MAX Lab

Laboratory Investigation Report

Patient Name		Centre	: 1)
Age/Gender		OP/IP No/UHID	: I
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Immunocytochemistry

SIN No:SP0744172

Endometrium CA Molecular Profiling*

Immunohistochemistry (IHC) Number:
IHC-4463/24 to IHC-4467/24 (SC-747/24)

Specimen Type:
Uterus

Histopathology Impression:
High grade adenocarcinoma favour serous carcinoma over an endometrioid adenocarcinoma, grade 3.
pT1bN0.
FIGO stage - IIC.

Interpretation of IHC for MMR, IHC for P53 and Sanger Sequencing for POLE mutation:

IHC For MMR MLH1, PMS2, MSH2, MSH6	IHC For P53	Pathogenic POLE mutation
Intact nuclear expression	Mutant expression pattern	Absent

Final Impression:
Endometrial carcinoma, P53 mutated

Immunohistochemistry (IHC) Test:
IHC – P53 and IHC MMR (MLH-1, PMS 2, MSH 2, MSH 6)

Immunohistochemistry P53 Result: Diffuse strong nuclear expression in the neoplastic cells, consistent with a mutant expression pattern.

Immunohistochemistry (IHC) MMR Result:

Marker	*Result (Nuclear expression)*
MLH-1	Intact nuclear expression
PMS 2	Intact nuclear expression
MSH 2	Intact nuclear expression
MSH 6	Intact nuclear expression

MMR Impression: No loss of nuclear expression of mismatch repair (MMR) proteins: low probability of microsatellite instability–high (MSI-H)* + _

Control: Both the internal as well as external control show appropriate immunoreactivity

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Application:

- The primary application of IHC for MMR Protein is to screen for lynch syndrome / to identify patient at higher risk for additional colonic and extra-colonic tumours and with at - risk family members.
- IHC for MMR and p53 proteins act as act as surrogate marker for classifying the endometrial carcinoma as TCGA microsatellite instability (MSI) (hypermutated) and copy number high subgroups respectively.

IHC Interpretation:

- **Interpretation of p53 IHC**
  - *Wild type (normal) – scattered nuclear staining (upto 1 to 5 % nuclei), mild epithelial (basal sparing)*
  - *Aberrant (mutational type) →80 % strong and diffuse nuclear staining, complete absence of nuclear staining in all cells, moderate to strong cytoplasmic staining.*

- **Interpretation of MMR IHC**
  - *No loss of nuclear expression of mismatch repair (MMR) proteins: low probability of microsatellite instability–high (MSI-H)* + _*
  - *Loss of nuclear expression of MLH1 and PMS2: testing for methylation of the + _ MLH1 promoter is indicated (the presence of MLH1 methylation suggests that the tumor is sporadic and germline evaluation is probably not indicated; absence of MLH1 methylation suggests the possibility of Lynch syndrome, and sequencing and/or large deletion/duplication testing of germline MLH1 may be indicated)**
  - *Loss of nuclear expression of MSH2 and MSH6: high probability of Lynch syndrome (sequencing and/or large deletion/duplication testing of germline + _ MSH2 may be indicated, and, if negative, sequencing and/or large deletion/duplication testing of germline MSH6may be indicated)**
  - *Loss of nuclear expression of MSH6 only: high probability of Lynch syndrome (sequencing and/or large deletion/duplication testing of germline + _ MSH6 may be indicated)**
  - *Loss of nuclear expression of PMS2 only: high probability of Lynch syndrome (sequencing and/or large deletion/duplication testing of germline + _ PMS2 may be indicated)**
  - *There are exceptions to the above IHC interpretations. These results should not be considered in isolation, and clinical correlation with genetic counseling is recommended to assess the need for germline testing.*
  - *Reference : Endometrium - Biomarkers Carcinoma Endometrium* Biomarkers (v1.1.0.0 CAP protocol)

Notes:

- ***Detection System:*** *Optiview DAB IHC Detection Kit - VENTANA (IVD).*
- ***Primary Antibodies:***

- MLH-1 – Anti-MLH1, Clone M1, Mouse Monoclonal Primary Antibody, RTU, IVD, VENTANA (Ref – 760-5091)
- PMS 2 – Anti-PMS2, Clone A16-4, Mouse Monoclonal Primary Antibody, RTU, IVD, VENTANA (Ref – 760-5094
- MSH 2 – Anti-MSH2, Clone G219-1129 Mouse Monoclonal Primary Antibody, RTU, IVD, VENTANA (Ref – 760-5093
- MSH 6 – Anti-MSH6, Clone SP93, Rabbit Monoclonal Primary Antibody, RTU, IVD, VENTANA (Ref – 760-5092
- p53 – Clone BP-53-12,Mouse Monoclonal Antibody, IVD, RTU, PathnSitu (REF – PM101)

TEST REQUESTED

POLE gene mutation analysis

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METHOD USED

PCR, Sanger sequencing

SAMPLE INFORMATION

FFPE Block (Block No.: SC-747/24 I, Tumor Content: ~ 15-20 %)

RESULT

Negative

Interpretation

Wild Type	Indicates absence of mutation in both of the alleles
Heterozygous	Indicates presence of mutation in one of the alleles
Homozygous	Indicates presence of mutation in both of the alleles

NOTE

1. Genetic Counselling is recommended.

2. This is an in-house developed assay.

3. Test conducted on tissue block.

4. The method used is Sanger sequencing.

5. POLE Mutations Detected by the Assay**:**

***POLE* variant**	**CDS Mutation**	***POLE* variant**	**CDS Mutation**
p.P286R	c.857C>G	p.P436L	c.1307C>T
p.V411L	c.1231G>T	p.L424I	c.1270C>A
p.A456P	c.1366G>C	p.M444K	c.1331T>A
p.S297F	c.890C>T	p.S297A	c.889T>G
p.P436R	c.1307C>G	p.A428T	c.1282G>A
p.S459F	c.1376C>T	p.S461L	c.1382C>T

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(CIN No.: U85100DL2021PLC381826)

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p.A465V	c.1394C>T	p.A426V	c.1277C>T
p.P286S	c.856C>T	p.D275V	c.824A>T
p.L424V	c.1270C>G	p.L424P	c.?
p.T278M	c.833C>T	p.P441L	c.1322C>T

6. The mutations p.P286R, p.V411L, p.S297F, p.A456P, p.S459F are pathogenic (ultramutated) as per PMID 31829447 and 31829442.

COMMENTS

Molecular analysis of endometrial carcinoma has identified four clinically significant molecular subgroups with differing clinical prognoses: POLE mutations, microsatellite instability-high (MSI-H), copy number low, and copy number high. Mutations in the exonuclease domain of POLE have been reported to improve progression-free survival in endometrial cancer.

Kindly correlate with clinical findings

***** End Of Report *****

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