ORIGINAL ARTICLE

Annals of Gastroenterology (2024) **37,** 1-8

Immune checkpoint inhibitor-associated gastrointestinal adverse events in patients with colorectal cancer

Antonio Pizuorno Machado[a#], Saltenat Moghaddam Adames[b#], Malek Shatila[c], Parvir Aujla[a], Ryan Huey[d], Yinghong Wang[c*], Anusha Thomas[c*]

The University of Texas Health Science Center, Houston, TX; The University of Texas MD Anderson Cancer Center, Houston, TX; Baylor College of Medicine, Houston, TX, USA

Abstract

Background Immune checkpoint inhibitors (ICI) target microsatellite instability-high (MSI-H) tumors with success. The incidence and characteristics of ICI-related colitis (IMC) in patients with MSI-H colorectal cancers (CRC) are unclear.

Methods We performed a retrospective analysis of adult patients with CRC who received ICI between June 1, 2014, and December 31, 2022, including data on IMC observed up to 3 months after the last dose of ICI. Patients' demographics, oncologic profile, endoscopic features, treatment and clinical outcomes were evaluated.

Results Of 474 patients with CRC receiving ICI during our study period, 18 developed IMC (3.8%). The majority were Caucasian (88.8%), male (61.1%), and their median age was 69.5 years. Of these patients, 50% received combination therapy with anti-PD-1/L1 and CTLA-4; 66.6% had MSI-H colorectal cancer, 11.1% had a second cancer-melanoma, while 61.2% and 66.7% had grade 1-2 colitis and diarrhea respectively. Endoscopic evaluation was used in 5 patients, of whom 2 had ulcerative inflammation necessitating selective immunosuppressive therapy with biologics. Therapy was withheld in 61.1% because of toxicity; 41.4% and 5.8% were noted to have median Common Terminology Criteria for Adverse Events grade 2 liver and pancreas toxicity respectively. The majority of our cohort received steroid therapy.

Conclusions The lower severity of IMC, compared to toxicity in other ICI-treated cancers, may be influenced by the tumor microenvironment in MSI-H colorectal cancer after ICI exposure. Larger prospective studies are necessary to determine the role of tumor biology and the gut microbiome in the disease profile and severity of IMC.

Keywords Immune checkpoint inhibitors, colorectal cancer, immune-related adverse event, colitis

Ann Gastroenterol 2024; 37 (XX): 1-8

Conflict of Interest: None

[#] Authors share co-first authorship

*Authors share co-senior authorship

Correspondence to: Anusha Thomas, Department of Gastroenterology, Hepatology, and Nutrition, The University of Texas MD Anderson Cancer Center, 1515 Holcombe Blvd Houston, Unit 1466, TX 77030, USA, e-mail: ASThomas1@mdanderson.org

Received 24 April 2024; accepted 16 October 2024; published online 12 December 2024

DOI: https://doi.org/10.20524/aog.2024.0935

Introduction

Immune checkpoint inhibitors (ICIs) are a potent and increasingly important treatment option for various malignancies. To date, more than 8 ICI agents have been approved. While conferring an appreciable survival benefit, these agents also predispose to unique immune-related gastrointestinal adverse events (irAEs), with diarrhea and colitis amongst the most common [1]. Immune-mediated colitis (IMC) has been reported in up to 40% of patients treated with ICIs. It varies widely in severity [2,3], and can be a cause for discontinuation of ICI therapy [4]. Failure in early recognition and delayed or suboptimal treatment early in the disease course can lead to an increased risk of complications such as bowel perforation [5].

The use of ICIs to treat microsatellite instability-high (MSI-H) colon cancer is a relatively recent development. One clinical trial showed that pembrolizumab can lead to significantly longer progression-free survival than chemotherapy, when received as first-line therapy for MSI-H/mismatch repair deficient (dMMR)-metastatic colorectal cancer (CRC), with fewer treatment-related adverse events [6]. Several studies have shown activity and clinical benefit for ICIs in CRC [6-9]. However, much remains to be learned about irAEs for this patient population. Given their novelty, our knowledge of ICI's potential irAEs in this setting is still limited. Ostensibly, the presence of malignancy in the bowel may uniquely impact the risk and severity of gastrointestinal irAEs specifically.

There have been limited large-scale studies investigating the safety of ICIs in patients with CRC in terms of irAE. In this retrospective study, we explored the incidence and clinical manifestations of IMC among patients with CRC.

Patients and methods

Study design and population

This retrospective chart review was a descriptive, single-center study that included adult patients diagnosed with CRC and treated with ICI at a tertiary cancer center between June 1, 2014 and December 31, 2022. This study was approved by the institutional review board with a waiver of patients' informed consent. We identified adult cancer patients 18 years or older who: (1) were treated with ICIs for CRC; and (2) had a diagnosis of IMC at least 3 months after the last ICI dose. Patients with preexisting inflammatory bowel disease, microscopic colitis, or other autoimmune gastrointestinal disorders were excluded.

Clinical data

Demographic and cancer-related information such as age, sex, primary cancer type, stage, cancer treatments received and doses, and Charlson Comorbidity Index score were collected. Also collected were data related to the onset of colitis, such as date, cycles of ICI before colitis, type of ICI, and peak Common Terminology Criteria for Adverse Events (CTCAE) grades for colitis and diarrhea. The diagnosis of colitis was based on the clinical presentation and endoscopic and histologic features, after the exclusion of other etiologies. Information about the treatment for colitis, such as steroids, infliximab and vedolizumab, including doses and start and end dates, was also obtained. Colonoscopy/sigmoidoscopy and pathology findings at the time of colitis diagnosis were reported if available.

[a]Department of Internal Medicine, The University of Texas Health Science Center, Houston, TX, USA (Antonio Pizuorno Machado, Parvir Aujla); [b]Baylor College of Medicine, Houston, Texas, USA (Saltenat Moghaddam Adames); [c]Department of Gastroenterology, Hepatology, and Nutrition, The University of Texas MD Anderson Cancer Center, Houston, TX, USA (Saltenat Moghaddam Adames, Malek Shatila, Yinghong Wang, Anusha Thomas); [d]Department of Cancer Medicine, The University of Texas MD Anderson Cancer Center, Houston, TX, USA (Ryan Huey)

Statistical analysis

The statistical analyses performed were descriptive in nature. The distributions of continuous variables were summarized as medians and interquartile ranges, and those of categorical variables as frequencies and percentages. These were calculated using SPSS 26 (2019, IBM Corporation, USA).

Results

Patient population, characteristics and oncologic history

We identified 474 patients with a diagnosis of CRC who had exposure to ICI between June 2014 and December 2022. Of these, only 18 patients met our inclusion criteria (Fig. 1). These patients had a median age of 69.5 years; 11 (61.1%) were male; and 16 (88.8%) were white (Table 1). Regarding oncological history, 18 patients (100%) were diagnosed with CRC, followed by overlapped melanoma in 2 patients (11.1%) and genitourinary cancer in 1 patient (5.5%). The majority of the patients (n=13, 72.2%), had stage IV cancer; 12 patients (66.6%) had an MSI-H CRC. With regard to the class of ICI that patients received, 9 (50%), 8 (44.4%), and 1 (5.5%) patients received a combination of PD-1/L1 and CTLA-4 combination therapy, PD-1/L1 inhibitor monotherapy and CTLA-4 monotherapy, respectively (Table 2). Patients underwent a median of 6 cycles of ICI. After the colitis event, 5 patients (27.7%) continued with ICI and 2 patients (5.8%) continued with other forms of cancer therapy.

Characteristics and treatment of colitis

The predominant symptom was diarrhea in all 18 patients (100%), and abdominal pain in 18 patients (100%) (Table 3);

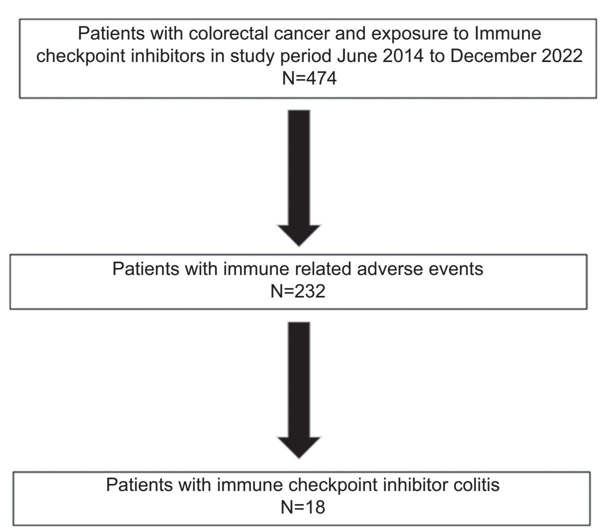

Figure 1 Patient selection diagram

Table 1 Patient characteristics, N=18

Characteristic	No (%)
Age, years, median (IQR)	69.5 (50-73)
Sex, male	11 (61.1)
Race, white	16 (88.8)
Cancer type	
Melanoma*	2 (11)
Genitourinary *	1 (5.5)
Gastrointestinal	18 (100)
Cancer stage	
III	5 (27.7)
IV	13 (72.2)
MSI status of tumor	
MSI - High	12 (66.6)
MSI - Stable†	3 (16.6)
Unavailable	3 (16.6)
Type of ICI	
Anti–CTLA-4 monotherapy	1 (5.5)
Anti–PD-1/L1 monotherapy	8 (44.4)
Combination anti–CTLA-4 and anti–PD-1/L1	9 (50)
Cycles of ICI, median (IQR)	6 (2.5-10)
Other gastrointestinal irAE	
Hepatitis	14 (41.1)
Pancreatitis	2 (5.8)
Active treatment with other chemotherapy	
Continued with ICI	5 (27.7)
Continued with other chemotherapy	2 (5.8)
Median time duration of ICI therapy, months (IQR)	3.5 (0-7)
Median time duration from gastrointestinal irAE diagnosis and first dose of ICI, days (IQR)	259 (70-444)

*2 patients were diagnosed with melanoma and 1 patient with genitourinary cancer and then subsequently diagnosed with colorectal cancer

†Patients in the MSI Stable group received ICI for alternative reasons, such as different clinical trials, as well as other secondary cancers where ICI was indicated

MSI, microsatellite instability; CTLA-4, cytotoxic T lymphocyte antigen 4; ICI, immune checkpoint inhibitor; IQR, interquartile range; PD-1/PD-L1, programmed cell death 1/programmed death ligand 1; irAE, immune-related adverse event

colitis presented at a median of 259 days after ICI initiation (Table 1). The median fecal calprotectin before treatment was 641 μg/g. The median peak CTCAE was 1 for colitis and 2 for diarrhea (Table 3). The majority of the patients had grade 1 colitis (55.5%). Hospitalization was required for 4 patients (22.2%). As regards the treatment of colitis, steroids were used in the entire cohort, and in conjunction with vedolizumab (1 patient, 5.5%) or infliximab alone (3 patients, 16.6%). A fecal microbiota transplant was performed in 1 patient (5.5%). Of the 18 patients who received corticosteroid treatment for IMC, the median duration of steroid use in the first year after colitis diagnosis was 30.5 days, and the median number of taper events was 3. For those who received biologics as treatment, the median number of doses was 2 and median duration of biologic use in the first year after colitis diagnosis was 9 days. Of the 18 patients who received treatment for IMC, 13 achieved remission and 5 had recurrence of IMC. The median CTCAE grade of diarrhea in those who had recurrence of IMC was 2.25 (Table 4).

Endoscopic and histology-related characteristics

At the time of colitis diagnosis, only 5 patients underwent an endoscopic procedure. Non-ulcer inflammation was found in 2 patients (40%), and ulcerative inflammation was also found in 2 patients (40%). On histology, the majority had active inflammation (4 patients, 80%) (Table 5).

Discussion

This study, to our knowledge is the first to explore the incidence and clinical presentation of lower gastrointestinal toxicity to ICI among patients with CRC. While our initial concern was that the presence of malignancy along the colon may predispose to locoregional inflammatory processes, particularly after immune checkpoint inhibition, surprisingly, we found that the incidence of gastrointestinal irAEs in our sample was substantially lower than that found in the literature

Table 2 Individual patient characteristics

Patients	1	2	3	4	5	6	7	8	9
Cancer stage	4	4	3	3	3	3	4	4	4
ICI Treatment (PD1/Pd-l-1, CTLA4, Combination)	PD-1/Pd-l-1	Combination	PD-1/Pd-l-1	PD-1/Pd-l-1	Combination	PD-1/Pd-l-1	PD-1/Pd-l-1	Combination	Combination
MSI/MMR status	Unavailable	Unavailable	MSI-H	MSI-H	Unavailable	MSI-S	MSI-H	MSI-H	MSI-H
CTCAE grade Grade of diarrhea	1	2	3	2	3	1	3	3	3
IMC treatment	Steroid	Steroid	Steroid	Steroid	Steroid	Steroid	Steroid	Steroid, biologic	Steroid
IMC recurrence after treatment	Remission	Recurrence	Remission	Remission	Recurrence	Remission	Remission	Recurrence	Recurrence (perforation)
Cancer status	Remission/ stable	Remission/ stable	Disease progression	Disease progression	Remission/ stable	Disease progression	Disease progression	Remission/ stable	Disease progression
Patients	**10**	**11**	**12**	**13**	**14**	**15**	**16**	**17**	**18**
Cancer stage	3	4	4	4	4	4	4	4	4
ICI Treatment (PD1/Pd-l-1, CTLA4, Combination)	Combination	Combination	PD-1/Pd-l-1	Combination	Combination	PD-1/Pd-l-1	Combination	CTLA-4	PD-1/Pd-l-1
MSI/MMR status	MSI-H	MSI-H	MSI-H	MSI-H	MSI-H	MSI-H	MSI-H	MSI-S	MSI-S
CTCAE grade Grade of diarrhea	2	2	1	4	2	2	1	2	2
IMC treatment	Steroid	Steroid, biologic	Steroid	Steroid	Steroid, biologic	FMT, steroid biologic	Steroid	Steroid	Steroid
IMC recurrence after treatment	Remission	Recurrence	Remission	Remission	Remission	Remission	Remission	Remission	Remission
Cancer status	Disease progression	Remission/ stable	Remission/ stable	Remission/ stable	Remission/ stable	Remission/ stable	Remission/ stable	Remission/ stable	Remission/ stable

ICI, immune checkpoint inhibitor; PD-1/PD-L1, programmed cell death 1/programmed death ligand 1; CTLA-4, cytotoxic T lymphocyte antigen 4; MSI, microsatellite instability; MMR, mismatch repair; CTCAE v5, Common Terminology Criteria for Adverse Events version 5; IMC, immune-mediated colitis.

Table 3 Characteristics of gastrointestinal irAE in patients with colorectal cancer, N=18

Colitis, N=18	No. (%)
Symptoms	
Diarrhea	18 (100)
Abdominal pain	18 (100)
Nausea/vomiting	1 (5.5)
Median fecal calprotectin before treatment (IQR), N=8	641 (347-1560.5)
Median CTCAE grade of colitis (IQR)	1 (0.5-2)
Median CTCAE grade of diarrhea (IQR)	2 (2-3)
Grade I-II diarrhea	12 (66.7)
Grade III- IV diarrhea	6 (33.3)
Hospitalization required, N (%)	4 (22.2)
Cancer treatment withheld because of toxicity, N (%)	11 (61.1)
All-cause mortality, N (%)	7 (38.8)

CTCAE v5, Common Terminology Criteria for Adverse Events version 5; ICI, immune checkpoint inhibitor; IMC, immune-mediated colitis; IQR, interquartile range; TNF, tumor necrosis factor; FMT, fecal microbiota transplantation; irAE, immune-related adverse event

Table 4 Treatment and outcomes in patients diagnosed with colitis, N=18

At the time of colitis diagnosis	No. (%)
Treatment of IMC	
Corticosteroids	18 (100)
Median days of steroid use in the first year after colitis diagnosis (IQR), N=13	30.5 (271-15.5)
Median steroid taper events (IQR), N=13	3 (5.5-1.5)
Biologic*	4 (22.2)
Median number of biologic doses (IQR), N=4	2 (3-1)
Median days of biologic use in the first year after colitis diagnosis (IQR), N=4	9 (117-1)
FMT	1 (5.5)
Treatment of IMC outcomes	
Remission	13 (72.2)
IMC recurrence	5 (27.8)
Median CTCAE grade of colitis recurrence (IQR), N=5	2.25 (3.5-1.25)
Median CTCAE grade of diarrhea recurrence (IQR), N=5	2.25 (3.5-1.25)
Perforation	1 (5.5)

*1 patient received Infliximab; 3 patients received vedolizumab

IMC, immune-mediated colitis; IQR, interquartile range; FMT, fecal microbiota transplant, CTCAE, Common Terminology Criteria for Adverse Events, CRP, C reactive protein

Table 5 Endoscopy-related characteristics for patients diagnosed with colitis, N=18

At the time of colitis diagnosis	No. (%)
Endoscopic findings	5 (27.7)
Ulcers	2 (40)
Non-ulcer inflammation	2 (40)
Normal	1 (20)
Histologic findings	
Active inflammation	4 (80)
Normal	1 (20)

for other tumor types (14-37%), while also being potentially less severe [10-12]. Furthermore, our sample demonstrated a delayed onset of toxicity (median of 259 days after ICI) in comparison to the reported time window of 2-3 months. These findings pose interesting questions regarding the mechanism of immune-mediated toxicity and the role of the tumor microenvironment, as well as the gut microbiome, in their development.

CRC refers to any tumor of the inner lining of the rectum or colon. It is the third most common cancer type, comprising 8% of new cancer cases annually, and although its incidence and mortality rates have declined in the past decade, it remains among the deadliest types of malignancy when metastatic [13,14]. In CRCs not amenable to resection, systemic treatments are available, the choice of which highly depends on the tumor's mutational profile. For instance, current guidelines from the National Comprehensive Cancer Network endorse the use of ICI for the treatment of dMMR/MSI-H CRC, which is predictive of response to ICIs [15-17]. Immunotherapy, however, comes with the risk of irAEs, of which gastrointestinal toxicities (primarily enterocolitis) are among the more common and severe [18]. This poses a unique situation where there is a regional overlap in cancer location and drug-related organ toxicity, a phenomenon that has yet to be studied adequately in the field of immunotherapy. Previous studies have suggested the existence of tumor-dependent irAE profiles. For instance, 1 study found that melanoma was associated with a higher incidence of gastrointestinal and cutaneous irAEs and a lower frequency of pulmonary irAEs [19]. Another study showed that patients with melanoma were more likely to develop cutaneous irAEs, while those with non-small cell lung cancer were more likely to develop pulmonary irAEs [20]. Together, these suggest the potential for locoregional tumor effects that influence the preponderance of inflammatory adverse events, highlighting the complexity of the tumor microenvironment. Though the specific immune phenotype varies greatly between types of cancers, depending on the interplay of increased immune activation in response to tumor neoantigens and the activation of immunosuppressive signaling pathways by the tumor to evade the body's immune surveillance [21], there is a disruption of immune cell functioning regardless. This conceivably impacts local predisposition to autoimmunity induced by checkpoint inhibitors, and is supported by 2 studies that found that patients who received ipilimumab for active metastatic disease had a lower rate of severe irAEs than those who received it as post-surgical, adjuvant treatment [22,23]. In our study, we found that CRC could potentially mitigate the risk for luminal gastrointestinal irAEs among patients receiving immunotherapy. While these results need to be validated through further studies, it raises an interesting question regarding the impact of tumor burden and location on the incidence of related organ toxicities.

ICIs are an effective means of treating cancer by enhancing the human body's natural immune defenses, allowing it to mount an anti-tumor response. Three classes of ICIs have FDA approval with different mechanisms. PD-1/L1 inhibitors block the activity of the programmed death-1/ligand 1 protein, which typically suppresses cytokine production and immune cell proliferation [24]. CTLA-4 inhibitors interfere with the activity of the cytotoxic T-lymphocyte antigen 4 protein, which serves the dual function of inhibiting T-cell costimulation while promoting the activity of regulatory T cells that dampen immune responses [25]. Finally, the recently approved lymphocyte activation gene 3 inhibitors help reconstitute the immune system after T-cell exhaustion [26]. These agents induce a potent antitumor immunity which, by the same mechanism, may also promote autoimmunity. Although the precise mechanism of development of these immune-related adverse events probably differs according to the class of ICI used and the system involved, T cells are heavily implicated in this process [27]. IrAEs can pose a significant obstacle to long-term treatment with ICIs, and extensive research is underway to elucidate the pathophysiology of irAEs and to identify predictive biomarkers for these toxicities [27]. Of all the risk factors explored, the aforementioned tumor microenvironment has received surprisingly little attention for its role in the pathogenesis of irAEs. Two tumor immunophenotypes are traditionally described, based on the degree of immune cell infiltration. Immunologically "hot" tumors are those with a preponderance of tumor-infiltrating lymphocytes, a strong immune signature, and activation of immune checkpoints by the tumor as a means of circumventing this inflammatory response [28]. "Cold" tumors, on the other hand, are those with sparse inflammatory infiltrate and typically dense, fibrotic stroma [29]. Some measure of immunosuppression is employed by both phenotypes to allow the cancer to escape immune surveillance, but their responsiveness to immunotherapy differs significantly [28]. Conceivably, this difference in tumor microenvironment may also impact the risk of irAEs. Microsatellite status is a well-known marker of genomic instability and has been associated with tumor immune phenotype—specifically, MSI-H tumors are considered "hot" and are responsive to ICIs [28,30]. As this remains an understudied phenomenon, future studies are needed to explore the influence of the tumor microenvironment on the risk of irAEs. The gut microbiome in particular closely interacts with the tumor microenvironment and is a promising avenue for future research.

The gut microbiome is a complex ecosystem consisting of symbiotic bacteria that has received immense attention in recent years concerning its influence on physiological functions. Bacterial metabolites such as short-chain fatty acids, bile acids and amino-acid derivatives have been implicated in various processes, including metabolism, inflammation, and immunity [31,32]. With the advent of immunotherapy, there is a growing body of research to show that the gut microbiome is also involved in carcinogenesis, and may modulate the effectiveness of cancer treatments, most notably immune checkpoint inhibition [33,34]. It does this by altering the composition of macrophages, natural killer cells, $CD4^+$ and $CD8^+$ T cells in the tumor immune microenvironment and enhancing tumor immunogenicity [33]. In this way, depending on the specific bacterial composition, the gut microbiome can potentially enhance antitumor immunity and increase cancer susceptibility to immunotherapy [35,36]. While beneficial in terms of cancer outcomes, this remodeling of the tumor immune microenvironment may also impact the risk of immune-related adverse events. One study by Chaput *et al* found that melanoma patients colonized with specific species of bacteria had significantly longer progression-free survival on ICIs, but had a much higher incidence of IMC [37]. Other studies since have demonstrated the impact of different microbial signatures on the incidence and severity of other irAEs. This is an especially important area to study in the realm of CRC, as gut dysbiosis is a key feature of the disease [38,39]. It is difficult to ascertain whether the altered microbiome in CRC precedes the cancer or results from it. Nonetheless, it opens up many new avenues in terms of diagnostics and therapeutics [40,41]. Fecal microbiota transplantation, in particular, has exploded in popularity, and is being explored in many clinical trials as a means to augment the efficacy of cancer therapy—especially immune checkpoint inhibition—and to mitigate toxicity to cancer medications [42-46]. It has proven to be highly effective in treating refractory IMC, leading to rapid symptom resolution in up to 85.1% of patients, and has had promising results as a first-line treatment for this irAE [47,48]. Furthermore, there are currently 2 trials underway to explore its utility in combination with ICI for treating CRC [49,50]. Its usefulness in CRC and as a means of preventing irAEs remains understudied, and could be an untapped vein for future research.

This study had several limitations. First, it was a retrospective study using electronic health records. As a result, missing information and subjective interpretation of medical records may have affected the accuracy of the data collected. Moreover, other information of interest, such as gut microbial composition, could not be collected. Given our small sample size, it is difficult to draw robust conclusions. Finally, the lack of a comparison group with different cancer types precludes the possibility of conducting further analysis beyond the descriptive findings presented.

Our study is among the first to explore the clinical manifestation of gastrointestinal irAEs in patients with CRC. We found that the CRC microenvironment may not necessarily predispose to more severe gastrointestinal irAEs compared to other cancer types. It is likely that multiple elements, such as tumor location, tumor microenvironment and gut microbiome, are all factors that affect the development of gastrointestinal irAEs in this population. More studies are needed to explore the complex interplay of these features and to further elucidate the mechanism of toxicity to checkpoint inhibition.

Acknowledgment

The authors would like to thank the Scientific Editor, Research Medical Library, The University of Texas MD Anderson Cancer Center.

Summary Box

What is already known:

- Immune checkpoint inhibitors (ICI) have recently been employed to treat microsatellite instability high colorectal cancer (CRC)
- The incidence and characteristics of inhibitor-related colitis and immune related adverse events have not been well studied in CRC
- Immune-related gastrointestinal adverse events, such as diarrhea and colitis, are one of the most common side-effects of ICI therapy

What the new findings are:

- Only 18 patients developed a gastrointestinal immune related adverse event, with an incidence of 3.8%
- Gastrointestinal immune-related adverse events in CRC patients treated with checkpoint inhibitors primarily had grade 1-2 colitis and grade 1-2 diarrhea
- In patients who had recurrence of gastrointestinal immune-related adverse events, the median grade was 2.25 for colitis and for diarrhea

References

1. Gong Z, Wang Y. Immune checkpoint inhibitor-mediated diarrhea and colitis: a clinical review. *JCO Oncol Pract* 2020;**16**:453-461.
2. Prieux-Klotz C, Dior M, Damotte D, et al. Immune checkpoint inhibitor-induced colitis: diagnosis and management. *Target Oncol* 2017;**12**:301-308.
3. Shivaji UN, Jeffery L, Gui X, et al. Immune checkpoint inhibitor-associated gastrointestinal and hepatic adverse events and their management. *Therap Adv Gastroenterol* 2019;**12**:1756284819884196.
4. Som A, Mandaliya R, Alsaadi D, et al. Immune checkpoint inhibitor-induced colitis: a comprehensive review. *World J Clin Cases* 2019;**7**:405-418.
5. Pizuorno Machado A, Shatila M, Liu C, et al. Characteristics, treatment, and outcome of patients with bowel perforation after immune checkpoint inhibitor exposure. *J Cancer Res Clin Oncol* 2023;**149**:5989-5998.
6. André T, Shiu KK, Kim TW, et al; KEYNOTE-177 Investigators. Pembrolizumab in microsatellite-instability-high advanced colorectal cancer. *N Engl J Med* 2020;**383**:2207-2218.
7. Le DT, Kim TW, Van Cutsem E, et al. Phase II open-label study of pembrolizumab in treatment-refractory, microsatellite instability-high/mismatch repair-deficient metastatic colorectal cancer: KEYNOTE-164. *J Clin Oncol* 2020;**38**:11-19.
8. Lenz HJ, Van Cutsem E, Luisa Limon M, et al. First-line nivolumab plus low-dose ipilimumab for microsatellite instability-high/ mismatch repair-deficient metastatic colorectal cancer: the phase II CheckMate 142 study. *J Clin Oncol* 2022;**40**:161-170.
9. Overman MJ, McDermott R, Leach JL, 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**:1182-1191.
10. Tran AN, Wang M, Hundt M, et al. Immune checkpoint inhibitor-associated diarrhea and colitis: a systematic review and meta-analysis of observational studies. *J Immunother* 2021;**44**:325-334.
11. Nielsen DL, Juhl CB, Chen IM, Kellermann L, Nielsen OH. Immune checkpoint Inhibitor-Induced diarrhea and Colitis: Incidence and Management. A systematic review and Meta-analysis. *Cancer Treat Rev* 2022;**109**:102440.
12. Das S, Johnson DB. Immune-related adverse events and anti-tumor efficacy of immune checkpoint inhibitors. *J Immunother Cancer* 2019;**7**:306.
13. Marley AR, Nan H. Epidemiology of colorectal cancer. *Int J Mol Epidemiol Genet* 2016;**7**:105-114.
14. American Cancer Society. Cancer Facts & Figures 2023. Cancer Statistics 2023. 2023. Available from: https://www.cancer.org/content/dam/cancer-org/research/cancer-facts-and-statistics/annual-cancer-facts-and-figures/2023/2023-cancer-facts-and-figures.pdf [Accessed 18 November 2024].
15. Benson AB, Venook AP, Al-Hawary MM, et al. Colon cancer, Version 2.2021, NCCN clinical practice guidelines in oncology. *J Natl Compr Canc Netw* 2021;**19**:329-359.
16. Chan TA, Yarchoan M, Jaffee E, et al. Development of tumor mutation burden as an immunotherapy biomarker: utility for the oncology clinic. *Ann Oncol* 2019;**30**:44-56.
17. Chang L, Chang M, Chang HM, Chang F. Microsatellite instability: a predictive biomarker for cancer immunotherapy. *Appl Immunohistochem Mol Morphol* 2018;**26**:e15-e21.
18. Michot JM, Bigenwald C, Champiat S, et al. Immune-related adverse events with immune checkpoint blockade: a comprehensive review. *Eur J Cancer* 2016;**54**:139-148.
19. Khoja L, Day D, Wei-Wu Chen T, Siu LL, Hansen AR. Tumour- and class-specific patterns of immune-related adverse events of immune checkpoint inhibitors: a systematic review. *Ann Oncol*

2017;**28**:2377-2385.
20. Rose LM, DeBerg HA, Vishnu P, et al. Incidence of skin and respiratory immune-related adverse events correlates with specific tumor types in patients treated with checkpoint inhibitors. *Front Oncol* 2020;**10**:570752.
21. Gajewski TF, Schreiber H, Fu YX. Innate and adaptive immune cells in the tumor microenvironment. *Nat Immunol* 2013;**14**:1014-1022.
22. Larkin J, Del Vecchio M, Mandalá M, et al. Adjuvant nivolumab versus ipilimumab in resected stage III/IV melanoma: 5-year efficacy and biomarker results from CheckMate 238. *Clin Cancer Res* 2023;**29**:3352-3361.
23. Ascierto PA, Del Vecchio M, Robert C, et al. Ipilimumab 10 mg/kg versus ipilimumab 3 mg/kg in patients with unresectable or metastatic melanoma: a randomised, double-blind, multicentre, phase 3 trial. *Lancet Oncol* 2017;**18**:611-622.
24. Keir ME, Butte MJ, Freeman GJ, Sharpe AH. PD-1 and its ligands in tolerance and immunity. *Annu Rev Immunol* 2008;**26**:677-704.
25. Bagchi S, Yuan R, Engleman EG. Immune checkpoint inhibitors for the treatment of cancer: clinical impact and mechanisms of response and resistance. *Annu Rev Pathol* 2021;**16**:223-249.
26. Aggarwal V, Workman CJ, Vignali DAA. LAG-3 as the third checkpoint inhibitor. *Nat Immunol* 2023;**24**:1415-1422.
27. Blum SM, Rouhani SJ, Sullivan RJ. Effects of immune-related adverse events (irAEs) and their treatment on antitumor immune responses. *Immunol Rev* 2023;**318**:167-178.
28. Wang L, Geng H, Liu Y, et al. Hot and cold tumors: immunological features and the therapeutic strategies. *MedComm (2020)* 2023;**4**:e343.
29. Bonaventura P, Shekarian T, Alcazer V, et al. Cold tumors: a therapeutic challenge for immunotherapy. *Front Immunol* 2019;**10**:168.
30. Yamamoto H, Imai K. Microsatellite instability: an update. *Arch Toxicol* 2015;**89**:899-921.
31. Jia W, Xie G, Jia W. Bile acid-microbiota crosstalk in gastrointestinal inflammation and carcinogenesis. *Nat Rev Gastroenterol Hepatol* 2018;**15**:111-128.
32. Dzutsev A, Goldszmid RS, Viaud S, Zitvogel L, Trinchieri G. The role of the microbiota in inflammation, carcinogenesis, and cancer therapy. *Eur J Immunol* 2015;**45**:17-31.
33. Lu Y, Yuan X, Wang M, et al. Gut microbiota influence immunotherapy responses: mechanisms and therapeutic strategies. *J Hematol Oncol* 2022;**15**:47.
34. Bhatt AP, Redinbo MR, Bultman SJ. The role of the microbiome in cancer development and therapy. *CA Cancer J Clin* 2017;**67**:326-344.
35. Gopalakrishnan V, Spencer CN, Nezi L, et al. Gut microbiome modulates response to anti-PD-1 immunotherapy in melanoma patients. *Science* 2018;**359**:97-103.
36. Routy B, Le Chatelier E, Derosa L, et al. Gut microbiome influences efficacy of PD-1-based immunotherapy against epithelial tumors. *Science* 2018;**359**:91-97.
37. Chaput N, Lepage P, Coutzac C, et al. Baseline gut microbiota predicts clinical response and colitis in metastatic melanoma patients treated with ipilimumab. *Ann Oncol* 2017;**28**:1368-1379.
38. Ahn J, Sinha R, Pei Z, et al. Human gut microbiome and risk for colorectal cancer. *J Natl Cancer Inst* 2013;**105**:1907-1911.
39. Song M, Chan A, Sun J. Influence of the gut microbiome, diet, and environment on risk of colorectal cancer. *Gastroenterology* 2020;**158**:322-340.
40. Zackular JP, Rogers MA, Ruffin MT 4th, Schloss PD. The human gut microbiome as a screening tool for colorectal cancer. *Cancer Prev Res (Phila)* 2014;**7**:1112-1121.
41. Zhou P, Yang D, Sun D, Zhou Y. Gut microbiome: new biomarkers in early screening of colorectal cancer. *J Clin Lab Anal* 2022;**36**:e24359.
42. Asan Medical Center. FMT with nivolumab in patients with advanced solid cancers who have progressed during anti-PD-(L)1 therapy. Available from: https://clinicaltrials.gov/study/NCT05533983 [Accessed 18 November 2024].
43. M.D. Anderson Cancer Center. Fecal microbiota transplantation in treating immune-checkpoint inhibitor induced-diarrhea or colitis in genitourinary cancer patients. Available from: https://clinicaltrials.gov/study/NCT04038619 [Accessed 18 November 2024].
44. Fondazione Policlinico Universitario Agostino Gemelli IRCCS. Fecal microbiota transplantation to improve efficacy of immune checkpoint inhibitors in renal cell carcinoma. Available from: https://clinicaltrials.gov/study/NCT04758507 [Accessed 18 November 2024].
45. Michael Scharl. Fecal microbiota transplantation in patients with malignancies not responding to immune checkpoint inhibitor therapy. Available from: https://clinicaltrials.gov/study/NCT05273255 [Accessed 18 November 2024].
46. The Netherlands Cancer Institute. FMT to convert response to immunotherapy. Available from: https://clinicaltrials.gov/study/NCT05251389 [Accessed 18 November 2024].
47. Wang Y, Varatharajalu K, Shatila M, et al. Effect of fecal transplantation on patients' reported outcome after immune checkpoint inhibitor colitis. *J Clin Oncol* 2023;**41**(16 Suppl):2645.
48. Wang Y, Varatharajalu K, Shatila M, et al. First-line treatment of fecal microbiota transplantation for immune-mediated colitis. *J Clin Oncol* 2023;**41** 16_suppl:2510.
49. Chinese Academy of Medical Sciences. FMT combined with immune checkpoint inhibitor and TKI in the treatment of CRC patients with advanced stage. Available from: https://clinicaltrials.gov/study/NCT05279677 [Accessed 18 November 2024].
50. M.D. Anderson Cancer Center. Fecal microbiota transplant and re-introduction of anti-PD-1 therapy (pembrolizumab or nivolumab) for the treatment of metastatic colorectal cancer in anti-PD-1 non-responders. Available from: https://clinicaltrials.gov/study/NCT04729322 [Accessed 18 November 2024].