

Mismatch Repair Protein Expression and its Association with Clinicopathological Parameters in Colorectal Carcinoma: A Cohort Study

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ABSTRACT

Introduction: Colorectal Carcinoma (CRC) ranks third in global cancer prevalence and is a major cause of cancer-related deaths. The pathogenesis of CRC involves complex genetic and molecular alterations, notably defects in the Deoxyribonucleic Acid (DNA) Mismatch Repair (MMR) pathway. The MMR system, which includes proteins like MLH1, MSH2, MSH6, and PMS2, corrects DNA replication errors. When deficient, it results in Microsatellite Instability (MSI), characterised by changes in repetitive sequence lengths, which are key biomarkers in CRC. High-MSI (MSI-H), defined as the deficiency of more than one MMR protein, is clinically significant due to its association with better prognosis, enhanced response to immunotherapy, and its link to Lynch syndrome.

Aim: To evaluate the expression of MMR proteins in CRC and its association with various clinicopathological parameters.

Materials and Methods: This was a cohort observational study conducted at the Department of Pathology, Kalinga Institute of Medical Sciences, Bhubaneswar, Odisha, India over a period of 24 months from June 2022 to May 2024. A total of 62 histologically confirmed CRC cases were examined using Immunohistochemistry (IHC) to assess MMR protein expression. MSI status was classified into three groups based on MMR protein expression: Microsatellite Stable (MSS) with

no loss of expression, MSI-H with the loss of more than one MMR protein, and low MSI (MSI-L) with the loss of one MMR protein. The associations with clinicopathological features such as age, gender, tumour site, size, histological type, grade, Lymphovascular Invasion (LVI), Perineural Invasion (PNI), and lymph node status were analysed using the Chi-square test through Statistical Packages of Social Sciences (SPSS) version 23.0, and results were presented as figures and tables.

Results: Of the 62 cases of CRC, 43 (69.35%) were MSS, 15 (24.19%) were MSI-H, and 4 (6.45%) were MSI-L. MSI-H tumours showed significant associations with right colon location ($p=0.04$), marked intratumoural lymphocytic infiltration ($p=0.003$), and peritumoural lymphocytic infiltration ($p=0.001$), indicating an enhanced immune response. Most patients with MSI-H status survived after a minimum follow-up of six months, with an average survival of 1,752 days, compared to MSS patients, who had an average survival of 1,141 days.

Conclusion: The MMR protein expression assessed by IHC is a reliable method to screen for MSI in CRC. The association of MSI-H with specific histological features, like marked intratumoural and peritumoural lymphocytes and right-sided tumour location, highlights its importance in guiding further genetic testing and management.

Keywords: Colorectal cancer, Immunohistochemistry, Microsatellite instability, Replication errors

INTRODUCTION

Colorectal Carcinoma (CRC) is the third most prevalent cancer worldwide, ranking second among cancer-related deaths [1]. This malignancy affects the colon and rectum and shows a rising trend among young individuals, particularly in developing countries like India [2]. The pathogenesis of CRC is complex and heterogeneous, involving various molecular and genetic alterations [3]. A critical pathway in CRC carcinogenesis is the defect in the global “DNA damage response” system, which ensures genomic stability and prevents the accumulation of mutations leading to cancer.

A key component of this system is the MMR pathway, which includes proteins such as MLH1, MSH2, MSH6, and PMS2 [3,4]. These nuclear enzymes identify and repair base-base mismatches occurring during DNA replication, which can result from DNA polymerase errors or environmental mutagens [5]. When the MMR system is functional, it maintains genetic code fidelity by correcting replication errors. However, in some cases of CRC, there is a loss or deficiency of MMR proteins [6], which can arise from inherited genetic mutations (e.g., Lynch syndrome) or acquired epigenetic changes (e.g., promoter hypermethylation of MMR genes) [7,8]. This deficiency leads to the accumulation of DNA replication errors

in microsatellites, which are short, repetitive nucleotide sequences in the genome, resulting in Microsatellite Instability (MSI). MSI involves insertions or deletions of nucleotides that alter the length of these repetitive sequences [5].

The presence of MSI in colorectal tumours has significant clinical implications, indicating a better prognosis for MSI-high CRC due to increased immunogenicity and a higher mutational burden, which presents more neoantigens to stimulate an anti-tumour immune response [9,10]. MMR status and MSI serve as predictive biomarkers for chemotherapy response [11]. MSI-high CRCs are resistant to conventional chemotherapy, particularly cisplatin-based protocols, likely due to the rapid development of drug-resistant clones resulting from increased mutational rates [7,12]. Consequently, understanding the MMR status of CRC can guide treatment modifications, with MSI-high cases responding better to immunotherapy with checkpoint inhibitors rather than traditional chemotherapy [13]. The US Food and Drug Administration recommends Pembrolizumab, an immune checkpoint inhibitor, for MSI-high/deficient-MMR (dMMR) solid tumours, including colorectal cancer [14].

Additionally, MSI identification in CRC has broader implications, such as screening family members of Lynch syndrome patients

who are at risk of developing CRC and other cancers [15]. Testing for MMR deficiency or MSI can be conducted through methods like Polymerase Chain Reaction (PCR)-based tests, which compare microsatellite sequence lengths in tumour DNA to those in normal tissue, as well as IHC, which detects MMR proteins in tumour tissue [16]. Numerous studies have linked specific clinical and histopathological parameters to the MMR status of CRCs [17].

The objectives of the present study were to study MMR protein expression in CRC and assess MSI and to associate the MSI status with age, gender, tumour location and size, histological type, histological grade, T stage, nodal metastasis, LVI, PNI, necrosis, mucinous component, signet ring cell component, and intratumoural and peritumoural lymphocytes.

The hypothesis of the present study was that the MMR protein expression status is associated with distinct clinicopathological parameters and may indicate probable response patterns to immunotherapy. Hence, the present study was aimed to evaluate the expression of MMR proteins in CRC and its association with various clinicopathological parameters.

MATERIALS AND METHODS

This cohort observational study was conducted over two years (June 2022 - May 2024) at the Department of Pathology, Kalinga Institute of Medical Sciences, Bhubaneswar, Odisha, India. Approval was granted by the Institutional Ethics Committee (KIIT/KIMS/IEC: 941/2022), following the ethical guidelines of the 1975 Helsinki Declaration, as updated in 2000. A total of 62 histologically confirmed cases of colonic adenocarcinoma, which met the inclusion and exclusion criteria, were retrieved from the Department archives spanning 2019 to 2024.

Inclusion and exclusion criteria: The inclusion criteria for the study were radical excision specimens of histopathologically confirmed CRCs, while the exclusion criteria included colonoscopic biopsies, poorly preserved specimens, and cases that had previously undergone neoadjuvant chemotherapy.

Study Procedure

Colectomy specimens were received in 10% Neutral Buffered Formalin (NBF), cut open along the antimesenteric border, and fixed overnight. The type of specimen (right or left hemicolectomy) was noted, and grossing was conducted according to the protocols of the College of American Pathologists (CAP) [18] and the American Joint Committee on Cancer (AJCC) [19]. Tumour sections and adjacent normal colon were processed using the laboratory's Standard Operating Procedures (SOPs). Sections of 3 µm thickness were cut, and manual staining was performed using Haematoxylin and Eosin (H&E) as per SOP.

Microscopically, the routine H&E sections were examined for tumour type, histologic grade (well differentiated, moderately differentiated, poorly differentiated, and undifferentiated), tumour extent, nodal status, LVI, PNI, necrosis, signet ring cell component, mucinous component, and lymphocytic infiltration. Intratumoural lymphocytic infiltration was classified into mild to moderate (≤ 3 Intraepithelial Lymphocytes (IEL) per High Power Field, HPF) and marked (> 3 IEL/HPF) [20]. A peritumoural lymphocytic response required two or more large lymphoid aggregates [20]. Pathological staging was conducted according to the AJCC 8th edition [19].

The IHC was employed to evaluate the expression of MSH-2, MLH-1, MSH-6, and PMS-2 using anti-rabbit monoclonal antibodies on 3-µm thick Formalin-fixed, Paraffin-embedded (FFPE) tissue sections. The IHC staining procedure was conducted in accordance with SOPs, with normal colon serving as the positive control. Both external and internal positive controls were included. The evaluation of MMR protein expression was based on nuclear expression in tumour cells. Even patchy nuclear staining was interpreted according to CAP protocols as 'Intact Nuclear Expression,' while

'Loss of Nuclear Expression' was interpreted only when there was an absolute lack of nuclear staining [21].

The MSI status was categorised into three groups based on MMR protein expression: MSS with intact nuclear expression of all four MMR proteins, MSI-H with loss of nuclear expression of more than one MMR protein, and MSI-L with loss of nuclear expression of one MMR protein [Table/Fig-1].

MSS	MSI-H (loss of more than one MMR protein)	MSI-L (loss of only one MMR protein)
No loss of expression	Loss of expression of all MMR proteins	Loss of only MSH-6
	Combined loss of MLH-1 and PMS-2	
	Combined loss of MSH-2 and MSH-6	

[Table/Fig-1]: Categorisation of MSI status.

The association of MSI status with clinicopathological parameters included both clinical and histopathological factors. Clinical parameters encompassed age, gender, tumour location (right or left), and survival status, while histopathological parameters included histological type, grade, tumour stage (T stage), nodal metastasis, LVI, PNI, necrosis, mucinous component, signet ring cell component, intratumoural lymphocytes, and peritumoural lymphocytes.

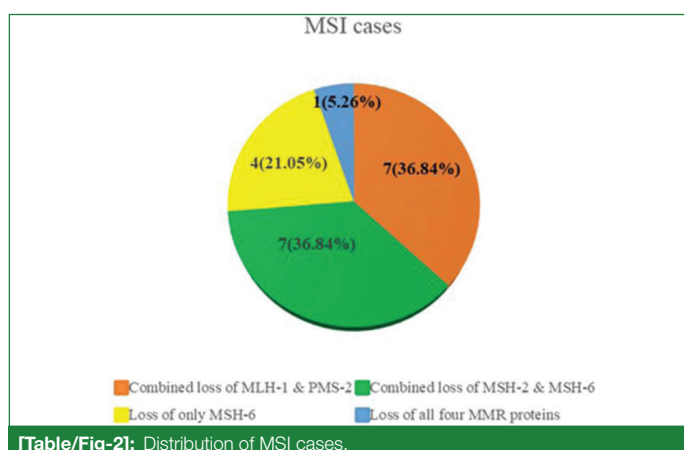
Patient follow-up data were collected through telephone conversations, in-person visits, Outpatient Department (OPD) visits, and radiological and clinical findings from the hospital database.

STATISTICAL ANALYSIS

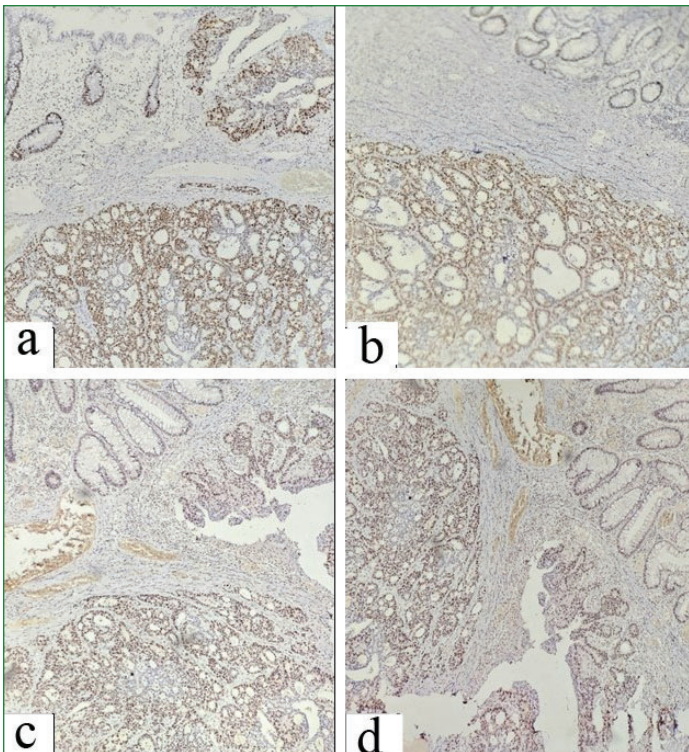
Statistical analysis was conducted to determine the percentages of MSS, MSI-H, and MSI-L tumours, as well as to associate MMR protein expression with clinicopathological parameters. SPSS version 23.0 was employed to evaluate the data after it was entered into Microsoft Excel 2016. Standard Deviation (SD), mean, and percentages were used to express results. Tests of association included the Chi-square test, with a p-value of less than 0.05 considered statistically significant. Survival analysis was performed using Kaplan-Meier curves.

RESULTS

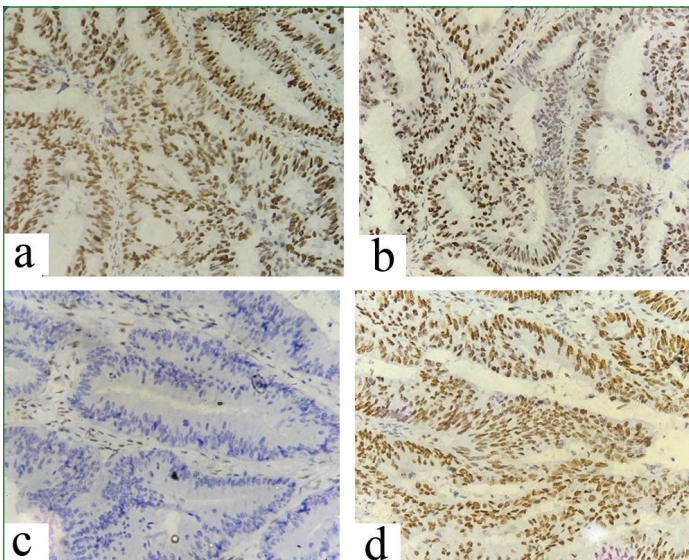
A total of 43 (69.35%) of the 62 cases of CRC were found to be MSS, 15 (24.19%) to be Microsatellite Instability-high (MSI-H), and 4 (6.45%) to be Microsatellite Instability-low (MSI-L). Among the 19 (30.64%) MSI cases, the most common patterns were the combined loss of MLH-1 and PMS-2, seen in 7 (36.84%) cases, and the combined loss of MSH-2 and MSH-6, also seen in 7 (36.84%) cases. Additionally, 1 (5.26%) case showed loss of all four MMR proteins, which were classified as MSI-H. Four (21.05%) cases exhibited loss of only MSH-6, which was categorised as MSI-L [Table/Fig-2]. There were no cases with a combined loss of MSH-6 and PMS-2, MLH-1 and MSH-2, or isolated loss of MLH-1, MSH-2, or PMS-2. [Tables/Fig-3-5] depict cases of MSS, MSI-L, and MSI-H, respectively.



[Table/Fig-2]: Distribution of MSI cases.



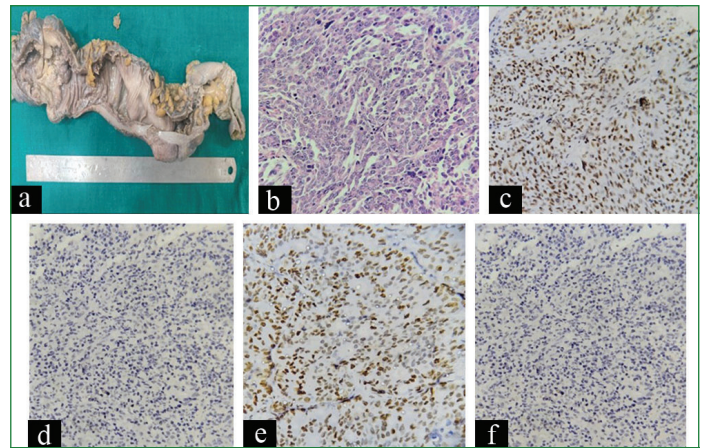
[Table/Fig-3]: A case of MSS colon cancer: a) Tumour cells showing no loss of expression (MSH-2 100x magnification); b) Tumour cells showing no loss of expression (MLH-1 100x magnification); c) Tumour cells showing no loss of expression (MSH-6 100x magnification); d) Tumour cells showing no loss of expression (PMS-2 100x magnification).



[Table/Fig-4]: A case of MSI-L colon cancer: a) Tumour cells showing no loss of expression (MSH-2 400x magnification); b) Tumour cells showing no loss of expression (MLH-1 400x magnification); c) Tumour cells showing loss of expression (MSH-6 400x magnification); d) Tumour cells showing no loss of expression (PMS-2 400x magnification).

Age distribution was similar across MSS and MSI groups, with 24 (38.70%) of the cases diagnosed in patients under 50 years of age. All MSI-L cases occurred in patients over 50. Although not statistically significant ($p=0.257$), MSI-H tumours were more likely to occur in women, with 8 (53.33%) of the 15 cases compared to 16 (37.20%) of the 43 cases in the MSS group.

The MSI-H tumours predominantly occurred in the right colon, accounting for 13 (86.66%) of the cases, while MSS tumours were nearly evenly split between the right and left-sides of the colon, with 21 (48.83%) and 22 (51.16%) cases, respectively. All MSI-L tumours were located in the left colon. Furthermore, MSI-H tumours were larger, with 11 (73.33%) of the 15 cases measuring over 7 cm. Tumour size was not statistically significant concerning MSI status ($p=0.551$). The clinical parameters were depicted in [Table/Fig-6].



[Table/Fig-5]: A case of MSI-H colon cancer: a) Hemicolectomy specimen showing an ulceroproliferative growth; b) Grade 3 colon cancer (H&E-400x); c) Tumour cells showing no loss of expression (MSH-2 400x magnification); d) Tumour cells showing loss of expression (MLH-1 400x magnification); e) Tumour cells showing no loss of expression (MSH-2 400x magnification); f) Tumour cells showing loss of expression (PMS-2 400x magnification).

Parameters		MSI-H	MSI-L	MSS	p-value
Total number of cases		15	4	43	
Age (in years)	<50	6 (40.00%)	0	18 (41.86%)	0.257
	≥50	9 (60.00%)	4 (100%)	25 (58.19%)	
Gender	Male	7 (46.67%)	2 (50%)	27 (62.79%)	0.522
	Female	8 (53.33%)	2 (50%)	16 (37.20%)	
Location	Right colon	13 (86.66%)	0	21 (48.84%)	0.004
	Left colon	2 (13.34%)	4 (100%)	22 (51.16%)	
Size	<4 cm	2 (13.33%)	2 (50%)	9 (20.90%)	0.551
	4-7 cm	2 (13.33%)	0	7 (16.27%)	
	>7 cm	11 (73.34%)	2 (50%)	27 (62.79%)	

[Table/Fig-6]: Association of MSI status with clinical parameters.

Adenocarcinomas constituted 52 (83.87%) of the tumours, evenly distributed between the MSI-H and MSS groups. Mucinous histology was more frequently associated with MSI-H tumours, comprising 3 (37.50%) out of 8 mucinous carcinomas, compared to 12 (23.08%) out of 52 non mucinous tumours. Both the signet ring cell carcinomas in the present study were MSS. All MSI-L cases were adenocarcinomas, with no significant association between tumour type and MSI status ($p=0.701$).

The MSI-H cases were more frequently poorly differentiated, with 8 (53.33%) cases, whereas MSS cases were more likely to be well or moderately differentiated. Histological grade did not significantly associate with MSI status ($p=0.283$). The majority of cases, 50 (80.06%), were diagnosed at stage T3, with no significant association between tumour stage and MSI status ($p=0.793$).

Lymph node metastasis was absent in 8 (53.33%) of MSI-H cases but present in 24 (55.81%) of MSS cases. All MSI-L cases had lymph node metastasis. MSI status did not significantly associate with metastasis to lymph nodes ($p=0.159$), LVI ($p=0.915$), or PNI ($p=0.281$). Most cases in all categories showed necrosis, with no significant association between necrosis and MSI status ($p=0.556$). A mucinous component was present in 6 (40.00%) of MSI-H cases and 9 (20.93%) of MSS cases, while none of the MSI-L cases had a mucinous component. This association was not significant ($p=0.168$).

Intratumoural lymphocytic infiltration was marked in 14 (93.33%) of MSI-H cases compared to 15 (34.88%) of MSS cases, constituting a statistically significant finding ($p=0.003$). Peritumoural lymphocytic infiltration was seen in 13 (86.67%) of MSI-H cases versus 8 (18.60%) of MSS cases, with no MSI-L cases exhibiting this feature, which was also statistically significant ($p=0.001$). The histopathological parameters are depicted in [Table/Fig-7].

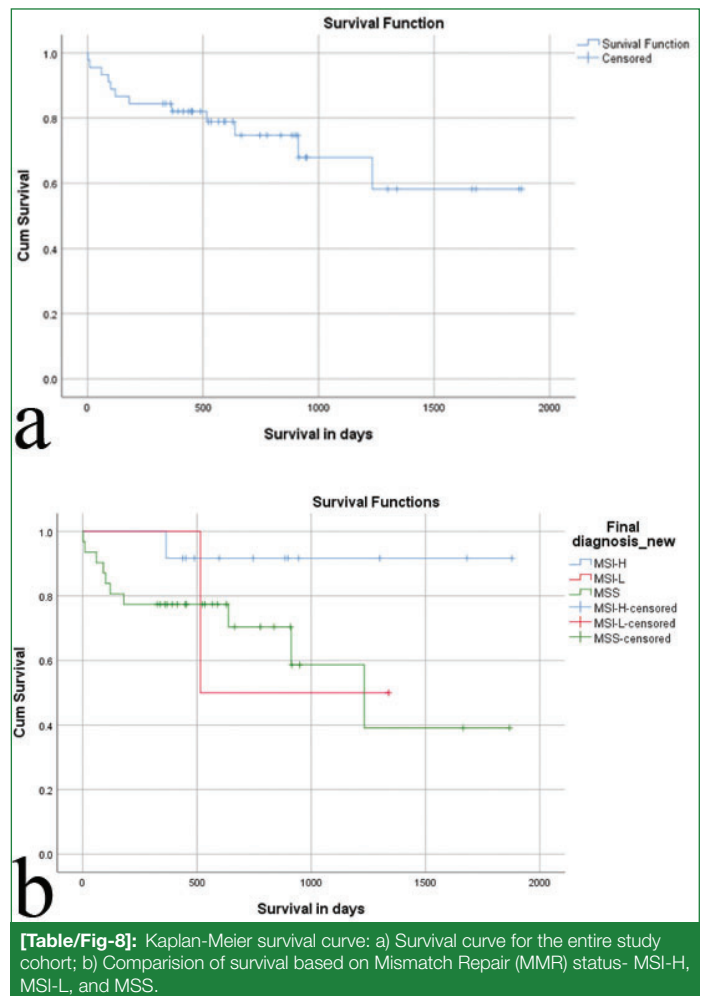
Parameters		MSI-H	MSI-L	MSS	p-value
Total number of cases		15	4	43	
Histological type	Adenocarcinoma	12 (80.00%)	4 (100%)	36 (83.72%)	0.701
	Mucinous carcinoma	3 (20.00%)	0 (0.00%)	5 (11.62%)	
	Signet ring cell carcinoma	0 (0.00%)	0 (0.00%)	2 (4.66%)	
Histological grade	Well-differentiated	2 (13.34%)	1 (25.00%)	13 (30.23%)	0.283
	Moderately differentiated	5 (33.33%)	2 (50.00%)	20 (46.51%)	
	Poorly differentiated	8 (53.33%)	1 (25.00%)	10 (23.25%)	
T stage	T1	0 (0.00%)	0 (0.00%)	3 (6.97%)	0.793
	T2	3 (20.00%)	0 (0.00%)	5 (11.62%)	
	T3	12 (80.00%)	4 (100%)	34 (79.06%)	
	T4	0 (0%)	0 (0%)	1 (2.32%)	
Lymph node metastasis	Absent	8 (53.33%)	0 (0.00%)	19 (44.18%)	0.159
	Present	7 (46.67%)	4 (100%)	24 (55.81%)	
LVI	Identified	7 (46.67%)	2 (50.00%)	18 (41.86%)	0.915
	Not identified	8 (53.33%)	2 (50.00%)	25 (58.13%)	
PNI	Identified	2 (13.33%)	2 (50.00%)	12 (27.90%)	0.281
	Not identified	13 (86.67%)	2 (50.00%)	31 (72.10%)	
Necrosis	Absent	7 (46.67%)	1 (25.00%)	14 (32.55%)	0.556
	Present	8 (53.33%)	3 (75.00%)	29 (67.45%)	
Mucinous component	Identified	6 (40.00%)	0 (0.00%)	9 (20.93%)	0.168
	Not identified	9 (60.00%)	4 (100%)	34 (79.07%)	
Signet ring cell component	Identified	0 (0.00%)	0 (0.00%)	4 (9.30%)	0.389
	Not identified	15 (100%)	4 (100%)	39 (90.70%)	
Intratumoural lymphocytes	Mild/Moderate	1 (6.66%)	3 (75.00%)	28 (65.11%)	0.003
	Marked	14 (93.34%)	1 (25.00%)	15 (34.89%)	
Peritumoural lymphocytes	Absent	2 (13.33%)	4 (100%)	35 (81.40%)	0.001
	Present	13 (86.67%)	0 (0.00%)	8 (18.60%)	

[Table/Fig-7]: Association of MSI status with histopathological parameters.

Follow-up data was available for 45 cases. Of the 12 cases followed up that were MSI-H, 11 (91.66%) were alive, and 1 (8.34%) was deceased. Among MSS patients, 21 (67.74%) were alive while 10 (32.26%) had died after a follow-up of atleast six months. At the time of follow-up, the mean survival time for all three groups (MSS, MSI-L, MSI-H) was 1339 days; the average survival for the MSI-H group was 1752 days, while the average survival for the MSS group was 1141 days. The Kaplan-Meier survival curves for the entire cohort, as well as based on MMR status, were plotted in [Table/Fig-8a,b], respectively. The first graph [Table/Fig-8a] shows the overall Kaplan-Meier survival curve for all 45 patients, with cumulative survival gradually decreasing over time. Censored cases (marked "+") represent patients who were alive at the last follow-up or lost to follow-up.

The second graph [Table/Fig-8b] compares survival rates between the MSI-H, MSI-L, and MSS groups. MSI-H (blue) maintains the

highest survival rate with minimal decline even beyond 1500 days, while MSS (green) shows the steepest drop, reaching about 40% survival by the end of the study. MSI-L (red) demonstrates an intermediate trajectory, although it is based on a very small sample size. The curves suggest a prognostic advantage for MSI-H tumours, consistent with the higher proportion of living patients in the clinical outcome data.



[Table/Fig-8]: Kaplan-Meier survival curve: a) Survival curve for the entire study cohort; b) Comparison of survival based on Mismatch Repair (MMR) status- MSI-H, MSI-L, and MSS.

DISCUSSION

The CRC poses a significant global public health issue owing to its high annual incidence rate. MSI is a crucial pathway in the carcinogenesis of CRC. MSI can be identified either directly through Polymerase Chain Reaction (PCR) techniques, which amplify specific microsatellite repeats, or indirectly by IHC, which detects the absence of MMR proteins. Both methods have demonstrated high concordance in numerous studies [10,22].

In present study, authors examined 62 histopathologically confirmed cases of CRC and performed IHC for the MMR proteins MSH-2, MLH-1, MSH-6, and PMS-2. Based on the nuclear expression of these proteins, the CRCs were categorised as MSI-H, MSI-L, or MSS. Various clinicopathological parameters were then evaluated.

Authors found that out of the 62 cases, 19 (30.64%) were MSI, of which 15 (24.19%) were classified as MSI-H and 4 (6.45%) as MSI-L. This finding is consistent with the results of Paulose RR et al., who documented 27.1% MSI cases, but is higher than those reported in Western and other Asian countries [23]. On the other hand, Soliman NA et al., reported a significantly higher percentage, with 67% of their CRC cases being MSI (47% MSI-H and 20% MSI-L) [20]. Such variations in MSI prevalence might be attributed to differences in study populations, geographic regions, or sample sizes.

Among the 15 (24.1%) MSI-H cases in present study, authors found an almost equal distribution of cases with a combined loss of MLH-1 and PMS-2, as well as MSH-2 and MSH-6. This contrasts

with other studies that reported the combined loss of MLH-1 and PMS-2 as the most dominant pattern of deficient MMR (dMMR). For PMS-2 and MSH-6 to maintain stable heterodimers, their binding partners are necessary. While deficient MSH-2 typically causes loss of both MSH-2 and MSH-6, defective MLH-1 is usually characterised by the coupled loss of MLH-1 and PMS-2. Nevertheless, MLH-1 and MSH-2 can each form heterodimers with other proteins; MLH-1 or MSH-2 labelling is generally unaffected by the loss of PMS-2 or MSH-6 activity, respectively [23].

Authors also encountered one case with the loss of all four MMR proteins, a rare occurrence that has also been documented by Soliman NA et al., and Paulose RR et al., [20,23].

The age range of present study population was from 20 to 85 years, with a mean age of 52.58 years. This aligns with the conclusions of Soliman NA et al., and Paulose RR et al., [20,23]. However, in contrast to the findings of Paulose RR et al., authors did not observe a significant association between younger age and MSI-H status, which may be attributable to the limited number of cases identified in patients under 50 years.

The gender distribution of CRC cases was similar across most studies. Present study did not find a statistically significant association between gender and MSI status, consistent with findings from Soliman NA et al., and Paulose RR et al., [20,23].

In terms of tumour laterality, present study included more cases in the right colon (35, 56.45%) compared to the left colon, similar to the findings of Soliman NA et al., [20]. Other studies have reported a higher occurrence of CRC in the left colon and rectum [23,24]. Notably, 13 (86.67%) of the MSI-H cases in the present study were located in the right colon, indicating a significant association, consistent with other authors.

There was no significant association between tumour size and MSI status. Other studies have not extensively considered size as a clinical parameter for MSI status association [20,23,24].

Regarding histological type, present study included 52 (83.87%) adenocarcinomas, 8 (12.90%) mucinous carcinomas, and 2 (3.23%) signet ring carcinomas. Amongst adenocarcinomas, 12 (23.07%) were MSI-H, while 3 (37.50%) of mucinous carcinomas were MSI-H, though this was not statistically significant. All signet ring cell carcinomas were MSS. The mucinous component was identified in 6 (40.00%) of MSI-H cases compared to only 9 (20.93%) of MSS cases, without statistical significance. Most studies have reported a higher percentage of mucinous CRC cases as Mismatch Repair-Deficient (MMRd) compared to non mucinous tumours. However, Paulose RR et al., found significant associations between histological type, mucinous component, and MSI status, unlike present study [23].

In terms of histological grade, more MSI-H tumours were poorly differentiated 8 (53.34%) compared to MSS tumours 10 (23.25%), though this was not statistically significant. Other authors have found a significant association between poorly differentiated CRC and MSI status [20,23,24].

Most cases in present study were diagnosed at the T3 stage, leading to an uneven distribution. There was no association between tumour stage and MSI status in present study, consistent with Soliman NA et al., [20]. However, Jung SB et al., showed a significant association between higher tumour stage and MSI status [24].

Nodal metastasis was absent in predominantly MSI-H cases 8 (53.34%) compared to 19 (44.18%) MSS cases. This was not statistically significant, in contrast to Soliman NA et al., who found a significant association between the absence of lymph node metastasis and MSI-H status [20].

Authors did not find any association between Lymphovascular Invasion (LVI) or Perineural Invasion (PNI) and MSI status, consistent with Soliman NA et al., and Paulose RR et al., [20,23]. Similarly,

there was no association between tumour necrosis and MSI status, aligning with Soliman NA et al., [20].

Intratumoural and peritumoural lymphocytic infiltration was markedly higher in MSI-H tumours in present study. Specifically, 14 (93.3%) of MSI-H tumours exhibited marked intratumoural lymphocytic infiltration, and 13 (86.7%) showed peritumoural lymphocytic infiltration, both significantly associated with MSI-H status. These findings agree with most other studies, which have found a higher frequency of Crohn's-like peritumoural reactions in MSI cases.

In terms of clinical outcomes, out of 62 cases, 45 were followed up, with 12 cases being MSI-H. The MSI-H cases showed a longer average survival compared to MSS cases, as described in the literature [7,17,23,25].

Limitation(s)

The limitations of present study include a small sample size with late-stage tumour diagnosis leading to uneven case distribution. Tissue preservation issues were observed in a few cases. As molecular analysis for MSI classification was not performed, cases could not be classified as hereditary or sporadic.

CONCLUSION(S)

The present study found that most cases were MSS, with the majority of MSI-H cases demonstrating a combined loss of MLH-1 and PMS-2, as well as MSH-2 and MSH-6. MSI status was significantly associated with right-sided colon tumours, as well as intratumoural and peritumoural lymphocytic infiltration. Universal MSI/MMR screening is limited in India; therefore, clinicopathological parameters may guide selective screening, potentially improving outcomes in resource-limited settings. Further studies are required to establish whether IHC for MMR proteins can altogether replace PCR in low-income countries.

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