Pharmacogenetics and Drug Safety

A. Pharmacogenetics vs Pharmacogenomics

1. *Pharmacogenetics* = inherited variations in drug effects; single gene interactions with drugs
   Includes:
   - Disposition
   - Safety
   - Tolerability
   - Efficacy

2. *Pharmacogenomics* = the effect of a drug on gene expression – OR – the use of genomic technologies to identify new drug targets. In the latter case, identifying a gene that is expressed very high in a disease tissue, yet very low expression is seen in the normal state, could be used to identify that gene as a drug target or a biomarker of the disease state. It is important to note that the terms “pharmacogenetics” and “pharmacogenomics” are sometimes used interchangeably, simply due to the fact that the term “genomics” is more contemporary “buzz word” than “genetics”.

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Rare vs. common variants

<table>
<thead>
<tr>
<th>Smaller effect; multiple variants</th>
<th>PHARMACOGENOMICS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Large single variant effect</td>
<td></td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Single gene</th>
<th>Small number of genes</th>
<th>Complex biologic pathway</th>
<th>Whole genome</th>
</tr>
</thead>
</table>

*Pharmacogenetics Research Network Investigators, Ann Int Med, 2006*
3. The promise of pharmacogenetics/genomics is PERSONALIZED MEDICINE

Drug therapy tailored to a patient’s unique genetic makeup, especially with regards to:

a. Choice of the drug
b. Choice of the dosing regimen

B. Human Genome Overview

1. Sequence completed in 2001
2. 3 billion bases of DNA
3. Divided into 23 chromosomes
4. In females, all diploid (two copies)
5. In males, X and Y are haploid (one copy each)
6. ~30,000 Genes

C. Chromosomes

1. Every human cell with the exception of gametes contains 23 chromosomes
2. Carry all the genetic coding for all the proteins in every cell
3. Consist of DNA tightly wound around special protein structures called histones

It is important to note that red blood cells (RBCs) and platelets do NOT have chromosomal DNA, since these “cells” are derived from progenitor cells in the bone marrow. Interestingly, it has been estimated that RBCs are made at a rate of 2 million per second in humans. Even though RBCs and platelets lack a nucleus and chromosomal DNA, a “DNA sample” can still be derived from a blood sample due to the presence of other nucleated white blood cells in the blood (i.e. neutrophils, eosinophils, lymphocytes, monocytes, etc.).
D. Structure of DNA
DNA is comprised of a string of 4 nucleotide bases (A, G, T and C) that are linked together in a double helix. Bases on opposite strands are always matched A-T and C-G.

E. Structure of Genes
A segment of DNA containing all of the information needed to encode for one protein is called a gene.
F. Transcription and Translation

G. Genomic variations

1. Single Nucleotide Polymorphisms (SNPs)
2. Variable Number Tandem Repeats (VNTR)
3. Insertions/Deletions

4. SNPS Upclose

   a. Polymorphisms:

      i. common variation in DNA, often defined as greater than 1% in general population
      ii. occur on average every 1331 bp, although frequency can be much greater in a given gene
      iii. estimated to be ~11 million polymorphisms in the human genome

   b. influence expression

      \[ 5'-\text{CATGTACCTGGGCCG-3'} \]
      \[ 3'-\text{GTACATGGACCCGGC-5'} \]

      \[ 5'-\text{CATGTACCTGGGCCG-3'} \]
      \[ 3'-\text{GTACATGGCCCGGC-5'} \]
c. Coding Polymorphisms are further classified as:

i. Non-synonymous (missense) – results in translation of a different amino acid

ii. Synonymous (sense) – results in the translation of the same amino acid

iii. Nonsense – results in the insertion of a stop codon

d. Mainly used to characterize genetic differences between individuals

H. SNP Application to Drug Therapy…. “Personalized Medicine”

Main goals of Personalized Medicine are to

Prevent adverse drug reactions (ADRs)
Improve drug efficacy

1. Adverse Drug Reactions (ADRs)

   i. accounts for nearly 7,000 deaths annually
   ii. 4th leading cause of death in the United states
   iii. Cost the U.S. health care system between $1.5 and $5.4 million per year
   iv. Agency for Health Care Quality (AHRQ) estimates that computerized systems could reduce medication up to 95%.

2. Drug Metabolism and Bioactivation
Table 1*: Representative Drug Metabolizing Enzymes SNPs

<table>
<thead>
<tr>
<th>CYP Family</th>
<th>Allele</th>
<th>Nucleotide Change</th>
<th>Enzyme Activity Change</th>
<th>Associated Drug Concentration Change</th>
</tr>
</thead>
<tbody>
<tr>
<td>1A2</td>
<td>CYP1A2*1C</td>
<td>-3860 G&gt;C</td>
<td>Decreases</td>
<td>Increases</td>
</tr>
<tr>
<td>2C9</td>
<td>CYP2C9*3A</td>
<td>1075 A&gt;C</td>
<td>Decreases</td>
<td>Increases</td>
</tr>
<tr>
<td>3A4</td>
<td>CYP3A4*18A</td>
<td>878 T&gt;C</td>
<td>Increases</td>
<td>Decreases</td>
</tr>
</tbody>
</table>

*Only three representative CYP families, a single SNP example. Complete list can be found at http://cypalleles.ki.se/

3. The CYP Families

Proportion of drugs metabolized by the major P450 enzymes

a. CYP 3A

- most abundantly expressed in human liver
- CYP 3A4 most abundant form
- CYP 3A7 most abundant in fetal liver & and thought to be involved in steroid metabolism

c. CYP 2D6 Polymorphisms

-Over 70 Single Nucleotide Polymorphisms (SNPs)
-Over 65 drugs metabolized by CYP2D6 inc. tricyclic antidepressants, neuroleptics, serotonin reuptake inhibitors, antiarrhythmics, b-adrenergic agonists, opiates
-4 Phenotypic Subpopulations
PM – poor metabolizers, adverse drug effects
IM – Intermediate metabolizers
EM – Extensive metabolizers
UM – Ultra-rapid metabolizers, usual drug doses ineffective; exaggerated response if metabolite is active (exaggerated response to codeine, formation of morphine increased)
- Frequency varies with racial background
- CYP2D6*4 is the most common variant allele in Caucasians with a population frequency of ~20%.

TCA Example

![Graph showing drug concentration over time for different CYP2D6 gene numbers.]

**d. CYP2C9**

- Encodes the p450 enzyme that metabolizes the anticoagulant warfarin

**i. Warfarin**

  a. antagonist at vitamin K epoxide reductase
     
     i. required to maintain levels of reduced vitamin K, which allows carboxylation of glutamate receptors on coagulation factors

  b. there is wide inter-individual variability in therapeutic efficacy
     
     i. 0.5 mg/day – 50 mg/day

**ii. Over 200 polymorphisms have been identified in CYP2C9**

  a. 30 coding region variants

     i. CYP2C9*2

     most common in Caucasian populations
ii. CYP2C9*3  
most common across all ethnicities  

b. individuals with either CYP2C9*2 or CYP2C9*3 exhibit a  
increasing sensitivity to warfarin  

i. greatest sensitivity if homozygous for CYP2C9*3  

iii. 23 haplotypes have been identified within CYP2C9, but only the *2 or  
*3 SNPs are associated with increased warfarin sensitivity  

e. CYP2C19  

i. Only two SNPs have been identified thus far  

CYP2C19*2 and CYP2C19*3  

ii. Each results in a non-functional protein product  

individuals homozygous for either CYP2C19*2 or  
CYP2C19*3 have no functional enzyme  

results in increased drug levels and improved therapeutic  
outcome
Table 2. Genetic Basis for Adverse Drug Responses in Drug Metabolism.

<table>
<thead>
<tr>
<th>Adverse Drug Response Type</th>
<th>Effect of SNP on Metabolic Enzyme</th>
<th>Effect on Peak Drug Plasma Concentration</th>
<th>Adverse Drug Response</th>
<th>Remediation of Adverse Drug Response Risk</th>
</tr>
</thead>
<tbody>
<tr>
<td>Decreased Clearance</td>
<td>(1) SNP in promoter region of gene causes decreased enzyme expression. (2) SNP in coding region of gene causes altered enzyme activity.</td>
<td>Upon normal dosing, peak plasma concentrations will exceed normal levels due to decreased metabolic capability of the patient.</td>
<td>Risk of drug-induced toxicity due to inadvertent overdosing of patient.</td>
<td>Decrease the drug dose or choose an alternate drug therapy.</td>
</tr>
<tr>
<td>Increased Clearance</td>
<td>SNP in promoter region of gene causes increased enzyme expression and/or inducibility.</td>
<td>Upon normal dosing, peak plasma concentrations will not reach efficacy levels normal due to increased metabolic capability of the patient.</td>
<td>Risk of undermedication due to increased drug metabolism.</td>
<td>Increase the drug dose or choose an alternate drug therapy.</td>
</tr>
<tr>
<td>Decreased Bioactivation</td>
<td>(1) SNP in promoter region of gene causes decreased enzyme expression. (2) SNP in coding region of gene causes altered enzyme activity.</td>
<td>The efficacy of the drug depends upon oxidative enzyme-mediated bioactivation to be effective.</td>
<td>Risk of undermedication due to the absence of bioactivation of the prodrug.</td>
<td>Choose an alternate drug therapy.</td>
</tr>
</tbody>
</table>

4. Improving drug efficacy

a. Drugs that require P450 bioactivation

<table>
<thead>
<tr>
<th>Drug</th>
<th>Active Metabolite</th>
<th>CYP450 Enzyme</th>
</tr>
</thead>
<tbody>
<tr>
<td>Codeine</td>
<td>Morphine</td>
<td>CYP2D6</td>
</tr>
<tr>
<td>Dihydrocodeine</td>
<td>Dihydromorphine</td>
<td>CYP2D6</td>
</tr>
<tr>
<td>Tramadol</td>
<td>O-desmethyltramadol</td>
<td>CYP2D6</td>
</tr>
<tr>
<td>Clopidogrel</td>
<td>Unidentified</td>
<td>CYP2C19</td>
</tr>
<tr>
<td>Amitriptyline</td>
<td>Nortriptyline</td>
<td>CYP2D6</td>
</tr>
<tr>
<td>Fluoxetine</td>
<td>Norfluoxetine</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Diazepam</td>
<td>Desmethyldiazepam</td>
<td>CYP2C19, CYP3A4</td>
</tr>
<tr>
<td>Carbamazepine</td>
<td>Carbamazepine-10,11-epoxide</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Chlordiazepoxide</td>
<td>Desmethylichlordiazepoxide</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Imipramine</td>
<td>Desipramine</td>
<td>CYP2C19, CYP2D6</td>
</tr>
<tr>
<td>Isosorbide Dinitrate</td>
<td>Isosorbide 5-mononitrate</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Morphine</td>
<td>Morphine-6-glucuronide</td>
<td>CYP2D6</td>
</tr>
<tr>
<td>Prazepam</td>
<td>Desmethyl-diazepam</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Prednisone</td>
<td>Prednisolone</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Primidone</td>
<td>Phenobarbital</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Verapamil</td>
<td>Norverapamil</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Zidovudine</td>
<td>Zidovudine triphosphate</td>
<td>CYP3A4, CYP2A6</td>
</tr>
<tr>
<td>Theophylline</td>
<td>1-methylxanthine and 3-methylxanthine</td>
<td>CYP1A2</td>
</tr>
<tr>
<td>Citalopram</td>
<td>desmethylcitalopram</td>
<td>CYP3A4, CYP2C19</td>
</tr>
<tr>
<td>Venlafaxine</td>
<td>O-desmethylenflaxine</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Quinidine</td>
<td>O-desmethyquinidine</td>
<td>CYP3A4</td>
</tr>
<tr>
<td>Cyclophosphamide</td>
<td>4-hydroxycyclo-phosphamide</td>
<td>CYP3A4, CYP2B6</td>
</tr>
</tbody>
</table>
b. $\beta_2$-adrenergic receptor ($\beta_2$AR)

- 12 polymorphisms reported in the ADRB2 coding region
- Gly16 allele predisposes patients to nocturnal asthma and asthma severity
- non-synonymous SNPs encoding for either Arg or Gly at position have been linked to altered responses to short acting $\beta_2$AR agonists (Gly better response than Arg)

c. Leukotriene receptors and 5-lipoxygenase

- polymorphisms in the 5-lipoxygenase gene (ALOX5) promoter are associated with differential responses to 5-lipoxygenase inhibitors
- polymorphisms in the leukotriene receptor have not been associated with reduced response

J. Limitations to implementation

1) High-Throughput DNA Analysis Technology: Costs, Data Standards and Future Technologies.
2) Information Management: Access, Security and System Structures.
3) Genomics & Genetics Education: Physicians, Pharmacists, Nurses and Consumers.
4) Point-of-Care Utilization of Genomics: Physician’s Office, Hospital, Pharmacy and Consumer.
6) Electronic Health Record Management and Utilization.
7) Translational Research: Establishing Linkages Between Allelic Information and Healthcare Outcomes.
K. References

4) http://cypalleles.ki.se/
9) [http://www.pharmgkb.org](http://www.pharmgkb.org)