

## Overview

### Useful For

Genetic confirmation of hereditary von Willebrand disease with the identification of alterations in the *VWF* gene known or suspected to cause the condition

Testing for close family members of an individual with a von Willebrand disease diagnosis

### Genetics Test Information

This test detects pathogenic alterations within the *VWF* gene to delineate the underlying molecular defect in a patient with a laboratory diagnosis of von Willebrand disease (VWD), a bleeding disorder of variable severity.

The gene target for this test includes the following:

Gene name (transcript): *VWF* (GRCh37 (hg19) NM\_000552)

Chromosomal location: 12p13.31

### Reflex Tests

Test ID	Reporting Name	Available Separately	Always Performed
CULAF	Amniotic Fluid Culture/Genetic Test	Yes	No
CULFB	Fibroblast Culture for Genetic Test	Yes	No
MATCC	Maternal Cell Contamination, B	Yes	No

### Testing Algorithm

A clinical and laboratory testing algorithm for von Willebrand disease (VWD) has been developed by the National Heart, Lung, and Blood Institute of the National Institutes of Health that is freely available at <https://www.nhlbi.nih.gov/health-pro/guidelines/current/von-willebrand-guidelines>.

The laboratory workup for VWD is complex and requires initial coagulation screening (including a CBC, platelet count, partial thromboplastin time (PTT), prothrombin time (PT), and fibrinogen or thrombin time) should be performed prior to any consideration of genetic testing. Genetic testing should not be performed until a definitive diagnosis of VWD has been made.

Prenatal genetic testing is **not** performed without the prior identification of familial *VWF* alterations.

For any cord blood or prenatal specimen that is received, maternal cell contamination studies will be added. A maternal whole blood sample is required to perform this test.

If amniotic fluid is received, amniotic fluid culture will be added and charged separately. If chorionic villus specimen is received, fibroblast culture will be added and charged separately.

### Special Instructions

- [Informed Consent for Genetic Testing](#)
- [Informed Consent for Genetic Testing \(Spanish\)](#)
- [von Willebrand Disease Patient Information](#)

**Method Name**

Custom Sequence Capture and Targeted Next-Generation Sequencing (NGS) followed by Polymerase Chain Reaction (PCR) and Sanger Sequencing when appropriate

**NY State Available**

Yes

**Specimen****Specimen Type**

Varies

**Ordering Guidance**

Genetic testing should only be considered if clinical and family history, initial coagulation screens, initial von Willebrand factor (VWF) tests (antigen, ristocetin cofactor, and factor VIII coagulant activity), and specialized nongenetic VWF studies indicate a diagnosis of VWD.

**Additional Testing Requirements**

**Due to the complexity of testing non-peripheral blood, consultation with the laboratory is required for all cord blood samples.** All cord blood specimens **must be accompanied** by a maternal blood specimen. Order this test on the cord blood specimen (only 1 sample tube required) and order MATCC / Maternal Cell Contamination, Molecular Analysis, Blood on the maternal specimen.

**Shipping Instructions**

**Prenatal Specimens:** Advise Express Mail or equivalent if not on courier service. Prenatal specimens can be sent Monday through Thursday and **must be received by 3 p.m. CST on Friday** in order to be processed appropriately.

**Blood:** Ambient and refrigerate specimens must arrive within 7 days and frozen specimens must arrive within 14 days .

Collect and package specimen as close to shipping time as possible.

**Necessary Information**

[von Willebrand Disease Patient Information](#) is required, see Special Instructions. Testing may proceed without the patient information, however, the information aids in providing a more thorough interpretation. Ordering providers are strongly encouraged to fill out the form and send with the specimen.

**Specimen Required**

Results will be reported and also telephoned or faxed, if requested.

**Submit only 1 of the following specimens:**

**Specimen Type:** Peripheral blood or cord blood

**Container/Tube:**

**Preferred:** Lavender top (EDTA)

**Acceptable:** Yellow top (ACD) or light-blue top (sodium citrate)

**Specimen Volume:** 3 mL

**Collection Instructions:**

1. Invert several times to mix blood.
2. Send specimen in original tube.

**Specimen Stability Information:** Ambient (preferred) 7 days/Refrigerated 7 days/Frozen 14 days

**Due to the complexity of prenatal testing, consultation with the laboratory is required for all prenatal testing.**

**Specimen Type:** Amniotic fluid

**Supplies:** Refrigerate/Ambient Mailer, 5 lb (T329)

**Container/Tube:** Amniotic fluid container

**Specimen Volume:** 10-20 mL

**Collection Instructions:**

1. Optimal timing for specimen collection is during 14 to 18 weeks of gestation, but specimens collected at other weeks of gestation are also accepted.
2. Discard the first 2 mL of amniotic fluid.
3. Place the tubes in a Styrofoam container.
4. Fill remaining space with packing material.
5. Unavoidably, about 1% to 2% of mailed-in specimens are not viable.
6. Bloody specimens are undesirable.
7. If the specimen does not grow in culture, you will be notified within 7 days of receipt.

**Additional Information:**

A separate culture charge will be assessed under CULAF / Amniotic Fluid Culture for Genetic Testing

**Specimen Stability Information:** Refrigerated (preferred) <24 hours/Ambient <24 hours

**Specimen Type:** Chorionic villi

**Supplies:** CVS Media (RPMI) and Small Dish (T095)

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**Container/Tube:** 15-mL tube containing 15 mL of transport media

**Specimen Volume:** 20-30 mg

**Collection Instructions:**

1. Collect specimen by the transabdominal or transcervical method.
2. Transfer the chorionic villi specimen to a Petri dish containing transport medium.
3. Using a stereomicroscope and sterile forceps, assess the quality and quantity of the villi and remove any blood clots and maternal decidua.

**Additional Information:**

A separate culture charge will be assessed under CULFB / Fibroblast Culture for Genetic Testing

**Specimen Stability Information:** Refrigerated (preferred) <24 hours/Ambient <24 hours

**Specimen Type:** Confluent cultured cells

**Container/Tube:** T-25 flask

**Specimen Volume:** 2 Flasks approximately 90% confluent

**Collection Instructions:** Submit confluent cultured cells from another laboratory

**Additional Information:** There will be no culture charge.

**Specimen Stability Information:** Ambient (preferred) <24 hours/Refrigerated <24 hours

**Forms**

1. [von Willebrand Disease Patient Information \(T825\)](#) is required, see Special Instructions.
2. **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 in Special Instructions:

-[Informed Consent for Genetic Testing](#) (T576)

-[Informed Consent for Genetic Testing-Spanish](#) (T826)

3. If not ordering electronically, complete, print, and send a [Coagulation Test Request](#) (T753) with the specimen.

**Specimen Minimum Volume**

Blood: 1 mL

Amniotic fluid: 10 mL

Chorionic villi: 10 mg

Confluent cultured cells: 2 full flasks

**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 and Interpretive**

**Clinical Information**

von Willebrand disease (VWD) is a bleeding diathesis that usually involves mucous membranes and skin sites. It is typically of mild to moderate severity, although life-threatening bleeding in the central nervous system or gastrointestinal (GI) tract can occur. The most common presenting symptoms in individuals affected by VWD include epistaxis, menorrhagia, bleeding after dental extraction, postoperative bleeding, ecchymoses, bleeding from minor cuts or abrasions, gingival bleeding, and hemarthrosis.(1)

VWD affects up to 1% of the general population. While VWD occurs with equal frequency among men and women, symptoms in women are more obvious because of increased bleeding during menstrual periods, pregnancy, and after childbirth.

VWD is a result of defects in the concentration, structure, or function of von Willebrand factor (VWF), leading to decreased factor VIII (FVIII) in circulation and/or impaired platelet adhesion and aggregation at the site of vascular injury. The *VWF* gene encodes for VWF, a protein that protects blood clotting FVIII from degradation in circulation and promotes platelet adhesion and aggregation at the site of vascular injury. In circulation, VWF assembles into linear strings called multimers, the size of which is biologically important, larger multimers being more reactive than smaller multimers.

In general, bleeding risk is typically proportional to severity of VWF deficiency or the degree to which it impairs VWF function. However, bleeding risk is highly variable in von Willebrand due to the complexity of the protein structure and function and the great heterogeneity in alteration types that cause this disease. Further, because VWD is an autosomal gene, biallelic combinations of different sequence variants also contribute to phenotypic variability.

VWD is classified into 3 types:

Type 1- Partial quantitative deficiency of VWF

Type 2- Qualitative VWF defects including

- Type 2A- Decreased VWF-dependent platelet adhesion and a selective deficiency of high-molecular-weight VWF multimers,

- Type 2B- Increased affinity for platelet glycoprotein Ib

- Type 2M- Decreased VWF-dependent platelet adhesion without a selective deficiency of high-molecular-weight VWF multimers

- Type 2N- Markedly decreased binding affinity for factor VIII

Type 3- Virtually complete deficiency of VWF

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VWD type 1 and type 2B are inherited in an autosomal dominant manner. VWD type 2A and 2M have been observed to be inherited in both autosomal dominant or autosomal recessive manners. VWD type 2N and 3 are inherited in an autosomal recessive manner.

Causes of acquired (nongenetic) VWD that should be excluded prior to genetic testing include:

- 1) Autoimmune clearance or inhibition of VWF, typically in association with lymphoproliferative diseases, monoclonal gammopathies, systemic lupus erythematosus, and some cancers
- 2) Increased shear-induced proteolysis of VWF, which can occur with cardiovascular lesions or with pulmonary hypertension
- 3) Increased binding of VWF to platelets and other cell surfaces, associated with myeloproliferative disorders
- 4) Nonimmune-related hypothyroidism
- 5) Use of valproic acid, ciprofloxacin, griseofulvin, hydroxyethyl starch, and other drugs.

### Reference Values

An interpretive report will be provided.

### Interpretation

An interpretive report will be provided.

Evaluation and categorization of variants is performed using the most recent published American College of Medical Genetics and Genomics (ACMG) recommendations as a guideline. Variants are classified based on known, predicted, or possible pathogenicity and reported with interpretive comments detailing their potential or known significance.

Consultations with the Mayo Clinic Special Coagulation Clinic, Molecular Hematopathology Laboratory, or Thrombophilia Center are available for DNA diagnosis cases. This may be especially helpful in complex cases or in situations where the diagnosis is atypical or uncertain.

### Cautions

#### [Clinical:](#)

Some individuals may have a mutation that is not identified by the methods performed. The absence of a variant, therefore, does not eliminate the possibility of von Willebrand disease (VWD). This assay does not distinguish between germline and somatic alterations, particularly with variant allele frequencies (VAF) significantly lower than 50%. Test results should be interpreted in context of clinical findings, family history, and other laboratory data. Misinterpretation of results may occur if the information provided is inaccurate or incomplete.

#### Technical Limitations:

Next-generation sequencing (NGS) may not detect all types of genetic variants. Additionally, rare alterations may be present that could lead to false negative or positive results. Therefore test results should be interpreted in the context of activity and antigen measurements, clinical findings, family history, and other laboratory data. If results do not match clinical findings, consider alternative methods for analyzing these genes, such as Sanger sequencing or large deletion/duplication analysis. Misinterpretation of results may occur if the information provided is inaccurate or incomplete.

If multiple alterations are identified, NGS is not able to distinguish between alterations that are found in the same

allele ("in cis") and alterations found on different alleles ("in trans"). This limitation may complicate diagnosis or classification and has implications for inheritance and genetic counseling. To resolve these cases, molecular results must be correlated with clinical history, activity and antigen measurements, and family studies.

Unless reported or predicted to cause disease, alterations found deep in the intron or alterations that do not result in an amino acid substitution are not reported. These and common polymorphisms identified for this patient are available upon request.

#### Reclassification of Variants Policy:

At this time, it is not standard practice for the laboratory to systematically review likely pathogenic variants or variants of uncertain significance that are detected and reported. The laboratory encourages health care providers to contact the laboratory at any time to learn how the status of a particular variant may have changed over time.

Contact the laboratory if additional information is required regarding the transcript and/or human genome assembly used for the analysis of this patient's results.

### Clinical Reference

1. Nichols WL, Hultin MB, James AH, et al: von Willebrand disease (VWD): evidence-based diagnosis and management guidelines, the National Heart, Lung, and Blood Institute (NHLBI) Expert Panel report (USA). *Haemophilia*. 2008;14(2):171-232

2. Goodeve AC: The genetic basis of von Willebrand disease. *Blood Rev* 2010;24(3):123-134

3. Springer TA: von Willebrand factor, Jedi knight of the bloodstream. *Blood* 2014;124(9):1412-1425

### Performance

#### Method Description

Next-generation sequencing and/or Sanger sequencing are performed.

Regions of homology, high guanine-cytosine (GC)-rich content, and repetitive sequences may not provide accurate sequence. Therefore, all reported alterations detected by next-generation sequencing in these regions are confirmed by an independent reference method. However, this does not rule out the possibility of a false-negative result in these regions.

Sanger sequencing is used to confirm alterations detected by next-generation sequencing when appropriate. (Unpublished Mayo method)

#### PDF Report

No

#### Day(s) Performed

Varies

#### Report Available

21 to 28 days

#### Specimen Retention Time

Whole Blood: 2 weeks; DNA: Indefinitely

**Performing Laboratory Location**

Rochester

**Fees and 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 Regional Manager. 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. This test has not been cleared or approved by the US Food and Drug Administration.

**CPT Code Information**

81408

**LOINC® Information**

Test ID	Test Order Name	Order LOINC Value
VWFNG	VWF Gene, Full Gene NGS	94219-3

Result ID	Test Result Name	Result LOINC Value
113117	VWFNG Result	50397-9
113111	Alterations Detected	82939-0
113109	Interpretation	69047-9
113112	Additional Information	48767-8
113113	Method	85069-3
113114	Disclaimer	62364-5
113115	Panel Gene List	40970-6
113116	Reviewed By	18771-6