Pharmacol. Exp. Ther.* 300, 355–360
39 de Wildt, S.N. *et al.* (1999) Glucuronidation in humans. Pharmacogentic and developmental aspects. *Clin. Pharmacokinet.* 36, 439–452
40 Duanmu, Z. *et al.* (2006) Developmental expression of aryl, estrogen, and hydroxysteroid sulfotransferases in pre- and postnatal human liver. *J. Pharmacol. Exp. Ther.* 316, 1310–1317
41 Miyagi, S.J. and Collier, A.C. (2007) Pediatric development of glucuronidation: the ontogeny of hepatic UGT1A4. *Drug Metab. Dispos.* 35, 1587–1592
42 Clewell, H.J. *et al.* (2004) Evaluation of the potential impact of age- and gender-specific pharmacokinetic differences on tissue dosimetry. *Toxicol. Sci.* 79, 381–393
43 Nong, A. *et al.* (2006) Modeling interchild differences in pharmacokinetics on the basis of subject-specific data on physiology and hepatic CYP2E1 levels: a case study with toluene. *Toxicol. Appl. Pharmacol.* 214, 78–87
44 McNamara, P.J. and Alcorn, J. (2002) Protein binding predictions in infants. *AAPS Pharm Sci.* 4, E4
45 Rudolph, A.M. (1983) Hepatic and ductus venosus blood flows during fetal life. *Hepatology* 3, 254–258
46 Elbarbry, F.A. *et al.* (2007) Ontogeny of hepatic CYP1A2 and CYP2E1 expression in rat. *J. Biochem. Mol. Toxicol.* 21, 41–50
47 Alcorn, J. *et al.* (2007) Evaluation of the assumptions of an ontogeny model of rat hepatic cytochrome P450 activity. *Drug Metab. Dispos.* 35, 2225–2231
48 Cocking, C. *et al.* (2006) Drug metabolism studies in the newborn infant and children using an oral erythromycin breath test. *Paediatr. Perinat. Drug Ther.* 7, 172–177
49 Allegaert, K. *et al.* (2007) Urinary metabolites to assess *in vivo* ontogeny of hepatic drug metabolism in early neonatal life. *Methods Find Exp. Clin. Pharmacol.* 29, 251–256
50 Blake, M.J. *et al.* (2007) Ontogeny of dextromethorphan *O*- and *N*-demethylation in the first year of life. *Clin. Pharmacol. Ther.* 81, 510–516
51 Ratanasavanh, D. *et al.* (1991) Intralobular distribution and quantitation of cytochrome P-450 enzymes in human liver as a function of age. *Hepatology* 13, 1142–1151
52 Gentry, P.R. *et al.* (2003) Evaluation of the potential impact of pharmacokinetic differences on tissue dosimetry in offspring during pregnancy and lactation. *Regul. Toxicol. Pharmacol.* 38, 1–16
53 Kearns, G.L. *et al.* (2003) Impact of ontogeny on linezolid disposition in neonates and infants. *Clin. Pharmacol. Ther.* 74, 413–422
54 Saldien, V. *et al.* (2003) Target-controlled infusion of rocuronium in infants, children, and adults: a comparison of the pharmacokinetic and pharmacodynamic relationship. *Anesth. Analg.* 97, 44–49, table of contents
55 Stephenson, T. (2005) How children's responses to drugs differ from adults. *Br. J. Clin. Pharmacol.* 59, 670–673
56 Faustman, E.M. *et al.* (2000) Mechanisms underlying Children's susceptibility to environmental toxicants. *Environ. Health Perspect.* 108 (Suppl. 1), 13–21
57 Tabacova, S.A. and Kimmel, C.A. (2001) Enalapril: pharmacokinetic/dynamic inferences for comparative developmental toxicity. A review. *Reprod. Toxicol.* 15, 467–478