History of Pediatric Pharmacogenetics: Adults are Just Big Children

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Progress in Pediatric Pharmacogenetics: Our Heritage and Vision for the Future

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Kansas City, MO
This is my wife, Donna’s first ASCPT meeting; it is likely to be her last because I have embarrassed her, but I am nonetheless deeply appreciative of her support over the years.

The content of this presentation reflects my personal perspective, and, as a consequence, is likely to be selective and biased.

In the past 12 months, I have no financial relationships with the manufacturer(s) of any commercial product(s) and/or providers of commercial services discussed in this presentation.
<table>
<thead>
<tr>
<th>Grant Number</th>
<th>Project Title</th>
</tr>
</thead>
<tbody>
<tr>
<td>R03 HD036783</td>
<td>Ontogeny in the first year of life</td>
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<tr>
<td>R01 ES010855</td>
<td>Pharmacogenetics of drug-induced birth defects</td>
</tr>
<tr>
<td>R43 CA110874</td>
<td>Dextromethorphan breath test</td>
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<tr>
<td>U01 HD044239</td>
<td>Ontogeny of drug bioactivation</td>
</tr>
<tr>
<td>R13 HD065386</td>
<td>Pediatric pharmacogenomics meeting</td>
</tr>
<tr>
<td>R01 HD058556</td>
<td>Longitudinal phenotyping study during puberty</td>
</tr>
<tr>
<td>R01 HD081299</td>
<td>Proteomics in pediatric liver (Prasad, PI)</td>
</tr>
<tr>
<td>T32 HD069038</td>
<td>Pediatric Clinical Pharmacology training program</td>
</tr>
<tr>
<td>U54 HD090258</td>
<td>GOLDILOKs (+ Philanthropic Support)</td>
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</tbody>
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Mentors and Colleagues

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Complex Problems, Multidisciplinary Teams

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Presentation Goals

- Explore the close ties between pediatric pharmacogenetics and pediatric clinical pharmacology as it matured as a discipline
- Review the role of genetic variation as a factor contributing to variability in drug disposition and response in pediatric patients in the context of progress made over the past 40 years
- Present some approaches to be considered to translate pediatric pharmacogenetics into precision therapeutics for children
In the Beginning (1962) ...
Pediatric Pharmacogenetics: The Early Days

- Who is this, and what is his relationship to pediatric pharmacogenetics?
Harry C. Shirkey, MD (1966)
Pharmacogenetics is a newly recognized discipline which is concerned with alterations in response to drugs that result from genetic differences among individuals. The genetic analysis of human variation has been refined and substantially accelerated by the addition of biochemical genetics to consideration of form and appearance. It has more recently been appreciated that some populations can be distinguished only by their response to drugs. Individuals with the trait in question may be perfectly normal in all other respects. It is only following the use of an exogenous chemical agent that it becomes apparent that they are different from others. Drug responses then provide an important additional tool for the geneticist. This is the particular significance of revealed by the effects of drugs. Some of the more common examples will be considered as models for many as yet unrecognized.

**THE DRUG-SENSITIVE ERYTHROCYTE (GLUCOSE-6-PHOSPHATE DEHYDROGENASE DEFICIENCY)**

(see also p. 703)

Drug-induced hemolytic anemia represents a heritable metabolic defect, in which instance the primary expression of the abnormal gene is in the abnormal function of an enzyme. However, the involved erythrocytes appear quite normal except when challenged from outside, in this case by the administration of certain drugs (Table 37, p. 704). The recognition of this abnormality is, therefore, of
Pediatric Pharmacogenetics: The Early Days

- Who is this, and what is his relationship to pediatric pharmacogenetics?
PART III. GENETIC VARIATIONS THAT MODIFY DRUG RESPONSE

VARIATIONS IN DETOXICATION ENZYMES DURING MAMMALIAN DEVELOPMENT*

Sumner J. Yaffo, Joseph Krasner and Charlotte S. Catz

Department of Pediatrics, School of Medicine
State University of N. Y. at Buffalo

It has been repeatedly demonstrated that the developing fetus and newborn infant are more sensitive than the adult to the effects of many pharmacologic agents. Differences in absorption, distribution, and excretion may be present in the newborn organism, but the principal factor in this response differential appears to be variation in the rates of detoxication. Drug metabolic processes have been studied in vitro in several animal species and, in most instances, activities are lower in the newborn organism compared with the adult. Variations in drug metabolism appear to be genetically determined and the enzymic activity observed at any given age is dependent upon the species and strain of organism as well as the substrate employed in the reaction. Our primary objective in our initial studies was to determine how soon after birth one could detect and differentiate the contribution to drug metabolism made by genetic endowment.
Sumner Yaffe: Early Recognition of Importance of Genetic Variation and Ontogeny (mostly ontogeny)

**Figure 1.** Strain variation in sleeping time. The height of each bar represents the mean value for sleeping time in minutes following i.p. injection of 100 mg/kg of hexobarbital. Twelve adult animals of each sex and strain were used in the assay procedure.

**Figure 2.** Strain and age variation in sleeping time. The height of each bar represents the mean value for sleeping time (in minutes) following i.p. administration of 50 mg/kg of hexobarbital. Twelve (or more) male mice of each age and strain were used for the determination.
Sumner J. Yaffe, MD

THE PEDIATRIC CLINICS OF NORTH AMERICA

Volume 19 / NUMBER 1
FEBRUARY 1972

SYMPOSIUM ON
PEDIATRIC PHARMACOLOGY

Sumner J. Yaffe, M.D., Editor
Pharmacogenetics may also affect children …

It is well known that among adult patients receiving isoniazid, the slow acetylators are at risk to develop a peripheral neuropathy. Such a reaction is so common in the general population that many physicians prescribe pyridoxine routinely for all patients taking isoniazid. It is also well known that the neurologic side-effects of this drug among childhood patients are so rare that they can be ignored as a hazard to a child who is taking isoniazid. The rarity of this troublesome pharmacogenetically determined side-effect among children has not been explained.
Malignant hyperthermia has not been observed in infants and children below the age of 3 years, but almost 25 per cent of all cases occurred prior to the fifth year of life and more than 30 per cent occurred during the first 10 years. Kalow and his associates have concluded that malignant hyperthermia is hereditary in a substantial number of cases but they cannot explain this apparent predisposition of young children (3 to 10 years of age) for the condition.
Increasing awareness of role for genetic factors contributing to variability in drug response, largely adverse drug reactions
- Sulfonamides and G6PD deficiency
- Speculation regarding risk of birth defects (Dan Nebert, 1981)

Children may also be affected - same genes, same drugs, but …

Observation that phenotypic traits/ADR risks may differ in children
- Less risk of isoniazid peripheral neuropathy
- Children at increased risk of drug-induced malignant hyperthermia

No investigations of the genetic basis of a drug-related phenotype in children
Investigating Pharmacogenetics in Children: 1980s and Beyond

Meyer, Nat Rev Genet 2004; 5:669-676
Weinshilboum and Sladek, 1980: first “phenotyping” study conducted in children -- thiopurine S-methyl transferase (TPMT) polymorphism

Study population included 115 children aged 13.0 ± 0.4 years

Wide variation in RBC TPMT activity segregated as a monogenic trait consistent with autosomal codominant inheritance

Lennard et al, 1983: Relationship between intracellular 6MP concentrations and neutropenia

Lennard, Weinshilboum collaboration, 1987: Correlation of RBC TPMT activity and 6-TGN concentrations, and other observations …
Thiopurine pharmacogenetics in leukemia: Correlation of erythrocyte thiopurine methyltransferase activity and 6-thioguanine nucleotide concentrations

Thiopurine methyltransferase (TPMT) catalyzes the S-methylation of thiopurine drugs such as 6-thioguanine (6-MP) and azathioprine. Human erythrocyte (RBC) TPMT activity is controlled by a common genetic polymorphism. On a genetic basis approximately one in every 300 subjects lacks TPMT activity, and 11% of subjects have intermediate activities. 6-Thioguanine nucleotides (6-TGN) are major metabolites of 6-MP and azathioprine in humans. RBC 6-TGN concentrations are correlated directly with risk for the development of leukopenia in patients treated with thiopurine drugs. Our studies were performed to determine whether there was a relationship between genetically controlled levels of RBC TPMT activity and RBC concentrations of 6-TGN. We found a significant negative correlation between RBC TPMT activity and 6-TGN concentrations in blood samples from 40 children with acute lymphoblastic leukemia receiving long-term therapy with 6-MP (r = −0.474; P < 0.005). In addition, RBC TPMT activities were significantly higher in blood samples from these patients than in blood samples from adult control subjects (P < 0.0001) or children with acute lymphoblastic leukemia who were in remission but were not receiving drug therapy (P < 0.0001). Finally, three adult patients were studied who developed very high RBC 6-TGN concentrations and thiopurine-induced leukopenia. Two of the three patients had no detectable RBC TPMT activity—presumably on a genetic basis. These results indicate that low TPMT activity may be a risk factor for the occurrence of elevated 6-TGN concentrations and for the development of severe leukopenia in patients treated with thiopurine drugs. Measurement of RBC TPMT activity might make it possible to predict this risk factor for the development of thiopurine drug toxicity. (CLIN PHARMACOL THER 1987;41:18-25.)

- Negative correlation between RBC TPMT activity and 6-TGN concentrations
- TPMT activity higher in children with ALL treated with 6MP than adult controls (ontogeny?)
- TPMT activity higher in children with ALL treated with 6MP than children with ALL in remission and not receiving drug (induction?)
- TPMT activity potential risk factor for development of severe leukopenia
Progress in Pediatric Pharmacogenetics: TPMT

- McLeod et al, 1995: TPMT activity >50% higher in full term newborns relative to race-matched adults; distribution of activity consistent with demonstrated genetic polymorphism
- Krynetski et al, 1995: Identification of TPMT*2 allele from cDNA
- Szumlanski et al, 1996: TPMT gene cloned and TPMT*3A and *3B alleles described
- Extensive literature in ALL and pediatric IBD in ‘80s, ‘90s and ‘00s
- CPIC guideline for TPMT and thiopurine dosing first published 2011
- NUDT15: 2014 (Crohn’s Disease; Korea), 2015 (ALL, St. Jude)
Clinical Pharmacogenetics Implementation Consortium Guideline for Thiopurine Dosing Based on TPMT and NUDT15 Genotypes: 2018 Update

Mary V. Relling¹, Matthias Schwab²,³,⁴, Michelle Whirl-Carrillo⁵, Guilherme Suarez-Kurtz⁶, Ching-Hon Pui⁷, Charles M. Stein⁸, Ann M. Moyer⁹, William E. Evans¹, Teri E. Klein⁴, Federico Guillermo Antillon-Klussmann¹⁰,¹¹, Kelly E. Caudle¹, Motohiro Kato¹², Allen E.J. Yeoh¹³,¹⁴, Kjeld Schmiegelow¹⁵,¹⁶ and Jun J. Yang¹
Progress in Pediatric Pharmacogenetics: TPMT

- **TPMT normal metabolizer (NM)**
  - NUDT15 NM
    - Use standard dose
  - NUDT15 IM
    - Dose reduction recommended\(^a\). See NUDT15 IM recommendation\(^b\).
  - NUDT15 PM
    - Consider dose reduction\(^a\). See NUDT15 PM recommendation\(^b\).

- **TPMT intermediate metabolizer (IM)**
  - NUDT15 NM
  - NUDT15 IM
    - Consider dose reduction\(^a\). See TPMT IM recommendation\(^b\).
  - NUDT15 PM
    - Consider dose reduction\(^a\). See TPMT IM and NUDT15 IM recommendation\(^b,c\).

- **TPMT poor metabolizer (PM)**
  - NUDT15 NM
  - NUDT15 IM
  - NUDT15 PM
    - Dose reduction recommended\(^a\). See TPMT PM recommendation\(^b\).
PREDISPOSITION TO PHENYTOIN HEPATOTOXICITY ASSESSED IN VITRO

Stephen P. Spielberg, M.D., Ph.D., Gary B. Gordon, M.D., Ph.D, David A. Blake, Ph.D., Daniel A. Goldstein, M.D., and H. Franklin Herlong, M.D.

Abstract  Arene oxide metabolites of phenytoin may be involved in the pathogenesis of drug-induced hepatotoxicity. We examined individual susceptibility to toxicity from such metabolites by exposing human lymphocytes to metabolites generated by a murine hepatic microsomal system. Cells from 17 controls showed no toxicity at concentrations of phenytoin from 31 to 125 μM. Cells from three patients with phenytoin hepatotoxicity manifested dose-dependent toxicity from the metabolites. Phenytoin alone was not toxic to cells. The patients' dose-response curves resembled the response of control cells in which epoxide hydrolase (a detoxification enzyme for arene oxides) was inhibited. Detoxification of non-arene oxide metabolites (e.g., of acetaminophen) was normal in patients' cells. Cells from parents of two patients had intermediate responses. Cells from a sibling of one patient showed no toxicity; a sibling of another patient had a response similar to that of the patient. A heritable defect in response to arene oxides thus may predispose some patients to phenytoin hepatotoxicity. (N Engl J Med. 1981; 305:722-7.)
Circuitous Route to Pediatric Pharmacogenetics

HSC
- Spielberg
  - Leeder

UofT
- Kalow
  - Grant

Basel
- Meyer
  - Grant
  - Gaedigk
  - NAT2
  - CYP2D6*5

Stuttgart
- Eichelbaum

HSC
- Leeder
- Grant
- Gaedigk
- CMH
Investigating Pharmacogenetics in Children: 1990s and Beyond

Meyer, Nat Rev Genet 2004; 5:669-676
Progress in Pediatric Pharmacogenetics: CYP2D6

- Evans et al, 1989: first CYP2D6 phenotyping study conducted in children (n=26, 3-21 y); dextromethorphan (DM) as the probe
- Evans and Relling, 1991: CYP2D6 genotype-phenotype concordance assessed in 116 subjects with median age of ~10 years
- Treluyer et al, 1991: Early investigation of CYP2D6 ontogeny in fetal and newborn liver
- Blake et al, 2007: Longitudinal phenotyping study in first year of life
Progress in Pediatric Pharmacogenetics: CYP2D6

- Longitudinal DM phenotyping study in healthy term infants
- "Well-baby" visits at 2 weeks, 1 mo, 2 mo, 4 mo, 6 mo, and 12 months of age
- 0.3 mg/kg; overnight urine collection
- Analysis for DM and metabolites (DX, 3MM and 3HM) by HPLC
- Genotype concordant with phenotype at 2 wk
Progress in Pediatric Pharmacogenetics: CYP2D6

- In the very young critically ill, ontogeny and genetic variation are important, among other factors.

- Genotyping may be of limited value in an acute neonatal setting due to the developmental changes in other factors, such as maturation of renal function.

- PK studies in extreme genotypes (0 vs >2 functional alleles) are required to determine the magnitude of effect on dose-exposure-response relationships.

Adapted from Allegaert et al., Pediatr Anesth 2011;21:266-73
FDA warning for nursing mothers issued Aug 17, 2007

FDA reviewing reports of SADRs in tonsil- and adenoinectomy issued Aug 15, 2012

Black box warning (contraindication) issued Feb 20, 2013

CPIC guideline updated (CPT 2014; 95:376-82)

Additional FDA updates for codeine and tramadol issued Apr 20, 2017 and Jan 11, 2018
Progress in Pediatric Pharmacogenetics: CYP2D6

- Studies of CYP2D6 pharmacogenetics in pediatric patients few in number and generally uninformative; most common phenotype parent/metabolite ratios or clinically obtained trough concentrations

- General conclusions:
  
  *its blood concentration after dose escalation. Although blood concentration is not related to adverse effects or clinical improvement, determination of plasma FLX concentrations could provide information about variability in clinical response.*

- Few PK studies assessing influence of CYP2D6 genotype on drug clearance in pediatric age groups; atomoxetine, tramadol and risperidone exceptions

Blázquez et al., J Clin Psychopharmacol 2014;34: 318-326
Progress in Pediatric Pharmacogenetics: CYP2D6

Clinical Pharmacogenetics Implementation Consortium (CPIC) Guideline for CYP2D6 and CYP2C19 Genotypes and Dosing of Selective Serotonin Reuptake Inhibitors

JK Hicks¹, JR Bishop², K Sangkuhl³, DJ Müller⁴, Y Ji⁵, SG Leckband⁶, JS Leeder⁷, RL Graham⁸, DL Chiulli⁹, A LLerena¹⁰, TC Skaar¹¹, SA Scott¹², JC Stingl¹³, TE Klein³, KE Caudle¹⁴ and A Gaedigk⁷
Few pediatric pharmacogenetics studies/data
Developmental changes in gene expression
Studies investigating the role of genetic variation as a factor contributing to variability in drug disposition and response have been reported in many areas:

<table>
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<tr>
<th>ADHD</th>
<th>Autism</th>
<th>Asthma</th>
<th>BMT</th>
<th>CHD</th>
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</thead>
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<td>Epilepsy</td>
<td>HIV</td>
<td>IBD, PPIs</td>
<td>JIA</td>
<td>Kawasaki</td>
<td>Oncology</td>
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<td>Neonatology</td>
<td>Pain</td>
<td>Transplant</td>
<td>Cisplatin</td>
<td>Morphine</td>
<td>Warfarin</td>
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Few studies have translated into clinically actionable, validated tests or models for routine application.
Pediatric Pharmacogenetics: Proposal for the Future

1. Genotype-stratified pharmacokinetic studies to establish population extremes and magnitude of pharmacogenetic effect on the dose→exposure relationship

2. Rich, intensive opportunistic sampling (aka “pragmatic pharmacology”)

3. Prospective validation and model refinement

4. Exposure-controlled/escalation studies to investigate the exposure→response relationship to establish therapeutically relevant exposure ranges, given knowledge of drug target expression and function

➢ Biomarkers of drug effect

- Previous participants in longitudinal DM study
- Primary diagnosis of ADHD
- Selected based on CYP2D6 genotype
  - 2 or more functional alleles: EM2
  - 1 functional allele: EM1
  - 0 functional alleles: PM
  - 0.5 functional alleles: IM
- Single oral dosage unit closest to 0.5 mg/kg
- Serial plasma sampling

Brown et al, CPT 2016 (PMID: 26660002)
Genotype-Stratified Pravastatin Pharmacokinetics

- AAP recommendations for universal lipid screening
- Variability in response to statins (LDL reduction) in pediatric clinical trials
- Dose-exposure relationship subject to genetic variation (SLCO1B1)
- Only pediatric PGx study to date reported higher pravastatin exposure in SLCO1B1 521TT

Wagner et al, CPT 2019 (PMID: 30549267)
Sources of Variability: Demographic Factors

- In the 521 TT group, PVA AUC associated with weight ($r^2=0.375, \ p_{13}=0.015$) and BMI ($r^2=0.390, \ p=0.013$)
- No association with any demographic variable in 521 TC
Sources of Variability: Conversion to 3α-PVA

- Also an OATP1B1 substrate based on genotype-phenotype association
- PVA AUC decreases with increase in 3α-PVA formation for 521TC group ($r^2=0.742$, $p<0.0001$), but not 521TT

Wagner et al, CPT 2019 (PMID: 30549267)

PDA Outcomes are Unpredictable

**Effective**
- Minimal Side Effects

**Effective**
- Acute Kidney Injury

**Not Effective**
- Minimal Side Effects

**Not Effective**
- Intestinal Perforation

No further treatment
Renal function recovers
Heart Surgery (25%)
Bowel surgery (and heart surgery later; 10%)

Courtesy of Tamorah Lewis, MD, PhD
Indomethacin in PDA: Sample Collection Strategy

Subject 15

Subject 14

Subject 13

Hours of Life

Urine Collection (to 229 hours)

Urine Collection (to 222 hours)

Urine Collection (to 260 hours)

△ = indomethacin dose
▽ = plasma sample
◇ = dried blood spot sample

Courtesy of Tamorah Lewis, MD, PhD
Distinct Patterns of Metabolite Formation and Excretion

Lewis et al, Pediatr Res (PMID: 29967531)
Proposal for the Future: 3. Prospective Model Validation

Development of dosing models incorporating PGx is encouraging; clinical application requires prospective validation as the authors of this study acknowledge.
Prospective Model Validation: Characterizing Individuals

Unpublished Data
Proposal for the Future: 4. Exposure-Controlled Dosing

Dose → Exposure → Response

Response → Exposure → Dose

What is the therapeutic goal of drug administration?

What exposure is required to achieve the desired response?

What dose must be administered to achieve that exposure?
Different drug exposures are required to achieve equivalent drug responses, depending on level of drug target expression (or function)

What should the target exposure be?

Is it the same across drug target expression/function levels?

Accessible surrogate markers of drug response?

McLaughlin et al, CTS 2019 (PMID: 30516322)
Summary and Conclusions

- Origins of pediatric pharmacogenetics lightly linked to the development of pediatric clinical pharmacology as a discipline
- Some cases of demonstrable impact (TPMT+NUDT15 in ALL; regulatory changes for codeine), but few examples of widespread integration into clinical practice
- Genotype-stratified PK studies have the potential to efficiently capture magnitude of PGx effect, and to identify additional sources of variability
- Future studies should utilize all available sources of new data, especially the value of opportunistic sampling for PK and PD studies, including biomarkers predictive of drug disposition and response (metabolomics)
- Generating more models is not sufficient; require prospective validation
Engaging Patients and Families

My genes say I am a: ____________

Courtesy of Susan Abdel-Rahman, PharmD and Jean Dinh, PharmD, PhD