Systematic review of combinations of targeted or immunotherapy in advanced solid tumors

Aaron C Tan,1,2 Stephen J Bagley,3 Patrick Y Wen,4 Michael Lim,5 Michael Platten,6,7 Howard Colman,8 David M Ashley,9 Wolfgang Wick,6 Susan M Chang,10 Evanthia Galanis,11 Alirea Mansouri,12 Simon Khagi,13 Minesh P Mehta,14 Amy B Heimberger,15 Vinay K Puduvali,16 David A Reardon,4 Solmaz Sahebjam,17 John Simes,18 Scott J Antonia,9 Don Berry,19 Mustafa Khasraw

ABSTRACT

With rapid advances in our understanding of cancer, there is an expanding number of potential novel combination therapies, including novel–novel combinations. Identifying which combinations are appropriate and in which subpopulations are among the most difficult questions in medical research. We conducted a Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA)-guided systematic review of trials of novel–novel combination therapies involving immunotherapies or molecular targeted therapies in advanced solid tumors. A MEDLINE search was conducted using a modified Cochrane Highly Sensitive Search Strategy for published clinical trials between July 1, 2017, and June 30, 2020, in the top-ranked medical and oncology journals. Trials were evaluated according to a criterion adapted from previously published Food and Drug Administration guidance and other key considerations in designing trials of combinations. This included the presence of a strong biological rationale, the use of a new established or emerging predictive biomarker prospectively incorporated into the clinical trial design, appropriate comparator arms of monotherapy or supportive external data sources and a primary endpoint demonstrating a clinically meaningful benefit. Of 32 identified trials, there were 11 (34%) trials of the novel–novel combination of anti-programmed death 1 (PD-1)/programmed death ligand 1 (PD-L1) and anti-cytotoxic T lymphocyte-associated antigen-4 (CTLA-4) therapy, and 10 (31%) trials of anti-PD-1/PD-L1 and anti-vascular endothelial growth factor (VEGF) combination therapy. 20 (62.5%) trials were phase II trials, while 12 (37.5%) were phase III trials. Most (72%) trials lacked significant preclinical evidence supporting the development of the combination in the given indication. A majority of trials (69%) were conducted in biomarker unselected populations or used pre-existing biomarkers within the given indication for patient selection. Most studies (66%) were considered to have appropriate comparator arms or had supportive external data sources such as prior studies of monotherapy. All studies were evaluated as selecting a clinically meaningful primary endpoint. In conclusion, designing trials to evaluate novel–novel combination therapies presents numerous challenges to demonstrate efficacy in a comprehensive manner. A greater understanding of biological rationale for combinations and incorporating predictive biomarkers may improve effective evaluation of combination therapies. Innovative statistical methods and increasing use of external data to support combination approaches are potential strategies that may improve the efficiency of trial design. Designing trials to evaluate novel–novel combination therapies presents numerous challenges to demonstrate efficacy in a comprehensive manner. A greater understanding of biological rationale for combinations and incorporating predictive biomarkers may improve effective evaluation of combination therapies. Innovative statistical methods and increasing use of external data to support combination approaches are potential strategies that may improve the efficiency of trial design.

INTRODUCTION

With rapid advances in our understanding of cancer genomics and immunobiology, an expanding number of novel therapies are being evaluated in clinical trials.1 As a consequence, there is an exponentially increasing number of mathematically possible drug combinations,2 including novel–novel combinations, in which two or more drugs are investigational and are not yet approved standards of care. Concordantly, there has been an increase in the number of clinical trials evaluating combination therapies.3 The use of combination therapies to improve efficacy has traditionally been a central tenet of medical oncology ever since the initial use of combination chemotherapy regimens in hematological malignancies and breast cancer.4 The underlying rationale is anchored in the synergistic or additive effects of drugs with differing and potentially complementary mechanisms of action to increase the likelihood of response, forestall or overcome resistance and minimize overlapping toxicities. However, patient-to-patient variability and
the independent action of drugs may also confer benefit for combination therapies without additive or synergistic interactions. Multarm, ‘pick-the-winner’ or factorial trial designs may often be used to evaluate more than one novel treatment individually and in combination in a single trial. However, it may be impractical to conduct such trials due to finite resources, particularly with an increasing number of novel therapies and potential combinations. Identifying which combinations are appropriate in which patient subpopulations are among the most difficult questions in medical research. Garnering regulatory approval requires demonstrating that each component of a combination contributes to its benefit. This creates numerous challenges for clinicians, industry and regulatory bodies alike in designing, conducting and interpreting trials of novel–novel combination therapies. Therefore, the rational, efficient and effective evaluation of novel–novel drug combinations is crucial. Regulatory agencies, such as the US Food and Drug Administration (FDA), have recognized these challenges and have released formal guidance for trial sponsors. Nevertheless, a fine balance between the level of evidence required to obtain regulatory approval of a novel–novel combination versus the practicality of conducting such trials is needed. Furthermore, as the drug development landscape in oncology continues to evolve, such as the advent of immunotherapeutic strategies, capabilities to adapt clinical trial design remain crucial.

We sought to conduct a focused systematic review of clinical trials of novel–novel combination therapies involving immunotherapies or molecular targeted therapies in advanced solid tumors in order to identify opportunities to improve paradigms for the drug development pathway and clinical trial evaluation of the efficacy of rational combination therapies. Selected studies were evaluated and assessed according to a set of criteria adapted from the aforementioned FDA guidance and other key considerations in designing trials of novel–novel combination therapies. This included the underlying biological rationale for the combination, the incorporation of an established or emerging predictive biomarker, and the clinical trial design in terms of comparator arms and the primary endpoint.

**METHODS**

**Search strategy**

The search strategy was conducted in MEDLINE according to a modified Cochrane Highly Sensitive Search Strategy to identify published clinical trials that evaluated novel combination therapies in advanced solid tumors. In order to maintain contemporary relevance and focus on the most innovative/promising agents, our search covered the time period from July 1, 2017, to June 30, 2020, and was restricted to articles published in eight selected high-tier peer-reviewed journals (New England Journal of Medicine, Lancet, Journal of the American Medical Association, Lancet Oncology, Journal of Clinical Oncology, Cancer Discovery, JAMA Oncology, and Annals of Oncology). These journals were selected as the top-ranked general medical or general oncology journals by impact factor that publish oncology clinical trials according to the Journal Citation Reports 2019.

**Study selection**

A priori inclusion criteria were established. To be eligible for inclusion, studies had to be primary research articles reporting the outcomes of a phase II or III clinical trial evaluating novel–novel combination therapy with programmed death 1 (PD-1)/programmed death ligand 1 (PD-L1) or cytotoxic T lymphocyte-associated antigen-4 (CTLA-4) immune checkpoint targeting agents or molecularly targeted agents. Novel–novel combination therapy was defined as two or more investigational drugs, of which none were approved or recommended by treatment guidelines for the given indication. Exclusion criteria included pediatric studies (subjects<18 years of age), observational studies, meta-analyses, publications using pooled data from two or more trials, dose finding or phase I trials, early stage or locally advanced solid tumor studies, and hematological studies. In cases of updated analyses after initial study publication, studies were included in this analysis only if prespecified additional analysis for mature data of primary endpoints was being reported. Board-certified or equivalent oncologists (ACT and MK) reviewed the articles for final eligibility, and disagreement was resolved by discussion and consensus.

**Data extraction**

Data extracted for each study included (1) study name/c clinical trial ID; (2) journal; (3) authors; (4) trial sponsor; (5) tumor type and study population (newly diagnosed vs recurrent); (6) drugs studied; (7) treatment arms; (8) trial phase; (9) treatment regimens; (10) Common Terminology Criteria for Adverse Events version used; (11) biomarker selection criteria; (12) trial endpoints; (13) response data including objective response rate (ORR); (14) survival data including progression-free survival (PFS) and overall survival (OS); (15) adverse event data, including number of total and severe adverse events and mortality; (16) FDA approval for the combination therapy as of September 2020.

**Study evaluation and statistical analysis**

The trial design of each study was evaluated according to a set of criteria adapted from the criteria outlined by the FDA in their guidance on the development of novel combination therapies. As this review consisted only of studies involving patients with advanced cancer, it was accepted that all studies fulfilled the first FDA criteria, in that studies were evaluating a combination treatment for a serious disease or condition. Further general criteria in the FDA guidance included (1) a strong biological rationale for use of the combination and (2) the combination may provide a significant therapeutic advance over...
available therapy and is superior to the individual agents. In addition, factors which contribute to the efficiency of trial design and subsequent clinical impact, such as the use of external data sources and clinically meaningful primary endpoints, were also evaluated.

Trial designs were assessed according to the following criteria:

1. A strong biological rationale, defined as any published in vitro or in vivo preclinical data demonstrating activity specifically for the combination therapy with class-specific agents over the individual agents alone in the given indication. The presence of a biological rationale was considered ‘limited’ if published preclinical data were conducted only in a single experimental model system.

2. Use of a new established or emerging predictive biomarker prospectively incorporated into the clinical trial design to define eligible patients for the combination therapy and for which there are no approved or recommended therapeutic options for the given biomarker.

3. Appropriate comparator arms where applicable, allowing for an evaluation of the efficacy and safety of the individual agents alone, or supportive external data sources such as prior studies of monotherapy.

4. Primary endpoint which demonstrates a clinically meaningful benefit in the given indication and according to the phase of trial, such as ORR, PFS and/or OS.

RESULTS

Study selection and characteristics

We identified 160 potentially eligible studies. After full-text review and applying the selection criteria, 32 studies were included in the final analysis (figure 1). The characteristics of the studies are listed in table 1. Studies were broadly classified based on the drug–target combination for further in-depth analysis according to the evaluation criteria (see online supplemental appendix for additional results).

Inhibition of PD-1/PD-L1 and CTLA-4 combination therapy

There were 11 trials of the novel–novel combination of anti-PD-1/PD-L1 and anti-CTLA-4 therapy, as shown in figure 2, table 2 and online supplemental appendix, including six (55%) phase II and five (45%) phase III trials. There were four studies conducted in non-small cell lung cancer (NSCLC), with a range of other tumor types in the remaining studies (figure 2).

Figure 1  Selection of studies evaluating a novel–novel combination therapy with a targeted therapy and/or immune checkpoint inhibitor.
Rationale for combinations

We first assessed for the presence of a strong biological rationale for the combination approach, according to preclinical data supporting superior efficacy of the combination compared with the individual agents alone, in the given indication. The combination of nivolumab and ipilimumab was first approved in unresectable or metastatic melanoma in 2015, while there is currently no approved indication for the combination of durvalumab and tremelimumab. Preclinical models in melanoma demonstrated the enhanced antitumor activity for combination checkpoint blockade,9 and development of the combination was also supported by synergistic activity observed in murine colorectal and ovarian tumor models.10 11 Anti-CTLA-4 therapy impacts the lymphoid compartment, resulting in an increase in the number as well as breadth of specificity of tumor antigen reactive T cells, whereas anti-PD-1 impacts the immunosuppression within the tumor microenvironment. Clinical activity was subsequently seen in a phase I trial of nivolumab and ipilimumab in patients with advanced melanoma.12 As a result, trials of combination checkpoint blockade were evaluated in numerous other cancers. Of the 11 trials investigating inhibition of PD-L1 and CTLA-4 combination therapy included in this review, there was a distinct absence of a strong biological rationale in most studied indications, with limited in vitro or in vivo data supporting the combination, although in many cases

Table 1  Study characteristics

| Characteristics (n=32) | n (%) |
|-----------------------|-------|
| Trial phase           |       |
| II                    | 20 (62.5) |
| III                   | 12 (37.5) |
| Trial sponsor         |       |
| Academic              | 10 (31) |
| Industry              | 22 (69) |
| Tumor type            |       |
| Breast                | 1 (3) |
| Colorectal            | 4 (13) |
| Endometrial           | 1 (3) |
| HCC                   | 1 (3) |
| HNSCC                 | 1 (3) |
| Melanoma              | 1 (3) |
| Mesothelioma          | 1 (3) |
| Multiple, including basket | 2 (6) |
| NSCLC                 | 5 (16) |
| Ovarian               | 2 (6) |
| Pancreatic            | 1 (3) |
| RCC                   | 7 (22) |
| Salivary              | 1 (3) |
| Sarcoma               | 3 (9) |
| Thyroid               | 1 (3) |
| Journal               |       |
| Annals of Oncology    | 4 (13) |
| Cancer Discovery      | 0 (0) |
| JAMA                  | 0 (0) |
| JAMA Oncology         | 7 (22) |
| Journal of Clinical Oncology | 5 (16) |
| Lancet                | 1 (3) |
| Lancet Oncology       | 8 (25) |
| New England Journal of Medicine | 7 (22) |
| Combination therapy   |       |
| Immuno therapy        | 13 (41) |
| Targeted therapy      | 8 (25) |
| Both immuno therapy and targeted therapy | 11 (34) |
| Drug targets          |       |
| PD-1/PD-L1+CTLA-4     | 11 (34) |
| PD-1/PD-L1+VEGF       | 10 (31) |
| BRAF+MEK (+EGFR)      | 4 (13) |
| HER2                  | 2 (6) |
| Other*                | 5 (16) |
| Biomarker selection   |       |
| Selected              | 15 (47) |

Continued

Table 1  Continued

| Characteristics (n=32) | n (%) |
|-----------------------|-------|
| Primary endpoint      |       |
| Unselected            | 17 (53) |
| Phase II trials (n=36) |       |
| ORR                   | 15 (75) |
| PFS                   | 3 (15) |
| OS                    | 1 (5)  |
| DCR                   | 1 (5)  |
| Phase III trials (n=12) |       |
| PFS                   | 2 (17) |
| OS                    | 3 (25) |
| PFS and OS            | 6 (50) |

*Chemotherapy+VEGF, chemotherapy+PARP, PD-1+HPV16 vaccine, PD-1+oncolytic virus, PD-L1+MEK.
CTLA-4, cytotoxic T lymphocyte-associated antigen-4; DCR, disease control rate; EGFR, epidermal growth factor receptor; HCC, hepatocellular carcinoma; HER2, human epidermal growth factor receptor 2; HNSCC, head and neck squamous cell carcinoma; JAMA, The Journal of the American Medical Association; MEK, mitogen-activated protein kinase kinase; NSCLC, non-small cell lung cancer; ORR, objective response rate; OS, overall survival; PARP, poly(ADP-ribose) polymerase; PD-1, programmed death 1 ; PD-L1, programmed death ligand 1 ; PFS, progression-free survival; RCC, renal cell carcinoma; VEGF, vascular endothelial growth factor.
there were preliminary signs of efficacy in early-phase trials, including phase I trials, emphasizing the need for better preclinical models to select rational combination immunotherapy approaches.13

Utility of biomarkers

Next, we assessed the incorporation of a newly established or emerging predictive biomarker into the trial design that may suggest efficacy of the combination superior to monotherapy. Of the 11 trials, 6 (55%) were conducted in unselected patient populations. Four (44%) trials selected patients based on PD-L1 status, a previously established but somewhat controversial biomarker to select patients for anti-PD-1/PD-L1 monotherapy. Only one (9%) trial incorporated a new established or emerging biomarker, with the CheckMate 227 trial including a coprimary endpoint of PFS in patients with high tumor mutation burden (TMB) for nivolumab plus ipilimumab versus chemotherapy.14

Comparator arms

The use of appropriate comparator arms that would allow for the comparison of the efficacy and safety of the individual agents alone versus the combination was also critically examined. This was evaluated in the context of the presence or absence of supportive external data sources, such as prior studies of monotherapy. Of the phase III trials, one trial was conducted in first-line renal cell carcinoma (RCC), with the remaining trials conducted in NSCLC. CheckMate 214 evaluated nivolumab plus ipilimumab versus sunitinib as first-line therapy in advanced RCC.15 There was prior monotherapy data for ipilimumab in a phase II trial that allowed pretreated and treatment-naïve patients.16 Nivolumab monotherapy, already approved in the treatment resistant setting, had also been evaluated in treatment-naïve patients in a multicohort phase Ib expansion study.17 CheckMate 227 was an open-label phase III randomized trial in untreated advanced NSCLC.14 19 The trial was designed to test multiple nivolumab-based regimens in different patient populations. Nivolumab monotherapy had been previously evaluated in treatment-naïve advanced NSCLC in the phase III CheckMate 026 study in patients with PD-L1 expression of 5% or more,20 while ipilimumab monotherapy had been evaluated in a previous phase II trial.21 Durvalumab plus tremelimumab was evaluated in two phase III trials for NSCLC, with the MYSTIC trial22 as first-line therapy and the ARCTIC trial23 as third-line or later line therapy. Durvalumab monotherapy had been investigated in the treatment-refractory setting in NSCLC,24 but neither durvalumab or tremelimumab monotherapy had been previously evaluated in the first-line setting.

Overall, the phase II and III trials of combination anti-PD-1/PD-L1 and anti-CTLA-4 therapy were appropriately designed with comparator arms of either individual agent alone and where there were no prior studies in the given indication.

Primary endpoints

The primary endpoints of the selected studies were also evaluated. In general, the phase II studies used ORR or DCR, while the phase III studies used PFS and/or OS. The primary endpoints were assessed overall as appropriate and clinically meaningful in all studies.

Inhibition of PD-1/PD-L1 and VEGF combination therapy

There were 10 trials of the novel–novel combination of anti-PD-1/PD-L1 and anti-vascular endothelial growth factor (VEGF) therapy, as shown in figure 3, online supplemental table 1 and online supplemental appendix.

Rationale for combinations

The combination of antiangiogenic therapy and immune checkpoint inhibitor therapy is based on evidence demonstrating that aberrant angiogenesis is a hallmark of many solid tumors, resulting in immune evasion.25 Therefore, normalization of abnormal tumor vasculature with antiangiogenic therapy may improve immune effector cell function by decreasing hypoxia, acidosis and nutrient deprivation and may increase the infiltration of immune effector cells into the tumor microenvironment in order to enhance the efficacy of immunotherapy. VEGF also has pleotropic immunosuppressive effects including impairment of dendritic cell function, as well as mobilization of immunosuppressive cells such as tumor associated macrophages, regulatory T cells, and myeloid-derived suppressor cells.26 Consequently, this has led to a proliferation of studies of novel–novel combinations of therapeutics from these two classes. Improved efficacy with combination inhibition of VEGF and immune checkpoint blockade has been shown in animal models for melanoma,27 colorectal cancer,28 breast cancer and

Figure 2 Characteristics of combination anti-PD-1/PD-L1 and anti-CTLA-4 therapy trials. (A) Combination therapy regimen, (B) tumor type, and (C) trial phase. HNSCC, head and neck squamous cell carcinoma; NSCLC, non-small cell lung cancer; RCC, renal cell carcinoma.
## Table 2 Combination anti-PD-1/PD-L1 and anti-CTLA-4 therapy trials

| Study name                          | Year, lead author | Tumor type          | Line of therapy | Combination regimen                     | Patients (total n) | Biomarker selection | Primary endpoint                                   | Trial design                                                                 | Primary endpoint met | FDA-approved therapy | Strong biological rationale | New biomarker | Comparator arms of monotherapy | Clinically meaningful primary endpoint |
|-------------------------------------|-------------------|---------------------|------------------|------------------------------------------|--------------------|---------------------|------------------------------------------------|------------------------------------------------|---------------------|----------------------|-------------------------------|----------------|---------------------------|--------------------------------------|
| **Phase II trials**                 |                   |                     |                  |                                          |                    |                     |                                                 |                                              |                     |                      |                              |               |                           |                                      |
| Alliance A091401                    | 2018, D’Angelo    | Sarcoma             | 2+               | Nivolumab plus ipilimumab               | 96                 | Unselected          | ORR (N and N+I)–non-comparative                  | N or N+I (1:1)         | Yes                  | No                    | No                   | No                         | Yes Yes                                                                            |
| CONDOR                              | 2019, Siu         | HNSCC               | 2+               | Durvalumab plus tremelimumab            | 267                | PD-L1               | ORR (D+T)–non-comparative                       | PD-L1<25%, D+Tor D 2 (1:1) | Yes                  | No                    | No                   | No                         | Yes Yes                                                                            |
| IFACT-1501 MAPS2                    | 2019, Scherpereel  | Mesothelioma        | 2+               | Nivolumab plus ipilimumab               | 125                | Unselected          | DCR at 12 weeks (N and N+I)–non-comparative     | N or N+I (1:1)         | Yes                  | No                    | No                   | No                         | Yes Yes                                                                            |
| NCT02558894                         | 2019, O'Reilly    | Pancreatic          | 2                | Durvalumab plus tremelimumab            | 65                 | Unselected          | ORR (D+T and D)–lead-in safety study, with expansion pending efficacy signal, non-comparative | D+Tor D (1:1)         | No                   | No                    | No                   | No                         | Yes Yes                                                                            |
| CO.26                               | 2020, Chen        | Colorectal          | 3+               | Durvalumab plus tremelimumab            | 180                | Unselected          | OS (D+T vs BSC)                                 | Yes                  | No                   | Limited               | No                    | No                         | Yes Yes                                                                            |
| NRG-GY003                           | 2020, Zamarin     | Ovarian             | 2+               | Nivolumab plus ipilimumab               | 100                | Unselected          | ORR at 6months (N vs N+I)                       | N or N+I (1:1)         | Yes                  | No                    | Limited              | No                         | Yes Yes                                                                            |
| **Phase III trials**                |                   |                     |                  |                                          |                    |                     |                                                 |                                              |                     |                      |                              |               |                           |                                      |
| CheckMate 214                       | 2018, Motzer      | RCC                  | 1                | Nivolumab plus ipilimumab               | 1096               | Unselected          | Coprimary: OS, ORR and PFS (in intermediate or poor prognostic risk) | N+I or sunitinib (1:1) | Yes                  | Yes                   | No                   | No                         | Yes Yes                                                                            |
| CheckMate 227                       | 2018, Hellmann    | NSCLC                | 1                | Nivolumab plus ipilimumab               | 1739               | TMB                 | Coprimary: PFS (N+I vs chemo in TMB high)       | PD-L1>1%, N+I or N or chemo (1:1:1) | No                   | Yes                   | No                    | Yes                   | Yes                        | Yes Yes                                                                            |
| CheckMate 227                       | 2019, Hellmann    | NSCLC                | 1                | Nivolumab plus ipilimumab               | 1739               | PD-L1               | Coprimary: OS (N+I vs chemo in PD-L1>1%)        | PD-L1>1%, N+I or N+chemo or chemo (1:1:1) | Yes                  | Yes                   | Yes                   | No                    | Yes                        | Yes Yes                                                                            |
| MYSTIC                              | 2020, Rizvi       | NSCLC                | 1                | Durvalumab plus tremelimumab            | 1118               | PD-L1               | Coprimary: OS (D vs chemo in PD-L1>25%), PFS and OS (D+T vs chemo in PD-L1>25%) | Yes                  | No                   | No                    | No                   | No                         | Yes Yes                                                                            |
| ARCTIC                              | 2020, Planchard   | NSCLC                | 3+               | Durvalumab plus tremelimumab            | 595                | PD-L1               | Coprimary: PFS and OS (D+T vs SOC in PD-L1>25%) | Study A: PD-L1>25%, D or SOC (1:1); Study B: PD-L1>25%, D+Tor SOC or D or T (3:2:2:1) | No                   | No                   | No                    | No                   | No                         | Yes Yes                                                                            |

BSC, best supportive care; Chemo, chemotherapy; D, durvalumab; DCR, disease control rate; HNSCC, head and neck squamous cell carcinoma; I, ipilimumab; N, nivolumab; NSCLC, non-small cell lung cancer; ORR, objective response rate; OS, overall survival; PD-L1, programmed death ligand 1; PFS, progression-free survival; RCC, renal cell carcinoma; SOC, standard of care; T, tremelimumab; TMB, tumor mutation burden.
pancreatic neuroendocrine tumors.\textsuperscript{29} Specifically, lenvatinib plus anti-PD-1 therapy has also been evaluated in murine hepatocellular, colorectal and melanoma models.\textsuperscript{30,31} However, preclinical data demonstrating the efficacy of other specific drug combinations are sparse. For example, the combination of axitinib plus avelumab in RCC had not been published in preclinical models, and the rationale for the combination as first-line therapy was based on the toxicity profile for axitinib, with lower hepatotoxicity compared with sunitinib.\textsuperscript{32} There were also no preclinical data published for the remaining combination therapies in the given studied indications.

Utility of biomarkers

Of the 10 trials evaluating combination anti-PD-1/PD-L1 and anti-VEGF therapy, a majority (80\%) were conducted in biomarker unselected populations of patients. The two (20\%) trials, which included biomarker selection criteria, selected patients on the basis of PD-L1 IHC for avelumab\textsuperscript{33} and atezolizumab,\textsuperscript{34} respectively, in advanced RCC. PD-L1 IHC had not been previously established as a biomarker for patient selection for anti-PD-1/PD-L1 therapy in advanced RCC and therefore represented a new predictive biomarker in this patient population. No robust biomarkers for anti-VEGF therapy have been identified, and consequently, no trial attempted to select a biomarker for this class of agents.

Comparator arms

Overall, the phase II trials of combination anti-PD-1/ PD-L1 and VEGF therapy were appropriately designed with comparator arms of either individual agent alone and where there were no prior studies in the given indication. Two phase III trials in patients with treatment-naive metastatic RCC involved axitinib combinations. Axitinib monotherapy is approved in the second-line setting\textsuperscript{35} but had not been previously evaluated as first-line therapy.

Primary endpoints

The primary endpoint in five of the six phase II trials was ORR, with the remaining trial of pembrolizumab plus axitinib in metastatic sarcoma using PFS rate at 3 months as the primary endpoint. Coprimary endpoints of PFS and OS were used in the four phase III trials of combination anti-PD-1/PD-L1 and anti-VEGF therapy. Overall, these endpoints were considered appropriate and clinically meaningful. It should be borne in mind that in tumors of the central nervous system, both pseudoprogression and pseudoresponse have been identified as limiting characteristics of immune checkpoint inhibitors and antiangiogenic inhibitors, respectively, and in certain extracranial settings, concern regarding post-treatment peritumoral inflammatory changes (‘pseudo-progression’) following checkpoint inhibition has been described. These could limit the validity of ORR as an endpoint in such studies.

Inhibition of BRAF and MEK combination therapy

There were four trials of the novel–novel combination of BRAF and mitogen-activated protein kinase kinase (MEK) inhibitor therapy, as shown in online supplemental table 2 and online supplemental appendix. In addition, the BEACON CRC trial evaluated epidermal growth factor receptor (EGFR) inhibitor therapy in combination with BRAF and MEK inhibition.\textsuperscript{36}

Rationale for combinations

In BRAF-mutated melanoma, the combination of BRAF and MEK-pathway inhibition had initially established clinical efficacy and tolerability compared with BRAF-inhibitor monotherapy with two combinations (dabrafenib plus trametinib, and vemurafenib plus cobimetinib).\textsuperscript{37,38} BRAF and MEK inhibitor combinations were originally developed based on preclinical data demonstrating that the combination could improve efficacy and delay the emergence of resistance.\textsuperscript{39,40} Reactivation of the MAPK pathway was a commonly reported mechanism of resistance to BRAF-inhibitor monotherapy.\textsuperscript{41,42} The combination of encorafenib and binimetinib was subsequently evaluated due to the increased potency of encorafenib compared with dabrafenib and vemurafenib, related to a greater dissociation half-life and improved pharmacodynamics.\textsuperscript{43} Furthermore, the combination with binimetinib, which ameliorated the toxicity of encorafenib monotherapy, allowed for high doses of encorafenib in the combination treatment.\textsuperscript{44}

The rationale for the inhibition of BRAF, MEK and EGFR in BRAF V600E mutated colorectal cancer was developed after extensive preclinical investigations characterizing these pathways. Rapid feedback activation through EGFR after BRAF inhibition alone explained the poor efficacy of BRAF monotherapy and led to the development of BRAF plus EGFR inhibitor combinations.\textsuperscript{44,45} Subsequently, the combination of BRAF and MEK inhibition to improve efficacy compared with BRAF plus EGFR inhibition was also demonstrated preclinically.\textsuperscript{46,47}

Tan AC, et al. J Immunother Cancer 2021;9:e002459. doi:10.1136/jitc-2021-002459
Utility of biomarkers
All four combination trials of BRAF plus MEK inhibition selected patients based on the presence of a BRAF mutation. For advanced melanoma, there were already approved therapies for patients harboring BRAF mutations. In the remaining tumor types, however, the combination therapies represented a new genotype-directed therapeutic option.

Comparator arms and primary endpoints
COLUMBUS was a three-arm, randomized phase III study evaluating combination encorafenib (450 mg daily dose) plus binimetinib versus encorafenib monotherapy (300 mg daily dose) versus vemurafenib monotherapy.46 Binimetinib monotherapy had previously been investigated in a phase II study for patients with NRAS or BRAF-mutated melanoma.49 BEACON CRC (Binimetinib, Encorafenib, and Cetuximab Combined To Treat BRAF-Mutant Colorectal Cancer) was also a three-arm, randomized phase III study, with study arms consisting of triplet therapy (encorafenib, binimetinib and cetuximab), doublet therapy (encorafenib and cetuximab) or investigator’s choice of cetuximab plus irinotecan or cetuximab plus FOLFIRI (control group).36 There had been no prior trials of encorafenib, binimetinib or cetuximab monotherapy in patients with advanced BRAF V600E mutated colorectal cancer. Cetuximab monotherapy had previously been studied in unselected patients with metastatic colorectal cancer.30 Retrospective analyses, however, suggested responses to cetuximab may be lower in patients with BRAF mutations.31 Additionally, vemurafenib monotherapy had been investigated in BRAF V600E mutated colorectal cancer in a small phase II trial and a basket trial.32 33 There was no meaningful clinical activity for vemurafenib monotherapy, with only one response out of 31 patients across the two trials. The primary endpoints in COLUMBUS and BEACON CRC were both considered clinically meaningful.

Inhibition of HER2 combination therapy
There were two studies reporting the novel–novel combination of human epidermal growth factor receptor 2 (HER2) inhibitor therapy with trastuzumab and pertuzumab, as shown in online supplemental table 3 and online supplemental appendix. Both studies were reports from the MyPathway basket trial for patients with colorectal cancer54 and salivary gland cancers,55 respectively.

Rationale for combinations
In colorectal cancer, pre-clinical data from HER2-amplified colorectal tumor grafts or xenografts, had demonstrated limited activity of single agent HER2 targeted therapy with trastuzumab, pertuzumab or lapatinib.56 57 Anti-tumor activity however, was increased with combination HER2 targeting regimens, although with trastuzumab plus lapatinib or pertuzumab plus lapatinib. The complementary mechanisms of action of trastuzumab and pertuzumab, and demonstrated efficacy in breast cancer,58 provided a strong rationale for this combination in HER2-amplified colorectal cancer.

Utility of biomarkers
As HER2 targeted therapies had not been approved in either tumor type, both studies were assessed as incorporating a new established or emerging biomarker.

Comparator arms and primary endpoints
As a phase Ia multiple basket trial of various targeted therapies in advanced solid tumors, ORR was considered an appropriate and clinically meaningful endpoint for the MyPathway trial. Understandably, as a basket trial, comparator arms of either trastuzumab or pertuzumab monotherapy were not included in this trial.

Other combination therapies
There were five trials evaluating other novel–novel combination therapies, including chemotherapy plus VEGF inhibitor therapy, chemotherapy plus poly(ADP-ribose) polymerase (PARP) inhibitor therapy, anti-PD-1 plus HPV16 vaccine therapy, anti-PD-1 plus an oncolytic virus therapy, and anti-PD-L1 plus MEK inhibitor therapy (online supplemental appendix and online supplemental table 4).

COTEZO IMblaze 370 was a three-arm randomized phase III study of atezolizumab plus cobimetinib versus atezolizumab monotherapy versus regorafenib in patients with previously treated metastatic colorectal cancer.59 In the initial phase I trial of cobimetinib monotherapy, there were 41 patients with metastatic colorectal cancer, but no responses were seen.60 The combination of anti-PD-L1 and MEK inhibitor in colorectal cancer was developed on the basis of preclinical data, suggesting MEK inhibition could affect the immune contexture in the tumor microenvironment.61 Cobimetinib had been shown to increase T-cell infiltration into tumors and downregulate immunosuppressive cytokines and receptors.62 Combination therapy of cobimetinib with anti-PD-L1 inhibition also resulted in synergistic and durable tumor regression in mice models.62

BROCADE was a three-arm phase II trial for patients with BRCA1/2-mutated recurrent or metastatic breast cancer.63 Patients were randomized to receive veliparib plus carboplatin/paclitaxel or veliparib plus temozolomide or carboplatin/paclitaxel. The primary endpoint was PFS comparing both veliparib containing arms with chemotherapy alone. Veliparib plus temozolomide was considered the novel–novel combination for this review. Veliparib monotherapy had been evaluated in a single-arm phase II trial for patients with germline BRCA1/2 associated metastatic breast cancer,54 while temozolomide monotherapy had previously shown a lack of activity in an unselected population of patients with metastatic breast cancer.64 The rationale for the combination of temozolomide and veliparib was from preclinical breast cancer models which demonstrated synergistic activity for the combination.66 67 At the time the study was developed,
there were no targeted therapies approved for patients BRCA1/2-mutated breast cancers.68

Overall evaluation of novel–novel combination therapies in oncology

The overall assessment and evaluation of trial designs of novel–novel combination therapies according to the four criteria are shown in figure 4.

Discussion

With an expanding number of novel therapies in clinical development and possible drug combinations, the fundamental challenge remains to evaluate rational combinations efficiently and effectively. In particular, a mechanistic understanding of the contribution of each drug to the treatment effect is needed, both from a scientific and regulatory perspective.69 Based on a systematic review of the top general medical and oncology journals over the past 3 years, we identified 32 recently published trials evaluating novel–novel combinations and evaluated each trial according to criteria adapted from FDA guidance on the development of novel combination therapies (box 1).

First, we assessed the strength of the biological rationale according to the presence of published preclinical data. Most trials (72%) lacked significant preclinical evidence supporting the development of the combination in the given indication. This was particularly evident in studies evaluating combination immune checkpoint blockade, for which there were only two studies with limited preclinical data in colorectal and ovarian cancers. This could, in part, be attributed to difficulties in developing preclinical animal models for immunotherapy, due to the inability of immunocompetent animal models to fully recapitulate the human immune system.70 Phase I studies were also excluded from our analysis, for which preclinical data may be especially relevant in the initial decision to evaluate a combination. Notably, however, significant clinical efficacy in tumor types such as NSCLC14 19 and RCC15 has been seen, despite the lack of supportive preclinical data. For some of these tumors (such as RCC), evidence of single agent activity or early efficacy in phase I trials served as the rationale for subsequent combination phase II and III studies. Coupled with the initial outcomes of trials in melanoma,12 this has resulted in a rapid increase in the number of combination immunotherapy trials.3 Nevertheless, the value of numerous trials with overlap and duplication in the combination partner targets, being conducted in unselected patients can be questioned,3 particularly in cancers for which single-agent immunotherapy has no activity. This observation highlights the importance of a greater understanding of tumor-specific immunity and the need to develop more effective biomarkers.71 Given the expanding number of potential combinations, rational selection of combinations based on mechanistic evidence and robust biological rationale is crucial.72 Additionally, for many trials in our review, preclinical data supporting the combination may have been demonstrated in other tumor types outside the given trial’s studied indication. Particularly for targeted therapies, there can be significant diversity across tumor types in the actionability of oncogenic driver mutations.73 BRAF V600E mutations are a prominent example, with a spectrum of activity for combination BRAF and MEK inhibition across histologies.74 This diversity is also exemplified in colorectal cancer, with the role for combined BRAF plus EGFR inhibition demonstrated elegantly in preclinical studies.75
### Box 1  What is specifically learned about combinations of novel–novel agents?

**Summary:** We searched MEDLINE according to a modified Cochrane Highly Sensitive Search Strategy to identify published clinical trials that evaluated novel combination therapies in advanced solid tumors. Our search covered the period from July 1, 2017, to June 30, 2020. We restricted our search to articles published in eight selected peer-reviewed journals (New England Journal of Medicine, Lancet, Journal of the American Medical Association, Lancet Oncology, Journal of Clinical Oncology, Cancer Discovery, JAMA Oncology and Annals of Oncology). We identified 160 potentially eligible studies. After full-text review and applying the selection criteria, 32 studies were included in the final analysis (figure 1). Studies were broadly classified based on the drug–target combination for further in-depth analysis. Below is a summary of the key messages and recommendation.

**A. Key Messages**

1. Combinations are the future of clinical trials and this is so in most diseases.
2. Identifying which combinations are appropriate and in which patient subpopulations are among the most difficult questions in medical research.
3. In 72% of the analyzed studies, there was no significant preclinical evidence supporting the development of the specific combination in the given indication. This was especially true in studies with combination immune checkpoint blockade. This, in part, may be due to difficulties in developing immunocompetent animal models to fully recapitulate the human immune system.
4. In 69% of the analyzed studies, trial populations were unselected and did not use pre-existing predictive biomarkers for the given indication for patient selection.
5. Sixty-six per cent of the trials analyzed had appropriate comparator arms or had supportive external data sources such as monotherapy prior studies.
6. A greater understanding of the biological rationale for the combination and incorporating novel biomarkers can improve the practical evaluation of novel–novel combination therapies. However, biomarker-driven clinical trials using combination therapies present additional challenges and require careful consideration of the biomarkers’ performance and clinical utility.
7. Actionability of oncogenic driver mutations and utility of a specific biomarker in one cancer type does not predict similar outcomes in other cancers, for example, EGFR in NSCLC versus CRC, TMB in NSCLC versus gliomas. Therefore, preclinical and clinical rationales for biomarker-driven combination therapies in one cancer cannot justify a combination trial in another histology.
8. There were no prior activity data for monotherapy in specific tumor cohorts in many basket trials (eg, MyPathway and ROAR).
9. Garnering regulatory approval requires demonstrating that each component of a combination contributes to its benefit. Such demonstrations may entail factorial designs and require interacting closely with regulators.
10. Partial factorial designs in adaptive clinical trials are likely to be the best and most efficient solutions for the future, especially in the context of precision medicine where biomarker-defined subpopulations are becoming the norm.

**B. Recommendations**

1. Avoid duplication in partner targets in unselected patients, particularly in cancers for which single-agent immunotherapy has no activity.

---

**Box 1  Continued**

2. In the design of a combination, consider whether other monotherapy arms are required when trial designs are conceived.
3. Consider if randomized arms of monotherapy are ethical; for example, if based on the mechanism of action, no clinical activity of monotherapy is expected.
4. Consider use of milestone survival or response endpoints to more efficiently generate early evidence.
5. Consider the incorporation of real-world evidence and novel hybrid designs, including in regulatory decision making.
6. Consider using (historical) data on single-agent efficacy, ideally to be obtained from multiple datasets and for all agents within the combination.
7. Consider introducing experimental therapies, including novel–novel combinations, into the trial at any time, allowing for rational combinations based on new biological or clinical insights.
8. Consider innovative but sound statistical methods, including use of historical or external data sources to support combination approaches.

CRC, colorectal cancer; EGFR, epidermal growth factor receptor; NSCLC, non-small cell lung cancer; TMB, tumor mutational burden.

We then evaluated the use of a newly established or emerging predictive biomarker incorporated into the clinical trial design. Similarly, we found that a majority of trials (69%) were conducted in biomarker unselected populations or used pre-existing biomarkers within the given indication for patient selection. No studies included separate novel predictive biomarkers for both novel agents. Biomarker-driven clinical trial designs and analysis plans present additional challenges and require careful consideration of the biomarkers’ performance and clinical utility. However, the use of biomarker-selected patients has the potential to both improve patient care and accelerate drug development with greater efficiency. CheckMate 227, which incorporated association of TMB with OS as a second coprimary endpoint after an amendment prior to the initial analysis, demonstrated the flexibility of a large phase III trial to adapt to the emerging science. The biomarker was also investigated as a subset of the overall trial population—an important consideration in prospective biomarker-driven clinical trial design. Ultimately, the role for TMB as a predictive biomarker in NSCLC remains unclear. Furthermore, despite tissue agnostic approval for TMB of ≥10 as a biomarker for pembrolizumab, there remains controversy over its use with numerous biological and practical considerations across tumor types. This illustrates the difficulties in identifying predictive biomarkers, which may lag behind the development of novel therapeutics. Dynamic biomarkers, which may change over time as tumors evolve, and the cost of developing biomarkers further complicate the development process. Nevertheless, particularly for immunotherapies, different agents may have pleiotropic effects on a variety of different cells and compartments where they are operational. There may also be heterogeneity in the relative contribution of
the various elements among patients. Therefore, without relevant biomarker selection-driven trial design, clinical efficacy may not be gleaned with an all-comers approach.

Trial design with appropriate comparator arms of monotherapy was also assessed for each trial. Most studies (66%) were considered to have appropriate comparator arms or had supportive external data sources such as prior studies of monotherapy. Noticeably, in basket trials, such as MyPathway54,55 and ROAR (Rare Oncology Agnostic Research),82 there were no data for monotherapy in certain tumor cohorts. However, basket trials in rare tumor types or uncommon molecular subsets clearly face practical challenges in terms of statistical design and patient recruitment. This emphasizes the need for a comprehensive understanding of tumor biology to identify optimal combinations.83 There is also increasing use and acceptance of real-world evidence in regulatory decision making.84 Historical data on single-agent efficacy would ideally come from multiple datasets and for all agents within the combination.85 There may also be situations in which randomized arms of monotherapy may be unethical, for example, if no clinical activity of monotherapy is expected based on the mechanism of action. This heightens the importance of careful consideration to determine whether additional arms of monotherapy are required when trial designs are conceived.

Finally, all studies were evaluated as selecting a primary endpoint, which demonstrated a clinically meaningful benefit. Nonetheless, there can be complexities in determining the validity of surrogate endpoints particularly for early-phase combination immuno-therapy trials. Milestone survival or response endpoints are increasingly used86 to more efficiently generate early evidence. KEYNOTE-14687 for example, had a primary endpoint of ORR at 24 weeks, and ultimately was the basis for FDA approval for the combination of pembrolizumab plus lenvatinib in endometrial cancer.86 This trial also provides important insights into the use of historical data to evaluate the treatment effect of lenvatinib and pembrolizumab monotherapy.87 Additional exploratory post hoc analyses using propensity score approaches were also conducted by the FDA to evaluate the contribution of each agent.88 Importantly though, confirmatory randomized trials evaluating the combination are still ongoing.

Collectively, our review has identified that improvements in the effective evaluation of novel–novel combination therapies are clearly needed. Many of these findings may also be applicable to other combination therapies, including a new combination of two previously approved agents or the addition of a new agent to an existing approved therapy. In box 1, we provide a series of recommendations on the efficient design of future clinical trials evaluating novel combination therapies. Novel adaptive trial designs represent one approach that may enhance the efficiency of trials. GBM AGILE, a phase II/III adaptive platform trial (NCT03970447), is an example which incorporates statistical innovations such as Bayesian Adaptive design in a seamless registration trial.91 Candidate biomarkers may be identified and validated under this single platform master protocol. Furthermore, experimental therapies, including novel–novel combinations, may be introduced into the trial at any time, allowing for rational combinations based on advances in our biological understanding of tumors. Pharmaceutical platforms are also increasingly using multiarm randomized trials, for example, MORPHEUS (NCT03193190, NCT03281369, NCT03280563, NCT03424005, and NCT03376988), with multiple combination therapy arms compared against a single standard-of-care control arm, and allow for the introduction of novel–novel combinations at any time.85 Additionally, seamless phase I/II trials are becoming more commonly used92 and highlight that many of the considerations we have outlined previously may become increasingly important in the strategic design of early-phase studies as well. In particular, this includes the introduction of experimental rational combinations based on emerging biological or clinical insights—to establish appropriate dosing and characterize safety.

There are several limitations to our review, including the restricted time period (July 2017–June 2020) and journal selection, which may have introduced inherent publication bias into the studies included in our review. However, there were still 6/32 (19%) trials, which did not meet the primary endpoint included, and our evaluation criteria were not dependent on the trial’s primary outcome. In selecting only published trials, which was required to assess our evaluation criteria, contemporary trials such as the aforementioned adaptive platform trials will have been excluded. Broader evaluation of ongoing and unpublished trials is therefore also warranted, although outside the scope of this study. The included trials also represent the data known at the time of initial publication, not necessarily the data known at the time of trial design, and may therefore not completely reveal the historical sequence of events at the time of study conception. Ultimately though, there also needs to be inherent flexibility to adapt to rapidly evolving clinical paradigms. Lastly, there is significant heterogeneity of the included trials, particularly with regard to tumor types, trial design and mechanism of action of drug combinations. Nevertheless, the primary focus of this review was to identify guiding principles to improve trial efficiency in evaluating novel–novel drug combinations. Our analysis provided key insights into the published literature with recommendations (box 1) to improve paradigms for drug development and future trial design.

CONCLUSIONS

Designing trials to evaluate novel–novel combination therapies presents numerous challenges to demonstrate efficacy in a comprehensive manner. Critically, a greater understanding of the biological rationale for the combination and incorporating novel predictive biomarkers may further improve the effective evaluation of novel–novel combination therapies. Innovative statistical methods and increasing the use of historical or external data sources to support combination
approaches are potential strategies that may improve the efficiency of trial design.

Author affiliations
1Division of Medical Oncology, National Cancer Centre Singapore, Singapore
2Duke-NUS Medical School, National University of Singapore, Singapore
3AAbramson Cancer Center and Perelman School of Medicine, University of Pennsylvania, Philadelphia, Pennsylvania, USA
4Center for Neuro-Oncology, Dana-Farber Cancer Institute, Boston, Massachusetts, USA
5Department of Neurosurgery, Stanford University, Stanford, California, USA
6Department of Neurology, Heidelberg University Hospital, Heidelberg, Germany
7DKTK CCU Neuroimmunology and Brain Tumor Immunology, German Cancer Research Center, Heidelberg, Germany
8Huntsman Cancer Institute, University of Utah, Salt Lake City, Utah, USA
9Duke Cancer Institute, Duke University, Durham, North Carolina, USA
10Department of Neurological Surgery, University of California San Francisco, San Francisco, California, USA
11Division of Medical Oncology, Mayo Clinic Rochester, Rochester, Minnesota, USA
12Department of Neurosurgery, Penn State Cancer Institute, Hershey, Pennsylvania, USA
13Division of Medical Oncology, University of North Carolina at Chapel Hill School of Medicine, Chapel Hill, North Carolina, USA
14Department of Radiation Oncology, Miami Cancer Institute, Miami, Florida, USA
15Department of Neurosurgery, Northwestern University, Chicago, Illinois, USA
16Department of Neurooncology, The University of Texas MD Anderson Cancer Center, Houston, Texas, USA
17Department of Neuro-oncology, H. Lee Moffitt Cancer Center and Research Institute, Tampa, Florida, USA
18NHMRC Clinical Trials Centre, University of Sydney, Camperdown, New South Wales, Australia
19Department of Biostatistics, University of Texas MD Anderson Cancer Center, Houston, Texas, USA

Twitter Michael Plattten @plattten_michael, Solmaz Sehejbam @SolmazSehejbam and Mustafa Khasraw @mkkhasraw

Contributors ACT, SJF, PYW, DB and MK conceived of the study and performed the data analysis. All authors performed data interpretation, contributed to the writing of the manuscript, and read and approved the final manuscript.

Funding The authors have not declared a specific grant for this research from any funding agency in the public, commercial or not-for-profit sectors.

Competing interests ACT reports consultant or advisory roles for Amgen. SJF reports research grants from Incyte, Eli Lilly, Novocure, GSK; consultant or advisory roles for Bayer and Novocure; and US patent for ‘Combination therapies of EGFRvIII chimeric antigen receptors and PD-1 inhibitors’. PYW reports consultant or advisory roles for Agios, Astra Zeneca, Bayer, Boston Pharmaceuticals, CNS Pharmaceuticals, Elevate Bio Immunotherapeutics, Inxva, Karyopharm, Merck, Novartis, Nuvation Bio, Vascular Biogenics, VBI Vaccines, Voyager and QED; research funding from Agios, Astra Zeneca/MedImmune, Beigene, Celgene, Eli Lilly, Genentech/Roche, Kazia, MediciNovca, Merck, Novartis, Nuvation Bio, Oncocentrix, Vascular Biogenics and VBI Vaccines; is an editor for UpToDate and Elsevier. ML reports consultant or advisory roles for Tocagen, SQZ Technologies, VBI, InCephalo Therapeutics and Pyramidal Bio; non-research consulting roles for Stryker; research support from Arbor, Bristol-Myers-Squibb, Accuray, DNAtrix, Tocagen, Biohaven and Kyrin-Kyowa; and has patents for focused radiation+checkpoint inhibitors and local chemotherapy+checkpoint inhibitors. MP reports consultant or advisory roles for non-financial support from Roche, personal fees and non-financial support from Bayer, personal fees from Novartis, personal fees from Apogenix, non-financial support from Pfizer, personal fees from Affirx outside the submitted work. In addition, MP has a patent EP2753315B1 licensed to Bayer, a patent EP2805080B1 issued, a patent US20180155403A1 pending, a patent US2018024611A1 pending, a patent US20170254803A1 pending, a patent WO201846100A1 licensed to Bayer, a patent WO2019101643A1 licensed to Bayer, a patent WO2019101647A1 licensed to Bayer, a patent WO2019101641A1 licensed to Bayer, and a patent WO2019101642A1 licensed to Bayer. HC reports consultant or advisory roles for BD-DNAtrix/Teladoc, Karyopharm Therapeutics, Private Health, Orbis, Bayer, Forma Therapeutics and Adstra Pharmaceuticals; research funding (Inst) from Newlink Genetics, Plexikon, Kadmon, Orbus, Merck, DNAtrix, Abivie, Beigene, Forma Therapeutics, GCAR, Array BioPharma, Karyopharm, Nuvation Bio, Bayer and Bristol Meyer Squibb. DMA reports stock and other ownership interests in Diverse Biotech; consulting or advisory roles for Istaro Oncology and Jackson Laboratory for Genomic Medicine; patents, royalties, other intellectual property for ‘Methods for predicting tumor response to immunotherapy, U.S. Provisional application no. 62/78778’ and ‘Methods for predicting tumor response to immunotherapy, U.S. Provisional application no. 62/620,577’; and expert testimony for Tannoury, Nauts, McKinney & Garbarino, PLLC. WW reports consultant or advisory roles (Inst) for Agios, Bayer, MSD, Novartis and Roche; research funding (Inst) from Apogenix, Boehringer Ingelheim, Pfizer and Roche. SMC reports research funding (Inst) from Agios. ES reports consultant or advisory roles (Inst) for MedImmune, Inc., Agios Pharmaceuticals and Karyopharm; consultant or advisory roles for Gradalis, Inc. and Kyatec, Inc.; research funding (Inst) from MedImmune, Inc., Tracor, Genentech and Bristol-Meyers Squibb. MPM reports consultant or advisory roles for Zap, Mevion, Karyopharm, Tocagen and Astra-Zeneca; and Board of Directors options from Oncocentrix. ABH reports consultant or advisory roles for Caris Life Sciences and WCG Oncology; royalties on licensed intellectual property from Celldex Therapeutics and DNAtrix; research funding from Celularity, Carethera, Codiak, and Moleculin. DAR reports consultant or advisory roles for Abbvie, Advantagene, Agens, Amgen, Bayer, Bristol-Myers Squibb, Celldex, DelMar, EMD Serono, Genentech/Roche, Inxva, Invivo, Medicenna Biopharma, Inc., Merck, Merck KGaA, Monteris, Novocure, Oncorus, Oxigene, Regeneron, Stemline, Sumitomo Dainippon Pharma and Taiho Oncology, Inc.; honoraria from Abbvie, Advantagene, Agens, Bristol-Meyers Squibb, Celldex, EMD Serono, Genentech/Roche, Invivo, Invivo, Medicenna Biopharma, Inc., Merck, Merck KGaA, Monteris, Novocure, Oncorus, Oxigene, Regeneron, Stemline, Sumitomo Dainippon Pharma and Taiho Oncology, Inc.; research funding (Inst) from Akerta Pharmaceuticals, Agens, Celldex, EMD Serono, Invivo, Omixon and Tragara. SS reports consultant or advisory roles for Merck and Boehringer Ingelheim; research funding from Merck, Bristol-Meyers Squibb, and Brooklyn ImmunoTherapeutics. RJS reports research funding (Inst) from Bayer, Astra Zeneca, Pfizer, Astella, Roche, Merck, Abbvie and Amgen. SJA reports consultant or advisory roles for Achilles, Amgen, AstraZeneca, Bristol-Myers Squibb, Caris Life Sciences, CBMG, Celsius Therapeutics, G1 Therapeutics, GliaxonSmithKline, Meren, Merck, Nektar, RAPT Therapeutics, Venn, Glysmpe and Samyang; data review committee for EMD Serano. DB is co-owner of Berry Consultants, LLC, a company that designs adaptive Bayesian clinical trials for pharmaceutical and medical device companies, NIH cooperative groups, patient advocacy groups, and international consortia. MK reports consultant or advisory roles for Janssen, Abbvie, Ipsen, Pfizer Roche, and Jackson Laboratory for Genomic Medicine; research funding from Abbvie, Bristol-Meyers Squibb, and Specialized Therapeutics. AM, SK and VKP report no conflicts of interest.

Patient consent for publication Not required.

Provenance and peer review Not commissioned; externally peer reviewed.

Supplemental material This content has been supplied by the author(s). It has not been vetted by BMJ Publishing Group Limited (BMJ) and may not have been peer-reviewed. Any opinions or recommendations discussed are solely those of the author(s) and are not endorsed by BMJ. BMJ disclaims all liability and responsibility arising from any reliance placed on the content. Where the content includes any translated material, BMJ does not warrant the accuracy and reliability of the translations (including but not limited to local regulations, clinical guidelines, terminology, drug names and drug dosages), and is not responsible for any error and/or omissions arising from translation and adaptation or otherwise.

Open access This is an open access article distributed in accordance with the Creative Commons Attribution Non Commercial (CC BY-NC 4.0) license, which permits others to distribute, remix, adapt, build upon this work non-commercially, and license their derivative works on different terms, provided the original work is properly cited, appropriate credit is given, any changes made indicated, and the use or non-commercial. See http://creativecommons.org/licenses/by-nc/4.0/.

ORCID ids
Aaron C Tan http://orcid.org/0000-0001-7292-1114
Michael Plattten http://orcid.org/0000-0002-4746-887X
Amy B Heimberger http://orcid.org/0000-0002-9970-8695
Mustafa Khasraw http://orcid.org/0000-0003-3249-9849

REFERENCES
1 Scarlett UK, Chang DC, Murtagh TJ, et al. High-Throughput testing of Novel-Novel combination therapies for cancer: an idea whose time has come. Cancer Discov 2016;6:956–62.
metastatic melanoma: an overview of rationale, preclinical evidence, and initial clinical data. *Front Oncol* 2015;5:202.

28 Yasuda S, Sho M, Yamato I, et al. Simultaneous blockade of programmed death 1 and vascular endothelial growth factor receptor 2 (VEGFR2) induces synergistic anti-tumour effect in vivo. *Clin Exp Immunol* 2013;172:500–6.

29 Allen E, Jabouille A, Rivera LB, et al. Combined antiangiogenic and anti-PD-L1 therapy stimulates tumor immunity through HEV formation. *Sci Transl Med* 2017;9(391):eaat0697. [Epub ahead of print: 12 Apr 2017]

30 Kato Y, Tabata K, Kimura T, et al. Lenvatinib plus anti-PD-1 antibody combination treatment activates CD8+ T cells through reduction of tumor-associated macrophage and activation of the interferon pathway. *PLoS One* 2019;14:e0212513.

31 Kimura T, Kato Y, Ozawa Y, et al. Immunomodulatory activity of lenvatinib contributes to antitumor activity in the Hepa1-6 hepatocellular carcinoma model. *Cancer Sci* 2018;109:3999–4002.

32 Choueiri TK, Larkin J, Oya M, et al. Preliminary results for avelumab plus axitinib as first-line therapy in patients with advanced clear-cell renal-cell carcinoma (javelin renal 100): an open-label, dose-finding and dose-expansion, phase 1b trial. *Lancet Oncol* 2018;19:451–60.

33 Motzer RJ, Penkov K, Haenen J, et al. Avelumab plus axitinib versus sunitinib for advanced renal-cell carcinoma. *N Engl J Med* 2019;380:1103–15.

34 Rini BI, Powles T, Atkins MB, et al. Atezolizumab plus bevacizumab versus sunitinib in patients with previously untreated metastatic renal cell carcinoma (IMmotion151): a multicentre, open-label, phase 3, randomised controlled trial. *Lancet* 2019;393:2404–15.

35 Rini BI, Escudier B, Tomczak P, et al. Comparative effectiveness of axitinib versus sorafenib in patients with previously untreated advanced renal cell carcinoma: (randomised phase 3 trial. *Lancet* 2011;378:1931–9.

36 Kopetz S, Grothey A, Yaeger R, et al. Encorafenib, Binimetinib, and Cetuximab in BRAF V600E-Mutated Colorectal Cancer. *N Engl J Med* 2019;381:1632–43.

37 Long GV, Straykovsky D, Gogas H, et al. Combined BRAF and MEK inhibition versus BRAF inhibition alone in melanoma. *N Engl J Med* 2014;371:1867–75.

38 Paraiso KHT, Fedorenko IV, Cantini LP, Munco AK, et al. Recovery of phospho-ERK activity allows melanoma cells to escape from BRAF inhibitor therapy. *Br J Cancer* 2010;102:1724–30.

39 King AJ, Arnone MR, Bleam MR, et al. Dabrafenib; preclinical characterization, increased efficacy when combined with trametinib, while BRAF/MEK combinatorial skin lesions. *PLoS One* 2013;8:e67583.

40 Rizos H, Menzies AM, Pupp GM, et al. BRAF inhibitor resistance mechanisms in metastatic melanoma: spectrum and clinical impact. *Clin Cancer Res* 2014;20:3815–27.

41 Van Allen EM, Wagle N, Sucker A, et al. The genetic landscape of clinical resistance to RAF inhibition in metastatic melanoma. *Cancer Discov* 2014;4:94–109.

42 Delord J-P, Robert N, Nakazawa Y, et al. Phase I Dose-Escalation and -Expansion Study of the BRAF Inhibitor Encorafenib (LGX818) in Metastatic BRAF-Mutant Melanoma. *Cancer Res* 2017;73:5339–48.

43 Prahallad A, Sun C, Huang S, et al. Unresponsiveness of colon cancer to BRAF(V600E) inhibition through feedback activation of EGFR. *Nature* 2012;483:100–3.

44 Mao T, Tian F, Mariadason JM, et al. Resistance to BRAF inhibition in BRAF-mutant colon cancer can be overcome with PI3K inhibition or demethylating agents. *Clin Cancer Res* 2013;19:657–67.

45 Corcoran RB, Dias-Santagata D, Bergethon K, et al. BRAF gene amplification can promote acquired resistance to MEK inhibitors in cancer cells harboring the BRAF V600E mutation. *Sci Signal* 2010;3:ra84.

46 Corcoran RB, Ebi H, Turke AB, et al. EGFR-Mediated re-activation of MAPK signaling contributes to in vivo resistance of BRAF mutant colorectal cancers to RAF inhibition with vemurafenib. *Cancer Discov* 2012;2:227–35.

47 Dummer R, Ascierto PA, Gogas HJ, et al. Encorafenib plus binimetinib versus vemurafenib or encorafenib in patients with BRAF-mutant colorectal cancer. *Lancet Oncol* 2019;20:19:603–15.

48 Ascierto PA, Schadendorf D, Berking C, et al. MEK162 for patients with advanced melanoma harbouring NRAS or V600 BRAF mutations: a non-randomised, open-label, phase 2 study. *Lancet* 2013;381:1497–505.

49 Jonker DJ, O’Callaghan CJ, Karapetis CS, et al. Cetuximab for the treatment of colorectal cancer. *N Engl J Med* 2007;357:2040–8.
Supplementary Appendix

Inhibition of PD-1/PD-L1 and CTLA-4 combination therapy

Comparator arms

All three phase II trials evaluating nivolumab plus ipilimumab, were randomized two-arm trials in the treatment resistant setting, with nivolumab monotherapy as the comparator arm. NRG GY003 compared nivolumab plus ipilimumab versus nivolumab alone in recurrent or persistent ovarian cancer [1]. Nivolumab and ipilimumab monotherapy had been evaluated in previous single-arm phase II trials and case series [2–4]. Alliance A09140111, for treatment-refractory metastatic sarcoma, and IFCT-1501 MAPS2, for platinum-pemetrexed resistant pleural mesothelioma, were both non-comparative studies – evaluating both the combination and nivolumab alone. There was a single small phase II trial of ipilimumab alone in patients with recurrent synovial sarcoma [5]; but no studies of nivolumab alone. There had been no previous trials of either nivolumab or ipilimumab monotherapy in malignant pleural mesothelioma.

There were three phase II trials evaluating the combination of durvalumab plus tremelimumab. CONDOR was a three-arm non-comparative randomized trial in PD-L1 low/negative (TC <25%) recurrent or metastatic head and neck squamous cell carcinoma (HNSCC).[6] There had been several prior phase II trials of durvalumab in HNSCC, from which greater response rates in patients with PD-L1 TC ≥25% had been seen [7,8]. However, there were no prior trials of tremelimumab monotherapy in HNSCC. The clinical trial CO.26 evaluated durvalumab plus tremelimumab versus best supportive care (BSC) for patients with metastatic treatment-refractory colorectal cancer (CRC) in a randomized two-arm study [9]. There had been no prior studies of either agent alone in treatment-refractory CRC. Finally, durvalumab plus tremelimumab was also evaluated in patients with metastatic pancreatic adenocarcinoma in a randomized, two-part, two-arm trial, with durvalumab monotherapy in the other arm [10]. Preliminary safety and efficacy data had been obtained from a phase 1 expansion cohort of durvalumab monotherapy [11]; however, there were no prior studies of tremelimumab monotherapy.

Inhibition of PD-1/PD-L1 and VEGF combination therapy

Comparator arms

There were five trials evaluating patients with metastatic RCC, including two phase II trials and three phase III trials. The phase II trials were single-arm studies evaluating atezolizumab plus bevacizumab in non-clear cell RCC [12] and pembrolizumab plus bevacizumab in pre-treated clear cell RCC [13] respectively. In each phase III trial,
Inhibition of BRAF and MEK combination therapy

Rationale for combinations

The clinical activity of combination BRAF and MEK inhibitor seen in metastatic BRAF-mutated melanoma resulted in the evaluation of this combination in other cancers known to harbor oncogenic BRAF V600E mutations. In BRAF V600E mutated NSCLC, pre-clinical studies had demonstrated the role for BRAF inhibition, and also the potential sensitivity to MEK inhibition [37–39]. The combination of BRAF and MEK inhibition, however, had not been extensively evaluated in the pre-clinical setting. In BRAF V600E mutated anaplastic thyroid cancer, pre-clinical mouse and cell line models demonstrated the improved efficacy of combination BRAF and MEK inhibition compared with BRAF inhibition alone [40].

Comparator arms and primary endpoints

The trial evaluating dabrafenib plus trametinib in metastatic BRAF mutated NSCLC was a multicohort phase II trial [41]. Cohort A investigated dabrafenib monotherapy in both pre-treated and untreated patients, cohort B investigated dabrafenib plus trametinib in pre-
treated patients, whilst cohort C investigated dabrafenib plus trametinib in first-line patients [42,43]. Trametinib monotherapy had not been previously evaluated for \(BRAF\) mutated NSCLC. ROAR was a basket trial of dabrafenib plus trametinib in nine different \(BRAF\) V600E mutated rare cancers, including anaplastic thyroid cancer. There had been no prior studies of dabrafenib or trametinib monotherapy in anaplastic thyroid cancer, although dabrafenib has been studied in \(BRAF\) V600E mutated papillary thyroid cancers [44]. The primary endpoint in the two phase II trials of dabrafenib and trametinib was ORR, assessed as appropriate and clinically meaningful.

**Inhibition of HER2 combination therapy**

**Rationale for combinations**

In salivary gland cancers, there was no pre-clinical data indicating the potential efficacy of the combination over monotherapy. This may be in part, due to the relative rarity of salivary gland cancers.

**Utility of biomarkers**

The MyPathway trial included a basket for patients with advanced solid tumors harboring HER2 alterations that included HER2 amplification, overexpression and/or mutation. Furthermore, these HER2 alterations could be locally assessed by any Clinical Laboratory Improvement Amendments (CLIA) certified test which would subsequently be assessed for eligibility by the medical monitor [45]. Specifically for colorectal cancer patients, HER2 alterations were limited to HER2 amplification, as assessed by a CLIA certified test. This could include fluorescence or chromogenic in situ hybridization (FISH/CISH), next-generation sequencing (NGS) or immunohistochemistry (IHC) [46].

**Comparator arms and primary endpoints**

There were also no prior studies of monotherapy treatment in either tumor type. However this should be considered in the context of pre-clinical data suggesting lack of efficacy for monotherapy in HER2-amplified colorectal cancer, as described above [47,48].

**Other combination therapies**

ISA 101 is a synthetic long-peptide HPV-16 vaccine, and was evaluated in combination with nivolumab in a single-arm phase II trial [49]. Eligible patients had incurable HPV-16-positive cancer, and included oropharyngeal, cervical, vulvar, vaginal, penile or anal primary tumors. The rationale for the combination therapy was on the basis of pre-clinical data demonstrating HPV-specific T-cell responses with ISA 101 in cervical cancer models [50,51]. However, in the early phase trials of ISA 101 monotherapy in patients with HPV-16 induced
gynecological cancers, there was no tumor regression [52,53]. It was hypothesized that this may be due to a tumor-induced immunosuppressive environment, and combination with anti-PD-1 therapy could improve efficacy [53]. Although there had been no pre-clinical models which evaluated the combination therapy. There were no prior studies of ISA 101 monotherapy in non-gynecological cancers, whilst nivolumab had previously been approved as monotherapy in unselected recurrent squamous cell carcinoma of the head and neck [54].

Pembrolizumab plus talimogene laherparepvec (T-VEC) combination therapy was investigated in a single-arm phase II trial for patients with pre-treated locally advanced or metastatic sarcoma [55]. T-VEC, an oncolytic immunotherapy, is approved in patients with advanced melanoma as monotherapy [56]. However, T-VEC monotherapy had not previously been evaluated in patients with sarcoma, whilst pembrolizumab monotherapy had been evaluated in patients with advanced sarcomas in a single-arm phase II trial [32]. There was also no pre-clinical data investigating the combination therapy, although the combination had been evaluated in a clinical trial for patients with metastatic melanoma [57].

References

1. Zamarin D, Burger RA, Sill MW, Powell DJJ, Lankes HA, Feldman MD, et al. Randomized Phase II Trial of Nivolumab Versus Nivolumab and Ipilimumab for Recurrent or Persistent Ovarian Cancer: An NRG Oncology Study. J Clin Oncol. 2020;38:1814–23.

2. Hamanishi J, Mandai M, Ikeda T, Minami M, Kawaguchi A, Murayama T, et al. Safety and Antitumor Activity of Anti-PD-1 Antibody, Nivolumab, in Patients With Platinum-Resistant Ovarian Cancer. J Clin Oncol. 2015;33:4015–22.

3. Gaillard SL, Secord AA, Monk B. The role of immune checkpoint inhibition in the treatment of ovarian cancer. Gynecologic Oncology Research and Practice. 2016;3:11.

4. Hodi FS, Butler M, Oble DA, Seiden MV, Haluska FG, Kruse A, et al. Immunologic and clinical effects of antibody blockade of cytotoxic T lymphocyte-associated antigen 4 in previously vaccinated cancer patients. Proc Natl Acad Sci USA. 2008;105:3005–10.

5. Maki RG, Jungbluth AA, Gnjatic S, Schwartz GK, D’Adamo DR, Keohan ML, et al. A Pilot Study of Anti-CTLA4 Antibody Ipilimumab in Patients with Synovial Sarcoma. Sarcoma. 2013;2013:168145.

6. Siu LL, Even C, Mesia R, Remenar E, Daste A, Delord J-P, et al. Safety and Efficacy of Durvalumab With or Without Tremelimumab in Patients With PD-L1-Low/Negative Recurrent or Metastatic HNSCC: The Phase 2 CONDOR Randomized Clinical Trial. JAMA Oncol. 2019;5:195–203.

7. Zandberg DP, Algazi AP, Jimeno A, Good JS, Fayette J, Bouganim N, et al. Durvalumab for recurrent or metastatic head and neck squamous cell carcinoma: Results from a single-arm, phase II study in patients with ≥25% tumour cell PD-L1 expression who have progressed on platinum-based chemotherapy. Eur J Cancer. 2019;107:142–52.

8. Segal NH, Ou S-HI, Balmanoukian A, Fury MG, Massarelli E, Brahmer JR, et al. Safety and efficacy of durvalumab in patients with head and neck squamous cell carcinoma: results from a phase I/II expansion cohort. Eur J Cancer. 2019;109:154–61.
9. Chen EX, Jonker DJ, Loree JM, Kennecke HF, Berry SR, Couture F, et al. 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;6:831–8.

10. O'Reilly EM, Oh D-Y, Dhani N, Renouf DJ, Lee MA, Sun W, et al. Durvalumab With or Without Tremelimumab for Patients With Metastatic Pancreatic Ductal Adenocarcinoma: A Phase 2 Randomized Clinical Trial. JAMA Oncol. 2019;5:1431–8.

11. Segal NH, Hamid O, Hwu W, Massard C, Butler M, Antonia S, et al. A Phase I Multi-Arm Dose-Expansion Study of the Anti-Programmed Cell Death-Ligand-1 (Pd-L1) Antibody Medi4736: Preliminary Data. Annals of Oncology. Elsevier; 2014;25:i365.

12. McGregor BA, McKay RR, Braun DA, Werner L, Gray K, Flaifel A, et al. Results of a Multicenter Phase II Study of Atezolizumab and Bevacizumab for Patients With Metastatic Renal Cell Carcinoma With Variant Histology and/or Sarcomatoid Features. J Clin Oncol. 2020;38:63–70.

13. Dudek AZ, Liu LC, Gupta S, Logan TF, Singer EA, Joshi M, et al. Phase Ib/II Clinical Trial of Pembrolizumab With Bevacizumab for Metastatic Renal Cell Carcinoma: BTCRC-GU14-003. J Clin Oncol. 2020;38:1138–45.

14. Motzer RJ, Penkov K, Haanen J, Rini B, Albige L, Campbell MT, et al. Avelumab plus Axitinib versus Sunitinib for Advanced Renal-Cell Carcinoma. N Engl J Med. 2019;380:1103–15.

15. Rini BI, Plimack ER, Stus V, Gafanov R, Hawkins R, Nosov D, et al. Pembrolizumab plus Axitinib versus Sunitinib for Advanced Renal-Cell Carcinoma. N Engl J Med. United States; 2019;380:1116–27.

16. Rini BI, Powles T, Atkins MB, Escudier B, McDermott DF, Suarez C, et al. Atezolizumab plus bevacizumab versus sunitinib in patients with previously untreated metastatic renal cell carcinoma (IMmotion151): a multicentre, open-label, phase 3, randomised controlled trial. Lancet. England; 2019;393:2404–15.

17. Rini BI, Escudier B, Tomczak P, Kaprin A, Szczyluk C, Hutson TE, et al. Comparative effectiveness of axitinib versus sorafenib in advanced renal cell carcinoma (AXIS): a randomised phase 3 trial. Lancet (London, England). 2011;378:1931–9.

18. Yang JC, Haworth L, Sherry RM, Hwu P, Schwartzenbruber DJ, Topalian SL, et al. A randomized trial of bevacizumab, an anti-vascular endothelial growth factor antibody, for metastatic renal cancer. N Engl J Med. 2003;349:427–34.

19. Bukowski RM, Kabinavaran FF, Figlin RA, Flaherty K, Srinivas S, Vaishampayan U, et al. Avelumab monotherapy as first-line or second-line treatment in patients with metastatic renal cell carcinoma: phase Ib results from the JAVELIN Solid Tumor trial. J Immunother Cancer. 2019;7:275.
24. Taylor MH, Lee C-H, Makker V, Rasco D, Dutcus CE, Wu J, et al. Phase IB/II Trial of Lenvatinib Plus Pembrolizumab in Patients With Advanced Renal Cell Carcinoma, Endometrial Cancer, and Other Selected Advanced Solid Tumors. J Clin Oncol. 2020;38:1154–63.

25. Makker V, Rasco D, Vogelzang NJ, Brose MS, Cohn AL, Mier J, et al. Lenvatinib plus pembrolizumab in patients with advanced endometrial cancer: an interim analysis of a multicentre, open-label, single-arm, phase 2 trial. Lancet Oncol. England; 2019;20:711–8.

26. Robert C, Schachter J, Long GV, Arance A, Grob JJ, Mortier L, et al. Pembrolizumab versus Ipilimumab in Advanced Melanoma. The New England journal of medicine. 2015;372:2521–32.

27. Motzer RJ, Hutson TE, Glen H, Michaelson MD, Molina A, Eisen T, et al. Lenvatinib, everolimus, and the combination in patients with metastatic renal cell carcinoma: a randomised, phase 2, open-label, multicentre trial. Lancet Oncol. 2015;16:1473–82.

28. Garon EB, Rizvi NA, Hui R, Leighl N, Balmanoukian AS, Eder JP, et al. Pembrolizumab for the treatment of non-small-cell lung cancer. The New England journal of medicine. 2015;372:2018–28.

29. Herbst RS, Baas P, Kim DW, Felip E, Perez-Gracia JL, Han JY, et al. Pembrolizumab versus docetaxel for previously treated, PD-L1-positive, advanced non-small-cell lung cancer (KEYNOTE-010): a randomised controlled trial. Lancet (London, England). 2016;387:1540–50.

30. Patnaik A, Kang SP, Rasco D, Papadopoulos KP, Elassaiss-Schaap J, Beeram M, et al. Phase I Study of Pembrolizumab (MK-3475; Anti-PD-1 Monoclonal Antibody) in Patients with Advanced Solid Tumors. Clin Cancer Res. 2015;21:4286–93.

31. Young RJ, Woll PJ. Anti-angiogenic therapies for the treatment of angiosarcoma: a clinical update. Memo. 2017;10:190–3.

32. Dankort D, Filenova E, Collado M, Serrano M, Jones K, McMahon M. A new mouse model to explore the initiation, progression, and therapy of BRAFV600E-induced lung tumors. Genes Dev. 2007;21:379–84.
40. McFadden DG, Vernon A, Santiago PM, Martinez-McFaline R, Bhutkar A, Crowley DM, et al. p53 constrains progression to anaplastic thyroid carcinoma in a Braf-mutant mouse model of papillary thyroid cancer. Proc Natl Acad Sci U S A. 2014;111:E1600-1609.

41. Planchard D, Smit EF, Groen HJM, Mazieres J, Besse B, Helland Å, et al. Dabrafenib plus trametinib in patients with previously untreated BRAFV600E-mutant metastatic non-small-cell lung cancer: an open-label, phase 2 trial. The Lancet Oncology. 2017;18:1307–16.

42. Planchard D, Kim TM, Mazieres J, Quoix E, Riely G, Barlesi F, et al. Dabrafenib in patients with BRAF(V600E)-positive advanced non-small-cell lung cancer: a single-arm, multicentre, open-label, phase 2 trial. Lancet Oncol. 2016;17:642–50.

43. Planchard D, Besse B, Groen HJM, Souquet P-J, Quoix E, Baik CS, et al. Dabrafenib plus trametinib in patients with previously treated BRAF(V600E)-mutant metastatic non-small-cell lung cancer: an open-label, multicentre phase 2 trial. Lancet Oncol. 2016;17:984–93.

44. Shah MH, Wei L, Wirth LJ, Daniels GA, De Souza JA, Timmers CD, et al. Results of randomized phase II trial of dabrafenib versus dabrafenib plus trametinib in BRAF-mutated papillary thyroid carcinoma. J Clin Oncol. American Society of Clinical Oncology; 2017;35:6022–6022.

45. Kurzrock R, Bowles DW, Kang H, Meric-Bernstam F, Hainsworth J, Spigel DR, et al. Targeted therapy for advanced salivary gland carcinoma based on molecular profiling: results from MyPathway, a phase IIa multiple basket study. Ann Oncol. England; 2020;31:412–21.

46. Meric-Bernstam F, Hurwitz H, Raghav KPS, McWilliams RR, Fakih M, VanderWalde A, et al. Pertuzumab plus trastuzumab for HER2-amplified metastatic colorectal cancer (MyPathway): an updated report from a multicentre, open-label, phase 2a, multiple basket study. Lancet Oncol. 2019;20:518–30.

47. Leto SM, Sassi F, Catalano I, Torri V, Migliardi G, Zanella ER, et al. Sustained Inhibition of HER3 and EGFR Is Necessary to Induce Regression of HER2-Amplified Gastrointestinal Carcinomas. Clinical Cancer Research: An Official Journal of the American Association for Cancer Research. 2015;21:5519–31.

48. Bertotti A, Migliardi G, Galimi F, Sassi F, Torti D, Isella C, et al. A molecularly annotated platform of patient-derived xenografts ("xenopatients") identifies HER2 as an effective therapeutic target in cetuximab-resistant colorectal cancer. Cancer Discovery. 2011;1:508–23.

49. Massarelli E, William W, Johnson F, Kies M, Ferrarotto R, Guo M, et al. Combining Immune Checkpoint Blockade and Tumor-Specific Vaccine for Patients With Incurable Human Papillomavirus 16-Related Cancer: A Phase 2 Clinical Trial. JAMA Oncol. 2019;5:67–73.

50. Bijker MS, van den Eeden SJF, Franken KL, Melief CJM, van der Burg SH, Offringa R. Superior induction of anti-tumor CTL immunity by extended peptide vaccines involves prolonged, DC-focused antigen presentation. Eur J Immunol. 2008;38:1033–42.

51. Rosalia RA, Quakkelhaar ED, Redeker A, Khan S, Camps M, Drijfhout JW, et al. Dendritic cells process synthetic long peptides better than whole protein, improving antigen presentation and T-cell activation. Eur J Immunol. 2013;43:2554–65.

52. Kenter GG, Welters MJ, Valentijn ARPM, Lowik MJG, Vloon APG, et al. Phase I immunotherapeutic trial with long peptides spanning the E6 and E7 sequences of high-risk human papillomavirus 16 in end-stage cervical cancer patients shows low toxicity and robust immunogenicity. Clin Cancer Res. 2008;14:169–77.

53. van Poelgeest MIE, Welters MJ, van Esch EMG, Stynenbosch LFM, Kerpershoek G, van Persijn van Meerten EL, et al. HPV16 synthetic long peptide (HPV16-SLP) vaccination therapy of patients with advanced or recurrent HPV16-induced gynecological carcinoma, a phase II trial. J Transl Med. 2013;11:88.
54. Ferris RL, Blumenschein G, Fayette J, Guigay J, Colevas AD, Licitra L, et al. Nivolumab for Recurrent Squamous-Cell Carcinoma of the Head and Neck. N Engl J Med. 2016;375:1856–67.

55. Kelly CM, Antonescu CR, Bowler T, Munhoz R, Chi P, Dickson MA, et al. Objective Response Rate Among Patients With Locally Advanced or Metastatic Sarcoma Treated With Talimogene Laherparepvec in Combination With Pembrolizumab: A Phase 2 Clinical Trial. JAMA Oncol. 2020;6:402–8.

56. Andtbacka RHI, Kaufman HL, Collichio F, Amatruda T, Senzer N, Chesney J, et al. Talimogene Laherparepvec Improves Durable Response Rate in Patients With Advanced Melanoma. J Clin Oncol. 2015;33:2780–8.

57. Ribas A, Dummer R, Puzanov I, VanderWalde A, Andtbacka RHI, Michielin O, et al. Oncolytic Virotherapy Promotes Intratumoral T Cell Infiltration and Improves Anti-PD-1 Immunotherapy. Cell. 2017;170:1109-1119.e10.
# Supplementary Tables

## Supplementary Table 1. Combination anti-PD-1/PD-L1 and anti-VEGF therapy trials

| Study Name | Year, Lead Author | Tumor Type | Line of Therapy | Combination Regimen | Total Patient Number | Biomarker Selection | Primary Endpoint | Trial Design | Primary Endpoint Met | FDA Approved Therapy | Strong Biologic Rational | New Biomarker | Comparator Arms of Monotherapy | Clinically Meaningful Primary Endpoint |
|------------|-------------------|------------|-----------------|---------------------|----------------------|---------------------|------------------|--------------|------------------------|----------------------|-----------------------|----------------|--------------------------------|----------------------------------|
| **Phase II trials** |
| KEYNOTE-146 | 2019, Makk er | Endometrial | 1 to 3 | Lenvatinib, pembrolizumab | 54 | Unselected | ORR at 24w | Pembro+lenvatinib (single-arm/basket) | Yes | Yes | No | No | Yes | Yes |
| NCT02636725 | 2019, Wilky | Sarcoma | 2+ | Axitinib, pembrolizumab | 33 | Unselected | PFS rate at 3m | Pembro+axitinib (single-arm) | Yes | No | No | No | Yes | Yes |
| NCT02673962 | 2019, Liu | Ovarian | 2+ | Bevacizumab, nivolumab | 38 | Unselected | ORR | Nivo+bevacizumab (single-arm) | Yes | No | No | No | Yes | Yes |
| NCT02724878 | 2020, McGr egor | RCC (non-clear cell) | 1+ | Atezolizumab, bevacizumab | 60 | Unselected | ORR | Atezo+bevacizumab (single-arm) | Yes | No | No | No | Yes | Yes |
| KEYNOTE-146 | 2020, Taylor | Multiple | 2+ | Lenvatinib, pembrolizumab | 137 | Unselected | ORR at 24w | Pembro+lenvatinib (single-arm/basket) | Yes | No | Limited | No | Yes | Yes |
| BTCRC-GU14-003 | 2020, Dude k | RCC | 2+ | Bevacizumab, pembrolizumab | 48 | Unselected | ORR | Pembro+bevacizumab (single-arm) | Yes | No | No | No | Yes | Yes |
| **Phase III trials** |
| JAVELIN Renal 101 | 2019, Motze r | RCC | 1 | Avelumab, axitinib | 560 | PD-L1 | Co-primary: PFS & OS (in PD-L1 positive) | Avelumab+axitinib or sunitinib (1:1) | Yes | Yes | No | Yes | No | Yes |
| Study     | Year | Disease | Phase | Treatment 1 | Treatment 2 | Co-primary: PFS, OS | Pembro+axitinib or sunitinib (1:1) | Yes | No | No | No | Yes |
|-----------|------|---------|-------|-------------|-------------|---------------------|-------------------------------------|-----|----|----|----|-----|
| KEYNOTE-426 | 2019 | RCC     | 1     | Axitinib, pembrolizumab | 861         | Unselected          | Co-primary: PFS, OS                  | Yes | Yes | No | No | Yes |
| IMmotion151 | 2019 | RCC     | 1     | Atezolizumab, bevacizumab | 362         | PD-L1               | Co-primary: PFS (in PD-L1 positive), OS | Yes | No | No | Yes | Yes |
| IMbrave150  | 2020 | HCC     | 1     | Atezolizumab, bevacizumab | 501         | Unselected          | Co-primary: PFS, OS                  | Yes | Yes | No | No | Yes |

**Abbreviations:** Atezo = atezolizumab; HCC = hepatocellular carcinoma; m = months; Nivo = nivolumab; ORR = objective response rate; OS = overall survival; Pembro = pembrolizumab; PFS = progression-free survival; RCC = renal cell carcinoma; w = weeks.

**Supplementary Table 2.** Combination anti-BRAF and anti-MEK therapy trials
| Study Name     | Year of Publication | Tumor Type | Combination Regimen | Total Patient Number | Biomarker Selection | Primary Endpoint | Trial Design | FDA Approved Therapy | Strong Biologic Rational | New Biomarker | Comparat or Arms of Monotherapy | Clinical Meaningful Primary Endpoint |
|---------------|---------------------|------------|---------------------|----------------------|---------------------|------------------|--------------|----------------------|---------------------------|---------------|-----------------------------|-----------------------------------|
| NCT01336634   | 2017, Planchard      | NSCLC      | Dabrafenib, trametinib | 36                   | BRAF V600E          | ORR              | D+T (single-arm/multiple cohort) | Yes                    | Yes                       | No            | Yes                        | No                                |
| ROAR          | 2018, Subbiah        | Thyroid    | Dabrafenib, trametinib | 16                   | BRAF V600E          | ORR              | D+T (single-arm/basket)           | Yes                    | Yes                       | Yes                       | Yes                        | Yes                                |
| COLUMBUS      | 2018, Dummer         | Melanoma   | Binimetinib, encorafenib | 383                  | BRAF V600E          | PFS (E+B vs V)    | E+B or E or V (1:1:1)              | Yes                    | Yes                       | Yes                       | Yes                        | Yes                                |
| BEACON CRC    | 2019, Kopetz          | Colorectal | Binimetinib, cetuximab, encorafenib | 445                 | BRAF V600E          | Co-primary: OS & ORR (E+B+C vs FOLFIrI/irinotecan+C) | E+B+C or E+C or FOLFIrI/irinotecan+C (1:1:1) | Yes | No* | Yes | Yes | No | Yes |

Abbreviations: B = binimetinib; C = cetuximab; D = dabrafenib; E = encorafenib; NSCLC = non-small cell lung cancer; ORR = objective response rate; OS = overall survival; PFS = progression-free survival; T = trametinib; V = vemurafenib.

*The combination of encorafenib plus cetuximab has been FDA approved

**Supplementary Table 3. Combination anti-HER2 therapy trials**
| Study Name | Year, Lead Author | Tumor Type | Line of Therapy | Total Patient Number | Biomarker Selection | Primary Endpoint | Trial Design | Primary Endpoint Met | FDA Approved Therapy | Strong Biologic Rational | New Biomarker | Comparator Arms of Monotherapy | Clinical Meaningful Primary Endpoint |
|------------|-------------------|------------|-----------------|----------------------|---------------------|------------------|-------------|----------------------|-----------------------|-------------------------|----------------|-----------------------------|---------------------------------|
| MyPathway  | 2019, Meric-Bernstam | Colorectal | 2+ Pertuzumab, trastuzumab | 57 | HER2 | ORR | T+P (single-arm/basket) | Yes | No | Yes | Yes | No | Yes |
| MyPathway  | 2020, Kurzrock | Salivary | 2+ Pertuzumab, trastuzumab | 19 | HER2 | ORR | T+P (single-arm/basket) | Yes | No | Yes | Yes | No | Yes |

Abbreviations: ORR = objective response rate; P = pertuzumab; T = trastuzumab.

**Supplementary Table 4.** Other combination therapy trials
| Study Name | Year, Lead Author | Tumor Type | Line of Therapy | Combination Regimen | Total Patient Number | Biomarker Selection | Primary Endpoint | Trial Design | Prim Endp Met | FDA Approve Therap | Strong Biologic Rational | New Biomarker | Compar Arms of Monothrapy | Clinical Meaningful Primary Endpnt |
|------------|------------------|------------|-----------------|---------------------|----------------------|--------------------|-----------------|--------------|--------------|----------------|---------------------|----------------|------------------------|-----------------------------|
| Phase II trials |
| NCT02187302 | 2017, Voss | RCC | 3 or 4 | Bevacizumab, CRLX101 | 111 | Unselected | PFS (in clear-cell RCC) | CRLX101+bevacizumab or SOC (1:1) | No | No | No | No | No | Yes |
| BROCADE | 2018, Han | Breast | 1 or 2 | Temozolomide, veliparib | 290 | BRCA1/2 | PFS (VCP vs CP, VT vs CP) | V+CP or V+T or CP (1:1:1) | No | No | Yes | Yes | Yes | Yes |
| NCT02426892 | 2019, Massarelli | Multiple | 1 or 2 | ISA 101, nivolumab | 24 | HPV-16 | ORR | ISA 101+nivolumab (single-arm) | Yes | No | No | Yes | No | Yes |
| NCT03069378 | 2020, Kelly | Sarcoma | 2+ | Pembrolizumab, talimogene laherparepvec | 20 | Unselected | ORR at 24w | Pembrolizumab+T-VEC (single-arm) | Yes | No | No | No | Yes | Yes |
| Phase III trials |
| COTEZO IMblaze370 | 2019, Eng | Colorectal | 3+ | Atezolizumab, cobimetinib | 363 | Unselected | Co-primary: OS (atezolizumab+cobimetinib vs regorafenib, atezolizumab vs regorafenib) | Atezolizumab+cobimetinib or atezolizumab or regorafenib (2:1:1) | No | No | Yes | No | Yes | Yes |

Abbreviations: CP = carboplatin/paclitaxel; ORR = objective response rate; OS = overall survival; PFS = progression-free survival; RCC = renal cell carcinoma; SOC = standard of care; T = temozolomide; T-VEC = talimogene laherparepvec; V = veliparib; w = weeks.
A systematic review of combinations of targeted or immunotherapy in advanced solid tumors

**Strong Biological Rationale**
- Yes (n=6, 19%)
- Limited (n=3, 9%)
- No (n=23, 72%)

**New Biomarker**
- Yes (n=10, 31%)
- No (n=22, 69%)

**Comparator Arms of Monotherapy**
- Yes (n=21, 66%)
- No (n=11, 34%)

**Authors**
Aaron C. Tan, Stephen J. Bagley, Patrick Y. Wen, Michael Lim, Michael Platten, Howard Colman, David M. Ashley, Wolfgang Wick, Susan M. Chang, Evanthia Galanis, Alireza Mansouri, Simon Khagi, Minesh P. Mehta, Amy B. Heimberger, Vinay K. Puduvalli, David A. Reardon, Solmaz Sahebjam, R. John Simes, Scott J. Antonia, Don Berry, Mustafa Khasraw

**Correspondence**
mustafa.khasraw@duke.edu

**In Brief**
Designing trials to evaluate novel-novel combination therapies presents numerous challenges to demonstrate efficacy in a comprehensive manner. A greater understanding of biological rationale for combinations and incorporating biomarkers may improve effective evaluation of combination therapies. Innovative statistical methods and increasing use of external data to support combination approaches are potential strategies that may improve the efficiency of trial design.