Pharmacogenomics and Clinical Practice:
Ready for Prime Time?

Manju T. Beier, Pharm D, CGP, FASCP
Chief Scientific Officer
Natural Molecular Testing Corp.
Adjunct Clinical Associate Professor of Pharmacy
The University of Michigan

The Human Genome Project

The original goals of the Human Genome Project are complete:
- The complete draft sequence of the human genome was finished in April of 2005
- The vast majority of the 20,000 - 25,000 human genes have been identified
- This information is publicly available

But the story is far from over...
- Functions are still unknown for nearly half of known human genes
- Research is still ongoing at many genome research centers across the world
Single Nucleotide Polymorphisms (SNPs)

- Occur when a single nucleotide (A, T, C, or G) in the genome sequence is altered
  - Variable gene expressions that are found in >1% of population
- Most common type of genetic variation among people
  - ATCGCCGGA\textcolor{red}{\text{A}}\text{ACCTAGAGAC}
  - ATCGCCGGA\textcolor{red}{\text{G}}\text{ACCTAGAGAC}
- In an entire human genome there are approximately 10 to 30 million potential SNPs
- Found in both coding (i.e., gene) and non-coding regions of the human genome

The Need for Improved Therapeutics

The effectiveness of prescribed medications ranges from 20-95%

- 80% - analgesics
- 25% - cancer chemotherapy
- 30% - Alzheimer’s disease
- 60% - depression (SSRIs)
- 40% - incontinence
- 50% - inflammatory arthritis
- 60% - schizophrenia
- 50% - migraine (prophylaxis)
- 60% - asthma
- 60% - cardiac arrhythmias
Pharmacogenomics combines pharmacogenetics with genomic studies. Uses large groups of patients to evaluate how candidate drugs interact with a range of genes and their protein products.

Relevant Genes/Biomarkers - Pharmacogenomics

- Pharmacogenomics focuses on the 225 genes responsible for the approximately 1,900 drug metabolizing enzymes, and receptors…….
- CYP450 enzymes responsible for 80% of phase I metabolism
Drug Metabolizing Enzymes

- Polymorphisms occur in transporters and receptors, causing inter-individual variability in drug response
- Numerous metabolizing enzymes are polymorphic
  - CYP2C9, CYP2C19, and CYP2D6 polymorphisms can be clinically significant and are well defined
  - CYP 3A4/5 has polymorphisms but not completely clinically defined

Examples of CYP450 Substrate Drugs

<table>
<thead>
<tr>
<th>1A2</th>
<th>2C9</th>
<th>2C19</th>
<th>2D6</th>
<th>3A4</th>
</tr>
</thead>
<tbody>
<tr>
<td>Caffeine</td>
<td>Glipizide</td>
<td>Clopidogrel</td>
<td>Amitriptyline</td>
<td>Alprazolam</td>
</tr>
<tr>
<td>Clozapine</td>
<td>Nateglinide</td>
<td>Phenotoin</td>
<td>Atomoxetine</td>
<td>CCBs</td>
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<tr>
<td>Olanzapine</td>
<td>S-Warfarin</td>
<td>PPI</td>
<td>Carvedilol</td>
<td>Cyclosporine</td>
</tr>
</tbody>
</table>

Theophylline

- Metoprolol
- Timolol
- Midazolam
- Ziprasidone
- Tamoxifen
- Quetiapine
- Certain TCAs
- Simvastatin
- Codeine
- Tacrolimus

3A4 Family: Inhibitors and Inducers

- Inhibitors:
  - Antifungals (itraconazole, ketoconazole)
  - Fluvoxamine
  - Calcium channel blockers (many)
  - Macrolide antibiotics (erythromycin, clarithromycin)
  - HIV antivirals

- Inducers:
  - Carbamazepine
  - Phenobarbital
  - Phenotoin
  - Rifampin
  - St. John’s Wort
CYP Metabolic Activity: Four Bin Phenotypic Model

- **Extensive metabolizer**: an individual typically with the wild type or "normal" phenotype
  - Will likely metabolize a drug as anticipated in the package insert
- **Intermediate metabolizer**: an individual who possesses one partially functional or non-functional allele coding for a metabolizing enzyme
  - Will metabolize a drug, but at a reduced rate
- **Ultrarapid metabolizer**: an individual with increased expression of a metabolizing enzyme
  - Will metabolize a drug at a more rapid rate than "normal"
  - Will buildup active metabolites with prodrugs
- **Poor metabolizer**: an individual with decreased or no expression of a metabolizing enzyme
  - Will metabolize a drug very slowly or not at all
  - Will not produce active metabolite with prodrugs

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Poor Metabolizers

- Can result in toxicity/extreme side effects
- *Will not produce active metabolite with prodrugs*
- To a lesser degree, intermediate metabolizers will experience similar effects

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Ultrarapid Metabolizers

- Unlikely to experience therapeutic effect at normal therapeutic doses
- *Prodrugs will accumulate metabolites/active drugs quickly*
Various Phenotypes:
Clinical Consequences

<table>
<thead>
<tr>
<th>Drug Type</th>
<th>Metabolizer Phenotype</th>
<th>Effect on Drug Metabolism</th>
<th>Potential Clinical Consequences</th>
</tr>
</thead>
<tbody>
<tr>
<td>Prodrug</td>
<td>Poor to Intermediate</td>
<td>Slow</td>
<td>• Decreased levels of active drug</td>
</tr>
<tr>
<td>Metabolized to active drug</td>
<td></td>
<td></td>
<td>• Patient at risk for therapy failure</td>
</tr>
<tr>
<td></td>
<td>Rapid to Ultra Rapid</td>
<td>Fast</td>
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</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>• Increased therapeutic effect</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td>• Accumulation of active drug may lead to drug-induced side effects</td>
</tr>
<tr>
<td>Active Drug</td>
<td>Poor to Intermediate</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td>• Increased doses may be needed</td>
</tr>
</tbody>
</table>

Caveats for CYP2D6

- >80 alleles defined
- Significant variations of frequency in ethnic backgrounds
  - CYP 2D6 in Caucasians (approx.):
    - PM: 7-10%
    - IM: 15%
    - EM: 70% (normal metabolizers)
    - UM: 7% affects
      - 29% of Ethiopians

http://medicine.iupui.edu/clinpharm/ddis/ClinicalTable.aspx.
Medication Issues in Psychiatry

- Variation in individual clinical response
- Few experience complete symptom remission
  - Efficacy takes 6-8 weeks, longer in older adults
  - Long lag time before alternative med considered
- Majority continue to experience significant psychiatric symptoms
- Most have medication-induced side effects
- Watch out for drug-drug interactions!
- High risk of morbidity and mortality

Antidepressants and Response

- Many medications in different therapeutic classes
- Remission rate 35% - 45%
- Variation in medication response dependent on a number of factors:
  - age, gender, renal and hepatic functioning, medical co-morbidity, nutrition, substance use, smoking (induces enzymes CYP1A2), prior response to therapy (if any), adherence, genetics

http://www.fda.gov/drugs/scienceresearch/researchareas/pharmacogenetics/ucm083378.htm
Specific Case Scenarios

JJ is a 66 year old Veteran

- presented to his clinician with depression
- otherwise physically healthy with a history of hypertension
  - Medications:
    - Amitriptyline 50 mg po hs for sleep
    - Metoprolol 50mg ER po daily
- doctor prescribed fluoxetine 40 mg daily for depression

CYP2D6: Inhibitors
Best to Remember

- The enzyme is inhibited by several common drugs
  - Strong inhibitors
    - Fluoxetine, paroxetine, quinidine, bupropion, haloperidol
  - Moderate inhibitors: duloxetine
  - Weak inhibitors: amiodarone, cimetidine, citalopram, escitalopram, fluvoxamine, risperidone
  - Dose dependent: sertraline
Case Scenario

• The patient (JJ) responded quickly to fluoxetine
• Several weeks later, the patient was found unconscious at home
• What happened??
  ➢ Fluoxetine is a powerful inhibitor of 2D6
    ▪ Means more of his meds were in the body
    ▪ Amitriptyline and metoprolol are both metabolized by 2D6
    ▪ Drop in BP and HR and increased sedation

2D6 Poor Metabolizers:
What happens with Prodrugs?

• Exceptions to the patterns of drug presentation occur when the drug is a Prodrug (needs to be converted to active drug)
  ➢ Tamoxifen converted to endoxifen (2D6)
  ➢ Codeine converted to morphine (2D6)
  ➢ Hydrocodone (Vicodin) converted to hydromorphone (2D6)

Analgesics and Pain:
Role of Genotyping
Medication Issues in Pain

- Variation in individual clinical response
- Med-induced side effects
- Drug-drug interactions
- Drug-herb interactions (St. John’s Wort)
- Pain does not exist in isolation
  - Coexists with anxiety, depression and mood disorders
  - Chronic pain often requires antidepressants and other mood altering meds
- Neuropathic pain management
  - Challenging and requires use of psychoactive meds

WHO 3-Step Ladder

3: Severe

- Morphine
- Hydromorphone
- Methadone
- Levorphanol
- Fentanyl
- Oxycodone

± Adjuvants


Adjuvant Medications

Pharmacotherapy

May be used in all 3 stages of the WHO analgesic ladder

Have been developed for clinical indications other than analgesia and are not as effective as opioid analgesics in relieving pain

- Antidepressants
- Anesthetics
- Anticonvulsants
- Muscle relaxants
- α-adrenergic agonists
- NMDA blockers
- Antipsychotics
- Corticosteroids
Mr. Jones, Age 79  
**Pneumonia**

- Patient w/ pneumonia treated w/ antibiotics and w/ codeine for cough  
- On treatment day 2 patient was found unresponsive  
- Transfer into ICU, life support: intubation, respirator  
- Supportive care was successful – patient recovered fully  
- What happened?

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**Codeine is a Substrate of CYP2D6**

Consider the variation in codeine’s metabolism among PM, IM, EM, UM individuals  
2D6 conversion accounts for it's analgesic activity

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**CYP2D6 and Phenotypes and Codeine**

- Consequence of variants for prodrugs that need to be converted to active drugs:  
  - PM: no active drug  
  - IM: less active drug – approximately 20% lower concentrations of morphine than in EM  
  - UM: more active drug – up to 800% higher concentrations of morphine than in EM  
- Dose Adjustment (change from standard dose):  
  - PM: select a different drug  
  - UM: dramatic decrease in dose or use a different drug

Codeine & CYP2D6:
FDA Guidance 2007 and new 2012

- Limited evidence suggests that individuals who are ultra-rapid metabolizers (those with a specific CYP2D6 genotype) may convert codeine to its active metabolite, morphine, more rapidly and completely than other people.
- In nursing mothers, this metabolism can result in higher than expected serum and breast milk morphine levels.
- In a nursing mother known to be or suspected to be an ultra-rapid metabolizer of codeine, consider other options for relieving pain or persistent cough.
- Implications for geriatric/pediatric patients prescribed opioids containing codeine for pain relief?

Phenotypes and Drug-drug Interactions

- An individual with an extensive (normal) or intermediate metabolic phenotype (for 2D6) can be converted to a poor metabolizer when treated with an SSRI such as fluoxetine

Top Drug-drug Interactions in Pain/Psychiatry

- Combinations involving fluoxetine, paroxetine, bupropion and 2D6 substrates
- Combinations involving opioids (codeine, methadone, oxycodone, tramadol) AND
  - 2D6 (fluoxetine, paroxetine, duloxetine) and/or
  - 3A4 inhibitors (antifungals, fluvoxamine)
- Carbamazepine and phenytoin and St. John’s Wort as 3A4/5 inducers when combined with 3A4 substrates especially
### Examples of CYP450 Substrate Drugs

*(Pain Meds in Red)*

<table>
<thead>
<tr>
<th>Enzyme</th>
<th>Pain Meds</th>
<th>Prodrug Metabolite</th>
<th>CYP2D6 Metabolite</th>
<th>CYP3A4 Metabolite</th>
</tr>
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<tr>
<td>1A2</td>
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<td>Glipizide</td>
<td>Clonazepam</td>
<td>Tramadol (prodrug)</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Hydralazine</td>
</tr>
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<td></td>
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<td>Tramadol (prodrug)</td>
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**Examples of 2D6 Mediated Opioids:**

- **Tramadol**: converted to more active drug
  - Paroxetine (2D6 inhibitor) decreased efficacy in one trial

- **Hydrocodone**: converted to hydromorphone (Dilaudid)
  - 10-33 times more potent than hydrocodone
  - Inhibition by 2D6 affects some efficacy
  - 3A4 also forms norhydrocodone (not active)

**What Other 2D6 Mediated Opioids?**

- **Oxycodone**: metabolized by 2D6 and 3A4
  - To Oxymorphone (only 11%, but more potent)
  - To Noroxycodone by 3A4 is major pathway (less active than oxycodone)
  - So, 2D6 inhibitors AND 3A4 inhibitors are relevant
Non 2D6 Mediated Opioids

- morphine
- oxymorphone
- buprenorphine
- fentanyl
- methadone
- hydromorphone

What about Morphine?

- Very effective analgesic
- Many dosage forms
- CYP450 do not play a role!
  - Very few drug interactions
- Liver converts to glucuronides (active)
- Need to reduce dose in decreased kidney function

What about Methadone?

- Highly effective analgesic
- Has long and variable half life
- Need special expertise in dosing
  - #1 opioid for deaths per CDC (July 2012)
- Does not depend on active metabolites
- Metabolized by 3A4 and 2B6 (minor 2D6)
  - Many drug interactions
  - Inducers and inhibitors of 3A4 especially
What about Fentanyl?

• Very potent opioid
• Does not depend on active metabolites
• Metabolized by 3A4
  ➢ Many drug interactions
  ➢ Inducers and inhibitors of 3A4 especially

The Case of Warfarin

Warfarin Monitoring Woes

• Therapeutic range: INR 2-3 (2.5-3.5 for prosthetic heart valves)
• INR <2: risk of thromboembolic event
• INR >3: risk of bleeding complications
• Huge Monitoring Hassle Factor!

Source of Variability in Warfarin Dose Requirements

- Many clinical and environmental factors can influence warfarin response
  - Age, gender, race, height, weight, concomitant medications, foods (vitamin K), herbal ingestion etc.
  - Wide inter-individual variability in therapeutic efficacy
  - Remember the “3Ds”
- Despite knowledge of these factors, a large proportion of variability in warfarin dose requirements remains uncertain


Warfarin Dosing
FDA Label Revisions

- 2 million patients start warfarin every year!
- Changed package insert for warfarin August 2007
  - Further revised Feb 2010 (includes genetic testing info)
- Label now provides information regarding altered metabolism in CYP2C9 and VKORC1 genetic variants (may account up to 40% in variability)
- Home INR self-tests
  - Increase percent of time in therapeutic window
    - http://inrselftest.com/content/home
    - http://www.ptinr.com


Warfarin Pharmacogenetics

- CYP2C9
  - Metabolizes >90% of active (S-Warfarin)
  - Variant alleles associated with increased sensitivity to warfarin (CYP2C9*2, *3)
- Vitamin K epoxide reductase (VKORC1)
  - Inhibited by warfarin (PD effect)
  - Important for replenishment of vitamin K
  - Variant alleles of VKORC1 gene associated with altered response to warfarin: G/G, G/A, A/A

CYP2C9 Variant Alleles

- CYP2C9*2, CYP2C9*3 – most common variants
  - Seen in 20-40% of Caucasians, <10% Asians and African Americans
  - Associated with reduced CYP2C9 enzyme activity
- Variant alleles associated with
  - Lower mean doses of warfarin
  - Longer times to stabilization of INR
  - Higher risk for bleeding events

Using pharmacogenetics in real time to guide warfarin initiation: a clinician update.
Prospective Warfarin Trials

- Clarification of Optimal Anticoagulation Through Genetics (COAG)
  - [www.clinicaltrials.gov/ct2/show/NCT00839657](http://www.clinicaltrials.gov/ct2/show/NCT00839657)
  - compare two approaches to warfarin dosing
  - Estimated Study Completion Date: July 2013

- Genetics Informatics Trial (GIFT) of Warfarin to Prevent DVT
  - Estimated Study Completion Date: August 2015

The Case of Clopidogrel

Clopidogrel and Pharmacogenetics: The Conundrum

- FDA Drug Safety Communication 3/12/10: Reduced effectiveness of clopidogrel in patients who are poor metabolizers of the drug
Effectiveness of Plavix depends on activation to an active metabolite by the cytochrome P450 (CYP) system, principally CYP2C19. Poor metabolizers treated with Plavix at recommended doses exhibit higher cardiovascular event rates following acute coronary syndrome (ACS) or percutaneous coronary intervention (PCI) than patients with normal CYP2C19 function. Tests are available to identify a patient’s CYP2C19 genotype and can be used as an aid in determining therapeutic strategy. Consider alternative treatment or treatment strategies in patients identified as CYP2C19 poor metabolizers.

FDA: Black Box Warning
Revised 2010

Clopidogrel and Pharmacogenetics
So, What’s the Problem?

• Clopidogrel is a prodrug: it has to be metabolized by the body (by CYP2C19) before it can be biologically active
• Clopidogrel metabolism shows genetic polymorphisms
• Possible drug interactions with proton pump inhibitors (PPIs)?
  ➢ Omeprazole is a 2C19 inhibitor, other PPIs too
  ➢ Conflicting data so far in clinical studies
  ➢ For those who require a PPI, pantoprazole is probably the best choice

Clopidogrel and Pharmacogenetics:
CYP2C19

• The CYP2C19*1 allele has fully functional metabolism
• The CYP2C19*2 and *3 alleles have no functional metabolism
• These two alleles account for most of the reduced function alleles in patients of Caucasian (85%) and Asian (99%) descent classified as poor metabolizers
• The CYP2C19*4, *5, *6, *7, and *8 and other alleles may be associated with absent or reduced metabolism of clopidogrel, but are less frequent than the CYP2C19*2 and *3 alleles
• A patient with two loss-of-function alleles (as defined above) will have poor metabolizer status
Clinical Event Rates in Studies

- Meta-analysis of data from nine pharmacogenetic studies of clopidogrel involving 9685 patients:
  - The hazard ratio for stent thrombosis was 2.67 (95% confidence interval 1.69–4.22) and 3.97 (95% confidence interval 1.75–9.02) in IMs and PMs, respectively, compared with NMs.
  - The risk for bleeding was greatest among URM, with an odds ratio of 3.27 (95% confidence interval 1.33–8.10) compared with NM.


Tamoxifen

- As a prodrug, tamoxifen is metabolized into its most active metabolite, endoxifen, by the CYP2D6
- Metabolism blocked in women who carry loss-of-function variant CYP2D6 alleles or who take drugs that inhibit CYP2D6 function
- Among the drugs that inhibit CYP2D6 are the SSRI antidepressants
  - Up to 25% of patients with breast cancer experience a major depressive disorder
  - Paroxetine and fluoxetine potently inhibit CYP2D6
  - Consider mirtazapine, venlafaxine, escitalopram

Tamoxifen and CYP2D6: What are the Facts?

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Fig 1: Risk of Breast Cancer Mortality Associated with Increasing Proportions of Antidepressant Use During Tamoxifen Treatment

<table>
<thead>
<tr>
<th>Drug</th>
<th>Adjusted Hazard Ratio for Death Compared to Placebo</th>
<th>P Value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Paroxetine</td>
<td>1.54</td>
<td>0.0028</td>
</tr>
<tr>
<td>Fluoxetine</td>
<td>1.32</td>
<td>0.05</td>
</tr>
<tr>
<td>Chlorpromazine</td>
<td>1.20</td>
<td>0.05</td>
</tr>
<tr>
<td>Sertaline</td>
<td>1.08</td>
<td>0.10</td>
</tr>
<tr>
<td>Venlafaxine</td>
<td>1.05</td>
<td>0.20</td>
</tr>
</tbody>
</table>

Potential Usefulness of Pharmacogenetic Testing

- Prediction of dose (initial and ongoing)
- Prediction of toxic side effects
- Prediction of therapeutic effects
- Prediction of drug-drug interactions
- Positive effects on measurable outcomes?
  - ER visits, hospitalizations, quality of life, cost
  - Some published, some ongoing

Preemptive Pharmacogenomic Testing

- Low cost sequencing
- Theoretically be available prior to any prescribing decision
- Considered for every patient just like allergy and age!!
- Storing the sequencing info for the future
  - Accessible in the future when needed
General Items to Ponder

- Cost Issues: Who Will Pay?
  - Potential emotional and financial liability associated with genetic information
- Consumer Protection and Privacy
  - In May 2008, the Genetic Information Nondiscrimination Act or GINA, was enacted
- Direct to Consumer Genome wide Profiling?
  - Has the time come?
  - Oversight of labs and quality control
    - FDA does not regulate most laboratory-developed tests
    - Evaluation of Genomic Applications in Practice and Prevention (EGAPP) http://www.eqppreviews.org/default.htm

Online Quick Databases

- Lexicomp http://www.lexi.com
- Epocrates https://online.epocrates.com/
- Clinical pharmacology http://www.clinicalpharmacology.com/

Select Useful Websites

- http://www.genome.gov The National Human Genome Research Institute (NHGRI)
  - http://www.youtube.com/user/GenomeTV
- http://www.pharmgkb.org
- http://www.pharmacogenomics.ucsd.edu PharmGenEd™
- http://www.fda.gov/drugs/scienceresearch/researchareas/pharmacogenetics/ucm083378.htm