World Journal of Gastrointestinal Oncology

Submit a Manuscript: https://www.f6publishing.com

World J Gastrointest Oncol 2024 March 15; 16(3): 643-652

DOI: 10.4251/wjgo.v16.i3.643

ISSN 1948-5204 (online)

MINIREVIEWS

Prognostic and predictive role of immune microenvironment in colorectal cancer

Olesya Kuznetsova, Mikhail Fedyanin, Larisa Zavalishina, Larisa Moskvina, Olga Kuznetsova, Alexandra Lebedeva, Alexey Tryakin, Galina Kireeva, Gleb Borshchev, Sergei Tjulandin, Ekaterina Ignatova

Specialty type: Oncology

Provenance and peer review: Invited article; Externally peer reviewed.

Peer-review model: Single blind

Peer-review report's scientific quality classification

Grade A (Excellent): 0
Grade B (Very good): 0
Grade C (Good): C, C
Grade D (Fair): D
Grade E (Poor): 0

P-Reviewer: Emran TB, Bangladesh; Liu Z, China; Wang PG, China

Received: November 23, 2023
Peer-review started: November 23, 2023
First decision: December 6, 2023
Revised: December 20, 2023
Accepted: January 22, 2024
Article in press: January 22, 2024
Published online: March 15, 2024

Olesya Kuznetsova, Mikhail Fedyanin, Alexey Tryakin, Sergei Tjulandin, Department of Chemotherapy, Federal State Budgetary Institution (N.N. Blokhin National Medical Research Center of Oncology) of the Ministry of Health of the Russian Federation, Moscow 115478, Russia

Larisa Zavalishina, Larisa Moskvina, Olga Kuznetsova, Department of Pathology, Russian Medical Academy of Continuous Professional Education, Moscow 123242, Russia

Alexandra Lebedeva, OncoAtlas LCC, Moscow 115478, Russia

Galina Kireeva, Gleb Borshchev, Federal State Budgetary Institution "National Medical and Surgical Center named after N.I. Pirogov" of the Ministry of Health of the Russian Federation, Moscow 105203, Russia

Ekaterina Ignatova, STOONCO: Science and Technology in Oncology, Moscow 115478, Russia

Corresponding author: Olesya Kuznetsova, MD, Academic Research, Doctor, Department of Chemotherapy, Federal State Budgetary Institution (N.N. Blokhin National Medical Research Center of Oncology) of the Ministry of Health of the Russian Federation, 23 Kashirskoye Highway, Moscow 115478, Russia. kuznetsova.o.md@gmail.com

Abstract

Colorectal cancer (CRC) represents a molecularly heterogeneous disease and one of the most frequent causes of cancer-related death worldwide. The traditional classification of CRC is based on pathomorphological and molecular characteristics of tumor cells (mucinous, ring-cell carcinomas, *etc.*), analysis of mechanisms of carcinogenesis involved (chromosomal instability, microsatellite instability, CpG island methylator phenotype) and mutational statuses of commonly altered genes (KRAS, NRAS, BRAF, APC, *etc.*), as well as expression signatures (CMS 1-4). It is also suggested that the tumor microenvironment is a key player in tumor progression and metastasis in CRC. According to the latest data, the immune microenvironment can also be predictive of the response to immune checkpoint inhibitors. In this review, we highlight how the immune environment influences CRC prognosis and sensitivity to systemic therapy.

Key Words: Immunoscore; Immune microenvironment; Colorectal cancer; Gastrointestinal cancers; Predictive biomarkers; Digital pathology

Core Tip: Here, we describe the current data and perspectives of integrating the immune microenvironment in risk assessment and treatment strategies of colorectal cancer. The identification of tumors with immunoscore showing sensitivity to immunotherapy provide insights into future clinical research.

Citation: Kuznetsova O, Fedyanin M, Zavalishina L, Moskvina L, Kuznetsova O, Lebedeva A, Tryakin A, Kireeva G, Borshchev G, Tjulandin S, Ignatova E. Prognostic and predictive role of immune microenvironment in colorectal cancer. *World J Gastrointest Oncol* 2024; 16(3): 643-652
URL: https://www.wjgnet.com/1948-5204/full/v16/i3/643.htm
DOI: https://dx.doi.org/10.4251/wjgo.v16.i3.643

INTRODUCTION

The American Joint Committee on Cancer/Union for International Cancer Control is the most common staging system used for malignant tumors. It allows for the ranking of patients by risk of progression depending on the size of the primary tumor (T), lymph node involvement (N), and the presence of distant metastases (M)[1]. However, the disease prognosis can vary significantly even within the same stage group[2]. This variability raises the need for further personalization of staging and treatment approaches. Thus, for certain tumors a list of molecular predictive and prognostic biomarkers has been proposed to choose the optimal treatment strategy[3,4].

The traditional classification of colorectal cancer (CRC) is based on pathomorphological and molecular characteristics of tumor cells (mucinous, ring-cell carcinomas, *etc.*), analysis of mechanisms of carcinogenesis involved [chromosomal instability (CIN), microsatellite instability (MSI), CpG island methylator phenotype (CIMP)] and mutational statuses of commonly altered genes (KRAS, NRAS, BRAF, APC, *etc.*), as well as expression signatures (CMS 1-4). The analysis of tumor microenvironment (TME) has been proposed as an alternative approach. This review highlights the influence of the immune environment on CRC prognosis and sensitivity to systemic therapy.

Immunological aspects of CRC

Immune evasion, or antigenic escape, occurs through various mechanisms. The production of cytokines that activate suppressor T lymphocytes and myeloid suppressor cell (MDSC) is a common mechanism of immune system evasion. It deactivates cytotoxic CD8+, CD3+, CD4+ lymphocytes and reduces the recognition of nonshared antigens. The other mechanism is the loss of major histocompatibility complex on tumor cells or programmed cell death-ligand 1 (PD-L1) activation. It leads to the depletion of peripheral T-cells and evasion of apoptosis, one of the hallmarks of cancer[5,6].

It is recognized that CRC has low immunogenicity, and the use of immunotherapy, specifically immune checkpoint inhibitors (ICIs), is only effective for a small subgroup of CRC patients. Specifically, immunotherapy is effective for tumors with impaired mechanisms of DNA mismatch repair (dMMR), which are characterized by high lymphocytic infiltration and high immunogenicity. The deficiency of MMR proteins (MLH1, PMS2, MSH2, MSH6), which are responsible for correcting mismatch errors during replication, leads to the accumulation of mutations in microsatellites (short non-coding nucleotide sequences), causing MSI. This tumor phenotype is characterized by multiple neoantigens that are recognized by the immune system. Some studies have demonstrated a positive correlation between MSI status and CD8+ T-cell infiltration[7-10]. However, MSI tumors also have peculiar mechanisms for evading the immune system, which is reflected in an increased expression of PD-L1, CTLA-4, LAG-3 and IDO ligands on myeloid cells along the tumor invasion margin (IM)[11]. Thus, PD-L1 is not only a component of the PD-1/PD-L1 system, but is also a marker of a more complex interaction between the tumor and microenvironment[12]. Droeser *et al*[13] demonstrated that high PD-L1 expression is more common for microsatellite stable/MMR proficient (MSS/pMMR) tumors (37%) than for MSI/dMMR tumors (29%). A univariate analysis found that PD-L1 expression in MSS/pMMR tumors was associated with a lower depth of invasion, absence of regional lymph node involvement and vascular invasion.

A meta-analysis by Fridman *et al*[14] demonstrated that high densities of cytotoxic CD3+, CD8+ and memory CD45RO+ T cells were associated with longer disease-free survival (DFS) after surgical resection of the primary tumor and/or overall survival (OS) in melanoma, lung, pancreatic and gastric cancers. However, there was no impact on survival for other immune cells [B-lymphocytes, natural killer cells, macrophages, T helper subsets (Th2, Th17, and Treg), MDSC]. The systematic review of 200 published trials describing the role of immune cell subsets in 20 different disease entities demonstrated that the infiltration of cytotoxic CD8+ lymphocytes was associated with a favorable prognosis in 97% of studies[15]. Additionally, Pagès *et al*[16] found that high infiltration densities of effector and memory T cells were associated with a low risk of lymphovascular and perineural invasion, as well as regional lymph node involvement among localized CRC patients. By using immunohistochemistry (IHC), the authors identified a cluster of disease characteristics negatively correlated with recurrence risk[17]. The density of CD3+, CD8+ positive cells, associated cytotoxic molecule granzyme B, CD45RO+ memory cells, in the tumor center (TC) and the IM made it possible to stratify patients into risk groups. In a multivariate analysis, the density of CD3 (TC)/CD3 (IM) lymphocytes was an independent prognostic factor [hazard ratio (HR) = 2.379; $P = 1.4 \times 10^{-6}$] in terms of DFS, and the only parameter associated with OS

(HR = 1.89; $P = 1.2 \times 10^{-5}$) after adjustment for tumor size (T), degree of differentiation, and lymph node status (N). Overall, the studies have shown that various immunological infiltrates can correlate with prognosis, yet these findings require further validation.

Role of the microenvironment in resectable colon cancer

The prognostic role of the TME, as well as its assessment, has long been a question of debate. To standardize the assessment of the TME's role, Pagès *et al*[16] created a prognostic scoring system, immunoscore (IS). This method is based on quantitative IHC of CD3+ and CD8+ lymphocyte populations in the TC and IM with the use of digital pathology for accurate assessment. This scale grades the distribution of CD3+ and CD8+ lymphocytes into five categories, with IS 0 (I0) corresponding to a low density of both cell types in the TC and IM and IS 4 (I4) corresponding to a high density. Notably, a higher IS value is associated with better patient survival.

The prognostic role of the IS system was validated in an international consortium for patients with stage I-III CRC based on the assessment of over 2500 patients[18]. Additionally, IS analysis showed a high level of reproducibility between centers and pathologists ($r = 0.97$ for primary tumor and $r = 0.97$ for IM; $P < 0.0001$). Formalin-fixed, paraffin-embedded blocks containing TC (at least 5% of the tissue) and IM (with surrounding tissues) are needed for the analysis [19]. To be applied in a research setting, two adjacent tumor slides are stained with antibodies against CD3 and CD8 using the automated BenchMark XT immunostainer (Ventana Medical System). The slides are then scanned with a Hamamatsu NanoZoomer (Hamamatsu Photonics, Japan) to convert the physical slides into digital images. This instrument applies a 20 scanning resolution mode (0.45 μm/pixel) on a single focus plane. The images are further uploaded into the software, which allows for the automatic detection of the tissue and its histological structure. After image processing, densities of CD3+ and CD8+ positive lymphocytes in the TC or IM are reported. According to an automated calculation, the density level of each marker in each region is translated to the density percentile defined previously by Pagès *et al*[16]. The mean density percentile is then calculated, categorizing IS into five classes from 0 to 4. IS classified from 0 to 1 corresponds to low (IS low), IS-2 to a moderate (IS Int) and IS-3-4 to a high CD3+ and CD8+ lymphocyte infiltrate (IS high).

According to the published data, the accuracy of predicting relapse-free survival (integrated area under the curve) for IS is comparable to staging based on T and N status. Multivariate analysis found that IS value, T and N status had a significant impact on patient survival, while the degree of tumor differentiation, perineural/lymphovascular invasion, MSI status and sex did not. The previous studies demonstrated that the prognosis of patients with MSI/dMMR locally advanced tumors depend on the degree of immune cell infiltration rather than on genomic (KRAS, NRAS, BRAF mutation status) or transcriptomic (CMS 1-4) signatures[20-22]. In a subgroup analysis of a phase III study, 600 patients with stage III CRC receiving adjuvant oxaliplatin-containing therapy were analyzed according to their IS statuses. In a group of low-risk patients (T1-3, N1), low IS was associated with poor 5-year PFS [77.5% *vs* 91.8%; HR = 1.70; 95% confidence interval (CI): 1.03-2.79; $P = 0.037$]. A similar trend was observed for high-risk patients (T4/N2) (5-year PFS for low IS 55.3% *vs* high IS 70.3%, HR = 1.65; 95%CI: 1.11-2.47; $P = 0.013$). When comparing low-risk patients with low IS and high-risk patients with high IS, long-term outcomes were similar ($P = 0.174$). In line with the previous results, a meta-analysis of 11 studies and 5718 patients by Padayao and Dy[23] confirmed the prognostic role of IS in a group of patients with localized CRC. The authors demonstrated that patients with low IS had worse PFS compared to patients with high IS (HR = 1.75, 95%CI: 1.53-2.49) and OS (HR = 1.87, 95%CI: 1.45-2.13). Taken together, these findings raise the question about the role of adjuvant chemotherapy (ACT) depending on tumor immunogenicity[24]. Overall, the results of multiple studies have validated the prognostic role of IS.

Despite the proven prognostic role of IS in early and locally advanced CRC, its impact in real clinical practice remains controversial. For instance, evidence for adjuvant treatment decisions based on IS value is still lacking. Given the disputable role of ACT in stage II CRC patients, more precise criteria are needed to determine the risk of recurrence for these patients. This question was addressed in several studies. In the analysis of quantitative infiltration of CD3+ and CD8+, Wang *et al*[25] evaluated 113 patients with stage II CRC and immune cell infiltration and demonstrated that it translated into a low (8%), intermediate (55%) or high (38%) IS value. The authors confirmed that ACT in patients with intermediate and low IS improved DFS compared with no systemic treatment after surgery (HR = 0.3; 95%CI: 0.1-0.92; $P = 0.026$)[25]. A small American study of the ACT prescribing practices for stage II CRC patients demonstrated clinicians' willingness to integrate IS data into clinical practice. The authors asked 25 practicing medical oncologists to review the clinical data of ten patients and decide whether the ACT recommendation was needed. An educational session was subsequently conducted, and the same patients' profiles were represented with added IS results. Except for a single participant (96%), all clinicians decided to change their management recommendation in more than a single case. Specifically, for the IS-high cases, recommendations for ACT decreased from 60% to 31%[26]. Several works provide conflicting data on the necessity, duration and specific regimen of ACT based on IS value[27-30] (Table 1). Therefore, the results of randomized trials are needed to further establish whether IS is ready to be introduced into routine clinical practice.

The assessment of IS using biopsy samples is crucial for timely decisions. Considering the prognostic significance of IS, a modified IS method has been developed to assess biopsy samples (ISb). The ISb method excludes the need to assess TC and considers the risk of locally advanced rectal cancer progression. Pagès *et al*[31] confirmed a positive correlation between ISb value and the degree of pathological response to neoadjuvant chemoradiotherapy (CRT). A lower risk of relapse after CRT and surgery was also demonstrated for patients with high rather than low ISb (HR = 0.21; 95%CI: 0.06-0.78; $P = 0.009$). The prognostic role of ISb for DFS was demonstrated in multivariate analysis; its value was more reliable than pre- and post-neoadjuvant radiological assessment[31]. The use of ISb was optimized to determine a cohort of patients suitable for Watch-and-Wait (WW) strategy. The patients with ISb-high had the lowest risk of recurrence when WW was chosen; the 5-year DFS in groups of high, intermediate, low ISb were 97%, 61% and 56%, respectively. In a

Table 1 Studies of immunoscore role in patients receiving adjuvant chemotherapy

Ref.	CRC stage	ACT	IS predictive role (DFS)
Church *et al* [27]	III	mFOLFOX, 3 *vs* 6 months	3-yr DFS: 3 months IS high/IS low HR = 1.80 (95%CI: 1.25-2.60); 6 months IS high/IS low HR = 2.00 (95%CI: 1.38-2.92)
Pagès *et al* [28]	III	mFOLFOX, 3 *vs* 6 months	DFS: IS Int + high: HR = 0.53; 95%CI: 0.37-0.75; *P* = 0.0004; IS low: HR = 0.84, *P* = 0.269
Mlecnik *et al* [29]	II, III	5-FU	IS 2-3 III stage: HR = 2.69 (1.02-7.11) *P* = 0.038; IS 2-3 II: HR = 1.47 (0.35-6.18) *P*: Non-significant; IS4/IS0-1: No effect of ACT was detected
Pagès *et al* [30]	III	mFOLFOX, 3 *vs* 6 months	3-yr DFS: T1-3, N1, IS high 91.4% *vs* 80.9%, *P* = 0.01; T1-3, N1, IS low 77.5% *vs* 74.5%, *P* = 0.56; T4/N2 IS high 72.0% *vs* 56.0%, *P* = 0.006; T4/N2 IS low 58.2% *vs* 52.6%, *P* = 0.2

ACT: Adjuvant chemotherapy; CI: Confidence interval; CRC: Colorectal cancer; DFS: Disease-free survival; HR: Hazard ratio; IS: Immunoscore.

multivariate analysis, ISb was independent of age, sex, and cTNM stage and was the only parameter that correlated with the time to recurrence[32]. In summary, in addition to magnetic resonance imaging data following neoadjuvant computed tomography, ISb has been demonstrated to be effective in identifying candidates for a WW strategy among patients with locally advanced rectal cancer. Therefore, the assessment of ISb might be potentially implemented into routine clinical practice.

Taken together, these data have influenced the recommendations of international societies for medical oncologists. IS has already been mentioned in European Society for Medical Oncology consensus and Pan-Asian guidelines adaptation [33,34]. It is proposed as an additional tool to TNM staging to determine prognosis and guide decision-making in ACT for low-risk stage II and stage III patients. In NCCN guidelines, IS is also discussed as a prognostic but not predictive factor in terms of the effectiveness of ACT. For this reason, as well as considering the financial costs, experts do not advise the use of this test in routine practice when assessing the risk of recurrence or when considering ACT[35].

Despite its extensive validation, the place of IS in patient management remains controversial. Moreover, the role of IS seems even more controversial due to the implementation of minimal residual disease assessment using circulating tumor DNA (ctDNA) analysis. In GALAXY, this approach was evaluated, leading to the conclusion that the most significant risk factor for recurrence was postsurgical ctDNA positivity (at 4 wk after surgery) for stage II-III patients (HR = 10.82, $P < 0.001$). Furthermore, postsurgical ctDNA positivity was identified in patients with stage II or III CRC who derived benefit from ACT (HR = 6.59, $P < 0.0001$)[36]. The subgroup analysis demonstrated that in addition to the ctDNA status, its dynamics should be considered, as the conversion of ctDNA from positive to negative from 4 to 12 wk after surgery might determine a more favorable prognosis (HR = 52.3, 95%CI: 7.2-380.5; $P < 0.001$)[37]. Polyanskaya *et al*[38] also confirmed the prognostic significance of ctDNA positivity after surgery in patients with stages I-III CRC. The 1-year PFS in the groups of positive and negative ctDNA status was 62% and 100%, respectively ($P < 0.001$). In stage II patients with negative ctDNA after surgery, disease did not progress in any case. Tie *et al*[39] demonstrated that compared to the standard-management group, a lower percentage of patients in the ctDNA-guided group received ACT (15% *vs* 28%; risk ratio = 1.82; 95%CI: 1.25-2.65). The 3-year recurrence-free survival was 86.4% among ctDNA-positive patients who received ACT and 92.5% among ctDNA-negative patients who did not. Thus, the importance of ctDNA as a factor for de-escalation of ACT was emphasized.

Despite the convincing evidence of the importance of ctDNA, clinical and morphological aspects cannot be ignored when considering ACT. Samaille *et al*[40] have demonstrated that carcinoembryonic antigen (CEA) of more than 2 ng/mL is an important prognostic factor in terms of PFS regardless of the ctDNA status and the disease stage. The authors proposed the classification using the characteristics identified in a multivariate analysis (ctDNA, CEA and stage), which, assuming the most accurate prediction of PFS, identifies patients who would benefit the most from 6 months of ACT[40]. Taken together, the question regarding adding IS to previously reported factors still needs answering. Wang *et al*[25] analyzed the correlation between IS and ctDNA in patients with stage II CRC. The authors confirmed that IS-high patients have the lowest risk of recurrence: Among 43 patients (15% - high-risk patients with T4/risk factors), there was no progression during 3 years of follow-up independent of ACT recommendation. In intermediate- and low-IS patients, there was a statistically significant difference in 3-year DFS (85% with ACT, 62% - without ACT, HR = 0.3; 95%CI: 0.1-0.92; $P = 0.026$). In 49 patients, ctDNA analysis was performed after surgery; positive status was associated with a higher risk of relapse (40% *vs* 2%, $P = 0.024$). A trend towards a higher rate of ctDNA detection in the case of low IS (tDNA positivity results in the high, intermediate and low IS was detected in 5%, 12%, 25% respectively, $P = 0.339$) was observed. The authors explain the lack of statistical significance by small sample size and propose the assessment of both IS and ctDNA to optimize approaches to the ACT. However, it is currently unclear how to implement both IS and ctDNA analysis into patient management and whether it is needed.

Microenvironment as a predictor of immunotherapy effectiveness

The predictive role of TME has been widely studied. TME plays an essential role in the efficacy of ICI therapy. Tumor infiltrating lymphocytes (TILs) are the main effectors of antitumor activity and are considered as predictive for immunotherapy response[41]. Although immunotherapy results in long-lasting anti-cancer responses in patients with advanced melanoma, lung cancer, and bladder cancer, its effectiveness is limited to a specific patient cohort[42]. Unfortunately,

there is currently no universal predictive biomarker to identify such patients, as the effectiveness of immunotherapy can be influenced by different microenvironment cell types. For example, Wang *et al*[43] confirmed that high CD4+ and CD8+ infiltration was associated with melanoma response to ipilimumab, and decreased CD8+ value in biopsies was associated with increased risk of relapse. In another prospective phase II study, the increase of TILs following ipilimumab treatment in metastatic melanoma was associated with a more pronounced response[44]. Tumeh *et al*[5] confirmed that high CD8+ infiltration in IM before treatment was associated with PD-1/PD-L1 expression and predicted a response to ICI in advanced melanoma patients. In 2015, based on data for melanoma and non-small cell lung cancer, a new classification of cancers was proposed considering the presence of TIL and PD-L1 expression[45]. The detection of TILs and PD-L1 expression in TME can be associated with a greater sensitivity to ICI. Conversely, patients with PD-L1 negative tumors without TILs usually have a poor prognosis due to low immunogenicity. However, given that the TME is a complex system, the density, location of cell distribution and lymphocyte subpopulations must also be considered. In a pan-cancer study, no correlation was observed between TILs levels prior to treatment and response to nivolumab therapy[46]. Despite convincing evidence supporting the predictive role of TME, its complexity is a limiting factor and requires standardization.

The role of TME is especially critical in dMMR/MSI disease. Immunotherapy plays a major role in advanced dMMR/MSI CRC patients[47,48]. Despite the high immunogenicity of these tumors, about 30%-50% are resistant to treatment with anti-PD1 ± anti-CTLA-4 antibodies[47-49]. In pMMR/MSS tumors, the lack of response to ICI was demonstrated in several studies. In a phase II study of 18 pretreated CRC patients, pembrolizumab monotherapy resulted in an objective response rate (ORR) of 0% and mPFS of 2.2 months. In the dMMR/MSI group, ORR was 40%, and mPFS and mOS were not reached[50]. Dual blockade with durvalumab and tremelimumab for 166 patients with pMMR/MSS CRC resulted in mPFS of 1.8 months in the treatment group and 1.9 months in the best supportive care group[51]. In a study of botensilimab and balstilimab, the ORR among 41 patients with pMMR/MSS tumors was 24%, which was higher (42%) in the absence of liver disease. Thus, these data suggest that the location of metastases has the potential to influence the response[52].

Differences in microenvironment provide a biological explanation for the unequal effectiveness of ICI in CRC among patients with MSI and MSS phenotypes. For example, pMMR/MSS tumors possess a greater number of tumor-associated macrophages, which was associated with a poor prognosis in most studies[53]. Another study demonstrated that increased activation of β-catenin by melanoma cells leads to a decrease in the population of CD8α+ and CD103+ dendritic cells, resulting in decreased recruitment of cytotoxic T-lymphocytes into the TME[54]. β-catenin is a known activator of Wnt pathway signaling in CRC. Furthermore, the APC gene, an important regulator of β-catenin, is mutated in over 70% of pMMR/MSS CRC cases[55]. Additionally, mutations altering the APC gene occur in 20% of dMMR/MSI CRC cases, which can partially influence ICI resistance. The increased Wnt/β-catenin activity in CRC is thought to be correlated with the absence of T-lymphocyte infiltration in the TME[56]. Transforming growth factor-β (TGF-β) pathway activation can provide an additional potential explanation of ICI resistance, as it promotes epithelial-to-mesenchymal transition. The tumors with this activated pathway belong to CMS4 (mesenchymal subtype)[57]. The role of TGF-β in the microenvironment regulation in *in vitro* studies was associated with an increased number of regulatory T-cells and suppression of antitumor immunity[58]. In particular, TGF-β activation was observed in liver metastases from CRC and was associated with the suppression of CD4+ and CD8+ lymphocytes[59]. Preclinical studies evaluating TGF-β tyrosine kinase inhibitors demonstrated a decrease in the incidence of CRC metastasis to the liver[60,61]. Despite the presence of other factors that explain the ineffectiveness of ICI in pMMR/MSS CRC[62], the accumulated data on the role of the microenvironment provide another promising area of application of IS in terms of candidate selection for ICI.

For instance, in CheckMate 9X8, the authors demonstrated no improvement in PFS [median 11.9 *vs* 11.9 months (HR = 0.81, 95%CI: 0.53-1.21) $P = 0.3$] or OS [median 29.1 months *vs* ND (HR = 1.03, 95%CI: 0.64-1.66)] while adding nivolumab to the first line FOLFOX + bevacizumab treatment. However, in subgroup analysis, ≥ 2% CD8+ cells in the TME identified patients with longer survival rates with the addition of ICI (mPFS 13.2 *vs* 11.8 months with the addition of nivolumab for CD8 ≥ 2 % and CD8 < 2%, respectively)[63].

In a randomized AtezoTRIBE trial comparing FOLFOXIRI, bevacizumab with FOLFOXIRI, bevacizumab and atezolizumab, an increase in PFS was demonstrated [median 13.1 *vs* 11.5 months (HR = 0.69, 95%CI: 0.56-0.85, $P = 0.012$)] but not in OS (33.0 *vs* 27.2 months, HR = 0.81, 95%CI: 0.63-1.04, $P = 0.136$) for the entire patient cohort[64,65]. The aim of the further analysis of AtezoTRIBE study was to characterize tumor immune cell infiltrate by assessing the TMB (in 65% of patients), TILs (83%), PD-L1 TPS expression (74%), IS (35%), IS IC (72%). The IS assessment methodology was based on a technique similar to the aforementioned one. For IS-IC, surgically resected specimens or biopsies from either primary tumor or metastatic sites were used, and IHC was performed with antibodies to PD-L1 and CD8. Stained slides were then scanned with a high-resolution scanner (NanoZoomer XR, Hamamatsu Photonics, Hamamatsu, Japan) to obtain 20 × digital images. The density of PD-L1+ and CD8+ cells in the tumor core were quantified by digital pathology using the HALO platform (Indica Labs, Corrales, New Mexico, United States). Five parameters, measured as linear values, were selected for inclusion into the IS-IC score density of total CD8, density of CD8-free (without PD-L1+ cell in proximity), density of CD8-cluster (CD8 cells in proximity of less than 20 micrometers of another CD8), density of PD-L1 cells, and distance between CD8-positive and PD-L1-positive cells). Depending on the described parameters, patients were divided into low risk (high CD8, PD-L1, density and small distance between them) and high-risk groups[66]. Overall, the predictive role of TME on the effect of immunotherapy has been recognized, despite a major limitation for its use in clinical practice due to the heterogeneity in its measurement.

There is some data in regard to the agreement of IS and immunogenic signatures. For instance, a poor agreement was observed between TILs and IS or IS-IC (K of Cohen < 0.20). The disadvantage of describing TILs is only a rough assessment of lymphocytes in a sample without defining T-cell populations by their function, which may incorrectly reflect the immunogenicity of the TME. Discordance of 48% between IS and TIL density was previously reported[17].

Table 2 Studies of immunoscore in different tumor types

Ref.	Tumor	Number of patients, *n*	Test	Result
Ghiringhelli *et al*[69], 2023	NSCLC	133	IS-IC	Correlation IS and PFS (HR = 0.39, 95%CI: 0.26-0.59, $P < 0.0001$), OS (HR = 0.42, 95%CI: 0.27-0.65, $P < 0.0001$). 36-month PFS IS-high 34%, IS-low 0%
Antoniotti *et al*[65], 2022	CRC	216	IS-IC	IS-high + ICI - OS HR = 0.39, 95%CI: 0.18-0.84, $P < 0.001$
Bifulco *et al*[70], 2014	Melanoma	190	IS-IC	Correlation CD8+, PD-L1- and OS $P = 0.04$

CI: Confidence interval; CRC: Colorectal cancer; HR: Hazard ratio; ICI: Immune checkpoint inhibitor; IS: Immunoscore; NSCLC: Non-small cell lung cancer; OS: Overall survival; PD-L1: Programmed cell death-ligand 1; PFS: Progression-free survival.

These differences may be attributed to the fact that IS and IS-IC are based on the assessment of predefined T-cell subtypes and describe their spatial distribution in specific tumor regions. TIL evaluation provides a semi-quantitative method for assessing unselected cell populations in randomly selected areas and is an operator-dependent technique. In a study by Boquet *et al*[67], visual scoring of CD3+ and CD8+ T-cell densities to IS performed by different pathologists were compared. The disagreement of the results with the reference IS was observed in almost half of the cases (48%). The agreement among pathologists was minimal with a Kappa of 0.34 and 0.57 before and after training, respectively. The standardized IS assay outperformed expert pathologist assessment in the clinical setting[67]. In AtezoTRIBE, among patients with pMMR tumors, there was no difference in PFS between patients with high and low TIL levels ($P = 0.36$) or with high and low PD-L1 expression ($P = 1.0$). However, regarding the impact of IS-IC, patients with a high value achieved longer PFS compared with patients with a low score (16.4 *vs* 12.2 months; HR = 0.55, 95%CI: 0.30-0.99 $P = 0.049$). Although the data on OS was immature, it was described that IS/IS-IC had no prognostic value in the case of treating patients without the use of ICI. However, these immunogenicity criteria were proven to be predictive of response to ICI. Thus, the IS-IC has the potential to identify approximately 30% of pMMR/MSS CRC patients with an activated immune microenvironment who can potentially exhibit longer-lasting response with the addition of immunotherapy. Nonetheless, this requires further validation *via* prospective randomized studies, one of which has been planned by the authors of AtezoTRIBE and IS for 2024. A few studies have indicated that this approach to selecting patients for ICI treatment can be universal for several other tumors[68] (Table 2).

CONCLUSION

The TME plays an important role in disease progression and response to antitumor treatment. Based on accumulated knowledge about TME properties, the developed IS methodology and its modifications might be implemented in clinical practice in the future. This test stratifies CRC patients into risk groups depending on the tumor infiltration by immune cells, which correlates with the prognosis in a locally advanced disease. However, introducing the IS test into routine practice to aid in ACT decision-making seems premature despite its confirmed prognostic role. Another application of immune environment data might help identify patients who are likely to respond to immunotherapy. Data from post-hoc analyses can provide valuable information to plan further studies to identify candidates for ICI treatment, even in pMMR/MSS tumors. Thus, supporting evidence from prospective studies designed with the participation of clinicians and pathologists is currently warranted to understand the possibility of incorporating characteristics of the TME into the treatment of the patients with locally advanced and metastatic disease.

FOOTNOTES

Author contributions: Kuznetsova O and Lebedeva A drafted the manuscript; Zavalishina L, Moskvina L, Kuznetsova O, and Kireeva G performed the literature search; Fedyanin M, Tryakin A, Borshchev G, Tjulandin S, and Ignatova E critically reviewed the manuscript.

Conflict-of-interest statement: All the authors report no relevant conflicts of interest for this article.

Country/Territory of origin: Russia

ORCID number: Olesya Kuznetsova 0000-0001-7753-3081; Mikhail Fedyanin 0000-0001-5615-7806; Larisa Zavalishina 0000-0002-0677-7991; Larisa Moskvina 0000-0001-8670-1366; Olga Kuznetsova 0000-0002-9721-6355; Alexandra Lebedeva 0000-0003-1920-5076; Alexey Tryakin 0000-0003-2245-214X; Galina Kireeva 0000-0002-4732-5895; Gleb Borshchev 0000-0002-8332-7521; Sergei Tjulandin 0000-0001-9807-2229; Ekaterina

Ignatova 0000-0002-8114-7885.

S-Editor: Wang JJ
L-Editor: Filipodia
P-Editor: Zhang XD

REFERENCES

1 **Amin MB**, Greene FL, Edge SB, Compton CC, Gershenwald JE, Brookland RK, Meyer L, Gress DM, Byrd DR, Winchester DP. The Eighth Edition AJCC Cancer Staging Manual: Continuing to build a bridge from a population-based to a more "personalized" approach to cancer staging. *CA Cancer J Clin* 2017; **67**: 93-99 [PMID: 28094848 DOI: 10.3322/caac.21388]
2 **Nagtegaal ID**, Quirke P, Schmoll HJ. Has the new TNM classification for colorectal cancer improved care? *Nat Rev Clin Oncol* 2011; **9**: 119-123 [PMID: 22009076 DOI: 10.1038/nrclinonc.2011.157]
3 **Koncina E**, Haan S, Rauh S, Letellier E. Prognostic and Predictive Molecular Biomarkers for Colorectal Cancer: Updates and Challenges. *Cancers (Basel)* 2020; **12** [PMID: 32019056 DOI: 10.3390/cancers12020319]
4 **Mosele F**, Remon J, Mateo J, Westphalen CB, Barlesi F, Lolkema MP, Normanno N, Scarpa A, Robson M, Meric-Bernstam F, Wagle N, Stenzinger A, Bonastre J, Bayle A, Michiels S, Bièche I, Rouleau E, Jezdic S, Douillard JY, Reis-Filho JS, Dienstmann R, André F. Recommendations for the use of next-generation sequencing (NGS) for patients with metastatic cancers: a report from the ESMO Precision Medicine Working Group. *Ann Oncol* 2020; **31**: 1491-1505 [PMID: 32853681 DOI: 10.1016/j.annonc.2020.07.014]
5 **Tumeh PC**, Harview CL, Yearley JH, Shintaku IP, Taylor EJ, Robert L, Chmielowski B, Spasic M, Henry G, Ciobanu V, West AN, Carmona M, Kivork C, Seja E, Cherry G, Gutierrez AJ, Grogan TR, Mateus C, Tomasic G, Glaspy JA, Emerson RO, Robins H, Pierce RH, Elashoff DA, Robert C, Ribas A. PD-1 blockade induces responses by inhibiting adaptive immune resistance. *Nature* 2014; **515**: 568-571 [PMID: 25428505 DOI: 10.1038/nature13954]
6 **Hanahan D**, Weinberg RA. Hallmarks of cancer: the next generation. *Cell* 2011; **144**: 646-674 [PMID: 21376230 DOI: 10.1016/j.cell.2011.02.013]
7 **Kim H**, Jen J, Vogelstein B, Hamilton SR. Clinical and pathological characteristics of sporadic colorectal carcinomas with DNA replication errors in microsatellite sequences. *Am J Pathol* 1994; **145**: 148-156 [PMID: 8030745]
8 **Dolcetti R**, Viel A, Doglioni C, Russo A, Guidoboni M, Capozzi E, Vecchiato N, Macrì E, Fornasarig M, Boiocchi M. High prevalence of activated intraepithelial cytotoxic T lymphocytes and increased neoplastic cell apoptosis in colorectal carcinomas with microsatellite instability. *Am J Pathol* 1999; **154**: 1805-1813 [PMID: 10362805 DOI: 10.1016/S0002-9440(10)65436-3]
9 **Smyrk TC**, Watson P, Kaul K, Lynch HT. Tumor-infiltrating lymphocytes are a marker for microsatellite instability in colorectal carcinoma. *Cancer* 2001; **91**: 2417-2422 [PMID: 11413533]
10 **Lal N**, Beggs AD, Willcox BE, Middleton GW. An immunogenomic stratification of colorectal cancer: Implications for development of targeted immunotherapy. *Oncoimmunology* 2015; **4**: e976052 [PMID: 25949894 DOI: 10.4161/2162402X.2014.976052]
11 **Llosa NJ**, Cruise M, Tam A, Wicks EC, Hechenbleikner EM, Taube JM, Blosser RL, Fan H, Wang H, Luber BS, Zhang M, Papadopoulos N, Kinzler KW, Vogelstein B, Sears CL, Anders RA, Pardoll DM, Housseau F. The vigorous immune microenvironment of microsatellite instable colon cancer is balanced by multiple counter-inhibitory checkpoints. *Cancer Discov* 2015; **5**: 43-51 [PMID: 25358689 DOI: 10.1158/2159-8290.CD-14-0863]
12 **Fusi A**, Festino L, Botti G, Masucci G, Melero I, Lorigan P, Ascierto PA. PD-L1 expression as a potential predictive biomarker. *Lancet Oncol* 2015; **16**: 1285-1287 [PMID: 26433815 DOI: 10.1016/S1470-2045(15)00307-1]
13 **Droeser RA**, Hirt C, Viehl CT, Frey DM, Nebiker C, Huber X, Zlobec I, Eppenberger-Castori S, Tzankov A, Rosso R, Zuber M, Muraro MG, Amicarella F, Cremonesi E, Heberer M, Iezzi G, Lugli A, Terracciano L, Sconocchia G, Oertli D, Spagnoli GC, Tornillo L. Clinical impact of programmed cell death ligand 1 expression in colorectal cancer. *Eur J Cancer* 2013; **49**: 2233-2242 [PMID: 23478000 DOI: 10.1016/j.ejca.2013.02.015]
14 **Fridman WH**, Pagès F, Sautès-Fridman C, Galon J. The immune contexture in human tumours: impact on clinical outcome. *Nat Rev Cancer* 2012; **12**: 298-306 [PMID: 22419253 DOI: 10.1038/nrc3245]
15 **Bruni D**, Angell HK, Galon J. The immune contexture and Immunoscore in cancer prognosis and therapeutic efficacy. *Nat Rev Cancer* 2020; **20**: 662-680 [PMID: 32753728 DOI: 10.1038/s41568-020-0285-7]
16 **Pagès F**, Berger A, Camus M, Sanchez-Cabo F, Costes A, Molidor R, Mlecnik B, Kirilovsky A, Nilsson M, Damotte D, Meatchi T, Bruneval P, Cugnenc PH, Trajanoski Z, Fridman WH, Galon J. Effector memory T cells, early metastasis, and survival in colorectal cancer. *N Engl J Med* 2005; **353**: 2654-2666 [PMID: 16371631 DOI: 10.1056/NEJMoa051424]
17 **Galon J**, Costes A, Sanchez-Cabo F, Kirilovsky A, Mlecnik B, Lagorce-Pagès C, Tosolini M, Camus M, Berger A, Wind P, Zinzindohoué F, Bruneval P, Cugnenc PH, Trajanoski Z, Fridman WH, Pagès F. Type, density, and location of immune cells within human colorectal tumors predict clinical outcome. *Science* 2006; **313**: 1960-1964 [PMID: 17008531 DOI: 10.1126/science.1129139]
18 **Pagès F**, Mlecnik B, Marliot F, Bindea G, Ou FS, Bifulco C, Lugli A, Zlobec I, Rau TT, Berger MD, Nagtegaal ID, Vink-Börger E, Hartmann A, Geppert C, Kolwelter J, Merkel S, Grützmann R, Van den Eynde M, Jouret-Mourin A, Kartheuser A, Léonard D, Remue C, Wang JY, Bavi P, Roehrl MHA, Ohashi PS, Nguyen LT, Han S, MacGregor HL, Hafezi-Bakhtiari S, Wouters BG, Masucci GV, Andersson EK, Zavadova E, Vocka M, Spacek J, Petruzelka L, Konopasek B, Dundr P, Skalova H, Nemejcova K, Botti G, Tatangelo F, Delrio P, Ciliberto G, Maio M, Laghi L, Grizzi F, Fredriksen T, Buttard B, Angelova M, Vasaturo A, Maby P, Church SE, Angell HK, Lafontaine L, Bruni D, El Sissy C, Haicheur N, Kirilovsky A, Berger A, Lagorce C, Meyers JP, Paustian C, Feng Z, Ballesteros-Merino C, Dijkstra J, van de Water C, van Lent-van Vliet S, Knijn N, Mușină AM, Scripcariu DV, Popivanova B, Xu M, Fujita T, Hazama S, Suzuki N, Nagano H, Okuno K, Torigoe T, Sato N, Furuhata T, Takemasa I, Itoh K, Patel PS, Vora HH, Shah B, Patel JB, Rajvik KN, Pandya SJ, Shukla SN, Wang Y, Zhang G, Kawakami Y, Marincola FM, Ascierto PA, Sargent DJ, Fox BA, Galon J. International validation of the consensus Immunoscore for the classification of colon cancer: a prognostic and accuracy study. *Lancet* 2018; **391**: 2128-2139 [PMID: 29754777 DOI: 10.1016/S0140-6736(18)30789-X]
19 **Marliot F**, Lafontaine L, Galon J. Immunoscore assay for the immune classification of solid tumors: Technical aspects, improvements and clinical perspectives. *Methods Enzymol* 2020; **636**: 109-128 [PMID: 32178816 DOI: 10.1016/bs.mie.2019.07.018]

20 **Williams DS**, Mouradov D, Jorissen RN, Newman MR, Amini E, Nickless DK, Teague JA, Fang CG, Palmieri M, Parsons MJ, Sakthianandeswaren A, Li S, Ward RL, Hawkins NJ, Faragher I, Jones IT, Gibbs P, Sieber OM. Lymphocytic response to tumour and deficient DNA mismatch repair identify subtypes of stage II/III colorectal cancer associated with patient outcomes. *Gut* 2019; **68**: 465-474 [PMID: 29382774 DOI: 10.1136/gutjnl-2017-315664]

21 **Lee H**, Sha D, Foster NR, Shi Q, Alberts SR, Smyrk TC, Sinicrope FA. Analysis of tumor microenvironmental features to refine prognosis by T, N risk group in patients with stage III colon cancer (NCCTG N0147) (Alliance). *Ann Oncol* 2020; **31**: 487-494 [PMID: 32165096 DOI: 10.1016/j.annonc.2020.01.011]

22 **Mlecnik B**, Bindea G, Angell HK, Maby P, Angelova M, Tougeron D, Church SE, Lafontaine L, Fischer M, Fredriksen T, Sasso M, Bilocq AM, Kirilovsky A, Obenauf AC, Hamieh M, Berger A, Bruneval P, Tuech JJ, Sabourin JC, Le Pessot F, Mauillon J, Rafii A, Laurent-Puig P, Speicher MR, Trajanoski Z, Michel P, Sesboüe R, Frebourg T, Pagès F, Valge-Archer V, Latouche JB, Galon J. Integrative Analyses of Colorectal Cancer Show Immunoscore Is a Stronger Predictor of Patient Survival Than Microsatellite Instability. *Immunity* 2016; **44**: 698-711 [PMID: 26982367 DOI: 10.1016/j.immuni.2016.02.025]

23 **Padayao J**, Dy C. The prognostic ability of immune scoring system Immunoscore in patients with localized colon cancer: A systematic review and meta-analysis. *J Clin Oncol* 2023; 41: 2565 [DOI: 10.1200/JCO.2023.41.16_suppl.2565]

24 **Sinicrope FA**, Shi Q, Catteau A, Poage GM, Zemla TJ, Mlecnik B, Benson AB, Gill S, Goldberg RM, Kahlenberg MS, Nair SG, Shields AF, Smyrk TC, Galon J, Alberts SR. Immunoscore Is Prognostic in Low-Risk and High-Risk Stage III Colon Carcinomas Treated With Adjuvant Infusional Fluorouracil, Leucovorin, and Oxaliplatin in a Phase III Trial. *JCO Precis Oncol* 2022; **6**: e2200010 [PMID: 35952316 DOI: 10.1200/PO.22.00010]

25 **Wang F**, Lu S, Cao D, Qian J, Li C, Zhang R, Wang F, Wu M, Liu Y, Pan Z, Wu X, Lu Z, Ding P, Li L, Lin J, Catteau A, Galon J, Chen G. Prognostic and predictive value of Immunoscore and its correlation with ctDNA in stage II colorectal cancer. *Oncoimmunology* 2023; **12**: 2161167 [PMID: 36632564 DOI: 10.1080/2162402X.2022.2161167]

26 **Kasi A**, Dotan E, Poage GM, Catteau A, Vernerey D, George M, Barzi A. Impact of Immunoscore on the Management of Stage II Colon Cancer Patients: A Physician Survey. *Cancers (Basel)* 2021; **13** [PMID: 34771628 DOI: 10.3390/cancers13215467]

27 **Church D**, Sansom O, Maka N, Edwards J, Iveson T, Saunders MP, Boukovinas I, Messaritakis I, Moustou E, Chondrozoumaki M, Georgoulias V, Kassambara A, Catteau A, Galon J, Dempsey L, Hay J, Kelly CA, Sougklakos I, Harkin A. Clinical performance of Immunoscore in stage III colorectal cancer patients in the SCOT and IDEA-HORG cohorts. *J Clin Oncol* 2022; **40**: 196 [DOI: 10.1200/JCO.2022.40.4_suppl.196]

28 **Pagès F**, André T, Taieb J, Vernerey D, Henriques J, Borg C, Marliot F, Ben Jannet R, Louvet C, Mineur L, Bennouna J, Desrame J, Faroux R, Kirilovsky A, Duval A, Laurent-Puig P, Svrcek M, Hermitte F, Catteau A, Galon J, Emile JF. Prognostic and predictive value of the Immunoscore in stage III colon cancer patients treated with oxaliplatin in the prospective IDEA France PRODIGE-GERCOR cohort study. *Ann Oncol* 2020; **31**: 921-929 [PMID: 32294529 DOI: 10.1016/j.annonc.2020.03.310]

29 **Mlecnik B**, Berger A, Pages F, Galon J. Immunoscore® as a predictor of response to chemotherapy in stage II and stage III colon cancer. *J Immunother Cancer* 2015; **3**: P89 [DOI: 10.1186/2051-1426-3-S2-P89]

30 **Pagès F**, Taieb J, Laurent-Puig P, Galon J. The consensus Immunoscore in phase 3 clinical trials; potential impact on patient management decisions. *Oncoimmunology* 2020; **9**: 1812221 [PMID: 32939329 DOI: 10.1080/2162402X.2020.1812221]

31 **Pagès F**, El Sissy C, Kirilovsky A, Custers P, Dizdarevic, Lagorce EC, Castillo-Martin M, van den Berg J, Iseas S, Loria FS, Gerard JP, Dimofte G, Perez RO, Habr-Gama A, Figueiredo N, Hansen T, Chalabi M, Galon J, Beets GL, Zeitoun G. International validation of the immunoscore-biopsy (is b) to guide selection and monitoring of patients treated with watch-and-wait (WW) strategy for rectal cancer. *J Clin Oncol* 2022; **40**: 3517 [DOI: 10.1200/JCO.2022.40.16_suppl.3517]

32 **El Sissy C**, Kirilovsky A, Van den Eynde M, Muşină AM, Anitei MG, Romero A, Marliot F, Junca A, Doyen J, Mlecnik B, Haicheur N, Fredriksen T, Lagorce C, Jouret-Mourin A, Leonard D, Bibeau F, Iseas S, Roca EL, Cabanne AM, Vaccaro CA, Santino JP, Huertas E, Tougeron D, Carvalho C, Figueiredo N, Perez RO, Habr-Gama A, Scripcariu V, Gerard JP, Galon J, Zeitoun G, Pagès F. A Diagnostic Biopsy-Adapted Immunoscore Predicts Response to Neoadjuvant Treatment and Selects Patients with Rectal Cancer Eligible for a Watch-and-Wait Strategy. *Clin Cancer Res* 2020; **26**: 5198-5207 [PMID: 32669377 DOI: 10.1158/1078-0432.CCR-20-0337]

33 **Argilés G**, Tabernero J, Labianca R, Hochhauser D, Salazar R, Iveson T, Laurent-Puig P, Quirke P, Yoshino T, Taieb J, Martinelli E, Arnold D; ESMO Guidelines Committee. Electronic address: clinicalguidelines@esmo.org. Localised colon cancer: ESMO Clinical Practice Guidelines for diagnosis, treatment and follow-up. *Ann Oncol* 2020; **31**: 1291-1305 [PMID: 32702383 DOI: 10.1016/j.annonc.2020.06.022]

34 **Yoshino T**, Argilés G, Oki E, Martinelli E, Taniguchi H, Arnold D, Mishima S, Li Y, Smruti BK, Ahn JB, Faud I, Chee CE, Yeh KH, Lin PC, Chua C, Hasbullah HH, Lee MA, Sharma A, Sun Y, Curigliano G, Bando H, Lordick F, Yamanaka T, Tabernero J, Baba E, Cervantes A, Ohtsu A, Peters S, Ishioka C, Pentheroudakis G. Pan-Asian adapted ESMO Clinical Practice Guidelines for the diagnosis treatment and follow-up of patients with localised colon cancer. *Ann Oncol* 2021; **32**: 1496-1510 [PMID: 34411693 DOI: 10.1016/j.annonc.2021.08.1752]

35 **National Comprehensive Cancer Network**. NCCN clinical practice guidelines in oncology. [cited 15 August 2023]. Available from: https://www.nccn.org/guidelines/guidelines-detail?category=1&id=1428

36 **Kotani D**, Oki E, Nakamura Y, Yukami H, Mishima S, Bando H, Shirasu H, Yamazaki K, Watanabe J, Kotaka M, Hirata K, Akazawa N, Kataoka K, Sharma S, Aushev VN, Aleshin A, Misumi T, Taniguchi H, Takemasa I, Kato T, Mori M, Yoshino T. Molecular residual disease and efficacy of adjuvant chemotherapy in patients with colorectal cancer. *Nat Med* 2023; **29**: 127-134 [PMID: 36646802 DOI: 10.1038/s41591-022-02115-4]

37 **Kotaka M**, Shirasu H, Watanabe J, Yamazaki K, Hirata K, Akazawa N, Matsuhashi N, Yokota M, Ikeda M, Kato K, Aleshin A, Sharma S, Kotani D, Oki E, Takemasa I, Kato T, Nakamura Y, Taniguchi H, Mori M, Yoshino T. Association of circulating tumor DNA dynamics with clinical outcomes in the adjuvant setting for patients with colorectal cancer from an observational GALAXY study in CIRCULATE-Japan. *J Clin Oncol* 2022; **40**: 9 [DOI: 10.1200/JCO.2022.40.4_suppl.009]

38 **Polyanskaya E**, Fedyanin M, Boyarskikh U, Kechin A, Moroz EA, Khrapov EA, Oskorobin IP, Shamovskaya DV, Aliev VA, Mammadli ZZ, Tryakin A, Filipenko ML, Tyulendin SA. The prognostic value of circulating in blood tumor DNA as a marker of minimal residual disease in stage I-III colorectal cancer. *Advances Molecul Oncol* 2022; **9**: 32-42 [DOI: 10.17650/2313-805X-2022-9-2-32-42]

39 **Tie J**, Cohen JD, Lahouel K, Lo SN, Wang Y, Kosmider S, Wong R, Shapiro J, Lee M, Harris S, Khattak A, Burge M, Harris M, Lynam J, Nott L, Day F, Hayes T, McLachlan SA, Lee B, Ptak J, Silliman N, Dobbyn L, Popoli M, Hruban R, Lennon AM, Papadopoulos N, Kinzler KW, Vogelstein B, Tomasetti C, Gibbs P; DYNAMIC Investigators. Circulating Tumor DNA Analysis Guiding Adjuvant Therapy in Stage II Colon Cancer. *N Engl J Med* 2022; **386**: 2261-2272 [PMID: 35657320 DOI: 10.1056/NEJMoa2200075]

40 **Samaille T**, Falcoz A, Cohen R, André T, Laurent-Puig P, Taieb J, Auclin E, Vernerey D. SO-17 A new risk classification integrating ctDNA,

CEA, and pTN stage for DFS prognosis and predictive value for treatment duration in stage III colon cancer. *Ann Oncol* 2023; **34**: S169 [DOI: 10.1016/j.annonc.2023.04.489]

41 **Presti D**, Dall'Olio FG, Besse B, Ribeiro JM, Di Meglio A, Soldato D. Tumor infiltrating lymphocytes (TILs) as a predictive biomarker of response to checkpoint blockers in solid tumors: A systematic review. *Crit Rev Oncol Hematol* 2022; **177**: 103773 [PMID: 35917885 DOI: 10.1016/j.critrevonc.2022.103773]

42 **Zito Marino F**, Ascierto PA, Rossi G, Staibano S, Montella M, Russo D, Alfano R, Morabito A, Botti G, Franco R. Are tumor-infiltrating lymphocytes protagonists or background actors in patient selection for cancer immunotherapy? *Expert Opin Biol Ther* 2017; **17**: 735-746 [PMID: 28318336 DOI: 10.1080/14712598.2017.1309387]

43 **Wang W**, Yu D, Sarnaik AA, Yu B, Hall M, Morelli D, Zhang Y, Zhao X, Weber JS. Biomarkers on melanoma patient T cells associated with ipilimumab treatment. *J Transl Med* 2012; **10**: 146 [PMID: 22788688 DOI: 10.1186/1479-5876-10-146]

44 **Hamid O**, Schmidt H, Nissan A, Ridolfi L, Aamdal S, Hansson J, Guida M, Hyams DM, Gómez H, Bastholt L, Chasalow SD, Berman D. A prospective phase II trial exploring the association between tumor microenvironment biomarkers and clinical activity of ipilimumab in advanced melanoma. *J Transl Med* 2011; **9**: 204 [PMID: 22123319 DOI: 10.1186/1479-5876-9-204]

45 **Teng MW**, Ngiow SF, Ribas A, Smyth MJ. Classifying Cancers Based on T-cell Infiltration and PD-L1. *Cancer Res* 2015; **75**: 2139-2145 [PMID: 25977340 DOI: 10.1158/0008-5472.CAN-15-0255]

46 **Taube JM**, Klein A, Brahmer JR, Xu H, Pan X, Kim JH, Chen L, Pardoll DM, Topalian SL, Anders RA. Association of PD-1, PD-1 ligands, and other features of the tumor immune microenvironment with response to anti-PD-1 therapy. *Clin Cancer Res* 2014; **20**: 5064-5074 [PMID: 24714771 DOI: 10.1158/1078-0432.CCR-13-3271]

47 **Diaz LA Jr**, Shiu KK, Kim TW, Jensen BV, Jensen LH, Punt C, Smith D, Garcia-Carbonero R, Benavides M, Gibbs P, de la Fourchardiere C, Rivera F, Elez E, Le DT, Yoshino T, Zhong WY, Fogelman D, Marinello P, Andre T; KEYNOTE-177 Investigators. Pembrolizumab versus chemotherapy for microsatellite instability-high or mismatch repair-deficient metastatic colorectal cancer (KEYNOTE-177): final analysis of a randomised, open-label, phase 3 study. *Lancet Oncol* 2022; **23**: 659-670 [PMID: 35427471 DOI: 10.1016/S1470-2045(22)00197-8]

48 **André T**, Lonardi S, Wong KYM, Lenz HJ, Gelsomino F, Aglietta M, Morse MA, Van Cutsem E, McDermott R, Hill A, Sawyer MB, Hendlisz A, Neyns B, Abdullaev S, Memaj A, Lei M, Dixon M, Kopetz S, Overman MJ. Nivolumab plus low-dose ipilimumab in previously treated patients with microsatellite instability-high/mismatch repair-deficient metastatic colorectal cancer: 4-year follow-up from CheckMate 142. *Ann Oncol* 2022; **33**: 1052-1060 [PMID: 35764271 DOI: 10.1016/j.annonc.2022.06.008]

49 **Le DT**, Kim TW, Van Cutsem E, Geva R, Jäger D, Hara H, Burge M, O'Neil B, Kavan P, Yoshino T, Guimbaud R, Taniguchi H, Elez E, Al-Batran SE, Boland PM, Crocenzi T, Atreya CE, Cui Y, Dai T, Marinello P, Diaz LA Jr, André T. 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 [PMID: 31725351 DOI: 10.1200/JCO.19.02107]

50 **Le DT**, Uram JN, Wang H, Bartlett BR, Kemberling H, Eyring AD, Skora AD, Luber BS, Azad NS, Laheru D, Biedrzycki B, Donehower RC, Zaheer A, Fisher GA, Crocenzi TS, Lee JJ, Duffy SM, Goldberg RM, de la Chapelle A, Koshiji M, Bhaijee F, Huebner T, Hruban RH, Wood LD, Cuka N, Pardoll DM, Papadopoulos N, Kinzler KW, Zhou S, Cornish TC, Taube JM, Anders RA, Eshleman JR, Vogelstein B, Diaz LA Jr. PD-1 Blockade in Tumors with Mismatch-Repair Deficiency. *N Engl J Med* 2015; **372**: 2509-2520 [PMID: 26028255 DOI: 10.1056/NEJMoa1500596]

51 **Chen EX**, Jonker DJ, Loree JM, Kennecke HF, Berry SR, Couture F, Ahmad CE, Goffin JR, Kavan P, Harb M, Colwell B, Samimi S, Samson B, Abbas T, Aucoin N, Aubin F, Koski SL, Wei AC, Magoski NM, Tu D, O'Callaghan CJ. Effect of Combined Immune Checkpoint Inhibition vs Best Supportive Care Alone in Patients With Advanced Colorectal Cancer: The Canadian Cancer Trials Group CO.26 Study. *JAMA Oncol* 2020; **6**: 831-838 [PMID: 32379280 DOI: 10.1001/jamaoncol.2020.0910]

52 **Bullock A**, Grossman J, Fakih M, Lenz H, Gordon M, Margolin K, Wilky BA, Mahadevan D, Trent J, Bockorny B, Moser JC, Balmanoukian A, Schlechter B, Feliu WO, Rosenthal K, Bullock B, Stebbing J, Godwin J, O'Day S, Tsimberidou A, El-Khoueiry A. LBA O-9 Botensilimab, a novel innate/adaptive immune activator, plus balstilimab (anti-PD-1) for metastatic heavily pretreated microsatellite stable colorectal cancer. *Ann Oncol* 2022; **33**: S376 [DOI: 10.1016/j.annonc.2022.04.453]

53 **Kang JC**, Chen JS, Lee CH, Chang JJ, Shieh YS. Intratumoral macrophage counts correlate with tumor progression in colorectal cancer. *J Surg Oncol* 2010; **102**: 242-248 [PMID: 20740582 DOI: 10.1002/jso.21617]

54 **Spranger S**, Bao R, Gajewski TF. Melanoma-intrinsic β-catenin signalling prevents anti-tumour immunity. *Nature* 2015; **523**: 231-235 [PMID: 25970248 DOI: 10.1038/nature14404]

55 **Sun X**, Liu S, Wang D, Zhang Y, Li W, Guo Y, Zhang H, Suo J. Colorectal cancer cells suppress CD4+ T cells immunity through canonical Wnt signaling. *Oncotarget* 2017; **8**: 15168-15181 [PMID: 28147310 DOI: 10.18632/oncotarget.14834]

56 **Luke JJ**, Bao R, Sweis RF, Spranger S, Gajewski TF. WNT/β-catenin Pathway Activation Correlates with Immune Exclusion across Human Cancers. *Clin Cancer Res* 2019; **25**: 3074-3083 [PMID: 30635339 DOI: 10.1158/1078-0432.CCR-18-1942]

57 **Guinney J**, Dienstmann R, Wang X, de Reyniès A, Schlicker A, Soneson C, Marisa L, Roepman P, Nyamundanda G, Angelino P, Bot BM, Morris JS, Simon IM, Gerster S, Fessler E, De Sousa E Melo F, Missiaglia E, Ramay H, Barras D, Homicsko K, Maru D, Manyam GC, Broom B, Boige V, Perez-Villamil B, Laderas T, Salazar R, Gray JW, Hanahan D, Tabernero J, Bernards R, Friend SH, Laurent-Puig P, Medema JP, Sadanandam A, Wessels L, Delorenzi M, Kopetz S, Vermeulen L, Tejpar S. The consensus molecular subtypes of colorectal cancer. *Nat Med* 2015; **21**: 1350-1356 [PMID: 26457759 DOI: 10.1038/nm.3967]

58 **Otegbeye F**, Ojo E, Moreton S, Mackowski N, Lee DA, de Lima M, Wald DN. Inhibiting TGF-beta signaling preserves the function of highly activated, in vitro expanded natural killer cells in AML and colon cancer models. *PLoS One* 2018; **13**: e0191358 [PMID: 29342200 DOI: 10.1371/journal.pone.0191358]

59 **Huang XM**, Zhang NR, Lin XT, Zhu CY, Zou YF, Wu XJ, He XS, He XW, Wan YL, Lan P. Antitumor immunity of low-dose cyclophosphamide: changes in T cells and cytokines TGF-beta and IL-10 in mice with colon-cancer liver metastasis. *Gastroenterol Rep (Oxf)* 2020; **8**: 56-65 [PMID: 32104586 DOI: 10.1093/gastro/goz060]

60 **Zhang B**, Halder SK, Zhang S, Datta PK. Targeting transforming growth factor-beta signaling in liver metastasis of colon cancer. *Cancer Lett* 2009; **277**: 114-120 [PMID: 19147275 DOI: 10.1016/j.canlet.2008.11.035]

61 **Ghahremanifard P**, Chanda A, Bonni S, Bose P. TGF-β Mediated Immune Evasion in Cancer-Spotlight on Cancer-Associated Fibroblasts. *Cancers (Basel)* 2020; **12** [PMID: 33291370 DOI: 10.3390/cancers12123650]

62 **Sahin IH**, Ciombor KK, Diaz LA, Yu J, Kim R. Immunotherapy for Microsatellite Stable Colorectal Cancers: Challenges and Novel Therapeutic Avenues. *Am Soc Clin Oncol Educ Book* 2022; **42**: 1-12 [PMID: 35658496 DOI: 10.1200/EDBK_349811]

63 **Lenz HJ**, Parikh A, Spigel DR, Cohn AL, Yoshino T, Kochenderfer MD, Elez E, Shao SH, Deming DA, Holdridge RC, Larson T, Chen E,

Mahipal A, Ucar A, Cullen D, Baskin-Bey E, Ledeine JM, Hammell A, Tabernero J. Nivolumab (NIVO) + 5-fluorouracil/Leucovorin/oxaliplatin (mfolfox6)/bevacizumab (BEV) vs mfolfox6/BEV for first-line (1L) treatment of metastatic colorectal cancer (mcrc): phase 2 results from checkmate 9X8. *J Clin Oncol* 2022; **40**: 8 [DOI: 10.1200/JCO.2022.40.4_suppl.008]

64 **Antoniotti C**, Rossini D, Pietrantonio F, Salvatore L, Marmorino F, Ambrosini M, Lonardi S, Cremolini C. FOLFOXIRI plus bevacizumab and atezolizumab as upfront treatment of unresectable metastatic colorectal cancer (mCRC): Updated and overall survival results of the phase II randomized AtezoTRIBE study. *J Clin Oncol* 2023; **41**: 3500 [DOI: 10.1200/JCO.2023.41.16_suppl.3500]

65 **Antoniotti C**, Rossini D, Pietrantonio F, Catteau A, Salvatore L, Lonardi S, Boquet I, Tamberi S, Marmorino F, Moretto R, Ambrosini M, Tamburini E, Tortora G, Passardi A, Bergamo F, Kassambara A, Sbarrato T, Morano F, Ritorto G, Borelli B, Boccaccino A, Conca V, Giordano M, Ugolini C, Fieschi J, Papadopulos A, Massoué C, Aprile G, Antonuzzo L, Gelsomino F, Martinelli E, Pella N, Masi G, Fontanini G, Boni L, Galon J, Cremolini C; GONO Foundation Investigators. Upfront FOLFOXIRI plus bevacizumab with or without atezolizumab in the treatment of patients with metastatic colorectal cancer (AtezoTRIBE): a multicentre, open-label, randomised, controlled, phase 2 trial. *Lancet Oncol* 2022; **23**: 876-887 [PMID: 35636444 DOI: 10.1016/S1470-2045(22)00274-1]

66 **Moretto R**, Rossini D, Catteau A, Antoniotti C, Giordano M, Boccaccino A, Ugolini C, Proietti A, Conca V, Kassambara A, Pietrantonio F, Salvatore L, Lonardi S, Tamberi S, Tamburini E, Poma AM, Fieschi J, Fontanini G, Masi G, Galon J, Cremolini C. Dissecting tumor lymphocyte infiltration to predict benefit from immune-checkpoint inhibitors in metastatic colorectal cancer: lessons from the AtezoT RIBE study. *J Immunother Cancer* 2023; **11** [PMID: 37085190 DOI: 10.1136/jitc-2022-006633]

67 **Boquet I**, Kassambara A, Lui A, Tanner A, Latil M, Lovera Y, Arnoux F, Hermitte F, Galon J, Catteau A. Comparison of Immune Response Assessment in Colon Cancer by Immunoscore (Automated Digital Pathology) and Pathologist Visual Scoring. *Cancers (Basel)* 2022; **14** [PMID: 35267475 DOI: 10.3390/cancers14051170]

68 **Hijazi A**, Antoniotti C, Cremolini C, Galon J. Light on life: immunoscore immune-checkpoint, a predictor of immunotherapy response. *Oncoimmunology* 2023; **12**: 2243169 [PMID: 37554310 DOI: 10.1080/2162402X.2023.2243169]

69 **Ghiringhelli F**, Bibeau F, Greillier L, Fumet JD, Ilie A, Monville F, Laugé C, Catteau A, Boquet I, Majdi A, Morgand E, Oulkhouir Y, Brandone N, Adam J, Sbarrato T, Kassambara A, Fieschi J, Garcia S, Lepage AL, Tomasini P, Galon J. Immunoscore immune checkpoint using spatial quantitative analysis of CD8 and PD-L1 markers is predictive of the efficacy of anti- PD1/PD-L1 immunotherapy in non-small cell lung cancer. *EBioMedicine* 2023; **92**: 104633 [PMID: 37244159 DOI: 10.1016/j.ebiom.2023.104633]

70 **Bifulco C**, Capone M, Feng Z, Madonna G, Simeone E, Curvietto M, Mozzillo N, Ciliberto G, Botti G, Fox BA, Ascierto PA. MISIPI study: Melanoma ImmunoScore evaluation in patients treated with IPIlimumab. *J Transl Med* 2014; **12**: P11 [DOI: 10.1186/1479-5876-12-S1-P11]

Published by **Baishideng Publishing Group Inc**
7041 Koll Center Parkway, Suite 160, Pleasanton, CA 94566, USA
Telephone: +1-925-3991568
E-mail: office@baishideng.com
Help Desk: https://www.f6publishing.com/helpdesk
https://www.wjgnet.com