ORIGINAL ARTICLE

Assessing MSI Status in GI Cancers (Rectal, Gastric, and Colon Cancer): A Cross-Sectional Study

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ABSTRACT

Background: Microsatellite instability (MSI), a marker of DNA mismatch repair (MMR) deficiency, plays a significant role in the prognosis and treatment of gastrointestinal (GI) cancers, including colorectal, gastric, and rectal malignancies. MSI-high (MSI-H) tumors, particularly in colorectal cancer (CRC), are associated with better prognosis and enhanced response to immunotherapy. However, data on MSI prevalence and its clinicopathological relevance across different GI cancer subtypes, especially in diverse populations, remain limited.

Objective: To establish the prevalence of MSI-H among patients with colorectal cancer in our region and to underscore its potential in shaping individualized treatment approaches across various stages of disease.

Methods: This cross-sectional study was conducted in the Oncology Department of Jinnah Postgraduate Medical Centre from October 2024 to March 2025. A total of 98 patients with histologically confirmed colon, rectal, or gastric cancer were included. MSI status was determined using PCR-based analysis of five mononucleotide markers and immunohistochemistry (IHC) for MMR proteins (MLH1, MSH2, MSH6, PMS2). Clinical and pathological data were collected and analyzed using appropriate statistical tests.

Results: The mean age of patients was 41.2 ± 13.1 years; 71.4% were male. Cancer types included colon (48%), rectal (47%), and gastric (5%). Clinical stages were: stage II (17.5%), stage III (37.1%), and stage IV (45.4%). MSI-H was detected in 11.2% of cases; 88.8% were microsatellite stable (MSS). Deficient MMR protein expression was found in 11.3% of patients. MSI-H was slightly more frequent in males, though the difference was not statistically significant.

Conclusion: MSI-H was observed in 11.2% of GI cancers, indicating a meaningful subset of patients who may benefit from immunotherapy and genetic counseling. Routine MSI testing may improve personalized treatment strategies and patient outcomes.

Keywords: *Microsatellite instability (MSI), mismatch repair (MMR), colorectal cancer, gastric cancer, personalized medicine.*

INTRODUCTION

Gastrointestinal (GI) cancers—including those of the colon, stomach, and rectum—pose a major global health burden due to their high incidence, morbidity, and mortality. Despite progress in diagnostics and therapeutic interventions, these malignancies continue to be a pressing public health issue, emphasizing the need for better biomarkers to enhance disease understanding and refine treatment strategies [1-3].

Microsatellite instability (MSI) refers to a genetic alteration caused by deficiencies in the DNA mismatch repair (MMR) system, resulting in mutations within short, repetitive DNA sequences known as microsatellites. MSI can arise sporadically or in the context of hereditary syndromes such as Lynch syndrome. It has gained prominence as a crucial molecular marker across several cancers, particularly GI malignancies, due to its influence on tumor biology and potential to guide therapeutic decisions [4, 5].

In colorectal cancer, MSI has been extensively studied, especially in MSI-high (MSI-H) tumors, which display unique clinicopathological traits. Evaluating MSI status in such cancers helps identify molecular subtypes and supports tailored treatment approaches, particularly since MSI-H tumors may exhibit distinct responses to therapies such as immunotherapy [6, 7].

This study focuses on determining the prevalence of MSI in rectal, gastric, and colon cancers. Gaining insight into MSI patterns in GI malignancies is essential for developing individualized treatment plans and optimizing clinical management. The findings will contribute valuable knowledge to the molecular profiling of GI cancers and support more informed treatment decision-making.

Recognizing the prevalence and clinical importance of microsatellite instability (MSI) in gastrointestinal cancers is vital for enhancing therapeutic strategies and overall patient management. In colorectal cancer (CRC), MSI-High (MSI-H) is a well-established molecular subtype known to influence tumor progression and responsiveness to certain treatments, particularly immune checkpoint inhibitors [6].

This study plays a crucial role by offering a broad evaluation of MSI status across colon, gastric, and rectal cancers. In early-stage CRC, MSI-H tumors are typically

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linked with a favorable prognosis and demonstrate limited responsiveness to adjuvant 5-fluorouracil (5-FU)-based chemotherapy. In contrast, in metastatic disease, MSI-H status serves as a predictive biomarker for successful outcomes with immune checkpoint inhibitors (ICIs) such as pembrolizumab and nivolumab [8, 9]. These therapies have shown durable responses and extended survival in patients with MSI-H/dMMR metastatic CRC, resulting in FDA approval of ICIs for use in unresectable or metastatic MSI-H solid tumors regardless of origin.

Despite these advances, MSI testing is not yet routinely implemented in many clinical settings, particularly in regions with limited healthcare resources. The scarcity of regional data on MSI prevalence further restricts the development of immunotherapy strategies tailored to specific populations. Therefore, assessing the frequency of MSI-H in our local population is imperative to guide treatment decisions, identify suitable candidates for immunotherapy, and support appropriate genetic counseling and surveillance programs.

By addressing this gap, the present study seeks to establish the prevalence of MSI-H among patients with colorectal cancer in our region and to underscore its potential in shaping individualized treatment approaches across various stages of disease.

MATERIALS AND METHODS

This cross-sectional study was conducted in the Department of Oncology at Jinnah Postgraduate Medical Center/JSMU. This study was conducted in compliance with the ethical standards outlined in the Declaration of Helsinki. Approval was granted by the IRB of Jinnah Postgraduate Medical Center (No.F.2-81/2024-GENL/275/JPMC). Patients diagnosed with colorectal, gastric, or rectal cancers were recruited between October 2024 and March 2025. Inclusion criteria encompassed adults aged 18 years and above with a histologically confirmed diagnosis of colon, rectal, or gastric cancer who were treatment-naïve—having received no prior chemotherapy, radiotherapy, or immunotherapy—and had adequate formalin-fixed, paraffin-embedded (FFPE) tumor specimens available for MSI testing. Written informed consent was mandatory for participation. Patients were excluded if they had previously undergone cancer treatment, had insufficient or missing tissue samples for MSI/MMR testing, had a known diagnosis of another primary malignancy, presented with recurrent or previously treated metastatic disease, or declined to provide consent.

A total of 98 patients were enrolled using a non-probability consecutive sampling method. The sample size was calculated based on the estimated prevalence of MSI-High (MSI-H) status in gastrointestinal cancers—especially colorectal cancer—reported to 15% [9]. This ensured that the study population was sufficient to detect meaningful clinicopathological associations. All eligible patients presenting to the Oncology Department within the designated timeframe who fulfilled the inclusion criteria and consented to participate were consecutively recruited until the target sample size was reached.

Sample size was calculated using an online sample size calculator for proportion available www.openepi.com version 3.01, after inserting 15% prevalence of MSI-High (MSI-H) status in gastrointestinal cancers [9] at a 7.1% margin of error and 95% confidence interval, we required at least N=98 cases for this study.

Clinical and pathological data were collected using a purpose-built standardized proforma. Information was obtained from hospital medical records, pathology reports, and, where needed, direct interviews with patients. The variables recorded included demographic details (age, gender, ethnicity), tumor location, histological type, grade of differentiation, TNM stage, and MSI status. All patient data were anonymized to preserve confidentiality and securely stored for statistical analysis.

MSI assessment involved validated dual techniques—PCR using a panel of microsatellite markers and IHC for detecting MMR protein loss. This structured approach ensured uniform and accurate data collection across all patients.

MSI status was determined using two standard approaches:

1. **PCR-Based Analysis**

   A panel of five mononucleotide markers (BAT-25, BAT-26, NR-21, NR-24, and MONO-27) was used for polymerase chain reaction (PCR)-based testing [10]. Tumors were categorized as follows:

   MSI-High (MSI-H): Instability detected in two or more markers

   MSI-Low (MSI-L): Instability in a single marker

   Microsatellite Stable (MSS): No marker instability detected

2. **Immunohistochemistry (IHC)**

   Immunohistochemical staining was performed to evaluate the expression of mismatch repair (MMR) proteins—MLH1, MSH2, MSH6, and PMS2. Loss of expression of one or more proteins was interpreted as MMR deficiency (dMMR), indicative of MSI-H. Intact nuclear staining of all four proteins was considered proficient MMR (pMMR), consistent with MSS.

Data analysis was performed using SPSS version 26. Descriptive statistics were employed to summarize demographic profiles and clinical characteristics. Frequencies and percentages were computed for categorical variables. Numerical variables were

expressed as mean ± standard deviation. MSI status was compared between the two genders using the Chi-square or Fisher's exact test. A p-value below 0.05 was considered statistically significant.

RESULTS

Table **1** reports the baseline characteristics of the Studied Patients. In the present study, there were ninety-eight cancer patients with a mean age of 41.2 (SD=±13.1) years, ranging from 17 to 70 years old; the majority (71.4%) were male, and 28.6% were female.

Table 1: Baseline characteristics of the study patients.

Characteristics		Frequency	Percentage
Gender	Female	28	28.6
	Male	70	71.4

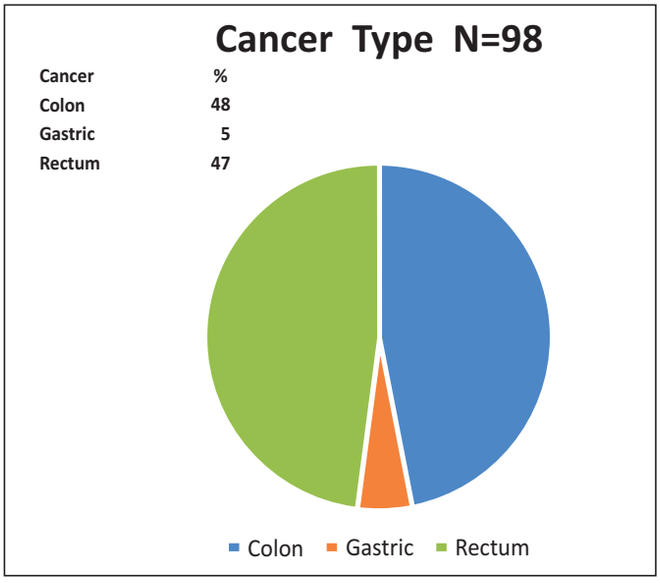

Fig. (1): Types of cancer.

Fig. (1) shows the cancer types of patients: Colon was (48%), Gastric was (5%), and Rectum was (47%). Table **2** reports the descriptive on clinical stage of cancer, MSI and MMR values of studied patients, for cancer type patients of Stage-II were (17.5%), Stage-III were (37.1%), and Stage-IV were (45.4%), in MSI values Stable were (88.8%), and Unstable were (11.2%), whereas for MMR values Intact nuclear expressions were (87., and Loss of nuclear expressions were (11.2%).

Table 2: Descriptive statistics on clinical stage of cancer, MSI, and MMR values.

Parameters		Frequency	Percentage
Clinical Stage of Cancer	Stage-II	17	17.5
	Stage-III	36	37.1
	Stage-IV	44	45.4
MSI Status	Stable	87	88.8
	Unstable	11	11.2
MMR Status	Intact nuclear expressions	87	88.8
	Loss of nuclear expressions	11	11.2

Table **3** reports the association of Cancer type, its clinical stage, MSI and MMR values with gender, among female samples for cancer type cases of Recto Sigmoid were (7.1%), Colon were (46.5%), Gastric were (7.1%), and Rectum were (39.3%), for clinical stages Stage-II were (10.7%), Stage-III were (32.1%), and Stage-IV were (57.1%), for MSI values Stable were (92.9%), Unstable were (7.1%), whereas for MMR values Intact nuclear expressions were (92.9%), and Loss of nuclear expressions were (7.1%), similarly among male patients, Cases of Sigmoid Colon were (1.4%), Cecum were (1.4%), Colon were (42.8%), Gastric were (4.3%), and Rectum were (50%), in clinical stages cases of Stage-II were (20.3%), Stage-III were (39.1%), Stage-IV were (40.6%), for MSI values Stable were (87.1%), Unstable were (12.9%), whereas for MMR values Intact nuclear expressions were (87.1%) and Loss of nuclear expressions were (12.9%). Fisher's exact test did not give any significant association of these parameters with gender (p>0.05).

Table 3: Association of cancer type, clinical stage of cancer, MSI, and MMR with gender.

Parameters		Gender				p-value
		Female		Male		
		n	%	n	%	
Cancer Type	Recto Sigmoid	2	7.1	0	0.0	0.270
	Sigmoid Colon	0	0.0	1	1.4	
	Cecum	0	0.0	1	1.4	
	Colon	13	46.5	30	42.8	
	Gastric	2	7.1	3	4.3	
	Rectum	11	39.3	35	50.0	
Clinical Stage of Cancer	Stage-II	3	10.7	14	20.3	0.310
	Stage-III	9	32.1	27	39.1	
	Stage-IV	16	57.1	28	40.6	
MSI Status	Stable	26	92.9	61	87.1	0.641
	Unstable	2	7.1	9	12.9	
MMR Status	Intact nuclear expressions	26	92.9	61	87.1	0.411
	Loss of nuclear expressions	2	7.1	9	12.9	
*p<0.05 was considered statistically Significant using Fisher's Exact test						

DISCUSSION

The normal tissue DNA repair system, called mismatch repair (MMR), can correct in the process of DNA replication errors. However, due to the lack of MMR genes in tumor cells or defects in the process of replication repair, the possibility of gene mutation is is increased. It can be seen that MSI H is an important factor in the occurrence and development of tumors. About 15% of all colorectal cancers have MSI High, with about 2.5% resulting from genetic inheritance and the remaining 12.5% being sporadic [11].

The primary objective of this cross-sectional study was to assess microsatellite instability (MSI) status in gastrointestinal (GI) cancers (rectal, gastric, and colon cancer). Among the 98 patients analyzed, the majority exhibited microsatellite stability (MSS), with 11.2% classified as MSI-High. This proportion aligns with global estimates reporting MSI-H in approximately 10-20% of colorectal cancers and a smaller fraction in gastric cancers.

Our findings reinforce the clinical relevance of MSI testing, particularly in colorectal cancers, where MSI status serves as both a prognostic and predictive biomarker [12, 13]. MSI-H tumors are generally associated with better prognosis in early-stage disease and a diminished response to conventional fluoropyrimidine-based chemotherapy. Furthermore, they have shown favorable responses to immune checkpoint inhibitors, particularly in advanced disease [14-16]. In our cohort, the proportion of MSI-H cases, identified either by PCR-based instability or by loss of mismatch repair (MMR) protein expression on immunohistochemistry, suggests a meaningful subset of patients who may benefit from immunotherapy.

Interestingly, although rectal and gastric cancers are less frequently associated with MSI-H status, the inclusion of these tumor sites provides a broader understanding of MSI distribution across the GI tract. Studies have shown that MSI-H gastric cancers are more likely to occur in older patients and have a distinct molecular and clinical profile [17]. Our study did not show a statistically significant association between MSI high status and gender, consistent with previous literature that suggests MSI H status has no gender preponderance [18]. The association between MSI and gastric cancer prognosis remains ambiguous [19].

The use of both molecular and immunohistochemical approaches to determine MSI high status added robustness to our assessment. Concordance between MSI-H and deficient MMR (dMMR) was noted, supporting the interchangeability of these methods in clinical settings when applied appropriately [20].

This study highlights the importance of routine MSI testing in GI cancers, not only for its prognostic implications but also for therapeutic stratification. With the advent of personalized medicine, identifying MSI-H/dMMR status has become essential in guiding treatment decisions, particularly with the increasing use of immunotherapy in MSI-H tumors.

LIMITATION

Due to a small gastric cohort, single-center sampling of gastric cancers is underrepresented. So, we need to conduct a further cohort study in the future to assess MSI status in gastric cancer. We acknowledge that a margin of error of 7.1% was used. However, we have mentioned this as one of the limitations of this study.

CONCLUSION

This study shows the prevalence of MSI-H in 11.2% of gastrointestinal cancers (rectal, gastric, and colon) in our population. While robust conclusions were drawn for colorectal cancer, this was due to an adequate sample size. The findings support the utility of MSI testing as a valuable biomarker for identifying patients who may benefit from immunotherapy. Incorporating routine MSI assessment into the prognostic evaluation of GI cancers can aid in personalized treatment planning and improve clinical outcomes.

ETHICS APPROVAL

Ethical approval was obtained from the Institutional Ethical Review Board (No.F.2-81/2024-GENL/275/JPMC). All procedures performed in studies involving human participants followed the ethical standards of the institution and the Helsinki Declaration.

CONSENT FOR PUBLICATION

Written informed consent was taken from the patients.

AVAILABILITY OF DATA

Data is available from the corresponding author on a reasonable request.

FUNDING

None.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

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AUTHORS' CONTRIBUTION

Dr. Sona designed the study. Dr. Sona and Dr. Perah collected data from medical records. Dr. Ghulam Haider analyzed the data. Dr. Sona and Dr. Priyanka drafted the manuscript. All authors have critically reviewed and approved the final draft and are responsible for the content.

REFERENCES

1. Ferlay J, Soerjomataram I, Dikshit R, Eser S, Mathers C, Rebelo M, *et al*. Cancer incidence and mortality worldwide: Sources, methods and major patterns in GLOBOCAN 2012. Int J Cancer2015; 136(5): E359-86.
DOI: https://doi.org/10.1002/ijc.29210

2. Christakis A, Papke D, Nowak J, Yurgelun M, Agoston A, Lindeman N, *et al*. Targeted cancer next-generation sequencing as a primary screening tool for microsatellite instability and Lynch syndrome in

upper gastrointestinal tract cancers. Cancer Epidemiol Biomarkers Prev 2019; 28(6): 1017-23.
DOI: https://doi.org/10.1158/1055-9965.EPI-18-1250

3. Siegel R, Miller KD, Jemal A. Cancer statistics, 2020. CA Cancer J Clin 2020; 70(1): 7-30.
DOI: https://doi.org/10.3322/caac.21590

4. Stadler Z, Battaglin F, Middha S, Yaeger R, Tran C, Cercek A, *et al*. Reliable detection of mismatch repair deficiency in colorectal cancers using mutational load in next-generation sequencing panels. J Clin Oncol 2016; 34(18_suppl): 103.
DOI: https://doi.org/10.1200/JCO.2016.34.18

5. Loupakis F, Depetris I, Biason P, Intini R, Prete AA, Leone F, *et al*. Prediction of benefit from checkpoint inhibitors in mismatch repair deficient metastatic colorectal cancer: Role of tumor infiltrating lymphocytes. Oncologist 2020; 25(6): 481-7.
DOI: https://doi.org/10.1634/theoncologist.2019-0626

6. Weisenberger D, Siegmund K, Campan M, Young J, Long T, Faasse M, *et al*. CpG island methylator phenotype underlies sporadic microsatellite instability and is tightly associated with BRAF mutation in colorectal cancer. Nat Genet 2006; 38(7): 787-93.
DOI: https://doi.org/10.1038/ng1834

7. Dudley J, Lin M, Le DT, Eshleman J. Microsatellite instability as a biomarker for PD-1 blockade. Clin Cancer Res 2016; 22(4): 813-20.
DOI: https://doi.org/10.1158/1078-0432.CCR-15-1678

8. Li K, Luo H, Huang L, Luo H, Zhu X. Microsatellite instability: A review of what the oncologist should know. Cancer Cell Int 2020; 20: 16.
DOI: https://doi.org/10.1186/s12935-019-1091-8

9. Hunzeker Z, Bhakta P, Gudipally S, Kavuri S, Venkatesan R, Nwanze C. Complete response of high microsatellite instability gastric cancer and synchronous microsatellite stability rectal cancer. Cureus 2022; 14(5): e25820.
DOI: https://doi.org/10.7759/cureus.25820

10. Lorenzi M, Amonkar M, Zhang J, Mehta S, Liaw K. Epidemiology of microsatellite instability high (MSI-H) and deficient mismatch repair (dMMR) in solid tumors: A structured literature review. J Oncol 2020; 2020: 1807929.
DOI: https://doi.org/10.1155/2020/1807929

11. Boland C, Goel A. Microsatellite instability in colorectal cancer. Gastroenterology 2010; 138(6): 2073-87.e3.
DOI: https://doi.org/10.1053/j.gastro.2009.12.064

12. Amato M, Franco R, Facchini G, Addeo R, Ciardiello F, Normanno N, *et al*. Microsatellite instability: From the implementation of the detection to a prognostic and predictive role in cancers. Int J Mol Sci 2022; 23(15): 8726.
DOI: https://doi.org/10.3390/ijms23158726

13. Middha S, Zhang L, Nafa K, Jayakumaran G, Wong D, Kim H, *et al*. Reliable pan-cancer microsatellite instability assessment by using targeted next-generation sequencing data. JCO Precis Oncol 2017; 2017: PO.17.00084.
DOI: https://doi.org/10.1200/PO.17.00084

14. Le D, Durham J, Smith K, Wang H, Bartlett B, Aulakh L, *et al*. Mismatch repair deficiency predicts response of solid tumors to PD-1 blockade. Science 2017; 357(6349): 409-13.
DOI: https://doi.org/10.1126/science.aan6733

15. Overman M, McDermott R, Leach J, Lonardi S, Lenz H, Morse M, *et al*. Nivolumab in patients with metastatic DNA mismatch repair-deficient or microsatellite instability-high colorectal cancer (CheckMate 142): An open-label, multicentre, phase 2 study. Lancet Oncol 2017; 18(9): 1182-91.
DOI: https://doi.org/10.1016/S1470-2045(17)30422-9

16. Latham A, Srinivasan P, Kemel Y, Shia J, Bandlamudi C, Mandelker D, *et al*. Microsatellite instability is associated with the presence of Lynch syndrome pan-cancer. J Clin Oncol 2019; 37(4_suppl): 91.
DOI: https://doi.org/10.1200/JCO.2019.37.4_suppl.91

17. Polom K, Marrelli D, Roviello G, Pascale V, Voglino C, Rho H, *et al*. Molecular key to understand the gastric cancer biology in elderly patients—The role of microsatellite instability. J Exp Clin Cancer Res 2020; 39: 94.
DOI: https://doi.org/10.1186/s13046-020-01768-z

18. Ashktorab H, Smoot D, Farzanmehr H, Fidelia-Lambert M, Momen B, Hylind L, *et al*. Clinicopathological features and microsatellite instability (MSI) in colorectal cancers from African Americans. Int J Cancer 2005; 116(6): 914-9.
DOI: https://doi.org/10.1002/ijc.21129

19. Zhu L, Li Z, Wang Y, Zhang C, Liu Y, Qu X. Microsatellite instability and survival in gastric cancer: A systematic review and meta-analysis. Mol Clin Oncol 2018; 9(2): 119-27.
DOI: https://doi.org/10.3892/mco.2018.1712

20. Saeed O, Mann S, Luchini C, Huang K, Zhang S, Sen J, *et al*. Evaluating mismatch repair deficiency for solid tumor immunotherapy eligibility: Immunohistochemistry *versus* microsatellite molecular testing. Hum Pathol 2021; 114: 29-36.
DOI: https://doi.org/10.1016/j.humpath.2021.05.009