Lynch Syndrome (HNPCC), Microsatellite Instability (MSI) with Immunohistochemistry (IHC)

**Test Code:** 4850

**Department:** Molecular Genetics

**Test Synonyms:**
- Hereditary Nonpolyposis Colorectal Cancer
- Mismatch repair genes and proteins
- Colorectal cancer
- MLH1, MSH2, MSH6, PMS2

**CPT Code(s):**
- 83890 x 2
- 83900
- 83901 x 12
- 83907 x 2
- 83909 x 14
- 88342 x 4

**Background:**
Microsatellites are short, tandemly repeated DNA sequences from 1-6 base pairs in length. Microsatellite markers can be used to detect a form of genetic instability called Microsatellite Instability (MSI)\(^1\,^2\). MSI is a consequence of germline or somatic inactivation of mismatch repair genes (including \(MSH2\), \(MSH6\), \(MLH1\) and \(PMS2\)). Loss or downregulation of the expression of the protein products of these genes results in failure of the DNA mismatch repair system. Due to their repetitive sequence composition, microsatellite sequences are especially vulnerable to faulty DNA mismatch repair genes, resulting in expansion or contraction of the microsatellite segment. Such changes are detectable by comparing microsatellite allele size variations between matching normal and tumor samples.

Further, loss of mismatch repair proteins can be detected by immunohistochemical staining (IHC) of tumor tissue. At OHSU, we have IHC assays for MLH1, MSH2, MSH6, and PMS2. IHC is performed in parallel with MSI PCR testing and consolidated into one report.

*All samples which are MSI high and MLH1 unstained by IHC can be reflexed to \(BRAF\) testing to aid in the determination of the genetic basis of the individual's colorectal cancer, i.e., hereditary vs. sporadic, for additional charge. (Please contact Client Services at (855) 535-1522 to add \(BRAF\) testing.)*

**Reasons for Referral:**
Lynch Syndrome MSI plus IHC is a tumor screening test for **all newly diagnosed colorectal cancer patients** to identify patients who may have an inherited form of colorectal cancer (Hereditary Nonpolyposis Colon Cancer or Lynch Syndrome). About 15% of patients having MSI tumors have Lynch Syndrome, while the remaining have a sporadic form of CRC; 95% of patients with Lynch Syndrome have MSI. Patients with MSI tumors and the absence of a specific mismatch repair gene can be tested by mutation analysis to identify the gene defect. Family members at risk for having inherited the familial mutation can be tested to determine whether they are at risk for developing CRC due to Lynch Syndrome and if so, undergo frequent surveillance screening.

**Methodology:**
- **PCR:** Fluorescently labeled primers for coamplification of seven markers including five mononucleotide repeat markers (**BAT-25**, **BAT-26**, **NR-21**, **NR-24** and **MONO-27**) and two pentanucleotide markers (Penta C and Penta D). The mononucleotide markers are used for MSI determination and the
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Pentanucleotide markers are used to confirm identity match in the tumor and normal tissue samples in each patient.

**IHC:** Standard immunohistochemical staining on sections of formal-fixed paraffin-embedded tissue, using a biotin-free detection system with primary antibodies MLH1-G168-728, MSH2-G219-1129, MSH6-clone 44, PMS2-A-16-2.

Samples with instability in two or more of these mononucleotide markers are designated MSI-High (MSI-H), whereas those with one unstable marker are designated MSI-Low (MSI-L). Samples with no detectable alterations are MSI-stable (MSS). These designations are in accordance with the National Cancer Institute’s Bethesda guidelines.

**Specimen Requirements:**

**Preferred:**
- 1 paraffin tissue block of tumor with minimal normal tissue AND patient blood in purple or yellow top tube.

**OR**
- 1 paraffin tissue block of tumor with minimal normal tissue AND 1 paraffin tissue block of normal tissue.

**If tissue blocks cannot be sent:**
- **Tumor:** 5 unstained slides of Tumor at 7 micron, 5 unstained slides of Tumor at 5 micron on positively charged slides+ 1 H&E AND
- **Normal tissue** (5 unstained slides at 7 micron and 5 unstained slides at 5 micron on positively charged slides OR patient blood in purple or yellow top tube)

**A REQUISITION FORM MUST ACCOMPANY ALL SAMPLES.** Please include detailed clinical information, including ethnicity, clinical history, and family history.

**Test Performed (Days):**
Weekly

**Turn Around Time:**
7 - 14 days

**Shipment Sensitivity Requirements:**
Package and ship specimen to remain cool, but not frozen. Use the cold pack provided in the kit. Ship via overnight express, using the FedEx priority overnight label provided. Contact Client Services for shipping kits and instructions at (855) 535-1522.

References: