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SCHOLARENA

Journal of Gynecology and Women Healthcare

RESEARCH ARTICLE

ISSN: 2639-7285

DNA Mismatch Repair Deficiency in Vietnamese Endometrial Carcinoma, Association with Histopathologic Parameters, One View from Single Institute in Vietnam

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Citation: : Pham Thi Han, Le Hai Giang, Nguyen Thuy Huong, Mai Thi Nhung, Nguyen Ngoc Duong (2023) DNA Mismatch Repair Deficiency in Vietnamese Endometrial Carcinoma, Association with Histopathologic Parameters, One View from Single Institute in Vietnam. J Gynecology Women Health care 4: 104

Abstract

Introduction: DNA mismatch repair deficiency (MMRd), leading to microsatellite instability (MSI), is the primary molecular feature of one over four molecular subgroups in endometrial carcinoma (EC). MSI, the second biggest group (accounting for 25-35% of all), was supposed to have associations with endometrioid carcinoma, high-grade tumor and tumor-infiltrating lymphocyte (TIL). This subgroup has an intermediate prognosis, worse than the POLE mutation but better than the p53 mutation. Prognosis stratification and adjuvant treatment planning for MSI patients was a significant challenge. Tumor budding, a microscopic feature, was expected to take a role in the story.

Objective: To determine MMRd incidence and review the relationship between MMRd and histopathologic features, including tumor budding.

Methods: 81 patients with endometrial carcinoma were evaluated by H&E and IHC (4 markers: MLH1, MSH2, MSH6, PMS2).

Results: MMRd status accounted for 44.4%. Loss of expression was observed the most commonly in PMS2 (34.6%), followed by MLH1 (18.5%), MSH6 (12.3%), and MSH2 (9.9%). There were no significant differences for MMRd cases in histopathological type, FIGO stage,lymphovascular space invasion (LVSI), tumor lymphocyte infiltration (TIL), and tumor budding status, except for histologic grade.

Conclusion: The MMRd group accounted for a high incidence rate (44.4%). MMRd status was associated with histologic grade 2 (p=0.026). The correlation between MMRd with histopathological type, FIGO stage, tumor infiltration lymphocyte,

and tumor budding status was not statistically confirmed.

Keywords: DNA Mismatch Repair Deficiency; Endometrial Carcinoma; Pathologic Parameters

Introduction

Endometrial cancer is the second most common (just behind the cervix) and the fourth leading cause of death due to gynecological cancers among women [1]. However, thanks to HPV vaccination and Pap test screening program, cervix cancer have decreased worldwide. While endometrial cancer has been increasing for 30 years, making it a common health problem in women that needs special attention [2].

In 2013, The Cancer Genome Atlas (TCGA) project performed an integrated genomic, transcriptomic, and proteomic characterization of 373 endometrial carcinomas, thereby classifying endometrial cancers into four subgroups with different prognoses: POLE mutation has the best prognosis, P53 mutation has the worst prognosis, microsatellite instability (MSI) and no specific molecular profile (NSMP) have an intermediate prognosis [3]. Molecular classification is one of the most important discoveries of endometrial cancer in recent years, and it will change the way we stage, risk stratify, and treat patients.

Microsatellite instability (MSI) is the occurrence of mutations in microsatellite regions; DNA segments contain 1-5 nucleotide pairs long that repeat throughout the genome. MSI is the second largest subgroup, accounting for 25-35% of four molecular groups of TCGA[3]. With the IHC classifier, deficiency of mismatch repair protein (MMRd), equivalent to the DNA microsatellite instability (MSI), defined by the loss of tumor nuclear expression of at least one of the four mismatch repair proteins (MLH1, PMS2, MSH6, and PMS2 [4]). The MSI/MMRd group has an intermediate prognosis, worse than the POLE mutation but better than the p53 mutation [5]. In particular, this group also has the potential for immunotherapy with PD-1 inhibitors [6].

However, scientists realized that MSI/MMRd is not a homogeneous group. The MSI cases related to germline mutations (in hereditary Lynch syndrome) and somatic mutations have different prognoses and treatment responses [7]. Prognostic stratification in the MSI/MMRd group is a significant challenge. Currently, assessing prognosis and planning adjuvant treatment for patients in this group is mainly based on histology, stage, and lymphovascular space invasion (LVSI) [8].

Some microscopic histopathological features are also used to support the prognostic stratification, one of which is receiving increasing attention: tumor budding. Tumor budding is defined as a cluster of less than four tumor cells appearing on the invasive area of the tumor [9]. It has been proven to be a poor prognostic factor in many types of cancer, such as colorectal, stomach, breast, pancreas, and squamous cell carcinoma of the head, face, and neck. However, evaluating this feature has not been common in endometrial cancer yet.

Methods

Patient selection: 81 endometrial carcinomas at K Hospital from 2020-2023

- Patients underwent total oophorohysterectomy.
- Diagnosis as one histopathological type of endometrial carcinoma, according to the 2020 World Health Organization (WHO) classification of female reproductive system tumors.
- Formalin-fixed, paraffin-embedded (FFPE) tissues are sufficient in quantity and quality to stain HE and IHC with four antibodies: MLH1, MSH2, MSH6, and PMS2.

Exclusion Criteria

- The specimens were curettage.
- Tumors that do not have a primary origin from the endometrium (e.g., spreading upward from the cervix, spreading from the pelvis).
- No complete information about survey characteristics.

IHC study

- Stain the sections with Roche's four antibodies MLH1, MSH2, MSH6, and PMS2 on Roche's BechmarkXT automated immunohistochemical staining system. On each slide, there were positive controls. Positive control was a pre-qualified endometrial tissue with a MMR status of intact. Negative controls were stained in separate slides. A species-matched negative control antibody was used to evaluate the presence of background in test samples and establish a staining intensity baseline. We evaluated the expression of antibodies on tumor cell nuclei, compared with internal control tissues (epithelial cells, endothelial cells, lymphocytes, fibroblasts), negative and positive control tissues.
- Expression of mismatch repair proteins, including MLH1, PMS2, MSH2, and MSH6, was evaluated by standard from The College of American Pathologists (CAP) Cancer Protocol (version June 2021) [10].
- Mismatch repair deficiency (MMRd) is determined when tumor cell nuclei lost >10% expression of at least one marker among MLH1, MSH2, MSH6, and PMS2 [11].
- Mismatch repair proficient (MMRp) is characterized by nuclear staining for all four MMR proteins.

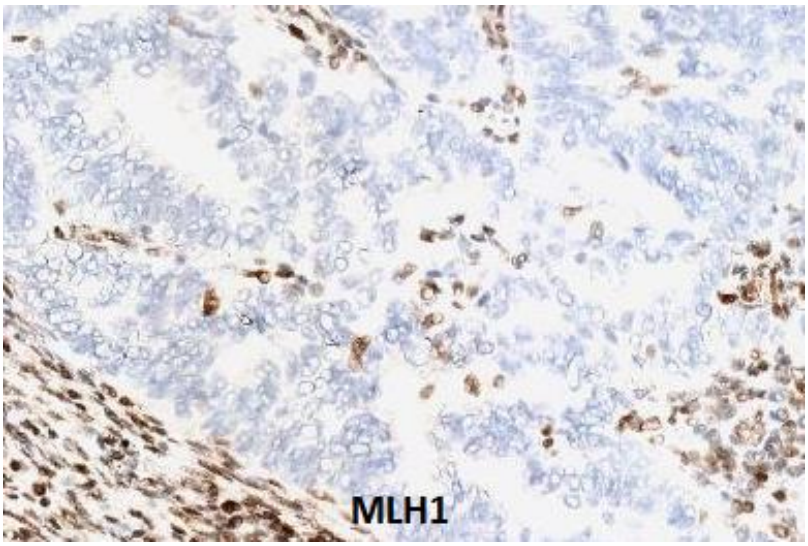

Figure 1: Loss of expression of MLH1. Tumor cell nuclei do not stain despite of the prominent positivity of internal control. (IHC, 400x)

Histopathological Features on H&E

Features were evaluated consistently according to these following standards.

- Histologic type, histologic grade: based on the World Health Organization's classification of tumors of the female genital system (WHO, 2020) [12].

- Staging: based on International Federation of Gynecology and Obstetrics (FIGO 2018) [13].
- Tumor-infiltrating lymphocyte (TIL): based on research "Assessing Tumor-Infiltrating Lymphocytes in Solid Tumors: A Practical Review for Pathologists and Proposal for a Standardized Method" from the InternationalImmuno-Oncology Biomarkers Working Group [14].
- Tumor budding: based on Stögbauer et al in research "Independent Tissue-Based Biomarkers in Endometrioid Endometrial Cancer: Tumor Budding in Microsatellite Instability and WHO Grading in Copy-Number-Low Patients" [15].

The endometrial cancers were immunostained at Vietnam National Cancer Hospital – one of the leading hospitals in Vietnam specializing in Oncology. Two pathologists examined all the sections separately, and a complete agreement was reached on the IHC diagnosis of defective MMR in the relevant cancers, thus considerably strengthening the IHC evidence base for this investigation with independent confirmation.

Results

Table 1: Overall patients characteristics

Feature		n	%
Age (years)	Range: 36-80	Average: 57.7 ± 8.8	
Stage	IA	47	58.0
	IB	18	22.2
	II	4	4.9
	III	9	11.1
	IV	3	3.7
Histopathologic type	Endometrioid	67	82.7
	Serous	9	11.1
	Clear cell	1	1.2
	Mixed	3	3.7
	Carcinosarcoma	1	1.2

A total of 81 Vietnamese patients with a diagnosis of endometrial carcinoma were selected for analysis. Patients ranged from 36 to 80 years old, with an average age of 57.68. The more detailed information is summarized in Table 1. Most patients were in the early stages of IA (47/81 cases, 58%) and IB (18/81 cases, 22.2%). More extensive stages II, III, and IV accounted for only 4.9%, 11.1%, and 3.7% sequentially. The most common histopathological type was endometrioid carcinoma (67/81 patients, 82.7%), followed by serous carcinoma (9 patients, 11.1%). Clear cell carcinoma, mixed carcinoma, and carcinosarcoma appeared to be much more rare.

Table 2: MMR protein expression status

MMR status	MLH1	PMS2	MSH2	MSH6	n (%)
Loss of 1 protein	-	+	+	+	0 (0)
	+	-	+	+	14 (17.3)
	+	+	-	+	0 (0)
	+	+	+	-	2 (2.4)
Loss of 2 proteins	-	-	+	+	11 (13.6)
	+	+	-	-	5 (6.2)
	-	+	-	+	1 (1.2)
Loss of 3 proteins	-	-	+	-	1 (1.2)
Loss of 4 proteins	-	-	-	-	2 (2.5)
Total, n (%)	15 (18.5)	28 (34.6)	8 (9.9)	10 (12.3)	36 (44.4)

((+) positive IHC; (-) lost of expression in IHC)

In this study, 36/81 patients had abnormal MMR protein expression (lost at least one or more protein expression), accounting for 44.4%. The rate of PMS2 loss was highest, accounting for 34.6%, followed by MLH1 18.5%, and then MSH6 and MSH2 (12.3% and 9.9%, respectively). Protein loss often occurred in pairs: MLH1 paired with PMS2 (11 cases, 13.6%) and MSH2 paired with MSH6 (5 cases, 6.2%). One case lost three proteins, and two lost all four proteins. Isolated loss of PMS2 was significantly high (14 cases, 17.3%).

Table 3: Association between MMR protein expression status and some pathological features

Features		MMR status			MLH1			PMS2			MSH2			MSH6		
		MMRp	MMRd	p	+	-	p	+	-	p	+	-	p	+	-	p
N (%)		**45 (55.6)**	**36 (44.5)**		**66 (81.5)**	**15 (18.5)**		**53 (65.4)**	**28 (34.6)**		**73 (90)**	**8 (10)**		**71 (87.7)**	**10 (12.3)**	
Histologic type	Endometrioid	34 (75.6)	33 (91.7)	0.057	53 (80.3)	14 (93.3)	0.448	40 (75.5)	27 (96.4)	0.028	61 (83.6)	6 (75)	0.621	59 (83.1)	8 (80)	0.681
	Non-endometrioid	11 (24.4)	3 (8.3)		13 (19.7)	1 (6.7)		13 (24.5)	1 (3.6)		12 (16.4)	2 (25)		12 (16.9)	2 (20)	
FIGO stage	IA	23 (51.1)	24 (66.7)	0.747	37 (56.1)	10 (66.7)	0.98	25 (47.2)	22 (78.6)	0.098	43 (58.9)	4 (50)	0.515	42 (59.2)	5 (50)	0.592
	IB	11 (24.4)	7 (19.4)		15 (22.7)	3 (20)		15 (28.3)	3 (10.7)		16 (21.9)	2 (25)		15 (21.1)	3 (30)	
	II	3 (6.7)	1 (2.8)		4 (6.1)	0 (0)		3 (5.7)	1 (3.6)		4 (5.5)	0 (0)		4 (5.6)	0 (0)	
	III	6 (13.3)	3 (8.3)		7 (10.6)	2 (13.3)		7 (13.2)	2 (7.1)		8 (11)	1 (12.5)		8 (11.3)	1 (10)	
	IV	2 (4.4)	1 (2.8)		3 (4.5)	0 (0)		3 (5.7)	0		2 (2.7)	1 (12.5)		2 (2.8)	1 (10)	
Histologic grade	1	14 (31.1)	6 (16.7)	0.026	20 (30.3)	0 (0)	0.019	14 (26.4)	6 (21.4)	0.23	20 (27.5)	0 (0)	0.247	20 (28.2)	0 (0)	0.081
	2	14 (31.1)	22 (61.1)		26 (39.4)	10 (66.7)		20 (37.7)	16 (57.1)		31 (42.5)	5 (62.5)		29 (40.8)	7 (70)	
	3	17 (37.8)	8 (22.2)		20 (30.3)	5 (33.3)		19 (35.8)	6 (21.4)		22 (30.1)	3 (37.5)		22 (31)	3 (30)	
LVSI	No LVSI	26 (57.8)	23 (63.9)	0.556	41 (62.1)	8 (53.3)	0.254	30 (56.6)	19 (67.9)	0.281	46 (63)	3 (37.5)	0.172	45 (63.4)	4 (40)	0.241

	Focal LVSI (<5)	10 (22.2)	9 (25)		13 (19.7)	6 (40)		12 (22.6)	7 (25)		15 (20.5)	4 (50)		16 (22.5)	3 (30)	
	Extensive LVSI (≥5)	9 (20)	4 (11.1)		12 (18.2)	1 (6.7)		11 (20.8)	2 (7.1)		12 (16.4)	1 (12.5)		10 (14.1)	3 (30)	
TIL	TIL	21 (46,7)	23 (63,9)	0.122	35 (53)	9 (60)	0.776	29 (54.7)	15 (53.6)	1.000	37 (50.7)	7 (87.5)	0.065	35 (49.3)	9 (90)	0.018
	No TIL	23 (53,3)	13 (36,1)		31 (47)	6 (40)		24 (45.3)	13 (46.4)		36 (49.3)	1 (12.5)		36 (50.7)	1(10)	
	No adenomyosis	35 (77.8)	25 (69.4)		51 (77.3)	9 (60)		42 (79.2)	18 (64.3)		54 (74)	6 (75)		52 (73.2)	8 (80)	
Tumor budding	Low grade	28 (62.2)	28 (77.8)	0.132	46 (69.3)	10 (66.7)	1.000	34 (64.2)	22 (78.6)	0.181	51 (69.9)	5 (62.5)	0.697	49 (69)	7 (70)	1.000
	High grade	17 (37.8)	8 (22.2)		20 (30.3)	5 (33.3)		19 (35.8)	6 (21.4)		22 (30.1)	3 (37.5)		22 (31)	3 (30)	

(MMR: mismatch repair protein, MMRd: mismatch repair deficiency; MMRp: mismatch repair proficient; LVSI: Lymphovascular space invasion)

Patients with mismatch repair proficient (MMRp) were compared with those with abnormal MMR protein expression cases (MMRd, MLH1 loss, MSH2 loss, MSH6 loss, PMS2 loss). The two groups MMRd and MMRp are both mainly endometrial carcinomas, at low stages (I and II). There were no significant differences for MMRd cases in histopathological type (p=0.057), FIGO stage (p=0.747). Compared with the MMRd group, MMRd tumors mainly have low histological grade (mainly grade 2) (p=0.026). Regarding LVSI, TIL and tumor budding, no association was found between these features and MSI (p=0.556; p=0.122; p=0.132). MLH1 deficiency tumor associated with histologic grade 2 (p=0.019). PMS2 deficiency tumor associated with endometrioid carcinoma (p=0.028).

Discussion

The development of molecular classification in endometrial cancer has opened a new era for oncologists to stage, prognosis, and treat patients more sufficiently. This fact also directly poses a challenge for pathologists to decide MSI status in endometrial cancer. However, in Vietnam, data concerning MSI status in endometrial carcinoma are sparse, while most studies have focused on patients with colorectal cancers. In the present study, we evaluated the proportion of MMRd status and the correlation between MSI phenotype and the various histologic parameters.

Evaluating MSI status in endometrial cancers has currently been validated in standardized guidelines [10]. Using IHC with 4 MMR protein markers is a fast, simple technique, low cost, and available in many facilities. Another advantage of this method is the ability to detect precisely the abnormal protein among MLH1, MSH2, MSH6, or PMS2 [4]. According to previous studies, the sensitivity, specificity, positive predictive, and negative predictive values using IHC were reported to be 100%, 86.1%, 58.3%, and 100%, respectively [16]. Furthermore, a direct comparison between the MSI-H genetic test and the IHC test identifying MMRd in endometrial cancer showed high similarity (93%) [17]. IHC test for MMRd combined with family/personal history allows screening for patients at high risk of hereditary Lynch syndrome, who would be directed for further tests to do.

David S. Guttery et al proved that, there was a significantly higher frequency of somatic mutations in DNA mismatch repair genes in Asian tumours, in particular PMS2 (p=0.0036) [18]. Studies on Asian ethnicity have reported MMRd rate detected by IHC ranging from 19% to 55%, which appeared to be higher than the data from Western community studies (Table 4). The frequency of MMRd observed in this study (44,4%) was similar to those performed on Asian races: Korean [19] (p=0.113), Japanese [20] (p=0.24), Turkish [21] (p=0.445), and Pakistani [22] (p=0.54). This result proved that MMRd status in endometrial carcinoma in Vietnamese people was similar to other Asian ethnicity.

Table 4: Research on Asian ethnicity

Researchers	Published year	Ethnicity	Method	Sample size(n)	MMRd/MSI-H(n)	MMRd/MSI-H (%)
Yin Ling Woo et al. [23]	2014	Malaysian, Chinese, Indian	IHC	77	15	19.4
Yunfeng Song et al. [24]	2021	Chinese	IHC	99	29	29
Joseph J. Noh et al. [19]	2021	Korean	IHC and PCR	373	139	37.3
Masafumi Kato et al. [20]	2014	Japanese	IHC	191	76	40
Aysun Fırat et al. [21]	2023	Turkish	IHC	44	19	43.1
Atif Ali Hashmi et al. [22]	2019	Pakistani	IHC	126	56	44.4
Siriwan Tangjitgamol et al. [25]	2017	Thai	IHC	385	212	55.1

Research by Joehlin Price and colleagues reported the same findings in 1049 endometrial carcinomas as PMS2 was the most common deficiency with 186 cases (17.7%), followed by MLH1, MSH6, MSH2 (15.7%, 4.9%, and 1.9%) [26]. Guttery et al proved that there was a significantly higher frequency of somatic mutations of PMS2 in Asian tumors (p=0.0036) [18]. However, due to insufficient data, larger studies need to be done for confirmation.

Our research also reported a significant rate of isolated loss of PMS2 (17.3%), much higher than findings from Joehlin Price's study with only 21/1049 cases (accounting for 2.0%) (p< 0.001) [26]. Surprisingly, single loss of MLH1 expression was not observed in any included patient, but that MLH1 was observed at the second highest expression rate among all markers. Kurpiel et al proved that double loss of MLH1 and PMS2 due to overmethylation of the promoter region of the MLH1 gene is the leading cause of MSI in endometrial carcinoma [27]. Perhaps the fact that MLH1 often lost expression along with PMS2 explains why our study did not record any cases of single loss of expression of MLH1.

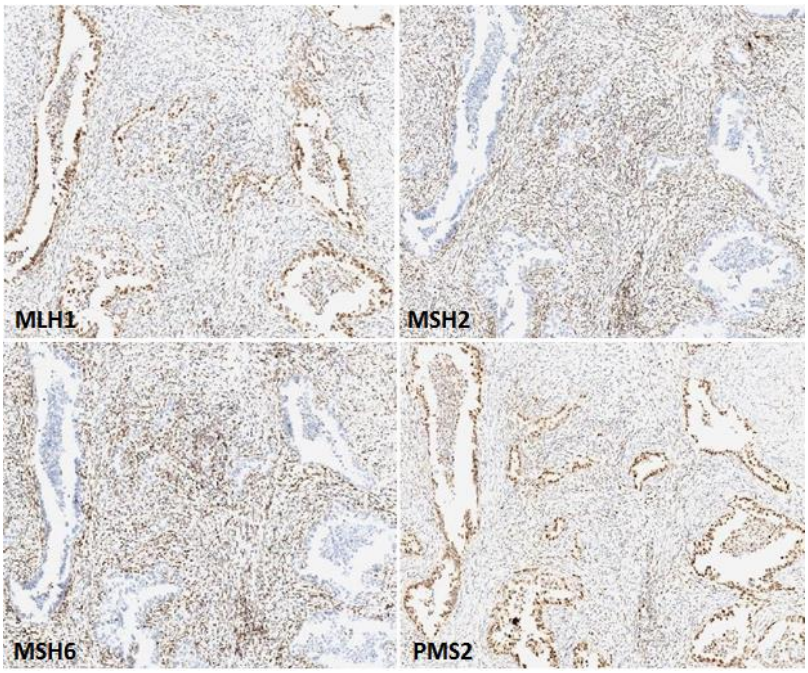

Figure 2 : A case loss of MSH2 and MSH6 expression, while MLH1 and PMS2 were retained. (IHC, 200x)

Histopathological Type

Previous studies in Asian countries reported that about 87% to 92% of MMRd cases were endometrioid; non-endometrioid cases included serous, clear cell, undifferentiated, and carcinosarcoma [20,25]. This study shared the same result, as the endometrioid type remains the primary histopathological type in MMRd group.

Stage and Grade

This study showed no significant differences for MMRd and MMRp cases in the FIGO stage (0.747) but revealed an association in the histological grade. MMRd tumor related to lower histological grade, especially grade 2 (p=0.026). Previous studies have reported inconsistent results regarding the correlation between MMRd status and tumor pathological features. Price et al concluded that there was no correlation between MMRd and disease stage or histological grade [26]. Other studies have found that the group of patients with MMRd often has the histopathological type of endometrioid and favorable pathological characteristics: earlier stage, low histological grade, and no lymph node metastasis [20,25,28]. However, McMeekin et al have demonstrated that the MMRd group is often associated with endometrioid type, high histological grade, deeper muscle invasion, more frequent LVSI, and advanced-stage [29].

Lymphovascular Space Invasion (LVSI)

According to research by Bosse et al., the presence of a lymphovascular space invasion did not have a significant effect on the prognosis, but the presence of extensive lymphovascular space invasion (≥5 vessels) was related to high incidence of nodal and distant metastasis [30]. Vermij et al also reported that extensive LVSI increased the risk of local recurrence or distant metastasis regardless of stage and histological type, so it was an independent prognostic factor [4]. Recently in June 2023, The International Federation of Gynecology and Obstetrics (FIGO) began to include LVSI features in the disease staging table, highlighting the significant value of LVSI in endometrial cancer [31].

The fact that we did not found any relationship between LVSI and MSI contrasted with McMeekin et al result, which reported an association between the presence of LVSI (especially extensive LVSI) and MSI. The difference may be due to limitations in sample size and sampling method, as well as geographical and racial characteristics.

Tumor-Infiltrating Lymphocytes (TIL)

Tumor infiltrating lymphocyte (TIL) is defined as the appearance of lymphocyte within tumor cells. TIL was first determined at low magnification to find hotspot region, then was counted on 10 high power fields. Only lymphocytes located within the boundary of tumor cell nests or glands were counted. Apoptotic bodies were discounted. According to Shia et al, number of TIL/10 HPFs ranged from 2 to 745, with a median value of 46. The mean number of TIL was significantly higher in the MSI-H group (94.5/10 HPFs) compared to with non-MSI-H group (60/10 HPFs) ($p = 0.002$). Using cut-off point of 40 TIL/10 HPFs, the sensitivity was 85% and the specificity was 46% [32]. When increasing the point, the specificity increases but the sensitivity decreases. Therefore, 40 TIL/10 HPFs was recommended by Hendry to have good sensitivity and specificity, suitable for detecting MSI status in endometrial carcinoma [14].

In our study, 32/81 (39.5%) patients had TIL. However, there have not been many studies using this cut-off as there is no official recommendation. Due to lack of standardization, many studies still preferred using a continuous parameter. 63,8% MMRd group had TIL, while this number in MMRp group was 47,7% , seeming to be lower. However, the Chi-square test revealed no difference between the two groups ($p = 0.122$, with 95% confidence).

TIL will be activated and cause tumor cell toxicity, leading to a better prognosis. However, this process is blocked by the PD1-PDL1 pathway, which controls the induction and maintenance of immune tolerance within the tumor microenvironment. If this

pathway is eliminated, the anti-cancer immune response will progress powerfully. This mechanism explains why MSI/MMRd tumors often have good responses to anti-PD1 drugs, typically Pembrolizumab [33].

Tumor Budding

Due to the characteristics of endometrial cancer, Stogbauer et al have developed a 2-level tumor budding classification scale to replace the 3-level classification scale for colorectal cancer developed by the National Consensus Conference on budding. According to Stögbauer, tumor budding in endometrial cancer has two levels: low-grade tumor budding (0-2 buds) and high-grade tumor budding (≥3 buds) on a high-power field (x400 magnification). High-grade tumor budding (≥3 buds) is an independent prognostic factor associated with poor prognosis and lymph node metastasis [15].

To date, no study has reported an association between tumor budding and MSI. Our Our study also did not record any correlation between two features. However, as an independent prognostic factor, tumor budding can be an amazing feature supporting prognostic stratification of MSI/MMRd groups more effectively. Therefore, further studies need to be done for unifying the way to assess tumor budding in clinical practice of endometrial cancer.

Conclusion

Through studying the characteristics on H&E and IHC stains with 4 MMR markers in 81 endometrial cancers, we draw the following conclusions:

The MMRd group accounts for 44.4%, with the highest rate of loss of expression being PMS2 (34.6%), followed by MLH1 (18.5%), MSH6 (12.3%), and MSH2 (9, 9%).

MMRd status was associated with histologic grade 2 (p=0.026). The correlation between MMRd with histopathological type, FIGO stage, lymphovascular space invasion (LVSI), tumor infiltration lymphocyte (TIL), and tumor budding status was not statistically confirmed.

References

1. Global Cancer Observatory (2022) https://gco.iarc.fr/

2. Rising Endometrial Cancer Rates Spur New Approaches to Prevention (2022) Division of Cancer Prevention.

3. Cancer Genome Atlas Research Network, Kandoth C, Schultz N, et al. (2013) Integrated genomic characterization of endometrial carcinoma. Nature 497: 67-73.

4. Vermij L, Smit V, Nout R, Bosse T (2020) Incorporation of molecular characteristics into endometrial cancer management. Histopathology 76: 52-63.

5. Ten-year results of the PORTEC-2 trial for high-intermediate risk endometrial carcinoma (2022) improving patient selection for adjuvant therapy.

6. Marcus L, Lemery SJ, Keegan P, Pazdur R (2019) FDA Approval Summary: Pembrolizumab for the Treatment of Microsatellite Instability-High Solid Tumors. Clinical Cancer Research 25: 3753-8.

7. Pakish JB, Zhang Q, Chen Z, et al. (2017) Immune Microenvironment in Microsatellite-Instable Endometrial Cancers: Hereditary or Sporadic Origin Matters. Clinical Cancer Research 23: 4473-81.

8. Concin N, Matias-Guiu X, Vergote I, et al. (2021) ESGO/ESTRO/ESP guidelines for the management of patients with endometrial carcinoma. Int J Gynecol Cancer 31: 12-39.

9. Lugli A, Kirsch R, Ajioka Y, et al. (2017) Recommendations for reporting tumor budding in colorectal cancer based on the International Tumor Budding Consensus Conference (ITBCC) 2016. Mod Pathol 30: 1299-311.

10. Bartley AN, Mills AM, Konnick E, et al. (2022) Mismatch Repair and Microsatellite Instability Testing for Immune Checkpoint Inhibitor Therapy: Guideline From the College of American Pathologists in Collaboration With the Association for Molecular Pathology and Fight Colorectal Cancer. Archives of Pathology & Laboratory Medicine 146: 1194-210.

11. Stelloo E, Jansen AML, Osse EM, et al. (2017) Practical guidance for mismatch repair-deficiency testing in endometrial cancer. Annals of Oncology 28: 96-102.

12. WHO (2022) Classification of Tumours Editorial Board. Endometrioid carcinoma of the uterine corpus. In: Vol Female Genital Tumours. 5th ed. WHO Classification of Tumours. World Health Organization 252-5.

13. Amant F, Mirza MR, Koskas M, Creutzberg CL (2018) Cancer of the corpus uteri. International Journal of Gynecology & Obstetrics 143: 37-50.

14. Hendry S, Salgado R, Gevaert T, et al. (2017) Assessing Tumor-Infiltrating Lymphocytes in Solid Tumors: A Practical Review for Pathologists and Proposal for a Standardized Method from the International Immuno-Oncology Biomarkers Working Group: Part 2: TILs in Melanoma, Gastrointestinal Tract Carcinomas, Non–Small Cell Lung Carcinoma and Mesothelioma, Endometrial and Ovarian Carcinomas, Squamous Cell Carcinoma of the Head and Neck, Genitourinary Carcinomas, and Primary Brain Tumors. Advances in Anatomic Pathology 24: 311-35.

15. Stogbauer F, Geß B, Brambs C, et al. (2023) Independent Tissue-Based Biomarkers in Endometrioid Endometrial Cancer: Tumor Budding in Microsatellite Instability and WHO Grading in Copy-Number-Low Patients. Cancers 15: 3832.

16. Ferguson SE, Aronson M, Pollett A, et al. (2014) Performance characteristics of screening strategies for Lynch syndrome in unselected women with newly diagnosed endometrial cancer who have undergone universal germline mutation testing. Cancer 120: 3932-9.

17. McConechy MK, Talhouk A, Li-Chang HH, et al. (2015) Detection of DNA mismatch repair (MMR) deficiencies by immunohistochemistry can effectively diagnose the microsatellite instability (MSI) phenotype in endometrial carcinomas. Gynecologic Oncology 137: 306-10.

18. Guttery DS, Blighe K, Polymeros K, Symonds RP, Macip S, Moss EL (2018) Racial differences in endometrial cancer molecular portraits in The Cancer Genome Atlas. Oncotarget 9: 17093-103.

19. Noh JJ, Kim MK, Choi MC, et al. (2022) Frequency of Mismatch Repair Deficiency/High Microsatellite Instability and Its Role as a Predictive Biomarker of Response to Immune Checkpoint Inhibitors in Gynecologic Cancers. Cancer Res Treat 54: 1200-8.

20. Kato M, Takano M, Miyamoto M, et al. (2015) DNA mismatch repair-related protein loss as a prognostic factor in endometrial cancers. J Gynecol Oncol 26: 40.

21. FìRat A (2023) Microsatellite instability (MSI) and p16/p53 protein status in different subtypes of endometrial carcinoma: with emphasis on tumor aggressiveness. J Cukurova Anesth Surg 6: 338-41.

22. Hashmi AA, Mudassir G, Hashmi RN, et al. (2019) Microsatellite Instability in Endometrial Carcinoma by Immunohistochemistry, Association with Clinical and Histopathologic Parameters. Asian Pac J Cancer Prev 20: 2601-6.

23. Woo YL, Cheah PL, Shahruddin SI, Omar SZ, Arends M (2014) The Immunohistochemistry Signature of Mismatch Repair (MMR) Proteins in a Multiethnic Asian Cohort With Endometrial Carcinoma: International Journal of Gynecological Pathology 33: 554-9.

24. Endometrial Tumors with MSI-H and dMMR Share a Similar Tumor Immune Microenvironment.

25. Tangjitgamol S, Kittisiam T, Tanvanich S (2017) Prevalence and prognostic role of mismatch repair gene defect in endometrial cancer patients. Tumour Biol 39: 101042831772583.

26. Joehlin-Price AS, Perrino CM, Stephens J, et al. (2014) Mismatch repair protein expression in 1049 endometrial carcinomas, associations with body mass index, and other clinicopathologic variables. Gynecologic Oncology 133: 43-7.

27. Kurpiel B, Thomas MS, Mubeen M, et al. (2022) MLH1/PMS2-deficient Endometrial Carcinomas in a Universally Screened Population: MLH1 Hypermethylation and Germline Mutation Status. Int J Gynecol Pathol 41: 1-11.

28. Jumaah AS, Al-Haddad HS, Salem MM, McAllister KA, Yasseen AA (2021) Mismatch repair deficiency and clinicopathological characteristics in endometrial carcinoma: a systematic review and meta-analysis. J Pathol Transl Med 55: 202-11.

29. McMeekin DS, Tritchler DL, Cohn DE, et al. (2016) Clinicopathologic Significance of Mismatch Repair Defects in Endometrial Cancer: An NRG Oncology/Gynecologic Oncology Group Study. J Clin Oncol 34: 3062-8.

30. Bosse T, Peters EEM, Creutzberg CL, et al. (2015) Substantial lymph-vascular space invasion (LVSI) is a significant risk factor for recurrence in endometrial cancer--A pooled analysis of PORTEC 1 and 2 trials. Eur J Cancer 51: 1742-50.

31. Berek JS, Matias-Guiu X, Creutzberg C, et al. (2023) FIGO staging of endometrial cancer: 2023. International Journal of Gynecology & Obstetrics 162: 383-94.

32. Shia J, Black D, Hummer AJ, Boyd J, Soslow RA (2008) Routinely assessed morphological features correlate with microsatellite instability status in endometrial cancer. Human Pathology 39: 116-25.

33. Eso Y, Shimizu T, Takeda H, Takai A, Marusawa H (2020) Microsatellite instability and immune checkpoint inhibitors: toward precision medicine against gastrointestinal and hepatobiliary cancers. Journal of Gastroenterology 55: 15.