

## Sequential testing for *CCM1*, *CCM2* and *CCM3*

Cerebral cavernous malformations (CCM) are congenital vascular anomalies of the brain that can cause significant neurological disabilities, including intractable seizures and hemorrhagic stroke. Three genes that contribute to autosomal dominant CCM have been identified. Using a genomic sequence-based positional cloning strategy, *KRIT1*, encoding a protein that interacts with the Krev-1/rap1a tumor suppressor, has been identified as the first CCM gene (*CCM1/KRIT1*).

The identical *CCM1/KRIT1* mutation is common to many Mexican-American families (+/-70%). This C1363T transition in exon 10 changes a *GLN* to a premature termination codon. Identification of a common Hispanic mutation has potential clinical significance for presymptomatic diagnosis of CCM in this population.

More recently the second gene detected that can cause CCMs was identified as *MGC4607*. *CCM2/MGC4607*, similar to the *KRIT1* binding partner ICAP1 $\alpha$ , encodes a protein with a PTB (phospho-tyrosine binding) domain. This protein may be part of the complex pathway of integrin signaling, that when perturbed, causes abnormal vascular morphogenesis in the brain leading to CCM formation.

The third gene was identified in 2005. Most recently mutations within the programmed cell death 10 (*CCM3/PDCD10*) gene were shown to cause cerebral cavernous malformations in some families. *CCM3/PDCD10* is highly conserved in both vertebrates and invertebrates. Its role in CCM vascular morphogenesis is yet to be defined.

It is thought based on family studies that mutations in the *CCM1/KRIT1* gene may account for up to 40% of familial CCM cases, mutations in the *CCM2/MGC4607* gene accounting for perhaps an additional 20% of familial cases and mutations in the recently identified *CCM3/PDCD10* gene ~10-20% and perhaps other still unidentified genes accounting for the remaining 20-30% of cases. A recent publication strongly supports a fourth CCM locus where the gene has yet to be determined. Identification of these CCM associated mutations has potential clinical significance for presymptomatic diagnosis of CCM in this population and their families.

Sequential testing for these genes is done in the following order: *CCM1* is tested first (with exon 10 being done first in all patients with documented Hispanic heritage: billed at \$210.00 if positive), if *CCM1* testing finds a likely causative mutation, the testing is stopped and billed at: \$1410.00; if *CCM1* testing is negative, *CCM2* is sequenced next, if testing finds a likely causative mutation in *CCM2*, the testing is stopped and billed at \$2390.00 (\$1410 + \$980) and if both *CCM1* and *CCM2* testing are negative; *CCM3* sequencing is done and billed at the discounted price of \$2,750.00 **You are only billed for the tests actually preformed.**

### Specimen Requirements

- Collect 2-5 ml of whole blood in EDTA (purple top tube) or ACD (yellow top tube). 5 ml is the preferred volume.
- Only one blood tube is required for multiple tests.
- Ship whole blood specimens at room temperature.
- Do not freeze blood.
- During hot weather, include a frozen ice pack in the shipping container. Do not allow the ice pack to come in direct contact with the specimen tube.
- In cold weather, include an unfrozen ice pack to help moderate extremes in temperature. The DNA in whole blood is stable for at least 48 hours at 21°C, 5-7 days at 4°C.

<i>CCM</i> single exon sequencing (any gene)	\$230.00	<i>CCM1/KRIT1</i> Mutation Test DNA Seq	\$1,410.00
Molec Diag, Ascertainment 83890	\$30	83890	\$30
Molec Diag, Isolation 83891	\$30	83891	\$30
Molecular Diag, Amplif 83898 x1 @ \$60	\$50	83898 x16 @ \$50	\$800
Mutat Id By Seq, Single Seg 83904 x1 @ \$40	\$30	83904 x16 @ \$30	\$480
Molecular Diag, Separation 83894	\$40	83894	\$40
Interpretation and Report 83912	\$30	83912	\$30

<i>CCM2/MGC4607</i> Mutation Test DNA Seq	\$980.00	<i>CCM3/PDCD10</i> Mutation Test DNA Seq	\$725.00
Molec Diag, Ascertainment 83890	\$30	83890	\$30
Molec Diag, Isolation 83891	\$30	83891	\$30
Molecular Diag, Amplif 83898 x10 @ \$50	\$500	83898 x7 @ \$50	\$350
Mutat Id By Seq, Single Seg 83904 x10 @ \$35	\$350	83904 x7 @ \$35	\$245
Molecular Diag, Separation 83894	\$40	83894	\$40
Interpretation and Report 83912	\$30	83912	\$30

*CCM1*, *CCM2*, *CCM3* sequential testing: Pricing variable: \$230.00, \$1,410.00, \$2,390.00, or \$2,750.00 (discounted from \$3,115.00 if ordered separately)

Molec Diag, Ascertainment 83890	\$35
Molec Diag, Isolation 83891	\$30
Molecular Diag, Amplif 83898 x33 @ \$45	\$1485
Mutat Id By Seq, Single Seg 83904 x33 @ \$30	\$990
Molecular Diag, Separation 83894 x3	\$120
Interpretation and Report 83912 x3	\$90

**Accreditation Info. CLIA ID #: 52D1027685** (expires 1/18/07) (CAP#: 7185561, AU ID: 1407125 expires 12/20/06)

**Ship to:**

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