Test Definition: 2D6Q

Cytochrome P450 2D6 Comprehensive Cascade, Varies

Overview

Useful For

Providing information relevant to tamoxifen, codeine, and tramadol, as well as other medications metabolized by cytochrome P450 2D6

Determining the exact genotype when other methods fail to generate this information or if genotype-phenotype discord is encountered clinically

Identifying precise genotype when required (eg, drug trials, research protocols)

Identifying novel variants that may interfere with drug metabolism (when reflex to sequencing is performed)

Reflex Tests

Test Id	Reporting Name	Available Separately	Always Performed
2D61Z	CYP2D6 Full Gene Sequence	No, (Bill Only)	No
2D62Z	CYP2D6 GEN CYP2D6-2D7 Hybrid	No, (Bill Only)	No
2D63Z	CYP2D6 GEN CYP2D7-2D6 Hybrid	No, (Bill Only)	No
2D64Z	CYP2D6 Nonduplicated Gene	No, (Bill Only)	No
2D65Z	CYP2D6 5' Gene DUP/MLT	No, (Bill Only)	No
2D66Z	CYP2D6 3' Gene DUP/MLT	No, (Bill Only)	No

Genetics Test Information

Testing is done in 2 tiers when needed. Tier 1 uses a polymerase chain reaction (PCR)-based 5'-nuclease assay to determine the variants present. All samples also have copy number determined by PCR-based 5'-nuclease assay. Testing in tier 1 allows for the detection of all common *CYP2D6* variants (eg, *2, *3, *4, *5, *6, *7, *8, *9, *10, *17, *29, *35, *41, *59) and rarer alleles such as *11, *12, *14, *15, and *114. Duplications and multiplications of alleles are also identified. Unitary and tandem *CYP2D7-2D6* (*13) alleles and *CYP2D6-2D7* (eg, *4N, *36, and *68) alleles can also be detected. Tier 2 testing involves sequencing using fluorescent dye-terminator chemistry and is only done if tier 1 testing results are ambiguous. Approximately 3% of samples require tier 2 testing.

Testing Algorithm

Tier 2 testing will be performed only if an ambiguous phenotype is identified by tier 1 testing. The number of sequencing tests needed to determine the phenotype will vary depending on the tier 1 result.

For more information see *CYP2D6* Comprehensive Cascade Testing Algorithm.

Special Instructions

- Informed Consent for Genetic Testing
- CYP2D6 Comprehensive Cascade Testing Algorithm
- Pharmacogenomic Association Tables
- Multiple Genotype Test List
- Informed Consent for Genetic Testing (Spanish)

Method Name

Tier 1: Real Time Polymerase Chain Reaction (PCR)

Tier 2: Polymerase Chain Reaction (PCR) followed by DNA Sequence Analysis

NY State Available

Yes

Specimen

Specimen Type

Varies

Ordering Guidance

This test is not for use in assessing for autoimmune hepatitis. Autoantibodies for the CYP2D6 enzyme are found in many cases of autoimmune hepatitis; order LKM / Liver/Kidney Microsome Type 1 Antibodies, Serum for autoimmune hepatitis assessment.

Testing is available as the single gene assay (this test) and as a part of a psychotropic or focused pharmacogenomics panel.

If multiple pharmacogenomic genotype testing is desired, order PGXQP / Focused Pharmacogenomics Panel, Varies.

If genotype testing for psychotropic medications is desired, order PSYQP / Psychotropic Pharmacogenomics Gene Panel, Varies.

Specimen Required

Multiple genotype tests can be performed on a single specimen after a single extraction. See Multiple Genotype Test List for a list of tests that can be ordered together.

Submit only 1 of the following specimens:

Specimen Type: Whole blood
Container/Tube: Lavender top (EDTA)
Specimen Volume: 3 mL
Collection Instructions:

1. Invert several times to mix blood.
2. Send whole blood specimen in original tube. **Do not aliquot.**
Specimen Stability Information: Ambient (preferred) 9 days/Refrigerated 30 days

Specimen Type: Saliva
Patient Preparation: Patient should not eat, drink, smoke, or chew gum 30 minutes prior to collection.
Supplies:
DNA Saliva Kit High Yield (T1007)
Saliva Swab Collection Kit (T786)
Container/Tube:
Preferred: High-yield DNA saliva kit
Acceptable: Saliva swab
Specimen Volume: 1 Tube if using T1007 or 2 swabs if using T786
Collection Instructions: Collect and send specimen per kit instructions.
Specimen Stability Information: Ambient (preferred) 30 days/Refrigerated 30 days
Additional Information: Saliva specimens are acceptable but not recommended. Due to lower quantity/quality of DNA yielded from saliva, some aspects of the test may not perform as well as DNA extracted from a whole blood sample. When applicable, specific gene regions that were unable to be interrogated will be noted in the report. Alternatively, additional specimen may be required to complete testing.

Specimen Type: Extracted DNA
Container/Tube: 2 mL screw top tube
Specimen Volume: 100 mcL (microliters)
Collection Instructions:
1. The preferred volume is 100 mcL at a concentration of 75 ng/mcL.
2. Include concentration and volume on tube.
Specimen Stability Information: Frozen (preferred) 1 year/Ambient/Refrigerated

Forms

1. **New York Clients-Informed consent is required.** Document on the request form or electronic order that a copy is on file. The following documents are available:
-Informed Consent for Genetic Testing (T576)
-Informed Consent for Genetic Testing-Spanish (T826)
2. If not ordering electronically, complete, print, and send 1 of the following forms with the specimen:
-Cardiovascular Test Request (T724)
-Neurology Specialty Testing Client Test Request (T732)
-Therapeutics Test Request (T831)

Specimen Minimum Volume

Whole blood: 1 mL
Saliva: 1 swab

Reject Due To

All specimens will be evaluated at Mayo Clinic Laboratories for test suitability

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Varies		

Clinical & Interpretive

Clinical Information

The cytochrome P450 (CYP) family of enzymes is a group of oxidative/dealkylating enzymes localized in the microsomes of many tissues including the intestines and liver. One of the CYP enzymes, CYP2D6, is wholly or partially responsible for the metabolism of many commonly prescribed drugs.

The *CYP2D6* gene is highly variable with over 100 named alleles. The gene may be deleted, duplicated, and multiplied and can have multiple sequence variations. In addition, some individuals have genes that are hybrids of *CYP2D6* and the *CYP2D7* pseudogene. Some individuals have *CYP2D6* variants that result in synthesis of an enzyme with decreased or absent catalytic activity. These individuals may process CYP2D6-metabolized medications more slowly. *CYP2D6* duplications and multiplications involving active alleles may result in ultrarapid metabolism of CYP2D6-metabolized drugs. *CYP2D6* genotype results are used to predict metabolizer phenotypes.(See Table 1)

Table 1. Enzyme Activity of Individual Star Alleles

Enzyme activity	Examples of *CYP2D6* star alleles
Normal (extensive) metabolism	**1, *2, *35*
Decreased activity	**9, *10, *14, *17, *29,* and **41, *59*
No or null activity	**3, *4, *4N, *5, *6, *7, *8, *11, *12, *13, *15, *36, *68, *114*

CYP2D6 phenotype is predicted based upon the number of functional, partially functional, and nonfunctional alleles present in a sample.

Phenotyping is derived from the Pharmacogene Variation Consortium website (1), the Clinical Pharmacogenetics Implementation Consortium website (2), published guidelines (3-8), and an exhaustive review of the CYP2D6 literature (9-10).

There are instances where a precise phenotype prediction is not possible, and in these instances, a range of possible phenotypes will be given. Individuals without a detectable gene alteration will have the predicted phenotype of an extensive drug metabolizer and are designated as *CYP2D6*1/*1*.

Drugs that are metabolized through CYP2D6 may require dosage adjustment based on the individual patient's genotype. Patients who are poor metabolizers may require lower than usual doses to achieve optimal response in the case of drugs that are inactivated by the CYP2D6 enzyme and higher than usual doses in the case of drugs that are activated by CYP2D6 enzyme. Alternatively, patients who are ultrarapid metabolizers may benefit from increased doses in the case of

drugs that are inactivated by CYP2D6 enzyme and lower doses in the case of drugs that are activated by CYP2D6. In the absence of clear guidance from the U.S. Food and Drug Administration on dosing for various metabolizer phenotypes, patients with either ultrarapid or poor metabolism may benefit by switching to comparable alternate medications not primarily metabolized by CYP2D6 or by therapeutic drug monitoring where applicable.

Overall, this test provides a comprehensive *CYP2D6* genotype result for patients, ensuring a more accurate phenotype prediction. This assay has clinical significance for patients taking or considering medications activated (eg, codeine, tramadol, and tamoxifen) or inactivated (eg, antidepressants and antipsychotics) by the CYP2D6 enzyme.

Sequential tier testing associated with this test will be initiated until the least ambiguous phenotype possible is determined.

Reference Values

A comprehensive interpretive report will be provided.

Interpretation

A comprehensive interpretive report will be provided, which combines the results of all tier testing utilized to obtain the final genotype.

The genotype, with associated star alleles, is assigned using standard allelic nomenclature as published by the Pharmacogene Variation (PharmVar) Consortium.(1)

For the *CYP2D6* copy number variation assay, the reportable copy number range is 0 to 4 copies for each of the *CYP2D6* region assessed.

Novel variants will be classified based on known, predicted, or possible effect on gene function and reported with interpretive comments detailing their potential or known significance.

For additional information regarding pharmacogenomic genes and their associated drugs, see Pharmacogenomic Associations Tables. This resource also includes information regarding enzyme inhibitors and inducers, as well as potential alternate drug choices.

Cautions

Rare variants may be present that could lead to false-negative or false-positive results. If results obtained do not match the clinical findings (phenotype), additional testing should be considered.

Specimens may contain donor DNA if obtained from patients who received non-leukoreduced blood transfusions or allogeneic hematopoietic stem cell transplantation. Results from specimens obtained under these circumstances may not accurately reflect the recipient's genotype. For individuals who have received blood transfusions, the genotype usually reverts to that of the recipient within 6 weeks. For individuals who have received allogeneic hematopoietic stem cell transplantation, a pretransplant DNA specimen is recommended for testing.

CYP2D6 genetic test results in patients who have undergone liver transplantation may not accurately reflect the patient's CYP2D6 status.

This method may not detect all *CYP2D6* variants that result in altered CYP2D6 activity. Therefore, absence of a detectable variant does not rule out the possibility that a patient has altered CYP2D6 metabolism due to other *CYP2D6* variants that cannot be detected with this method. Furthermore, when 2 or more variants are identified, the cis-/trans-status (whether the variants are on the same or opposite chromosomes) is not always known.

A complicating factor in correlating *CYP2D6* genotype with phenotype is that many drugs or their metabolites are inhibitors of CYP2D6 catalytic activity. Selective-serotonin reuptake inhibitors (SSRI), as well as some tricyclic antidepressants (TCA) and other drugs, may reduce CYP2D6 catalytic activity. Patients in all metabolizer categories, except poor metabolizer, may have CYP2D6 enzyme activity inhibited by a variety of medications or their metabolites. Consequently, an individual may require a lower medication dose than predicted by genotyping alone. It is important to interpret the results of testing in the context of other coadministered drugs.

CYP2D6 alleles with decreased function may metabolize different drugs at different rates, ranging from normal to poor, but the literature on this is incomplete at this time.

This test is not designed to provide specific dosing or drug selection recommendations and is to be used as an aid to clinical decision making only. Drug-label guidance should be used when dosing patients with medications regardless of the predicted phenotype.

Clinical Reference

1. PharmVar: Pharmacogene Variation Consortium. Updated November 5, 2024. Accessed November 14, 2024. Available at www.pharmvar.org/
2. Clinical Pharmacogenetics Implementation Consortium (CPIC). Accessed November 14, 2024. https://cpicpgx.org/
3. Brown JT, Bishop JR, Sangkuhl K, et al. Clinical pharmacogenetics implementation consortium guideline for cytochrome P450 (CYP)2D6 genotype and atomoxetine therapy. Clin Pharmacol Ther. 2019;106(1):94-102. doi:10.1002/cpt.1409
4. Crews KR, Gaedigk A, Dunnenberger HM, et al. Clinical pharmacogenetics implementation consortium guidelines for cytochrome P450 2D6 genotype and codeine therapy: 2014 update. Clin Pharmacol Ther. 2014;95(4):376-382. doi:10.1038/clpt.2013.254
5. Bell GC, Caudle KE, Whirl-Carrillo M, et al. Clinical pharmacogenetics implementation consortium (CPIC) guideline for CYP2D6 genotype and use of ondansetron and tropisetron. Clin Pharmacol Ther. 2017;102(2):213-218. doi:10.1002/cpt.598
6. Goetz MP, Sangkuhl K, Guchelaar HJ, et al. Clinical pharmacogenetics implementation consortium (CPIC) guideline for CYP2D6 and tamoxifen therapy. Clin Pharmacol Ther. 2018;103(5):770-777. doi:10.1002/cpt.1007
7. Hicks JK, Bishop JR, Sangkuhl K, et al. Clinical pharmacogenetics implementation consortium (CPIC) guideline for CYP2D6 and CYP2C19 genotypes and dosing of selective serotonin reuptake inhibitors. Clin Pharmacol Ther. 2015;98(2):127-134. doi:10.1002/cpt.147
8. Hicks JK, Sangkuhl K, Swen JJ, et al. Clinical pharmacogenetics implementation consortium guideline (CPIC) for CYP2D6 and CYP2C19 genotypes and dosing of tricyclic antidepressants: 2016 update. Clin Pharmacol Ther. 2017;102(1):37-44. doi:10.1002/cpt.597
9. Black JL 3rd, Walker DL, O'Kane DJ, Harmandayan M. Frequency of undetected CYP2D6 hybrid genes in clinical samples: impact on phenotype prediction [published correction appears in Drug Metab Dispos. 2012 Jun;40(6):1238]. Drug Metab Dispos. 2012;40(1):111-119. doi:10.1124/dmd.111.040832
10. Kirchheiner J, Nickchen K, Bauer M, et al. Pharmacogenetics of antidepressants and antipsychotics: the contribution

of allelic variations to the phenotype of drug response. Mol Psychiatry. 2004;9(5):442-473. doi:10.1038/sj.mp.4001494

11. Crews KR, Monte AA, Huddart R, et al: Clinical pharmacogenetics implementation consortium guideline for CYP2D6, OPRM1, and COMT genotypes and select opioid therapy. Clin Pharmacol Ther. 2021 Jan 2. doi:10.1002/cpt.2149

Performance

Method Description

Genomic DNA is extracted from whole blood or saliva.

Genotype Assay (Tier 1):
Genotyping is performed using a polymerase chain reaction (PCR)-based 5'-nuclease assay. Fluorescently labeled detection probes anneal to the target DNA. PCR is used to amplify the section of DNA that contains the variant. If the detection probe is an exact match to the target DNA, the 5'-nuclease polymerase degrades the probe, the reporter dye is released from the effects of the quencher dye, and a fluorescent signal is detected. Genotypes are assigned based on the allele-specific fluorescent signals that are detected.(Unpublished Mayo method)

Copy Number Assay (Tier 1):
This assay utilizes a duplex real-time PCR, which includes 1 copy number probe and a reference assay per reaction. Each copy number probe detects the genomic sequence of interest and the reference assay detects a sequence that is known to be present in 2 copies in a diploid genome. Relative quantitation is used to determine the relative copy number of the target of interest in a genomic DNA (gDNA) sample normalized to 10 ng/mcL for each probe. Each probe is normalized to the known copy number of the reference sequence, and compared to a calibrator sample with known copies of the target sequence included with each run.(Package insert: Taqman Copy Number Assays. Applied Biosystems; Revision D, 02/2019)

Sequencing Assays (Tier 2):
The *CYP2D6* allele of interest is amplified by PCR. The PCR product is then purified and sequenced in both directions using fluorescent dye-terminator chemistry. Sequencing products are separated on an automated sequencer and trace files analyzed for variations in the exons and intron/exon boundaries of all 9 exons using mutation detection software and visual inspection.(Unpublished Mayo method)

PDF Report

No

Day(s) Performed

Monday through Thursday

Report Available

3 to 16 days

Specimen Retention Time

Whole blood/Saliva swab: 2 weeks; Extracted DNA: 2 months

Performing Laboratory Location

Mayo Clinic Laboratories - Rochester Main Campus

Fees & Codes

Fees

- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

Test Classification

This test was developed and its performance characteristics determined by Mayo Clinic in a manner consistent with CLIA requirements. It has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information

0070U
0071U (if appropriate)
0076U (if appropriate)

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
2D6Q	CYP2D6 Genotype Cascade, V	47403-1

Result ID	Test Result Name	Result LOINC® Value
610103	CYP2D6 Genotype	40425-1
610104	CYP2D6 Phenotype	79715-9
610569	CYP2D6 Activity Score	104669-7
610105	Interpretation	69047-9
610106	Additional Information	48767-8
610107	Method	85069-3
610108	Disclaimer	62364-5
610109	Reviewed by	18771-6