Identifying Lynch Syndrome in Patients with Newly Diagnosed Gynecologic Cancer

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GYNECOLOGIC ONCOLOGY
CARE RESEARCH & EDUCATION

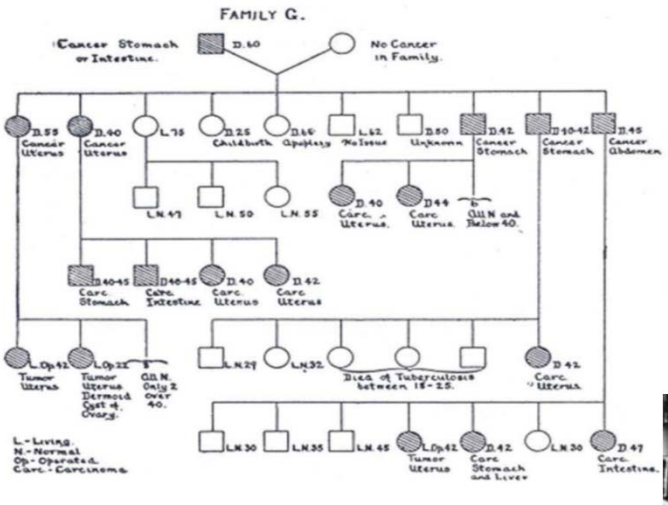
UHN Princess Margaret Cancer Centre

Disclosures

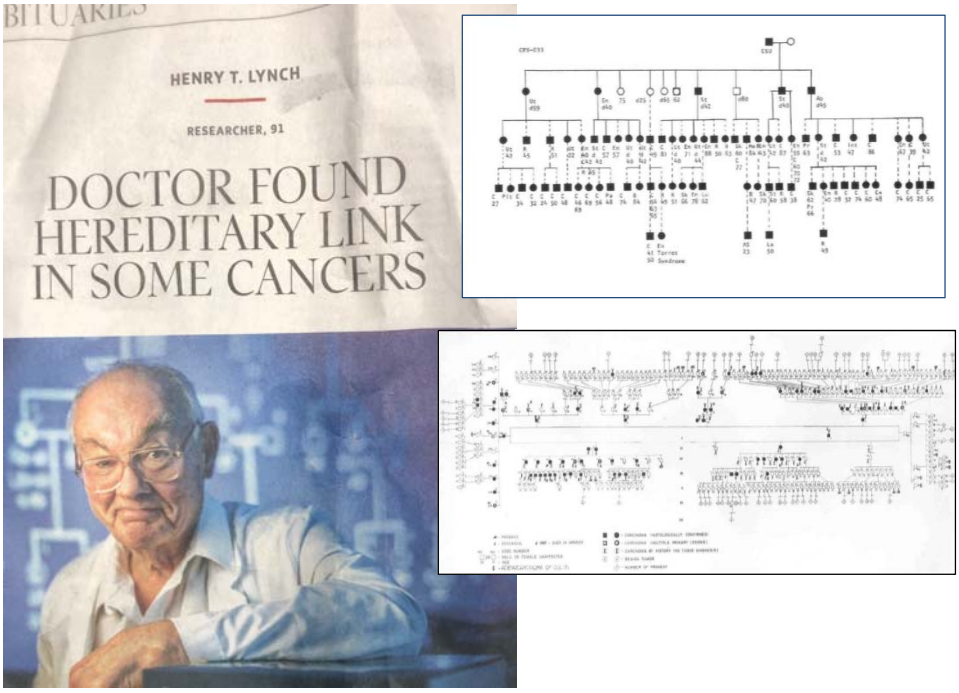
- I do not have an affiliation (financial or otherwise) with a pharmaceutical, medical device or communications organization.
- I do not intend to make therapeutic recommendations for medications that have not received regulatory approval (i.e. “off-label” use of medication).

Objectives

1. To review lifetime risk of gynecologic cancer in women with Lynch Syndrome (LS)
2. To understand the role of “reflex” IHC for mismatch repair (MMR) proteins in newly diagnosed endometrial cancer (EC) to identify LS
3. To discuss CCO guidelines for screening for LS in patients with newly diagnosed EC and ovarian cancer



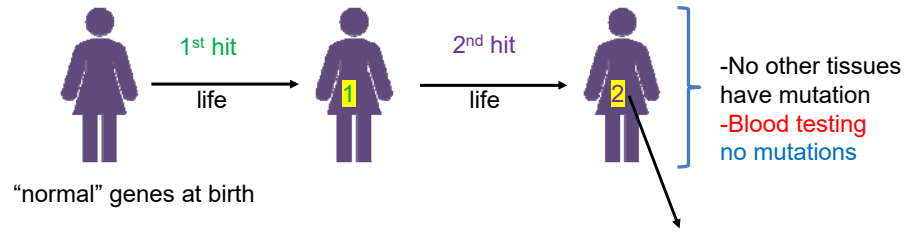
Dr. AS Warthin, *Archives Int Medicine* 1913



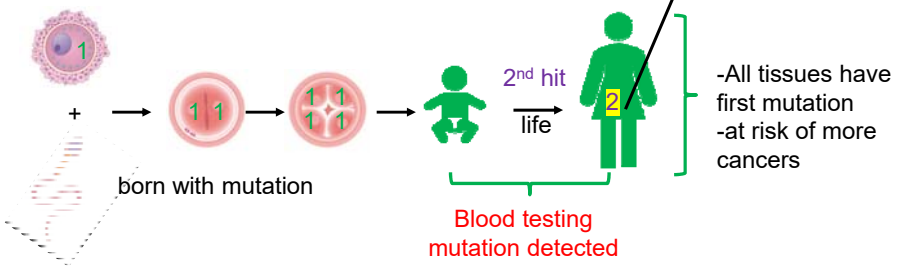
Lynch Syndrome

- Lynch Syndrome (LS) is a hereditary cancer susceptibility syndrome
- Familial clustering of cancers (e.g. CRC, EC and ovarian cancer)
- Autosomal dominant defects in ***mismatch repair (MMR)*** genes:
  - *MLH1 (42%)*
  - *MSH2 (33%)*
  - *MSH6 (18%)*
  - *PMS2 (7.5%)*
  - *EPCAM* -regulates *MSH2* – EPCAM deletion causes inactivation (by methylation) of MSH2 – an epigenetic mechanism
- 2-6% of unselected CRC and EC population germline mutation

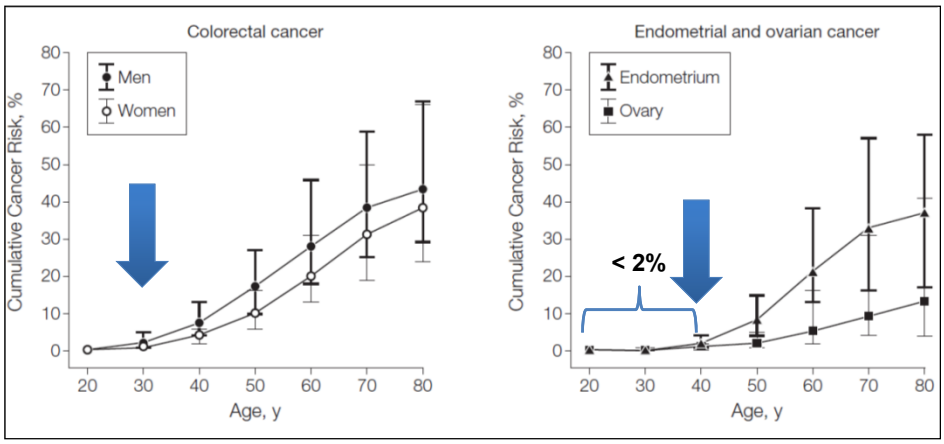
Sporadic/acquired cancer (90%)



Hereditary cancer (10%)



Cumulative Risk of Cancer by Age 70: *MLH1, MSH2, MSH6*



Bonadona *et al* 2011, *JAMA*

Cumulative Cancer Risk by Age 70

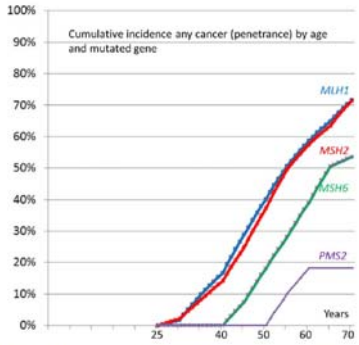


Figure 1 Calculated cumulative incidences by age and mutated gene for any cancer.

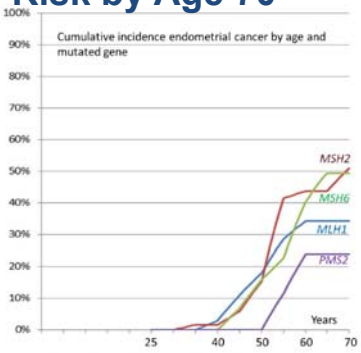


Figure 3 Calculated cumulative incidences by age and mutated gene for endometrial cancer as the first cancer by gene.

Gene	Total	EC	CRC	OC
MLH1	80% (71-88)	34%	45%	11%
MSH2	75% (65-85)	51%	33%	15%
MSH6	71% (52-90)	49%	26%	0
PMS2	24% (0-50)	24%	0	0

Moller *et al* 2015, *Gut*

Cumulative cancer incidence: 75% in females and 58% in males:

Missed Opportunity

- **Case:**
  - 52 year presents with PMB to family MD
  - Refers to general gynecology
  - Endometrial biopsy – grade 2 endometrioid adenocarcinoma
  - Referred to gynecologic oncology centre
  - No significant family history; maternal aunt had hysterectomy for cancer
  - Surgery – TLH/BSO/ pelvic lymphadenectomy
  - Final pathology: grade 2 endometrioid stage 1a (< 50% invasion) no LVSI
  - NO adjuvant therapy
  - On routine F/U describes rectal bleeding x 2 episodes - 3 years after surgery
  - Colonoscopy – right-sided colon tumour- biopsy + adenocarcinoma
  - CT multiple liver metastases and diffuse mesenteric adenopathy
  - ***Sent for Genetic assessment:***
    - Endometrial and Colon cancer MSH2 deficient on IHC for MMR;
    - Germline positive for pathogenic *MSH2* variant
  - Treated with chemotherapy
  - Died of disease 2 years later.

Why is it Important to Identify LS?

- **General gynecologist in Ontario:**
  - Manage 1/3 of the 2500 endometrial cancers in the province
- **EC is often sentinel cancer in LS**
  - 97% gynecologic cancer sentinel cancer
  - 82% sentinel cancer for the family
  - Median age: EC = 45 and OC = 39.5
- **Significant lead time before 2nd cancer**
  - 11 years (1-39) after diagnosis of EC developed 2nd cancer
  - 5.5 years (2-17) after diagnosis of OC developed 2nd cancer
- **Opportunity for cancer prevention in patient and FDR**
  - CRC screening - colonoscopy q 1-2 years:
    - Cancer Incidence – 57-69% ↓
    - Cancer Death – 65-83 % ↓
  - risk-reducing hysterectomy/BSO

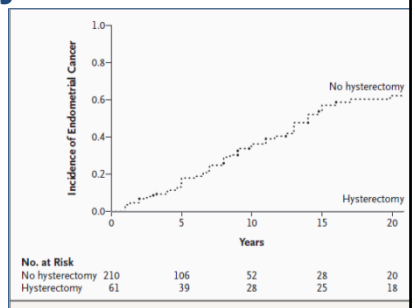
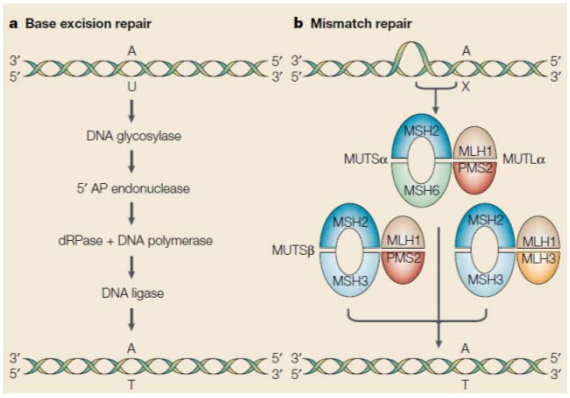


Figure 1. Cumulative Incidence of Endometrial Cancer among Women with the Lynch Syndrome Who Underwent Prophylactic Hysterectomy and Those Who Did Not.

Kim and Ferguson, Cancer 2021; Lu, Ob/Gyn 2005

Defects in MMR

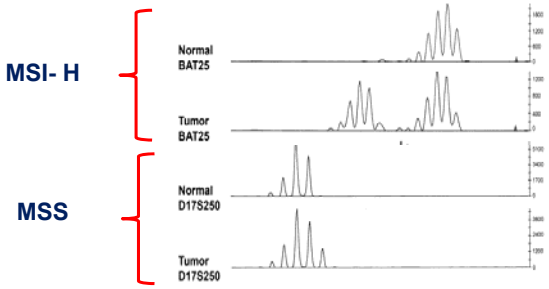
- MMR proteins correct errors that occur during DNA replication
- ***Impaired*** MMR leads to predisposition to mutations (***hypermutability***) – accumulation of errors
- Areas of nucleotide repeats (microsatellites) prone to mismatch = ***microsatellite instability*** (**MSI**)
- The presence of MSI represents ***phenotypic*** evidence that MMR is not functioning normally.
- **MMR defects due to:**
  1. Germline pathogenic variant;
  2. Somatic pathogenic variant;
  3. Epigenetic (hypermethylation)



Friedberg 2001, *Nature Reviews Cancer*

How Do We Identify MMR Defects?

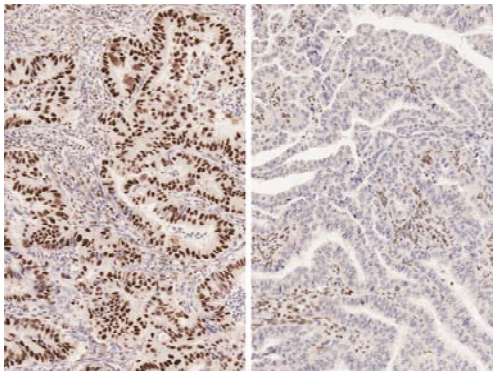
- **Microsatellite Instability (MSI)**
  - DNA based – compare tumour to normal
  - Series of loci examined (> 5)
  - ***25% of EC*** (15% CRC)
  - More expensive and more difficult to operationalize



How Do We Identify MMR Defects?

- **Immunohistochemistry**
  - 25% EC loss of protein expression (Deficient)
  - Direct germline testing
  - Easier to operationalize
  - 90-95 % concordant with MSI testing
  - More accurate for *MSH6* germline mutations in EC (EC often MSS)

MLH1 Intact



MLH1 Deficient

LS in Unselected Endometrial Cancer

- 562 incident cases EC
- 21.7% were MSI positive
- 2.3% positive germline mutation
- 15% missed by MSI testing; *MSH6*
- 38% diagnosed > age 50
- 62% did not meet Amsterdam II or Bethesda

Current screening guidelines for LS not adequate for EC population

Hampel *et al* 2006, *Cancer Research*

Lynch syndrome – Endometrial Cancer

- Prospective study N = 118 EC all histologies/stages < 70 yo
- 29% (N =34)) MMR deficient (MMRd)
- 5.9% (N = 7) positive pathogenic variant; 4 *MLH1*, 2 *MSH6*, 1 *MSH2*
- Of those MMRd, 28% had pathogenic variant- ***high pretest probability***
- ***Only 55% went for genetic assessment***

TABLE 4. Performance Characteristics of Screening Strategies for Identifying Mismatch Repair Germline Mutations (Lynch Syndrome) in Women With Newly Diagnosed Endometrial Cancer

Screening Strategy	No.	Sensitivity (95% CI)	Specificity (95% CI)	PPV (95% CI)	NPV (95% CI)
IHC	89	100 (59-100)	78.1 (67.5-86.4)	28 (12.1-49.4)	100 (94.4-100)
IHC age <60 y	43	100 (59-100)	86.1 (70.5-95.3)	58.3 (27.7-84.8)	100 (88.8-100)
MSI testing[a]	87	100 (54.1-100)	81.5 (71.3-89.3)	28.6 (11.3-52.2)	100 (94.6-100)
MSI testing[b]	89	85.7 (42.1-99.6)	81.7 (71.6-89.4)	28.6 (11.3-52.2)	98.5 (92.1-100)
MSI testing age <60 y[a]	41	100 (54.1-100)	88.6 (73.3-96.8)	60 (26.2-87.8)	100 (88.8-100)
MSI testing age <60 y[b]	43	85.7 (42.1-99.6)	88.9 (73.9-96.9)	60 (26.2-87.8)	97 (84.2-99.9)
eFHQ	82	71.4 (29-96.3)	86.7 (76.8-93.4)	33.3 (11.8-61.6)	97 (89.6-99.6)
Tumor morphology	83	71.4 (29-96.3)	42.1 (30.9-54)	10.2 (3.4-22.2)	94.1 (80.3-99.3)

Ferguson, *Cancer*, 2014

Lynch syndrome – Endometrial Cancer

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Tumor morphology	83	71.4 (29-96.3)	42.1 (30.9-54)	10.2 (3.4-22.2)	94.1 (80.3-99.3)

Recommended Reflex IHC on all EC < age 70

Ferguson, Cancer, 2014

Lynch syndrome – Genetic Navigation

- Prospective study N = 846 EC all histologies/stages & nonserous/ nonmucinous OC < 70 yo
- Navigated program – pathology reporting with directives
- 23% (N =194)) MMRd
- 4% (N = 34) positive pathogenic variant; 5 *MLH1*, 15 *MSH6*, 9 *MSH2, 5 PMS2*
- High pretest probability for MSH2/6 deficient; MSH6 deficient;
- ***91 % went to genetic assessment***

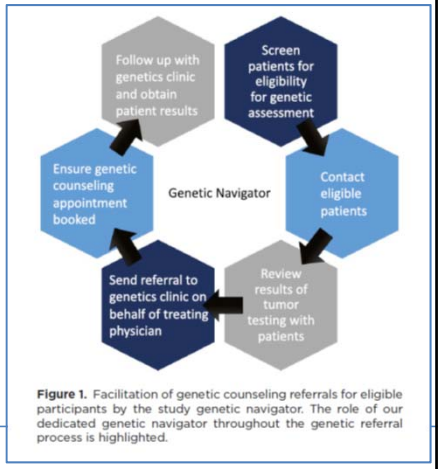


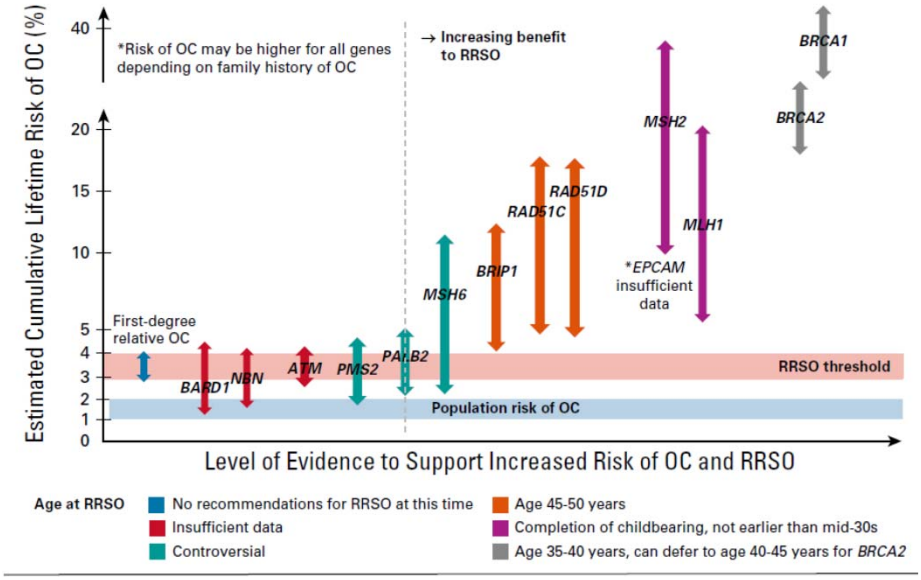
Figure 1. Facilitation of genetic counseling referrals for eligible participants by the study genetic navigator. The role of our dedicated genetic navigator throughout the genetic referral process is highlighted.

TABLE 4. Pretest Probability by Case Type and Immunohistochemistry Results

Case Type	LS+ Among MMRd	LS+ Among MLH1−/ PMS2−	LS+ Among MSH2−/ MSH6−	LS+ Among MSH6−	LS+ Among PMS2−	LS+ Among All Proteins Deficient
EC only	23/168 (14%)	2/123 (2%)	10/25 (40%)	6/13 (46%)	4/6 (67%)	1/1 (100%)
OC only[a]	6/15 (40%)	1/7 (14%)	1/4 (25%)	3/3 (100%)	1/1 (100%)	0/0
Sync EC component	4/11 (36%)[b]	1/7 (14%)	0/1	3/3 (100%)	0/0	0/0
Total	33/194 (17%)	4/137 (3%)	11/30 (37%)	12/19 (63%)	5/7 (71%)	1/1 (100%)

Kim and Ferguson, *Cancer*, 2021

Estimated Cumulative Risk of Ovarian Cancer



Liu et al, *JCO*, 2021

Lynch syndrome - Ovarian Cancer

- Prospective study N = 215 nonserous/nonmucinous ovarian cancer
- 13% (N =28)) MMRd
- 7% (N = 13) positive pathogenic variant; **7 *MSH6***, 3 *MLH1*, 2 *PMS2*, 1 *MSH2*
- Of those MMRd, 39% had pathogenic variant- ***high pretest probability***

TABLE 3. Performance Characteristics of Screening Strategies and Family History for Identifying Mismatch Repair Germline Pathogenic Variants (Lynch Syndrome) in Women with Newly Diagnosed Nonserous and/or Nonmucinous Ovarian Cancer

Screening Strategy	Total No.	Sensitivity (95% CI), %	Specificity (95% CI), %	PPV (95% CI), %	NPV (95% CI), %	No. of LS Patients Missed (95% CI)
IHC						(0-6)
IHC with *MLH1* pro methylation analys						(0-6)
MSI						(0-6)
OMOH family histor						(2-8)
IHC + MSI[b]						(0-7)
IHC with *MLH1* promoter methylation analysis + MSI[c]	188	92.3 (64.0-99.8)	97.7 (94.2-99.4)	75.0 (47.6-92.7)	99.4 (96.8-99.9)	1 (0-7)

Recommended Reflex IHC on all OC OR direct germline testing

Kim and Ferguson, Cancer, 2020

Recommendations for Screening for LS

Society of Gynecologic Oncology, March 2014:

"All women who are diagnosed with EC should undergo systematic clinical screening for LS (review of personal and family history) and/or molecular screening. Molecular screening of EC for LS is the ***preferred strategy*** when resources are available"

CCO Guideline 2015:

– Recommending reflex testing using IHC for all CRC and EC < age 70

CCO Funding Dec 2018:

– Reflex IHC on all newly diagnosed CRC and EC < 70 .
– Hypermethylation on all MLH1 deficient EC

CCO Funding August 2021:

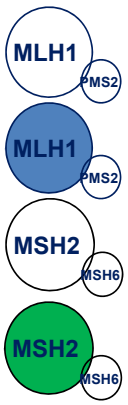
– Reflex IHC on all newly diagnosed CRC and EC ***regardless of age***
– Hypermethylation on all MLH1 deficient EC

CCO Funding August 2021:

– Germline testing on ***all epithelial ovarian cancer*** (breast and ovarian cancer panel test)
– Previously just high grade serous ovarian cancer

MMR IHC Interpretation

IHC Staining Pattern (Deficient)	Interpretation (Possible Variant)
MLH1 ***deficient*** and PMS2 ***deficient***	*MLH1*
MLH1 **intact** and PMS2 ***deficient***	*PMS2*
MSH2 ***deficient*** and MSH6 ***deficient***	*MSH2*
MSH2 **retained** and MSH6 ***deficient***	*MSH6*



MMR IHC Reporting: Intact Expression

Mismatch repair immunohistochemistry has been performed on this patient's endometrial cancer with the following results:

MLH1:	NORMAL - Intact nuclear expression
MSH2:	NORMAL - Intact nuclear expression
MSH6:	NORMAL - Intact nuclear expression
PMS2:	NORMAL - Intact nuclear expression

IHC interpretation – These findings indicate that it is ***unlikely*** that this carcinoma is associated with Lynch syndrome (LS) since tumours in patients with LS typically show abnormal mismatch repair protein expression. These immunohistochemical findings cannot entirely exclude the possibility of LS and ***family/personal history of cancers*** is still important.

Clinical Guidance - If there is a clinical suspicion of LS based on personal or family history referral to a genetic counsellor is indicated according to Ontario Ministry of Health guidelines. Features associated with LS include young age of onset (age < 50), strong family history of certain cancer types (including colorectal, small bowel, endometrial, ureter and renal pelvis and ovarian cancer) and multiple primary LS cancers in one individual.

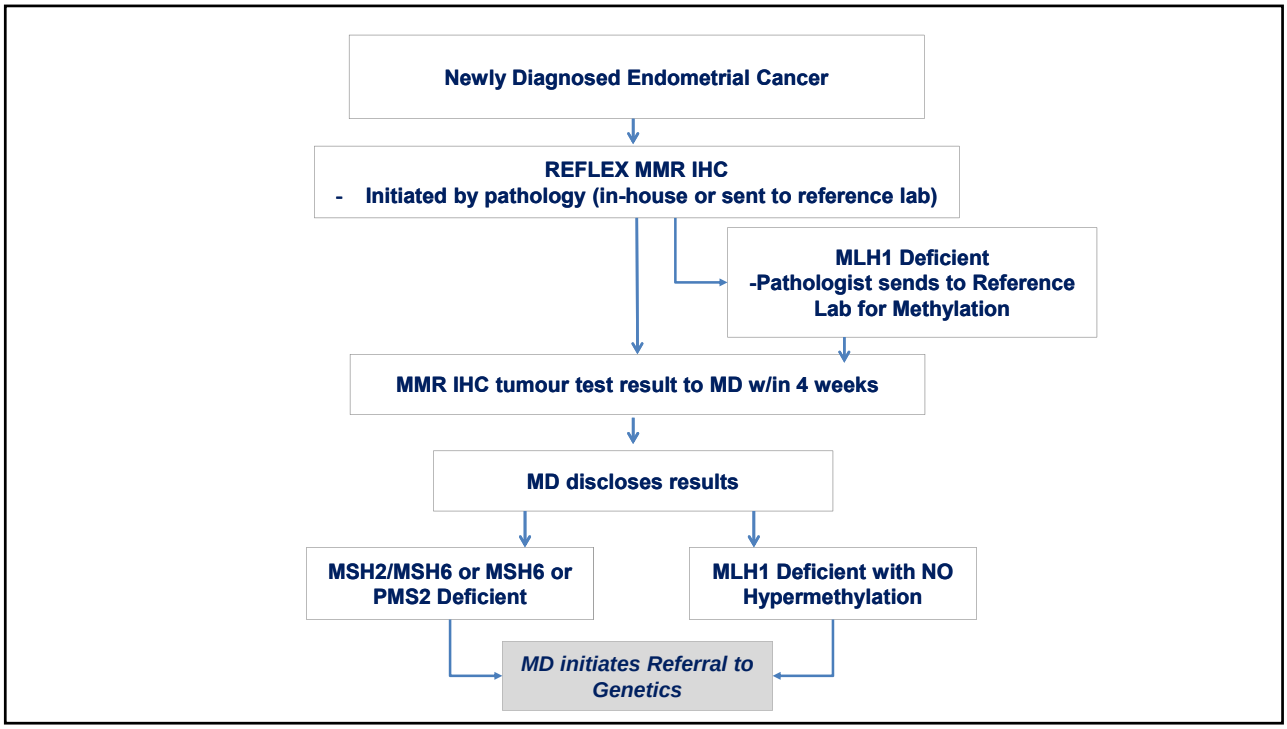
MMR IHC Reporting: Deficient MSH2/MSH6

Mismatch repair immunohistochemistry has been performed on this patient's endometrial cancer with the following results:

MLH1:	NORMAL - Intact nuclear expression.
MSH2:	***ABNORMAL – Loss of nuclear expression***
MSH6:	***ABNORMAL – Loss of nuclear expression***
PMS2:	NORMAL - Intact nuclear expression.

IHC interpretation- The patient's tumour has abnormal MSH2 expression (MSH2 Deficient tumour). Abnormal MSH2 expression can be associated with Lynch syndrome (LS). The expression of MSH6 is lost in MSH2 deficient tumours. As a result, loss of MSH6 expression by immunohistochemistry in this patient is not informative of possible alteration of the MSH6 gene.

Clinical Guidance - Referral to a genetic counsellor for Lynch syndrome assessment is indicated according to Ontario Ministry of Health guidelines.



Thank you

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GYNECOLOGIC ONCOLOGY
CARE RESEARCH & EDUCATION

HAMILTON HEALTH SCIENCES
Juravinski Cancer Centre
A Cancer Care Ontario regional partner

Reducing Gynecologic Cancer Risk in Unaffected Women with LS

- Role of Screening
- Chemoprevention
- Risk-reducing surgery

Rationale for Screening for Gynecologic Cancer in Unaffected Women with LS

Endometrial Cancer

- In premenopausal women symptoms less objective
- Potential beneficial to identify precancerous pathology and early stage disease:
  - Decrease burden of treatment; Up to 1/3 of women with EC require adjuvant therapy
  - Improve survival – unlikely to get this data

Ovarian Cancer

- Majority have isolated ovarian mass
- Majority early stage
  - 85% stage I/II
- Improved survival due to stage at presentation
  - 5 year OS 80%
  - 5-year OS only 40% for stage III/IV

Recommendations for Screening in Unaffected Women with LS

- **National Comprehensive Cancer Network guidelines 2019:**
  - Education of recognition and prompt ***reporting of symptoms*** with evaluation
    - Endometrial biopsy
  - No evidence to support screening for EC by U/S
  - However annual endometrial biopsy is sensitive and specific
  - Data ***do not support ovarian cancer screening*** but TV U/S and CA 125 maybe considered at clinician's discretion

Risk-Reducing Surgery in Lynch Syndrome

- Retrospective cohort study N = 315 (*MLH1, MSH2, MSH6*)
- No endometrial, ovarian cancer in surgery group
- 33% w/o hysterectomy developed EC (median age, 46)
- 5% w/o BSO developed OC (median age, 42)
- Recommended hysterectomy/BSO - age 40 or when done having children

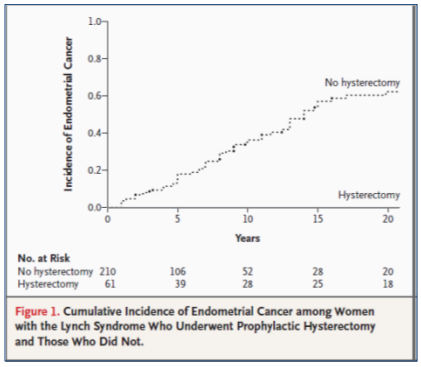


Figure 1. Cumulative Incidence of Endometrial Cancer among Women with the Lynch Syndrome Who Underwent Prophylactic Hysterectomy and Those Who Did Not.

Schmeler *et al N Engl J Med*, 2006

Chemoprevention – Lynch Syndrome

	No. (%) of Women		Incidence		
	With Available Data	Diagnosed With Endometrial Cancer	Incidence per 100 Person-Years	Difference (95% CI)	Hazard Ratio (95% CI)
Age at menarche					
≥13 Years	639	70 (11)	0.27	−0.04 (−0.15 to 0.05)	0.70 (0.44 to 1.11)
<13 Years (reference)	454	57 (12.6)	0.31		
Risk per year					0.85 (0.73 to 0.99)
Parity					
≥1 Live births	815	88 (10.8)	0.25	−0.18 (−0.32 to −0.04)	0.21 (0.10 to 0.42)
Nulliparous (reference)	278	40 (14.4)	0.43		
Hormonal contraceptive use					
≥1 Year	803	70 (8.7)	0.22	−0.23 (−0.36 to −0.1[illegible])	0.39 (0.23 to 0.64)
<1 Year (reference)	297	57 (19.2)	0.45		
Risk per year					0.93 (0.89 to 0.97)

There was no statistically significant association between endometrial cancer and age at first and last live birth, age at menopause, and postmenopausal hormone use.

Ghazaleh *JAMA* 2015

What Should Gynecologists Do to Reduce Gynecologic Cancer Risk?

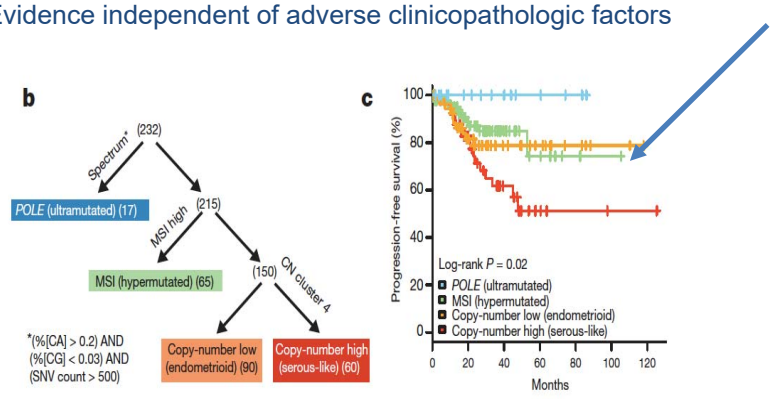
- ***No role for routine screening***: Refer to centre with expertise who follows these patients
- Should be performed by someone who ***understands Lynch syndrome*** in ***research setting:***
  - Only for women ***not complete child-bearing*** – discussion with patient
  - Understands limitations of the tests and
  - Can reduce delays in investigation/diagnosis
- Could be causing harm:
  - ***False sense of security*** – delay risk-reducing surgery
  - Distress
  - Over investigation for equivocal findings

What Should Gynecologists Do to Reduce Gynecologic Cancer Risk?

- Educate/screen patients about ***possible symptoms*** of endometrial and ovarian cancer
  - Postmenopausal bleeding
  - Abnormal bleeding
  - Urinary symptoms
  - Bowel symptoms – bloating, abdominal pain
- ***Investigate*** patients when they have these symptoms
  - Endometrial biopsy – 1st diagnostic test if abnormal bleeding
  - TV Ultrasound
- Discuss family planning and not to delay child-bearing
- Strongly recommend ***risk-reducing surgery*** at age 40 & done childbearing
- For women not done childbearing consider:
  - OCP if there is no contraindication- better for OC and EC prevention
  - Levonorgestrel IUS
- Ensure up to date on colonoscopy – facilitate referral

Other Benefit of Reflex IHC Beyond LS

- Mismatch repair deficient (MMRd) with intermediate prognostic –20 % recur
- Evidence independent of adverse clinicopathologic factors



Other Benefit of Reflex IHC Beyond LS

- **Cancer Treatment:**
  - Evidence of benefit of Pembrolizumab (immunotherapy) - recurrent cancer with evidence of MMR defect
  - 1st time disease site “agnostic” therapy
  - Health Canada approved April 2019