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REVIEW ARTICLE

# Pharmacogenetic Aspects of Drug Metabolizing Enzymes and Transporters in Pediatric Medicine: Study Progress, Clinical Practice and Future Perspectives

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## Abstract

As the activity of certain drug metabolizing enzymes or transporter proteins can vary with age, the effect of ontogenetic and genetic variation on the activity of these enzymes is critical for the accurate prediction of treatment outcomes and toxicity in children. This makes pharmacogenetic research in pediatrics particularly important and urgently needed, but also challenging. This review summarizes pharmacogenetic studies on the effects of genetic polymorphisms on pharmacokinetic parameters and clinical outcomes in pediatric populations for certain drugs, which are commonly prescribed by clinicians across multiple therapeutic areas in a general hospital, organized from those with the most to the least pediatric evidence among each drug category. We also further discuss the research status of the gene-guided dosing regimens and clinical implementation of pediatric pharmacogenetics. More and more drug–gene interactions are demonstrated to have clinical validity for children, and pharmacogenomics in pediatrics have shown evidence-based benefits to enhance the efficacy and precision of existing drug dosing regimens in several therapeutic areas. However, the most important limitation to the implementation is the lack of high-quality, rigorous pediatric prospective clinical studies, so adequately powered interventional clinical trials that support incorporation of pharmacogenetics into the care of children are still needed.

**Key Points**

Recently, pharmacogenomics in pediatrics has shown evidence-based benefits to enhance the efficacy and precision of existing drug dosing regimens of several drugs such as voriconazole, tacrolimus, and thiopurines.

The frequencies of specific genotypes and haplotypes such as *CYP2C19*, *CYP2C9*, and *CYP2D6* vary across ethnic populations, which might impact the dosage recommendations and limit the clinical utility of some pharmacogenomics tests; a clear understanding of the inter-ethnic genetic differences is therefore essential to guide effective global drug prescribing.

The biggest limitation to the implementation of pharmacogenomics is the lack of adequately powered pediatric prospective studies, so the generation of high-quality and validated evidence is still necessary for pediatric patients to support clinical implementation of pharmacogenomics into pediatric practice.

## 1 Introduction

Pharmacogenomics (PGx) is a critical component of precision medicine with the aim of individualizing drug therapy through genetic tests. Pharmacogenomic variants can affect the absorption, distribution, metabolism, and excretion of drugs through pharmacokinetic (PK) mechanisms. Genetic polymorphisms influencing the expression level or functional activity of drug metabolizing enzymes or transporters (DMETs) can lead to major differences in drug exposure, potentially affecting efficacy and drug safety. In addition, gene expression and function may change with age, and developmental changes within the pediatric age range can result in specific drug exposures and effects in children [1]. Therefore, PGx clinical testing maybe particularly important in pediatric practice to help guide whether to select drugs for therapeutic alternatives and to adjust the initial dose of target drugs [2].

It is important to validate pharmacogenomic associations in children rather than extrapolating data solely from adults. Although pharmacogenetic studies are much

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harder to conduct in children for well-known reasons such as ethical considerations, there are several drugs with some pediatric evidence revealing significant and commonly available drug–gene interactions. This review will focus on pediatric evidence for associations of pharmacogenetics in DMET genes with PK characteristics and clinical outcomes for certain drugs that are commonly prescribed by clinicians across multiple therapeutic areas in the pediatric field, organized from the most to the least pediatric evidence among each drug category. From the perspective of a general hospital, we excluded several specialty drugs such as psychotropic drugs that are usually prescribed more by psychiatrists. More critically, pharmacogenetics in child psychopharmacology have been summarized in more detail in the corresponding specialized reviews [3, 4]. In fact, dose adjustment recommendations for escitalopram and sertraline based on CYP2C19 metabolizer status are well supported by some studies in adolescents and recommended by the Clinical Pharmacogenetics Implementation Consortium (CPIC) [4, 5]. Genetic studies of gene pharmacodynamics and other non-genetic factors contribute to risk prediction, but are beyond the scope of this review. We will also review examples of how pharmacogenomic discoveries in children have demonstrated clinical utility in guiding drug dosing, discuss how to implement these PGx testing results clinically, and finally provide a broad overview of recommendations for gene-based dose optimization for children.

## 2 Pharmacogenomics Research Progress and Clinical Implementation in Children

Here, based on limited and potentially underpowered pediatric-specific evidence, we focus on the role of pharmacogenetics and gene-based dose adjustment regimens in pediatric personalized medicine, and the current situation of clinical application. The summary of these original studies focusing on children with several details, including authors, research design, sample size, and ethnicity, is reported in Table 1. Specific studies are described below in the Sects. 2.1. to 2.5. focusing on the genes coding for transporters, metabolizing among various drugs.

### 2.1 Anti-Infective Drugs

#### 2.1.1 Voriconazole (VCZ)

Voriconazole (VCZ) is primarily inactivated by the CYP2C19 metabolic enzyme, with minor contributions by CYP3A and CYP2C9. The genetic polymorphisms of *CYP2C19, CYP2C9,* and *CYP3A4* were closely related to the large variations of the VCZ plasma concentrations in adults [6]. And genes encoding for drug transporters *ABCC2*, *ABCG2*, and *SLCO1B3* have a suspected role in voriconazole pharmacokinetics [7].

**2.1.1.1 Pharmacogenetics of VCZ Affecting Pharmacokinetics and Clinical Outcomes** The *CYP2C19* genetic variants affecting CYP2C19 enzymatic activity are significantly related to the high variability in VCZ trough concentrations ($C_0$). Specifically, $C_0$ of patients with **2* or **3* allele [6] were significantly higher than that with wild-type carriers and the *CYP2C19*17* polymorphism accelerated the effect on the PK parameters of VCZ [8]. In addition, CYP2C19 intermediate metabolizers (IMs) and poor metabolizers (PMs) have elevated VCZ plasma concentrations and ultra-rapid metabolizers (UMs) have decreased concentrations when compared with normal metabolizers (NMs) [9–11]. However, a population pharmacokinetic (PPK) analysis with a small sample size ($n = 21$) has suggested that the *CYP2C19* phenotype did not have a clinically relevant effect on voriconazole exposure in Japanese pediatric subjects. And yet, as the number of subjects ($n = 2$) with CYP2C19 PM status was limited and the information on which alleles were genotyped or how authors interpreted genotype to phenotype was unclear in the study, the evidence may not be powered to support the negative result [12].

The variability of VCZ plasma concentrations was also related to *CYP2C9*2* and *CYP2C9*3* alleles [6]. The evidence that *CYP2C9*3* was strongly associated with differences in voriconazole plasma concentrations was reported in adults but not in children. The *CYP2C9*2* allele was not tested in several studies because of the low frequency in Asian populations (< 0.1%) [6, 13]. Similarly, the *CYP3A4*22* (with a frequency of 5–7% in the Caucasian population [14]) and rs4646437,which were discovered through the association with low CYP3A4 activity, are associated with higher plasma VCZ $C_0$ in adults, while little data is available in pediatric patients [6, 15]. Additionally, Allegra et al. [7] suggested that the genotype groups of *SLCO1B3* rs4149117 and *ABCB1* rs1045642 significantly influenced VCZ $C_0$. Effects on $C_0$ were also significant for the variants *ABCG2* rs2231142 (enhancing) and *ABCC2* rs2273697 (reducing) in a retrospective study with limited subjects ($n = 36$) [16]. However, this remains to be further elucidated because the subgroup or overall sample size of these studies is relatively small, and there were also some negative results that did not support the effect of these genetic polymorphisms [15, 17].

Clinically, different studies showed inconsistent association between voriconazole $C_0$ and clinical outcomes. Most studies reported no statistically significant association between mean $C_0$ values and treatment response and severe toxicity in children, for example, the majority of patients

showed clinical improvement regardless of voriconazole trough levels [11, 18, 19]. However, the correlation is still noteworthy as Hicks et al. ($n = 33$) found that adverse effects such as neurotoxicity and hepatotoxicity were more common in patients with higher trough plasma concentrations [9].

**2.1.1.2 Gene-Guided Dosing Regimens and Clinical Implementation** Age is an important consideration in the administration of VCZ as physiological development is associated with the level of maturation of certain enzymes or transporter activities. Therefore, until now, it has been difficult to accurately predict voriconazole dosing due to the tremendous variability in VCZ pharmacokinetics in children. The CPIC [20] and Royal Dutch Association for the Advancement of Pharmacy Pharmacogenetics Working Group (DPWG) [21] have developed clinical guidelines for VCZ dose adjustment based on *CYP2C19* genotype. Clinical decision support for CYP2C19-based VCZ dosing is

**Table 1** Evidence for gene–drug interactions in pediatrics

| Drug | Study | Sample size of target populations, *n* | Ethnicity/race/nationality, *n* | Study design |
|---|---|---|---|---|
| **Anti-infective drugs** | | | | |
| Voriconazole | Fan et al. [6] | 68 | Chinese | Prospective |
| | Espinoza et al. [8] | Group 1: 232;<br>Group 2: 33 | Chilean | NA |
| | Hicks et al. [9] | 33 | African, 6<br>European, 23<br>Hispanic, 2<br>Multiple races, 2 | Retrospective |
| | Chen et al. [10] | 91 | Chinese | Retrospective |
| | Narita et al. [11] | 37 | Japanese | Retrospective |
| | Muto et al. [12] | 21 | Japanese | PPK analysis |
| | Chuwongwattana et al. [15] | Age <12 years: 31;<br>Age ≥12 years: 146 | Thai | Retrospective |
| | Tilen et al. [16] | 36 | NA | Retrospective |
| | Takahashi et al. [17] | 58 | White, 46<br>Black, 5<br>Native American, 3<br>Mixed/unknown, 4 | PPK analysis |
| | Liu et al. [18] | 107 | Chinese | Retrospective |
| | Tian et al. [19] | Pilot study: 25;<br>Genotype-directed study: 20 | NA | Prospective |
| | Teusink et al. [22] | 28 | Hispanic | Retrospective |
| | Karlsson et al. [24] | 82 | Caucasian, 57<br>Black, 6<br>Asian, 4<br>Other, 15 | PPK analysis |
| Efavirenz | Salem et al. [26] | 96 | Non-Hispanic, White, 12<br>Non-Hispanic, Black, 54<br>Hispanic, 28<br>Native American, 1<br>Other, 1 | Prospective |
| | Luo et al. [27] | 168 | White, 56<br>Black, 88<br>Other, 24 | PPK analysis |
| | Bienczak et al. [28] | 169 | African | PPK analysis |
| | Sinxadi et al. [29] | 54 | Black South African | NA |
| | Liu et al. [30] | 39 | Chinese | NA |
| | Saitoh et al. [31] | 71 | Non-Hispanic, White, 8<br>Non-Hispanic, Black, 46<br>Hispanic, 16<br>Native American, 1<br>Other, 1 | Retrospective |
| | Puthanakit et al. [32] | 63 | Thai | Retrospective |

**Table 1** (continued)

| Drug | Study | Sample size of target populations, *n* | Ethnicity/race/nationality, *n* | Study design |
|---|---|---|---|---|
| **Immunosuppressive agents** | | | | |
| Tacrolimus | Andrews et al. [34] | 46 | Caucasian, 34<br>Asian, 2<br>Black, 6<br>Other, 4 | PPK analysis |
| | Billing et al. [35] | 37 | NA | Prospective |
| | Prytuła et al. [36] | 54 | Caucasian, 38 | PPK analysis |
| | Guy-Viterbo et al. [37] | 114 | Black, 4<br>Caucasian, 110 | PPK analysis |
| | Mo et al. [38] | 167 | Chinese | Retrospective |
| | Li et al. [39] | 108 | Chinese | PPK analysis |
| | Gijsen et al. [40] | 43 | NA | Retrospective |
| | Knops et al. [41] | 43 | Caucasian, 39<br>Black-African, 2<br>North-African, 1<br>Asian, 1 | Retrospective |
| | Yang et al. [42] | 136 | Chinese | Retrospective |
| | Min et al. [46] | 53 | White/Caucasian, 38<br>Asian, 11<br>Black, 1<br>Mixed, 3 | Prospective |
| MMF | Krall et al. [48] | 104 | Chilean | Prospective |
| | Zhao et al. [49] | 89 | NA | PPK analysis |
| | Fukuda et al. [50] | 32 | Caucasian, 29<br>African American, 3 | Prospective |
| | Ohmann et al. [51] | 59 | White, non-Hispanic, 51<br>White, Hispanic, 2<br>Black, non-Hispanic, 6 | Retrospective |
| | Burckart et al. [52] | 290 | White, 215<br>Black, 54<br>Other, 21 | NA |
| | Prausa et al. [53] | Matched cases 38;<br>Controls 38 | White, 59<br>Black, 13<br>Native American, 1<br>Other, 4 | Retrospective |
| | Varnell et al. [54] | 38 | NA | Prospective |
| Cyclosporine | Li et al. [55] | 86 | Chinese | PPK analysis |
| | Cvetković et al. [56] | 47 | Serbian | NA |
| | Turolo et al. [57] | 87 | Italian | Retrospective |
| | Fanta et al. [58] | 104 | Caucasian, 103 | PPK analysis |
| | Yanagimachi et al. [59] | 63 | NA | Retrospective |
| **Chemotherapeutic agents** | | | | |
| Thiopurine | Relling et al. [62] | 180 | NA | NA |
| | Moriyama et al. [64] | 5 | East Asian, 3;<br>African, 1;<br>European, 1 | NA |
| | Tanaka et al. [65] | 92 | Japanese | Retrospective |
| | Lennard et al. [69] | 1334 | White, 1160;<br>Other, 174 | Prospective |

**Table 1** (continued)

| Drug | Study | Sample size of target populations, *n* | Ethnicity/race/nationality, *n* | Study design |
|---|---|---|---|---|
| Anthracycline | Visscher et al. [71] | Dutch-EKZ 128;<br>Canadian-CPNDS 90 | NA | Retrospective |
| | Visscher et al. [72] | discovery cohort 156;<br>replication cohort 188;<br>Dutch-EKZ 96 | NA | Retrospective |
| | Krajinovic et al. [73] | 251 | Caucasian | NA |
| | Semsei et al. [74] | 235 | NA | Retrospective |
| | Visscher et al. [75] | Discovery 344;<br>Replication 218 | NA | Retrospective |
| Busulfan | Nava et al. [77] | 112 | Caucasian, 94<br>African, 12<br>Other, 6 | PPK analysis |
| | Ansari et al. [78] | 44 | Middle East, 44 | Retrospective |
| | Ten Brink et al. [79] | 84 | NA | Retrospective |
| | Yuan et al. [80] | Model-Building Cohort, 69;<br>External Validation Cohorts, 14 | Chinese | PPK analysis |
| | Elhasid et al. [81] | 18 | Caucasian | Retrospective |
| | Nava et al. [82] | 101 | White, 80<br>Black, 11<br>Other, 10 | Retrospective |
| Vincristine | Ceppi et al. [83] | 339 | Caucasian | NA |
| | Guilhaumou et al. [84] | 26 | Caucasian | PPK analysis |
| | Moore et al. [85] | 50 | Australian | PPK analysis |
| | Egbelakin et al. [86] | 107 | Caucasians, 105<br>African-American, 1<br>Asian, 1 | NA |
| | Renbarger et al. [87] | 113 | Caucasians, 92<br>African-Americans, 21 | Retrospective |
| | Aplenc et al. [88] | 533 | African–American, 31;<br>Other, 502 | Prospective |
| | Wright et al. [89] | Cases 167;<br>Controls 57 | European, 175 | Retrospective |
| **Antiepileptic drugs** | | | | |
| VPA | Budi et al. [90] | Control group 47;<br>CYPtest group 51 | NA | Prospective |
| | Guo et al. [91] | 98 | Chinese | Retrospective |
| | Algharably et al. [92] | 48 | Egyptian | Prospective |
| | Xu et al. [93] | 264 | Chinese | PPK analysis |
| | Nandith et al. [94] | 99 | South Indian | Prospective |
| | Banawalikar et al. [95] | 65 | South Indian | Prospective |
| | Feng et al. [96] | 174 | Chinese | Prospective |
| | Tóth et al. [98] | 50 | Caucasian | Retrospective |
| | Noai et al. [99] | 85 | Japanese | Retrospective |
| | Mei et al. [100] | 290 | Chinese | PPK analysis |
| | Inoue et al. [101] | 78 | Japanese | PPK analysis |
| CBZ | Kim et al. [106] | 146 | NA | NA |
| | Zhang et al. [108] | 117 | Chinese | Retrospective |
| | Hung et al. [109] | 91 | NA | NA |
| | Ueda et al. [110] | 192 | Japanese | Retrospective |

**Table 1** (continued)

| Drug | Study | Sample size of target populations, *n* | Ethnicity/race/nationality, *n* | Study design |
|---|---|---|---|---|
| **Gastrointestinal drugs** | | | | |
| PPI | Kearns et al. [112] | Study 1: 24<br>Study 2: 19 | White, 27<br>Black, 8<br>Hispanic, 6<br>Other, 2 | NA |
| | Shakhnovich et al. [113] | 40 | White, 20<br>Black/African American, 12<br>Asian, 1<br>Other, 7 | Prospective |
| | Gumus et al. [114] | 244 | Turkish | Retrospective |
| | Franciosi et al. [115] | 74 | White, 53 | Retrospective |
| | Bernal et al. [116] | 670 | White, 553<br>African American, 76<br>Asian American, 11<br>Other or unknown, 30 | Retrospective |
| | Lima et al. [117] | 279 | White, 107<br>African American, 139<br>Asian, 5<br>Other, 28 | Prospective |
| | Kearns et al. [118] | Pantoprazole cohort 40;<br>Omeprazole cohort 23 | NA | Retrospective |
| | Cicali et al. [119] | Conventional dosing, 30;<br>Genotype-guided dosing, 30 | White, 47<br>African American, 4<br>Other, 9 | Prospective |
| | Tang et al. [120] | 64 | NA | Prospective |

also available via the Pharmacogenomics Knowledge Base (PharmGKB) website (https://www.pharmgkb.org/). Moreover, limited and inconsistent studies are available describing the relationship between *CYP2C19* polymorphisms and dose requirements in pediatric patients [9, 17, 19, 22]. And the researchers who completed these studies all developed various algorithms with genotype-directed initial administration of VCZ. For example, Takahashi et al. suggested the following doses to attain target $C_0$: 16 mg/kg (weight of < 15 kg) for NMs, 33 to 50% lower for PMs and 25 to 50% higher for UMs. These pediatric-specific recommendations remain incomplete, which is partly due to a lack of sufficient data demonstrating a difference between *CYP2C19* normal and rapid metabolizers in children [20].

*CYP2C19* genotype-guided voriconazole dosing has been implemented by some medical centers to study clinical suitability, and the results demonstrated benefit [9, 22, 23]. For example, adjusting voriconazole dosing based on CYP2C19 metabolizer status in pediatric patients resulted in a significant reduction in the time required to achieve target drug concentrations [22]. However, whether the preemptive genotype-directed dosing should be recommended in pediatric patients in clinical practice needs further study. A population pharmacokinetics (PPK) analysis reported that, individually, dose adjustments based only on *CYP2C19* genotype appear to offer no improvement in terms of exposure distribution over the unadjusted dose [24], similar to the findings of Tian et al. [19]. Some researchers have concluded that the *CYP2C19* genotyping status alone does not accurately forecast voriconazole plasma concentrations in patients, therefore, a combination of PGx and therapeutic drug monitoring (TDM) strategies for VCZ individualization can be of great benefit for patients [23, 25].

### 2.1.2 Efavirenz (EFV)

Efavirenz (EFV) is cleared primarily in the liver and these metabolic reactions are catalyzed exclusively by the CYP2B6 enzyme. It has been demonstrated that activation levels of CYP2B6 show broad inter-individual variation.

**2.1.2.1 Pharmacogenetics of EFV Affecting Pharmacokinetics and Clinical Outcomes** Most studies demonstrated that the TT genotype of *CYP2B6* rs3745274 had a significant elevated systemic exposure and reduced effect on the clearance (CL) of EFV compared with the GG and GT genotypes, and subsequently some researchers have constructed PPK models to quantify the effects on EFV clearance of

the *CYP2B6* rs3745274 (occurring in 3–6% of Caucasians and 16–20% of African-Americans) and rs28399499 (a less frequent variant found almost exclusively in Africans) variants [26–29]. Limited data are currently available on antiretroviral pharmacogenomics in pediatric Asian populations [30]. In addition, it has been demonstrated that a composite *CYP2B6* genotype based on *CYP2B6* rs3745274, rs28399499, and rs4803419 best described EFV exposure in HIV-infected African adults and children [29]. No effect of any variant of the drug transporter p-glycoprotein (P-gp/MDR1, encoded by *ABCB1*) was observed in related studies [26, 30].

Clinically, no association was found between *CYP2B6* rs3745274 polymorphisms and virologic or immunologic responses, toxicity, side effects, or the development of viral resistance against EFV [31, 32].

**2.1.2.2 Gene-Guided Dosing Regimens and Clinical Implementation** The CPIC guideline has provided therapeutic recommendations for efavirenz prescribing based on *CYP2B6* genotypes in three groups of children (aged $<3$ years, aged $>3$ years and weighing $<40$ kg, weighing $\geq 40$ kg) [33]. Also, some PPK studies have established models to construct dosing guidelines taking into account genotype and other factors such as body weight (the most influential covariate for CL in Luo's model), age, and prior antiretroviral therapy [26, 27]. In particular, Bienczak et al. [28] proposed a dosing optimization strategy for African children between the four metabolic subgroups based on optimal ratios of 1:0.66:0.33:0.1, by analyzing existing data from 169 children. To our knowledge, feedback on the current dosing recommendations clinically used in specific pediatric cases is not yet known. Thus, as shown in the abovementioned studies, genotype appeared to be an important potential predictor for many children, but it could not be considered an absolute indicator of dosing needs.

## 2.2 Immunosuppressive Agents

### 2.2.1 Tacrolimus (TAC)

Tacrolimus is primarily metabolized by the cytochromes 3A4 and 3A5 (CYP3A4 and CYP3A5), of which CYP3A5 is the most important metabolic enzyme. P450 oxidoreductase (POR) is the protein that enables the activity of CYP enzymes through a certain mechanism. TAC is also a substrate for the drug transporter P-gp/MDR1.

**2.2.1.1 Pharmacogenetics of TAC Affecting Pharmacokinetics and Clinical Outcomes** It is clear from the data that the *CYP3A5* genetic variants are significantly related to the variability of TAC plasma concentrations in pediatric patients [34–36]. More specifically, the carriers of the *CYP3A5*1* allele (*CYP3A5* expressers) have lower dose-corrected tacrolimus $C_0$ compared with non-carriers (*CYP3A5*3/*3* genotype) [34]. Of note, the effect of *CYP3A5*1* polymorphism and weight on TAC $C_0$ is cumulative [36]. Additionally, the impact of *CYP3A4* genetic polymorphism on the PK of TAC remains unclear due to limited evidence. *CYP3A4*22* SNP frequency is relatively low in whites (3–8%), but nonetheless this allele is an interesting candidate for the exploration of potentially altered CYP3A4 expression since T allele carriers significantly increased TAC plasma $C_0$ in children (with PharmGKB level 1B evidence) [37]. The *CYP3A4*1G* allele is associated with a higher TAC exposure in adults whereas no correlation was found in pediatrics [34, 38, 39].

Some studies have described the interactions between the effect of the *CYP3A5* gene and other genotypes. Interestingly, *POR*28* single nucleotide polymorphisms (SNPs) appear to exert isoform-specific effects on CYP activity. A small cohort study of pediatric kidney transplant recipients ($n = 43$) reported that the *CYP3A5* expressers carrying at least one *POR*28* allele had lower tacrolimus concentrations compared with the *CYP3A5* expressers carrying *POR*1/*1* [40], but this finding was not replicated, probably because the sample size of subjects with *POR*28* was limited ($n = 1$) and the authors did not have enough observations to study its effect [41]. Also, *CYP3A5* combined with a CC genotype of *ABCB1* rs1128503 showed a higher overall contribution to the attributable variance in TAC pharmacokinetics than *CYP3A5* alone because an *ABCB1* rs1128503 genotype may heighten the effects of *CYP3A5* [42].

Whether these gene polymorphisms are associated with transplant-related clinical outcomes remains controversial. The last serum creatinine (SCr) levels, graft function, or the incidence of adverse drug events (ADEs) did not significantly differ between *CYP3A5* expressers and non-expressers, nor between carriers of at least one *POR*28* allele or *POR*1/*1* allele [35, 40]. Yang et al. found that those recipients with the CC genotype of *ABCB1* rs1128503 presented with a high incidence of acute rejection and transplant-related infection, despite their much lower TAC concentrations [42].

**2.2.1.2 Gene-Guided Dosing Regimens and Clinical Implementation** A CPIC guideline exists for *CYP3A5*-based dosing of tacrolimus, recommending a 1.5- to 2-fold increase in dose for children and adolescents with at least one *CYP3A5*1* allele, similar to the recommendations for adults [43]. We need to be careful to consider the patient's ethnic background when administering medication because the frequency of *CYP3A5*3* SNPs is highly ethnicity dependent, presenting in the majority of Africans (45–73%) and in a minority of Caucasians (5–15%) [44]. For Asians, the

prevalence of *CYP3A5*3* occurred at around 15–35% [44]. Multiple studies have given specific dosing recommendations based on *CYP3A5* polymorphism and other non-genetic factors such as body weight, age, and donor type (deceased/living) for children [34, 36, 37, 39, 41], but these proposals are inconsistent. Among them, Li et al. were the first to distinguish the effects of wild type (*1*1), heterozygous (*1*3), and homozygous (*3*3) on the PK of TAC in pediatric subjects, and provided guidance on the daily dose of TAC based on each of the three genotypes [39]. Moreover, Elens et al. [45] suggested that the combination of *CYP3A4* and *CYP3A5* alleles may predict tacrolimus dose requirements better than either gene alone, which should be further studied.

Limited studies described whether the dose adjustments recommended in the abovementioned studies are clinically applicable. The PPK models in some studies have been externally validated in an independent dataset, providing a preliminary validation of the utility [36, 38]. Moreover, a prospective trial of 53 children demonstrated that *CYP3A5* genotype-guided dosing was safe and resulted in earlier attainment of target therapeutic concentrations with significantly fewer out-of-range concentrations than with standard dosing [46].

In general, researchers do recommend using *CYP3A5* genotype-guided dosing for patients with a known *CYP3A5* genotype to individualize initial tacrolimus treatment, especially in Chinese children [39]. However, the current evidence is limited to the effect of *CYP3A5* on tacrolimus pharmacokinetic parameters, with no direct and high-quality evidence for improved clinical outcome or toxicity [43]. Thus, few studies recommended whether or not to test for the *CYP3A5* genotype in advance.

#### 2.2.2 Mycophenolate Mofetil and Mycophenolate Sodium

Mycophenolate mofetil (MMF) is a prodrug that is rapidly metabolized to mycophenolic acid (MPA). MPA is mainly metabolized by uridine diphospho-glucuronosyltransferase (UGT) family members (particularly UGT1A9, UGT1A8 and UGT2B7) [47]. The efflux transporter MRP2 (multidrug-resistance protein 2 /ABCC2) is involved in the enterohepatic circulation of MPA.

There is less evidence regarding the effect of genetic variants on MPA pharmacokinetics than that associated with TAC pharmacokinetics, which might be partly attributed to the low frequency of genes encoding enzymes or transporters. In pediatric patients, polymorphisms in *UGT1A9*, *UGT2B7*, and *UGT1A8* may influence the metabolism and clearance [48–50]. The apparent oral clearance (CL/F) was significantly lower in patients with CC genotype of *UGT2B7* rs7439366 compared with patients with CT and TT genotypes, and this effect was independent of body weight [49]. In addition, Fukuda et al. [50] found that combined *UGT1A9*-440T>C, *UGT2B7* rs7438135, and *ABCC2* rs717620 polymorphisms might be important predictors of inter-individual variability in MPA exposure in Caucasian pediatric kidney transplant recipients aged 2–19 years ($n = 32$). However, a larger cohort is needed to continue validation of *UGT* polymorphisms since the results were not replicated in the same ethnic populations (Caucasians) in a pharmacogenetic substudy ($n = 37$) of Billing et al.'s randomized controlled trial (RCT) [35], and the PharmGKB level of evidence focusing on pediatrics is unavailable for the MMF-UGT association.

As for the transporters, studies on the impact of *ABCC2* polymorphisms have yielded mixed results, which need to be further studied. The effect of *ABCC2* polymorphisms on the pharmacokinetics of MPA is unclear. An evaluation of *ABCC2* variants (rs717620, rs2273697, rs8187694, and rs3740066) in an RCT study and a PPK analysis ($n = 89$) found no associations with MMF pharmacokinetics [35, 49]. The long-term implications of *ABCC2* polymorphisms on transplant outcome have rarely been reported previously. Some studies reported that the GG genotype of *ABCC2* rs717620 was protective against MMF discontinuation secondary to gastrointestinal side effects [51] and conferred increased risk of rejection and late rejection with hemodynamic compromise [52]. Additionally, MMF-related adverse events (e.g., leukopenia and diarrhea) in pediatric recipients were found to be associated with *UGT2B7* rs7438135 or *UGT1A9*-331C>T (with an increased risk with both) polymorphism [50, 53, 54]. Compared with adults, children and adolescents may be more susceptible to MMF-related leukopenia and/or the effects of SNPs due to maturation of these enzymes.

#### 2.2.3 Cyclosporine

Cyclosporine (CsA) is a substrate for CYP3A4 and CYP3A5 and P-gp. Most studies have focused on the impact of genetic variation in the genes which encode CYP3A4, CYP3A5, and P-gp on the pharmacokinetics of CsA, but the results are still contradictory. In this PPK study with 86 Chinese pediatric subjects, they reported that the *CYP3A4**1G genotype significantly influenced the cyclosporine CL with the result that the clearance rate of CsA in *CYP3A4**1GT allele carriers increased compared with that in *CYP3A4**1G CC carriers [55]. Meanwhile, similar to the effect of *CYP3A5* SNPs on TAC, it was suggested that overall CsA $C_0$/dose was significantly lower in CYP3A5 expressers compared with non-expressers (PharmGKB Level 3) in pediatric renal transplant recipients, including age as a covariate [56]. However, conflicting evidence has been reported. Some cohort studies with a larger sample size (including 87 teenagers and 104 pediatric patients) reported that *CYP3A5**3 was not

associated with variation in cyclosporine pharmacokinetics [57, 58]. In addition, some pediatric studies reported that the *ABCB1* genotype significantly influenced cyclosporine concentrations and the effect is age-dependent [57, 58], although in Li et al.'s study [55] the effect was inconsistent. The allele frequency of ABCB1 is greatly influenced by ethnicity, and the inter-racial influences in the Italian population are not as frequent as in other Europeans [57].

Clinically, *ABCB1* rs1128503 and rs2032582 were each independent risk factors for CsA-related neurotoxicity in children and adults, especially the CC genotype at *ABCB1* rs1128503, but its association was not statistically significant in children [58]. Also, no relationship between CsA-related neurotoxicity and the *CYP3A5* expresser genotype was detected [59]. More studies are required on the predictive value of genotyping for individualization of cyclosporine dosing in children.

## 2.3 Chemotherapeutic Agents

### 2.3.1 Thiopurine Drugs

Azathioprine activation involves conversion to mercaptopurine via metabolism by the glutathione S-transferase (GST) family. Then, thiopurine *S*-methyltransferase (TPMT) catalyzes the methylation of 6-mercaptopurine (6-MP) and its downstream metabolites. NUDT15 dephosphorylates the active 6-thioguanine nucleotide (6-TGN) back to the less toxic forms, therefore reducing thiopurine cytotoxicity [60].

**2.3.1.1 Pharmacogenetics of Thiopurines Affecting Pharmacokinetics and Clinical Outcomes** Pharmacogenomics of thiopurines (6MP and 6TG) with *TPMT* is probably the most studied drug–gene interaction in pediatric medicine. Alleles *2, *3A,*3B, and *3C of *TPMT* are by far the most common variants and represent approximately 90% of low and intermediate TPMT activity in Caucasians. The *3A allele was more common in Caucasians (3.9%) and the *3C allele more common in Africans (3.5%) and Asians (0.7–2.5%) [61]. It is relatively clear that this drug–gene association has important clinical implications because the treatment outcome of childhood acute lymphoblastic leukemia (ALL) with 6MP is highly associated with genetic polymorphism in *TPMT* [62].

The association of certain *NUDT15* alleles with adverse reactions to thiopurines in children has been replicated in some studies [63–65]. The NUDT15 PM phenotype is largely restricted to Asian people with a frequency of about one in every 50 patients, which had hitherto been rarely reported in European or African patients [63, 66]. For example, the *NUDT15**2 allele, a 6-MP toxicity-related locus discovered in Asians, was associated with thiopurine-related hematopoietic toxicity in several studies of 6MP [63, 65], and further studies identified a similar toxicity profile for azathioprine and 6TG. Additionally, data on numerous novel potential pharmacogenomic markers relevant for optimization of thiopurine treatment are still controversial, such as *ABCC4* and *GSTM1* [67].

**2.3.1.2 Gene-Guided Dosing Regimens and Clinical Implementation** Multiple studies in pediatric populations have shown that the evidence in children does not deviate from that seen in adults [68]. Currently, *TPMT* and *NUDT15* pharmacogenomic testing is applied in pediatric care, contributing to the reduction of thiopurine-induced toxicity [67]. Thiopurine pharmacogenomics has been demonstrated to be one of the best examples of successful application of pharmacogenomics in pediatrics [67]. The CPIC guideline [63] on thiopurines provided recommendations for initial dose selections as a function of both *TMPT* and *NUDT15* genotypes, which recommended that TPMT IMs receive 30 to 70% of the full dose, and PMs receive 10% of the full dose three times per week to avoid ADRs. The *NUDT15* recommendations parallel those for *TPMT*: for NUDT15 IMs and those with variants with uncertain functional activity, reduced dosing is also recommended.

Preemptive testing of *TPMT* and *NUDT15* genes has been clinically implemented to achieve a balance between efficacy and toxicity [60]. All active Children's Oncology Group protocols for acute lymphocytic leukemia currently recommend testing for *TPMT* variants at diagnosis and adjusting initial doses of thiopurine drugs accordingly [69]. Given the comparable impact of these variants with risk alleles in *TPMT,* similar benefits are expected with pre-emptive *NUDT15* genotyping, especially for Asian patients, while *NUDT15*-guided thiopurine dosing was considered to be of limited importance in Caucasians owing to the lower frequency of the known risk variant *NUDT15**2 [63, 64]. In addition, it is crucial to continuing to monitor patients treated with thiopurines for toxicity rather than interpreting normal metabolizer status for TPMT or NUDT15 as a guarantee against encountering significant toxicity [60, 62].

### 2.3.2 Anthracyclines

Anthracyclines are an important component of childhood cancer treatment, including doxorubicin and daunorubicin. However, their use is limited by anthracycline-induced cardiotoxicity (ACT). The solute carrier transporters (SLCs) play an essential role in the absorption and excretion and members of the ABC transporter family regulate the distribution of anthracyclines.

**2.3.2.1 Pharmacogenetics of Anthracyclines Affecting Pharmacokinetics and Clinical Outcomes** Some studies found that polymorphisms in genes encoding for ABC transport-

ers were associated with ACT. A recent review described the pharmacogenomic markers related to the development of ACT in childhood cancer patients [70] and it concluded that eight variants in five genes *(ABCB1, ABCB4, ABCC1, ABCC2, ABCC5)* were identified as predictors of risk [70]. Specifically, the findings were validated in some internal replication cohorts. For instance, Visscher et al. [71] reported two gender-dependent associations in the *ABCB4* gene (rs1149222 and rs4148808) that appear to be significant only among females. And it has been reported that *ABCB1* (rs2235047 and rs4148808) [71, 72], *ABCC5* rs7627754 [73], as well as *ABCC1* (rs3743527 and rs246221) [74] gene variants were associated with increased risk of ACT in pediatric ALL.

Also, variations in genes encoding SLC and UGT were found to be associated with anthracycline cardiotoxicity. Visscher et al. [72] have further replicated the association of the *SLC28A3* SNPs in an independent replication cohort ($n = 188$). The genetic variants in *SLC28A3* (rs7853758 and rs4877847), *SLC10A2* (rs9614091), and *SLC22A17* (rs4982753 and rs4149178) appear to confer a significant decreased risk of ACT and improve a genotype-guided risk prediction model, with replication in the second cohort [71, 72, 75]. Conversely, the rs6759892 and rs17863783 gene variants in *UGT1A6* have been found to be significantly associated with an increased risk of ACT [71, 72]. However, we should be cautious about the low-level evidence for these drug–gene associations (PharmGKB level 3 or 4). Together, combining these genetic variants with clinical risk factors may be possible to distinguish between those at higher and lower risk for development of ACT. If replicated in other populations, these findings may provide the basis for safer dosing of this widely used drug.

**2.3.2.2 Gene-Guided Dosing Regimens and Clinical Implementation** A number of genes have been identified through association studies of ACT. *SLC28A3* rs7853758 and *UGT1A6*4* currently have the strongest evidence as pharmacogenomic markers for ACT [70, 72]. These findings may altogether lead to prediction models to identify patients who might be highly susceptible to ACT and require treatment adjustment. Also these pharmacogenomics data have been utilized to develop evidence-based clinical practice recommendations by the Canadian Pharmacogenomics Network for Drug Safety, which recommends pharmacogenomic testing should be performed in childhood cancer patients with doxorubicin or daunorubicin therapy for *UGT1A6**4 (rs17863783) and *SLC28A3* rs7853758 variants [76]. To date, all association studies in ACT have been retrospective. Prospective studies are needed to better implement practice guidelines to mitigate the risk of ACT. Also, the limitations of the lack of data on ethnicity require further refinement.

### 2.3.3 Busulfan (Bu)

The only known metabolic pathway of busulfan (Bu) is its conjugation to glutathione, a reaction that is mainly catalyzed by the hepatic enzyme GST. Part of the PK variability of busulfan results from genetic variations in the enzyme-coding gene *GSTA1*.

**2.3.3.1 Pharmacogenetics of Bu Affecting Pharmacokinetics and Clinical Outcomes** Several studies reported that polymorphic expression of metabolic enzymes (especially GSTA1) influenced busulfan CL [77, 78]. The potential effect of a haplotype in CYP39A1 in busulfan PK is newly discovered, but the role in busulfan metabolism needs further clarification [79]. A study with 84 pediatric patients reported that both *GSTA1* and *CYP39A1* genotypes were associated with busulfan CL and they together could account for up to 17% of the variability in pediatric patients [79]. To be specific, patients who were heterozygous for *GSTA1**A/*B or homozygous *B/*B had a lower busulfan CL compared with the wild-type genotype *GSTA1**A/*A [80], and patients who were carriers of one of the variant *CYP39A1**TC alleles or homozygous patients had a lower CL compared with *CYP39A1**WT/*WT patients [79]. Also, the effect of *GSTA1* haplotype on CL may be dependent on age, with the *GSTA1* haplotype having a larger influence in younger children [79].

Clinically, some studies concluded that patients with *GSTA1* genotypes (*GSTA1**B, *GSTA1**B/*B, and *GSTA1**B1/*B1) had increased risk of SOS (sinusoidal obstructive syndrome) [80, 81]. Ansari et al. reported that GSTA1 slow metabolizers had increased TRT (transplant-related toxicity) [78]. Nevertheless, these associations still need to be confirmed by more studies as a PharmGKB level of evidence has not been given, probably due to the low quality of evidence.

**2.3.3.2 Gene-Guided Dosing Regimens and Clinical Implementation** Currently, depending on a patient's *GSTA1* diplotype group, busulfan first-dose tailoring can be estimated from doses obtained from currently available weight- and/or age-based guidelines. The inclusion of the *GSTA1* diplotype groups as a covariate in a novel pharmacogenetics-based PPK model is recommended to improve dose prediction in many studies [77, 82]. Interestingly, *GST* polymorphism frequencies are highly heterogeneous in the Israeli population. *GSTA1*A/*A* wild-type variant was less common in individuals of Muslim descent compared with those of Jewish or Druze, while mutant allele *GSTA1*B* was more common in Moslems than in Jews and Druze [81]. However, to our knowledge, no studies give specific recommendations for gene-based dosing. Meanwhile, the high burden of concomitant medication interactions and the cost

of performing genomic testing may limit the utility of pharmacogenomic testing.

#### 2.3.4 Vincristine

To date, most pharmacogenomics studies on the effect of vincristine-induced peripheral neuropathy (VIPN) in children have emphasized DNA sequence variations. Given that vincristine is predominantly metabolized by CYP3A5, several groups have investigated the role of the CYP3A subfamily in VIPN in terms of inter-individual differences in pharmacogenetics related to drug bioavailability, clearance, efficacy, and toxicity. The effect of gene polymorphism on patient susceptibility to developing neurotoxicity is not clear and data have been mostly controversial. A majority of the studies disagree with regards to the association of lower vincristine clearance in CYP3A5 poor metabolizers with increased risk for VIPN [83–85], but this association was observed definitely in other studies [86–88]. Egbelakin et al. [86] reported CYP3A5 expressers have a significantly reduced risk of VIPN compared with CYP3A5 non-expressers in children with ALL. Also, there was a study showing patients with the *CYP3A4**1B and *CYP3A5*3* genotypes had a decreased risk of peripheral neuropathy that was statistically significant on univariate analysis [88]. Besides, considering that 70% of African Americans (vs 20% of Caucasians) express *CYP3A5*, a hypothesis could be proposed that vincristine is metabolized more efficiently in those of African American ethnicity, leading to reduced vincristine exposure and associated toxicity [87]. This has been subsequently proven by Egbelakin et al., who reported that vincristine-related neurotoxicity was much more frequent and more severe in Caucasians than in African–Americans [86].

Beyond the impact of the CYP3A subfamily on vincristine metabolism, significant associations between VIPN and variants of *ABCB1* [83], *ABCC1* (rs3784867), and *SLC5A7* (rs1013940) [89] have been found in multiple studies. But the genetic variants in the *ABCB1* gene alone cannot explain the large variability in vincristine pharmacokinetics. For dose optimization of vincristine, additional studies are required to reconcile these inconsistencies, and replication studies with the higher level of evidence are needed to implement genotype testing combining *CYP3A5*, *SLCs* and *ABCs* as the level of current evidence is low.

### 2.4 Antiepileptic Drugs

#### 2.4.1 Valproic Acid

Children have a lower glucuronidase activity than adults as UGTs are developmentally regulated; therefore, a larger part of valproic acid (VPA) is liable to undergo CYP-dependent metabolism in children [90]. The main catalyst is CYP2C9, with minor contributions from CYP2A6 and CYP2B6. In addition, VPA needs to be transported by various transporters, such as ABCB1 and ABCC2.

**2.4.1.1 Pharmacogenetics of VPA Affecting Pharmacokinetics and Clinical Outcomes** Different studies have yielded different observations regarding the UGT gene polymorphisms and their effect on VPA concentrations. Several studies indicated that genetic variants in *UGT1A3/1A4/1A6/1A9* and *UGT2B7* may influence the pharmacokinetics of VPA in children [91, 92]. Specifically, the most common SNPs of *UGT1A6* (rs6759892, rs2070959, and rs1105879) [92] and *UGT2B7* (rs7668258 and rs7439366) have been reported to be associated with lower adjusted plasma VPA concentrations in epileptic children [91]. However, the associations remained controversial with low evidence (PharmGKB level 3) for pediatric patients as some studies reported that there was no significant effect of *UGT* gene variants on serum valproate concentrations [93, 94]. One reason for this contradictory result may be due to the fact that the pattern of *UGT1A6* gene polymorphisms varies slightly in different populations. For example, in the Chinese pediatric epilepsy population, the predominant pattern of genetic polymorphism observed is wild-type, whereas in the Indian population it is mutant type [95]. Little literature to date has explained the influence of *UGT* genetic variants on the clinical outcome of VPA monotherapy. A study of 174 Chinese children indicated that genetic polymorphisms of *UGT2B7* rs7668282 were associated with seizure reduction and resulted in a greater incidence of VPA-induced hepatotoxicity in younger children [96]. Evidence for the influence of *UGT1A6* polymorphic variants at rs2070959 and rs1105879 loci on ADR and seizure severity is conflicting [92, 95].

Some studies suggest it is children's CYP2C9-status rather than *CYP2C9* gene polymorphisms alone that better explains the variation in the serum VPA concentrations and susceptibility of some adverse reactions. Loss-of-function mutations in the *CYP2C9* gene or low CYP2C9 expression can be identified as risk factors for certain side effects [97, 98]. Of note, the *CYP2C9* *2 allele is present predominantly in Caucasians (16%), and the *3 allele is present in East Asians but is absent from the African population [61], which is particularly useful to identify ethnic groups with a higher proportion of reduced-function variants when VPA is prescribed. Additionally, whether *CYP2C19* gene polymorphisms have a significant impact on the PK profile of VPA in children remains unclear. Some researchers speculate that CYP2C19 heterozygous EM and PM genotypes may be associated with increased testosterone or progesterone levels, thus leading to weight gain, which is frequently reported as a side effect of VPA [99].

Of note, the effect of a single gene polymorphism on VPA metabolism is limited. It has been confirmed that the combined genotype *UGT-CYP* (such as the *UGT2B7* rs7668258 and the *CYP2C9**1/*1 genotype) has significant influence on VPA PK in some PPK models [100, 101], while no significant relationships between genetic variants in *CYPs* and *UGTs* and VPA CL/F were found in Xu's PPK model using data from 264 epileptic pediatric patients [93].

Taken together, the abovementioned studies suggested that the patients' CYP2C9 status plays a clinically relevant role in VPA efficacy and safety. The impact of genetic variants on VPA PK and clinical outcomes remains unclear to date.

**2.4.1.2 Gene-Guided Dosing Regimens and Clinical Implementation** Currently, CPIC guidelines are not available to guide the use of CYP2C9 metabolizer status for VPA selection and dose. Extrapolation of the valproate metabolic phenotype from *CYP2C9* genotype may lead to false predictions as non-genetic factors can significantly alter the drug metabolizing phenotype [98]. Tailored valproate therapy adjusted to the pediatric patients' CYP2C9 status (determined by *CYP2C9* genotype and *CYP2C9* expression) may be superior in reducing the risk of serious adverse reactions and predicting specific dose requirements [90, 97]. Monostory et al. [97] concluded that CYP2C9-status–guided valproic acid therapy can be recommended for children with at least one or two wild-type *CYP2C9* alleles. For example, dose reductions of valproic acid are recommended for the children with heterozygous genotypes (CYP2C9*1/*2 or CYP2C9*1/*3) or for low CYP2C9 expressers, whereas increased dose is proposed for high expresser patients with CYP2C9*1/*1 genotype [97]. Furthermore, CYP2C9-status–guided therapy has been successfully applied for VPA dose optimization in 99 pediatric patients, which significantly reduced the number of patients outside of the therapeutic range of serum VPA concentrations and those with toxic symptoms [90, 102]. However, the sample size of the CYP test group in this study seems to be too small ($n = 51$) to make firm conclusions that CYP2C9-status–guided VPA treatment is preferable to non–CYP2C9-guided treatment [90]. These findings still require replication.

### 2.4.2 Carbamazepine

The major route of carbamazepine (CBZ) metabolism depends on the activity of CYP3A4, CYP3A5, and CYP2C8. Also, several minor metabolic pathways of CBZ metabolism are catalyzed by CYP2B6 and CYP2A6, with a smaller contribution from CYP1A2 [103]. While CYP3A4 seems to be the most important player in this reaction, it is not considered very polymorphic [104]. On the other hand, CYP3A5 exhibits high genetic variability, with *CYP3A5**3 (rs776746) representing the most frequent and best studied variation leading to severely decreased enzyme activity [105]. Of note, absent from the PharmGKB evaluation is consideration of the CBZ-CYP3A5 association in pediatric patients. Although carbamazepine transport is not yet fully clarified, there are several transporters suggested to affect CBZ crossing the blood-brain barrier, including MDR1 and MRP2 [106].

Both CYP1A2 and CYP2C19 show considerable polymorphism. The association of *CYP1A2**1F and *ABCC2* rs2273697 with pharmacokinetics of CBZ has been reported (PharmGKB level 3 or 4) [105]. Genetic variations such as *CYP2C19**2, *CYP2C19**3, *UGT2B7**2, *ABCB1* rs1045642, and *ABCC2* rs717620 and rs3740066 have been deemed not to be important for response to CBZ in children, while the observed effect of *CYP3A5**3 and *CYP2C8*3* polymorphisms required additional studies to further clarify [105, 107, 108].

Clinically, early studies reported that no polymorphisms involved in CBZ metabolism were identified as pathogenic in adult patients [109]. A retrospective study of Japanese patients with epilepsy showed that the *GSTM1* null genotype was a risk factor for mild carbamazepine-induced hepatotoxicity [110]. However, no relevant results have been reported in the pediatric population to date.

## 2.5 Gastrointestinal Drugs

### 2.5.1 Proton Pump Inhibitors

Most first-generation proton pump inhibitors (PPIs) are primarily metabolized by CYP2C19, with CYP3A4 playing a minor role. The second-generation PPIs esomeprazole and rabeprazole are less CYP2C19 dependent in their metabolism [111].

PPI use in children is common and continues to increase, and some studies have suggested increased CYP2C19 function in children compared with adults [5]. Thus, the impact of *CYP2C19* genetic variability on PPI exposure and clinical outcomes should be carefully considered in the pediatric population. For pediatric patients, emerging evidence identified the *CYP2C19* genotype as a significant covariate for pantoprazole PK, which demonstrated that PMs and IMs have higher exposure, reduced clearance, and longer drug half-life compared with NMs [112, 113]. Furthermore, multiple clinical studies of children taking PPIs have shown the associations of *CYP2C19* function with efficacy and adverse events. *CYP2C19* RMs or UMs have been associated with reduced PPI efficacy compared with PMs and NMs, thus the *CYP2C19*2* and *17 variants should be taken into consideration in predicting the clinical outcome of therapy with PPIs in the pediatric population [114, 115]. Some studies showed that increased gastrointestinal and respiratory

infections were observed in PMs or NMs compared with those with increased *CYP2C19* function [116, 117], but additional studies are required to replicate these findings. The data for omeprazole is lacking compared with pantoprazole and some studies have shown no association between *CYP2C19* genotype and drug exposure for omeprazole [117, 118]. We can see that the results of studies with omeprazole and pantoprazole in pediatric patients may be inconsistent, but the reasons for this are not entirely clear. Relatively few subjects of a certain genotype may be a factor contributing to the lack of association between *CYP2C19* genotype and omeprazole exposure.

Some prospective clinical studies of CYP2C19 genotype-guided pediatric dosing of PPI therapy have confirmed that this genotype tailored treatment is promising and acceptable in a clinical pediatric setting and may reduce PPI-associated adverse effects [119, 120]. The CPIC guideline contains pediatric-specific dosing recommendations for PPIs based on CYP2C19 phenotype, which are the same as those for adults [111]. Taken together, these data support *CYP2C19* gene-guided dosing optimization of PPI therapy in children. Most interestingly, the distribution of *CYP2C19* alleles and genotypes shows wide interethnic differences. For example, the most prevalent genotype of CYP2C19 PM in Asians is *CYP2C19*2*, and *CYP2C19*3* with loss function is typically rare in the general population such as Caucasians and Africans (8% vs <1%), except Asians [20, 61]. In addition, considering the relatively high frequency of the *CYP2C19*17* allele in a Chilean population (12%) [8], RM/UM genotypes should be taken into consideration as well when genotyping *CYP2C19* to predict enzyme activity in individualized drug therapy.

## 3 Conclusion

Pharmacogenomics has shown evidence-based benefits in several therapeutic areas such as infectious disease, organ transplantation and immunosuppression, oncology, and neurology. To date, gene-guided dosing recommendations in pediatric patients have been provided for several drugs such as VCZ, EFV, TAC, VPA, PPI, and thiopurines, whereas the association of pharmacogenomics with PK variability for CsA, CBZ, MMF, and busulfan remains controversial with only low-level evidence (PharmGKB level 3 or 4). Furthermore, given the high likelihood that pediatric patients would benefit from pharmacogenomic-guided dosing, CPIC publishes guidelines covering pediatric-specific recommendations, and several guidelines have separate recommendations for children and adults when a high level of PharmGKB literature evidence (level 1 or 2) for the drug–gene association is available (e.g., VCZ, TAC, and PPI, etc.). Furthermore, researchers have attempted to apply the genotype-guided dosing regimens to real-world situations. Gene-guided VCZ dosing has been successfully applied in several medical institutions, and the clinical utility of *CYP3A5* gene-guided TAC dosing has been demonstrated in prospective trials, and preemptive testing of *TPMT* and *NUDT15* genes for thiopurines is proven to be one of the best examples of successful application of pharmacogenomics in pediatrics.

### 3.1 Future Perspectives

The complex physiologic and metabolic changes that occur during childhood provide the opportunity to discover pediatric-specific drug–gene interactions and complicate the implementation of these PGx testing results in pediatrics clinically. For example, some enzymes such as CYP2B6 and TPMT are expressed at relatively constant levels throughout childhood and into adulthood [121]. Therefore, it is worth considering whether genetic variation in these metabolic enzymes may have similar effects on the degree of PK variability in children and adults. Adult data are therefore frequently extrapolated to the use of drugs in children of specific developmental stages. Nevertheless, a simple extrapolation is not always suitable. CYP2C9, 2C19, 2D6, 3A4, as well as most of the UGTs have negligible activity at birth and reach adult activity within a few weeks (e.g., 2D6) to several years (until post-puberty for 2C9) after birth, so genotyping neonates and toddlers may not be meaningful [122]. We should reasonably extrapolate findings from adult studies to children, taking into account the maturation characteristics of each metabolic enzyme and transporter protein, and conduct more high-quality studies to validate pharmacogenomic associations in children.

It is well known that the frequencies of specific genotypes and haplotypes vary across ethnic populations, which might impact the dosage recommendations and limit the clinical utility of some PGx tests. Genetic variation affecting drug exposure has been most widely studied for genes in the CYP family, such as *CYP2C19*, *CYP2C9*, and *CYP2D6*. A similar ethnic difference was certainly seen in other metabolism genes (TPMT, UGT), as well as the SLCO1B1 transporters. These findings have wide-ranging clinical implications for adjusting dosing because variations in DMET genes are closely associated with response to drug therapy. A clear and systematic understanding of the inter-ethnic genetic differences in target genes is also therefore essential to guide effective global drug prescribing.

Recommendations for the implementation of pharmacogenomic tests appear to vary for different drugs, and challenges to clinical implementation of PGx in pediatrics still need further discussion. Probably the most important limitation to the implementation is the lack of adequately powered pediatric clinical trials of PGx-guided dosing for well-known reasons (e.g., ethical considerations, lack of

clinical utility, differences in gene expression and ontogeny across developmental stages), in addition to the high cost of genetic testing combined with the lack of accessibility, the limited training of prescribers, and the characteristics of the drug itself, etc. Preemptive genetic testing is essential for thiopurine drugs, which has been applied clinically in children as the potential adverse events (life-threatening cytopenias) are severe. Several studies mentioned herein have identified some SNPs associated with ACT in children with cancer, highlighting the value of pre-exposure genetic testing as well. However, pre-prescription pharmacogenetic testing appears non-urgent for most children receiving treatment with certain drugs (e.g., VCZ) whose therapeutic and toxic concentrations are well known to achieve target therapeutic outcomes through TDM strategies. Overall, this practice will ultimately help reduce healthcare costs when patients are initially treated with the drug most likely to be effective for them based on the results of PGx testing.

It is critical for medical practitioners to assess the evidence critically as more drug–gene interactions are demonstrated to have clinical validity for children. It can be seen that the clinical use of PGx testing in children remains uncommon due to various conflicting findings. Differences in study population, sample size, ethnicities, and concomitant medication might be possible confounding factors. Clinicians may adjust the dose based on other information (e.g., age, concurrent medications, renal and liver function) when the data on pharmacogenomics in children is unavailable, which is almost as important as adjusting based on PGx [123]. Additionally, to facilitate the implementation of pharmacogenomics into clinical practice, Practitioners should be professionally trained, and appropriate procedural tools need to be developed to help clinicians properly integrate pharmacogenomics into their clinical practice, such as CPIC and PharmGKB. Focusing on pediatric PGx and encouraging cost reductions in genotyping is urgent for individualized medicine in children. Future directions include the incorporation of additional genetic, epigenetic, and clinical risk factors to guide the frequency of dosing and biomarker monitoring of various medications, with the goal of eventually implementing practice guidelines for pediatric individualized administration.

Additionally, the clinical implementation of pediatric pharmacogenetics for drug dosing remains challenging due to a general lack of evidence-based recommendations. Our review reveals that, in most cases, few studies with clinically meaningful endpoints have been published in children and the majority of them have relatively small cohorts. We can't simply translate the results of these observational studies into clinical practice. In order to make use of such information, any identified association must be replicated in a validation cohort and the effect size must be convincing [124]. Real-world interventional data generated from the application of gene-guided pharmacotherapy regimens during routine care of pediatric patients can validate known pharmacogenetic findings better, propelling the field of pediatric pharmacogenetics towards clinical implementation. Adequately powered interventional clinical studies that support incorporation of pharmacogenetics into the care of children are strongly needed.

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## References

1. Marsot A, Boulamery A, Bruguerolle B, et al. Population pharmacokinetic analysis during the first 2 years of life: an overview. Clin Pharmacokinet. 2012;51:787–98. https://doi.org/10.1007/s40262-012-0015-8.
2. Stevens A, Hanson D, Whatmore A, et al. Human growth is associated with distinct patterns of gene expression in evolutionarily conserved networks. BMC Genom. 2013;14(1):547. https://doi.org/10.1186/1471-2164-14-547.
3. Namerow LB, Walker SA, Loftus M, et al. Pharmacogenomics: an update for child and adolescent psychiatry. Curr Psychiatry Rep. 2020;22(5):26. https://doi.org/10.1007/s11920-020-01145-4.
4. Ramsey LB, Namerow LB, Bishop JR, et al. Thoughtful clinical use of pharmacogenetics in child and adolescent psychopharmacology. J Am Acad Child Adolesc Psychiatry. 2021;60(6):660–4. https://doi.org/10.1016/j.jaac.2020.08.006.
5. Hicks JK, Bishop JR, Sangkuhl K, et al. Clinical pharmacogenetics implementation consortium (CPIC) guideline for CYP2D6 and CYP2C19 genotypes and dosing of selective serotonin

reuptake inhibitors. Clin Pharmacol Ther. 2015;98(2):127–34. https://doi.org/10.1002/cpt.147.
6. Fan X, Zhang H, Wen Z, et al. Effects of CYP2C19, CYP2C9 and CYP3A4 gene polymorphisms on plasma voriconazole levels in Chinese pediatric patients. Pharmacogenet Genom. 2022;32(4):152–8. https://doi.org/10.1097/fpc.0000000000000464.
7. Allegra S, Fatiguso G, Francia S, et al. Pharmacogenetic of voriconazole antifungal agent in pediatric patients. Pharmacogenomics. 2018;19(11):913–25. https://doi.org/10.2217/pgs-2017-0173.
8. Espinoza N, Galdames J, Navea D, et al. Frequency of the CYP2C19*17 polymorphism in a Chilean population and its effect on voriconazole plasma concentration in immunocompromised children. Sci Rep. 2019;9(1):8863. https://doi.org/10.1038/s41598-019-45345-2.
9. Hicks JK, Crews KR, Flynn P, et al. Voriconazole plasma concentrations in immunocompromised pediatric patients vary by CYP2C19 diplotypes. Pharmacogenomics. 2014;15(8):1065–78. https://doi.org/10.2217/pgs.14.53.
10. Chen J, Wu Y, He Y, et al. Combined effect of CYP2C19 genetic polymorphisms and c-reactive protein on voriconazole exposure and dosing in immunocompromised children. Front Pediatr. 2022;10: 846411. https://doi.org/10.3389/fped.2022.846411.
11. Narita A, Muramatsu H, Sakaguchi H, et al. Correlation of CYP2C19 phenotype with voriconazole plasma concentration in children. J Pediatr Hematol Oncol. 2013;35(5):e219–23. https://doi.org/10.1097/MPH.0b013e3182880eaa.
12. Muto C, Shoji S, Tomono Y, et al. Population pharmacokinetic analysis of voriconazole from a pharmacokinetic study with immunocompromised Japanese pediatric subjects. Antimicrob Agents Chemother. 2015;59(6):3216–23. https://doi.org/10.1128/aac.04993-14.
13. Yang JQ, Morin S, Verstuyft C, et al. Frequency of cytochrome P450 2C9 allelic variants in the Chinese and French populations. Fundam Clin Pharmacol. 2003;17(3):373–6. https://doi.org/10.1046/j.1472-8206.2003.00148.x.
14. Elens L, van Gelder T, Hesselink DA, et al. CYP3A4*22: promising newly identified CYP3A4 variant allele for personalizing pharmacotherapy. Pharmacogenomics. 2013;14(1):47–62. https://doi.org/10.2217/pgs.12.187.
15. Chuwongwattana S, Jantararoungtong T, Prommas S, et al. Impact of CYP2C19, CYP3A4, ABCB1, and FMO3 genotypes on plasma voriconazole in Thai patients with invasive fungal infections. Pharmacol Res Perspect. 2020;8(6): e00665. https://doi.org/10.1002/prp2.665.
16. Tilen R, Paioni P, Goetschi AN, et al. Pharmacogenetic analysis of voriconazole treatment in children. Pharmaceutics. 2022;14:6. https://doi.org/10.3390/pharmaceutics14061289.
17. Takahashi T, Mohamud MA, Smith AR, et al. CYP2C19 phenotype and body weight-guided voriconazole initial dose in infants and children after hematopoietic cell transplantation. Antimicrob Agents Chemother. 2021;65(9): e0062321. https://doi.org/10.1128/aac.00623-21.
18. Liu L, Zhou X, Wu T, et al. Dose optimisation of voriconazole with therapeutic drug monitoring in children: a single-centre experience in China. Int J Antimicrobial Agents. 2017;49(4):483–7. https://doi.org/10.1016/j.ijantimicag.2016.11.028.
19. Tian X, Zhang C, Qin Z, et al. Impact of CYP2C19 phenotype and drug-drug interactions on voriconazole concentration in pediatric patients. Antimicrobial Agents Chemotherapy. 2021;65(9): e0020721. https://doi.org/10.1128/aac.00207-21.
20. Moriyama B, Obeng AO, Barbarino J, et al. Clinical pharmacogenetics implementation consortium (CPIC) guidelines for CYP2C19 and voriconazole therapy. Clin Pharmacol Ther. 2017;102(1):45–51. https://doi.org/10.1002/cpt.583.
21. Swen JJ, Nijenhuis M, de Boer A, et al. Pharmacogenetics: from bench to byte–an update of guidelines. Clin Pharmacol Ther. 2011;89(5):662–73. https://doi.org/10.1038/clpt.2011.34.
22. Teusink A, Vinks A, Zhang K, et al. Genotype-directed dosing leads to optimized voriconazole levels in pediatric patients receiving hematopoietic stem cell transplantation. Biol Blood Marrow Transplant. 2016;22(3):482–6. https://doi.org/10.1016/j.bbmt.2015.11.011.
23. García-García I, Dapía I, Montserrat J, et al. Experience of a strategy including CYP2C19 preemptive genotyping followed by therapeutic drug monitoring of voriconazole in patients undergoing allogenic hematopoietic stem cell transplantation. Front Pharmacol. 2021;12: 717932. https://doi.org/10.3389/fphar.2021.717932.
24. Karlsson MO, Lutsar I, Milligan PA. Population pharmacokinetic analysis of voriconazole plasma concentration data from pediatric studies. Antimicrob Agents Chemother. 2009;53(3):935–44. https://doi.org/10.1128/aac.00751-08.
25. Hyland R, Jones BC, Smith DA. Identification of the cytochrome P450 enzymes involved in the N-oxidation of voriconazole. Drug Metab Dispos. 2003;31(5):540–7. https://doi.org/10.1124/dmd.31.5.540.
26. Salem AH, Fletcher CV, Brundage RC. Pharmacometric characterization of efavirenz developmental pharmacokinetics and pharmacogenetics in HIV-infected children. Antimicrob Agents Chemother. 2014;58(1):136–43. https://doi.org/10.1128/aac.01738-13.
27. Luo M, Chapel S, Sevinsky H, et al. Population pharmacokinetics analysis to inform efavirenz dosing recommendations in pediatric HIV patients aged 3 months to 3 years. Antimicrob Agents Chemother. 2016;60(6):3676–86. https://doi.org/10.1128/aac.02678-15.
28. Bienczak A, Cook A, Wiesner L, et al. The impact of genetic polymorphisms on the pharmacokinetics of efavirenz in African children. Br J Clin Pharmacol. 2016;82(1):185–98. https://doi.org/10.1111/bcp.12934.
29. Sinxadi PZ, Leger PD, McIlleron HM, et al. Pharmacogenetics of plasma efavirenz exposure in HIV-infected adults and children in South Africa. Br J Clin Pharmacol. 2015;80(1):146–56. https://doi.org/10.1111/bcp.12590.
30. Liu X, Ma Q, Zhao Y, et al. Impact of single nucleotide polymorphisms on plasma concentrations of efavirenz and lopinavir/ritonavir in Chinese children infected with the human immunodeficiency virus. Pharmacotherapy. 2017;37(9):1073–80. https://doi.org/10.1002/phar.1988.
31. Saitoh A, Fletcher CV, Brundage R, et al. Efavirenz pharmacokinetics in HIV-1-infected children are associated with CYP2B6-G516T polymorphism. Journal of acquired immune deficiency syndromes (1999). 2007;45(3):280-5. http://doi.org/https://doi.org/10.1097/QAI.0b013e318040b29e.
32. Puthanakit T, Tanpaiboom P, Aurpibul L, et al. Plasma efavirenz concentrations and the association with CYP2B6-516G>T polymorphism in HIV-infected Thai Children [Article]. Antivir Ther. 2009;14(3):315–20.
33. Desta Z, Gammal RS, Gong L, et al. Clinical Pharmacogenetics implementation consortium (CPIC) guideline for CYP2B6 and efavirenz-containing antiretroviral therapy. Clin Pharmacol Ther. 2019;106(4):726–33. https://doi.org/10.1002/cpt.1477.
34. Andrews LM, Hesselink DA, van Gelder T, et al. A population pharmacokinetic model to predict the individual starting dose of tacrolimus following pediatric renal transplantation. Clin Pharmacokinet. 2018;57(4):475–89. https://doi.org/10.1007/s40262-017-0567-8.
35. Billing H, Höcker B, Fichtner A, et al. Single-nucleotide polymorphism of CYP3A5 impacts the exposure to tacrolimus in pediatric renal transplant recipients: a pharmacogenetic substudy

of the TWIST trial [Article]. Ther Drug Monit. 2017;39(1):21–8. https://doi.org/10.1097/FTD.0000000000000361.
36. Prytuła AA, Cransberg K, Bouts AHM, et al. The effect of weight and CYP3A5 genotype on the population pharmacokinetics of tacrolimus in stable paediatric renal transplant recipients [Article]. Clin Pharmacokinet. 2016;55(9):1129–43. https://doi.org/10.1007/s40262-016-0390-7.
37. Guy-Viterbo V, Baudet H, Elens L, et al. Influence of donor-recipient CYP3A4/5 genotypes, age and fluconazole on tacrolimus pharmacokinetics in pediatric liver transplantation: a population approach. Pharmacogenomics. 2014;15(9):1207–21. https://doi.org/10.2217/pgs.14.75.
38. Mo X, Li J, Liu Y, et al. Kidney podocyte-associated gene polymorphisms affect tacrolimus concentration in pediatric patients with refractory nephrotic syndrome. Pharmacogenom J. 2020;20(4):543–52. https://doi.org/10.1038/s41397-019-0141-x.
39. Li L, Zhu M, Li DY, et al. Dose tailoring of tacrolimus based on a non-linear pharmacokinetic model in children with refractory nephrotic syndrome. Int Immunopharmacol. 2021;98: 107827. https://doi.org/10.1016/j.intimp.2021.107827.
40. Gijsen VMGJ, van Schaik RHN, Soldin OP, et al. P450 oxidoreductase*28 ( POR*28) and tacrolimus disposition in pediatric kidney transplant recipients—a pilot study. Ther Drug Monit. 2014;36(2):152–8. https://doi.org/10.1097/FTD.0b013e3182a3f282.
41. Knops N, Herman J, van Dyck M, et al. Tacrolimus dose requirements in paediatric renal allograft recipients are characterized by a biphasic course determined by age and bone maturation. Br J Clin Pharmacol. 2017;83(4):863–74. https://doi.org/10.1111/bcp.13174.
42. Yang TH, Chen YK, Xue F, et al. Influence of CYP3A5 genotypes on tacrolimus dose requirement: age and its pharmacological interaction with ABCB1 genetics in the Chinese paediatric liver transplantation [Article]. Int J Clin Pract. 2015;69(S183):53–62. https://doi.org/10.1111/ijcp.12667.
43. Birdwell KA, Decker B, Barbarino JM, et al. Clinical pharmacogenetics implementation consortium (CPIC) guidelines for CYP3A5 genotype and tacrolimus dosing. Clin Pharmacol Ther. 2015;98(1):19–24. https://doi.org/10.1002/cpt.113.
44. Lamba JK, Lin YS, Schuetz EG, et al. Genetic contribution to variable human CYP3A-mediated metabolism. Adv Drug Deliv Rev. 2002;54(10):1271–94. https://doi.org/10.1016/s0169-409x(02)00066-2.
45. Elens L, Bouamar R, Hesselink DA, et al. A new functional CYP3A4 intron 6 polymorphism significantly affects tacrolimus pharmacokinetics in kidney transplant recipients. clin Chem. 2011;57(11):1574–83. https://doi.org/10.1373/clinchem.2011.165613.
46. Min S, Papaz T, Lafreniere-Roula M, et al. A randomized clinical trial of age and genotype-guided tacrolimus dosing after pediatric solid organ transplantation. Pediatr Transplant. 2018. https://doi.org/10.1111/petr.13285.
47. Van Driest SL, Webber SA. Pharmacogenomics personalizing pediatric heart transplantation. Circulation. 2015;131(5):503–12. https://doi.org/10.1161/Circulationaha.114.001382.
48. Krall P, Yañez D, Rojo A, et al. CYP3A5 and UGT1A9 polymorphisms influence immunosuppressive therapy in pediatric kidney transplant recipients. Front Pharmacol. 2021;12: 653525. https://doi.org/10.3389/fphar.2021.653525.
49. Zhao W, Fakhoury M, Deschênes G, et al. Population pharmacokinetics and pharmacogenetics of mycophenolic acid following administration of mycophenolate mofetil in de novo pediatric renal-transplant patients [Article]. J Clin Pharmacol. 2010;50(11):1280–91. https://doi.org/10.1177/0091270009357429.
50. Fukuda T, Goebel J, Cox S, et al. UGT1A9, UGT2B7, and MRP2 genotypes can predict mycophenolic acid pharmacokinetic variability in pediatric kidney transplant recipients. Ther Drug Monit. 2012;34(6):671–9. https://doi.org/10.1097/FTD.0b013e3182708f84.
51. Ohmann EL, Burckart GJ, Brooks MM, et al. Genetic polymorphisms influence mycophenolate mofetil-related adverse events in pediatric heart transplant patients. J Heart Lung Transplant. 2010;29(5):509–16. https://doi.org/10.1016/j.healun.2009.11.602.
52. Burckart GJ, Figg WD 2nd, Brooks MM, et al. Multi-institutional study of outcomes after pediatric heart transplantation: candidate gene polymorphism analysis of ABCC2. J Pediatr Pharmacol Therap. 2014;19(1):16–24. https://doi.org/10.5863/1551-6776-19.1.16.
53. Prausa SE, Fukuda T, Maseck D, et al. UGT genotype may contribute to adverse events following medication with mycophenolate mofetil in pediatric kidney transplant recipients. Clin Pharmacol Ther. 2009;85(5):495–500. https://doi.org/10.1038/clpt.2009.3.
54. Varnell CD, Fukuda T, Kirby CL, et al. Mycophenolate mofetil-related leukopenia in children and young adults following kidney transplantation: Influence of genes and drugs. Pediatr Transplant. 2017. https://doi.org/10.1111/petr.13033.
55. Li TF, Hu L, Ma XL, et al. Population pharmacokinetics of cyclosporine in Chinese children receiving hematopoietic stem cell transplantation. Acta Pharmacol Sin. 2019;40(12):1603–10. https://doi.org/10.1038/s41401-019-0277-x.
56. Cvetković M, Zivković M, Bundalo M, et al. Effect of age and allele variants of CYP3A5, CYP3A4, and POR genes on the pharmacokinetics of cyclosporin A in pediatric renal transplant recipients from Serbia. Ther Drug Monit. 2017;39(6):589–95. https://doi.org/10.1097/ftd.0000000000000442.
57. Turolo S, Tirelli AS, Ferraresso M, et al. Frequencies and roles of CYP3A5, CYP3A4 and ABCB1 single nucleotide polymorphisms in Italian teenagers after kidney transplantation. Pharmacol Rep. 2010;62(6):1159–69. https://doi.org/10.1016/s1734-1140(10)70378-9.
58. Fanta S, Niemi M, Jönsson S, et al. Pharmacogenetics of cyclosporine in children suggests an age-dependent influence of ABCB1 polymorphisms. Pharmacogenet Genom. 2008;18(2):77–90. https://doi.org/10.1097/FPC.0b013e3282f3ef72.
59. Yanagimachi M, Naruto T, Tanoshima R, et al. Influence of CYP3A5 and ABCB1 gene polymorphisms on calcineurin inhibitor-related neurotoxicity after hematopoietic stem cell transplantation. Clin Transplant. 2010;24(6):855–61. https://doi.org/10.1111/j.1399-0012.2009.01181.x.
60. Ramsey LB, Brown JT, Vear SI, et al. Gene-based dose optimization in children. Annu Rev Pharmacol. 2020;60:311–31. https://doi.org/10.1146/annurev-pharmtox-010919-023459.
61. Man M, Farmen M, Dumaual C, et al. Genetic variation in metabolizing enzyme and transporter genes: comprehensive assessment in 3 major East Asian subpopulations with comparison to Caucasians and Africans. J Clin Pharmacol. 2010;50(8):929–40. https://doi.org/10.1177/0091270009355161.
62. Relling MV, Hancock ML, Rivera GK, et al. Mercaptopurine therapy intolerance and heterozygosity at the thiopurine S-methyltransferase gene locus. J Natl Cancer Inst. 1999;91(23):2001–8. https://doi.org/10.1093/jnci/91.23.2001.
63. Relling MV, Schwab M, Whirl-Carrillo M, et al. Clinical pharmacogenetics implementation consortium guideline for thiopurine dosing based on TPMT and NUDT15 genotypes: 2018 update. Clin Pharmacol Ther. 2019;105(5):1095–105. https://doi.org/10.1002/cpt.1304.

64. Moriyama T, Yang YL, Nishii R, et al. Novel variants in NUDT15 and thiopurine intolerance in children with acute lymphoblastic leukemia from diverse ancestry. Blood. 2017;130(10):1209–12. https://doi.org/10.1182/blood-2017-05-782383.
65. Tanaka Y, Kato M, Hasegawa D, et al. Susceptibility to 6-MP toxicity conferred by a NUDT15 variant in Japanese children with acute lymphoblastic leukaemia. Br J Haematol. 2015;171(1):109–15. https://doi.org/10.1111/bjh.13518.
66. Yang JJ, Landier W, Yang W, et al. Inherited NUDT15 variant is a genetic determinant of mercaptopurine intolerance in children with acute lymphoblastic leukemia. J Clin Oncol. 2015;33(11):1235–42. https://doi.org/10.1200/jco.2014.59.4671.
67. Pavlovic S, Kotur N, Stankovic B, et al. Clinical application of thiopurine pharmacogenomics in pediatrics. Curr Drug Metab. 2020;21(1):53–62. https://doi.org/10.2174/1389200221666200303113456.
68. Relling MV, Gardner EE, Sandborn WJ, et al. Clinical pharmacogenetics implementation consortium guidelines for thiopurine methyltransferase genotype and thiopurine dosing. Clin Pharmacol Ther. 2011;89(3):387–91. https://doi.org/10.1038/clpt.2010.320.
69. Lennard L, Cartwright CS, Wade R, et al. Thiopurine dose intensity and treatment outcome in childhood lymphoblastic leukaemia: the influence of thiopurine methyltransferase pharmacogenetics. Br J Haematol. 2015;169(2):228–40. https://doi.org/10.1111/bjh.13240.
70. Petrykey K, Andelfinger GU, Laverdière C, et al. Genetic factors in anthracycline-induced cardiotoxicity in patients treated for pediatric cancer. Expert Opin Drug Metab Toxicol. 2020;16(10):865–83. https://doi.org/10.1080/17425255.2020.1807937.
71. Visscher H, Ross CJ, Rassekh SR, et al. Validation of variants in SLC28A3 and UGT1A6 as genetic markers predictive of anthracycline-induced cardiotoxicity in children. Pediatr Blood Cancer. 2013;60(8):1375–81. https://doi.org/10.1002/pbc.24505.
72. Visscher H, Ross CJD, Rassekh SR, et al. Pharmacogenomic prediction of anthracycline-induced cardiotoxicity in children [Article]. J Clin Oncol. 2012;30(13):1422–8. https://doi.org/10.1200/JCO.2010.34.3467.
73. Krajinovic M, Elbared J, Drouin S, et al. Polymorphisms of ABCC5 and NOS3 genes influence doxorubicin cardiotoxicity in survivors of childhood acute lymphoblastic leukemia. Pharmacogenom J. 2016;16(6):530–5. https://doi.org/10.1038/tpj.2015.63.
74. Semsei AF, Erdelyi DJ, Ungvari I, et al. ABCC1 polymorphisms in anthracycline-induced cardiotoxicity in childhood acute lymphoblastic leukaemia. Cell Biol Int. 2012;36(1):79–86. https://doi.org/10.1042/cbi20110264.
75. Visscher H, Rassekh SR, Sandor GS, et al. Genetic variants in SLC22A17 and SLC22A7 are associated with anthracycline-induced cardiotoxicity in children [Article]. Pharmacogenomics. 2015;16(10):1065–76. https://doi.org/10.2217/pgs.15.61.
76. Aminkeng F, Ross CJ, Rassekh SR, et al. Recommendations for genetic testing to reduce the incidence of anthracycline-induced cardiotoxicity. Br J Clin Pharmacol. 2016;82(3):683–95. https://doi.org/10.1111/bcp.13008.
77. Nava T, Kassir N, Rezgui MA, et al. Incorporation of GSTA1 genetic variations into a population pharmacokinetic model for IV busulfan in paediatric hematopoietic stem cell transplantation. Br J Clin Pharmacol. 2018;84(7):1494–504. https://doi.org/10.1111/bcp.13566.
78. Ansari M, Huezo-Diaz P, Rezgui MA, et al. Influence of glutathione S-transferase gene polymorphisms on busulfan pharmacokinetics and outcome of hematopoietic stem-cell transplantation in thalassemia pediatric patients. Bone Marrow Transplant. 2016;51(3):377–83. https://doi.org/10.1038/bmt.2015.321.
79. Ten Brink MH, Van Bavel T, Swen JJ, et al. Effect of genetic variants GSTA1 and CYP39A1 and age on busulfan clearance in pediatric patients undergoing hematopoietic stem cell transplantation [Article]. Pharmacogenomics. 2013;14(14):1683–90. https://doi.org/10.2217/pgs.13.159.
80. Yuan J, Sun N, Feng X, et al. Optimization of busulfan dosing regimen in pediatric patients using a population pharmacokinetic model incorporating gst mutations [Article]. Pharmacogenom Personal Med. 2021;14:253–68. https://doi.org/10.2147/PGPM.S289834.
81. Elhasid R, Krivoy N, Rowe JM, et al. Influence of glutathione S-transferase A1, P1, M1, T1 polymorphisms on oral busulfan pharmacokinetics in children with congenital hemoglobinopathies undergoing hematopoietic stem cell transplantation. Pediatr Blood Cancer. 2010;55(6):1172–9. https://doi.org/10.1002/pbc.22739.
82. Nava T, Rezgui MA, Uppugunduri CRS, et al. GSTA1 genetic variants and conditioning regimen: missing key factors in dosing guidelines of busulfan in pediatric hematopoietic stem cell transplantation. Biol Blood Marrow Transpl. 2017;23(11):1918–24. https://doi.org/10.1016/j.bbmt.2017.07.022.
83. Ceppi F, Langlois-Pelletier C, Gagné V, et al. Polymorphisms of the vincristine pathway and response to treatment in children with childhood acute lymphoblastic leukemia [Article]. Pharmacogenomics. 2014;15(8):1105–16. https://doi.org/10.2217/pgs.14.68.
84. Guilhaumou R, Simon N, Quaranta S, et al. Population pharmacokinetics and pharmacogenetics of vincristine in paediatric patients treated for solid tumour diseases. Cancer Chemother Pharmacol. 2011;68(5):1191–8. https://doi.org/10.1007/s00280-010-1541-4.
85. Moore AS, Norris R, Price G, et al. Vincristine pharmacodynamics and pharmacogenetics in children with cancer: a limited-sampling, population modelling approach. J Paediatr Child Health. 2011;47(12):875–82. https://doi.org/10.1111/j.1440-1754.2011.02103.x.
86. Egbelakin A, Ferguson MJ, MacGill EA, et al. Increased risk of vincristine neurotoxicity associated with low CYP3A5 expression genotype in children with acute lymphoblastic leukemia. Pediatr Blood Cancer. 2011;56(3):361–7. https://doi.org/10.1002/pbc.22845.
87. Renbarger JL, McCammack KC, Rouse CE, et al. Effect of race on vincristine-associated neurotoxicity in pediatric acute lymphoblastic leukemia patients [Article]. Pediatr Blood Cancer. 2008;50(4):769–71. https://doi.org/10.1002/pbc.21435.
88. Aplenc R, Glatfelter W, Han P, et al. CYP3A genotypes and treatment response in paediatric acute lymphoblastic leukaemia. Br J Haematol. 2003;122(2):240–4. https://doi.org/10.1046/j.1365-2141.2003.04430.x.
89. Wright GEB, Amstutz U, Drögemöller BI, et al. Pharmacogenomics of vincristine-induced peripheral neuropathy implicates pharmacokinetic and inherited neuropathy genes [article]. Clin Pharmacol Ther. 2019;105(2):402–10. https://doi.org/10.1002/cpt.1179.
90. Budi T, Toth K, Nagy A, et al. Clinical significance of CYP2C9-status guided valproic acid therapy in children. Epilepsia. 2015;56(6):849–55. https://doi.org/10.1111/epi.13011.
91. Guo YJ, Hu C, He XJ, et al. Effects of UGT1A6, UGT2B7, and CYP2C9 genotypes on plasma concentrations of valproic acid in chinese children with epilepsy. Drug Metab Pharmacokinet. 2012;27(5):536–42. https://doi.org/10.2133/dmpk.DMPK-11-NT-144.
92. Algharably EA, El Hamamsy M, Hassanein SM, et al. The effect of UGT1A6 polymorphism at two loci on the clinical response to valproic acid in epileptic children [Article]. Int J Pharmac Sci

Res. 2016;7(10):3986–94. https://doi.org/10.13040/IJPSR.0975-8232.7(10).3986-94.
93. Xu S, Chen Y, Zhao M, et al. Population pharmacokinetics of valproic acid in epileptic children: effects of clinical and genetic factors. Eur J Pharmac Sci. 2018;15(122):170–8. https://doi.org/10.1016/j.ejps.2018.06.033.
94. Nandith PB, Adiga U, Shenoy V, et al. UGT1A6 and UGT2B7 gene polymorphism and its effect in pediatric epileptic patients on sodium valproate monotherapy. Indian J Pediatr. 2021;88(8):764–70. https://doi.org/10.1007/s12098-020-03565-9.
95. Banawalikar N, Adiga S, Adiga U, et al. Association of UGT1A6 gene polymorphism with clinical outcome in pediatric epileptic patients on sodium valproate monotherapy. Braz J Med Biol Res. 2021;54(9): e11097. https://doi.org/10.1590/1414-431X2021e11097.
96. Feng W, Mei S, Zhu L, et al. Effects of UGT2B7, SCN1A and CYP3A4 on the therapeutic response of sodium valproate treatment in children with generalized seizures [Article]. Seizure. 2018;58:96–100. https://doi.org/10.1016/j.seizure.2018.04.006.
97. Monostory K, Nagy A, Tóth K, et al. Relevance of CYP2C9 function in valproate therapy. Curr Neuropharmacol. 2019;17(1):99–106. https://doi.org/10.2174/1570159x15666171109143654.
98. Tóth K, Bűdi T, Kiss Á, et al. Phenoconversion of CYP2C9 in epilepsy limits the predictive value of CYP2C9 genotype in optimizing valproate therapy. Per Med. 2015;12(3):199–207. https://doi.org/10.2217/pme.14.82.
99. Noai M, Soraoka H, Kajiwara A, et al. Cytochrome P450 2C19 polymorphisms and valproic acid-induced weight gain. Acta Neurol Scand. 2016;133(3):216–23. https://doi.org/10.1111/ane.12473.
100. Mei S, Feng W, Zhu L, et al. Effect of CYP2C19, UGT1A8, and UGT2B7 on valproic acid clearance in children with epilepsy: a population pharmacokinetic model. Eur J Clin Pharmacol. 2018;74(8):1029–36. https://doi.org/10.1007/s00228-018-2440-6.
101. Inoue K, Suzuki E, Yazawa R, et al. Influence of uridine diphosphate glucuronosyltransferase 2B7 -161C>T polymorphism on the concentration of valproic acid in pediatric epilepsy patients. Ther Drug Monit. 2014;36(3):406–9. https://doi.org/10.1097/ftd.0000000000000012.
102. Monostory K, Budi T, Toth K, et al. In response: commentary on clinical significance of CYP2C9-status-guided valproic acid therapy in children. Epilepsia. 2016;57(8):1339–40. https://doi.org/10.1111/epi.13451.
103. Kerr BM, Thummel KE, Wurden CJ, et al. Human liver carbamazepine metabolism: role of CYP3A4 and CYP2C8 in 10,11-epoxide formation. Biochem Pharmacol. 1994;47(11):1969–79. https://doi.org/10.1016/0006-2952(94)90071-X.
104. Zanger UM, Schwab M. Cytochrome P450 enzymes in drug metabolism: regulation of gene expression, enzyme activities, and impact of genetic variation. Pharmacol Ther. 2013;138(1):103–41. https://doi.org/10.1016/j.pharmthera.2012.12.007.
105. Djordjevic N, Jankovic SM, Milovanovic JR. Pharmacokinetics and pharmacogenetics of carbamazepine in children [review]. Eur J Drug Metab Pharmacokinet. 2017;42(5):729–44. https://doi.org/10.1007/s13318-016-0397-3.
106. Kim WJ, Lee JH, Yi J, et al. A nonsynonymous variation in MRP2/ABCC2 is associated with neurological adverse drug reactions of carbamazepine in patients with epilepsy. Pharmacogenet Genom. 2010;20(4):249–56. https://doi.org/10.1097/FPC.0b013e328338073a.
107. Hu T, Zeng X, Tian T, et al. Association of EPHX1 polymorphisms with plasma concentration of carbamazepine in epileptic patients: Systematic review and meta-analysis. J Clin Neurosci. 2021;91:159–71. https://doi.org/10.1016/j.jocn.2021.07.009.
108. Zhang H, Zhang W, Li Y, et al. Correlations between UGT2B7∗2 gene polymorphisms and plasma concentrations of carbamazepine and valproic acid in epilepsy patients. Brain Develop. 2018;40(2):100–6. https://doi.org/10.1016/j.braindev.2017.09.004.
109. Hung SI, Chung WH, Jee SH, et al. Genetic susceptibility to carbamazepine-induced cutaneous adverse drug reactions. Pharmacogenet Genom. 2006;16(4):297–306. https://doi.org/10.1097/01.fpc.0000199500.46842.4a.
110. Ueda K, Ishitsu T, Seo T, et al. Glutathione S-transferase M1 null genotype as a risk factor for carbamazepine-induced mild hepatotoxicity. Pharmacogenomics. 2007;8(5):435–42. https://doi.org/10.2217/14622416.8.5.435.
111. Lima JJ, Thomas CD, Barbarino J, et al. Clinical pharmacogenetics implementation consortium (CPIC) guideline for CYP2C19 and proton pump inhibitor dosing. Clin Pharmacol Ther. 2021;109(6):1417–23. https://doi.org/10.1002/cpt.2015.
112. Kearns GL, Blumer J, Schexnayder S, et al. Single-dose pharmacokinetics of oral and intravenous pantoprazole in children and adolescents. J Clin Pharmacol. 2008;48(11):1356–65. https://doi.org/10.1177/0091270008321811.
113. Shakhnovich V, Brian Smith P, Guptill JT, et al. A population-based pharmacokinetic model approach to pantoprazole dosing for obese children and adolescents. Paediatr Drugs. 2018;20(5):483–95. https://doi.org/10.1007/s40272-018-0305-1.
114. Gumus E, Karaca O, Babaoglu MO, et al. Evaluation of lansoprazole as a probe for assessing cytochrome P450 2C19 activity and genotype-phenotype correlation in childhood. Eur J Clin Pharmacol. 2012;68(5):629–36. https://doi.org/10.1007/s00228-011-1151-z.
115. Franciosi JP, Mougey EB, Williams A, et al. Association between CYP2C19*17 alleles and ph probe testing outcomes in children with symptomatic gastroesophageal reflux. J Clin Pharmacol. 2018;58(1):89–96. https://doi.org/10.1002/jcph.977.
116. Bernal CJ, Aka I, Carroll RJ, et al. CYP2C19 phenotype and risk of proton pump inhibitor-associated infections. Pediatrics. 2019;144:6. https://doi.org/10.1542/peds.2019-0857.
117. Lima JJ, Lang JE, Mougey EB, et al. Association of CYP2C19 polymorphisms and lansoprazole-associated respiratory adverse effects in children. J Pediatr. 2013;163(3):686–91. https://doi.org/10.1016/j.jpeds.2013.03.017.
118. Kearns GL, Leeder JS, Gaedigk A. Impact of the CYP2C19*17 allele on the pharmacokinetics of omeprazole and pantoprazole in children: evidence for a differential effect. Drug Metab Dispos. 2010;38(6):894–7. https://doi.org/10.1124/dmd.109.030601.
119. Cicali EJ, Blake K, Gong Y, et al. Novel implementation of genotype-guided proton pump inhibitor medication therapy in children: a pilot, randomized, multisite pragmatic trial. Cts-Clin Transl Sci. 2019;12(2):172–9. https://doi.org/10.1111/cts.12589.
120. Tang M, Blake KV, Lima JJ, et al. Genotype tailored treatment of mild symptomatic acid reflux in children with uncontrolled asthma (GenARA): rationale and methods. Contemp Clin Trials. 2019;78:27–33. https://doi.org/10.1016/j.cct.2019.01.009.
121. Hines RN. Developmental expression of drug metabolizing enzymes: impact on disposition in neonates and young children [short survey]. Int J Pharm. 2013;452(1–2):3–7. https://doi.org/10.1016/j.ijpharm.2012.05.079.
122. De Wildt SN, Tibboel D, Leeder JS. Drug metabolism for the paediatrician [review]. Arch Dis Child Educ Pract Ed. 2014;99(12):1137–42. https://doi.org/10.1136/archdischild-2013-305212.
123. Relling MV, Altman RB, Goetz MP, et al. Clinical implementation of pharmacogenomics: overcoming genetic exceptionalism.

Lancet Oncol. 2010;11(6):507–9. https://doi.org/10.1016/s1470-2045(10)70097-8.
124. Manolio TA. Bringing genome-wide association findings into clinical use. Nat Rev Genet. 2013;14(8):549–58. https://doi.org/10.1038/nrg3523.



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