Medication DNA Insight

Technical Bulletin

Abacavir hypersensitivity

(DC:TB-0224.001 06DEC2012)

Report Type: Pharmacogenetics

About: Abacavir, which is used to treat HIV infection, is well tolerated by most people, but some individuals become hypersensitive with symptoms that include fever, skin rash, fatigue, gastrointestinal problems and respiratory problems. Hypersensitivity can be severe and, in rare cases, fatal.

Genetics: The mechanism of abacavir hypersensitivity is not known, but clinical symptoms suggest an immunological reaction influenced by genetic factors. Human leukocyte antigen-B (HLA-B) genetic variants have been associated with immunological response. Many studies have demonstrated an increased risk for abacavir hypersensitivity in patients with a variant HLA-B allele, HLA-B*5701.[1,2,3,4] Genetic testing for this allele can indicate whether an individual may be hypersensitive to abacavir.

Genetic testing for HLA-B*5701 prior to abacavir therapy has been shown to reduce the incidence of abacavir hypersensitivity.[5] In a 2008 *New England Journal of Medicine* study, screening HIV-1 infected subjects for the presence of HLA-B*5701 resulted in a 60% reduction in the incidence of clinically-suspected hypersensitivity compared to standard of care with no genetic screening. Suspected abacavir hypersensitivity reactions were reported in 7.8% (66/847) of subjects who received no HLA-B*5701 screening prior to abacavir treatment. In contrast, of patients who received abacavir only if they were negative for HLA-B*5701, only 3.4% (27/803) ($p<0.0001$) had suspected abacavir hypersensitivity reactions. These results predicted that 61% of HLA-B*5701 positive subjects would develop abacavir hypersensitivity during treatment with abacavir as compared to 4.5% of HLA-B*5701 negative subjects. Several other studies have replicated the clinical utility of screening for the presence of HLA-B*5701 prior to abacavir therapy.[6,7,8]

Recommendations: FDA's boxed warning on the Ziagen label states that patients with the HLA-B*5701 allele are at high risk for experiencing hypersensitivity reactions. The label also contains a recommendation for screening for the HLA-B*5701 allele before the initiation of abacavir treatment, regardless of whether or not the patient has tolerated abacavir in the past.[9]

The NIH Clinical Pharmacogenetics Implementation Consortium Guidelines strongly recommend HLA-B*5701 screening before abacavir is administered and do not recommend its use in HLA-B*5701-positive individuals. Moreover, the NIH also notes that patients testing negative for HLA-B*5701 who are administered abacavir should also be carefully monitored for hypersensitivity because they still have a low (3%) risk of developing a hypersensitivity reaction.[10] These recommendations are consistent with those of the U.S. Department of Health and Human Services and the European Medicines Agency.[10]

Possible Outcomes: Hypersensitive, Typical Risk

Markers or Alleles Tested: HLA-B [HLA-B*5701]

The test includes rs2395029, which is a marker for the HLA-B*5701 allele in Caucasians and Hispanics.[11,12,13,14] The linkage between rs2395029 and HLA-B*5701 has not been investigated in African or Asian populations (see Limitations and Warnings).

Specificity and sensitivity of the test: The presence of HLA-B*5701 has a positive predictive value of approximately 47% to 61% for either immunologically confirmed (IM) or clinically diagnosed (CL) abacavir hypersensitivity. Thus, 47% to 61% of patients who have the HLA-B*5701 allele would be predicted to develop a hypersensitive reaction to abacavir. The negative predictive value of the test is 96% to 100%. Thus, almost all patients who do not have the HLA-B*5701 allele will not develop a hypersensitive reaction to abacavir.[5,10,15]

Measurement	Method of Diagnosis	Percentage of Patients
Sensitivity - % of people with hypersensitivity who will get a positive test result	IM*	100%
Sensitivity - % of people with hypersensitivity who will get a positive test result	CL*	46% - 50%
Specificity - % of people without hypersensitivity who will get a negative test result	IM/CL	97%
Positive Predictive Value - % of people with a positive test result who will become hypersensitive	IM/CL	47% - 61%
Negative Predictive Value - % of people with a negative test result who will not become hypersensitive	IM/CL	96% - 100%

*IM, immunologically confirmed by initial clinical evaluation, followed by a positive result on an abacavir skin patch test; CL, clinically diagnosed by reported symptoms.

Ethnic Distribution of Tested Alleles: Abacavir hypersensitivity is observed in about 8% of individuals of Western European ancestry[5,6] but is much less common in East Asian populations.[16,17,18] Correspondingly, the prevalence of HLA-B*5701 is about 6% to 7% in Western Europe and U.S. Caucasians but less than 1% in East Asian (Korean, Chinese, Japanese, Taiwanese) and African populations. The prevalence of the risk allele is higher in South and Southeast Asian populations; the frequency is about 5% to 20% in Asian Indian populations and about 4% among Thais.[10]

Limitations and Warnings: The marker tested for HLA-B*5701 applies to patients of Caucasian or Hispanic ethnicity and may not apply to other ethnic populations. Approximately 6% of patients that receive a positive test result may not be HLA-B*5701-positive and, thus, may not be at increased risk of a hypersensitivity reaction to abacavir.[10,12,13] If clinically indicated, patients of non-Hispanic or non-Caucasian ethnicity could be advised to undergo HLA sequencing to assess their risk of abacavir hypersensitivity. Patients testing negative for HLA-B*5701 may develop abacavir hypersensitivity.

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
995.27 Other drug allergy Applies to: • Allergic reaction NEC (due) to correct medical substance properly administered • Drug allergy NOS • Drug hypersensitivity NOS • Hypersensitivity (due) to correct medical substance properly administered	N/A

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Aminoglycoside antibiotics-induced hearing loss

(DC:TB-0225.001 06DEC2012)

Report Type: Pharmacogenetics

About: For more than 60 years, aminoglycoside antibiotics such as streptomycin, gentamicin, neomycin, paromomycin, kanamycin, spectinomycin, amikacin, netilmicin and tobramycin have been widely used. They continue to be used, especially in developing countries, for the treatment of severe bacterial infections. However, aminoglyoside use also carries the risk of both nephrotoxicity and ototoxicity.[1] Certain genetic variants further increase the risk of aminoglycoside-induced ototoxicity.

Genetics: The most frequent cause of inherited aminoglycoside-induced ototoxicity is the 1555A>G mutation in the mitochondrial MT-RNR1 gene.[2] Mitochondria, which are organelles that provide energy for the cell, have DNA molecules (mtDNA) that are distinct from the chromosomal DNA in the nucleus of the cell. The MT-RNR1 gene encodes an RNA component of the mitochondrial ribosome called 12S rRNA.[3] The mechanism by which the 1555A>G mutation leads to the death of hair cells in the inner ear after administration of aminoglycoside antibiotics is not completely understood, but may involve decreased mitochondrial protein synthesis, decreased energy production and increased reactive oxygen species formation.[1]

Individuals with the 1555A>G mutation are at risk because even a single course (a standard multi-dose regimen prescribed by a physician) of treatments with aminoglycoside antibiotics will cause severe hearing loss. In every known case, individuals always suffer significant and irreversible hearing loss within a few days to weeks after aminoglycoside treatment.[2] Since mitochondrial genes are maternally inherited, as all the cytoplasm, which includes mitochondria, comes from the egg, every child of a female carrier of the mutation is likely to inherit the 1555A>G mutation, while the children of male mutation carriers will not.[2]

Additionally, individuals carrying the 1555A>G mutation are at risk for late-onset sensorineural hearing loss even without exposure to aminoglycoside antibiotics. Approximately 40% and 70% of 1555A>G carriers will develop hearing loss by age 30 and 65, respectively.[2,4,5]

Each cell contains thousands of mtDNA molecules because there are multiple copies of the mtDNA molecule in each of the hundreds of mitochondria that are found in the cell.[4] The 1555A>G mutation is always homoplasmic, meaning that all copies of the mtDNA in the cell will carry the mutation.[2,4,5]

Mode of Inheritance: Maternal[2]

Possible Outcomes: Typical Risk, Do Not Prescribe

Markers or Alleles Tested: MT-RNR1 [1555A>G]

Recommendations: Patients with the 1555A>G mutation should avoid treatment with aminoglycoside antibiotics, and alternative antibiotics should be prescribed when necessary. However, these patients may still be at risk of hearing loss even without exposure to aminoglycoside antibiotics. These patients should also avoid noise exposure, and regular audiometric assessment may be indicated.[2]

Female patients with the 1555A>G mutation who have or are planning to have children should inform their pediatrician so that aminoglycoside antibiotics can be avoided. Patients with the 1555A>G mutation should be counseled that the mutation is transmitted maternally and that other family members may also be at risk of aminoglycoside-induced hearing loss.[2]

Ethnic distribution of tested alleles: The 1555A>G mutation is found in all ethnic groups, is carried by approximately 1 in 500 people in Western countries and is found in 20-30% of deaf individuals from Spain and Asia.[2,6,7]

Limitations and Warnings: Hearing loss is always a possible side effect of aminoglycoside use, even in the absence of the 1555A>G mutation.[1,2,4,8]

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
976.6 Poisoning by anti-infectives and other drugs and preparations for ear, nose, and throat	N/A

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Beta-blockers

(DC:TB-0233.002 07MAY2013)

Report Type: Pharmacogenetics

About: Beta-blockers (carvedilol, metoprolol, etc.) are used as a standard therapy for heart failure.[1] This class of drugs antagonizes beta-adrenergic receptors and down-regulates the effects of catecholamine hormones (epinephrine and norepinephrine). Genetic variants that affect beta-adrenergic signaling may influence outcomes in heart failure patients treated with beta-blockers.

Genetics: G-protein-coupled receptor kinases (GRKs) desensitize signaling of beta-adrenergic receptors. A gain-of-function variant in GRK5 (Gln41Leu) is associated with survival benefits in heart failure patients.[1,2] The Gln41Leu mutation is thought to provide a “natural” beta block, producing a more rapid desensitization of the beta-adrenergic receptor. In patients without the Gln41Leu mutation, beta-blockers provided patients with a typical survival benefit. In patients with the Gln41Leu mutation, beta-blockers were unlikely to provide additional survival benefit to heart failure patients. However, it is important to note that in heart failure patients who are not treated with beta-blockers, individuals with Gln41Leu had increased survival compared to those without.[1] The Gln41Leu mutation is more common in individuals of African ancestry, which may explain why African-Americans have a lower chance of responding to beta-blockers compared to other groups. This genetic effect is found in African-American patients;[1] it is not known if the tested mutation has the same effect in patients of non-African ancestry.

Recommendations: NA

Possible Outcomes: Reduced Therapeutic Benefit, Typical Therapeutic Benefit

Markers or Alleles Tested: GRK5 [rs17098707]

Ethnic Distribution of Tested Alleles: The minor allele (Leu41) frequency is 1.7% in Caucasians and 23.1% in African-Americans.[2]

Limitations and Warnings: The results of this test are based on a study of heart failure patients. Thus, they may not apply to patients being treated with beta-blockers for other conditions.

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
995.29 Unspecified adverse effect of other drug, medicinal and biological substance Applies to: • Unspecified adverse effect of medicinal substance NEC properly administered	N/A

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Clopidogrel metabolism

(DC:TB-0237.001 06DEC2012)

Report Type: Pharmacogenetics

About: Clopidogrel (Plavix) is an oral anti-platelet agent used to inhibit blood clots in patients with coronary artery disease, peripheral vascular disease, and cerebrovascular disease. Clopidogrel is a prodrug that must be metabolized to an active form to be effective. The CYP2C19 enzyme, a member of the cytochrome P450 superfamily, metabolizes clopidogrel to the active metabolite via a two-step reaction. Multiple cytochrome P450 enzymes contribute to the metabolism of clopidogrel, but CYP2C19 acts in both steps of the reaction, accounting for 45% of the first oxidation step and for 20% of the second step.[1]

Genetics: Variants of the CYP2C19 gene that lead to reduced enzyme function have been shown to be associated with reduced metabolism of clopidogrel to its active form in many ethnic populations.[2,3,4,5,6] Individuals can be classified based on their CYP2C19 enzyme activity into four metabolizer groups: Ultrarapid Metabolizer (higher than normal enzyme activity), Extensive Metabolizer (normal enzyme activity), Intermediate Metabolizer (intermediate enzyme activity) and Poor Metabolizer (low or no enzyme activity).[7]

In 2010, FDA added a boxed warning to the Plavix label indicating that clopidogrel can be less effective in CYP2C19 poor metabolizers and that genetic tests can help define a therapeutic strategy. In 2011, the NIH Clinical Pharmacogenetics Implementation Consortium issued guidelines describing clinical actions that can be implemented based on metabolizer status (see “Recommendations” below).[7]

The evidence is strongest for patients who are being treated with clopidogrel and receive percutaneous coronary intervention (PCI). Poor or intermediate metabolizers who receive PCI are at significantly increased risk of stent thrombosis, which can result in myocardial infarction and death.[8,9,10] For other indications, many studies have shown that poor metabolizers (2% to 15% of patients) and intermediate metabolizers (18% to 45% of patients) may be at risk for adverse cardiac events, such as myocardial infarction and stroke, when treated with clopidogrel;[7,11] however, recent studies dispute this claim.[9,12,13]

Recommendations: FDA’s boxed warning on the Plavix label recommends that alternative treatments should be considered for patients identified as CYP2C19 poor metabolizers.[14] The NIH Clinical Pharmacogenetics Implementation Consortium Guidelines recommend that prasugrel or another alternative be considered for intermediate and poor metabolizers.[7] The Royal Dutch Association for the Advancement of Pharmacy’s Pharmacogenetics Working Group also recommends alternative therapies, such as prasugrel.[15]

Concurrent use of clopidogrel with CYP2C19 inhibitors may affect clopidogrel response, particularly in extensive and ultra-rapid metabolizers. In these individuals, concurrent use of clopidogrel with CYP2C19 inhibitors (see "Known CYP2C19

Inhibitors" table below) may result in a poor metabolizer phenotype; as such, these individuals may have increased risk for cardiac adverse events when being treated with clopidogrel.[14]

Concurrent use of clopidogrel with omeprazole or esomeprazole should be avoided.[14] Concurrent use of clopidogrel with other CYP2C19 substrates (see "Known CYP2C19 Substrates" table below) may affect clopidogrel response.[16]

A patient's CYP2C19 metabolizer status may result in unexpected responses to other drugs, such as benzodiazepines, phenytoin, barbiturates and others[17] (see "Known CYP2C19 Substrates" table below).

Possible Outcomes: Poor Metabolizer, Intermediate Metabolizer, Extensive Metabolizer, Ultrarapid Metabolizer

Markers or Alleles Tested: CYP2C19 [CYP2C19*2, CYP2C19*3, CYP2C19*4, CYP2C19*5, CYP2C19*6, CYP2C19*8, CYP2C19*17]

Ethnic Distribution of Tested Alleles

The CYP2C19 panel detects alleles that have a combined frequency of over 99% in major ethnic groups.[7]

CYP2C19	Allele	Caucasian	African	East Asian	Middle Eastern	Enzyme activity
Wild-type	*1	63%	68%	60%	87%	Normal
rs4244285	*2	15%	15%	29%	12%	None
rs4986893	*3	0.42%	0.52%	8.9%	1.1%	None
rs28399504	*4	0.25%	0.093%	0.049%	ND[a]	None
rs56337013	*5	0.0073%	ND	0.062%	ND	Reduced
rs72552267	*6	0.017%	0%	0%	ND	None
rs41291556	*8	0.35%	0%	0%	ND	Reduced
rs12248560	*17	21%	16%	2.7%	ND	Increased

[a]Not Determined

Predicted CYP2C19 Metabolizer Status[7]

CYP2C19 Diplotype	Predicted Metabolizer Status
*2-*8/*2-*8	Poor Metabolizer
*1/*2-*8, *17/*2-*8	Intermediate Metabolizer
*1/*1	Extensive Metabolizer
*1/*17, *17/*17	Ultrarapid Metabolizer

Limitations and Warnings: Many rare CYP2C19 variants have been identified, but are not part of this test. It is possible, but unlikely, that the patient may have a variant that is not included in this test.

In addition to the genetic variants included in this test, other genetic and nongenetic factors can influence the effective dose of clopidogrel, including variants in other genes, age, sex, nutrition, lifestyle, other medications and route of administration.[16]

CYP2C19 genotype and metabolizer status may also affect responses to other drugs[17].

Known CYP2C19 Inhibitors and Substrates Tables

Known CYP2C19 Substrates[18]	Known CYP2C19 Inhibitors[18]
Proton-pump Inhibitors	Proton-pump Inhibitors
lansoprazole	lansoprazole
omeprazole	omeprazole
pantoprazole	pantoprazole
rabeprazole	rabeprazole
Anti-epileptics	Other
diazepam	chloramphenicol
phenytoin	cimetidine
S-mephenytoin	felbamate
phenobarbitone	fluoxetine
	fluvoxamine
Other	indomethacin
amitriptyline	ketoconazole
carisoprodol	modafinil
citalopram	oxcarbazepine
chloramphenicol	probenicid
clomipramine	ticlopidine
clopidogrel	topiramate
cyclophosphamide	
hexobarbital	
imipramine N-deME	
indomethacin	
R-mephobarbital	
moclobemide	
nelfinavir	
nilutamide	
primidone	
progesterone	
proguanil	
propranolol	
teniposide	
R-warfarin	

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Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
995.29 Unspecified adverse effect of other drug, medicinal and biological substance Applies to: • Unspecified adverse effect of medicinal substance NEC properly administered	N/A

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Estrogen supplementation

(DC:TB-0228.001 06DEC2012)

Report Type: Pharmacogenetics

About: Combined hormonal contraceptives and post-menopausal hormone replacement therapy products contain estrogen. These medications by themselves pose an increased risk of blood clots (venous thrombosis), but the risk is even greater when they are used by individuals with certain inherited blood clotting disorders. Relatively common variants in two genes that code for components of the blood clotting cascade confer this increased risk.[1]

Genetics: The Factor V Leiden variant in the F5 gene, which encodes the factor V coagulation cofactor, and the G20210A mutation in the F2, or prothrombin gene, are associated with an increased risk of venous thrombosis in women using combined oral contraceptives or hormone replacement therapy.[2,3,4] Both the Factor V Leiden and the G20210A variants have dominant inheritance patterns, meaning that inheriting only one copy of the variant places a woman at increased risk of experiencing venous thrombosis if she uses estrogen supplementation.

By themselves, combined oral contraceptives increase the risk for blood clots in women 4-fold. Women who carry the Factor V Leiden mutation alone have an 8-fold increase in the risk for blood clots. Women who carry the Factor V Leiden mutation and who use combined oral contraceptives increase their risk for blood clots 35-fold.[1,5] In a meta-analysis of six case-control studies and one cohort study, presence of Factor V Leiden alone increased the risk of venous thromboembolism in combined oral contraceptive users over non-users.[1] In three case-control studies and one cohort study, the F2 G20210A variant alone also increased the risk of venous thromboembolism in combined oral contraceptive users over non-users.[6]

Post-menopausal hormone replacement therapy with oral estrogen also presents a risk to women with the Factor V Leiden and G20210A mutations. In women undergoing estrogen hormone replacement therapy, the Factor V Leiden mutation is associated with increased risk for deep vein thrombosis.[1,7] Additionally, a meta-analysis of six independent studies found that the Factor V Leiden and G20210A mutations increased the risk of blood clots in women undergoing estrogen hormone replacement therapy.[4] In an observational study of postmenopausal women carrying the Factor V Leiden or G20210A mutations, oral estrogen but not transdermal estrogen conferred additional risk of venous thromboembolism.[8]

Recommendations: According to the World Health Organization (WHO),[9] the U.K. Medical Eligibility Criteria[10] and the U.S. Centers for Disease Control and Prevention (CDC),[11] the use of combined oral contraceptives, the combined contraceptive patch or the combined contraceptive vaginal ring in individuals with known thrombogenic mutations (e.g., Factor V Leiden, prothombin mutation, protein S, protein C and antithrombin deficiencies) is an unacceptable health risk (level 4). The WHO also concludes that the evidence for combined oral contraceptives applies to combined injectable contraceptives. The Royal Dutch Association for the Advancement of Pharmacy's Pharmacogenetics Working Group recommends selecting alternative therapies to estrogen-containing oral contraceptives when a patient has the Factor V Leiden mutation and a family history of thrombotic events.[12]

The North American Menopause Society[13] and the Endocrine Society[14] acknowledge that thrombogenic mutations, such as Factor V Leiden, can increase the risk of venous thromboembolism, but they make no recommendations against using hormone replacement therapy in women with thrombogenic mutations. They do recommend thrombophilia screening prior to hormone replacement therapy use for women with a personal or family history of venous thromboembolism. For women with thromboembolic risk factors, the International Menopause Society suggests non-oral routes of estrogen or tibolone may be used if hormone replacement therapy is considered appropriate.[15]

The prevalence of individuals with one copy of both the Factor V Leiden and G20210A mutations is 1 in 1,000.[11]

Possible Outcomes: Increased Risk of Venous Thrombosis, Normal Risk of Venous Thrombosis

Markers and Alleles Tested: F5 [Factor V Leiden/R506Q]; F2 [G20210A]

Ethnic Distribution of Tested Alleles: The carrier rate for Factor V Leiden is 1 in 19 U.S. Caucasians, 1 in 45 Hispanic Americans, 1 in 83 African Americans, 1 in 222 Asian Americans and 1 in 80 Native Americans. In Europe, the mutation is particularly common with a carrier rate of 1 in 6 to 1 in 10 in southern Sweden and Greece and 1 in 33 to 1 in 50 in Italy and Spain. Similar high numbers have been found in many Middle Eastern countries.[16]

The carrier rate for G20210A is 2% to 5% in U. S. Caucasians. The mutation is found in 2% to 4% of healthy individuals in southern Europe, which is twice as high as the prevalence in northern Europe. G20210A is rare in Far Eastern populations, in Africa, and in indigenous populations of Australia and the Americas.[16]

Limitations and Warnings: Genetic variants in other proteins, such as protein S, protein C and antithrombin are known to increase the risk of venous thrombosis, but are not part of this test. Non-genetic factors known to increase the risk of venous thrombosis include age, obesity, trauma/surgery, smoking, pregnancy and airplane travel.[4]

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
E932.2 Ovarian hormones and synthetic substitutes causing adverse effects in therapeutic use Applies to: • Contraceptives, oral • Estrogens • Estrogens and progestogens combined • Progestogens	V78.9 Screening for unspecified disorder of blood and blood-forming organs

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Metoprolol metabolism

(DC:TB-0217.001 06DEC2012)

*Report Type: Pharmacogenetics

About: Metoprolol (MET) is one of the most commonly used beta-blockers for treatment of heart failure.[1] This cardioselective beta(1)-adrenergic blocker is also used for treatment of acute myocardial infarction, angina pectoris and mild to moderate hypertension.[2] Metoprolol is metabolized to inactive forms by a member of the cytochrome P450 superfamily, the CYP2D6 enzyme, which is responsible for 70% to 80% of the metabolism of metoprolol.[3]

Genetics: CYP2D6 variants play a major role in the pharmacokinetics of metoprolol, which translates into relevant clinical outcomes.[3,4,5,6] A wide range of variants in the CYP2D6 gene affect the enzyme activity, and individuals can be classified based on this activity into three metabolizer groups: Extensive Metabolizer (normal enzyme activity), Intermediate Metabolizer (intermediate enzyme activity) and Poor Metabolizer (low or no enzyme activity).

CYP2D6 poor and intermediate metabolizers treated with standard doses of metoprolol have increased plasma concentration of the drug and increased risk for bradycardia.[7,8,9,10,11,12,13] They may also have greater reductions in heart rate, diastolic blood pressure, and arterial pressure at typical doses.[3,6,14,15,16,17,18]

The largest and most representative study on CYP2D6-metabolized beta-blockers demonstrates that the CYP2D6 poor metabolizer status is associated with lower heart rate and blood pressure.[3] The association between CYP2D6 genotype and metoprolol response or side effects was not confirmed in some much smaller studies.[7,11,19,20]

Recommendations: The FDA-approved labels of metoprolol[12,13] indicate that concomitant use of CYP2D6 inhibitors (see "Known CYP2D6 Inhibitors" table below) with metoprolol may lead to higher than normal plasma levels of metoprolol at standard doses. CYP2D6 inhibitors are also likely to decrease the cardioselectivity of metoprolol. The Royal Dutch Association for the Advancement of Pharmacy's Pharmacogenetics Working Group also suggests dose adjustment or alternative medications for CYP2D6 poor metabolizers and intermediate metabolizers.[21] CYP2D6 genotype and metabolizer status may also affect a patient's response to other drugs (see "Known CYP2D6 Substrates" table below).

Possible Outcomes: Poor Metabolizer, Intermediate Metabolizer, Extensive Metabolizer

Markers or Alleles Tested: CYP2D6 alleles are classified as non-functional (*3, *4, *6 and *8), reduced-function (*9, *10, *17, *29 and *41), and normal-function (*1, *2 and *35). CYP2D6 phenotype prediction follows conventional guidelines (See below).

Predicted CYP2D6 Metabolizer Status

CYP2D6 Diplotype	Predicted Metabolizer Status
Two non-functional alleles	Poor metabolizer
One non-functional allele plus one reduced-function allele, or two reduced-function alleles	Intermediate metabolizer
One or two normal-function copies of the CYP2D6 gene	Extensive metabolizer

Ethnic distribution of CYP2D6 phenotypes

Ethnicity	Poor Metabolizer	Intermediate Metabolizer	Extensive Metabolizer
African American	2-8%	~30%	60-70%
Caucasian	5-10%	10-17%	70-80%
East Asian	<2%	50-60%	40-50%
Hispanic	3-10%	no data	no data

Limitations and Warnings: This test detects common CYP2D6 alleles that have a combined frequency of over 90% in major ethnic groups. Other alleles that may also affect CYP2D6 enzyme activity are not tested. For example, the test does not include alleles that are associated with higher than normal enzyme activity. As such, a small percentage of patients with the test result of CYP2D6 "Extensive Metabolizer" may metabolize metoprolol at higher than normal rates. This may result in failure to achieve optimal plasma concentrations at standard doses and an increased risk of therapeutic failure.[4,7,16]

Known CYP2D6 Inhibitors and Substrates Tables

Known CYP2D6 Inhibitors[22,23,24]		
amiodarone	escitalopram	paroxetine
bupropion	flecainide acetate	perphenazine
celecoxib	fluoxetine	primaquine phosphate
chloroquine phosphate	fluphenazine	propafenone
chlorpheniramine	fluvoxamine maleate	propoxyphene
chlorpromazine	halofantrine	quinacrine
cimetidine	haloperidol	quinidine
cinacalcet	histamine H1 receptor antagonists	ranitidine
citalopram	hydroxychloroquine	reduced haloperidol
clemastine	hydroxyzine	ritonavir
clomipramine	labetalol	sertraline
cocaine	levomepromazine	terbinafine
codeine	lomustine	thioridazine
darifenacin hydrobromide	lumefantrine	ticlopidine
delavirdine mesylate	methadone	tripelennamine
desipramine	metoclopramide	valdecoxib
diphenhydramine	mibefradil	vinblastine sulfate
doxepin	midodrine	vincristine sulfate
doxorubicin	moclobemide	vinorelbine tartrate
dronedarone	norfluoxetine	yohimbine
duloxetine		

Known CYP2D6 Substrates[22,24,25]		
alprenolol	fenfluramine	oxycodone
amitriptyline	fentanyl	paliperidone
amoxapine	flecainide	paroxetine
amphetamine	fluoxetine	penbutolol sulfate
aripiprazole	fluphenazine	pentazocine lactate
atomoxetine	fluvoxamine	perhexiline
betaxolol	formoterol	perphenazine
bisoprolol	galantamine	phenacetin
bufuralol	haloperidol	phenformin
captopril	hydrocodone	pindolol
carvedilol	hydrocortisone	promethazine
cevimeline	hydroxyamphetamine hydrobromide	propafenone
chlorpheniramine	iloperidone	propoxyphene
chlorpromazine	imipramine	propranolol
chlorpropamide	labetalol	protriptyline
cinacalcet	lidocaine	quetiapine
clomipramine	loratadine	ranolazine
clonidine	maprotiline	risperidone
clozapine	meperidine	ropivacaine
codeine	methadone	selegiline
cyclobenzaprine	methamphetamine	S-metoprolol
cyclophosphamide	methoxyamphetamine	sparteine
darifenacin hydrobromide	metoclopramide	tamoxifen
debrisoquine	metoprolol	tamsulosin
delavirdine mesylate	mexiletine	thioridazine
desipramine	minaprine	tiagabine
dexfenfluramine	mirtazapine	timolol
dextromethorphan	molindone	tolterodine
diphenhydramine	morphine	tramadol
dolasetron	nebivolol	trazodone
donepezil	nortriptyline	trimipramine maleate
doxazosin mesylate	olanzapine	venlafaxine
doxepin	ondansetron	yohimbine
duloxetine	orphenadrine citrate	zuclopenthixol
encainide		

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
N/A	V58.83 Encounter for therapeutic drug monitoring

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Interferon-alpha/ribavirin

(DC:TB-0218.001 06DEC2012)

Report Type: Pharmacogenetics

About: Hepatitis C virus (HCV) infection is the most common chronic, blood-borne infection in the US and affects 170 million worldwide.[1] The standard treatment for HCV infection, particularly in resource-limited settings, is a combination of pegylated (polyethylene glycol) IFN-alpha (PEG-IFN-alpha) and ribavirin (RBV).[2] IFN-alpha stimulates the natural defense against the viral infection, and pegylation enhances its antiviral potency.[2] Ribavirin is a nucleoside antimetabolite that inhibits replication of the virus.[3] Telaprevir and boceprevir are HCV NS3/4A protease inhibitors that require co-administration with PEG-IFN-alpha/RBV.[4,5]

IFN-based therapies are expensive and can induce severe side effects; therefore, it would be beneficial to distinguish responders from non responders prior to treatment screening.[6,7]

Genetics: A variant in the rs12979860 marker near the IL28B gene is a strong predictor of response to PEG-IFN-alpha/RBV.[4,5] The IL28B gene encodes a cytokine called interleukin 28B (IL28B), also known as IFN-lambda-3.

A meta-analysis of seven studies containing a total of approximately 4,700 patients found that variants at rs12979860 near the IL28B gene could be used as a predictor of treatment response,[8] as measured by sustained virological response (SVR). SVR is defined as an absence of hepatitis C viral RNA six months after therapy. SVR was higher in patients who were homozygous for the C allele of rs12979860 compared to patients who were heterozygous. Additionally, rs12979860 can explain some of the variability in the response rate between different ethnic groups.[8] Individuals of European ancestry have a higher chance of successful treatment than African Americans,[9] consistent with higher C allele frequency in European Americans (70%) compared to African Americans (40%) (see “Allele Frequency” table below).[9] Moreover, East Asians have a higher C allele frequency and higher SVR rate than European patients.[10]

Patients who are homozygous for the C allele also have higher rates of clearing HCV without treatment[11] and may have an increased SVR rate after liver transplantation.[12] Conversely, carriers of the T allele are at an increased risk of therapeutic failure,[13] and being homozygous for the T allele is associated with a more severe recurrence of hepatitis after transplantation.[14]

Precisely how rs12979860 variation affects response to PEG-IFN-alpha/RBV is not well understood.[15] Other variants near the IL28B gene have been shown to alter the expression, stability or receptor binding of IL28B.[15] Rs12979860 is located in a DNA methylation site, and it has been speculated that the C variant may decrease expression of IL28B.[16]

The association of IL28B variants with treatment response varies with the strain of the HCV virus. The association is strongest for patients infected with the HCV-1 virus, the viral strain most commonly found worldwide. In patients infected

with HCV-2, -3 or -6, however, IL28B variants are weakly associated with response to PEG-IFN-alpha/RBV.[15,17,18] For patients infected with HCV-5 virus, IL28B variants have not been shown to be associated with response to PEG-IFN alpha/RBV.

Telaprevir and boceprevir are newly approved drugs for treatment of Hepatitis C. They are inhibitors of the viral NS3/4A protease and are indicated for patients with a null or partial response to PEG-IFN-alpha/RBV treatment.[4,5] These drugs require co-administration with PEG-IFN-alpha/RBV. In addition, many candidate drugs for treating hepatitis C are in various phases of clinical trials.[2] Thus, genotyping may help determine the best treatment strategy for HCV-infected patients.[7]

Recommendations: NA

Possible Outcomes: Poor Responder, Responder

Markers or Alleles Tested: IL28B [rs12979860]

Ethnic Distribution of Tested Alleles:

Population	C allele frequency (rs12979860)[1]
Biaka Pygmies	23.5%
Zaramo	37.2%
Yoruba	31.2%
Hungarians	65.1%
Irish	73.9%
European-American	67.4%
Druze	77.6%
Yemenite Jews	69.5%
Indians	65.5%
Laotians	93.6%
Chinese, San Francisco	97.5%
Koreans	93.5%
Micronesians	98.6%
Pima, Mexico	55.5%
Mayans	37.5%
Ticuna	20.2%
Karitiana	82.4%

Limitations and Warnings: In patients infected with HCV-2, -3 or -6 strains, the IL28B variants are weakly associated with treatment response.[15,17,18]

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
995.29 Unspecified adverse effect of other drug, medicinal and biological substance Applies to: • Unspecified adverse effect of medicinal substance NEC properly administered	N/A

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Beta-blockers, LVEF response

(DC:TB-0208.002 07MAY2013)

Report Type: Pharmacogenetics

About: Beta-blockers (carvedilol, metoprolol, etc.) are used as a standard therapy for heart failure.[1] This class of drugs antagonizes beta-adrenergic receptors and down-regulates the effects of catecholamine hormones (epinephrine and norepinephrine). Genetic variants that affect beta-adrenergic signaling may influence outcomes in heart failure patients treated with beta-blockers.

Genetics: In heart failure patients, the benefits of treatment with beta-blockers for left ventricular ejection fraction (LVEF) are associated with variants in the ADRB1 gene, which encodes the beta(1)-adrenergic receptor.[2] A meta-analysis of three pharmacogenetic studies in heart failure patients found that individuals who are homozygous for the Arg389 allele have a greater increase in LVEF in response to beta-blockers (carvedilol, metoprolol or bisoprolol) compared to individuals who carry the Gly389 allele.[2] This genetic effect was also reported in a study involving patients who underwent 1.5 years of carvedilol treatment.[3]

In a number of small studies, the Arg389 allele has been studied in relation to other phenotypes, including exercise heart rate, response to rate-control therapy in atrial fibrillation, changes in heart rate and blood pressure in patients with essential hypertension and LV hypertrophy, and transplant-free survival in heart failure patients.[4,5,6,7] However, it is still controversial whether Arg389 modifies the outcomes of beta-blocker therapy. Some studies showed no association, while others reported significant associations but do not always agree on the direction of the genetic effect.

Recommendations: NA

Possible Outcomes: Enhanced Benefit, Beneficial

Markers or Alleles Tested: ADRB1 [rs1801253]

Ethnic Distribution of Tested Alleles:

Ethnicity	Arg389 allele (C allele) frequency[8]
Caucasian	~70%
East Asians	75-85%
Africans	~60%

Limitations and Warnings: NA

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test,

nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
995.29 Unspecified adverse effect of other drug, medicinal and biological substance Applies to: • Unspecified adverse effect of medicinal substance NEC properly administered	N/A

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Methotrexate toxicity

(DC:TB-0226.001 06DEC2012)

Report Type: Pharmacogenetics

About: Methotrexate (MTX) is a chemotherapeutic agent used in the treatment of lymphoma and leukemia, as well as uterine, breast, skin, ovarian and other cancers. MTX is also used to treat very severe and disabling psoriasis or in hematopoetic stem cell transplantation to prevent graft-versus-host disease. Some patients taking MTX may experience many and/or severe side effects, which are often referred to as MTX toxicity.[1]

Genetics: The T allele of the rs1801133 marker (C677T variant) in the MTHFR (5,10-methylenetetrahydrofolate reductase) gene, which is important for folate metabolism, was shown to be associated with MTX toxicity in patients with rheumatoid arthritis. The T allele results in an amino acid change that leads to reduced enzyme activity. Homozygotes for the T allele have approximately 30% of the expected MTHFR enzyme activity, and heterozygotes have approximately 65% activity, compared to the most common genotype, C allele homozygotes. Reduced MTHFR enzyme activity may result in reduced elimination of MTX, thus resulting in higher than expected MTX plasma concentrations and increasing the likelihood of MTX toxicity.[2]

While other MTHFR mutations are associated with MTHFR deficiency, only the C677T variant has shown significant association with methotrexate toxicity. In a meta-analysis of eight small studies, individuals with a T allele were shown to have a 1.7-fold increased risk for MTX-induced side effects.[3] These studies included patients from India, Japan, South Korea, Israel and the Netherlands. Additionally, a meta-analysis of 14 studies demonstrated that the T allele was associated with an increased risk of MTX-induced toxicity (liver toxicity, myelosuppression, oral mucositis, gastrointestinal toxicity and skin toxicity) in patients with acute lymphoblastic leukemia (ALL).[4]

Most studies with statistically significant data indicate an association between the T allele with MTX-induced side effects in patients with rheumatoid arthritis and ALL. It should be noted, however, that a 2011 meta-analysis did not identify a significant association between the C677T variant and MTX toxicity in patients with rheumatoid arthritis.[5] Association of the T allele with MTX toxicity has also been observed in patients undergoing hematopoetic cell transplantation and in patients with high-grade non-Hodgkin's lymphoma, acute leukemia, ovarian cancer, breast cancer, or juvenile idiopathic arthritis.[2] However, these studies are relatively small and controversial. In addition to MTX toxicity, the T allele has been associated with lowered efficacy of MTX, such as reduced anti-tumor activity or reduced survival in some studies but not others. The T allele has also been shown to be associated with therapeutic response to a different chemotherapy, fluorouracil (5-FU), in some studies but not others.[6,7,8,9]

Recommendations: Varying the MTX dose or supplementing with folic or folinic acid (leucovorin) has been shown to reduce the risk of toxicity-related discontinuation of MTX treatment in patients with and without the T allele.[10,11,12]

Please also see the related tests: MTHFR deficiency and Genetic risk for decreased folate.

Possible Outcomes: Increased Risk, Typical Risk

Markers or Alleles Tested: MTHFR [rs1801133]

Ethnic Distribution of Tested Alleles: The minor allele frequency was approximately 29.4% to 33.5% in Caucasians.[5]

Limitations and Warnings: Some variants not reported in the test also result in altered MTHFR activity. Therefore, a negative result for the reported MTHFR variant does not rule out the presence of additional variants that can cause altered MTHFR activity related adverse effects upon MTX treatment.

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
995.29 Unspecified adverse effect of other drug, medicinal and biological substance Applies to: • Unspecified adverse effect of medicinal substance NEC properly administered	N/A

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Phenytoin hypersensitivity

(DC:TB-0221.001 06DEC2012)

Report Type: Pharmacogenetics

About: Phenytoin is used to treat epilepsy, accounting for approximately 52% of all prescriptions for antiepileptic drugs.[1] It acts by blocking voltage-sensitive sodium channels in neurons.[2] Monitoring phenytoin levels is often necessary due to the drug's narrow therapeutic range and large individual variability in clearance.[1] Use of phenytoin can lead to cutaneous adverse reactions that vary from mild maculopapular eruption with increasing severity to hypersensitivity syndrome, Stevens-Johnson syndrome (SJS) and toxic epidermal necrolysis (TEN). SJS and TEN are rare but life-threatening.[3]

Genetics: The HLA-B*1502 allele of the HLA-B gene has been shown to be associated with phenytoin-induced cutaneous adverse reactions in Asians. The association between HLA-B*1502 and phenytoin-induced SJS/TEN has been found in both Thai and Chinese populations.[4,5,6] The HLA-B gene encodes a human leukocyte antigen protein that is involved in immune response. The HLA-B*1502 allele has also been shown to be associated with increased risk of SJS/TEN for antiepileptic drugs that are related to phenytoin, such as carbamazepine and oxcarbazepine.[4,5,6,7]

Recommendations: FDA advises that alternative therapies should be considered for HLA-B*1502-positive patients, who are at an increased risk of phenytoin-induced adverse reactions.[8]

Possible Outcomes: Hypersensitive, Typical, Unknown

Markers or Alleles Tested: HLA-B [HLA-B*1502]

This test includes rs3909184 and rs2844682, which together tag the HLA-B*1502 allele in Han Chinese.[9] Patients with one or two HLA-B*1502 alleles are assigned a "Hypersensitive" result. Patients with no HLA-B*1502 alleles are assigned a "Typical" result.

For patients with a particular genotype (rs3909184 (G/C), rs2844682 (C/T)), the HLA-B*1502 status cannot be determined, and thus these patients are assigned an "Unknown" result. These patients may require further evaluation.

Ethnic Distribution of Tested Alleles: The HLA-B*1502 allele is more prevalent in individuals of Asian ancestry.[10] The HLA-B*1502 allele has been observed in about 10% to 15% of patients in parts of China, Thailand, Malaysia, Indonesia, the Philippines and Taiwan. The frequency of this allele in South Asian individuals, such as Indians, is about 2% to 4%, but the frequency may be higher in some groups. The frequency of HLA-B*1502 is much lower (less than 1%) in Japan and Korea.[11] The HLA-B*1502 allele is also less frequently found (less than 1%) in those of African, European, Hispanic or Native American descent.[10]

Limitations and Warnings: The markers tested for HLA-B*1502 are most applicable to patients of Han Chinese descent.[9] If clinically indicated, patients of other Asian ethnicities could be advised to undergo HLA sequencing to assess their risk of

phenytoin hypersensitivity. There are insufficient data to associate HLA-B*1502 with phenytoin hypersensitivity in other non-Asian ethnicities. Many HLA-B*1502-positive Asian patients treated with phenytoin will not develop SJS/TEN. Conversely, these reactions can still occur infrequently in HLA-B*1502-negative patients of any ethnicity. Therefore, healthcare professionals are advised to watch for symptoms in all patients.

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
E936.1 Hydantoin derivatives causing adverse effects in therapeutic use Applies to: • Phenytoin	N/A

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Phenytoin metabolism

(DC:TB-0222.001 06DEC2012)

Report Type: Pharmacogenetics

About: Phenytoin is used to treat epilepsy, accounting for approximately 52% of all prescriptions for antiepileptic drugs.[1] Monitoring phenytoin levels is often necessary due to the drug's narrow therapeutic range and large individual variability in clearance.[1] Phenytoin acts by blocking voltage-sensitive sodium channels in neurons[2] and is primarily metabolized to hydroxyphenytoin (p-HPPH)[3] by the CYP2C9 enzyme, which accounts for up to 90% of the drug's metabolism.[4] p-HPPH has no anticonvulsant properties but is associated with some side effects, such as gingival hyperplasia, somnolence, dry month and fatigue.[5]

Genetics: The most common reduced function CYP2C9 variants are the CYP2C9*2 and CYP2C9*3 alleles. *In vitro* studies have shown that CYP2C9*2 and CYP2C9*3 are associated with approximately 29% and 95% reductions in phenytoin clearance, respectively, as compared to the wild-type allele, CYP2C9*1[2]. The CYP2C9*6 allele is a rare loss-of-function variant found in African populations.[6,7] Different combinations of CYP2C9 alleles give rise to different levels of CYP2C9 enzyme activity. For example, individuals with two wild-type alleles (e.g., CYP2C9*1/ CYP2C9*1) are extensive metabolizers. Individuals with one wild-type and one reduced function allele (e.g., CYP2C9*1/ CYP2C9*2) are intermediate metabolizers. Individuals with two reduced function alleles (e.g., CYP2C9*2/ CYP2C9*2 or CYP2C9*2/ CYP2C9*3) are poor metabolizers. These genetically determined differences in phenytoin metabolism can lead to toxicity and variable efficacy.[8,9]

CYP2C9 intermediate and poor metabolizers have increased plasma concentrations of phenytoin[10,11,12,13,14] and increased risk of phenytoin-induced neurological toxicity.[4,8,15,16,17,18,19] Symptoms of neurological toxicity may include dizziness, nystagmus (lateral and vertical), ataxia, slurred speech, lethargy and mental confusion.[8]

Recommendations: The Pharmacogenetics Working Group of the Royal Dutch Association for the Advancement of Pharmacy recommends a 25% dose decrease of phenytoin for CYP2C9*1/CYP2C9*2 and CYP2C9*1/CYP2C9*3 individuals and a 50% dose decrease of phenytoin for CYP2C9*2/CYP2C9*2, CYP2C9*3/CYP2C9*3 and CYP2C9*2/ CYP2C9*3 individuals.[20] The German AGNP (Arbeitsgemeinschaft für Neuropsychopharmakologie und Pharmakopsychiatrie) strongly recommends therapeutic drug monitoring (TDM) of plasma phenytoin in all patients, regardless of CYP2C9 genotype, in their Consensus Guidelines for Therapeutic Drug Monitoring.[21] According to the Guidelines, the therapeutic reference range for phenytoin is 10 to 20 µg/ml and the alert level is 25 µg/ml, indicating a narrow therapeutic range.[21]

CYP2C9 metabolizer status may affect response to other drugs, such as warfarin, tolbutamide, glipizide, celecoxib and fluvastatin.[22,23]

Possible Outcomes: Poor Metabolizer, Intermediate Metabolizer, Extensive Metabolizer

Markers or Alleles Tested: CYP2C9 [CYP2C9*2, CYP2C9*3, CYP2C9*6]

Ethnic Distribution of Alleles:[24]

Gene	Allele	African	Caucasian	Japanese	Chinese
CYP2C9	*1	80.6	77.9	96.5	95.8
CYP2C9	*2	1.2	15.9	0	0.1
CYP2C9	*3	0	5.7	3.4	4.1
CYP2C9	*6	1.2	0	0	0

Limitations and Warnings: NA

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
995.29 Unspecified adverse effect of other drug, medicinal and biological substance Applies to: • Unspecified adverse effect of medicinal substance NEC properly administered	N/A

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Proton pump inhibitors

(DC:TB-0219.001 06DEC2012)

Report Type: Pharmacogenetics

About: Proton pump inhibitors (PPIs), such as omeprazole, esomeprazole (S-isomer of omeprazole), lansoprazole, rabeprazole and pantoprazole, are potent gastric acid inhibitors that are used to treat gastroesophageal reflux disease, duodenal and gastric ulcers, erosive esophagitis, and pathological hypersecretory conditions, such as Zollinger-Ellison syndrome.[1] PPIs are converted to sulfenamides by the acidic environment of the stomach. The sulfenamide is the active form of the drug and inhibits gastric acid secretion by forming a disulfide bond with the gastric acid pump H K -ATPase enzyme.[2] PPIs, with the exception of rabeprazole, are primarily metabolized by the CYP2C19 enzyme in the liver prior to elimination from the body.[2]

Genetics: The CYP2C19 genotype is the most important genetic determinant for PPI treatment.[1] Individuals can be classified based on their CYP2C19 enzyme activity into four metabolizer groups: Ultrarapid Metabolizer (UM, higher than normal enzyme activity), Extensive Metabolizer (EM, normal enzyme activity), Intermediate Metabolizer (IM, intermediate enzyme activity) and Poor Metabolizer (PM, low or no enzyme activity).[3]

CYP2C19 PMs and IMs have lower clearance rates and higher plasma concentrations of PPIs than CYP2C19 EMs. Therefore, they are likely to benefit more from the PPI treatment and may experience stronger suppression of gastric acid secretion, an improved eradication rate of *Helicobacter pylori* infection, and improved cure rates of gastroesophageal reflux disease, esophagitis or ulcers.[1,2,4] PPIs are very safe drugs, even in CYP2C19 PMs, who are expected to have elevated plasma concentrations of PPIs.[1,4] In CYP2C19 EMs, the clearance of omeprazole, lansoprazole and pantoprazole is significantly increased, resulting in lower plasma concentrations, compared to PMs.[1] Thus, EMs may experience insufficient suppression of gastric acid secretion and a decreased therapeutic effect in some cases.[1,2,4] CYP2C19 UMs have decreased drug exposure and are at increased risk of therapeutic failure.[5,6]

For *H. pylori* eradication, PPIs are typically used in dual and triple therapies together with the antibiotics amoxicillin and/or clarithromycin;[1,7,8] the PPIs make the antibiotics more stable by reducing intragastric acidity.[2] Genetic testing for CYP2C19 has been shown to predict efficacy of *H. pylori* eradication treatment. For example, one study reported that the cure rates for *H. pylori* infection were 29%, 60% and 100%, respectively, for CYP2C19 EM, IM and PM individuals.[9] However, the success of eradication depends on additional critical factors, including patient compliance, the choice of PPI regimen, type of therapy (i.e., dual or triple) and antibiotic resistance of *H. pylori*.[1] Thus, the choice of PPI and regimen is considered to be the most practical approach for *H. pylori* eradication.[1]

Recommendations: The Pharmacogenetics Working Group of the Royal Dutch Association for the Advancement of Pharmacy recommends dose increases for CYP2C19 UM individuals using omeprazole, esomeprazole, lansoprazole or pantoprazole for *H. pylori* eradication treatment.[10]

The FDA-approved label for Plavix warns against concurrent use of omeprazole or esomeprazole with clopidogrel because omeprazole can inhibit CYP2C19 activity and reduce the effectiveness of clopidogrel.[11] Thus, patients who are at risk for heart attack or stroke will not receive the full anti-clotting effect of clopidogrel if they are also using omeprazole.[12]

Possible Outcomes: Poor Metabolizer, Intermediate Metabolizer, Extensive Metabolizer, Ultrarapid Metabolizer

Markers or Alleles Tested: CYP2C19 [CYP2C19*2, CYP2C19*3, CYP2C19*4, CYP2C19*5, CYP2C19*6, CYP2C19*8, CYP2C19*17]

Ethnic Distribution of Tested Alleles

The CYP2C19 panel detects alleles that have a combined frequency of over 99% in major ethnic groups.[3]

CYP2C19	Allele	Caucasian	African	East Asian	Middle Eastern	Enzyme activity
Wild-type	*1	63%	68%	60%	87%	Normal
rs4244285	*2	15%	15%	29%	12%	None
rs4986893	*3	0.42%	0.52%	8.9%	1.1%	None
rs28399504	*4	0.25%	0.093%	0.049%	ND[a]	None
rs56337013	*5	0.0073%	ND	0.062%	ND	Reduced
rs72552267	*6	0.017%	0%	0%	ND	None
rs41291556	*8	0.35%	0%	0%	ND	Reduced
rs12248560	*17	21%	16%	2.7%	ND	Increased

Limitations and Warnings: Many rare CYP2C19 variants have been identified, but are not part of this test. It is possible, but unlikely, that the patient may have a variant that is not included in this test.

Other critical factors that may influence the therapeutic effect of PPIs include the following: patient compliance, choice of PPI regimen, type of therapy (i.e., dual, triple) and antibiotic used, age, sex, nutritional status, liver and kidney function, concomitant diseases and medications, pharmacogenetics of CYP3A4, and IL-1 beta polymorphisms.[1,13,14,15,16,17]

Though the CYP2C19 genotype is associated with therapeutic efficacy for omeprazole, lansoprazole and pantoprazole, it is considered to have little or no effect on the therapeutic efficacy of rabeprazole, which is mainly metabolized by a nonenzymatic reduction.[1,18]

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
995.29 Unspecified adverse effect of other drug, medicinal and biological substance Applies to: • Unspecified adverse effect of medicinal substance NEC properly administered	N/A

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Simvastatin-induced myopathy

(DC:TB-0229.001 06DEC2012)

Report Type: Pharmacogenetics

About: Simvastatin is a member of the statins, a class of cholesterol-lowering drugs whose major adverse effect is skeletal muscle toxicity. Approximately 5% to 10% of patients taking statins experience muscle pain (myalgia).[1] A small portion of patients, (1.5% to 5.0%) may develop more severe symptoms indicating muscle degradation (myopathy).[1] In rare cases (0.1 to 0.2 cases per 1,000 person-years), severe muscle damage leads to acute, potentially lethal kidney failure (rhabdomyolysis).[1,2]

Genetics: Simvastatin-induced myopathy has been shown to be influenced by variation in the SLCO1B1 gene.[3] Approximately 60% of myopathy cases in a simvastatin (80 mg/day) clinical trial were attributed to the C allele of the rs4149056 marker in the SLCO1B1 gene.[3] SLCO1B1 encodes the organic anion-transporting polypeptide 1B1 (OATP1B1, also known as OATP-C or OATP2), which regulates the hepatic uptake of statins and other drugs. The C allele at rs4149056 reduces the activity of the OATP1B1 transporter,[4] leading to increased blood simvastatin levels and the potential for increased toxicity to the muscles. However, available clinical data are insufficient to show whether the SLCO1B1 variant also alters myopathy risk associated with the use of statins other than simvastatin.[5]

The risk of myopathy varies with the type of statin and is dose-related. Some statins are associated with lower risk of myopathy compared with others,[1] and the pharmacokinetic effects of variants of rs4149056 are not uniform for different statins.[6,7] The incidences of myopathy and rhabdomyolysis while taking 80 mg simvastatin daily are disproportionally higher than those with lower doses.[8]

Genetic variation in SLCO1B1 also affects pharmacokinetics of other drugs, such as methotrexate and HIV protease inhibitors.[9]

Recommendations: The NIH Clinical Pharmacogenetics Implementation Consortium (CPIC) published guidelines for SLCO1B1 genotyping and simvastatin-induced myopathy, recommending reduced dose or alternative statins for patients with the C allele at rs4149056. The CPIC also recommends routine surveillance of serum creatine kinase levels for those patients.[5]

Possible Outcomes: Increased Risk, Typical Risk

Markers and Alleles Tested: SLCO1B1 [rs4149056]

Ethnic Distribution of Tested Allele

Frequency of C allele of the rs4149056 marker in major ethnic groups.[5]

Ethnicity	C allele frequency (rs4149056)
Caucasian	15%
African	3%
Middle Eastern	20%
Asian	13%

Limitations and Warnings: Although the tested SLCO1B1 variant has the most significant genetic effect on the risk of simvastatin-induced myopathy in clinical studies, rarer mutations in SLCO1B1 that may also affect the function of the encoded protein are not screened in this test. Current knowledge is limited on the involvement of other genes in the metabolism and clinical effects of simvastatin. In addition to genetic effects, the risk of simvastatin-induced myopathy varies with the patient's age, gender, body mass index, ethnicity and other clinical factors.[5]

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Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
359.4 Toxic myopathy	N/A

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Voriconazole metabolism

(DC:TB-0220.002 29JAN2013)

Report Type: Pharmacogenetics

About: Voriconazole is a triazole antifungal drug that is used to treat a wide variety of fungal infections, such as *Candida*, *Cryptococcus*, *Aspergillus*, most dimorphic fungi and other yeasts and hyaline molds.[1] Therapeutic drug monitoring is recommended for optimizing voriconazole therapy and preventing toxicity due to the drug's high interpatient variability and nonlinear pharmacokinetic properties in adults.[1,1,2,3] Voriconazole serum concentrations less than 1 mg/L are associated with therapeutic failure, whereas levels greater than 5.5 mg/L have been linked to encephalopathy without improvement of therapeutic efficacy.[4]

Though the metabolic pathways of voriconazole are not completely understood, voriconazole is mainly metabolized to N-oxide,[5] which has low antifungal activity.[3] Metabolism to N-oxide is primarily mediated by enzymes encoded by CYP2C19 and CYP3A4, and to a lesser extent by CYP2C9,[6,7] though voriconazole has the highest affinity for CYP2C19.[8]

Genetics: Variants in the CYP2C19 gene have been associated with variability in voriconazole metabolism and exposure.[3] Individuals can be classified based on their CYP2C19 enzyme activity into four metabolizer groups: Ultrarapid Metabolizer (UM, higher than normal enzyme activity), Extensive Metabolizer (EM, normal enzyme activity), Intermediate Metabolizer (IM, intermediate enzyme activity) and Poor Metabolizer (PM, low or no enzyme activity).[9]

Variants of the CYP2C19 gene account for approximately half of the variability in oral clearance.[10,11] UMs have decreased plasma concentrations of voriconazole with exposure reduced by up to approximately 50% compared to EMs.[10,12,13] CYP2C19 PMs display decreased metabolism of voriconazole, increased plasma concentrations and approximately four-fold higher voriconazole exposure than EMs. Pharmacokinetic effects in IMs are between those of PMs and EMs.[3,5,10,11,13,14,15]

There is not yet sufficient evidence for an association of CYP2C19 genotype and pharmacodynamic effects such as treatment efficacy or adverse events in patients treated with voriconazole.[16,17] However, there are data linking VOR plasma concentrations to treatment failure or risk of adverse events. Low plasma concentrations of voriconazole, as found in some UMs, are associated with an increased risk of treatment failure and mortality in some non-genetic studies.[18,19,20,21] In a 2011 study, the UM genotype was found to be more prevalent in patients with extremely low plasma concentrations of voriconazole (0.03 mg/mL).[12] In another study, 5 patients with persistently low VOR concentrations (<0.35 mg/mL) died from invasive fungal infection.[18] In another study, 1 patient who had low plasma concentrations of voriconazole experienced therapeutic failure, which was reversed with a higher dose.[19]

In addition, high plasma concentrations of voriconazole have been shown to be associated with liver toxicity,[18,21,22,23] although a recent study indicates no association of plasma VOR levels with liver toxicity.[24]

Recommendations: The Royal Dutch Association for the Advancement of Pharmacy's Pharmacogenomics Working Group recommends therapeutic drug monitoring (TDM) for voriconazole treatment of CYP2C19 PMs and IMs.[25]

The FDA-approved label indicates that when CYP2C19 is inhibited, dose adjustment or monitoring for adverse events or lack of efficacy may be needed.

Co-administration of inhibitors of CYP2C19, CYP2C9 and CYP3A4 may increase the voriconazole plasma concentrations.[3] According to one study, the administration of ritonavir, a potent CYP3A4 inhibitor, resulted in a higher exposure with voriconazole, which might increase the risk of adverse reactions, particularly in CYP2C19 PMs.[15]

Possible Outcomes: Poor Metabolizer, Intermediate Metabolizer, Extensive Metabolizer, Ultrarapid Metabolizer

Markers Tested: CYP2C19 [CYP2C19*2, CYP2C19*3, CYP2C19*4, CYP2C19*5, CYP2C19*6, CYP2C19*8, CYP2C19*17]

Ethnic Distribution of Tested Alleles:

CYP2C19	Allele	Caucasian	African	East Asian	Middle Eastern	Enzyme activity
Wild-type	*1	63%	68%	60%	87%	Normal
rs4244285	*2	15%	15%	29%	12%	None
rs4986893	*3	0.42%	0.52%	8.9%	1.1%	None
rs28399504	*4	0.25%	0.093%	0.049%	ND[a]	None
rs56337013	*5	0.0073%	ND	0.062%	ND	Reduced
rs72552267	*6	0.017%	0%	0%	ND	None
rs41291556	*8	0.35%	0%	0%	ND	Reduced
rs12248560	*17	21%	16%	2.7%	ND	Increased

[a]Not Determined

Predicted CYP2C19 Metabolizer Status[9]

CYP2C19 Diplotype	Predicted Metabolizer Status
*2-*8/*2-*8	Poor Metabolizer
*1/*2-*8, *17/*2-*8	Intermediate Metabolizer
*1/*1	Extensive Metabolizer
*1/*17, *17/*17	Ultrarapid Metabolizer

Limitations and Warnings: Many rare CYP2C19 variants have been identified, but are not part of this test. It is possible, but unlikely, that the patient may have a variant that is not included in this test.

CYP2C19 genotype and metabolizer status may also affect responses to other drugs.[26]

Known CYP2C19 Substrates[27]	Known CYP2C19 Inhibitors[27]
Proton-pump Inhibitors	Proton-pump Inhibitors
lansoprazole	lansoprazole
omeprazole	omeprazole
pantoprazole	pantoprazole
rabeprazole	rabeprazole
Anti-epileptics	Other
diazepam	chloramphenicol
phenytoin	cimetidine
S-mephenytoin	felbamate
phenobarbitone	fluoxetine
	fluvoxamine
Other	indomethacin
amitriptyline	ketoconazole
carisoprodol	modafinil
citalopram	oxcarbazepine
chloramphenicol	probenicid
clomipramine	ticlopidine
clopidogrel	topiramate
cyclophosphamide	
hexobarbital	
imipramine N-deME	
indomethacin	
R-mephobarbital	
moclobemide	
nelfinavir	
nilutamide	
primidone	
progesterone	
proguanil	
propranolol	
teniposide	
R-warfarin	

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
N/A	V58.83 Encounter for therapeutic drug monitoring

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Warfarin

(DC:TB-0230.002 31JUL2013)

Report Type: Pharmacogenetics

About: Warfarin is the most frequently used oral anticoagulant worldwide, prescribed for prophylaxis and treatment of thrombotic disorders and thromboembolic events. Such indications include venous thrombosis, pulmonary embolism, atrial fibrillation and cardiac valve replacement. Warfarin is highly efficacious, but its narrow therapeutic index and large interindividual dosing variability lead to a high incidence of adverse events. Improper warfarin dosing is the second leading cause of drug-related emergency room visitation[1] and the number one cited reason for drug-related mortality.[2]

Warfarin acts as an anticoagulant through its ability to inhibit reduction of vitamin K by the vitamin K epoxide reductase complex subunit 1 (VKORC1) enzyme complex. Reduced vitamin K is an essential cofactor of gamma-glutamyl carboxylase, the enzyme that activates coagulation factors II, VII, IX and X. By decreasing the amount of reduced vitamin K available, warfarin depresses the activation of factors II, VII, IX and X into functional, coagulant proteins, and, therefore, decreases the ability of blood to clot. The primary metabolizing enzyme of warfarin is cytochrome P450 2C9 (CYP2C9).[3]

In 2010, the United States Food and Drug Administration (FDA) released a table of dosing recommendations for initiation of warfarin therapy based on VKORC1 and CYP2C9 genotypes. This pharmacogenetics-based dosing table significantly improved accuracy of therapeutic dose prediction compared to the traditional strategy of empirically determined dose.[4]

Genetics: The A allele of the -1639G>A mutation in the VKORC1 gene has been shown to decrease hepatic expression of VKORC1 and, therefore, increase patient sensitivity to warfarin.[5,6,7] Research studies have shown that the therapeutic dose of warfarin in patients with two copies of the A allele was less than the dose of patients with two copies of the G allele, with a difference up to 2.0 to 4.5-fold.[7,8,9]

Individuals carrying *2 or *3 genetic variants of CYP2C9 clear warfarin at a 30% to 50% or 80% to 90% slower rate, respectively, and exhibit increased serum levels of warfarin compared to carriers of only the reference wild-type variant *1.[10,11] CYP2C9*2 and CYP2C9*3 variants may decrease the dose required for effective anticoagulation and may increase the time necessary to achieve stable, therapeutic effect.[9,12] The CYP2C9*6 variant may also reduce metabolic activity and the dose required for effective anticoagulation.[11,13]

Customizing initial warfarin dose to VKORC1 and CYP2C9 genotypes may decrease patient risk of bleeding complications and may reduce the time required to achieve a stable, therapeutic effect.[12,14,15]

Recommendations: The FDA-approved label for warfarin recommends initial dosing based on VKORC1 and CYP2C9 genotypes in addition to clinical factors.[16] The National Institutes of Health (NIH) Clinical Pharmacogenetics Implementation Consortium guidelines recommend initial dosing based on VKORC1 and CYP2C9 genotypes.[11]

Standard doses of warfarin may cause bleeding complications in patients whose genotypes indicate increased or substantially increased sensitivity to warfarin. These patients may require lower initial doses of warfarin. Increased laboratory monitoring may be appropriate.

Classes of drugs that potentially interact with warfarin include the following:[16] inhibitors or inducers of CYP2C9, CYP1A2 and/or CYP3A4, anticoagulants, antiplatelet agents, nonsteroidal anti-inflammatory agents, serotonin reuptake inhibitors, antibiotics, antifungals, and botanical (herbal) products and foods. This list is not complete. Consult the warfarin drug label[16] and the labels of all concurrently used drugs for more specifics about warfarin drug interactions.

Possible Outcomes: Substantially Increased Sensitivity, Increased Sensitivity, Typical Sensitivity

Markers or Alleles Tested: VKORC1 -1639G>A [rs9923231]; CYP2C9 [CYP2C9*2/rs1799853, CYP2C9*3/rs1057910, CYP2C9*6/rs9332131]

Ethnic Distribution of Tested Alleles

Frequency of VKORC1 and CYP2C9 alleles differs significantly between racial and ethnic groups.[17]

Gene	Allele	Caucasian	African American	Asian	Hispanic
VKORC1	-1639: G	59.4%	89.2%	33.3%	56.4%
VKORC1	-1639: A	40.6%	10.8%	66.7%	43.6%
CYP2C9	*1	78.8%	86.7%	92.2%	82.2%
CYP2C9	*2	15.1%	2.8%	2.9%	6.9%
CYP2C9	*3	5.7%	2.0%	3.9%	6.4%
CYP2C9	*6	0.0%	1.0%	0.0%	0.5%

Predicted Warfarin Sensitivity Status

	VKORC1 -1639G>A genotype		
CYP2C9 genotype (below)	G/G	G/A	A/A
*1/*1	Typical sensitivity	Typical sensitivity	Increased sensitivity
*1/*2	Typical sensitivity	Increased sensitivity	Increased sensitivity
*1/*3	Increased sensitivity	Increased sensitivity	Substantially increased sensitivity
*1/*6	Increased sensitivity	Increased sensitivity	Substantially increased sensitivity
*2/*2	Increased sensitivity	Increased sensitivity	Substantially increased sensitivity
*2/*3	Increased sensitivity	Substantially increased sensitivity	Substantially increased sensitivity
*2/*6	Increased sensitivity	Substantially increased sensitivity	Substantially increased sensitivity
*3/*3	Substantially increased sensitivity	Substantially increased sensitivity	Substantially increased sensitivity
*3/*6	Substantially increased sensitivity	Substantially increased sensitivity	Substantially increased sensitivity
*6/*6	Substantially increased sensitivity	Substantially increased sensitivity	Substantially increased sensitivity

Limitations and Warnings: Warfarin can cause major or fatal bleeding. Additional genetic variants within VKORC1, CYP2C9, and other genes not included in this test are known to affect warfarin sensitivity. Not all genetic factors influencing warfarin sensitivity have been identified. Regular monitoring of INR (international normalized ratio) should be performed on all treated patients.

Not all factors influencing warfarin response are known. Important non-genetic factors include age, sex, weight, height, race, ethnicity, comorbidities, warfarin indication, target INR, and use of tobacco and interacting medications.[11]

Compatible ICD-9 Codes: Insurance coverage is not required for genetic testing. The diagnostic code (ICD-9) information provided herein is for insurance information purposes only and does not guarantee insurance coverage for any genetic test, nor is it intended to be a definitive list of diagnosis codes that may be applicable for any individual patient. The testing laboratory will pursue reimbursement directly from the patient should the patient's insurance carrier refuse to provide coverage.

Primary ICD-9 Code(s)	Screening ICD-9 Code(s)
E934.2 Anticoagulants causing adverse effects in therapeutic use	N/A

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