



World Journal of Current Medical and Pharmaceutical Research

Content available at www.saap.org.in

ISSN: 2582-0222



EVALUATION OF MISMATCH REPAIR PROTEIN EXPRESSION AND ITS ASSOCIATION WITH HISTOPATHOLOGICAL FEATURES IN COLORECTAL CARCINOMA: A PROSPECTIVE STUDY

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DOI: <https://doi.org/10.37022/wjcmpr.v8i3.454>

ARTICLE HISTORY	ABSTRACT
Received on: 24-08-2026 Revised on: 11-09-2026 Accepted on: 16-10-2026	Background: Mismatch repair deficiency represents a biologically distinct subset of colorectal carcinoma with diagnostic and therapeutic significance. The present study aimed to assess mismatch repair (MMR) protein expression and its correlation with histopathological features.
Keywords: Colorectal carcinoma; mismatch repair; dMMR; immunohistochemistry; tumour-infiltrating lymphocytes.	Methods: This prospective study included 150 colorectal carcinoma resection specimens, which were evaluated for MLH1, PMS2, MSH2, and MSH6 expression using immunohistochemistry. The clinicopathological features were analysed and compared between deficient mismatch repair (dMMR) and proficient mismatch repair (pMMR) tumours.
Corresponding Author Enoch Raju Kanikella	Results: A total of 30 cases (20.0%) showed dMMR. The most frequent pattern was loss of MLH1/PMS2 expression (60.0% of dMMR cases), followed by loss of MSH2/MSH6 expression. High levels of tumour-infiltrating lymphocytes (TILs) were significantly more common in dMMR than in pMMR tumours (40.0% vs. 16.7%; OR = 3.33; p = 0.005) and were independently associated with dMMR (aOR = 3.85; p = 0.005). Positive but statistically non-significant adjusted associations were observed for right-sided tumour location and mucinous/medullary morphology.
	Conclusion: dMMR was identified in one-fifth of colorectal carcinoma cases and was most strongly associated with prominent lymphocytic infiltration.

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INTRODUCTION

The molecular heterogeneity of colorectal carcinoma is well documented and disruption of the DNA mismatch repair (MMR) system is an important pathway of tumorigenesis. The MMR proteins MLH1, PMS2, MSH2 and MSH6 are normally involved in the recognition and correction of replication errors, and the loss of functional MMR leads to the accumulation of mutations and often to a microsatellite instability-high (MSI-H) phenotype. The MMR deficient colorectal carcinomas are a biologically distinct subgroup that has implications for hereditary cancer assessment, prognosis and treatment selection [1,2]. Immunohistochemistry is a practical approach for assessing the MMR status in routine pathology. Loss of nuclear expression of one or more MMR proteins can identify tumours with defective MMR, and can also help determine which part of the MMR pathway is defective, in the presence of appropriate internal controls. Immunohistochemical assessment has been shown to be highly sensitive and specific when compared to molecular MSI testing [3]. The importance of MMR testing in all

tumours has therefore increased as it is not possible to identify a proportion of patients who require further evaluation for Lynch syndrome based on age, family history or other clinical selection criteria [4]. In addition, there are characteristic, though not exclusive, morphological patterns that are seen in MMR deficient colorectal carcinomas. Pathological studies of these patients have found that microsatellite instability is linked to the location of the tumour in the colon, mucinous differentiation, atypical differentiation, expansive growth pattern, Crohn-like peritumoral lymphoid reaction, and especially to the presence of more tumour infiltrating lymphocytes [5]. Greenon et al. showed that intratumoral lymphocytes were among the most powerful histological markers of MSI, while Alexander et al. found that a combination of tumour morphology and host immune response was helpful as a pathological clue to the MSI phenotype [5,6]. These features are clinically relevant because they will help identify tumours that are likely to be MMR deficient, but their low sensitivity and specificity means that morphology cannot be used to replace direct MMR testing. The determination of

MMR status is not only relevant to morphological classification, but also has a wider scope of application. There have been differences in clinical outcome and response to conventional adjuvant therapy between microsatellite-unstable and microsatellite-stable colorectal cancers [7]. More recently, MMR deficiency is emerging as an important predictive biomarker for immune-checkpoint inhibition, as PD-1 blockade has been shown to be much more active in MMR-deficient tumours than in MMR-proficient tumours [8]. Considering the close relationship between molecular phenotype, tumour morphology and host immune response, the expression of MMR may be useful in addition to the routine histopathological parameters to provide clinically relevant information and aid in the recognition of tumors with dMMR. The present study thus investigated the immunohistochemical expression of MLH1, PMS2, MSH2, and MSH6 in colorectal carcinoma, and the correlation between MMR status and tumour location, histological subtype, grade, stage, invasive characteristics, tumor budding and lymphocytic response.

METHODS

Study design and case selection

This was a prospective observational histopathology study that was carried out for 12 months in 2025 and involved 150 consecutive cases of primary colorectal carcinoma. Eligible were adults aged ≥ 18 years who had been diagnosed with primary colorectal carcinoma (CRC) by histology, had a sufficient amount of viable tumour for mismatch repair (MMR) immunohistochemistry, and had complete histopathological data. Resection specimens were preferentially included to allow for accurate evaluation of depth of invasion, lymph-node status, lymphovascular invasion, perineural invasion and pathological stage. Recurrent colorectal carcinoma, metastatic tumours from another primary site involving the colorectum, inadequate fixation and/or insufficient tissue, extensive necrosis or technical artifact that precluded reliable immunohistochemical interpretation, incomplete essential pathology data, and post-neoadjuvant specimens in which pretreatment morphology could not be reliably assessed were excluded.

Histopathological evaluation

The anatomical location, gross pattern, size, histological type, grade, depth of invasion, lymph-node involvement and pathological stage of the tumours were evaluated. Histological subtypes were conventional, mucinous, signet-ring cell, medullary and other known variants of adenocarcinoma. Mucinous adenocarcinoma was characterized as having $>50\%$ extracellular mucin. Other morphological parameters observed were lymphovascular invasion, perineural invasion, tumour budding, tumour-infiltrating lymphocytes (TIL), Crohn-like lymphoid reaction, tumour-border configuration, necrosis, and desmoplastic response. The tumour site was classified as right-sided, left-sided or rectal/rectosigmoid based on the pre-defined anatomical classification. Pathological T category was determined by the depth of invasion and pathological N category by the involvement of the lymph nodes, while the overall stage was based on pathological T category, pathological N category and documented distant metastasis status.

MMR immunohistochemistry

Immunohistochemistry was carried out on MLH1, PMS2, MSH2 and MSH6. In the presence of appropriate internal non-neoplastic controls, nuclear staining was considered as a positive result in tumor cells. Tumours with nuclear expression of all four proteins were considered proficient MMR (pMMR) and those with the loss of one or more proteins were considered deficient MMR (dMMR). The loss patterns were classified as MLH1/PMS2 loss, MSH2/MSH6 loss, isolated PMS2 loss, or isolated MSH6 loss. The etiology (hereditary or sporadic) was not assigned exclusively by MMR immunophenotype.

Statistical analysis

Data for continuous variables were presented as mean $\pm$ SD or median [IQR] and for categorical variables, as n (%). For continuous variables, the dMMR and pMMR tumors were compared using the Welch's t test or the Mann-Whitney U test, and for categorical variables, using chi-square or Fisher's exact tests, as appropriate. The following univariable logistic regression analyses were performed to identify predictors of dMMR, which were subsequently entered into a parsimonious multivariable model that included age, right-sided location, mucinous/medullary morphology, and high TIL. Odds ratios, 95% confidence intervals and p-values were reported for associations. A receiver operating characteristic (ROC) analysis and area under the curve (AUC) were used to evaluate an exploratory morphology-based model. All tests used were two-tailed and $p < 0.05$ was considered significant.

RESULTS

Clinicopathological characteristics

A total of 150 colorectal carcinoma resection specimens were included. The mean age was 58.8 ± 12.1 years, and 83 (55.3%) patients were male. The overall distribution of tumor site, histological subtype, grade, pathological stage, and major prognostic features is summarized in Table 1. Conventional adenocarcinoma was the predominant subtype, and stage III disease represented the largest stage group.

Table 01: Clinicopathological characteristics of the study population

Characteristic	Overall (N=150)
Age, years	58.8 $\pm$ 12.1
Sex	
Male	83 (55.3%)
Female	67 (44.7%)
Tumor site	
Cecum	15 (10.0%)
Ascending colon	21 (14.0%)
Hepatic flexure	10 (6.7%)
Transverse colon	9 (6.0%)
Splenic flexure	5 (3.3%)
Descending colon	14 (9.3%)
Sigmoid colon	37 (24.7%)
Rectosigmoid	11 (7.3%)
Rectum	28 (18.7%)
Histological type	
Conventional adenocarcinoma	121 (80.7%)
Mucinous adenocarcinoma	15 (10.0%)
Signet-ring cell carcinoma	7 (4.7%)
Medullary carcinoma	4 (2.7%)

Other rare adenocarcinoma subtype	3 (2.0%)
Tumor grade	
Well differentiated	16 (10.7%)
Moderately differentiated	109 (72.7%)
Poorly differentiated	25 (16.7%)
pT stage	
pT1	7 (4.7%)
pT2	28 (18.7%)
pT3	81 (54.0%)
pT4a	27 (18.0%)
pT4b	7 (4.7%)
pN stage	
pN0	78 (52.0%)
pN1	65 (43.3%)
pN2	7 (4.7%)
AJCC stage	
I	21 (14.0%)
II	47 (31.3%)
III	66 (44.0%)
IV	16 (10.7%)
Lymphovascular invasion, present	56 (37.3%)
Perineural invasion, present	49 (32.7%)
Tumor budding	
Low	33 (22.0%)
Intermediate	62 (41.3%)
High	55 (36.7%)
Tumor-infiltrating lymphocytes	
Low	60 (40.0%)
Moderate	58 (38.7%)
High	32 (21.3%)
Crohn-like lymphoid reaction	
Absent	104 (69.3%)
Present	46 (30.7%)
MMR status	
pMMR	120 (80.0%)
dMMR	30 (20.0%)

Values are mean $\pm$ SD or n (%), as appropriate. Indented entries are categories within the preceding characteristic. AJCC, American Joint Committee on Cancer; MMR, mismatch repair; TIL, tumor-infiltrating lymphocytes.

Mismatch repair protein expression

Deficient MMR was identified in 30 (20.0%) tumors, while 120 (80.0%) were pMMR. Among dMMR tumors, MLH1/PMS2 loss was the most frequent pattern, followed by MSH2/MSH6 loss; isolated PMS2 and isolated MSH6 loss were uncommon (Table 2). The overall prevalence of pMMR and dMMR is shown in Figure 01.

Table 02: Distribution of mismatch repair protein-expression patterns

MMR expression pattern	n	% of all cases	% among dMMR	95% CI
pMMR (all four proteins retained)	120	80.0%	—	72.9–85.6%
dMMR (loss of $\geq$ 1 MMR protein)	30	20.0%	—	14.4–27.1%
MLH1/PMS2 loss	18	12.0%	60.0%	42.3–75.4%
MSH2/MSH6 loss	7	4.7%	23.3%	11.8–40.9%
Isolated PMS2 loss	3	2.0%	10.0%	3.5–25.6%
Isolated MSH6 loss	2	1.3%	6.7%	1.8–21.3%

Percentages for individual loss patterns are shown both for the entire cohort and among the 30 dMMR tumors. For overall pMMR/dMMR prevalence, the 95% CI is based on N=150; for loss patterns, the 95% CI is based on n=30 dMMR tumors. CI, confidence interval; dMMR, deficient mismatch repair; pMMR, proficient mismatch repair.

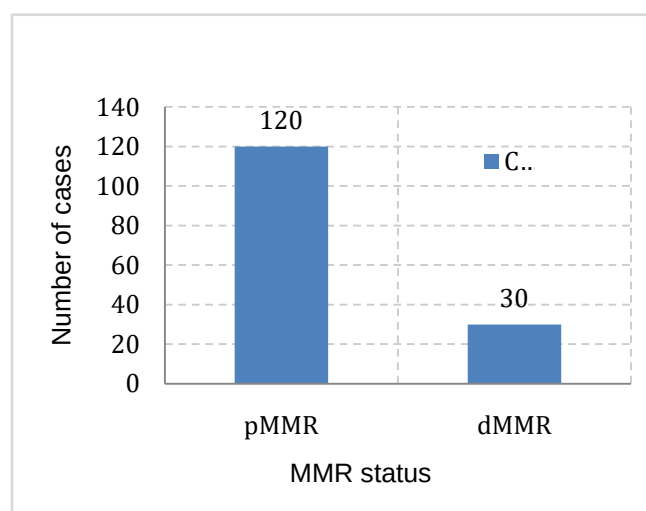


Figure 01: Prevalence of proficient and deficient mismatch repair in colorectal carcinoma.

Association of MMR status with histopathological features

dMMR tumors showed enrichment for right-sided location and mucinous/medullary morphology, although these associations were not independently conclusive at the conventional significance threshold. A prominent lymphocytic response was the clearest morphological correlate: high TIL was present in 12 (40.0%) dMMR tumors compared with 20 (16.7%) pMMR tumors (OR 3.33, 95% CI 1.39–7.99; $p=0.005$). Tumor grade, pT category, nodal status, overall stage, lymphovascular invasion, perineural invasion, and tumor budding showed substantial overlap between the two MMR groups (Table 03).

Table 03: Association of MMR status with clinicopathological characteristics

Characteristic	pMMR (n=120)	dMMR (n=30)	Effect estimate	Test statistic	p-value
Age, years	59.3 ± 12.5	57.0 ± 10.0	Mean difference -2.3 (-6.6 to 2.0)	$t=-1.06$	0.293
Male sex	69 (57.5%)	14 (46.7%)	OR 0.65 (0.29–1.44)	$\chi^2=1.14$	0.286
Right-sided location	40 (33.3%)	15 (50.0%)	OR 2.00 (0.89–4.50)	$\chi^2=2.87$	0.090
Mucinous/medullary histology	12 (10.0%)	7 (23.3%)	OR 2.74 (0.97–7.71)	$\chi^2=3.86$	0.050
Poor differentiation	19 (15.8%)	6 (20.0%)	OR 1.33 (0.48–3.69)	$\chi^2=0.30$	0.584
Lymphovascular invasion	48 (40.0%)	8 (26.7%)	OR 0.55 (0.22–1.33)	$\chi^2=1.82$	0.177
Perineural invasion	39 (32.5%)	10 (33.3%)	OR 1.04 (0.44–2.43)	$\chi^2=0.01$	0.931
High tumor budding	44 (36.7%)	11 (36.7%)	OR 1.00 (0.44–2.29)	$\chi^2=0.00$	1.000
High TIL	20 (16.7%)	12 (40.0%)	OR 3.33 (1.39–7.99)	$\chi^2=7.79$	0.005
Crohn-like lymphoid reaction	34 (28.3%)	12 (40.0%)	OR 1.69 (0.73–3.87)	$\chi^2=1.54$	0.215
Expanding/pushing border	52 (43.3%)	16 (53.3%)	OR 1.49 (0.67–3.34)	$\chi^2=0.97$	0.325
Histological type				$\chi^2=10.55$; FFH exact (MC)	0.052
Conventional adenocarcinoma	98 (81.7%)	23 (76.7%)			
Mucinous adenocarcinoma	11 (9.2%)	4 (13.3%)			
Signet-ring cell carcinoma	7 (5.8%)	0 (0.0%)			
Medullary carcinoma	1 (0.8%)	3 (10.0%)			
Other rare adenocarcinoma subtype	3 (2.5%)	0 (0.0%)			
Tumor grade				$\chi^2=0.69$; FFH exact (MC)	0.632
Well differentiated	12 (10.0%)	4 (13.3%)			
Moderately differentiated	89 (74.2%)	20 (66.7%)			
Poorly differentiated	19 (15.8%)	6 (20.0%)			
pT stage				$\chi^2=1.23$; FFH exact (MC)	0.826

pT1	5 (4.2%)	2 (6.7%)			
pT2	23 (19.2%)	5 (16.7%)			
pT3	64 (53.3%)	17 (56.7%)			
pT4a	23 (19.2%)	4 (13.3%)			
pT4b	5 (4.2%)	2 (6.7%)			
pN stage				$\chi^2=3.31$; FFH exact (MC)	0.155
pN0	61 (50.8%)	17 (56.7%)			
pN1	55 (45.8%)	10 (33.3%)			
pN2	4 (3.3%)	3 (10.0%)			
AJCC stage				$\chi^2=0.45$; FFH exact (MC)	0.913
I	17 (14.2%)	4 (13.3%)			
II	37 (30.8%)	10 (33.3%)			
III	54 (45.0%)	12 (40.0%)			
IV	12 (10.0%)	4 (13.3%)			
Tumor budding				$\chi^2=0.05$	0.977
Low	26 (21.7%)	7 (23.3%)			
Intermediate	50 (41.7%)	12 (40.0%)			
High	44 (36.7%)	11 (36.7%)			
Tumor-infiltrating lymphocytes				$\chi^2=8.07$	0.018
Low	52 (43.3%)	8 (26.7%)			
Moderate	48 (40.0%)	10 (33.3%)			
High	20 (16.7%)	12 (40.0%)			

Age was compared using Welch's *t* test. Binary categorical variables were assessed using Pearson chi-square or Fisher's exact test, as appropriate. For multi-category variables, the global test statistic and *p*-value are shown once on the characteristic row; indented rows show category-specific distributions. Sparse multi-category tables used the Fisher-Freeman-Halton exact test with Monte Carlo estimation (FFH exact [MC]). OR, odds ratio; CI, confidence interval; TIL, tumor-infiltrating lymphocytes.

Predictors of dMMR status

On univariable logistic regression, high TIL was associated with increased odds of dMMR. Right-sided location and mucinous/medullary morphology showed positive associations of lesser precision, whereas age, sex, differentiation, tumor budding, lymphovascular invasion, and perineural invasion were not significant predictors (Table 4).

Table 04: Univariable logistic regression for predictors of dMMR status

Predictor	Crude OR	95% CI	Test statistic	p-value
Age, per 10-year increase	0.85	0.61–1.19	Wald $\chi^2=0.86$	0.353
Male sex	0.65	0.29–1.44	Wald $\chi^2=1.13$	0.288
Right-sided location	2.00	0.89–4.50	Wald $\chi^2=2.81$	0.094
Mucinous/medullary histology	2.74	0.97–7.71	Wald $\chi^2=3.64$	0.056
Poor differentiation	1.33	0.48–3.69	Wald $\chi^2=0.30$	0.585
High TIL	3.33	1.39–7.99	Wald $\chi^2=7.29$	0.007
Crohn-like reaction	1.69	0.73–3.87	Wald $\chi^2=1.52$	0.218
High tumor budding	1.00	0.44–2.29	Wald $\chi^2=0.00$	1.000
Lymphovascular invasion	0.55	0.22–1.33	Wald $\chi^2=1.79$	0.181
Perineural invasion	1.04	0.44–2.43	Wald $\chi^2=0.01$	0.931

The dependent variable was dMMR status. ORs >1 indicate higher odds of dMMR. OR, odds ratio; CI, confidence interval.

In the parsimonious multivariable model, high TIL remained independently associated with dMMR (aOR 3.85, 95% CI 1.52–9.78; *p*=0.005). Right-sided location retained a positive association (aOR 2.18, 95% CI 0.91–5.20; *p*=0.080), while the adjusted association for mucinous/medullary morphology was less precise. The overall model was significant (likelihood-ratio $\chi^2=14.19$, *df*=4; *p*=0.007), with an AUC of 0.690 and acceptable calibration (Hosmer-Lemeshow *p*=0.628) (Table 5 and Figure 2).

Table 05: Multivariable logistic regression for predictors of dMMR status

Predictor	Adjusted OR	95% CI	Test statistic	p-value
Age, per 10-year increase	0.82	0.57–1.17	Wald $\chi^2=1.25$	0.264
Right-sided location	2.18	0.91–5.20	Wald $\chi^2=3.06$	0.080
Mucinous/medullary histology	2.27	0.76–6.81	Wald $\chi^2=2.15$	0.142
High TIL	3.85	1.52–9.78	Wald $\chi^2=8.05$	0.005

Model: age, right-sided location, mucinous/medullary histology, and high TIL. Likelihood-ratio $\chi^2=14.19$ (*df*=4), *p*=0.007; McFadden *R*²=0.095; AUC=0.690; Brier score=0.141; Hosmer-Lemeshow *p*=0.628. aOR, adjusted odds ratio; CI, confidence interval.

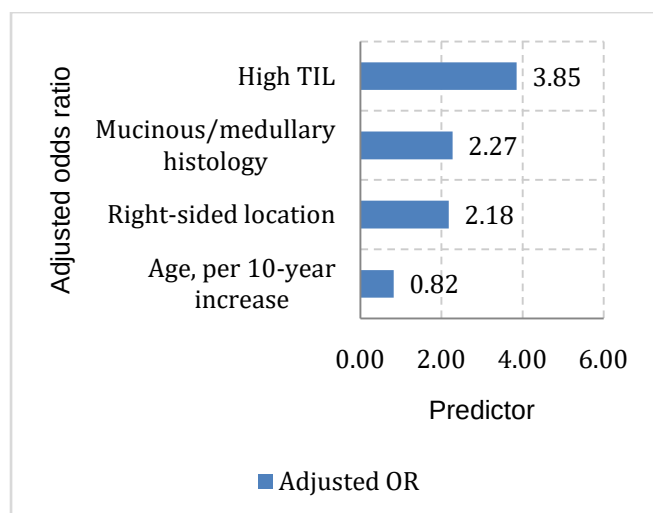


Figure 02: Adjusted predictors of deficient mismatch repair status

Exploratory morphology-based discrimination

An exploratory morphology-only model incorporating right-sided location, mucinous/medullary histology, poor differentiation, high TIL, and Crohn-like lymphoid reaction yielded an AUC of 0.689 (95% CI 0.578–0.797). At the exploratory Youden cut-off, sensitivity was 0.533 and specificity was 0.767, indicating only moderate discrimination.

DISCUSSION

Deficient mismatch repair (dMMR) was found in 20.0% of colorectal carcinomas in the present prospective series of 150 cases. The most common abnormal pattern was the loss of MLH1/PMS2 (60.0% of dMMR cases), followed by MSH2/MSH6 loss. High TIL was most strongly associated with dMMR (aOR 3.85), and was independently associated with dMMR after multivariable adjustment (high TIL in 40.0% of dMMR tumours, 16.7% of pMMR tumours). The location (right side) and the morphology (mucinous/medullary) had positive, but less clear, associations, whereas tumour grade, pT and pN category, overall stage, lymphovascular invasion, perineural invasion and tumour budding were broadly similar between groups. The prevalence of dMMR observed is similar to that reported in some Indian and international cohorts, but there is significant geographic variation. Nayak et al. have analysed 231 consecutive colorectal carcinomas in India and found that 22.9% of these showed MMR protein loss, 13.9% had loss of MLH1/PMS2, 7.4% had loss of MSH2/MSH6, and 1.7% had isolated loss of PMS2 [9]. Age <60 years, location at the right side, and intratumoral lymphocytic infiltration were independently associated with MMR deficiency on multivariable analysis. These findings are very similar to our prevalence and the independent association with high TIL. Hashmi et al. reported a higher frequency (34%) of loss of one or more MMR proteins in 100 colorectal carcinomas from Pakistan. Loss of MLH1/PMS2 was found in 16%, loss of MSH2/MSH6 in 6%, and loss of all four proteins in 7%. The location on the right side and the presence of intratumoral lymphocytosis were significantly associated with dMMR, whereas age, sex, grade and stage were not significantly different [10]. Our results indicate that the lymphocytic response is similar and there are no consistent stage or grade associations, suggesting that host immune

morphology might be more reproducible than several conventional tumour parameters as a clue to dMMR.

Also, larger molecular-pathological series support the predominance of MLH1/PMS2 loss in our cohort. Ye et al. analyzed 502 colorectal cancers and found that dMMR was detected in 14.3% and MSI-high in 12.7% of cases, with 98.4% agreement between the two methods. In the dMMR tumors, paired loss of MLH1/PMS2 was seen in 59.7%, which is very similar to the rate we observed [11]. Right-sided and mucinous tumours were more common in their MSI-high cases, and MSI-high tumours were also associated with poor differentiation, earlier T category, and absence of nodal or distant metastasis. We found the same direction of enrichment for right-sided and mucinous/medullary tumours, but this was not significant for stage or nodes.

The incomplete predictive value of morphology is important. Singh et al. analysed 74 Indian colorectal carcinomas and found that 40.6% were MSI/MMR abnormal, with MLH1/PMS2 loss being the most common pattern (27%) followed by MSH2/MSH6 loss (6.8%). In univariable analysis, younger age, colonic location and the absence of nodal metastasis were significant, but only age below 63 years remained independently associated with MSI, and none of the histological features were independently predictive of MSI status [12]. The age-based ROC model only had an AUC of 0.65. Likewise, the morphology-based model in our study did not discriminate well (AUC 0.689), although it incorporated right-sided location, mucinous/medullary morphology, poor differentiation, TIL, and Crohn-like lymphoid reaction. The results highlight the importance of morphology to complement case selection but not to replace direct MMR testing.

In our series, the correlation between dMMR and immune-rich morphology was even stronger. Maraqa et al. studied 155 CRC cases in patients 45 years or younger and found that 29 cases (19%) were dMMR. Proximal location, composite of pathological features associated with MSI (mucinous change, TIL, Crohn-like reaction) and lower stage were significantly associated with dMMR tumors, and nodal metastasis was less frequent. Loss of MSH2/MSH6 was correlated with a positive family history and overall survival was better in the dMMR patients [13]. Our study was not limited to young patients or survival but the frequency of dMMR (20%) and enrichment of TIL and proximal tumours were very similar.

Other cohorts have shown more variability in the association between MMR status and conventional histological variables. Yassen et al. analysed 50 colorectal carcinomas that were resected and found that 28% were MSI-high, 20% MSI-low and 52% microsatellite stable. MSI status was associated with tumor size, T category, and stage, while MSI-high tumours more frequently occurred in the right colon, showed poorer differentiation, and had fewer distant metastases [14]. However, there was significant overlap between dMMR and pMMR tumours in our pT category, nodal status and overall AJCC stage. These inconsistencies between studies could be due to variations in cohort size, case selection, MSI/MMR classification, and stage distribution.

There is also significant variation in prevalence rates measured in different cohorts and using different methods in India. Pandey et al. evaluated 46 colorectal carcinomas, and found MSI in 10.8% (4.3% MSI-high and 6.5% MSI-low). Seven cases were found to have lost expression of MLH1,

and only one case had loss of expression of MSH2 [15]. The lower frequency observed than the 20% dMMR rate in our study might be partly due to the small number of patients, the limited number of antibodies used for MMR, and the difference between molecular MSI classification and four-protein immunohistochemical screening.

Prospective universal testing data also support the utility of routine immunohistochemistry. Lemos Garcia et al. studied 275 CRC patients under 70 years of age and found abnormal MMR expression in 24 patients (8.7%), the most common being the loss of both MLH1 and PMS2 (15 cases), followed by the loss of PMS2 alone (four cases) [16]. Importantly, none of the patients with abnormal IHC met Amsterdam criteria, 13 met revised Bethesda criteria and 1 genetic Lynch syndrome case would not have been identified without routine tumour screening. It serves to emphasize that patient selection should not be based on clinical and/or morphological criteria only, and highlights the need for systematic MMR evaluation regardless of the presence of classic pathological features.

The strong TIL response in dMMR tumours is likely biologically explainable. Loss of MMR function leads to many insertion-deletion mutations and neoantigens derived from frameshift mutations that can increase immune recognition and recruitment of lymphocytes. After multivariable adjustment, high TIL was the only independently significant morphological predictor of dMMR, with Crohn-like reaction and pushing border having weaker associations.

Several strengths of this study include its prospective design, use of all four major MMR proteins, standardized internal-control interpretation, and detailed assessment of tumour morphology, invasive characteristics, and host lymphocytic response. The study also examined both univariable and adjusted associations and quantified the discriminatory performance of a morphology-based model. Limitations include the single-centre design and relatively modest number of dMMR tumours, which reduced precision for less frequent histological subtypes and individual loss patterns. Molecular MSI testing, BRAF V600E analysis, MLH1 promoter methylation, and germline testing were not part of the reported dataset; therefore, dMMR cases cannot be classified as sporadic or Lynch-associated. The moderate AUC of the morphology-only model further indicates that histopathological features should be regarded as supportive rather than definitive predictors of MMR deficiency.

Overall, dMMR represented one-fifth of colorectal carcinomas in this cohort, with MLH1/PMS2 loss being the dominant immunophenotype. A prominent tumour-infiltrating lymphocytic response showed the strongest independent association with dMMR, while right-sided location and mucinous/medullary morphology provided additional but less definitive clues. These findings support careful morphological evaluation while reinforcing the need for direct MMR immunohistochemistry rather than selective testing based solely on tumour appearance.

CONCLUSION

Deficient mismatch repair was identified in 20% of colorectal carcinomas, with MLH1/PMS2 loss being the most frequent pattern. High tumour-infiltrating lymphocytes showed the strongest independent association with dMMR, while right-sided location and

mucinous/medullary morphology showed weaker positive associations. Histopathological features may help raise suspicion for dMMR, but direct MMR immunohistochemistry remains necessary for reliable classification.

FUNDING

Nil.

ACKNOWLEDGEMENT

Not Declared.

AUTHOR CONTRIBUTION

All Authors are Contributed Equally.

CONFLICT OF INTEREST

Not Declared.

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