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Lynch Syndrome Screening in Gynecological Cancers: Results of an International Survey with Recommendations for Uniform Reporting Terminology for Mismatch Repair Immunohistochemistry Results

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Abstract

Aim: Lynch Syndrome (LS) is associated with an increased risk of developing endometrial carcinoma (EC) and ovarian carcinoma (OC). There is considerable variability in current practices and opinions related to screening newly diagnosed patients with EC/OC for LS. An online survey was undertaken to explore the extent of these differences.

Methods: An online questionnaire was developed by a panel of experts and sent to all members of the British Association of Gynaecological Pathologists (BAGP) and the International Society of Gynecological Pathologists (ISGyP). Anonymised results were received and analyzed.

Results: Thirty-six BAGP and 44 ISGyP members completed the survey. More than 90% of respondents were aware of the association of LS with both EC and OC, but 34% were not aware of specific guidelines for LS screening. Seventy-one percent of respondents agreed that universal screening for LS should be carried out in all newly diagnosed EC cases, with immunohistochemistry (IHC) alone as the preferred approach. Only 36% of respondents currently performed IHC or microsatellite instability testing on all newly diagnosed EC, with most of the remaining respondents practicing selective screening, based on clinical or pathological features or both. A significant minority of respondents (35%) believed that patient consent was required before performing MMR IHC. Almost all respondents favored use of standardized terminology for reporting MMR staining results and this is proposed herein.

Conclusion: There is wide support for universal LS screening in patients with EC, but this survey highlights areas of considerable variation in practice.

Key words: Lynch Syndrome, endometrial carcinoma, mismatch repair, screening, TCGA, immunohistochemistry, terminology, consent

Introduction:

Lynch Syndrome (LS) is an inherited predisposition to a variety of cancers caused by a pathological mutation within the mismatch repair (MMR) genes (*MLH1*, *MSH2*, *MSH6*, *PMS2*) (1). LS is a grossly under-diagnosed condition with an estimated 95% of cases being unrecognised (2). Women with LS have a lifetime risk of approximately 50-60% of developing each of colorectal carcinoma (CRC) and endometrial carcinoma (EC), with a lower risk of ovarian carcinoma (OC) of 8-10%; OC is the third most frequent malignancy in women with LS (3-5). EC is the first or sentinel malignancy in more than half of those women with LS who develop cancer (6). A significant proportion of women with LS and EC go on to develop a second or further malignancies (7), the risks of which have been shown to be up to 55% and 15% respectively (8). Surveillance and preventative strategies aimed at the patient and susceptible family members results in a decreased incidence and substantially reduced mortality from LS-related cancers (7, 9-11).

Screening for LS in all newly diagnosed EC patients therefore offers an opportunity to detect undiagnosed LS. EC has an excellent 5-year disease specific survival (12). As EC is often a sentinel event, this allows diagnosis before the development of potentially fatal malignancies such as CRC. Early diagnosis enables colorectal surveillance which, is known to improve the disease specific survival in CRC (3, 9, 10). In addition, cascade testing of family members allow the diagnosis of healthy pathological MMR mutation carriers and their subsequent enrolment into suitable surveillance programmes.

Pathological MMR mutations lead to dysfunctional and structurally abnormal MMR proteins. This in turn brings about a detectable tumour phenotype with loss of MMR protein expression and a hypermutated tumour demonstrating microsatellite instability (1, 13). Screening for pathological MMR mutations can be carried out easily using readily available immunohistochemistry (IHC) for MMR proteins (Figures 1 and 2), or by MSI testing, or both (14-16). Overall only 2-3% of ECs occur in patients with LS, and clinical and morphological factors are not sensitive enough to allow targeted screening as cases would be missed regardless of the selection criteria used (17). Guidelines from several professional bodies and expert consensus groups therefore advocate universal screening for LS in all newly diagnosed EC cases (18-22) however there is wide variation in practice and a lack of universal agreement amongst pathologists (23, 24).

The identification of MMR defective (MMRd) EC has implications beyond the detection of LS. ECs with acquired somatic pathological MMR mutations (alongside those with germline mutations) are susceptible to check point

inhibition treatments (25). Furthermore, the publication of the Cancer Genome Atlas (TCGA) Study and subsequent validation studies have shown that EC can be subclassified into 4 different molecular categories based on their mutational profiles, including one associated with MMRd that accounts for approximately 25% of EC (26-28). Tumors within the 4 molecular categories differ with respect to prognosis, and response to treatment. Therefore, universal screening of ECs enables tailored management and is of benefit despite LS screening.

LS screening in all or selected cases of OC is more controversial. However, the risk of LS in patients with ovarian endometrioid or clear cell carcinoma is identical to that of patients with CRC or EC (29, 30); the risk of LS in patients with other OC histotypes is much lower (29, 31).

Given the factors above, we undertook a survey amongst members of two large gynaecological pathology societies in order to assess the current understanding of LS in relation to gynaecological malignancy and the views of pathologists regarding screening for LS. We hoped this would provide critical information about the current clinical landscape with an international perspective.

Methods:

A collaborative multidisciplinary group was formed from members of the British Association of Gynaecological Pathologists (BAGP), International Society of Gynecological Pathologists (ISGyP) and invited experts, to design questions that would explore variation in attitudes and current practices related to LS screening in gynecological cancers. The group included 4 pathologists (NS, WGM, MJA, CBG), 2 clinical geneticists (DGE, LS), 3 gynaecological surgeons (NR, EC, RM) and 1 molecular geneticist (IF). We also utilised Lynch syndrome UK (a patient advocate group) to ensure our questionnaire content captured the key concerns of both clinical and patient stake holders. After taking opinions from the experts, the questions were formulated and sent for review to the panel for comments. These were then sent to members of the council of the BAGP and ISGyP, finalised by two senior authors (EC and NS) and approved by the panel for sending out to the memberships of BAGP and ISGyP. The link to the electronic survey hosted on the University of Manchester website was sent to all members of the BAGP and the ISGyP between January and June 2017.

The survey was composed of 36 questions and included a combination of dichotomous, multiple choice and open-ended format questions. The questions were separated into those relating to the respondent, those assessing background knowledge, those assessing current practice and lastly those exploring the opinions on future guidelines. The survey questions are tabulated in Supplementary Table 1.

Responses from the two groups surveyed were compiled and the results analyzed. Any survey that was returned incomplete was excluded from analysis.

Results:

A total of 52 ISGyP members and 48 BAGP members responded. These represented a minority of the 177 (27%) BAGP and 345 (14%) ISGyP members to whom the link was emailed. Members of ISGyP received the survey after the BAGP survey had been closed; while some participants may have had the opportunity to respond on both occasions, given the low response rate overall, and the differences in the responses as below, this seems unlikely. Twenty responses were excluded due to incomplete data (15 of the responders reported having no knowledge LS and did not offer responses to further questions). There were 44 complete surveys from ISGyP and 36 from BAGP members giving a total of 80 completed datasets.

Background information about the respondent

Responses were received from the UK and mainland Europe, Asia, North and South America, Africa and Australia. Of the BAGP respondents, who had obtained their primary medical qualification in a variety of countries worldwide, 30/36 had obtained their higher specialist training in the UK, and all of the remainder in mainland Europe. Of the included ISGyP responses, 24 (55%) were from North America and 15 (34%) from Europe, with the remainder being from Australia (3, 7%), South America and Africa (1 respondent (2%) each). Half of responders were based in an academic centre/teaching hospital affiliated to a medical school or tertiary referral center, with smaller numbers based in private hospitals or laboratories or district general hospitals. Thirty nine percent (n=31) were based in a cancer center or unit. The majority of responders (74%, n=59) described themselves as practicing in many areas with a special interest/responsibility in gynaecological pathology. Twenty five percent (n=20) solely practiced gynaecological pathology.

Half of pathologists (n=40) reported diagnosing over 100 new cases of EC annually (including referrals).

Baseline knowledge about Lynch Syndrome

Of note, fifteen percent of those pathologists who had initially responded, had no knowledge of LS and were therefore removed from further analysis. Those included in the analysis were aware of LS. When asked to select associated malignancies, 100% included CRC. Only 1% (n=1) and 3% (n=2) did not include EC and OC respectively.

The majority of pathologists (70%, n=56) were aware of guidelines such as those of the Society of Gynecologic Oncologists(31) relating to the screening

of LS. Thirty percent (n=24) of the practitioners questioned reported being unfamiliar with any guidelines. Fourteen percent (n=11) were not aware of the recently published TCGA molecular classification of EC.

When asked 'roughly what proportion of sporadic or germline (LS) DNA mismatch repair gene defects can be detected by IHC alone for mismatch repair gene proteins', 81% of respondents (n=65) opted for a proportion above 75% with 43% (n=34) responders choosing >90%.

Current practice regarding screening for mismatch repair defects and microsatellite instability

Only three percent (n=2) respondents never carried out MMR IHC on EC cases, and 36% (n=29) tested all new cases. There was a striking difference in the proportion of pathologists currently carrying out universal MMR testing in all cases of EC between respondents from North America (19/24, 79%) and those from all other regions (9/56, 16%). Amongst respondents who carried out LS screening in selected cases, the indications varied widely as summarized in Table 1. There was an overall majority amongst BAGP and ISGyP members in favor of universal screening for LS in new cases of EC, with less agreement on reflex screening in OC (Figure 3).

Of the pathologists who undertook MMR IHC for initial screening, 81% (n=65) requested all four markers (MLH1, PMS2, MSH2, MSH6) in the first instance. Far fewer (8%, n=10) initially requested 2 markers with a third marker or full panel subsequently performed if there is loss of staining or equivocal staining with any marker.

Nearly half of those questioned had access to *MLH1* promoter methylation testing (if IHC revealed a loss of MLH1), either within their institute or regionally (25% (n=20) and 36% (n=29) respectively). Fourteen percent (n=11) respondents had no access to this testing. Seventy percent (n=56) had access to MSI testing regionally and/or within their institution. These respondents were employed in both University and non-University hospitals.

When questioned further regarding initiating testing for MSI in their institution, 32 of 56 (57%) pathologists with access to testing did so only in those cases where the initial IHC was normal, equivocal or difficult to interpret. Only four of 56 (7%) sent all samples for MSI regardless of the IHC result.

A majority of respondents (62%, n=60,) had germline MMR gene sequencing available to them at least on a regional level. Eight percent (n=6) of responders had no access, and 29% (n=23) were unsure.

Gross specimen handling

Over two thirds (68% n=54) of the pathologists were based in departments that had received prophylactic hysterectomy and salpingo-oophorectomy specimens for LS cases. The majority of these (60%, n=48) reported that there was a departmental standard protocol for dissecting and sampling these specimens; in the remainder, no protocol was in place.

Consensus regarding universal screening of new EC for Lynch Syndrome

There was overwhelming agreement for universal screening of all new EC cases for LS with 71% (n=57) in favor of this approach (41% strongly agreeing). When the response was 'neutral' or 'disagree', reasons cited included cost implications and the relative low incidence of LS-related EC. Of note, those who disagreed with universal screening were employed in university and non-university hospitals. When questioned about the ideal method for universal screening of new cases of EC, 59% (n=47) advocated the use of IHC alone while 29% (n=23) favored combined IHC and MSI testing (the remainder were unsure about a preferred strategy). The majority of those that advocated IHC and MSI were employed in a University hospital (n=14). The preferred method for initial screening of new cases of EC varied according to the group surveyed with ISGyP members favoring IHC alone whereas, amongst BAGP members, a combination of IHC and MSI testing was the preferred option (Figure 4).

Cost effectiveness was again provided as a rationale for IHC (n=28) only while others were guided by previous experience (n=7).

On the question of whether consent for IHC testing was required in new cases of EC, 38% (n=28) held the opinion that consent should be gained from the patient before MMR IHC testing.

The pathologists were then asked 'if universal MMR IHC was recommended for EC which of the following would serve as an initial screening test in your opinion?'. Sixty four percent (n=51) responded that testing for all four MMR markers should be undertaken in the first instance. All of those who had a preference for 2 protein (n=2) IHC screening (MSH6 and PMS2 with reflex four protein IHC for those found to be deficient) both worked in a University hospital. When asked if their respective laboratories had the capability to absorb the time and cost demands of performing MMR immunohistochemistry on all new EC, over half (56% n=45) stated that they would. Thirty three percent (n=26) however felt that this would not be practical with 11% (n=9) strongly stating that their laboratory did not have this capacity. Figure 5 shows the percentage of BAGP and ISGyP respondents who agreed and disagreed with the statement regarding laboratory capability in the event of the introduction of universal LS testing for new EC cases.

There was a divergence of opinion regarding the centralization of IHC screening and molecular analysis (e.g. microsatellite instability analysis) in the diagnostic pathway in the event of introduction of universal screening. Almost equal numbers agreed (44% n=35) or disagreed (39% n=31) with centralization, interestingly those who agreed with centralization mostly (n=23) employees of a University hospital.

There was almost universal acceptance of the need for standardized reporting terminology for MMR immunohistochemistry with only one responder indicating neutrality when asked about the utility of this approach. Multiple answers were provided when asked about who should lead on referring EC patients with abnormal screening results for LS germline testing. In the majority of cases, multiple responses were given with the gynecological oncology multidisciplinary team /tumor board meeting widely identified as the preferred option. Ten responders felt that the pathologist should be solely responsible for referral.

Universal screening of new ovarian carcinomas for LS

Attitudes towards the screening of women with a new diagnosis of OC were also assessed. There was a tendency towards disagreement regarding the introduction of universal screening amongst BAGP and ISGyP members surveyed (figure 1). Overall, 83% (n=66) of responders were neutral, disagreed or strongly disagreed with the introduction of universal screening in this specific context. When questioned further, responders who disagreed cited reasons relating to incidence, cost-effectiveness and the need to base decisions on tumor morphology (i.e. endometrioid and clear cell types) rather than employ a universal screening approach.

Discussion

This survey has offered insights into current knowledge and practice with respect to screening EC and OC for LS among ISyGP and BAGP members. As with similar studies in the past, this shows continuing variation in attitudes and practices (23, 24). It is acknowledged that the survey had a poor response overall from both societies, a reflection itself perhaps of the lack of awareness in regard to this issue. Considering that these responses are from those who are aware of the implications of screening, a particularly striking finding is the difference between responses obtained from the two societies with respect to universal screening for LS in EC. This is likely related to the lack of clear guidelines and resources for testing. In the United Kingdom, universal LS screening for colorectal cancers has been recommended by the National Institute of Health and Care Excellence (32) since February 2017, however a similar approach for EC is currently under assessment with a

decision anticipated before the end of 2020. In the absence of mandatory testing, UK laboratories have to absorb the cost of additional immunohistochemistry or molecular testing, while differences in adoption of guidelines and/or funding may explain the easier access and greater support of universal testing in other countries.

Responders had a good awareness of the association between LS and the subsequent development of EC; less encouraging however was the finding that 7% of those who completed the survey were unaware of published guidelines such as those produced by The Society of Gynecologic Oncologists, the National Comprehensive Cancer Network and the Royal College of Pathologists of UK. It is also important to note that 15 responses were excluded because the respondents reported having 'no knowledge of LS'. Sixteen percent of those completing the survey were unaware of the recently published TCGA molecular classification of EC. This suggests that there is a need to further disseminate 'best practice' recommendations regarding these hereditary cancers. Furthermore, it highlights the disconnect between research and clinical practice; the identification of MMRd tumors has key prognostic and therapeutic implications which could be denied to patients if clinicians are unaware of them (25, 26).

A high percentage of the pathologists who undertook the survey were involved in screening for LS in new cases of EC and they were overwhelmingly likely to use IHC with the four markers MLH1, PMS2, MSH2, MSH6. Over 70% were in favor of universal screening using IHC as the preferred technique; cost and previous experience with this technique were given as reasons for this choice over the combination of a combined IHC/MSI approach. Interpretation of MMR IHC is relatively straightforward provided that standardised and external quality assured protocols are in use and there is due regard to pitfalls in interpretation (16). There was strong support for standardized reporting of MMR IHC. As a result of this and as part of a larger consensus project on LS screening, standardised terminology is recommended which is presented in table 2 (33). Of note was the disparity between access to testing due to working in a non-University hospital setting. This health inequality remains a key barrier to universal screening of EC for LS. It is potentially an argument for centralization of such testing so as to assure access for all. In addition, it may prove cost effective to have central referral and therefore efficient resource use, although further research exploring this is required.

An interesting finding was the significant proportion of pathologists who believed that individual patient consent should be granted before initiating MMR IHC testing. In the event of universal screening, approximately one third of pathologists held the opinion that consent should be sought before proceeding with IHC. This has been an area of controversy for years, with

advocates for universal screening without consent arguing that the results of MMR IHC only indicate an increased likelihood of LS, and affording the patient the option of undergoing definitive testing. It is the opinion of the authors that testing that yields somatic and not germline information, gives information about the biology of the tumour and not that of the patient. In this way, it is arguably no different from reporting an EC arising in the lower uterine segment, with a prominent peritumoral and intratumoral lymphocytic infiltrate, in that such a patient would also be at increased risk of LS. With the constantly changing face of genomic medicine, ethical considerations regarding the implications of these investigations are likely to become more contentious. This is an area where guidance and formulation of a standardized approach may be of benefit. With only 2-3% of EC cases occurring in LS patients, taking individual consent prior to a screening test would place an unnecessary burden on the patient and the clinical team; it is far more preferable to only seek consent for definitive germline testing in women who are positive on their screening test. A positive screening test does not equate to LS; approximately 50% of cases who are screen-positive following MMR IHC combined with *MLH1* promoter methylation testing will show positive results on germline testing, the remainder being the result of false negative MMR IHC, bi-allelic somatic mutations and epigenetic phenomena, as well as false negative germline testing results (13, 34, 35). From the viewpoint of TCGA disease classification, the situation is analogous to performing p53 IHC testing to help delineate serous-like EC; the presence of *TP53* mutation is sought in the tumour and while this could be indicative of Li-Fraumeni syndrome, consent is not taken *a priori* from the patient.

The practical implications of universal testing in cases of newly diagnosed EC were investigated with questions regarding access to IHC and relevant genomic testing. An encouraging finding was that the majority of respondents reported that they believed their laboratory would be able to cope with the practical and financial impact of the introduction of universal screening. This was not so evident outside University hospital settings. Thirty-one responders reported having no access to, or being unaware of arrangements for *MLH1* promoter methylation testing. MSI testing, however, was more likely to be available with 70% having access. In regions with access to *MLH1* promoter methylation testing, an equivocal and/ or difficult to interpret IHC result was often given as the rationale for requesting MSI testing. Eight pathologists had no access to germline MMR sequencing and a further 23 reported being unaware of the availability of this service in their region. These results highlight the variation in molecular services available to pathologists involved in the diagnosis and management of women with hereditary endometrial and ovarian malignancies and the barriers to testing posed by global health inequality.

Most pathologists who responded to the survey were receiving risk-reducing hysterectomy and salpingo-oophorectomy specimens and while the majority had an agreed protocol in place for the processing of these surgical

specimens, nearly 20% had no protocol. It could be argued that if these specimens are handled as routine 'benign' cases, they are likely to be less widely sampled and this may have implications for the detection of occult malignancy. This warrants the adoption of specific guidance. The recently published ISGyP recommendations for specimen handling in endometrial carcinoma, carried out as part of a project on endometrial carcinoma reporting, includes guidance on LS cases (36). Similar recommendations have also been published previously (37) and these include sampling the entire endometrium, including the lower uterine segment, fallopian tubes and ovaries with representative sampling of the cervix.

There was less agreement when the pathologists were questioned about universal screening of newly diagnosed OC with divergent opinions expressed. The disagreement, at least in part, can be explained by the differing likelihood of finding MMR deficiency in different OC histotypes, with a very low prevalence in the most common tumor type high-grade serous carcinomas. Many of the respondents specifically stated that screening this subset of OC patients was not indicated. Resource issues were also cited as a reason why this should not be adopted as a strategy. Given the increasing availability and diminishing costs of next generation sequencing, in which panels of genes are tested simultaneously, together with the 15-20% possibility of patients with high-grade serous carcinoma having germline mutations in genes associated with Hereditary Breast and Ovarian Cancer syndrome, it may be preferable to proceed directly to this testing in all newly diagnosed OC patients, using a gene panel that includes genes associated with LS and Hereditary Breast and Ovarian Cancer.

As with all surveys, this one has important limitations. Only a minority of members of both societies responded to the survey. The respondents are self-selected with a likelihood of having an interest in this field; it is therefore possible that this is not a true representation of current practice and that current coverage of and support for LS screening in EC is even lower than the results suggest. The BAGP survey preceded that within ISGyP and some questions therefore slightly differed; for example ISGyP members were asked in which country they practiced while BAGP members largely work within the UK and were asked their country of primary and specialist qualification; the data were insufficient to draw meaningful comparisons between practices in different nations.

Conclusions

Despite good evidence that screening for LS in EC has high potential for reducing morbidity, mortality and healthcare costs, and the wide availability of screening tests, there remains huge variation in the approach to screening. Pathologists need to be aware of the implications of LS screening for the

patient and her family. Standardised and external quality assured protocols should be developed and adopted for LS screening in EC. The ISGyP has recently recommended that all newly diagnosed cases of EC undergo testing for LS using IHC or molecular testing as the initial modality (21, 22). We also suggest that all newly diagnosed patients with ovarian endometrioid or clear cell carcinoma are similarly screened for LS.

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All authors have contributed to this manuscript:

Questionnaire design: NS, NR, EJC, MA, IF, R Manchanda, WGM, LS, DGE, CBG

BAGP council approval and amendment of questionnaire: SA, AF, RG, YLH, PM, GvS, BT

Analysis of results: NR, JW, NS

MMR IHC reporting terminology: MA, TB, IF, R McMahon, DGE, CBG, NS

Manuscript preparation: All authors contributed to the preparation of the manuscript.

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Figure Legends

Figure 1a. Normal expression of mismatch repair proteins in well-fixed specimen. This example of MLH1 staining in a biopsy serves to demonstrate typical MMR protein expression as diffuse strong nuclear staining, typically stronger than the background stromal nuclei that serve as an internal control.

Figure 1b. MSH2 loss in a case of known germline *MSH2* mutation.

Figure 2a. MMR protein staining is fixation sensitive; in this example of normal MLH1 staining the nuclei stain more weakly than in the biopsy demonstrated in Figure 1a. The stroma and normal non-neoplastic endometrium serve as internal controls.

Figure 2b. In poorly fixed specimen due attention should be paid to the internal control which is weaker. In this example of germline *MSH2* mutation, staining for MSH2 protein is absent in the presence of positive, albeit weak staining in background stromal and endometrial glandular epithelial cells.

Figure 3. Bar chart showing responses amongst BAGP and ISGyP members regarding the introduction of universal screening for Lynch syndrome for new cases of endometrial and ovarian malignancy.

Figure 4. Bar chart showing the favored screening test for new cases of endometrial malignancy amongst BAGP and ISGyP members.

Figure 5. Bar chart showing the responses of BAGP and ISGyP members when questioned about whether their laboratory would be able to absorb (agree) or unable to absorb (disagree) the demands of universal testing for Lynch Syndrome for new cases of endometrial carcinoma.

References

1. Lynch HT, Lynch PM, Lanspa SJ, Snyder CL, Lynch JF, Boland CR. Review of the Lynch syndrome: history, molecular genetics, screening, differential diagnosis, and medicolegal ramifications. *Clin Genet.* 2009;76(1):1-18.
2. Singh H, Schiesser R, Anand G, Richardson PA, El-Serag HB. Underdiagnosis of Lynch syndrome involves more than family history criteria. *Clin Gastroenterol Hepatol.* 2010;8(6):523-9.
3. Moller P, Seppala T, Bernstein I, Holinski-Feder E, Sala P, Evans DG, et al. Cancer incidence and survival in Lynch syndrome patients receiving colonoscopic and gynaecological surveillance: first report from the prospective Lynch syndrome database. *Gut.* 2017;66(3):464-72.
4. Ryan NAJ, Evans DG, Green K, Crosbie EJ. Pathological features and clinical behavior of Lynch syndrome-associated ovarian cancer. *Gynecol Oncol.* 2017;144(3):491-5.
5. Ryan NAJ, Morris J, Green K, Lalloo F, Woodward ER, Hill J, et al. Association of Mismatch Repair Mutation With Age at Cancer Onset in Lynch Syndrome: Implications for Stratified Surveillance Strategies. *JAMA Oncol.* 2017;3(12):1702-6.
6. Lu KH, Dinh M, Kohlmann W, Watson P, Green J, Syngal S, et al. Gynecologic cancer as a "sentinel cancer" for women with hereditary nonpolyposis colorectal cancer syndrome. *Obstet Gynecol.* 2005;105(3):569-74.
7. Moller P, Seppala T, Bernstein I, Holinski-Feder E, Sala P, Evans DG, et al. Incidence of and survival after subsequent cancers in carriers of pathogenic MMR variants with previous cancer: a report from the prospective Lynch syndrome database. *Gut.* 2017;66(9):1657-64.
8. Win AK, Lindor NM, Winship I, Tucker KM, Buchanan DD, Young JP, et al. Risks of colorectal and other cancers after endometrial cancer for women with Lynch syndrome. *J Natl Cancer Inst.* 2013;105(4):274-9.
9. Moller P, Seppala TT, Bernstein I, Holinski-Feder E, Sala P, Gareth Evans D, et al. Cancer risk and survival in path_MMR carriers by gene and gender up to 75 years of age: a report from the Prospective Lynch Syndrome Database. *Gut.* 2018;67(7):1306-16.
10. Burn J, Gerdes AM, Macrae F, Mecklin JP, Moeslein G, Olschwang S, et al. Long-term effect of aspirin on cancer risk in carriers of hereditary colorectal cancer: an analysis from the CAPP2 randomised controlled trial. *Lancet.* 2011;378(9809):2081-7.
11. Schmeler KM, Lynch HT, Chen LM, Munsell MF, Soliman PT, Clark MB, et al. Prophylactic surgery to reduce the risk of gynecologic cancers in the Lynch syndrome. *N Engl J Med.* 2006;354(3):261-9.
12. Amant F, Mirza MR, Koskas M, Creutzberg CL. Cancer of the corpus uteri. *Int J Gynaecol Obstet.* 2018;143 Suppl 2:37-50.
13. Haraldsdottir S, Hampel H, Tomsic J, Frankel WL, Pearlman R, de la Chapelle A, et al. Colon and endometrial cancers with mismatch repair deficiency can arise from somatic, rather than germline, mutations. *Gastroenterology.* 2014;147(6):1308-16 e1.
14. Djordjevic B, Broaddus RR. Laboratory Assays in Evaluation of Lynch Syndrome in Patients with Endometrial Carcinoma. *Surg Pathol Clin.* 2016;9(2):289-99.
15. Stelloo E, Jansen AML, Osse EM, Nout RA, Creutzberg CL, Ruano D, et al. Practical guidance for mismatch repair-deficiency testing in endometrial cancer. *Ann Oncol.* 2017;28(1):96-102.
16. Mills AM, Longacre TA. Lynch Syndrome Screening in the Gynecologic Tract: Current State of the Art. *Am J Surg Pathol.* 2016;40(4):e35-44.
17. Bartosch C, Clarke B, Bosse T. Gynaecological neoplasms in common familial syndromes (Lynch and HBOC). *Pathology.* 2018;50(2):222-37.
18. NCCN Clinical Practice Guidelines in Oncology: Uterine Neoplasms [Internet]. 2017 [cited 6 January 2018].

19. Colombo N, Creutzberg C, Amant F, Bosse T, Gonzalez-Martin A, Ledermann J, et al. ESMO-ESGO-ESTRO Consensus Conference on Endometrial Cancer: diagnosis, treatment and follow-up. *Ann Oncol.* 2016;27(1):16-41.
20. Crosbie EJ RN, Arends M, Bosse T, Burn J, Cornes JM, Crawford R, Eccles D, Frayling IM, Ghaem-Maghani S, Hampel H, Kauff ND, Kitchener HC, Kitson SJ, Manchanda R, McMahon RFT, Monahan KJ, Menon U, Moller P, Moeslein G, Rosenthal A, Sasieni P, Seif MW, Singh N, Skarrott P, Snowsill TM, Steele R, Tischkowitz M, Manchester International Consensus Group, Evans DG. . The Manchester International Consensus Group Recommendations for the Management of Gynecological Cancers in Lynch Syndrome. . *Genetics in Medicine.* 2019;accepted for publication.
21. Cho KR, Cooper K, Croce S, Djordevic B, Herrington S, Howitt B, et al. International Society of Gynecological Pathologists (ISGyP) Endometrial Cancer Project: Guidelines From the Special Techniques and Ancillary Studies Group. *Int J Gynecol Pathol.* 2019;38 Suppl 1:S114-S22.
22. Singh N, Hirschowitz L, Zaino R, Alvarado-Cabrero I, Duggan MA, Ali-Fehmi R, et al. Pathologic Prognostic Factors in Endometrial Carcinoma (Other Than Tumor Type and Grade). *Int J Gynecol Pathol.* 2019;38 Suppl 1:S93-S113.
23. Kalloger SE, Allo G, Mulligan AM, Pollett A, Aronson M, Gallinger S, et al. Use of mismatch repair immunohistochemistry and microsatellite instability testing: exploring Canadian practices. *Am J Surg Pathol.* 2012;36(4):560-9.
24. Mascarenhas L, Shanley S, Mitchell G, Spurdle AB, Macrae F, Pachter N, et al. Current mismatch repair deficiency tumor testing practices and capabilities: A survey of Australian pathology providers. *Asia Pac J Clin Oncol.* 2018;14(6):417-25.
25. Le DT, Uram JN, Wang H, Bartlett BR, Kemberling H, Eyring AD, et al. PD-1 Blockade in Tumors with Mismatch-Repair Deficiency. *N Engl J Med.* 2015;372(26):2509-20.
26. Cancer Genome Atlas Research N, Kandoth C, Schultz N, Cherniack AD, Akbani R, Liu Y, et al. Integrated genomic characterization of endometrial carcinoma. *Nature.* 2013;497(7447):67-73.
27. Talhouk A, McConechy MK, Leung S, Li-Chang HH, Kwon JS, Melnyk N, et al. A clinically applicable molecular-based classification for endometrial cancers. *Br J Cancer.* 2015;113(2):299-310.
28. Stelloo E, Nout RA, Osse EM, Jurgenliemk-Schulz IJ, Jobsen JJ, Lutgens LC, et al. Improved Risk Assessment by Integrating Molecular and Clinicopathological Factors in Early-stage Endometrial Cancer-Combined Analysis of the PORTEC Cohorts. *Clin Cancer Res.* 2016;22(16):4215-24.
29. Chui MH, Gilks CB, Cooper K, Clarke BA. Identifying Lynch syndrome in patients with ovarian carcinoma: the significance of tumor subtype. *Adv Anat Pathol.* 2013;20(6):378-86.
30. Chui MH, Ryan P, Radigan J, Ferguson SE, Pollett A, Aronson M, et al. The histomorphology of Lynch syndrome-associated ovarian carcinomas: toward a subtype-specific screening strategy. *Am J Surg Pathol.* 2014;38(9):1173-81.
31. Ryan NAJ, Bolton J, McVey RJ, Evans DG, Crosbie EJ. BRCA and lynch syndrome-associated ovarian cancers behave differently. *Gynecol Oncol Rep.* 2017;22:108-9.
32. Espinosa I, Jose Carnicer M, Catusus L, Canet B, D'Angelo E, Zannoni GF, et al. Myometrial invasion and lymph node metastasis in endometrioid carcinomas: tumor-associated macrophages, microvessel density, and HIF1A have a crucial role. *Am J Surg Pathol.* 2010;34(11):1708-14.
33. Crosbie EJ, Ryan NAJ, Arends MJ, Bosse T, Burn J, Cornes JM, et al. The Manchester International Consensus Group recommendations for the management of gynecological cancers in Lynch syndrome. *Genet Med.* 2019.
34. Mensenkamp AR, Vogelaar IP, van Zelst-Stams WA, Goossens M, Ouchene H, Hendriks-Cornelissen SJ, et al. Somatic mutations in MLH1 and MSH2 are a frequent

cause of mismatch-repair deficiency in Lynch syndrome-like tumors. *Gastroenterology*. 2014;146(3):643-6 e8.

35. Mills AM, Sloan EA, Thomas M, Modesitt SC, Stoler MH, Atkins KA, et al. Clinicopathologic Comparison of Lynch Syndrome-associated and "Lynch-like" Endometrial Carcinomas Identified on Universal Screening Using Mismatch Repair Protein Immunohistochemistry. *Am J Surg Pathol*. 2016;40(2):155-65.

36. Malpica A, Euscher ED, Hecht JL, Ali-Fehmi R, Quick CM, Singh N, et al. Endometrial Carcinoma, Grossing and Processing Issues: Recommendations of the International Society of Gynecologic Pathologists. *Int J Gynecol Pathol*. 2019;38 Suppl 1:S9-S24.

37. Downes MR, Allo G, McCluggage WG, Sy K, Ferguson SE, Aronson M, et al. Review of findings in prophylactic gynaecological specimens in Lynch syndrome with literature review and recommendations for grossing. *Histopathology*. 2014;65(2):228-39.