Journal of
*Clinical Medicine*

MDPI

*Article*

# Treatment Efficacy of Immune Checkpoint Inhibitors for Patients with Advanced or Metastatic Colorectal Cancer: A Systematic Review and Meta-Analysis

**Junhee Pyo [1] and Hyo-Jung Park [2,*]**

[1] Asan Medical Center, Department of Biomedical Engineering, College of Medicine, University of Ulsan, Seoul 05505, Korea; stdpjh@mail.ulsan.ac.kr
[2] Asan Medical Center, Department of Radiology and Research Institute of Radiology, College of Medicine, University of Ulsan, Seoul 05505, Korea
* Correspondence: happyeahj@gmail.com; Tel.: +82-230-104-399

**Abstract:** The treatment efficacy of immune checkpoint inhibitors (ICIs) in colorectal cancer (CRC) has been reported heterogeneously across clinical trials. We conducted a systematic review and meta-analysis to evaluate the efficacy of ICIs in patients with advanced/metastatic CRC. Ovid-Medline was searched to identify clinical trials providing the efficacy outcomes of overall response rate (ORR) or disease control rate (DCR). The pooled ORR and DCR were estimated across all studies and subgroups. Meta-regression was performed to find the influencing factors for treatment efficacy. A total of thirty studies (1870 patients) were eligible. The overall ORR and DCR were 20.1% and 58.5%, respectively, but these results were heterogeneous across studies. Multivariate meta-regression revealed that microsatellite phenotype (odds ratio of MSI-H/dMMR versus MSS/pMMR: 1.67, $p < 0.001$) and drug regimen (odds ratio of monotherapy versus combination therapy: 1.07, $p = 0.019$) were the source of heterogeneity and also significantly influenced factors for the efficacy of the treatment. Although the efficacy of ICIs as a first-line therapy was higher than that of ICIs as the second- or more-line therapy (ORR: 51.5% vs. 13.4%, DCR: 85% vs. 49.5%), multivariate regression showed that the line of therapy was not a significant factor for the treatment efficacy. Our study suggests that the microsatellite phenotype and drug regimen, rather than the line of treatment, are the primary factors influencing the treatment response among advanced/metastatic CRC patients treated with an ICI-based regimen.

**Keywords:** colorectal cancer; immune checkpoint; microsatellite instability; systematic review; treatment response assessment



**Citation:** Pyo, J.; Park, H.-J. Treatment Efficacy of Immune Checkpoint Inhibitors for Patients with Advanced or Metastatic Colorectal Cancer: A Systematic Review and Meta-Analysis. *J. Clin. Med.* **2021**, *10*, 3599. https://doi.org/10.3390/jcm10163599

Academic Editors: Shailendra Anoopkumar-Dukie and Catherine McDermott

Received: 19 June 2021
Accepted: 12 August 2021
Published: 16 August 2021



## 1. Introduction

Colorectal cancer (CRC) remains a leading cause of cancer-related death worldwide, with a 5-year survival rate for 14.3% for patients with metastatic CRC [1,2]. In addition to the development of chemotherapies, biologics, and targeted therapies, the introduction of immunotherapy with immune checkpoint inhibitors (ICIs) including anti-programmed death 1 (PD-1), anti-programmed death-ligand 1 (PD-L1), or anti- cytotoxic T-lymphocyte antigen 4 (CTLA-4) has led to changes in the management of metastatic CRC, thereby leading to meaningful improvements in survival and radiological response [3].

The activity of ICIs has been noted for CRC with microsatellite instability (MSI) or mismatch repair deficiency (dMMR), which leads to the high mutational load and upregulated expression of multiple immune checkpoints. MSI-H/dMMR CRCs display a good response to ICI, owing to their hyper-mutated nature. Nivolumab (anti PD-1) or pembrolizumab (anti PD-1) monotherapy as well as the combination of nivolumab and ipilimumab (anti CTLA-4 agent) were found to result in a good treatment response and improved survival outcomes among patients with refractory MSI-H/dMMR metastatic CRC [4–6] and were

approved by the Food and Drug Administration (FDA) for the treatment of MSI-H/dMMR metastatic CRC that experienced disease progression following chemotherapy. Not only as a second- or more-line therapy, pembrolizumab was also demonstrated to be superior to the conventional chemotherapy when used as the first-line treatment in the phase III KEYNOTE 177 trial [7], which led to the FDA approval of pembrolizumab as the first-line treatment for patients with unresectable or metastatic MSI-H/dMMR CRC. Since then, many trials have been conducted or are ongoing to investigate the treatment effect of various ICI-based regimens in patients with untreated MSI-H/dMMR metastatic CRC [8–10]. However, no attempt has been made to generate a systematic summary on the overall treatment efficacy of ICI treatment on those patients yet.

On the other hand, microsatellite-stable (MSS) or proficient MMR (pMMR) CRC displays a low mutational load, and many ICI treatment results have been disappointing for MSS/pMMR CRC [11–13]. However, conflict results do exist regarding the treatment efficacy of combination therapy with ICIs and other agents with different mechanisms of action [14–17]. The treatment efficacy of an ICI-based treatment for patients with MSS/pMMR CRC remains to be elucidated.

With the anticipated availability of several ICI-based regimens for the management of advanced/metastatic CRC, how to plan a specific treatment course with ICI agents is an important question. Rapidly accumulating evidence from trials evaluating the treatment efficacy of ICIs warrants a summary, which would allow a more evidence-based management of patients with advanced/metastatic CRC. The aim of this study was to conduct a systematic review and meta-analysis to evaluate the treatment efficacy of ICIs for patients with advanced/metastatic CRC using clinical trial data.

## 2. Materials and Methods

### 2.1. Literature Search

The literature review and meta-analysis were conducted following the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) guidelines. A systematic computerized search of Ovid-Medline and EMBASE databases was conducted to identify relevant studies published before November 2020, with restriction to articles written in English. The following search terms were used: ((“response”[Title/Abstract] OR “response rate”[Title/Abstract] OR “overall response rate”[Title/Abstract] OR “ORR”[Title/Abstract] OR “control rate”[Title/Abstract] OR “DCR”[Title/Abstract]) AND ((“colorectal cancer” [Title/Abstract] OR “colon cancer”[Title/Abstract] OR “rectal cancer”[Title/Abstract]) AND (“immune checkpoint inhibitor”[Title/Abstract] OR “checkpoint inhibitor”[Title/Abstract] OR “checkpoint”[Title/Abstract] OR “PD-1”[Title/Abstract] OR “PD-L1”[Title/Abstract] OR “CTLA-4”[Title/Abstract] OR “pembrolizumab”[Title/Abstract] OR “nivolumab” [Title/Abstract] OR “atezolizumab”[Title/Abstract] OR “avelumab”[Title/Abstract] OR “durvalumab”[Title/Abstract])))). We also reviewed the bibliographies of the selected studies to ensure that other eligible articles were included.

### 2.2. Eligibility Criteria and Quality Assessment

Studies were eligible for inclusion if patients with CRC were treated with ICIs and were evaluated based on the efficacy outcome measures of overall response rate (ORR) or disease control rate (DCR) according to Response Evaluation Criteria for Solid Tumors (RECIST) v1.1 [18]. ORR, a direct measure of tumoricidal activity of treatment, is defined as the proportion of patients who achieve a complete response (CR) or partial response (PR) per RECIST v1.1. DCR, an index that is used to measure the tumoristatic effects of treatment, is defined as the proportion of patients who achieved a CR, PR, and stable disease (SD) per RECIST v1.1. Studies were excluded if they were animal/in vitro studies, reviews and editorials, case reports, study protocols, conference proceedings, included no CRC patients, included no ICI use, and included no interest of the study purpose. The risk of bias and methodologic quality were evaluated using the Cochrane Risk of Bias 2.0 [19]

for randomized clinical trials and the Newcastle–Ottawa scale [20] for nonrandomized trials.

### 2.3. Data Extraction and Synthesis

There were two reviewers who independently reviewed the articles based on a standardized protocol; any disagreement was resolved in a meeting where a consensus was established. The following information was extracted from the eligible studies: (a) study characteristics: authors, publication year, trial phase, and enrollment periods; (b) patient characteristics: number of patients, tumor stage, microsatellite phenotype, drug type, and treatment line; and (c) study outcomes: number of overall response and diseases controlled. To explore the treatment efficacy as measured by ORR and DCR, pooled ORR and DCR were adopted as metameters for our data synthesis.

### 2.4. Statistical Analysis

DerSimonian–Laird random-effect models were constructed to synthesize the pooled ORR and DCR percentage with 95% confidence intervals (CIs) [21]. To assess publication bias, we plotted funnel plots and conducted Begg's test to detect asymmetry [22]. To investigate the heterogeneity, we performed Cochran's Q test and $I^2$ statistics, with significance identified if the $I^2$ statistics were greater than 50% and if the *p*-value of Cochran's Q test was less than 0.10 [23].

Due to the heterogeneity of the studies included, random effects models were used to estimate the pooled effect. Univariate and multivariate meta-regression analyses were conducted to explore the influencing factors for treatment efficacy and revealed the source of heterogeneity. In the meta-regression analysis, the Knapp and Hartung adjustments were applied, which are typically adopted in the meta-regression mixed effect model to control the type 1 error rate of 0.05; these values were reported as multiplicity-adjusted *p*-value and a 95% CI [24–26]. Additionally, we performed the leave-one-out sensitivity analysis by iteratively removing one study at a time to verify the dependency of the result on a single study. Subgroup analysis was performed by calculating the pooled ORR and DCR according to each category of microsatellite phenotype and drug regimen (Category 1, monotherapy and MSI-H/dMMR; Category 2, monotherapy and MSS/pMMR; Category 3, combination therapy and MSI-H/dMMR; and Category 4, combination therapy and MSS/pMMR) in patients treated with ICIs as second- or more-line therapy. R version 4.0.3 (R foundation for Statistical Computing, Vienna, Austria) was used for analysis with the "meta" packages.

## 3. Results

### 3.1. Literature Search

Following an electronic search and a review of bibliographies, 10,290 studies were identified. Of these, 693 were excluded after a review of the study type; reviews/editorials, conference presentations, study protocols, and case reports were excluded. A review of the title and the abstract of remaining studies was conducted. Thereafter, 78 studies were retained after excluding animal/in vitro studies, studies without CRC patients, studies with no ICI use, and studies with no interest of the study purpose. After a full text review of the 78 studies, 48 studies were excluded: observational studies (n = 30); cohort overlap (n = 13); insufficient information provided on treatment response according to RECIST v1.1 (n = 3); and neoadjuvant setting (n = 2). Finally, a total of 30 eligible clinical trials were included in this systematic review and meta-analysis [5–10,14–17,27–46] (Figure 1).

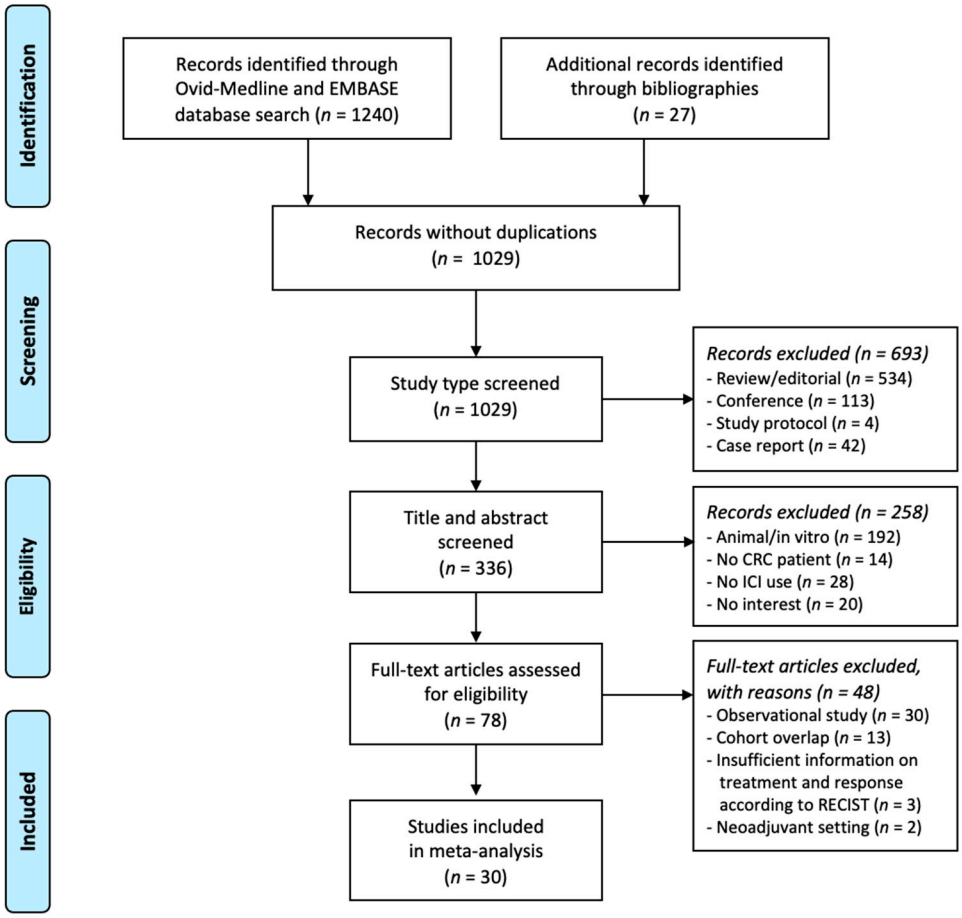



**Figure 1.** Flow diagram of the study selection process. RECIST, Response Evaluation Criteria for Solid Tumors.

### 3.2. Study Characteristics

Table 1 lists the characteristics of the 30 included clinical trials with subgroup data. The study population included patients with advanced or metastatic CRC.

**Table 1.** Characteristics of the studies included in the meta-analysis.

| Author (Year) | Phase | Sample Size | Disease State | Drug Regimen | Treatment Line | Microsatellite Phenotype | ORR [1] | DCR [1] |
|---|---|---|---|---|---|---|---|---|
| Brahmer JR et al. (2010) [28] | I | 14 | Metastatic CRC | Nivolumab | 2L+ | NR | 1 (7.1) | NR |
| Brahmer JR et al. (2012) [29] | I | 18 | Advanced/ metastatic CRC | Nivolumab | 2L+ | NR | 0 (0) | NR |
| Topalian SL et al. (2012) [44] | I | 19 | Advanced CRC | Nivolumab | 2L+ | NR | 0 (0) | NR |
| Wallin J et al. (2016) [45] | I | 23 | Metastatic CRC | Atezolizumab + BEV + FOLFOX | 1L | NR | 12 (0.5) | NR |
| Segal NH et al. (2016) [40] | II | 26 | Metastatic CRC | Pembrolizumab + (RT or RFA) | 3L+ | MSS/pMMR | 1 (3.8) | NR |
| Overman MJ et al. (2017) [5] | II | 74 | Metastatic CRC | Nivolumab | 2L+ | MSI-H/dMMR | 23 (31.1) | 51 (68.9) |
| Shahda S et al. (2017) [42] | II | 30 | Advanced CRC | Pembrolizumab + mFOLFOX6 | 1L | NR | 16 (53.3) | 30 (100) |
| Lee JJ et al. (2017) [36] | II | 30 | Metastatic CRC | Pembrolizumab + Azacitidine | 2L+ | MSS/pMMR | 1 (3.3) | 4 (13.3) |

**Table 1.** *Cont.*

| Author (Year) | Phase | Sample Size | Disease State | Drug Regimen | Treatment Line | Microsatellite Phenotype | ORR [1] | DCR [1] |
|---|---|---|---|---|---|---|---|---|
| O'Neil et al. (2017) [38] | I | 23 | Advanced/ metastatic CRC | Pembrolizumab | 2L+ | 22 MSS/pMMR, 1 MSI-H/dMMR | 1 (4.3) | 5 (21.7) |
| | | 22 | | Pembrolizumab | 2L+ | MSS/pMMR | 0 (0) | 4 (18.2) |
| | | 1 | | Pembrolizumab | 2L+ | MSI-H/dMMR | 1 (100) | 1 (100) |
| Segal NH et al. (2017) [41] | I | 14 | Metastatic CRC | Atezolizumab + CEA-CD3 TCB | 2L+ | NR | 3 (21.4) | 8 (57.1) |
| Overman MJ et al. (2018) [6] | II | 119 | Recurrent/ metastatic CRC | Nivolumab + Ipilimumab | 2L+ | MSI-H/dMMR | 65 (54.6) | 102 (85.7) |
| Eng C et al. (2019) [31] | III | 273 | Advanced/ metastatic CRC | Atezolizumab mono or combined with cobimetinib | 3L+ | NR | 7 (2.6) | 67 (24.5) |
| | | 183 | | Atezolizumab + Cobimetinib | 3L+ | NR | 5 (2.7) | 48 (26.2) |
| | | 90 | | Atezolizumab | 3L+ | NR | 2 (2.2) | 19 (21.1) |
| Floudas CS et al. (2019) [32] | I | 15 | Metastatic CRC | AMP-224 (anti pd-1) + Cy + RT | 2L+ | NR | 0 (0) | 3 (20.0) |
| Hellmann MD et al. (2019) [33] | I | 84 | Metastatic CRC | Atezolizumab + Cobimetinib | 2L+ | NR | 7 (8.3) | 26 (31.0) |
| Mettu NB et al. (2019) [37] | II | 82 | Metastatic CRC | Capecitabine + BEV + Atezolizumab | 2L+ | mostly MSS/pMMR (85.7%) | 7 (8.5) | NR |
| Cousin S et al. (2019) [17] | II | 48 | Metastatic CRC | Avelumab + Regorafenib | 2L+ | MSS/pMMR | 0 (0) | 23 (47.9) |
| Parikh AR et al. (2019) [39] | II | 40 | Metastatic CRC | Nivolumab + Ipilimumab + RT | 3L+ | MSS/pMMR | 3 (7.5) | 7 (17.5) |
| André T et al. (2020) [7] | III | 153 | Metastatic CRC | Pembrolizumab | 1L | MSI-H/dMMR | 67 (43.8) | 99 (64.7) |
| Cohen R et al. (2020) [30] | II | 57 | Metastatic CRC | Nivolumab + Ipilimumab | 2L+ | MSI-H/dMMR | 34 (59.6) | 51 (89.5) |
| Fukuoka S et al. (2020) [14] | I | 25 | Advanced/ metastatic CRC | Nivolumab + Rigorafenib | 3L+ | mostly MSS/pMMR | 9 (36.0) | 21 (84.0) |
| | | 24 | | Nivolumab + Rigorafenib | 3L+ | MSS/pMMR | 8 (33.3) | 20 (83.3) |
| | | 1 | | Nivolumab + Rigorafenib | 3L+ | MSI-H/dMMR | 1 (100) | 0 (0) |
| Kawazoe A et al. (2020) [15] | II | 50 | Metastatic CRC | Pembrolizumab + Napabucasin | 2L+ | MSI-H/dMMR or MSS/pMMR | 9 (18.0) | 27 (54.0) |
| | | 40 | Metastatic CRC | Pembrolizumab + Napabucasin | 2L+ | MSS/pMMR | 4 (10.0) | 18 (45.0) |
| | | 10 | Metastatic CRC | Pembrolizumab + Napabucasin | 2L+ | MSI-H/dMMR | 5 (50.0) | 9 (90.0) |
| Kim JH et al. (2020) [34] | II | 33 | Metastatic CRC | Avelumab | 2L+ | MSI-H/dMMR or POLE mutations | 8 (24.2) | 26 (78.8) |
| | | 21 | Metastatic CRC | Avelumab | 2L+ | MSI-H/dMMR | 6 (28.6) | 19 (90.5) |
| | | 3 | Metastatic CRC | Avelumab | 2L+ | pole | 0 (0) | 0 (0) |
| Le DT et al. (2020) [35] | II | 124 | Advanced/ metastatic CRC | Pembrolizumab | 2L+ | MSI-H/dMMR | 41 (33.1) | 67 (54.0) |
| | | 61 | | Pembrolizumab | 3L+ | MSI-H/dMMR | 20 (32.8) | 31 (50.8) |
| | | 63 | | Pembrolizumab | 2L+ | MSI-H/dMMR | 21 (33.3) | 36 (57.1) |
| Taylor K et al. (2020) [43] | II | 15 | Metastatic CRC | CC-486 + Durvalumab | 4L+ | MSS/pMMR | 0 (0) | NR |
| Yarchoan M et al. (2020) [46] | II | 17 | Metastatic CRC | GVAX + Cy + Pembrolizumab | 3L+ | MSS/pMMR | 0 (0) | 3 (17.6) |
| NCT01876511 (2020) [2] | II | 66 | Metastatic CRC | Pembrolizumab | 2L+ | MSI-H/dMMR or MSS/pMMR | | |
| | | 41 | | Pembrolizumab | 2L+ | MSI-H/dMMR | 22 (54.0) | 33 (80.0) |
| | | 25 | | Pembrolizumab | 2L+ | MSS/pMMR | 0 (0) | 4 (16.0) |
| Grothey A et al. (2018) [8] | II | 297 | Metastatic CRC | FOLFOX + BEV + Atezolizumab | 1L | NR | 49 (16.5) | 227 (76.4) |
| Lenz HJ et al. (2020) [9] | II | 45 | Metastatic CRC | Nivolumab + Ipilimumab | 1L | MSI-H/dMMR | 31 (68.9) | 38 (84.4) |

**Table 1.** *Cont.*

| Author (Year) | Phase | Sample Size | Disease State | Drug Regimen | Treatment Line | Microsatellite Phenotype | ORR [1] | DCR [1] |
|---|---|---|---|---|---|---|---|---|
| Stein A et al. (2020) [10] | II | 39 | Metastatic CRC | Avelumab + mFOLFOX6 + Cetuximab | 1L | NR | 31 (79.5) | 36 (92.3) |
| Kim R et al. (2020) [16] | I | 17 | Metastatic CRC | Nivolumab + Regorafenib | 2L+ | MSS/pMMR | 10 (58.8) | 10 (58.8) |

[1] Data are number with percentage in parentheses. [2] Available from clinicaltrials.gov/ct2/show/NCT01876511 (accessed on 17 January 2021). CRC, colorectal cancer; DCR, disease control rate; ORR, overall response rate.

There were 18 phase 2 studies, ten phase 1 studies, and two phase 3 studies. ICIs were administered as a monotherapy in ten studies or as combination therapy with other ICIs or other agent(s) with different mechanisms of action in 19 studies. There was one study that included both monotherapy and combination therapy with ICIs [31]. Patients were treated with ICIs as the first-line therapy in six studies and as the second- or more-line therapy in 24 studies. The microsatellite phenotype of CRC was MSI-H/dMMR in six studies, MSS/pMMR in seven studies, and MSI-H/dMMR or POLE mutation in one study. There were six studies that included both MSI-H/dMMR and MSS/pMMR tumors. There were ten studies that did not report the microsatellite phenotype of CRC. As an efficacy measure, the ORR and DCR were extracted for each trial.

Among the five randomized clinical trials [7,8,31,37,45], the risk of bias based on the Cochrane Risk of Bias 2.0 tool was identified to be low for the three studies, and there were some concerns for overall bias in two studies (Figure 2). For 25 non-randomized trials, the Newcastle–Ottawa scale score ranged from 6 to 8 points, indicating the high quality of the included studies (Table 2).

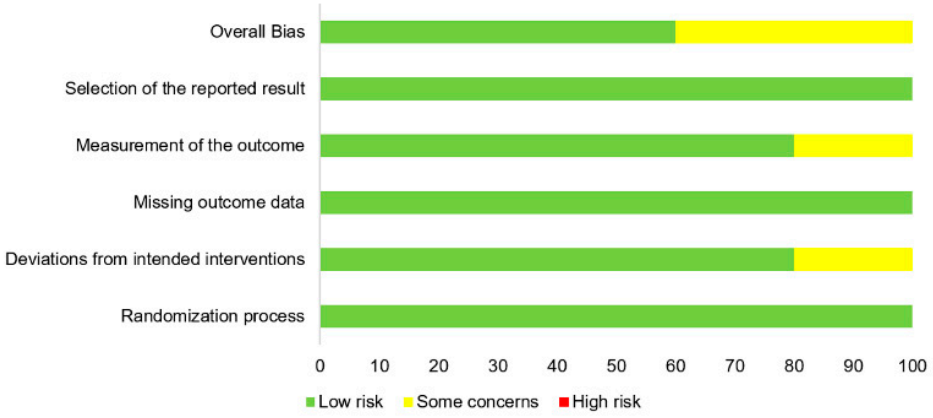



**Figure 2.** Quality assessment of the five randomized clinical trials.

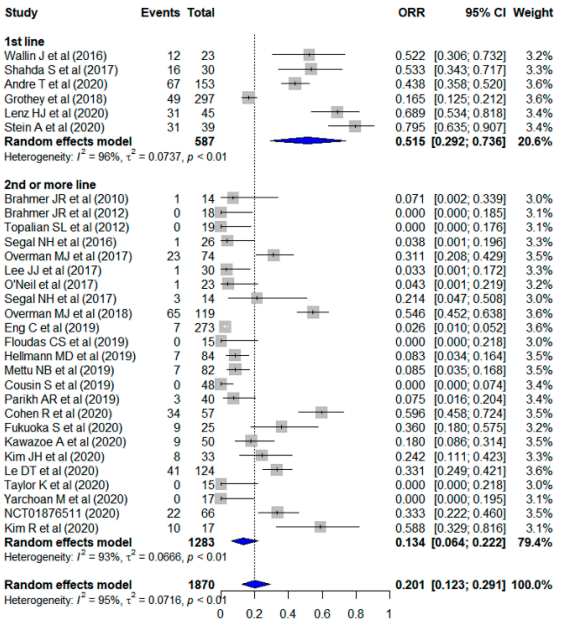
**Table 2.** The Newcastle–Ottawa scale quality assessment for the 25 non-randomized clinical trials.

| Study | Selection | | | | Comparability | Outcome | | | Total [1] |
|---|---|---|---|---|---|---|---|---|---|
| | Representativeness of the Exposed Cohort | Selection of the Non-Exposed Cohort | Ascertainment of Exposure | Demonstration That Outcome of Interest Was Not present at the Start of the Study | Comparability on the Basis of Design and Analysis | Ascertainment of Outcome | Adequate Follow-Up | Adequacy of Follow-Up of Cohorts | |
| Brahmer JR et al. (2010) [28] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Brahmer JR et al. (2012) [29] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Topalian SL et al. (2012) [44] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Segal NH et al. (2016) [40] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Overman MJ et al. (2017) [5] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Shahda S et al. (2017) [42] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Lee JJ et al. (2017) [36] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| O'Neil et al. (2017) [38] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Segal NH et al. (2017) [41] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Overman MJ et al. (2018) [6] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Floudas CS et al. (2019) [32] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Hellmann MD et al. (2019) [33] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Cousin S et al. (2019) [17] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Parikh AR et al. (2019) [39] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Cohen R et al. (2020) [30] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Fukuoka S et al. (2020) [14] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Kawazoe A et al. (2020) [15] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Kim JH et al. (2020) [34] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Le DT et al. (2020) [35] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Taylor K et al. (2020) [43] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Yarchoan M et al. (2020) [46] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| NCT01876511 (2020) | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Lenz HJ et al. (2020) [9] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |
| Stein A et al. (2020) [10] | 1 | 1 | 1 | 1 | 2 | 1 | 0 | 0 | 7 |
| Kim R et al. (2020) [16] | 1 | 1 | 1 | 1 | 2 | 1 | 1 | 1 | 9 |

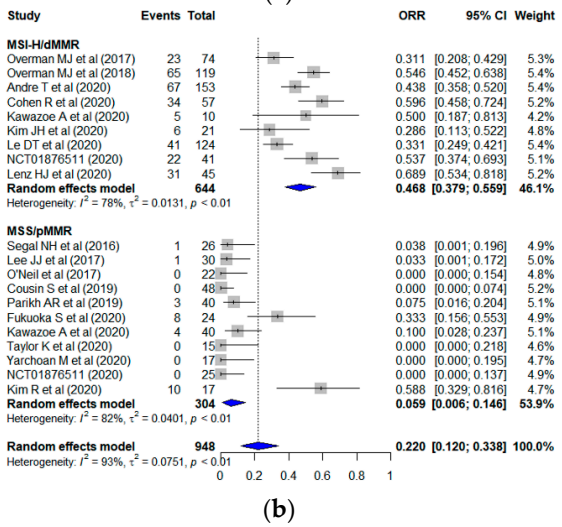
[1] Each study could be awarded a maximum of 9: a maximum of 2 for the item regarding comparability, and a maximum of 1 for the other seven items.

### 3.3. Efficacy Endoints: ORR and DCR

The pooled ORR from all of the studies was 20.1% (95% CI, 12.3–29.1%) (Figure 3a). The leave-one-out sensitivity analysis revealed that the pooled ORR ranged from 18.3% to 21.3% when each study was excluded. The funnel plot revealed no publication bias across the studies ($p = 0.475$) (Figure S1). The pooled estimates of ORR in the subgroups are shown in Figure 3a–c. The pooled ORR among patients treated with ICIs as the first-line and second- or more-line was 51.5% (95% CI, 29.2–73.6%) and 13.4% (95% CI, 6.4–22.2%), respectively; significant difference ($p = 0.003$) was observed between the two values. According to the microsatellite phenotype, the pooled ORR was 46.8% (95% CI, 37.9–55.9%) in MSI-H/dMMR tumors and 5.9% (95% CI, 0.6–14.6%) in MSS/pMMR tumors ($p < 0.001$). Regarding drug regimen, the pooled ORR of patients treated with the ICI monotherapy was 14.2% (95% CI, 5.3–26.0%); which was slightly lower than that of patients treated with the combination therapy (22.4%; 95% CI, 11.8–35.0%) ($p = 0.332$). Heterogeneity was observed for all of the studies ($I^2 = 94\%$) and each subgroup (first line, $I^2 = 96\%$; second- or more-line, $I^2 = 93\%$; MSI-H/dMMR, $I^2 = 83\%$; MSS/pMMR, $I^2 = 94\%$; ICI monotherapy, $I^2 = 92\%$; combination therapy, $I^2 = 95\%$).

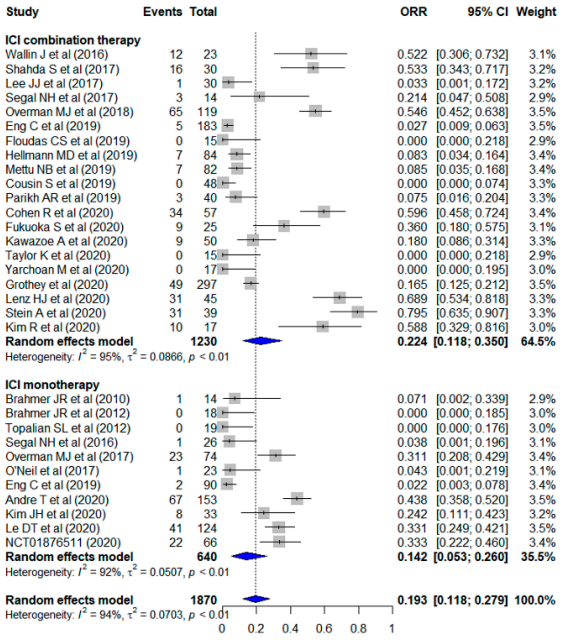



(**a**)




(**b**)

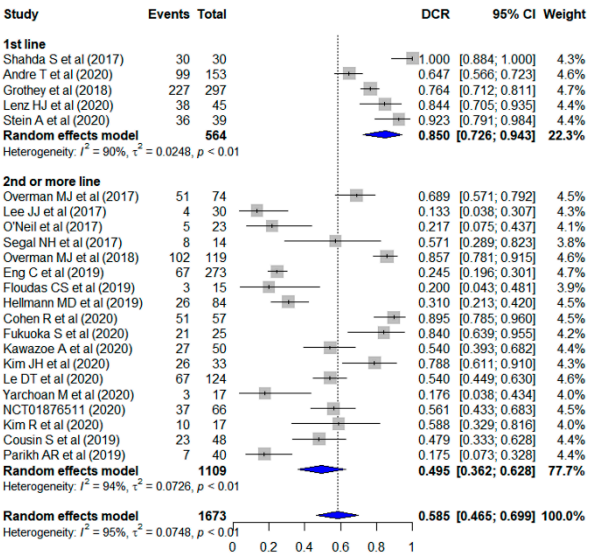
**Figure 3.** *Cont.*



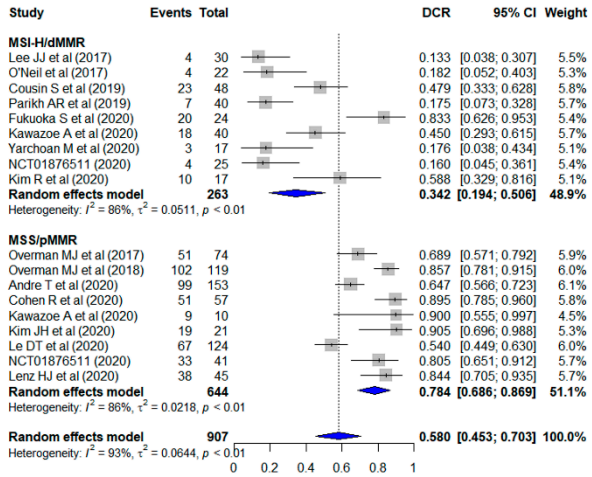

(**c**)

**Figure 3.** Forest plots displaying the pooled ORR according to (**a**) treatment line, (**b**) microsatellite phenotype, and (**c**) drug regimen. The blue quadrilaterial indicates the pooled incidence and its 95% confidence interval.

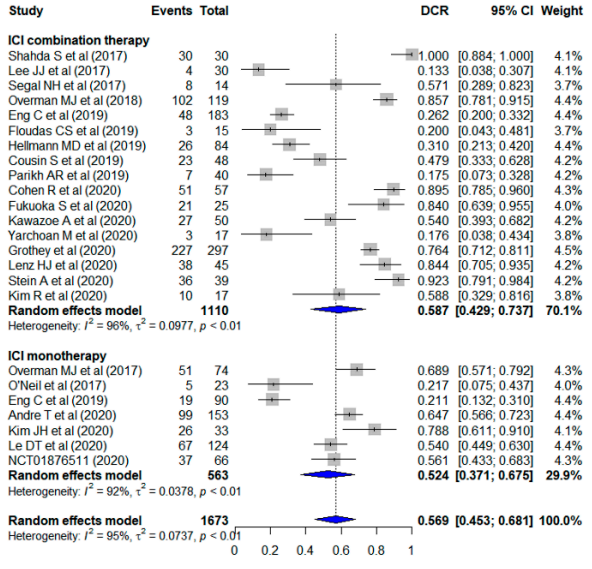
The pooled DCR from all of the studies was 58.5% (95% CI, 46.5–70.0%) (Figure 4a). When the leave-one-out sensitivity analysis was conducted, the pooled DCR was found to range from 55.8% to 60.7%. The funnel plot showed no publication bias across studies based on the Begg's test ($p$ = 0.508), which was conducted to test the funnel plot asymmetry (Figure S2). The pooled estimates of DCR in the subgroups are shown in Figure 4a–c. The pooled DCR was 85.0% (95% CI, 72.6–94.3%) for patients treated with ICI as the first-line therapy and 49.5% (95% CI, 36.2–62.8%) for those treated with ICI as the second- or more-line therapy ($p$ = 0.006). According to the microsatellite phenotype, the pooled DCR was 78.4% (95% CI, 68.6–86.9%) in tumors with MSI-H/dMMR and 34.2% (95% CI, 19.4–50.6%) in tumors with MSS/pMMR ($p$ < 0.001). Regarding drug regimen, the pooled DCR of the patients treated with the ICI monotherapy was 52.4% (95% CI, 37.1–67.5%) and that of the patients treated with the combination therapy was 58.7% (95% CI, 42.9–73.7%); no significant difference was identified ($p$ = 0.632). Heterogeneity was observed for all studies ($I^2$ = 94%) and each treatment line group (first-line, $I^2$ = 90%; second- or more-line, $I^2$ = 94%), microsatellite phenotype (MSI-H/dMMR, $I^2$ = 86%; MSS/pMMR, $I^2$ = 86%), and drug regimen (ICI monotherapy, $I^2$ = 92%; combination therapy, $I^2$ = 96%).




**(a)**




**(b)**




**(c)**

**Figure 4.** Forest plots displaying the pooled DCR according to (**a**) treatment line, (**b**) microsatellite phenotype, and (**c**) drug regimen.

In three studies in which patients with MSS/pMMR tumors were treated with ICI combined with regorafenib as the second- or more-line therapy [14,16,17], the DCR ranged from 47.9% to 83.3%, while the other regimens ranged from 13.3% to 45.0%. Among the three studies with ICI plus regorafenib, two studies performed by Fukuoka et al. [14] and Kim et al. [16] also showed markedly higher ORR (33.3% and 58.8%, respectively) than any other regimen (range, 0–07.5%).

### 3.4. Meta-Regression Analysis

We explored the influencing factors for the ORR and DCR by performing a meta-regression. In the univariate analysis, patients treated with ICI as the second- or more-line had a significantly lower ORR than those treated with ICI as the first-line treatment (Odds ratio [OR], 0.67; 95% CI, 0.52–0.86). Patients with MSI-H/dMMR tumors had significantly higher ORR (OR, 1.61; 95% CI, 1.37–1.90) than those with MSS/pMMR tumors. Slightly higher ORR was observed for patients treated with ICI combination therapy than for those treated with the ICI monotherapy (OR, 1.11; 95% CI, 0.89–1.39). In the multivariate analysis with covariates of the treatment line, microsatellite phenotype and drug regimen, and microsatellite phenotype and drug regimen appeared to significantly influence ORR. The OR of patients with MSI-H/dMMR tumors was 1.67 (95% CI, 1.42–1.98) with patients having MSS/pMMR tumors as the reference, and the OR of patients with ICI combination therapy was 1.24 (95% CI, 1.02–1.49) with patients who received ICI monotherapy as the reference (Table 3). The treatment line showed no significant difference between the first-line and the second- or more-line (OR, 0.90; 95% CI, 0.71–1.15 with the first-line as the reference).

**Table 3.** Results of the univariate and multivariate meta-regression analyses.

| Treatment Efficacy | Parameter | Pooled Estimate (%) | Univariate Analysis | | Multivariate Analysis | |
|---|---|---|---|---|---|---|
| | | | Odds Ratio | *p* Value | Odds Ratio | *p* Value |
| ORR | Treatment line | | | 0.003 | | 0.394 |
| | First-line | 51.5 (29.2–73.6) | Reference | | Reference | |
| | Second- or more-line | 13.4 (6.4–22.2) | 0.67 (0.52–0.86) | | 0.90 (0.71–1.15) | |
| | Microsatellite phenotype | | | <0.001 | | <0.001 |
| | MSS/pMMR | 5.9 (0.6–14.6) | Reference | | Reference | |
| | MSI-H/dMMR | 46.8 (37.9–55.9) | 1.61 (1.37–1.90) | | 1.67 (1.42–1.98) | |
| | Drug regimen | | | 0.332 | | 0.019 |
| | ICI monotherapy | 14.2 (5.3–26.0) | Reference | | Reference | |
| | ICI combination therapy | 22.4 (11.8–35.0) | 1.11 (0.89–1.39) | | 1.21 (1.04–1.42) | |
| DCR | Treatment line | | | 0.006 | | 0.613 |
| | First-line | 85.0 (72.6–94.3) | Reference | | Reference | |
| | Second- or more-line | 49.5 (36.2–62.8) | 0.68 (0.52–0.88) | | 1.07 (0.81–1.41) | |
| | Microsatellite phenotype | | | <0.001 | | <0.001 |
| | MSS/pMMR | 34.2 (19.4–50.6) | Reference | | Reference | |
| | MSI-H/dMMR | 78.4 (68.6–86.9) | 1.57 (1.29–1.91) | | 1.72 (1.41–2.10) | |
| | Drug regimen | | | 0.632 | | 0.032 |
| | ICI monotherapy | 52.4 (37.1–67.5) | Reference | | Reference | |
| | ICI combination therapy | 58.7 (42.9–73.7) | 1.07 (0.81–1.39) | | 1.24 (1.02–1.49) | |

Numbers in parenthesis are 95% confidence intervals. DCR, disease control rate; ICI, immune checkpoint inhibitor; MSI-H, microsatellite instability-high; MSS, microsatellite instability-stable; ORR, overall response rate.

The univariate and multivariate meta-regression analyses for DCR showed a similar trend to those for ORR. In the univariate analysis, the cohorts treated with ICIs as the second- or more-line treatment had significantly lower DCR than those treated with ICIs as the first-line treatment (OR, 0.68; 95% CI, 0.52–0.88). Patients with MSI-H/dMMR tumors had significantly higher DCR (OR, 1.57; 95% CI, 1.29–1.91) than those with MSS/pMMR tumors. Patients treated with ICI combination therapy had slightly higher DCR than those treated with ICI monotherapy (OR, 1.07; 95% CI, 0.81–1.39). In the multivariate analysis, the microsatellite phenotype and the drug regimen appeared to be the significant

factor influencing DCR (OR, 1.72 (95% CI, 1.41–2.10) for MSI-H/dMMR; OR, 1.24 (95% CI, 1.02–1.49) for ICI combination therapy) (Table 3).

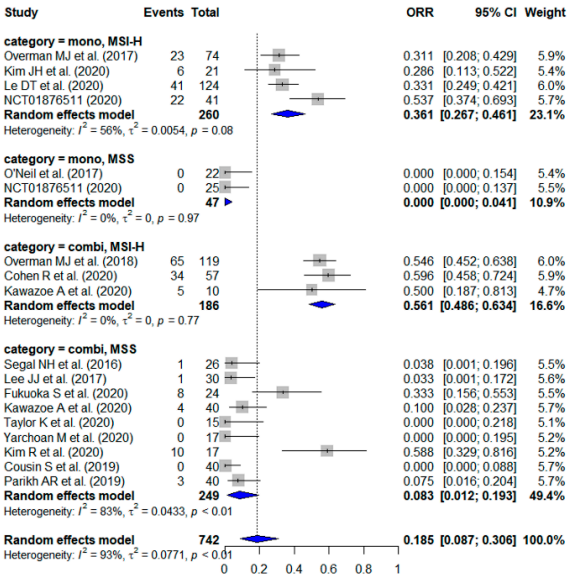
### 3.5. Subgroup Analysis

The pooled incidence of ORR and DCR in subgroups according to the microsatellite phenotype and drug regimen in patients treated with ICIs as second- or more-line therapy are provided in Table 4 with forest plots in Figure 5. Regarding ORR, the pooled ORR of Category 3 (combination therapy, MSI-H) was the highest (56.0%), followed by Category 1 (monotherapy, MSI-H; 36.2%), Category 4 (combination therapy, MSS; 10.9%), and Category 2 (monotherapy, MSS; 2.0%). With the exception of Category 2 versus Category 4 ($p = 0.318$), all inter-category comparisons showed significant differences ($p \leq 0.017$). As for DCR, the same trend was observed as in ORR. The pooled DCR was the highest in Category 3 (combination therapy, MSI-H; 87.3%), followed by Category 1 (monotherapy, MSI-H; 73.0%), Category 4 (combination therapy, MSS; 41.5%), and Category 2 (monotherapy, MSS; 17.0%). With the exception of Category 1 vs. Category 3 ($p = 0.32$) and Category 2 vs. Category 4 ($p = 0.142$), all of the comparisons showed significant differences ($p \leq 0.011$). Heterogeneity decreased in Category 1 and cCtegory 4 in pooling the of the ORR ($I^2 = 56\%$ and 83%, respectively) and the DCR ($I^2 = 84\%$ and 88%, respectively), and there was no heterogeneity in the pooling of both estimates in Category 2 and Category 3 ($I^2 = 0\%$).

**Table 4.** Subgroup analyses according to the microsatellite phenotype and drug regimen in patients treated with ICIs as second- or more-line therapy.

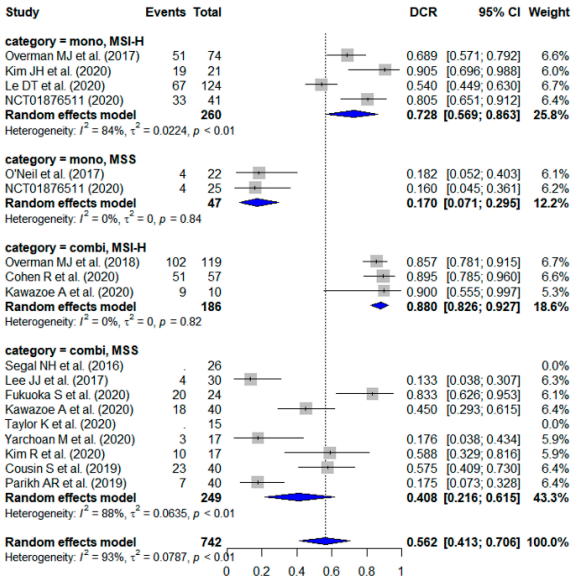
| Treatment Efficacy | Category | Pooled Estimate (%) | *p* Value | | | |
|---|---|---|---|---|---|---|
| | | | vs. Category 1 | vs. Category 2 | vs. Category 3 | vs. Category 4 |
| ORR | Category 1 (mono, MSI-H) | 36.1 (26.7–46.1) | - | <0.001 | 0.017 | <0.001 |
| | Category 2 (mono, MSS) | 0.0 (0.5–4.1) | <0.001 | - | <0.001 | 0.318 |
| | Category 3 (combi, MSI-H) | 56.1 (48.6–63.4) | 0.017 | <0.001 | - | <0.001 |
| | Category 4 (combi, MSS) | 8.3 (1.2–19.3) | <0.001 | 0.318 | < 0.001 | - |
| DCR | Category 1 (mono, MSI-H) | 72.8 (56.9–86.3) | - | 0.001 | 0.320 | 0.011 |
| | Category 2 (mono, MSS) | 17.0 (7.1–29.5) | 0.001 | - | <0.001 | 0.142 |
| | Category 3 (combi, MSI-H) | 88.0 (82.6–92.7) | 0.320 | <0.001 | - | <0.001 |
| | Category 4 (combi, MSS) | 40.8 (21.6–61.5) | 0.011 | 0.142 | <0.001 | - |

DCR, disease control rate; ORR, overall response rate.




**(a)**

**Figure 5.** *Cont.*




**(b)**

**Figure 5.** Forest plots of the (**a**) pooled ORR and (**b**) DCR of each subgroup according to the microsatellite phenotype and drug regimen in patients treated with ICIs as second- or more-line therapy.

## 4. Discussion

In this study, we evaluated the treatment efficacy of ICI-based therapy for patients with advanced/metastatic CRC using the data from 30 clincial trials. The findings of this meta-analysis indicate that the microsatellite phenotype of tumor and drug regimen significantly influence the treatment response of patients with advanced/metastatic CRC administered ICI. In MSS/pMMR tumors, a durable response was noted in the second- or more-line treatment when ICI was administered as part of a combination treatment.

We used ORR and DCR as the primary endpoints of our analysis. Although survival time such as overall survival has been regarded as the most reliable metric for assessing the efficacy of anticancer treatment, overall survival has several drawbacks when used a primary endpoint in clinical trials; the sample size needs to large enough, a much longer follow-up is required than other endpoints as the time to event (i.e., death) is much longer, and the analysis may be much more confounded than other endpoints by the effect of salvage therapies used after disease progression [47]. Since the ORR and DCR are the most commonly used primary or secondary endpoints in clinical trials, we could summarize the results using these endpoints from the most clinical trials to acquire more generalizable summary estimates.

Our study showed that the microsatellite phenotype significantly affected treatment efficacy, irrespective of the treatment line (i.e., first-line or second- or more-line) and drug regimen (i.e., ICI monotherapy or combination), as revealed by multivariate meta-regression analysis. Treatment efficacy was markedly higher in patients with MSI-H/dMMR tumors than in those with MSS/pMMR tumors. The pooled ORR and DCR of patients with MSI-H/dMMR tumors were 46.8% and 78.4%, respectively, while the pooled ORR and DCR of those with MSS/pMMR tumors were 5.9% and 34.2%, respectively. This finding is consistent with previous studies where a remarkable ICI efficacy was observed for patients with advanced or metastatic CRC and other solid tumors that are MSI-H or dMMR [5,48]. In addition to the MSI status, high mutational load (i.e., tumor mutational burden) and upregulated expression of PD-1/PD-L1 have been reported to be associated with an increased response rate to ICI treatment [49]. The association between MSI status and tumor mutation burden or the upregulated expression of multiple immune checkpoints has been suggested [50,51]. However, the relationship among these biomarkers is still unclear and needs further investigation.

Regarding the drug regimen, ICI combination therapy (i.e., ICI with other ICI or non-immunotherapy drugs) resulted in a higher treatment response rate than ICI monotherapy. The pooled ORR and DCR of patients treated with the ICI combination therapy were 22.4% and 58.7%, respectively, while the pooled ORR and DCR of those treated with ICI monotherapy were 14.2% and 52.4%, respectively. Although the overall pooled ORR was not found to differ between ICI monotherapy and the combination therapy in univariate analysis, the combination therapy resulted in a significantly higher ORR than the monotherapy (OR, 1.21; 95% CI, 1.04–1.42) when stratified by the line of treatment and microsatellite phenotype. The combination therapy also resulted in a significantly higher DCR than monotherapy in the multivariate analysis (OR, 1.24; 95% CI, 1.02–1.49).

The explicitly higher treatment efficacy of combination therapy including ICI and regorafenib [14,16,17] than the other regimens administered to patients with MSS/pMMR tumors administered ICI as a second- or more-line treatment is worth recognizing. The DCR of three studies on ICI plus rigorafenib ranged from 47.9% to 83.3%, while the DCR of other regimens ranged from 13.3% to 45.0%. Among the three studies, two studies performed by Fukuoka et al. [14] and Kim et al. [16] also showed markedly higher ORR (33.3% and 58.8%, respectively) than other regimens (range, 0–07.5%). This is surprising, considering the widespread concept of poor treatment efficacy and outcome in patients with MSS/pMMR tumors. Regorafenib is a multi-kinase inhibitor that targets a wide range of tyrosine kinases associated with oncogenesis, angiogenesis, and tumor microenvironment control [52]. The clinical potential of kinase inhibitors in combination with ICIs has been reported [53], which possibly results from the role of the kinase inhibitor in increasing tumor immunogenicity. Although low treatment efficacy has been reported [11–13], the long-lasting treatment response owing to the above combination therapies suggests the potential of ICI treatment for patients with MSS/pMMR CRC. The combination effect of ICI and other drugs with different mechanisms of action in MSS/pMMR CRC patients is an understudied topic [54] and is worth of further exploration.

Based on the univariate meta-regression analysis, both ORR and DCR were significantly higher in patients who were administered ICI as the first-line treatment than in those who were administered ICI as the second- or more-line treatment. However, it was revealed not to be an influencing factor for the treatment efficacy in multivariate analysis. Such findings might be due to the more frequent use of the ICI combination therapy as the first-line treatment instead of as the second- or more-line treatment. However, the number of studies that administered combination therapy as the first-line treatment was small, which limits inference on the efficacy of first-line ICI treatment for patients with untreated advanced/metastatic CRC. Therefore, further trials are needed to determine the treatment efficacy according to the line of ICI treatment. The results of the ongoing trials of ICIs administered to patients with untreated metastatic CRC are highly anticipated.

Our study had some limitations. First, the number of included trials with ICI as the first-line therapy was small. Nevertheless, all of the available information for the clinical trials performed to date has been included herein. The results of our study could serve as a basis for future studies when sufficent new data become available. Further, our findings will contribute to finding biologically meaningful combination therapies containing ICIs. Second, the included studies were highly heterogeneous, and this heterogeneity precluded us from acquiring a solid meta-analytic summary estimate of ORR and DCR across all 30 studies. When pooling all of the studies, the DerSimonian and Laird method was used, which is based on the normal approximation to result in mean value. However, this might be quite a strong assumption even if 30 studies were included, as the included studies were very heterogeneous. To reveal and explain the study heterogeneity and its reason for using a systematic approach, we explored the influencing factors for ORR and DCR by performing the meta-regression and subgroup analysis. Although the heterogeneity substantially decreased in subgroup analyses according to the microsatellite phenotype and drug regimen, significant heterogeneity still existed in the pooled ORR and DCR in Category 1 (monotherapy, MSI-H) and Category 4 (combination therapy, MSS; Table 4), and

there could be other unknown source of heterogeneity. Third, our analysis to determine the cause of heterogeneity and influencing factors for treatment efficacy was limited by only useing the information that was available in the included studies (i.e., microsatellite phenotype, treatment line, and drug regimen (monotherapy vs. Combination)), and our finding that the microsatellite phenotype and drug regimen are the influencing factors of the treatment response in CRC patients after ICI treatment has been previously proposed. However, we validated those results through a comprehensive review and meta-analysis using the available clinical trial data to date by extracting all of the available and categorizable data from the included studies. Additionally, one of our findings that combination therapy with ICI and regorafenib as a second- or more-line treatment showed high efficacy in patients with MSS/pMMR tumors is worth noticing, which suggests the potential of ICI treatment for patients with MSS/pMMR CRC, and further research is required to figure out the nature of antitumoral response and an effective ICI regimen in MSS/pMMR CRC. Considering that the objectives of the systematic review and meta-analysis include obtaining more valid and generalizable values of the estimates of interest and identifying areas for further research, we believe our study is pertinent.

## 5. Conclusions

We evaluated the treatment efficacy of the ICI-based therapy for patients with advanced/metastatic CRC by pooling the currently available clinical trial data. While the treatment efficacy was heterogeneous across the trials, the microsatellite phenotype and drug regimen were the primary factors influencing the treatment response. While most regimens showed low treatment efficacy for MSS/pMMR CRC, a durable response was noted in the second- or more-line treatment when ICI was administered as part of a combination treatment, which suggests the potential of ICI treatment for MSS/pMMR CRCs and indicates the need for further research.

**Supplementary Materials:** The following are available online at https://www.mdpi.com/article/10.3390/jcm10163599/s1, Figure S1: Funnel plot for the overall response rate, Figure S2: Funnel plot for the disease control rate.

**Author Contributions:** Conceptualization, H.-J.P.; methodology, J.P. and H.-J.P.; formal analysis, J.P.; investigation, J.P. and H.-J.P.; resources, H.-J.P.; data curation, J.P. and H.-J.P.; writing—original draft preparation, J.P. and H.-J.P.; writing—review and editing, H.-J.P.; supervision, H.-J.P.; project administration, H.-J.P.; funding acquisition, H.-J.P. All authors have read and agreed to the published version of the manuscript.

**Funding:** This research was funded by the National Research Foundation of Korea (NRF) grant funded by the Korea government (MSIT) (No. 2021R1C1C1010138).

**Institutional Review Board Statement:** Not applicable.

**Informed Consent Statement:** Not applicable.

**Data Availability Statement:** Data sharing is not applicable to this article as no new data were created or analyzed in this study.

**Conflicts of Interest:** The authors declare no conflict of interest.

## References

1. National Cancer Institute. Surveillance, Epidemiology, and End Results Program: Cancer Stat Facts: Colon and Rectum Cancer. Available online: https://seer.cancer.gov/statfacts/html/colorect.html (accessed on 20 January 2021).
2. Arnold, M.; Rutherford, M.; Lam, F.; Bray, F.; Ervik, M.; Soerjomataram, I. ICBP SURVMARK-2 Online Tool: International Cancer Survival Benchmarking. Lyon, France: International Agency for Research on Cancer. Available online: http://gco.iarc.fr/survival/survmark (accessed on 20 January 2021).
3. Havel, J.J.; Chowell, D.; Chan, T.A. The evolving landscape of biomarkers for checkpoint inhibitor immunotherapy. *Nat. Rev. Cancer* **2019**, *19*, 133–150. [CrossRef]
4. Le, D.T.; Uram, J.N.; Wang, H.; Bartlett, B.R.; Kemberling, H.; Eyring, A.D.; Skora, A.D.; Luber, B.S.; Azad, N.S.; Laheru, D.; et al. PD-1 Blockade in Tumors with Mismatch-Repair Deficiency. *N. Engl. J. Med.* **2015**, *372*, 2509–2520. [CrossRef] [PubMed]

5. Overman, M.J.; McDermott, R.; Leach, J.L.; Lonardi, S.; Lenz, H.J.; Morse, M.A.; Desai, J.; Hill, A.; Axelson, M.; Moss, R.A.; 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. [CrossRef]
6. Overman, M.J.; Lonardi, S.; Wong, K.Y.; Lenz, H.J.; Gelsomino, F.; Aglietta, M.; Morse, M.A.; Van Cutsem, E.; McDermott, R.; Hill, A.; et al. Durable Clinical Benefit with Nivolumab Plus Ipilimumab in DNA Mismatch Repair-Deficient/Microsatellite Instability-High Metastatic Colorectal Cancer. *J. Clin. Oncol.* **2018**, *36*, 773–779. [CrossRef] [PubMed]
7. André, T.; Shiu, K.K.; Kim, T.W.; Jensen, B.V.; Jensen, L.H.; Punt, C.; Smith, D.; Garcia-Carbonero, R.; Benavides, M.; Gibbs, P.; et al. Pembrolizumab in Microsatellite-Instability-High Advanced Colorectal Cancer. *N. Engl. J. Med.* **2020**, *383*, 2207–2218. [CrossRef] [PubMed]
8. Grothey, A.; Tabernero, J.; Arnold, D.; De Gramont, A.; Ducreux, M.P.; O'Dwyer, P.J.; Van Cutsem, E.; Bosanac, I.; Srock, S.; Mancao, C.; et al. Fluoropyrimidine (FP) + bevacizumab (BEV) + atezolizumab vs FP/BEV in BRAFwt metastatic colorectal cancer (mCRC): Findings from Cohort 2 of MODUL—A multicentre, randomized trial of biomarker-driven maintenance treatment following first-line induction therapy. *Ann. Oncol.* **2018**, *29*, viii714–viii715.
9. Lenz, H.-J.; Lonardi, S.; Zagonel, V.; Van Cutsem, E.; Limon, M.L.; Wong, K.Y.; Hendlisz, A.; Aglietta, M.; Garcia-Alfonso, P.; Neyns, B.; et al. Nivolumab plus low-dose ipilimumab as first-line therapy in microsatellite instability-high/DNA mismatch repair deficient metastatic colorectal cancer: Clinical update. *J. Clin. Oncol.* **2020**, *38*, 11. [CrossRef]
10. Stein, A.; Binder, M.; Goekkurt, E.; Lorenzen, S.; Riera-Knorrenschild, J.; Depenbusch, R.; Ettrich, T.J.; Doerfel, S.; Al-Batran, S.E.; Karthaus, M.; et al. Avelumab and cetuximab in combination with FOLFOX in patients with previously untreated metastatic colorectal cancer (MCRC): Final results of the phase II AVETUX trial (AIO-KRK-0216). *J. Clin. Oncol.* **2020**, *38*, 96. [CrossRef]
11. Hegde, P.S.; Karanikas, V.; Evers, S. The Where, the When, and the How of Immune Monitoring for Cancer Immunotherapies in the Era of Checkpoint Inhibition. *Clin. Cancer Res.* **2016**, *22*, 1865–1874. [CrossRef]
12. O'neil, B.H.; Wallmark, J.; Lorente, D.; Elez, E.; Raimbourg, J.; Gomez-Roca, C.; Ejadi, S.; Piha-Paul, S.A.; Moss, R.A.; Siu, L.L. 502 Pembrolizumab (MK-3475) for patients (pts) with advanced colorectal carcinoma (CRC): Preliminary results from KEYNOTE-028. *Eur. J. Cancer* **2015**, *51*, 30304–30305. [CrossRef]
13. Picard, E.; Verschoor, C.P.; Ma, G.W.; Pawelec, G. Relationships Between Immune Landscapes, Genetic Subtypes and Responses to Immunotherapy in Colorectal Cancer. *Front. Immunol.* **2020**, *11*, 369. [CrossRef]
14. Fukuoka, S.; Hara, H.; Takahashi, N.; Kojima, T.; Kawazoe, A.; Asayama, M.; Yoshii, T.; Kotani, D.; Tamura, H.; Mikamoto, Y.; et al. Regorafenib Plus Nivolumab in Patients with Advanced Gastric or Colorectal Cancer: An Open-Label, Dose-Escalation, and Dose-Expansion Phase Ib Trial (REGONIVO, EPOC1603). *J. Clin. Oncol.* **2020**, *38*, 2053–2061. [CrossRef]
15. Kawazoe, A.; Kuboki, Y.; Shinozaki, E.; Hara, H.; Nishina, T.; Komatsu, Y.; Yuki, S.; Wakabayashi, M.; Nomura, S.; Sato, A.; et al. Multicenter Phase I/II Trial of Napabucasin and Pembrolizumab in Patients with Metastatic Colorectal Cancer (EPOC1503/SCOOP Trial). *Clin. Cancer Res.* **2020**, *26*, 5887–5894. [CrossRef] [PubMed]
16. Kim, R.; Imanirad, I.; Carballido, E.; Strosberg, J.; Kim, Y.; Kim, D. Phase I/IB study of regorafenib and nivolumab in mismatch repair proficient advanced refractory colorectal cancer. *Ann. Oncol.* **2020**, *31*, 239. [CrossRef]
17. Cousin, S.; Bellera, C.A.; Guégan, J.P.; Gomez-Roca, C.A.; Metges, J.P.; Adenis, A.; Pernot, S.; Cantarel, C.; Kind, M.; Toulmonde, M.; et al. REGOMUNE: A phase II study of regorafenib plus avelumab in solid tumors—Results of the non-MSI-H metastatic colorectal cancer (mCRC) cohort. *J. Clin. Oncol.* **2020**, *38*, 4019. [CrossRef]
18. Eisenhauer, E.A.; Therasse, P.; Bogaerts, J.; Schwartz, L.H.; Sargent, D.; Ford, R.; Dancey, J.; Arbuck, S.; Gwyther, S.; Mooney, M.; et al. New response evaluation criteria in solid tumours: Revised RECIST guideline (version 1.1). *Eur. J. Cancer* **2009**, *45*, 228–247. [CrossRef] [PubMed]
19. Sterne, J.A.; Savović, J.; Page, M.J.; Elbers, R.G.; Blencowe, N.S.; Boutron, I.; Cates, C.J.; Cheng, H.Y.; Corbett, M.S.; Eldridge, S.M.; et al. RoB 2: A revised tool for assessing risk of bias in randomised trials. *BMJ* **2019**, *366*, 14898. [CrossRef] [PubMed]
20. Wells, G.A.; Shea, B.; O'Connell, D.; Peterson, J.; Welch, V.; Losos, M.; Tugwell, P. The Newcastle-Ottawa Scale (NOS) for Assessing the Quality of Nonrandomised Studies in Meta-Analyses. Available online: http://www.ohri.ca/programs/clinical_epidemiology/oxford.asp (accessed on 11 August 2021).
21. DerSimonian, R.; Laird, N. Meta-analysis in clinical trials. *Control. Clin. Trials* **1986**, *7*, 177–188. [CrossRef]
22. Egger, M.; Smith, G.D.; Schneider, M.; Minder, C. Bias in meta-analysis detected by a simple, graphical test. *BMJ* **1997**, *315*, 629–634. [CrossRef] [PubMed]
23. Higgins, J.P.T.; Thompson, S.G.; Deeks, J.J.; Altman, D.G. Measuring inconsistency in meta-analyses. *BMJ* **2003**, *327*, 557–560. [CrossRef]
24. Higgins, J.P.; Thompson, S.G. Controlling the risk of spurious findings from meta-regression. *Stat. Med.* **2004**, *23*, 1663–1682. [CrossRef]
25. Knapp, G.; Hartung, J. Improved tests for a random effects meta-regression with a single covariate. *Stat. Med.* **2003**, *22*, 2693–2710. [CrossRef]
26. IntHout, J.; Ioannidis, J.P.A.; Borm, G.F. The Hartung-Knapp-Sidik-Jonkman method for random effects meta-analysis is straightforward and considerably outperforms the standard DerSimonian-Laird method. *BMC Med. Res. Methodol.* **2014**, *14*, 25. [CrossRef]
27. Study of MK-3475 in Patients with Microsatellite Unstable (MSI) Tumors (Cohorts A, B and C). Available online: https://www.clinicaltrials.gov/ct2/show/NCT01876511 (accessed on 17 January 2021).

28. Brahmer, J.R.; Drake, C.G.; Wollner, I.; Powderly, J.D.; Picus, J.; Sharfman, W.H.; Stankevich, E.; Pons, A.; Salay, T.M.; McMiller, T.L.; et al. Phase I study of single-agent anti-programmed death-1 (MDX-1106) in refractory solid tumors: Safety, clinical activity, pharmacodynamics, and immunologic correlates. *J. Clin. Oncol.* **2010**, *28*, 3167–3175. [CrossRef]
29. Brahmer, J.R.; Tykodi, S.S.; Chow, L.Q.; Hwu, W.J.; Topalian, S.L.; Hwu, P.; Drake, C.G.; Camacho, L.H.; Kauh, J.; Odunsi, K.; et al. Safety and activity of anti-PD-L1 antibody in patients with advanced cancer. *N. Engl. J. Med.* **2012**, *366*, 2455–2465. [CrossRef] [PubMed]
30. Cohen, R.; Bennouna, J.; Meurisse, A.; Tournigand, C.; De La Fouchardière, C.; Tougeron, D.; Borg, C.; Mazard, T.; Chibaudel, B.; Garcia-Larnicol, M.L.; et al. RECIST and iRECIST criteria for the evaluation of nivolumab plus ipilimumab in patients with microsatellite instability-high/mismatch repair-deficient metastatic colorectal cancer: The GERCOR NIPICOL phase II study. *J. Immunother. Cancer* **2020**, *8*, e001499. [CrossRef] [PubMed]
31. Eng, C.; Kim, T.W.; Bendell, J.; Argilés, G.; Tebbutt, N.C.; Di Bartolomeo, M.; Falcone, A.; Fakih, M.; Kozloff, M.; Segal, N.H.; et al. Atezolizumab with or without cobimetinib versus regorafenib in previously treated metastatic colorectal cancer (IMblaze370): A multicentre, open-label, phase 3, randomised, controlled trial. *Lancet Oncol.* **2019**, *20*, 849–861. [CrossRef]
32. Floudas, C.S.; Brar, G.; Mabry-Hrones, D.; Duffy, A.G.; Wood, B.; Levy, E.; Krishnasamy, V.; Fioravanti, S.; Bonilla, C.M.; Walker, M.; et al. A Pilot Study of the PD-1 Targeting Agent AMP-224 Used with Low-Dose Cyclophosphamide and Stereotactic Body Radiation Therapy in Patients with Metastatic Colorectal Cancer. *Clin. Colorectal Cancer* **2019**, *18*, e349–e360. [CrossRef]
33. Hellmann, M.D.; Kim, T.W.; Lee, C.B.; Goh, B.C.; Miller, W.H., Jr.; Oh, D.Y.; Jamal, R.; Chee, C.E.; Chow, L.Q.; Gainor, J.F.; et al. Phase Ib study of atezolizumab combined with cobimetinib in patients with solid tumors. *Ann. Oncol.* **2019**, *30*, 1134–1142. [CrossRef]
34. Kim, J.H.; Kim, S.Y.; Baek, J.Y.; Cha, Y.J.; Ahn, J.B.; Kim, H.S.; Lee, K.W.; Kim, J.W.; Kim, T.Y.; Chang, W.J.; et al. A Phase II Study of Avelumab Monotherapy in Patients with Mismatch Repair-Deficient/Microsatellite Instability-High or POLE-Mutated Metastatic or Unresectable Colorectal Cancer. *Cancer Res. Treat.* **2020**, *52*, 1135–1144. [CrossRef] [PubMed]
35. Le, D.T.; Kim, T.W.; Van Cutsem, E.; Geva, R.; Jäger, D.; Hara, H.; Burge, M.; O'Neil, B.; Kavan, P.; Yoshino, T.; 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. [CrossRef]
36. Lee, J.J.; Sun, W.; Bahary, N.; Ohr, J.; Rhee, J.C.; Stoller, R.G.; Marks, S.M.; Lembersky, B.C.; Beasley, H.S.; Drummond, S.; et al. Phase 2 study of pembrolizumab in combination with azacitidine in subjects with metastatic colorectal cancer. *J. Clin. Oncol.* **2017**, *35*, 3054. [CrossRef]
37. Mettu, N.B.; Twohy, E.; Ou, F.S.; Halfdanarson, T.R.; Lenz, H.J.; Breakstone, R.; Boland, P.M.; Crysler, O.; Wu, C.; Grothey, A.; et al. BACCI: A phase II randomized, double-blind, multicenter, placebo-controlled study of capecitabine (C) bevacizumab (B) plus atezolizumab (A) or placebo (P) in refractory metastatic colorectal cancer (mCRC): An ACCRU network study. *Ann. Oncol.* **2019**, *30*, v203. [CrossRef]
38. O'Neil, B.H.; Wallmark, J.M.; Lorente, D.; Elez, E.; Raimbourg, J.; Gomez-Roca, C.; Ejadi, S.; Piha-Paul, S.A.; Stein, M.N.; Abdul Razak, A.R.; et al. Safety and antitumor activity of the anti-PD-1 antibody pembrolizumab in patients with advanced colorectal carcinoma. *PLoS ONE* **2017**, *12*, e0189848. [CrossRef]
39. Parikh, A.R.; Clark, J.W.; Wo, J.Y.; Yeap, B.Y.; Allen, J.N.; Blaszkowsky, L.S.; Ryan, D.P.; Giantonio, B.J.; Weekes, C.D.; Zhu, A.X.; et al. A phase II study of ipilimumab and nivolumab with radiation in microsatellite stable (MSS) metastatic colorectal adenocarcinoma (mCRC). *J. Clin. Oncol.* **2019**, *37*, 3514. [CrossRef]
40. Segal, N.H.; Kemeny, N.E.; Cercek, A.; Reidy, D.L.; Raasch, P.J.; Warren, P.; Hrabovsky, A.E.; Campbell, N.; Shia, J.; Goodman, K.A.; et al. Non-randomized phase II study to assess the efficacy of pembrolizumab (Pem) plus radiotherapy (RT) or ablation in mismatch repair proficient (pMMR) metastatic colorectal cancer (mCRC) patients. *J. Clin. Oncol.* **2016**, *34*, 3539. [CrossRef]
41. Segal, N.H.; Saro, J.; Melero, I.A.; Ros, W.; Argiles, G.; Marabelle, A.; Ruiz, M.R.; Albanell, J.; Calvo, E.; Moreno, V.; et al. Phase I studies of the novel carcinoembryonic antigen T-cell bispecific (CEA-CD3 TCB) antibody as a single agent and in combination with atezolizumab: Preliminary efficacy and safety in patients (pts) with metastatic colorectal cancer (mCRC). *Ann. Oncol.* **2017**, *28*, v134. [CrossRef]
42. Shahda, S.; Noonan, A.M.; Bekaii-Saab, T.S.; O'Neil, B.H.; Sehdev, A.; Shaib, W.L.; Helft, P.R.; Loehrer, P.J.; Tong, Y.; Liu, Z.; et al. A phase II study of pembrolizumab in combination with mFOLFOX6 for patients with advanced colorectal cancer. *J. Clin. Oncol.* **2017**, *35*, 3541. [CrossRef]
43. Taylor, K.; Yau, H.L.; Chakravarthy, A.; Wang, B.; Shen, S.Y.; Ettayebi, I.; Ishak, C.A.; Bedard, P.L.; Razak, A.A.; Hansen, A.R.; et al. An open-label, phase II multicohort study of an oral hypomethylating agent CC-486 and durvalumab in advanced solid tumors. *J. Immunother. Cancer* **2020**, *8*, e000883. [CrossRef] [PubMed]
44. Topalian, S.L.; Hodi, F.S.; Brahmer, J.R.; Gettinger, S.N.; Smith, D.C.; McDermott, D.F.; Powderly, J.D.; Carvajal, R.D.; Sosman, J.A.; Atkins, M.B.; et al. Safety, activity, and immune correlates of anti-PD-1 antibody in cancer. *N. Engl. J. Med.* **2012**, *366*, 2443–2454. [CrossRef] [PubMed]
45. Wallin, J.; Pishvaian, M.J.; Hernandez, G.; Yadav, M.; Jhunjhunwala, S.; Delamarre, L.; He, X.; Powderly, J.; Lieu, C.; Eckhardt, S.G.; et al. Clinical activity and immune correlates from a phase Ib study evaluating atezolizumab (anti-PDL1) in combination with FOLFOX and bevacizumab (anti-VEGF) in metastatic colorectal carcinoma. *Cancer Res.* **2016**, *76*, 2651.

46. Yarchoan, M.; Huang, C.Y.; Zhu, Q.; Ferguson, A.K.; Durham, J.N.; Anders, R.A.; Thompson, E.D.; Rozich, N.S.; Thomas, D.L.; Nauroth, J.M.; et al. A phase 2 study of GVAX colon vaccine with cyclophosphamide and pembrolizumab in patients with mismatch repair proficient advanced colorectal cancer. *Cancer Med.* **2020**, *9*, 1485–1494. [CrossRef]
47. Saad, E.D.; Buyse, M. Statistical controversies in clinical research: End points other than overall survival are vital for regulatory approval of anticancer agents. *Ann. Oncol.* **2016**, *27*, 373–378. [CrossRef]
48. Le, D.T.; Durham, J.N.; Smith, K.N.; Wang, H.; Bartlett, B.R.; Aulakh, L.K.; Lu, S.; Kemberling, H.; Wilt, C.; Luber, B.S.; et al. Mismatch repair deficiency predicts response of solid tumors to PD-1 blockade. *Science* **2017**, *357*, 409–413. [CrossRef]
49. Luchini, C.; Bibeau, F.; Ligtenberg, M.J.; Singh, N.; Nottegar, A.; Bosse, T.; Miller, R.; Riaz, N.; Douillard, J.Y.; Andre, F.; et al. ESMO recommendations on microsatellite instability testing for immunotherapy in cancer, and its relationship with PD-1/PD-L1 expression and tumour mutational burden: A systematic review-based approach. *Ann. Oncol.* **2019**, *30*, 1232–1243. [CrossRef] [PubMed]
50. Schrock, A.B.; Ouyang, C.; Sandhu, J.; Sokol, E.; Jin, D.; Ross, J.S.; Miller, V.A.; Lim, D.; Amanam, I.; Chao, J.; et al. Tumor mutational burden is predictive of response to immune checkpoint inhibitors in MSI-high metastatic colorectal cancer. *Ann. Oncol.* **2019**, *30*, 1096–1103. [CrossRef]
51. Zhang, Y.; Sun, Z.; Mao, X.; Wu, H.; Luo, F.; Wu, X.; Zhou, L.; Qin, J.; Zhao, L.; Bai, C. Impact of mismatch-repair deficiency on the colorectal cancer immune microenvironment. *Oncotarget* **2017**, *8*, 85526–85536. [CrossRef] [PubMed]
52. Fondevila, F.; Méndez-Blanco, C.; Fernández-Palanca, P.; González-Gallego, J.; Mauriz, J.L. Anti-tumoral activity of single and combined regorafenib treatments in preclinical models of liver and gastrointestinal cancers. *Exp. Mol. Med.* **2019**, *51*, 1–15. [CrossRef] [PubMed]
53. Ahn, R.; Ursini-Siegel, J. Clinical Potential of Kinase Inhibitors in Combination with Immune Checkpoint Inhibitors for the Treatment of Solid Tumors. *Int. J. Mol. Sci.* **2021**, *22*, 2608. [CrossRef]
54. Hermel, D.J.; Sigal, D. The Emerging Role of Checkpoint Inhibition in Microsatellite Stable Colorectal Cancer. *J. Pers. Med.* **2019**, *9*, 5. [CrossRef] [PubMed]