Sonora Quest Laboratories™
A Subsidiary of Laboratory Sciences of Arizona

Table 2. Approximate CF Carrier Risk Based on a Negative CFvantage Screen and Ethnicity [a,b]

Racial or Ethnic Group	Detection Rate, %	Prior Risk	Approximate Risk After Negative CFvantage Screen
Ashkenazi Jewish	95	1/24	1/461
Non-Hispanic Caucasian	90	1/25	1/241
Hispanic American	88	1/46	1/376
African American	78	1/65	1/292
Asian American	53	1/94	1/199

[a] Detection rates and residual risk estimates are based on a subset of 78 mutations detectable by the panel,[6-14] including the 23 ACMG/ACOG recommended mutations; exact data are currently unavailable for all mutations in the CFvantage Cystic Fibrosis Expanded Screen.
[b] Risks are based on the assumption that there is no family history of CF.

Risk Calculation for a CF-affected Fetus

A couple's risk of having a CF-affected fetus is the same for each pregnancy, regardless of the outcomes of prior pregnancies. For assistance calculating this risk, contact Genomics Client Services at 866.GENE.INFO (866.436.3463), a service available through one of our parent companies, Quest Diagnostics.

References

1. American College of Medical Genetics. Technical standards and guidelines for CFTR mutation testing. 2006 ed. http://www.acmg.net/Pages/ACMG_Activities/stds-2002/cf.htm 2006. Accessed August 11, 2016.
2. Committee on Genetics, American College of Obstetricians and Gynecologists. ACOG Committee Opinion. Number 486, April 2011. Update on carrier screening for cystic fibrosis. *Obstet Gynecol.* 2011;117:1028-1031.
3. Cystic Fibrosis Foundation Patient Registry. 2014 Annual Data Report.https://www.cff.org/Our-Research/CF-Patient-Registry/CF-Patient-Registry-Reports/. Accessed August 11, 2016.
4. Cystic Fibrosis Mutation Database. Toronto, Ontario, Canada: Cystic Fibrosis Centre at the Hospital for Sick Children.http://www.genet.sickkids.on.ca/Home.htmlUpdated April 25, 2011. Accessed August 11, 2016.
5. SosnayPR, Siklosi KR,Van GoorF, et al. Defining the disease liability of variant in the cystic fibrosis transmembrane conductance regulator gene. *Nat Genet*. 2013;45:1160-1167.
6. Committee on Genetics, American College of Obstetricians and Gynecologists. ACOG Committee Opinion. Number 325, December 2005. Update on carrier screening for cystic fibrosis. *Obstet Gynecol.* 2005;106:1465-1468.
7. Watson MS, Cutting GR, Desnick RJ, et al. Cystic fibrosis population carrier screening: 2004 revision of American College of Medical Genetics mutation panel. *Genet Med.* 2004;6:387-391.
8. Heim RA, Sugarmann EA, Allitto BA. Improved detection of cystic fibrosis mutations in the hetergeneous U.S. popultion using an expanded, pan-ethnic mutation panel. *Genet Med*. 2001;3:168-176.
9. Sugarmann EA, Rohlfs EM, Silverman LM, et al. *CFTR* mutation distribution among U.S. Hispanic and African American individuals: evaluation in cystic fibrosis patient and carrier screening populations. *Genet Med*. 2004;6:392-399.
10. Schrijver I, Ramalingam S, Sankaran R, et al. Diagnostic testing by *CFTR* gene mutation analysis in a large group of Hispanics: novel mutations and assessment of a population-specific mutation spectrum. *J Mol Diagn*. 2005;7:289-299.
11. Shoshani T, Augarten A, Yahav J, et al. Two novel mutations in the *CFTR* gene: W1089X in exon 17B and 4010delTATT in exon 21. *Hum Mol Genet*. 1994;3:657-658.
12. Friedman KJ, Leigh MW, Czarnecki P, et al. Cystic fibrosis transmembrane-conductance regulator mutations among African Americans. *Am J Hum Genet.* 1998;62:195-196.
13. Macek M Jr, Mackova A, Hamosh A, et al. Identification of common cystic fibrosis mutations in African-Americans with cystic fibrosis increases the detection rate to 75%. *Am J Hum Genet.* 1997;60:1122-1127.
14. Wong LJ, Wang J, Zhang YH, et al. Improved detection of *CFTR* mutations in Southern California Hispanic CF patients. *Hum Mutat.* 2001;18:296-307.

* The CPT code provided is based on AMA guidelines and is for informational purposes only. CPT coding is the sole responsibility of the billing party. Please direct any questions regarding coding to the payer being billed.

This test was developed and its analytical performance characteristics have been determined by Quest Diagnostics Nichols Institute. It has not been cleared or approved by FDA. This assay has been validated pursuant to the CLIA regulations and is used for clinical purposes.

Test Summary

Sonora Quest Laboratories™
A Subsidiary of Laboratory Sciences of Arizona

CFvantage® Cystic Fibrosis Expanded Screen

Test Code: 906672

Specimen Requirements: 5 mL room-temperature whole blood (EDTA, lavender-top tube): 3 mL minimum

CPT Code*: 81220

CLINICAL USE

- Detect cystic fibrosis (CF) carriers
- Determine a couple's risk of having a child with CF
- Identify familial mutations in affected individuals
- Diagnose CF postnatally

CLINICAL BACKGROUND

CF is one of the most common autosomal recessive diseases affecting Caucasians, with an incidence of approximately 1 in 3,000 births and a carrier rate of 1 in 25 in this population.[1,2] It also occurs in other ethnic groups at a lower frequency. The disorder may be characterized by pulmonary disease, pancreatic insufficiency, liver disease, and congenital absence of the vas deferens (CAVD), leading to male infertility.[1] Median predicted survival for CF patients is approximately 39 years,[3] with lung damage causing the majority of deaths.[2,3] A diagnosis of CF is confirmed by a positive sweat chloride test and/or detection of a CF-associated mutation on both chromosomes.[1]

CF is caused by mutations in th cystic fibrosis transmembrane conductance regulator (*CFTR*) gene, located on the long arm of chromosome 7 (7q31.2).[1] Mutations result in a defective CFTR protein that, in turn, results in defective cellular chloride transport. Over 1,900 mutations, most of which occur at frequencies of <0.1%, have been identified.[4] One mutation, F508del (delta F508), accounts for approximately 70% of all *CFTR* mutations in many, but not all, ethnic groups.[1,4] In 2014, 86% of patients in the Cystic Fibrosis Foundation patient registry were reported to have at least 1 copy of F508del, and 46% had 2 copies.[3]

The American College of Medical Genetics (ACMG) and the American Congress of Obstetricians and Gynecologists (ACOG) recommend screening for 23 common *CFTR* mutations that were chosen primarily based on their frequency in Ashkenazi Jewish and non-Hispanic Caucasian populations. CF is more common in these ethnic groups, and the 23 mutations account for 94% of mutant alleles in Ashkenazi Jews and 88% in non-Hispanic Caucasians.[1,2] Although the 23 mutations account for a lower percentage of mutant alleles in other ethnicities (eg, <50% in Asians), the ACMG and ACOG consider the panel of mutations pan-ethnic and recommend offering screening to all patients.[1,2]

The growing proportion of mixed-ethnicity individuals and the increasing ethnic diversity of the population are cause for larger panels with greater detection rates. In addition to the 23 ACMG/ACOG-recommended mutations, the panel includes mutations that met 2 important criteria in a recent study: 1) association with a clinical diagnosis of CF (patient sweat chloride concentration ≥60 mM), and 2) allele frequency ≥0.01% in North America and Europe.[5] Compared to the ACMG/ACOG panel, the CFvantage Cystic Fibrosis Expanded Screen detects a higher percentage of CF-causing mutations across ethnicities.[6-14]

INDIVIDUALS SUITABLE FOR TESTING

- Individuals with a family history of CF or *CFTR* mutations
- Symptomatic children and adults
- Males with CAVD
- Patients with chronic or idiopathic pancreatitis
- Previously unscreened, reproductively-active individuals or couples

METHOD

- Sequence-specific multiplex PCR followed by next-generation sequencing analysis with proprietary bioinformatics
- Detects mutations shown in **Table 1**
- Report form specifies mutations screened, mutations identified, and interpretive information

INTERPRETIVE INFORMATION

The following information will help with interpretation of test results.To discuss a patient's relevant family history or receive additional assistance, please contact Genomics Client Services at 866.GENE.INFO (866.436.3463), a service available through one of our parent companies, Quest Diagnostics.

Sonora Quest Laboratories™
A Subsidiary of Laboratory Sciences of Arizona

Table 1. CF Mutations Detected

Conventional Name (HGVS cDNA Nomenclature)	Conventional Name (HGVS cDNA Nomenclature)	Conventional Name (HGVS cDNA Nomenclature)
296+2T>A (c.164+2T>A)	1898+5G>T (c.1766+5G>T)	A455E (c.1364C>A)[a]
394delTT (c.262_263delTT)	2043delG (c.1911delG)	A559T (c.1675G>A)
405+1G>A (c.273+1G>A)	2055del9>A (c.1923_1931del9insA)	C524X (c.1572C>A)
406-1G>A (c.274-1G>A)	2105del13ins5 (c.1973_1985del13insAGAAA)	CFTRdele2,3 (c.54-5940_273+10250del21kb)
444delA (c.313delA)	2108delA (c.1976delA)	CFTRdele22,23 (c.3964-78_4242+577del)
457TAT>G (c.325_327delTATinsG)	2143delT (c.2012delT)	D110H (c.328G>C)
574delA (c.442delA)	2183AA>G (c.2051_2052delAAinsG)	D579G (c.1736A>G)
621+1G>T (c.489+1G>T)[a]	2184delA (c.2052delA)[a]	E60X (c.178G>T)
663delT (c.531delT)	2184insA (c.2052_2053insA)	E92K (c.274G>A)
711+1G>T (c.579+1G>T)[a]	2307insA (c.2175_2176insA)	E92X (c.274G>T)
711+3A>G (c.579+3A>G)	2347delG (c.2215delG)	E585X (c.1753G>T)
711+5G>A (c.579+5G>A)	2585delT (c.2453delT)	E822X (c.2464G>T)
712-1G>T (c.580-1G>T)	2622+1G>A (c.2490+1G>A)	E831X (c.2491G>T)
852del22 (c.720_741del22)	2711delT (c.2583delT)	E1104X (c.3310G>T)
935delA (c.803delA)	2789+5G>A (c.2657+5G>A)[a]	F311del (c.933_935delCTT)
936delTA (c.805_806delAT)	2869insG (c.2737_2738insG)	F508del (c.1521_1523delCTT)[a]
1078delT (c.948delT)	3007delG (c.2875delG)	G85E (c.254G>A)[a]
1154insTC (c.1022_1023insTC)	3120+1G>A (c.2988+1G>A)[a]	G91R (c.271G>A)
1161delC (c.1029delC)	3120G>A (c.2988G>A)	G178R (c.532G>A)
1213delT (c.1081delT)	3121-1G>A (c.2989-1G>A)	G330X (c.988G>T)
1248+1G>A (c.1116+1G>A)	3171delC (c.3039delC)	G480C (c.1438G>T)
1259insA (c.1127_1128insA)	3199del6 (c.3067_3072delATAGTG)	G542X (c.1624G>T)[a]
1288insTA (c.1153_1154insAT)	3272-26A>G (c.3140-26A>G)	G551D (c.1652G>A)[a]
1341+1G>A (c.1209+1G>A)	3659delC (c.3528delC)a	G970R (c.2908G>C)
1461ins4 (c.1329_1330insAGAT)	3667del4 (c.3535_3538delACCA)	G1244E (c.3731G>A)
1525-1G>A (c.1393-1G>A)	3791delC (c.3659delC)	H199Y (c.595C>T)
1548delG (c.1418delG)	3821delT (c.3691delT)	I336K (c.1007T>A)
1609delCA (c.1477_1478delCA)	3849+10kbC>T (c.3717+12191C>T)[a]	I507del (c.1519_1521delATC)[a]
1677delTA (c.1545_1546delTA)	3876delA (c.3744delA)	I1234V (c.3700A>G)
1717-1G>A (c.1585-1G>A)[a]	3905insT (c.3773_3774insT)	K710X (c.2128A>T)
1717-8G>A (c.1585-8G>A)	4005+1G>A (c.3873+1G>A)	L206W (c.617T>G)
1811+1.6kbA>G (c.1679+1.6kbA>G)	4016insT (c.3884_3885insT)	L467P (c.1400T>C)
1812-1G>A (c.1680-1G>A)	4209TGTT>AA (c.4077_4080delTGTTinsAA)	L732X (c.2195T>G)
1898+1G>A (c.1766+1G>A)[a]	4382delA (c.4251delA)	L927P (c.2780T>C)
1898+1G>T (c.1766+1G>T)		
1898+3A>G (c.1766+3A>G)		

(*Continued*)

Table 1. CF Mutations Detected (*Continued*)

Conventional Name (HGVS cDNA Nomenclature)	Conventional Name (HGVS cDNA Nomenclature)	Conventional Name (HGVS cDNA Nomenclature)
L1065P (c.3194T>C)	R117C (c.349C>T)	S466X (c.1397C>A or c.1397C>G)
L1077P (c.3230T>C)	R117H (c.350G>A)[a,b]	S489X (c.1466C>A)
L1093P (c.3278T>C)	R334W (c.1000C>T)[a]	S492F (c.1475C>T)
M1V (c.1A>G)	R347H (c.1040G>A)	S549N (c.1646G>A)
M1101K (c.3302T>A)	R347P (c.1040G>C)[a]	S549R (c.1645A>C or c.1647T>G)
N1303K (c.3909C>G)[a]	R352Q (c.1055G>A)	S945L (c.2834C>T)
P67L (c.200C>T)	R553X (c.1657C>T)[a]	S1196X (c.3587C>G)
P205S (c.613C>T)	R560K (c.1679G>A)	S1251N (c.3752G>A)
P574H (c.1721C>A)	R560T (c.1679G>C)[a]	S1255X (c.3764C>A)
Q39X (c.115C>T)	R709X (c.2125C>T)	T338I (c.1013C>T)
Q98X (c.292C>T)	R764X (c.2290C>T)	V520F (c.1558G>T)
Q220X (c.658C>T)	R851X (c.2551C>T)	W401X (c.1202G>A or c.1203G>A)
Q493X (c.1477C>T)	R1066C (c.3196C>T)	W846X (c.2537G>A)
Q525X (c.1573C>T)	R1066H (c.3197G>A)	W1089X (c.3266G>A)
Q552X (c.1654C>T)	R1128X (c.3382A>T)	W1145X (c.3435G>A)
Q890X (c.2668C>T)	R1158X (c.3472C>T)	W1204X (c.3611G>A or c.3612G>A)
Q1238X (c.3712C>T)	R1162X (c.3484C>T)[a]	W1282X (c.3846G>A)[a]
Q1313X (c.3937C>T)	R1283M (c.3848G>T)	Y122X (c.366T>A)
R75X (c.223C>T)	S341P (c.1021T>C)	Y1092X (c.3276C>A or c.3276C>G)

HGVS, Human Genome Variation Society.
[a] ACMG/ACOG-recommended mutation.[1]
[b] The variants 5T/7T/9T are included as needed (see Interpretive Information). 5T/7T/9T may be of clinical significance in congenital absence of the vas deferens (CAVD) and other disorders.

Diagnosis

Detection of 2 mutant alleles in conjunction with positive clinical findings or family history is consistent wiht CF. Failure to detect 2 mutant alleles in a symptomatic patient, however, does not exclude a diagnosis of CF. Not all individuals with CF

Carrier Detection

The presence of a single CF mutation in an asymptomatic individual identifies that person as a carrier. Absence of a CF mutation significantly reduces, but does not eliminate, the risk of being a carrier. The residual risk of being a carrier (ie, of having a CF mutation not screened for in this assay) is influenced by the individual's ethnicity and clinical and family history. The residual risk data in **Table 2** are based on a subset of mutations detectable by the panel[6-14]; thus, detection rates may be slightly higher and the residual risks may be slightly lower than shown in the table. If clinically indicated, additional testing is available.

IVS8 5T/7T/9T Variant

- A single 5T variant with an R117H mutation on the same chromosome (in *cis*) acts as a classic CF mutation. Thus, an individual with this genotype is a CF carrier.[1] A 5T variant occurring in in *trans* (on the opposite chromosome) with an R117H mutation may result in CAVD.[1]
- A 7T or 9T variant in *cis* with an R117H mutation on the as a mild CF mutation.[1] Thus, an individual with this genotype is a CF carrier. When coupled with a classic CF mutation, male patients may have CAVD.

The status of the 5T/7T/9T variant in intron 8 is reported only when the R117H mutation is detected. If a 5T variant is identified, testing of family members id required to determine if the variant is in *cis* or *trans*.[1] Genetic counseling is recommended.