2015

Pharmacokinetics in neonatal prescribing: evidence base, paradigms and the future

Kate O'Hara
University of Newcastle

Ian M. R Wright
University of Wollongong, iwright@uow.edu.au

Jennifer J. Schneider
University of Newcastle

Alison L. Jones
University of Wollongong, alisonj@uow.edu.au

Jennifer H. Martin
University of Newcastle

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Paediatric patients, particularly preterm neonates, present many pharmacological challenges. Due to the difficulty in conducting clinical trials in these populations dosing information is often extrapolated from adult populations. As the processes of absorption, distribution, metabolism and excretion of drugs change throughout growth and development extrapolation presents risk of over or underestimating the doses required. Information about the development these processes, particularly drug metabolism pathways, is still limited with weight based dose adjustment presenting the best method of estimating pharmacokinetic changes due to growth and development. New innovations in pharmacokinetic research, such as population pharmacokinetic modelling, present unique opportunities to conduct clinical trials in these populations improving the safety and effectiveness of the drugs used. More research is required into this area to ensure the best outcomes for our most vulnerable patients.

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Title Page

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Kate O'Hara
School of Medicine and Public Health
University of Newcastle
Australia

Ian M. R. Wright
University of Wollongong and Illawarra Health & Medical Research Institute,
Wollongong, Australia
University of Newcastle and Hunter Medical Research Institute, Newcastle, Australia
Clinical lead, Newborn services, Division of Paediatrics, Illawarra Shoalhaven Local
Health District, NSW, Australia

Jennifer J. Schneider
School of Biomedical Sciences and Pharmacy, University of Newcastle

Alison L. Jones
Executive dean faculty science, medicine and health, University of Wollongong
Deputy director, Illawarra Health and Medical research institute, Wollongong
School of Medicine and Public Health, University of Newcastle

Jennifer H. Martin
School of Medicine and Public Health
University of Newcastle

Corresponding Author
Kate O'Hara
Kate.ohara@uon.edu.au
Department of Clinical Pharmacology
School of Medicine and Public Heath
University of Newcastle
New South Wales
Australia

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Abstract

Paediatric patients, particularly preterm neonates, present many pharmacological challenges. Due to the difficulty in conducting clinical trials in these populations dosing information is often extrapolated from adult populations. As the processes of absorption, distribution, metabolism and excretion of drugs change throughout growth and development extrapolation presents risk of over or underestimating the doses required. Information about the development these processes, particularly drug metabolism pathways, is still limited with weight based dose adjustment presenting the best method of estimating pharmacokinetic changes due to growth and development. New innovations in pharmacokinetic research, such as population pharmacokinetic modelling, present unique opportunities to conduct clinical trials in these populations improving the safety and effectiveness of the drugs used. More research is required into this area to ensure the best outcomes for our most vulnerable patients.

Introduction

Due to the difficulties of conducting traditional pharmacokinetic studies in children and neonates extrapolation of dosing information from adult studies has persisted, despite increasing evidence that this is both inappropriate and at times ineffective. Much of the difficulty advising clinicians in dosing paediatric patients has come from a lack of knowledge of development of physiological processes and the quantification of this at each stage of development throughout infancy, childhood and adolescence. Further, the lack of an appropriate method to determine drug doses for clinical use from this knowledge has made this task more difficult. We propose that urgent investment in paediatric pharmacokinetic knowledge, validated against real pharmacodynamic outcomes, is required to ensure the best outcome for this vulnerable group of patients.

Both the European Medicines Agency (EMA), as part of the European Unions Paediatric Regulation in 2007, and Food and Drug Administration (FDA), by the Best Pharmaceuticals for Children Act (last amended 2007) have acknowledged that more work is required to ensure the safe use of medicines in children. While improvements have occurred, progress has been slow (1). A study conducted by the FDA in the United States examined different methods of predicting paediatric clearance of drugs based on adult values and concluded that no single method of prediction is suitable for all drugs or age groups (2)

Drug doses used in children have generally been extrapolated from adult data on a mg/kg basis. However, human growth and development is not a linear process and such an approach is known to be problematic (3). Size alone is not adequate for determining doses across the range of developmental processes that occur throughout childhood as it can not fully explain clearance, even when considered with other variables such as age and renal function (4). Despite this, extrapolation from adult data continues to occur for the majority of medications (5). Weight-based and surface area-based dosing regimens, extrapolated from adult data, are used in most clinical situations.

It is difficult to determine appropriate doses and intervals while still lacking basic information on metabolism and excretion in different age groups (6). This problem is particularly compounded in prescribing for neonates. While age and weight are the two

variables most commonly used when determining doses for neonates it is known that there is a non-linear relationship between metabolism and weight (7). Body surface area has been investigated as an appropriate alternative to weight when determining neonatal drug doses but has not been shown to increase accuracy or safety (7). Unfortunately no current method of dose estimation can replace clinical studies.

Premature birth can result in serious health complications including chronic lung disease, necrotizing enterocolitis, retinopathy of prematurity, intraventricular haemorrhage and cerebral palsy (8). Neonates born preterm or requiring intensive care treatment are frequently prescribed medications from a range of pharmacological classes, including analgesics, antimicrobials and diuretics, which are not licensed for use in this population (9, 10) .

The four major pharmacokinetic processes of absorption, distribution, metabolism and excretion each present important differences between the responses of neonates and adults to drug treatment. In neonates, these processes reach adult activity at different stages of growth and development (6). The developmental changes in absorption, distribution, metabolism and excretion will each be addressed below.

Absorption

Absorption is the ability of drugs administered by extravascular routes to overcome chemical, mechanical and physical barriers to be distributed to their site of action. The developmental differences in these barriers between neonates and adults can change the rate and extent of drug absorption (7). Intravenous administration of drugs is not affected by developmental changes in absorption as drugs are delivered directly to the bloodstream. However, compared to oral, intravenous administration is associated with additional risks, such as infections. Once a neonate has developed a functioning gut and is tolerating oral feeds oral administration is commonly used. Gastrointestinal absorption can be affected by many factors including gastric pH (11), rate of gastric emptying (12) and intestinal motility (13) , and the type of infant milk diet (14).

Bioavailability is also affected by the development of intestinal CYP enzymes and p-glycoprotein efflux pumps (15).

The absorption of drugs via other routes also changes throughout infancy. For example, changes in skin thickness during development can affect topical absorption (16). The larger body surface area to weight ratio (17) increases the amount of absorption through the skin of children as compared with adults. Extremely low birth weight newborn infants have very thin skin which does not provide an effective barrier (16) placing these infants at risk of adverse effects from topical exposures (18). Rectal (19), pulmonary (20), sublingual (21) and buccal (22) administration are not well studied in neonates and although potentially appealing from an administration perspective are likely to lead to unpredictable absorption via these routes.

Distribution

Body composition is an important determinant of drug distribution. Drugs which are hydrophilic mainly distribute into body water, while lipophilic drugs will preferentially distribute into fat. The increased percentage of total body water and the ratio of intra to extracellular fluid in neonates compared to adults will also influence the distribution of drugs into tissues (6). The corresponding decrease in body fat stores with an increase total body water in neonates compared with adults results in a change to the distribution of both hydrophilic and lipophilic drugs (23). Due to the lower percentage of fat and muscle mass in neonates drugs that rapidly distribute into muscle, like fentanyl, remain in the plasma compartment for longer (23). Both these factors are more pronounced in the premature neonate (7) with the total body water decreasing from 85% in preterm neonates to 75% in term neonates (24). Neonates and infants have less circulating plasma proteins which, for highly protein bound drugs, influences the amount of free drug available for distribution in the body and pharmacological effect. Neonates also have higher circulating bilirubin and free fatty acids which can displace drugs from albumin binding sites (6). Changes in the volume of distribution are also related to changes in blood flow, tissue perfusion, membrane permeability and cardiac output , all affected in conditions such as sepsis (25).

Metabolism

The metabolism of drugs is an essential step in both drug activity and clearance. The ability to metabolise drugs is present in the fetus and newborn and changes throughout early growth and development (26). Inter-individual variation in drug metabolism is dependent on a number of factors, including disease, environment and genes (7, 28) along with growth and development. Clinically, the development of drug metabolising enzymes is an important factor in determining drug selection. For example codeine is not commonly used in the first month of life as conversion to morphine via CYP2D6 is low, limiting effectiveness (3) while midazolam is cleared by CYP3A4 at a slower rate causing increased duration of sedation (29).

Drug metabolism enzymes are divided into phase I and phase II enzymes. Phase I enzymes are involved with primary oxidation, reduction and hydrolysis processes (30). Phase II enzymes conjugate drug molecules to allow excretion (30). The most important group of enzymes involved in phase I metabolism are cytochrome (CYP) P450 enzymes (7). A lack of activity of these enzymes can be responsible for the extreme toxicity syndromes that have been seen in premature infants, such as grey sickness or grey baby syndrome with chloramphenicol and gasping syndrome with benzyl alcohol(18, 31, 32).

Some CYP450 enzymes are active *in-utero* while others do not demonstrate activity until some time after birth. When corrected for weight the content of CYP enzymes in fetal livers is 30-60% of adult values (7) and full CYP activity is usually achieved by 2 years of age (6)..

Maturation rates are difficult to generalise and enzyme-specific information needs to be determined for an accurate estimate of drug metabolism (7). Polymorphisms, diseases, such as sepsis, and complex surgery can all increase the variability of drug metabolism (33). A diet based on infant formula rather than breast-milk (14) and antenatal exposure to cigarette smoke (34) can increase the rate of CYP enzyme development . Further, the routes of metabolism seen in adults may not be mirrored in neonates due to the activity or inactivity of particular CYP enzymes (3), summarized in Table 1.

Looking specifically at CYP enzymes known to be important in drug metabolism pathways, CYP1A2 has been shown to have negligible activity before birth suggesting that postnatal events are required to stimulate development of this enzyme (14). Using caffeine metabolism as a marker of CYP1A2 activity, increases are seen over the first eight months of life. Adult metabolism patterns are seen between 7 and 8 months of age (35). CYP1A2 is induced rapidly after birth with postnatal age rather than postmenstrual age correlating with changes in half-life and clearance (36). This rapid induction fits clinically with the lack of toxicity to caffeine seen in even the most premature infants started on caffeine for the prevention or treatment of apnoea of prematurity (37).

CYP2C9 has been detected in fetal livers from 8 weeks of gestation and in increasing amounts from 24 weeks (28). CYP2C9 begins to increase, to 10% of adult values, during the third trimester remaining constant throughout gestation until birth when levels increase to approximately 25% of adult values during the first month after birth. No further increases are seen in the next year of life (38).

CYP2C19 is also detected in fetal livers from 8 weeks of gestation with levels remaining constant throughout gestation (28). CYP2C19 reached 20% of adult values by the end of the second trimester and is the dominant CYP2C enzyme during gestation. At birth and during the neonatal period there is no significant change to the level of expression. (38). Increases are seen after 5 months of age when mature levels are reached (28).

CYP2D6 enzyme maturation is thought to be complete after 1 year with significant increases in the first month of life. Activity, using tramadol metabolism as a marker, has been shown to increase from 27 weeks postmenstrual age to reach 84% of mature values by one month of age (39). CYP2D6 is subject to genetic differences in activity levels. Toxicities from drugs metabolised via this pathway can be related to development of the pathway along with genetic predisposition to being a poor or ultra rapid metaboliser (33). The poor metaboliser genotype has been thought to be in part responsible for toxicities shown following perinatal exposure to paroxetine (40). Differences in toxicity related to codeine exposure in via breast milk have been linked to genetic polymorphisms in CYP2D6 activity (41).

CYP2E1 has not been detected in the first trimester of pregnancy. In some samples it was detected during the second trimester but only in very low levels, around 1% of adult values (38). Levels increase throughout the third trimester (42). During the neonatal period there is a rapid increase to around 25% of adult values. Adult values have been reached around 1 year of age (38) but possibly earlier, within 90 days of life (42).

The CYP3A family of enzymes is important in the metabolism of a large number of drugs and endogenous substances including steroids (43). CYP3A4 is expressed at low levels within the liver throughout gestation (38). CYP3A4 is functionally immature after birth and begins to increase after 2 weeks postnatal age (29). CYP3A4 begins to develop after birth to reach 30-40% of adult values by 4 weeks of age. Changes in expression of CYP3A4 are not seen in the first year of life and is thought to reach adult levels by around three years of age (38).

CYP3A7 activity is most prominent in perinatal life and at birth. Its activity declines rapidly following birth throughout the first year of life. This pattern of enzyme development is different to that of other CYP enzymes (7). CYP3A7 is active during the antenatal period reaching a peak in the first week after birth following which it begins to decline to reach the very low levels found in fully developed adult livers (44). Oestrogen and progesterone at birth increases activation of CYP3A4 enzymes (45). While CYP3A7 activity halves in the neonatal period it is still the most abundant CYP3A enzyme expressed at this time.

Less is known about the development of phase II enzymes in the immature liver (6). Phase II reactions include glucuronidation, sulphation, methylation and acetylation and so are an important part of drug metabolism and the biotransformation of endogenous compounds including steroids and bilirubin. The largest group of enzymes involved in these reactions are uridine diphosphate glucuronosyltransferase (UDP) isoenzymes (46). The development of UDP-glucuronosyltransferase (UGT) has both pharmacokinetic (as in chloramphenicol toxicity) and pharmacodynamic (as part of morphine glucuronidation) importance to newborn care (7). While postmenstrual age

seems to be the most important factor in the development of the cytochrome pathways both postmenstrual and postnatal age are relevant to the development of glucuronidation pathways, as demonstrated by the metabolism of tramadol (47).

Glucuronidation reactions are not thought to reach adult levels for at least three years (27). Conjugation, however, increases from minimal levels to almost adult levels within two weeks post birth in most cases. Conjugation is important for detoxifying products of both metabolism and drugs, particularly lipophilic compounds. Delays in achieving normal levels are seen in septic and preterm babies (48).

The activity of UGT1A1 can be measured using bilirubin conjugation as a marker. Factors affecting the development of this pathway include: postnatal and postmenstrual age, other medications, genetic polymorphisms, co-morbidities and maternal smoking status (46). Phenobarbitone administration has been reported to induce UGT1A1 activity (49).

UGT2B7 activity has been demonstrated, based on morphine glucuronidation, from 24 weeks post-menstrual age. Activity remains low in the first ten days of postnatal life and then begins to increase in both term and preterm babies (50). Again co-morbidities, surgery and gene polymorphisms can affect this pattern of development (46). The capacity of morphine metabolism by UGT2B7 is closely related to body weight as opposed to surface area and postnatal age after the first ten days of life. During the first two weeks of life capacity increases quickly followed by 2 years of gradual increases to adult levels (50).

Sulphation pathway enzymes have been found in fetal livers with SULT1A1 activity appearing to be expressed at consistent levels through antenatal and postnatal life with other enzymes (SULT1A3 and SULT1E1) declining in activity at the end of gestation (51). Sulphation is an effective metabolic pathway from birth (52).

N-acetyltransferases consist of 2 different enzymes NAT1 and NAT2. Genetic polymorphisms are known to affect NAT2 activity. Low levels are seen in fetal and newborn livers and only limited acetylation is possible. Development continues to between two (53) and four years of age (54).

Table 1: Summary of cytochrome P450 enzyme expression, substrates, inhibitors and inducers.

Enzyme	Becomes active at	Substrates	Inhibitors	Inducers
CYP 1A2	1-3 months	Caffeine Paracetamol	Ciprofloxacin	Tobacco Insulin Omeprazole
CYP 2D6	Hours-days	Amphetamines Codeine Flecainide Lignocaine Metoclopramide	Cocaine Methadone Ranitidine	Phenobarbitone Phenytoin
CYP 2C9	First weeks	Ibuprofen Phenytoin	Fluconazole Sulfamethoxazole	Rifampicin
CYP 2C19	First weeks	Omeprazole Phenytoin Indomethacin	Omeprazole Indomethacin	Carbamazepine Prednisone
CYP 3A4	First weeks	Steroids Clarithromycin Midazolam	Fluconazole Grapefruit Juice	Phenobarbitone Phenytoin
CYP 2E1	Hours	Ethanol Paracetamol		Ethanol Isoniazid

Pharmacogenetics influences the development of drug metabolism pathways. Genetic polymorphisms are well documented to result in drug metabolism variability in adults, and similar effects are expected in neonates although observations of these are currently limited (3, 7). The maturation of drug metabolism pathways can make it difficult to determine the effects of genetic polymorphisms. The phenotype may not be immediately apparent, as the gene may not be fully expressed or the pathway too poorly developed to discriminate differences. Despite this, genetic differences may be important from very early on in antenatal life and have been implicated in changes in drug metabolism resulting in birth defects or malformations (51). For all these reasons the impact of individual polymorphisms on drug metabolism in neonates is dependant on the rate of maturation of the enzyme pathway (55).

Finally, non-enzyme dependant mechanisms can influence the extent of drug metabolism, for example the rate of hepatic blood flow affecting the rate of hepatic drug metabolism. Hepatic blood flow increases throughout neonatal development. This is particularly important for drugs with a high hepatic extraction ratio (23) such as propranolol. Following birth blood flow through the liver changes rapidly, with the ductus venous closing . Following the first feed, within hours of birth, portal blood flow increases and bacterial colonisation of the gut begins, leading to a rapid rise in hepatic processing functions required and thus induction of a number of enzyme groups (48). These acute changes make standard pharmacological models of hepatic extraction, metabolism and blood flow initially invalid in neonates. These models can only be used once the ductus venosus is fully closed (45), up to a week after birth (56).

Excretion

Excretion is the important step of final removal of a drug and/or its metabolites from the body. Excretion is usually via renal or hepatic routes but it is possible for drugs to leave the body by many other routes.

Nephrogenesis is complete at 34 weeks of gestation although growth retardation, nephrotoxic drugs administered during pregnancy and congenital renal malformations

can have a negative effect on kidney development (57, 58). Drugs that affect kidney function, such as ibuprofen, given in the neonatal period can have ongoing effects beyond the period of administration (59, 60). Glomerular filtration rate increases to half adult value by three months of age and reaches adult levels by 2 years of age (23). Premature neonates have a slower rate of kidney development (61) and smaller kidney volume (62) although the administration of betamethasone has positive effects on kidney development (63). Babies who are born small for gestational age have been shown to have lower renal excretion than appropriate for gestational age counterparts (64). Creatinine at birth is not a reliable marker of glomerular filtration rate and largely reflects maternal renal function for the first few hours. It becomes a more accurate marker of renal function during the first weeks of postnatal life (65).

Organic anion transporters in the kidney are responsible for the final excretion of compounds formed during stage II biotransformation. Organic anion transporters have low activity at birth then increase rapidly to high levels, higher than those seen in adults, over the first few weeks of life and then begin to decline to adult levels. This change in secretion happens independently of changes in renal mass. Exposure to some hormones (66) and substrates, such as penicillin (67) also increase organic anion transporter activity.

Excretion is particularly important for drugs that do not undergo any biotransformation, such as gentamicin. In order to limit the toxicity of gentamicin the dose is adjusted according to the kidney development of each stage of growth throughout childhood.

The excretion of excipients, which are co-administered with drugs, is often not considered. Excipients have been associated with adverse effects and large amounts can be administered to hospitalised neonates (68-70) and are difficult to study (71). The neonatal administration of propylene glycol (72) and benzyl alcohol (32) in injectable drug formulations has been associated with serious adverse effects, including death.

Population Pharmacokinetic Models

The development of dosing information in children is more difficult than in adults and has been hampered by a lack of suitable methods for investigating pharmacokinetics in these populations. Most early clinical trial data comes from young, healthy adult males who are usually different from the target population for a drug (73). Children, especially neonates, are unable to provide the large number of blood samples required to produce individual pharmacokinetic models (74). Allometric scaling has been suggested as a method of estimating paediatric dosing from adult studies, however, no single method of allometric scaling has been shown to work for all drugs or all age groups (2). Population pharmacokinetic modelling is particularly useful in the neonatal population (75, 76), where repeat sampling of blood is too high a volume for the infant and ethically inappropriate (74). Population modelling also allows the study of more co-variates than allometric scaling which only takes into account size. Anti-infectives are the most common drugs studied in this population using this method (77).

Population pharmacokinetic models use concentration time points from a number of clinical subjects to determine the pharmacokinetics of drugs in that population. Data from patients who are receiving the drug clinically further strengthens the model by taking into account disease-specific changes in pharmacokinetics that may influence drug dosing. This is important in neonates for whom small dose changes may have large clinical effects. The data required include, but are not limited to, age (both postnatal and postmenstrual), weight, height and renal function. There are a number of ways to perform these studies in children including the naïve pooled data approach, standard two-stage approach and mixed effect models (78).

Adherance to specific sampling times is not important for population pharmacokinetic studies and samples taken as part of routine clinical care can be used (73). This limits the number of samples required from each subject. This is an important ethical consideration when working with neonates. The fact that these studies are conducted under real life conditions is an additional benefit (77). The use of population pharmacokinetic models in preterm neonates is limited by the knowledge of the development of drug metabolism pathways and physiological changes following birth.

This can limit the ability of the model to predict inter-patient variability and pharmacodynamic effects (79).

The introduction of these modelling techniques can reduce the burden to the patient of being involved in a pharmacokinetic study. While not perfect and still an evolving field population pharmacokinetic modelling has allowed prediction of the effects of drugs on the very smallest of neonates. Currently it appears that population pharmacokinetic studies are not followed up by the clinical studies required to fully evaluate the safety and effectiveness of dose recommendations made (77) and this remains a clear area of research need.

Conclusion

More information about the maturation of pharmacokinetic processes is required to ensure safe and effective use of drugs in children. The development of population pharmacokinetic modelling systems, which reduce the burden to test subjects, represent a significant improvement and offer large areas of future research. We suggest that neonatal population pharmacokinetic studies be conducted for commonly used drugs as an immediate research priority. Particularly they need to be developed by linking pharmacology, medicine and mathematics to ensure accurate, clinically relevant endpoints and correlations are measured. The models will need to be continually revised and updated as new physiological and pharmacological information becomes apparent to ensure the best possible outcome for these vulnerable patients.

All authors have completed the Unified Competing Interest form at www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author) and declare: no support from any organisation for the submitted work; no financial relationships with any organisations that might have an interest in the submitted work in the previous 3 years; no other relationships or activities that could appear to have influenced the submitted work.

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