Lynch Syndrome/Hereditary Nonpolyposis Colorectal Cancer

Colorectal cancer (CRC) exhibits the characteristics of familial clustering in ~10-15% of cases. The most common cause of hereditary CRC is Lynch syndrome (LS), also known as hereditary nonpolyposis colorectal cancer (HNPCC). LS is caused by a germline mutation in one of the genes within the DNA mismatch repair (MMR) system.

Indications for Ordering

Microsatellite instability (MSI) and/or immunohistochemistry (IHC) tumor analyses

- Universal screening for LS in all individuals with newly diagnosed CRC[1]
- Universal screening for LS in individuals with endometrial cancer[2]

Germline MMR gene testing

- Order following abnormal screening test result
- Confirms diagnosis of LS

Disease Overview

Incidence

- Accounts for 2-4% of CRC and ~1-2% of endometrial cancers[2,3]
- Most common hereditary CRC syndrome
- Most common cause of hereditary endometrial cancer

Risk Estimates

The following lifetime risk estimates apply to individuals with *MLH1* and *MSH2* pathogenic variants; risks for variants in *MSH6* or *PMS2* may be lower[1,2]:

- CRC: 52-82%
- Endometrial: 25-60%
- Prostate: ~30%
- Ovarian: 11-24%
- Gastric: 6-13%
- Hepatobiliary tract: 1-4%
- Urinary tract: 1-7%
- Small bowel: 3-6%
- Brain/central nervous system: 1-3%
- Sebaceous neoplasms: 1-9%
- Pancreatic: 1-6% (*MLH1* and *MSH2* only)

Genetics

Genes Tested

MLH1, *MSH2*, *MSH6*, *PMS2*, and *EPCAM*

Inheritance

Tests to Consider

Screening Studies (Requires Pathological Tissue)

Mismatch Repair by Immunohistochemistry 0049302

Method: Qualitative Immunohistochemistry

- First-line screening test for newly diagnosed CRC, endometrial carcinoma, and LS
- Highly recommended prior to ordering germline MMR gene testing
  - Directs subsequent genetic diagnostic testing
- Testing for CRC and other solid tumors to qualify patients for certain immune checkpoint inhibitor treatment

Microsatellite Instability (MSI), HNPCC/Lynch Syndrome, by PCR 0051740

Method: Capillary Electrophoresis

- First-line screening test for newly diagnosed CRC, endometrial carcinoma, and LS
- Directs subsequent genetic diagnostic testing for LS
- Testing for CRC and other solid tumors to qualify patients for certain immune checkpoint inhibitor treatment

Mismatch Repair by Immunohistochemistry with Reflex to BRAF Codon 600 Mutation and MLH1 Promoter Methylation 2002327

Method: Qualitative Immunohistochemistry/Qualitative Real-time Polymerase Chain Reaction

- Screening for HNPCC/LS[1]
- If MLH1 IHC is abnormal, evaluations of *BRAF* codon 600 and, possibly, *MLH1* methylation are performed
- Definitive diagnosis of LS requires additional targeted MMR germline molecular studies
- Do not use in endometrial cancer

BRAF Codon 600 Mutation Detection with Reflex to MLH1 Promoter Methylation 0051750

Method: Polymerase Chain Reaction/Pyrosequencing

- Recommended reflex test to differentiate between LS and sporadic CRC in tumors showing loss of MLH1
- If no *BRAF* variant is detected, *MLH1*

Autosomal dominant

Test Interpretation

Microsatellite Instability (MSI), HNPCC/Lynch Syndrome, by PCR

Sensitivity/Specificity

- Clinical sensitivity: 90%[1,2]
- Analytical sensitivity/specificity: >99%

Results

- High MSI (MSI-H)
  - MMR deficiency, either sporadic or LS
- Indeterminate MSI (MSI-I)
  - Instability in even one mononucleotide repeat can be associated with LS.
  - Follow-up IHC studies are recommended.
- Microsatellite stable (MSS)
  - LS unlikely

Limitations

- 10-15% of sporadic CRCs are also MSI-H.[1,2]
- Preoperative chemoradiation of rectal cancer:
  - May complicate IHC interpretation and/or decrease tumor mass
  - May make MSI testing difficult
  - Evaluation of pretreatment biopsies will avoid this limitation
- Screens for LS only and does not evaluate other hereditary causes of CRC or endometrial cancer.

Mismatch Repair by IHC

Sensitivity/Specificity

- Clinical sensitivity: 90%[1,2]

Results

- Normal: MMR proteins are normally expressed.
  - MMR deficiency is unlikely.
  - LS unlikely
- Abnormal: MMR protein expression is abnormal.
  - Loss of expression of one or more proteins is highly predictive of MMR deficiency.
  - Absence of both MLH1 and PMS2:
    - *MLH1* germline pathogenic variant is possible.
    - Consider *MLH1* methylation ± *BRAF* V600E studies.
    - If methylation and *BRAF* studies are negative, follow with *MLH1* germline genetic testing.
  - Absence of PMS2 only:
    - *PMS2* germline pathogenic variant likely
    - Consider *PMS2* germline testing.
    - If *PMS2* testing does not identify a germline pathogenic variant, consider *MLH1* germline testing.
  - Absence of both MSH2 and MSH6:
    - *MSH2* germline pathogenic variant likely
    - Consider *MSH2* germline testing.
  - Absence of MSH6 only:
    - *MSH6* germline pathogenic variant likely

promoter methylation is evaluated

Mismatch Repair by Immunohistochemistry with Reflex to MLH1 Promoter Methylation 2005270

Method: Qualitative Immunohistochemistry/Qualitative Real-time Polymerase Chain Reaction

- Reflex screening test for LS in non-CRC tumors (eg, endometrial carcinoma)
- If *MLH1* expression is lost, *MLH1* methylation is performed

MLH1 Promoter Methylation, Paraffin 2002499

Method: Real-Time Polymerase Chain Reaction/Fluorescence Resonance Energy Transfer

Recommended test to distinguish between LS and sporadic non-CRC tumors with loss of MLH1

Diagnostic Germline Genetic Studies

- Specimen: peripheral blood
- Germline genetic testing is available for all 4 MMR genes known to cause LS, either separately or as part of the hereditary gastrointestinal (GI) cancer panel (see Related Tests)

HNPCC/Lynch Syndrome (MLH1) Sequencing and Deletion/Duplication 0051650

Method: Polymerase Chain Reaction/Sequencing/Multiplex Ligation-dependent Probe Amplification

- Detect germline *MLH1* variants
- Use in MMR-deficient carcinoma with suggestive IHC results (loss of MLH1 and PMS2 proteins), negative for the *BRAF* codon 600 pathogenic variant, and with normal *MLH1* promoter methylation studies

HNPCC/Lynch Syndrome (MSH2) Sequencing and Deletion/Duplication 0051654

Method: Polymerase Chain Reaction/Sequencing/Multiplex Ligation-dependent Probe Amplification

- Detect germline *MSH2* variants
- Use in MMR-deficient carcinoma with suggestive IHC results (loss of MSH2 and MSH6 proteins)
- Includes evaluation of *EPCAM* exon 9 deletions and 10 Mb inversion of *MSH2* exons 1-7

HNPCC/Lynch Syndrome (MSH6) Sequencing and Deletion/Duplication 0051656

Method: Polymerase Chain Reaction/Sequencing/Multiplex Ligation-dependent Probe Amplification

- Detect germline *MSH6* variants
- Use in MMR-deficient carcinoma with suggestive IHC results (isolated loss of

- Consider *MSH6* germline testing.
- If *MSH6* testing does not identify a germline pathogenic variant, consider *MSH2* germline testing.

Limitations

- ~10% of individuals with LS will have IHC tests that show normal staining of the MMR proteins.
- Because the correlation of MSI with IHC is not 100%, direct testing of MSI by PCR may be helpful.
- Screens for LS only and does not evaluate other hereditary causes of CRC or endometrial cancer.

BRAF Codon 600 Mutation Detection with Reflex to *MLH1* Promoter Methylation

Sensitivity/Specificity

- Analytical sensitivity: Methylation levels >10% are reported as positive.

Results

- *BRAF* V600E detected
  - Presence in MMR-deficient CRC indicates the tumor is probably sporadic and not associated with LS.
  - Further germline testing is not typically indicated.
- *MLH1* promoter methylation detected
  - Presence in an MSI CRC indicates the tumor is probably sporadic and not associated with LS.
  - Further germline testing is not typically indicated.
- No variants detected
  - In MSI-H tumors with loss of MLH1 protein by IHC, *MLH1* germline testing is indicated.

Germline Genetic Studies (*MLH1*, *MSH2*, *MSH6, PMS2*, or *EPCAM*)

Sensitivity/Specificity

- Proportion of LS attributed to pathogenic variants in specific MMR gene:
  - *MLH1*: 50%[4]
  - *MSH2*: 40%[4]
  - *MSH6*: 7-10%[5,6,7]
  - *PMS2*: <5%[8]
  - *EPCAM*: ~1-3%[9]
- Analytical sensitivity/specificity: 99%

Results

- Positive: one pathogenic variant detected
  - Predicted to be causative for LS
- Negative: no pathogenic variants detected
  - Diagnosis of LS unlikely, but not excluded
- Inconclusive: variant detected, but whether it is benign or pathogenic is unknown

Limitations

- Not evaluated:
  - Regulatory region and deep intronic variants
  - Sequence variants and large deletion/duplications in genes other than *MLH1*, *MSH2*, *MSH6*, and *PMS2*
  - Sequence variants in *EPCAM*
  - Large deletions/duplications in *EPCAM*, other than exon 9
  - Large gene inversions, other than the *MSH2* 10 Mb exons 1-7 inversion
  - Causes of hereditary CRC or endometrial cancer other than LS

MSH6 protein)

HNPCC/Lynch Syndrome (PMS2) Sequencing and Deletion/Duplication 0051737

Method: Polymerase Chain Reaction/Sequencing/Multiplex Ligation-dependent Probe Amplification

- Detect germline *PMS2* variants
- Use in MMR-deficient carcinoma with suggestive IHC results (isolated loss of PMS2 protein)

Familial Mutation, Targeted Sequencing 2001961

Method: Polymerase Chain Reaction/Sequencing

- Useful when a pathogenic familial variant identifiable by sequencing is known
- A copy of a relative's lab report is REQUIRED

Deletion/Duplication Analysis by MLPA 3003144

Method: Multiplex Ligation-dependent Probe Amplification

- Use to assess for large deletion/duplication previously identified in a family member
- A copy of a relative's lab report is REQUIRED

See Related Tests

- Diagnostic errors can occur due to rare sequence variations.

References

1. NCCN clinical practice guidelines in oncology, genetic/familial high-risk assessment: colorectal, Version 1.2018. National Comprehensive Cancer Network. [Updated: Jul 2018; Accessed: Jan 2019]
2. NCCN clinical practice guidelines in oncology, uterine neoplasms, Version 2.2019. National Comprehensive Cancer Network. [Updated: Dec 2018; Accessed: Dec 2019]
3. Chadwick RB, Pyatt RE, Niemann TH, et al. Hereditary and somatic DNA mismatch repair gene mutations in sporadic endometrial carcinoma. J Med Genet. 2001;38(7):461-466. PubMed
4. Smith MJ, Urquhart JE, Harkness EF, et al. The contribution of whole gene deletions and large rearrangements to the mutation spectrum in inherited tumor predisposing syndromes. Hum Mutat. 2016;37(3):250-256. PubMed
5. Miyaki M, Konishi M, Tanaka K, et al. Germline mutation of MSH6 as the cause of hereditary nonpolyposis colorectal cancer. Nat Genet. 1997;17(3):271-272. PubMed
6. Berends MJW, Wu Y, Sijmons RH, et al. Molecular and clinical characteristics of MSH6 variants: an analysis of 25 index carriers of a germline variant. Am J Hum Genet. 2002;70(1):26-37. PubMed
7. Peltomäki P. Role of DNA mismatch repair defects in the pathogenesis of human cancer. J Clin Oncol. 2003;21(6):1174-1179. PubMed
8. Senter L, Clendenning M, Sotamaa K, et al. The clinical phenotype of Lynch syndrome due to germ-line PMS2 mutations. Gastroenterology. 2008;135(2):419-428. PubMed
9. Kuiper RP, Vissers LE, Venkatachalam R, et al. Recurrence and variability of germline EPCAM deletions in Lynch syndrome. Hum Mutat. 2011;32(4):407-414. PubMed

Additional Resources

Chen JM. The 10-Mb paracentric inversion of chromosome arm 2p in activating MSH2 and causing hereditary nonpolyposis colorectal cancer: re-annotation and mutational mechanisms. Genes Chromosomes Cancer. 2008;47(6):543-545. PubMed

Kohlmann W, Gruber SB. Lynch syndrome. In: Adam MP, Ardinger HH, Pagon RA, et al, editors. GeneReviews, University of Washington; 1993-2021. [Last revision: Apr 2018; Accessed: Feb 2020]

Stoffel EM, Mangu PB, Gruber SB , et al. Hereditary colorectal cancer syndromes: American Society of Clinical Oncology Clinical Practice Guideline endorsement of the familial risk-colorectal cancer: European Society for Medical Oncology Clinical Practice Guidelines. J Clin Oncol. 2015;33(2):209-217. PubMed

Related Information

Colorectal Cancer
Lynch Syndrome - Hereditary Nonpolyposis Colorectal Cancer (HNPCC)
Lynch Syndrome (HNPCC) Testing Algorithm

Related Tests

Hereditary Gastrointestinal Cancer Panel, Sequencing and Deletion/Duplication 2013449

Method: Massively Parallel Sequencing/Exonic Oligonucleotide-based CGH Microarray/Sequencing/Multiplex Ligation-dependent Probe Amplification

ARUP Laboratories is a nonprofit enterprise of the University of Utah and its Department of Pathology. 500 Chipeta Way, Salt Lake City, UT 84108
(800) 522-2787 | (801) 583-2787 | aruplab.com | arupconsult.com
Content Review November 2018 | Last Update February 2021