Clinical Chemistry Trainee Council
Pearls of Laboratory Medicine
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TITLE: Introduction to Pharmacogenetics

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Slide 1:
Hello, my name is Ping Wang. I am Director of Clinical Chemistry at The Methodist Hospital in Houston TX and Assistant Professor at Weill Cornell Medical College. Welcome to this Pearl of Laboratory Medicine on "Introduction to Pharmacogenetics."

Slide 2:
In this presentation, I will first introduce the concept of pharmacogenetics. The traditional way of drug dosing has its limitations as demonstrated by the prevalence of adverse drug reactions and limited drug efficacy. This leads us to the benefits of pharmacogenetics-based dosing. I will then talk about the influence of genetics on drug metabolism and action.

The concepts of pharmacokinetics and pharmacodynamics are first introduced, followed by common genes involved in pharmacogenetics. I will briefly go through pharmacogenetic biomarkers in FDA-approved drug labels, then discuss current testing methodologies used in this field. Finally, I would like to introduce you to a few online resources that I found valuable if you would like to learn more about pharmacogenetics.

Slide 3:
The traditional way of "trial-and-error" dosing has significant limitations. One such limitation is demonstrated by the prevalence of adverse drug reactions. Adverse drug reactions (or ADRs) account for 3-11% of total U.S. hospital admissions, according to a meta-analysis study. The total number of ADRs is estimated to be 2 million/year, among which 110,000 cases are fatal. They are the 4-6th leading cause of death in the United States.

Slide 4:
This slide shows an incomplete list of drugs that were withdrawn from the U.S. market due to severe adverse drug reactions. Examples include thalidomide, phenformin, buformin, Fen-fen, etc. The question to be asked is whether targeted drug administration or dosing based on individual patient's genetics would help avoid these withdrawals.

Slide 5:
The limitations of the traditional ways of drug dosing are also demonstrated by limited drug efficacy in various drug classes. As shown below, the response rate of patients to cancer drugs is about 25%. The response rate to selective serotonin inhibitors is about 62%. For asthma, diabetes, or migraine drugs, the response rate is between 50 to 60%.

Slide 6:
Pharmacogenetics, or sometimes used interchangeably with pharmacogenomics, is a science that examines variations in genes that dictate drug response. In a broad sense, the genetic variation may be inherited or acquired. Classical pharmacogenetics deals with inherited germline variations, while broadly defined pharmacogenetics also includes acquired somatic variations in tumors, as both may dictate drug responses, especially responses to cancer therapies.

Pharmacogenetics explores the ways these variations can be used to predict whether an individual patient will have a good response, a bad response, or no response to a drug at all. In some cases, it can also help determine what dosage of the drug should be given to an individual. The goal is to give the right patient the right drug at the right time with the right dose.

The genetic variations we talk about here include single nucleotide polymorphisms, or SNPs, insertions, deletions, duplications, and translocations.

Slide 7:
The genetic variations listed on the previous slide are used to determine the genotype and haplotype of an individual for that specific gene of interest. It would be useful to introduce and define these two concepts as they are central to the discipline of pharmacogenetics.

Genotype refers to the specific allele inherited at a gene locus. For example, the gene *CYP2C9*, or cytochrome P450 2C9, has a single nucleotide polymorphism, or SNP, at nucleotide number 430, which may be changed to a T from a more prevalent C. This is described as *c.430C*, followed by an arrow, then T. *CYP2C9* therefore has two possible genotypes at this locus: *c.430 C* and *c. 430T*. The prevalence of each genotype may vary in different ethnicities. Historically, during the process of CYP450 variant research, different alleles were given different "*" designations. The nomenclature of cytochrome P450 genes is now defined by a committee, and can be accessed online. The website correlates each "*" designated alleles to its genetic variations and the original literature describing them. The web address is listed on Slide 16, the "Pharmacogenetics Resources" slide.

Haplotype refers to the collective genotypes of a number of closely linked loci on a chromosome. For example, the gene *VKORC1*, or vitamin K epoxide reductase complex subunit 1, has ten loci in its sequence that are polymorphic. The combination of certain nucleotides at each locus is called a haplotype. For example, the combination of CCGATCTCTG at each of the 10 listed loci is called haplotype *H1*. The prevalence of each haplotype may vary in different ethnicities.

Slide 8:
Pharmacogenetics-based drug selection and dosing has many advantages. It helps to achieve the optimal dose more quickly by avoiding the traditional "trial-and-error" method. It also helps to avoid drugs that individual patients would suffer severe side effects from, or would not benefit from. In addition, it can be used to stratify patients for clinical trial. Only patients who are likely to respond are included in the trial, thereby increasing the success rate and speeding up the pipeline. Companion diagnostics, which involve genotyping patients before drug prescriptions, are required today for several drugs. Finally, decreased adverse effects and increased drug efficacy may help improve the likelihood that patients adhere to their drug regimens.

Slide 9:
This slide shows examples of companion diagnostic tests approved by FDA to be used before prescriptions of certain drugs. For the complete list of all FDA-approved companion diagnostic tests, go to the website listed at the bottom of the page (http://www.fda.gov/MedicalDevices/ProductsandMedicalProcedures/InVitroDiagnostics/ucm301431.htm).

Slide 10:
Genetics can influence drug response, and then, clinical outcome, by influencing pharmacokinetics and/or pharmacodynamics of the drug.

Pharmacokinetics refers to what the human body does to the drug. This is described by the classical ADME pathway—absorption, distribution, metabolism, and excretion. Organs such as stomach, intestine, liver, and kidney are important in this process. Genetic variations in genes encoding drug transporters and metabolizing enzymes may affect function and quantity of the encoded protein, and thereby affecting drug concentration the body is exposed to.

Pharmacodynamics refers to what the drug does to the human body. Genetic variations in genes encoding drug targets and downstream molecules involved in mechanism of action of the drug may affect protein function and quantity, and thereby affecting drug response.

Slide 11:
In this slide, I give some examples of common genes frequently studied in pharmacogenetics. First, and the most commonly involved, are the genes encoding phase I metabolizing enzymes. The purpose of phase I metabolism is to introduce or expose reactive or polar groups to the usually lipophilic drugs. Phase I enzymes are usually oxidation, reduction, and hydrolysis enzymes. The most important phase I enzyme is the Cytochrome P450s. They are a superfamily of monooxygenases that typically oxidize drugs. Besides Cyp450s, Phase I enzymes also include esterases and hydrolases. These reactions may convert inactive prodrugs to active metabolites. For example, in the case of clopidogrel, the metabolizing enzyme cytochrome P450 2C19, together with several other CYP450s, is required to convert inactive clopidogrel to its active metabolite for the platelet inhibition effect. Phase I metabolism may also convert active drugs to inactive metabolites. For example, *CYP2C9* converts warfarin to its inactive metabolite, hydroxywarfarin.

Slide 12:
Genes encoding phase II metabolizing enzymes are also frequently involved in pharmacogenetics. Phase II metabolizing enzymes are conjugation enzymes that attach glucuronic acid, sulfate, glutathione, and methyl moieties,etc., to the products of phase I metabolism, to make the metabolites more water-soluble and readily excreted. Examples are *UGT1A1* for irinotecan and *TPMT* for azathioprine and 6-mercaptopurine.

The most commonly studied gene encoding drug transporter is the *ABCB1* or *MDR1* gene, encoding p-glycoprotein. Genes encoding other enzymes may include the *G6PD* and *AchE* genes. An example of drug receptor or target gene is *VKORC1* for warfarin.

Slide 13:
The FDA has incorporated some pharmacogenetics information into approved drug labels. This slide lists drugs whose labels indicate the need for pharmacogenetic characterization before the drugs can be used correctly for their indications. Most of these drugs are targeted cancer therapies. The pharmacogenetic genes involved are usually drug targets that characterize the specific molecular form of the cancer. It is important that pharmacogenetic characterization of the tumor is performed before the drug treatment is given. Clinical guidelines are available or are being developed for some of these drugs.

Slide 14:
This table lists drugs whose labels recommend pharmacogenetic characterization in order to prescribe or dose the drugs correctly to avoid severe adverse effects. Some of these adverse effects are emphasized in the format of black box warnings on the package inserts. For some of these drugs, such as warfarin, dosing guidelines have been developed to adjust dosage according to pharmacogenetic test results and clinical factors. For other drugs, such as abacavir, the drug should be completely avoided when pharmacogenetic result matches certain genotype.

I want to point out that these two tables in Slides 13 and 14 are by no means a complete list of all drugs with pharmacogenetics information on labels. For a complete list, please refer to the FDA website shown at the bottom of the page (http://www.fda.gov/drugs/scienceresearch/researchareas/pharmacogenetics/ucm083378.htm).

Slide 15:
I would like to devote this slide to briefly talk about testing methodologies in pharmacogenetics. Metabolism status may be obtained by phenotyping methods such as using probe drugs, western blot, or immunohistochemistry. These may reveal the function and/or quantity of the protein encoded by relevant genes. Alternatively, we may elect to directly probe the DNA sequence. Fluorescence in situ hybridization may detect the presence of specific sequences within cells. PCR is frequently used to amplify the target sequences, followed by mutation-specific enzyme digestion, electrophoresis, allele specific hybridization, various sequencing methods, or mass spectrometry analysis. Fluorescence conjugated probes may be used in some technologies to detect specific DNA sequences. Many of these methods are available as commercial assays. Some are FDA-approved and marketed as companion diagnostic tests for clinical laboratory use.

Slide 16:
Lastly, I have listed a few online resources that I found valuable if you are interested in learning more about pharmacogenetics:

- The "Science Primer" is a website from NCBI that introduces background information on NCBI genetic resources, including pharmacogenetics (http://www.ncbi.nlm.nih.gov/About/primer/pharm.html).
- The PharmGKB, or Pharmacogenetics and Pharmacogenomics Knowledge Base, is a resource that curates knowledge in this field, for both clinicians and researchers (http://www.pharmgkb.org).
- The Pharmacogenomics Research Network is organized by NIH, and is geared towards researchers to provide and coordinate research funding in this field (http://www.nigms.nih.gov/Research/FeaturedPrograms/PGRN).
- The SNP Consortium (http://snp.cshl.org) is the home website for the International HapMap Project, and has links to data deposits and SNP databases, such as the dbSNP database housed at NCBI (http://www.ncbi.nlm.nih.gov/snp).
- Finally, I have listed the CYP allele nomenclature database website, which correlates each "*" designated alleles to its genetic variations and the original literature describing them (http://www.cypalleles.ki.se).

Slide 17: References

Slide 18: Disclosures

Slide 19: Thank You from www.TraineeCouncil.org
Thank you for joining me on this Pearl of Laboratory Medicine on "Introduction to Pharmacogenetics." I am Ping Wang.