Chapter 3
Developmental Changes in the Processes Governing Oral Drug Absorption

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Abstract Pharmacotherapy in children often consists of oral medication. Effectiveness of oral prescriptions may be influenced by extrinsic (formulation, nutrition, and co-medication) and intrinsic factors (physiological and disease-related variation).

During development the GI characteristics change: swallowing reflexes, excretion of digestive enzymes, intestinal motility, transit time and intestinal transporters and drug metabolizing enzymes. For example, changes in drug efflux transporters result in a decrease or increase in expelling drugs back into the intestinal lumen and thereby in variation in oral bioavailability.

Closing the main information gaps on the ontogeny of GI processes governing oral drug absorption would allow for more accurate prediction of the oral disposition of drugs in children of all ages. Different ex- and in vitro study designs, as drug dissolution/solubility tests, in vitro drug metabolism and transporter studies and in vivo drug-microdosing can be used to elucidate the age-related changes in GI processes to better understand oral drug disposition in children. Using these data in PB-PK models may further guide individualized pediatric drug therapy.

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3.1 Introduction

Pharmacotherapy in children will usually consist of oral medication [1]. Absorption in the gastro-intestinal (GI) tract—and thereby effectiveness of oral medication—may be influenced by drug formulation, food intake, co-medication, and physiological factors. The oral route is characterized by changing environments from the oral cavity with saliva to the GI tract with interplay of digestive enzymes, intestinal motility, transit time and, moreover, intestinal transporters and drug metabolizing enzymes on the cellular level [2]. In children, the interaction between the oral drug and the developmental continuum will influence the systemic exposure to and effectiveness of the medication. Few studies are available on changes in bioavailability and other oral absorption parameters in the pediatric age range, a selection of which is presented in Fig. 3.1.

3.1.1 Swallowing Reflex

Swallowing is a multifactorial mechanism transporting food or liquid from the oral cavity to the esophagus. Swallowing involves coordination of neurologic and aerodigestive systems from the oral cavity to the esophagus.
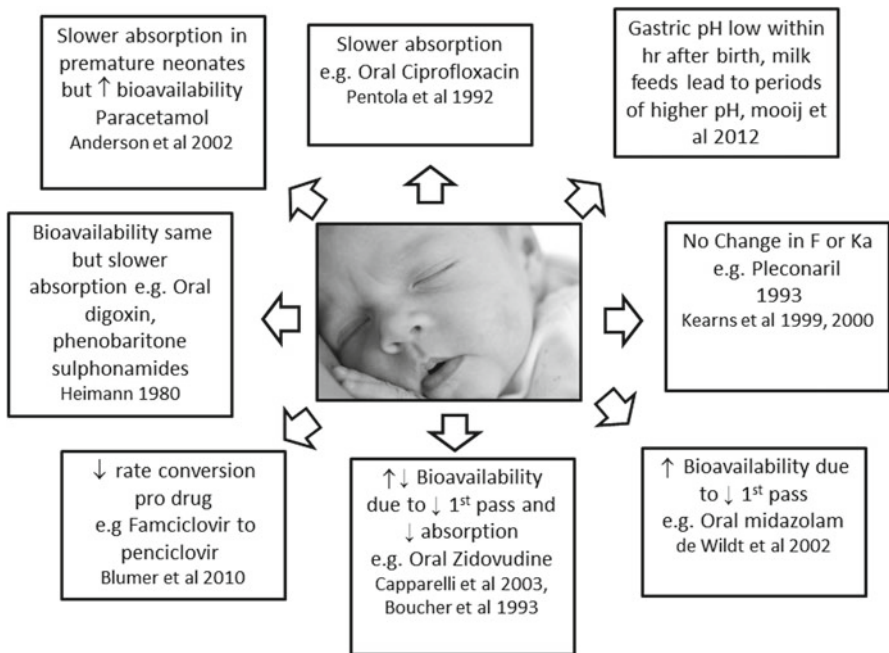

Fig. 3.1 Pediatric drug absorption—evidence from the literature

Quantitative measures such as time at which a (feeding) bolus reaches a specific location and time for a bolus to move from one place to another during a swallow may serve to determine if the process is successful [3]. Besides the voluntary component of swallowing, three different reflexes are also involved in the process; the pharyngeal swallowing reflex, the pharyngo-upper esophageal sphincter contractile reflex, and the pharyngoglottal closure reflex. Reflexive swallowing is crucial to airway protection, as it prevents food or fluid from directly entering the trachea [4], or returning from the esophagus to the trachea. Moreover, all reflexes have a distinct reaction on air and water stimuli [4, 5].

The first swallowing activity appears at around 11 weeks gestation, and further matures over time. Non-nutritive swallowing is well patterned by 34 weeks postmenstrual age. The suction and expression/compression component of sucking in preterm infants can be assessed on a five-point scale [6]. Recently an objective indicator of infants' feeding ability has been developed assessing feeding skills and endurance [7] as an expression of proficiency (number of ml taken during the first 5 min/total ml prescribed × 100) and milk transfer (ml/min).

Overall transfer (percentage of total volume transferred/volume to be taken) and rate of transfer (ml/min) of the feeding bolus is positively correlated with increased gestational age. So, although preterm babies could have developed similar oral motor skills as term babies, term babies can drink faster and more [8].

An important part of oral feeding is pharyngeal and upper esophageal sphincter function but this is not easy to assess. Compared with older neonates who are adapted to feeding, most preterm infants demonstrate poor pharyngeal pressures at the laryngeal inlet coupled with poor coordination of the pharyngeal propulsion with upper esophageal sphincter relaxation [9].

Research does not indicate that growth of the oropharyngeal cavity during childhood may influence swallowing [9], although the position of the feeding bolus in relationship to the laryngeal closure changes from the age of 2 to 48 months [10]. Furthermore, with increasing age or initiation of cup feeding, laryngeal vestibule closure occurs later in the swallowing process.

To our knowledge, salivary content has never been studied in relation to development. Two studies have addressed salivary flow rates in young fasting children and found that the mean salivary flow went up from 28.2 to 39.6 ml/min between 18 and 42 months of age, these changes are not statistically significant [11, 12].

On a more molecular level, recent studies on the salivary transcriptome revealed up-regulation of specific genes involved in the different aspects of oral feeding in newborns. The genes in question are involved in the sensory development, neurodevelopment, cartilage and bone development, cranial nerve development, feeding behavior, and muscular development [13].

Oral medication intake (tablets, capsules, liquids) is also influenced by the maturation state of normal feeding, unless liquid formulations are given via a nasogastric tube. Although liquid formulations overcome the problem of difficulty of swallowing solids, in children who are not fully accustomed to oral feeds, oral suspensions may also present a problem.

Moreover, children's taste sensation can negatively affect liquid ingestion. Sweet tastes are innately preferred by most or all herbivores and omnivores, presumably since sweetness reflects the presence of caloric sugars in plants. Bitter taste signals the presence of potentially toxic compounds and hence substances that are bitter are generally disliked and avoided. Neonates and infants therefore react adversely to bitter taste. A sensitivity to and preference for salty substances also appears to have an innate component, which develops at around 4 months of age. By 2 years of age, children's preferences for salty foods are even greater than those of adults [14]. These preferences in children should be accounted for by masking the taste of drugs, either by formulation or by dissolving the drug in a suitable vehicle. The latter is often challenged by the increased volume of the bottle feeding. However, this problem is not restricted to the (preterm) infant. Also older children and even adolescents (age between 11 and 20 years) often have difficulty swallowing tablets and capsules [15]. In a Danish study 8.5 % of the documented oral medication had been interrupted [16]. This problem might be explained by the smaller dimensions of the puerile pharynx and the developing oropharyngeal musculature [17] and by lack of experience in swallowing drugs. Children can be trained in swallowing oral medication, it was shown that training was successful from 3 years of age [18, 19].

Moreover, adapting formulations from a tablet or capsule to a liquid formulation may negatively influence drug efficacy [1]. Tablets and capsules cannot be divided multiple times without losing dosing accuracy. Regrettably, pediatric formulations for many drugs are lacking, and children often receive unlicensed and off-label drugs of which safety and efficacy are unknown. It is important, therefore, to speed up the licensing of drugs for children. Recently, much research effort was spent on pediatric medicines formulation with a view to facilitating easier oral drug administration. Especially the development of micro-particulate solid dose formulations and mini-tablets seems promising [20, 21] as it may overcome some of the inherent stability issues of oral liquid formulations.

3.1.2   pH

Gastric acid has a main role in food digestion and is an important barrier in gastrointestinal defense. Gastric pH is usually measured by intermittent gastric fluids aspirates or by continuous intragastric pH monitoring (24 h).

In all age ranges gastric pH is strongly acidic. Only in newborn children just seconds after delivery the mean is around 7, but dropping to 2–3 a few hours after birth [22, 23]. This phenomenon is explained by the swallowing of amniotic fluid during birth [22]. Other studies have subsequently shown that the gastric pH remains low at values between 2 and 3 in children of all ages [24–40].

In continuous measurement of gastric pH we usually consider the proportion of time in 24 h that values above 4 occur. In preterm infants this proportion ranged from 46 to 70 % [41, 42]. The age of 2 years seems to be a turning point as this proportion decreased from 51 in children under the age of 2 years to 34 % in older children [43]. This effect is most likely caused by the buffering capacity of the

formula or breast milk with which the younger children are fed, seeing that the older children receive more solids [37, 38, 43]. The influence of the buffering capacity of feeding was also seen in preterm neonates, as the postprandial pH of 7 dropped to 2 within 3 h after feeding [37, 38]. The buffering capacity of milk feeding is often believed to produce a non-acidic gastric environment in neonates. This is true to some extent; however, gastric pH reaches low adult values in between the feeds.

Little is known about the maturation of intestinal pH. One small study using a radio-transmitting pH-sensitive capsule showed an increase from 6.4 to 7.4 from the duodenum to the distal part of the small intestine in children aged between 8 and 14 years. The pH profile was almost identical to that found in healthy adults [44].

Different pH values can induce different absorption profiles to orally given drugs. For example, the antifungal drug ketoconazole is less effectively absorbed in preterm infants, most likely because the gastric fluid is not acidic during drug absorption [45].

3.1.3 pH Changes in GI tract

A drug's chemical nature determines the effect of prolonged periods of elevated gastric pH on its absorption. Weakly basic drugs such as ketoconazole [45] may be absorbed less well, but weakly acidic drugs somewhat better as they are more soluble at higher pH values. In addition, drugs that are unstable at acidic pH may be absorbed better at higher gastric pH values [46].

The effect of changes in gastric pH is probably most evident with drugs from Biopharmaceutical Classification System (BCS) classes II (low solubility, extensive metabolism) and IV (low solubility, low metabolism) [47]. It may be difficult to predict the effect of an age-related change in gastric pH (due to buffering effect of frequent oral feeds in neonates) on oral drug absorption as it is the result of interplay between changing feeding regimens and other developmental changes in oral drug absorption.

3.1.4 Gastric Emptying

The duration of a drug's exposure to a highly acidic environment is determined by the gastric emptying time.

Various techniques are available to establish gastric emptying time, including the gastric emptying breath test, scintigraphic procedure by Technetium-99M liquid gastric emptying scan, and the paracetamol absorption test. The results are expressed in various ways: gastric emptying time, gastric half-emptying time, or residual gastric activity at 1 h.

Gastric emptying time is influenced by stomach content. The L-glycine-1-^{13}C breath test performed in four healthy children (age range 12.1–16.0 years) showed different gastric half-emptying times after fructose or glucose intake (45.5 and 64.3 min, respectively). After ingestion of both sugars the gastric half-emptying time increased significantly to 85.3 min [48].

A meal of solids can increase the gastric half-emptying time even more as shown by a ^{13}C-octanoic acid breath test in nine healthy control patients (mean age 9 years; age range 4–16 years)after eating a meal of bread, ham, juice, and eggs [49]. The mean gastric half-emptying time was 121 min.

The influence of GI reflux on gastric emptying time was evaluated with the ^{13}C-octanoic acid breath test in 22 patients, mean age 13.2 year, with symptoms of gastroesophageal- or duodenogastroesophageal reflux. Surprisingly, gastric empty- ing time did not significantly differ from that in healthy controls [50]. Celiac disease, which affects the small bowel mucosa villi, was associated with a longer gastric emptying time, but the effect disappeared after initiating a gluten free diet [49]. The authors speculate that mucosal inflammation of the duodenum, with impaired smooth muscle contraction or neurotransmitter release, leads to motor abnormalities in the stomach-duodenal passage and thus longer emptying time. There are no data on gastric emptying in very young children established with breath tests.

Scintigraphic imaging can help establish gastric emptying time, gastric half- emptying time, and residual gastric activity.

This method showed a gastric half-emptying time of 60 min in preterm infants (median gestational age 28.9 weeks; range 26–33) at a median postnatal age of 9 days [51]. They were fed hourly although not using a standard meal size. The residual gastric activity at 1 h was 37.5 % (range 19–100 %) [51].

The influence of GERD (based on pH and/or scintigraphic imaging) on gastric emptying was studied in 477 patients aged 0–18 years [52]. In children without bolus or acidic gastroesophageal reflux, gastric residual activity at 1 h declined with increasing age: 65, 51, and 45 % in the age group up to 3 years, 4–6 years, and over 6 years, respectively. Gastric emptying was significantly delayed only in those chil- dren over 6 years suffering from reflux, these results were in line with findings from another study in children suffering from GERD; residual gastric activity was 52 % in 44 infants up to 23 months of age, and 49 % in eight children up to 14 years of age [53]. In yet another study, residual gastric activity in a healthy control group of 11 children with a mean age of 5.6 years was almost similar at 43.3 % [54].

Yahav et al. [55] reported a mean gastric emptying time of 87.8 min in a control group with a mean age 10.4 months. Gastric emptying times in healthy adult con- trols ranged between 56 (32–85) and 104 (49–126) min, for liquid and solid mark- ers, respectively [56, 57].

In the paracetamol absorption test a pharmacological tracer is applied for mea- suring the rate of gastric emptying. This technique has been rarely applied in chil- dren, therefore age-related changes in outcomes are not known. A small study in 15 critically ill but food-tolerant children (median age 5.3 years) showed a median 1.5 (interquartile range 0.7–2.2) ratio of time to reach paracetamol peak to the maxi- mum paracetamol concentration (T_{max}/C_{max}) [58]. The influence of diet was shown in adolescent participants, as the paracetamol absorption ratio was 1.4 for high-fat meals and 0.5 for low-fat meals [59].

Viewed from a different angle, a population pharmacokinetic study in newly born children showed a low oral paracetamol absorption rate in the first days of life, which then increased and stabilized after 1 week [60]. Gastric emptying time seems to

influence paracetamol absorption as a lag time, which is the time to reach and permeate the absorbing surface of the intestine [2], only occurs after oral paracetamol administration and not after rectal administration. This suggests that gastric emptying time may be the primary determinant of a lag time for oral absorption of paracetamol.

A recent meta-analysis of gastric emptying data from 49 studies covering 1991 healthy subjects ranging from preterm birth to adulthood showed that postnatal and gestational age were not significant covariates for changes in gastric emptying. The only significant influence was meal type; gastric emptying was faster in the order: breast milk > formula milk > semi-solid meal > solid meal [61]. A separate analysis of the data did not reveal a significant relationship between volume of feeds and gastric emptying time.

3.1.5 Antroduodenal Contractions

The rate of gastric emptying is determined by an orchestrated combination of antroduodenal motor activity, fundic contraction, pyloric sphincter relaxation, and intestinal motor activity. Contraction and relaxation of the distal stomach and proximal small intestine can be measured by antroduodenal manometry.

Both in the fasting and intraduodenally fed state, antral motor activity does not differ between preterm and term neonates [62]. Yet, in preterm neonates the proportion of antral clusters upon duodenal activity is much lower than that in term neonates. With increasing gestational age not only the degree of association of antral and duodenal activity [63] rises but also the effectiveness of the contractions on motility [64, 65].

Duodenal cluster activity during fasting lasted shorter in preterm neonates than in term infants but duodenal motor activity in response to feeding increased similarly in both preterm and term infants [62]. Maturation of the duodenal activity is dependent on the timing of the introduction of oral food, as maturation was more pronounced after early (days 3–4) rather than after late (days 10–14) introduction [66]. Moreover, duodenal motor activity response to bolus feeding shows a more immature pattern in preterm infants compared to term infants [67].

In contrast to antral motor activity, proximal intestinal (duodenal) motor activity matures throughout the first weeks of life, with increasing frequency, amplitude, and duration of propagating contractions. No data are available on children beyond the neonatal period.

3.1.6 Intestinal Transit Time

The effectiveness of gastro-intestinal motility is reflected by the orocecal transit time (OCTT), which can be measured with the hydrogen breath test, ^{13}C-ureide breath test, radio-transmitting capsule, red carmine marker test, or scintigraphy. However, each of these techniques has its limitations.

The hydrogen breath test with lactulose as non-absorbable carbohydrate substrate has limited use in the general population, which may include hydrogen-non-responders. Moreover, lactulose may accelerate transit time by its osmotic laxative effect, this was clearly shown in healthy subjects: OCTT after a meal was significantly longer than after lactulose [68]. Studies in the pediatric population using the hydrogen breath test to measure OCTT did not reveal an association with age [68–72]. The transit time differs from 60 to 110 min in children from 1 to 17 years of age, which is the same as in adults [72].

OCTT measured with the lactose-^{13}C-ureide breath test had a mean of 255 min in children from 3 to 17 years of age [69]. In adults the lactose-^{13}C-ureide test was validated against scintigraphy [73]. This test is unsuitable in infants below 6 months because they lack the required enzymatic activity to convert lactose ureide. The significantly longer OCTT with the labeled ureide test than the lactulose-H_2 breath test is thought to be caused by the laxative effect of lactulose [74].

In healthy children aged 4–14 years, intestinal transit times have been established using a radio-transmitting capsule, the mean values were 7.5 and 17.2 h in the small bowel and colon, respectively [44]. From the number of observations it was estimated that the capsule resided in the duodenum for 8 %, in the proximal part for 5 %, the mid part for 12 %, and the distal part for 75 % of the small intestinal passage time. The small intestinal transit time of 7.5 h established with this method is considerably longer than that established by the breath tests. The fact that the capsule, which was larger than 2 mm, moved through the distal part of the terminal ileum for three quarters of the time suggests a longer ileo-cecal transit for large particles.

Lastly, scintigraphy performed in premature neonates (gestational age 29 weeks) showed a mean OCTT of 3.1 h [51].

All techniques show a wide range of intestinal transit times with no clear age-association. Differences in reported transit times seem more or less related to the specific test properties.

Gastric emptying and intestinal transit time are the primary determinants of the rate at which drugs are presented to and dispersed along the mucosal surface of the small intestine. This rate is further influenced by intestinal disease. The time to reach maximal plasma levels of an orally absorbed drug could therefore be prolonged in the very young sick child.

3.1.7   Bile Salts and Pancreatic Enzymes

Bile, secreted from the liver, aids the digestion and absorption of lipids by the intestine.

A study in preterm neonates established intestinal bile concentration in the first few weeks postnatally at 4.55 mmol/l, for small- and appropriate-for-gestational-age neonates alike [75]. Another study showed that type of feeding influenced bile acid concentration, as it was higher in breast-fed infants than in formula-fed infants, but this difference is not statistically significant [76]. Measurements at 2, 7, and 10

days to 7 months postnatally made clear that the total bile acid concentration increases during the first few days to months, reaching comparable adult levels between 10 days and 7 months of age [77]. Digestive enzymes secreted by the exocrine pancreas aid in the digestion of nutrients, his digestive function is measured by the fecal Elastase-1 concentration, which is highly specific for the pancreas and is not degraded during the intestinal passage.

The fecal Elastase-1 concentration was abnormal in all of a group of preterm infants for the first 2 days after birth; while concentrations were normal in 43 % of a group of term infants. This discrepancy may be due to immaturity or insufficiency of the exocrine pancreatic function in premature neonates. Other than this there are no age-related changes in fecal Elastase-1 concentrations [78]. In both preterm and term neonates adult levels of fecal Elastase had been reached after 2 weeks [79].

The body's ability to solubilize and absorb lipophilic drugs is influenced by the effectiveness of the biliary function. Immature conjugation, decreased intestinal–hepatic-loop, and transport defects of bile salts into the intestinal lumen may reduce uptake of fat-soluble vitamins and lipophilic drugs.

3.1.7.1 Drug "First Pass" Metabolism in the Intestine

High levels of drug transporters such as multidrug resistance protein 1 (MDR1/MRP1/P-glycoprotein) in the villus tips on the apical side of small bowel enterocytes, along with CYP3A4 within the cells, form a concerted "first pass" defense mechanism limiting the oral bioavailability of drugs, dietary mycotoxins and other xenobiotics [80].

3.1.8 Development of Intestinal Transporters

Intestinal transporters are quite important to oral drug availability. Drug efflux transporters expelling drugs back into the intestinal lumen may reduce their availability. MDR1 is one of the most important efflux transporters [81]. Found in the brush border of the small intestine, this glycoprotein is genetically controlled by the *ABCB1* gene [82].

MDR1 ontogeny can be described by mRNA expression and protein content (total and glycosylated) and localization in the gut wall can be determined by immunohistochemistry. In duodenal biopsies from children aged from 1 month to 17 years, MDR1 mRNA expression was highly variable and not related to age [83, 84]. This observation was backed up Konieczna et al. [85] who investigated the differential expression of ABC transporters MDR1, MRP1, and BCRP in the intestinal epithelium of developing human embryos. Expression of all three transporters had reached adult levels after 12 weeks of intrauterine development. In contrast, Miki et al. showed an age relationship: mRNA expression was low in fetuses and neonates (14–20 weeks, 1–24 days post delivery) but generally higher in the adult group

(15–38 years) [86]. Van Kalken et al. failed to detect MDR1 expression in the intestine until 28 weeks gestational age [87]. Immunohistochemistry found the MDR1 protein on the apical surface of all enterocytes. In children younger than 3 years, MDR1 was also found on a small upper part of the lateral surface [88].

Variant alleles will often lower the activity of transporters. However, the effect of various alleles for MDR1 on activity of transporters on specific substrates is not always clear cut.

Pediatric post-renal transplant patients (age 0.36–16.3 years) carrying the *ABCB1* c.1236C>T or c.2677G>T variant allele showed higher oral bioavailability and lower pre-hepatic extraction ratios of the MDR1 and CYP3A4 substrate cyclosporine than did over 8-year-old non-carriers [89]. There is some evidence linking genotype of MDR1 with CYP3A4 mRNA expression, suggesting it is a compounded result of altered MDR1 and CYP3A4 activity [90].

Moreover, local or systemic inflammation may influence intestinal transporter activity. MDR1 mRNA expression in non-inflamed duodenal biopsies of children with Crohn's disease was significantly higher than that in normal biopsies [91]. The authors speculate that the discrepancy is due to the systemic inflammation present in Crohn's disease. Other studies, however, have shown down-regulation of drug transporter expression in inflammatory states [92].

Little is known about the postnatal development of the other members of the ATP binding cassette transporters found in the small bowel, such as multidrug resistance protein 2 (MRP2/ABCC2) or breast cancer resistance protein (BCRP/ABCG2) [81, 93–95].

3.1.9   Development of Intestinal Metabolism

Our understanding of the ontogeny of intestinal metabolism is far from complete. The 3A (CYP3A) subfamily of cytochrome P450 is probably most studied. This enzyme subfamily is abundantly expressed in the gut and is involved in the first pass metabolism of numerous orally administered drugs in adults [96].

The ontogeny of CYP3A can be described as changes in mRNA expression, protein expression, or activity level. A striking discrepancy is seen between intestinal CYP3A mRNA and protein expression, which may reflect the influence of a post-transcriptional regulatory mechanism. CYP3A protein expression increases with age [97], whereas CYP3A4 and CYP3A5 mRNA expressions are high in the first year of life and then drop to adult levels [88].

Immunohistochemistry in intestinal biopsies showed that CYP3A protein was present in only half of the enterocytes in children younger than 6 months. In the older children (up to 17 years of age) CYP3A protein was expressed in all enterocytes [88]. This suggests that CYP3A ontogeny is determined by the proportion of enterocytes expressing the enzyme rather than by a gradual turning on of enzyme expression in individual enterocytes. Further study should confirm this assumption as the manner of specimen collection, storage and pre-treatment to immunohistochemistry may have been of influence. Dissociation between protein and mRNA levels during maturation has also been reported for hepatic CYP2D6 [98].

The age-related increase in CYP3A protein levels is mirrored with increasing CYP3A4 activity, which can be measured by the degree of formation of 6beta-hydroxytestosterone from testosterone. This method did not detect CYP3A activity in fetal samples [97] butneonates showed much lower CYP3A activity compared to children older than 5 years [97].

Both mean intestinal CYP3A4 and CYP3A5 mRNA expression did not differ between young (age 0.1–15 years) and adult liver transplant recipients [81]. This finding suggests that intestinal CYP3A mRNA expression does not change beyond childhood. The authors did not study the effect of age within the pediatric cohort. However, these data suggest no age-related changes in CYP3A mRNA expression, although this cannot be excluded [83].

The influence of the CYP3A5 gene polymorphism has been studied in the transplant population. For children and adults alike, *CYP3A5*1* gene carriers express higher levels of intestinal CYP3A5 mRNA levels than do *CYP3A5*3* homozygous patients. In *CYP3A5*1* gene carriers, CYP3A5 mRNA accounted for 20–30 % of all CYP3A mRNA detected [83, 84].

Pediatric clinical trials on the oral bioavailability of CYP3A substrates are rare. Midazolam is a validated probe drug for CYP3A4/5 activity. In agreement with its age-related intestinal expression, the oral bioavailability of Midazolam in preterm infants (28–32 weeks, <10 days of age) is significantly higher than in adults (50 versus 30 %) [99, 100].

Of great clinical interest is evidence that type of feeding (breast milk or formula) seems to impact the developmental pattern of combined intestinal and hepatic CYP3A in neonates. CYP3A4 activity, expressed as the urinary metabolite/dextromethorphan ratio, increased in between two weeks and 6 months of age, but the repeated measurements showed that this increase was faster for formula- versus breast milk-fed children [101].

It is important to reiterate that intestinal MDR1 and CYP3A appear to work in concert to potentially limit oral drug bioavailability [90]. Hence, age-associated variation in intestinal MDR1/CYP3A4 activity may differentially impact substrates depending on their affinity for MDR1 and/or CYP3A4.

Increases in MDR1/P-glycoprotein causes increased efflux and therefore a decrease in substrate uptake. Consequently a lesser amount of intraepithelial drug is presented to the metabolizing enzyme. Decreased efflux can consequently cause greater risk on drug toxicity. However, as the MDR-1 substrate subsequently is presented to the liver the same efflux mechanism can have different consequences.

Increased CYP3A4 expression and activity with age, consequently causes higher oral bioavailability of, e.g. midazolam in premature children compared to adults.

3.1.10 Challenges in Research

To gain more insight in the ontogeny of oral drug absorption we will need to use a study design in which specific factors that are subject to change can be elucidated. After all, oral drug absorption is influenced by the interplay of age, genetic, and disease-related changes and co-medication, in addition to ethnicity and gender.

Nevertheless, the limited number of patients and reluctant willingness of parents and patients to cooperate is a challenge for pediatric studies. Moreover, breath tests are not feasible in all age groups and radioactivity of probe medication in the developing child raises ethical concerns, as does the invasive nature of tissue harvesting.

3.1.11 *Research Options*

3.1.11.1 GI Tract Model

To gain more insight in physiological influences on oral substrates, different parameters could be tested in an in vitro drug dissolution and solubility model. The Dutch Institute of Innovative Research has developed the TNO Gastro-Intestinal Tract model (TIM), a computer-controlled dynamic system which mimics the physiological human conditions in stomach and intestines [102, 103]. This system allows researchers to measure possible changes in the effective dose of the drug presented to the intestinal mucosa.

3.1.11.2 Modeling and Simulation: PB-PK Models and Population PK

The available data on age-related changes in relevant GI processes can be incorporated into population-based pharmacokinetics (PB-PK) software programs such as Simcyp® or PKsim®. These programs can then simulate the fate of drugs given to children of different ages and provide guidance for age-appropriate dosing. Modeling pediatric drug absorption by this approach still has a long way to go, however, it becomes more feasible as more research data becomes available and will eventually enable the prediction of oral absorption rate and bioavailability in children. However, in the meantime such a modeling approach can be used in terms of "what if" scenarios to investigate the effects of changing parameters on the prediction of absorption parameters.

The usefulness of these programs is limited by the lack of data on changing physiological and biochemical parameters across the pediatric age range. The current data availability is shown in Fig. 3.2, which also indicates areas that require further research including intestinal transporter ontogeny, intestinal fluid dynamics, and characteristics of the intestinal unstirred boundary layer. Moreover, validation of the models is still challenging as pharmacokinetic data on neonates and infants are scarce [104]. Opportunistic sampling and PK analysis in leftover blood drawn for clinical purposes from all patients receiving medication could provide more data. And then, more pediatric population pharmacokinetics (POPPK) studies involving oral drugs are needed, aimed at quantifying drug absorption parameters across the age spectrum rather than using fixed values for oral bioavailability (F) and absorption rate constant (ka).

AVAILABLE DATA

☑ Stomach volume

☑ pH

  ☑ Gastric: ↑ in early postnatal period after feeds but low in between

☑ Gastric emptying

  ☑ No significant effect of age on mean gastric residence time

  ☑ Food type effects significant

☑ Intestinal length/diameter

  ☑ ↑ as function of age

☑ Transit times

  ☑ No change with age

☑ Permeability

  ☑ Neonatal period of enhanced permeability to large hydrophilic molecules (prob. paracellular)

☑ Bile production and composition

☑ Intestinal CYP3A ontogeny

  ☑ ↑ in expression and activity with age

LACKING DATA

❏ Intestinal UGT/other drug-metabolizing enzyme ontogeny

❏ Intestinal transporter ontogeny

❏ Unstirred water layer characteristics

❏ Fluid dynamics

Fig. 3.2 Data availability for pediatric absorption models

3.1.11.3   In Vitro Drug Metabolism and Transporter Studies

The ontogeny of drug metabolizing enzymes and transporters can be studied in intestinal samples from children of all ages. Methods like RT-PCR for drug transporter expression (mRNA) and sensitive LC–MS–MS to measure protein content are used more widespread. The disconnect between drug transporter mRNA and activity should be considered by researchers especially where the solute carriers are involved, e.g. OATP1B1. Leftover tissue from surgical procedures should be collected consistently over a long time to provide enough samples for research purposes.

3.1.11.4   Microdosing

Ontogeny of drug absorption can also be addressed by a mechanism-based approach [105], e.g. studying one specific drug which represents a specific (intestinal) drug metabolizing enzyme. Pharmacokinetic studies in children of all ages may provide valuable information on the ontogeny of that specific pathway. For example, the plasma clearance of midazolam is a validated and widely used method to study variation in CYP3A4/5 activity in both adults and children [106].

Full PK studies to determine oral bioavailability for a probe drug using a multi-day cross-over design are hardly feasible in children for ethical and practical reasons. Children will not benefit from the drug but rather experience the drug effect and run the risk of adverse events.

Alternatively a stable-labeled isotope or a (very weak) radioactive-labeled microdose can be used [107, 108]. Both stable and radioactive approaches make it possible to administer a labeled probe drug in addition to an intravenous therapeutic dose. Parent compound and metabolites can therefore be traced in serum and urine. This enables simultaneous determination of the pharmacokinetics of therapeutic IV and the labeled oral dose. It eliminates the risk of therapeutic effect and toxicity. A prerequisite for the use of microdosing in this context, however, is dose-linearity across the dosing range.

3.1.12   Concluding Remarks

Closing the main information gaps on the ontogeny of GI processes governing oral drug absorption would allow for more accurate prediction of the oral disposition of drugs in children of all ages. Suggested approaches, both in vitro and in vivo, could provide more understanding of oral drug absorption in children. Clinical trials on the influence of age on drug absorption and thereby effectiveness are indispensable to formulate age-dependent drug dosing protocols.

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