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Journal
Seminars in Immunology, 28(1)

ISSN
1044-5323

Authors
Robert, Lidia
Ribas, Antoni
Hu-Lieskovan, Siwen

Publication Date
2016-02-01

DOI
10.1016/j.smim.2016.01.001

Peer reviewed

HHS Public Access

Author manuscript

Semin Immunol. Author manuscript; available in PMC 2017 February 06.

Published in final edited form as:

Semin Immunol. 2016 February ; 28(1): 73–80. doi:10.1016/j.smim.2016.01.001.

Combining targeted therapy with immunotherapy. Can 1+1 equal more than 2?

Lidia Robert[a], **Antoni Ribas**[b], and **Siwen Hu-Lieskovan**[c]

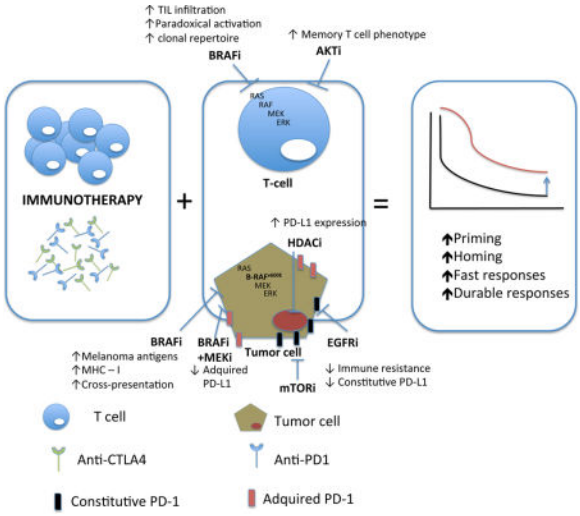
[a]Division of Hematology-Oncology, Department of Medicine, University of California Los Angeles (UCLA), Los Angeles, CA 90095, USA

[b]Division of Hematology-Oncology, Department of Medicine, University of California Los Angeles (UCLA), Los Angeles, CA 90095, USA; Department of Surgery, University of California Los Angeles (UCLA), Los Angeles, CA 90095, USA; Department of Medical and Molecular Pharmacology, University of California Los Angeles (UCLA), Los Angeles, CA 90095, USA; Jonsson Comprehensive Cancer Center (JCCC) at UCLA, Los Angeles, CA 90095, USA

[c]Division of Hematology-Oncology, Department of Medicine, University of California Los Angeles (UCLA), Los Angeles, CA 90095, USA; Jonsson Comprehensive Cancer Center (JCCC) at UCLA, Los Angeles, CA 90095, USA

Graphical abstract

How can targeted therapy synergize immunotherapy?



1. Introduction

Recent advances in the understanding of anti-tumor immunity and tumor immune escape have facilitated the design of new immunotherapy agents for the treatment of cancer. The clinical development of these agents in clinical trials has resulted in long-term survival in a subset of patients with different cancer histologies and has opened the door to combinatorial therapies that could improve outcomes. In parallel, the discovery of oncogenic driver pathways in different tumor types and the development of inhibitory molecules targeting these pathways signified another major development in the treatment of metastatic cancer. Hence, there is considerable interest in testing the combination of both treatment modalities in ongoing early stage clinical trials.

The treatment of metastatic melanoma is at the center stage of this research effort. Prior to 2011, very limited treatments with demonstrated survival benefits were available. However, since the launch of ipilimumab in 2011, the US FDA has approved eight different single or combinations of agents, which has led to significant improvement in response rates and survival of patients with melanoma. These new agents are either targeted inhibitors of the mitogen-activated protein kinase (MAPK) oncogenic signaling pathway, or immune modulatory agents. This review will summarize currently available evidence and explain the rationale that supports the combination of immunotherapy and targeted therapy for the treatment of melanoma, and describe how this approach is being extended to patients with other histological types of cancer.

1.1. Immunotherapy

Studies have indicated the association of tumor T cell infiltrates with clinical benefit of immunotherapy in several tumor types(1–8). In addition, these immune infiltrates have been shown to include specific T cell clones that target somatic point mutations (also called neo-antigens (9)), as well as overexpressed cancer-testis antigens(10) or lineage-specific antigens(11–13). Rosenberg and colleagues at the National Cancer Institute (NCI) have been conducting clinical trials using *ex vivo* expanded autologous tumor-infiltrating lymphocytes (TILs) for adoptive cell transfer (14) (15). Thus far, the results have been reproducible and have demonstrated durable and relatively high complete remission (CR) rates(15). The newer generation of ACT, utilizing autologous T cells engineered to express chimeric antigen receptor (CAR) directed against CD19, has been highly successful in acute or chronic lymphoblastic leukemia and non-Hodgkin lymphomas(16, 17). However, less activity has been observed when engineered T cell receptors (TCR) were directed against solid tumor antigens, including melanoma antigen recognized by T cells 1 (MART1) and NY-ESO-1(18, 19).

The development of immune checkpoint inhibitors has been revolutionary in the field of cancer immunotherapy. Blockade of cytotoxic T lymphocyte antigen 4 (CTLA4) (20, 21) and programmed cell death protein 1 (PD-1) have demonstrated durable responses across different tumor types(22–24). In addition, the combination of these two checkpoint inhibitors has resulted in unprecedented high response rates in melanoma (nearly 60%), but has been associated with increased frequency of toxicities(25). Subsequently, pipelines of newer checkpoint inhibitors and other immunomodulatory agents are being developed. Most

recently, FDA has approved intratumoral injection of talimogene laherparepvec (T-VEC), a genetically modified oncolytic virus, for the treatment of unresectable melanoma (26).

The success of these modern immunotherapy strategies has created great excitement in the cancer research field because it offers tumor specific response with durability due to the memory of effector cells. However, frequency of immunotherapy responses are relatively low in most cases, likely due to the tumor escape mechanisms that are different between individual patients and tumor types. Strategies to improve the response rate have been of high interest.

1.2. Targeted therapy

Small molecule inhibitors of driver mutation pathways, such as epidermal growth factor receptor (27) inhibitors for EGFR mutant lung cancer(28) or anaplastic lymphoma kinase (ALK) inhibitors for lung cancer patients who harbor the echinoderm microtubule-associated protein-like 4 (EML4)-ALK translocation(29), have been successfully developed for several cancer subtypes and can induce high response rates in tumors with underlying genetic alterations. Similarly, antibodies of human epidermal growth factor receptor 2 (HER2) have significantly improved survival in women with HER2 amplified breast cancer in both the adjuvant and metastatic settings(30). The identification of a prevalent driver mutation in *BRAF* has also led to the development of selective BRAF inhibitors and MEK inhibitors that shut down the MAPK pathway in melanomas(31–33). The initial response rates to targeted therapies have been high but the long-term effectiveness of these therapies has unfortunately been limited by the development of acquired resistance in the majority of patients (34–39).

2. The potential mechanisms of combined benefits of targeted therapy and immunotherapy

Targeted therapy can not only direct killing of tumor cells, but also have effects on the different components of the immune system, so called “immunesensitization”, suggesting a potentially synergistic benefit of combining targeted therapy and immunotherapy beyond the expected additive effect of two effective treatments (40).

2.1. Direct effects on tumor cells

The direct effects of BRAF and MEK inhibitors are achieved by the induction of cytotoxicity in melanoma cells through inhibition of the MAPK pathway, and subsequent cell death can create a more immunogenic environment in which tumor antigens can be cross-presented to T cells. Prior studies have demonstrated that decreased signaling through the MAPK pathway by BRAF and MEK inhibitors is correlated with increase in melanocyte differentiation antigens in both melanoma cell lines and clinical tumor samples from melanoma patients(41–43). Further, when resistance to BRAF inhibition occurs, it can be associated with loss of tumor antigen expression (41). BRAF inhibition has also been shown to cause the upregulation of major histocompatibility complex (MHC) class I molecules in tumor cells, which improves antigen presentation and recognition (44). In different preclinical models it has been described that both the apoptotic effect of inhibiting BRAF is

being enhanced (45) or blocked (46) by cytokines produced by T cells such as TNF or IFN-γ. Several groups are actively studying to better characterize this interaction. These results have justified the clinical testing of a combinatorial approach between BRAF inhibition and ACT for BRAF-mutant metastatic melanoma.

In glioblastoma where PTEN loss is found in 60–80% of cases (47), a study showed that this loss of PTEN and subsequent up-regulation of the PI3K-AKT pathway resulted in increased constitutive expression of PD-L1 and is associated with immune resistance(48). This study also showed a decrease in the transcription of PD-L1 in cells treated with rapamycin, a mTOR inhibitor. Currently there is a clinical trial testing the efficacy of PI3K/mTOR inhibitors vs anti-PD-1 antibodies (NCT02430363) for patients with glioblastoma; however, testing a combination of these two agents would be of high interest. A similar justification has been extended to EGFR mutant non-small cell lung cancer (NSCLC) (approximately 10% of all NSCLC cases (49)), where this driver mutation has been shown to mediate immune escape through the upregulation of PD-1 and PD-L1(50). Treatment with EGFR inhibitors, on the other hand, has reduced PD-1 and PD-L1 expression (51). Based on this knowledge, clinical trials focusing on the combination of EGFR and PD-1 inhibitors have been initiated (NCT02039674). Both of these situations are examples of constitutive expression of PD-L1 as a result of an oncogenic event as opposed to adaptive immune expression. Blocking these driver mutations might downregulate the expression of PD-L1 at the tumor level. However, it remains unclear the consequence of inhibiting these mutations in tumors that lack infiltrating lymphocytes, which is more common in glioblastoma with PTEN loss and EGFR mutant NSCLC (52).

Epigenetic alterations are common in cancer cells, including global hypomethylation or hypermethylation of CpG islands in promoter regions. This hypermethylation can result in gene silencing of tumor suppressor genes, oncogenes, tumor associated antigens (TAA), antigen presentation machinery (APM) and co-stimulatory signaling (53, 54), leading to immune escape(55). Hypomethylating agents such as 5-azacitidine (AZA) have shown activity in up-regulating tumor associated antigens and increasing expression of HLA-A1 and other components of the antigen presenting machinery (APM) such as Transporter Associated with Antigen Processing 1 (TAP-1) (56) and -2(56–59). Another epigenetic process, histone acetylation, is a reversible mechanism enabling access of chromatin and transcription of genes (60). Histone deacetylase (HDAC), on the other hand, removes acetyl groups and suppresses the gene transcription process. HDAC inhibitors have been shown to restore expression of TAA and cancer-testis antigens such as NY-ESO-1 (61–63), enhance MHC class I and II expression(64, 65), and upregulate PD-L1 expression(66, 67). In preclinical models, *Kim et al* reported tumor eradication in 80% of mice bearing either CT26 colon cancer or 4T1 breast cancer, treated with combination of epigenetic-modulators (5-AZA plus entinostat, a HDAC inhibitor) and checkpoint inhibitors (anti-CTLA4 plus anti-PD1), but not checkpoint inhibitors alone, accomplished by depletion of myeloid derived suppressive cells (MDSC) (68).

2.2. Effects on effector T lymphocytes

Besides inhibiting oncogenic events in tumor cells, targeted therapy exerts important effects on normal cells that reply on the targeted pathway regulation, especially the cytotoxic T cells. For example, the selective inhibitors of *BRAF* V600 mutation could serve as activators of the MAPK pathway in cells with a wild-type BRAF genotype but a strong upstream signal, including keratinocytes or lymphocytes. This effect has been defined as paradoxical activation of the MAPK pathway (69) that results in the immune-sensitization effects of increased T cell function(70–72), and contributes to the skin toxicity of squamous cell carcinoma of BRAF inhibitors. Preclinical studies involving fully immunocompetent mice bearing *BRAF*-mutant melanoma tumors (as well as *CDKN2A* mutation and *BRAF* and *MITF* amplification) have shown that CD8 cells are critical to the benefits of BRAF inhibitors, as depletion of CD8 cells but not CD4 or NK cells in mice, can partially abrogate the anti-tumor effects of BRAF inhibition(70, 73). Furthermore, it was determined that despite the potential detrimental effects to the T effector cell function by directly inhibiting the MAPK pathway in cultured T cells *in vitro* (41, 74), addition of a MEK inhibitor in the *in vivo* setting could be synergistic to combined BRAF inhibition and immunotherapy, with maintained T cell function and a more immune permissive tumor microenvironment, and dampen the unwanted toxicity associated with the paradoxical activation of the MAPK pathway by BRAF inhibitors alone (75, 76). In addition, BRAF and MEK inhibitors can upregulate tumor PD-L1 expression and this has translated into a superior antitumor response when combining with PD-L1 antibodies (75, 76).

The number of TILs also appears to be affected by the inhibition of the MAPK pathway(73, 75, 76). A greater number of TILs has been described in tumor samples from patients treated by BRAF and/or MEK inhibitors, with increased TIL clonality in BRAF inhibitor treated tumors (42, 77, 78). Because increased tumoral or peritumoral infiltration of CD8 lymphocytes with high clonality has been shown to predict responses to PD-1 blockade(79), this clonal expansion towards a more specific repertoire induced by BRAF inhibition, along with the observed increase in PD-1 and PD-L1 soon after BRAF and/or MEK inhibition(42, 80), provide a sound rationale for this combination of targeted therapy and immunotherapy.

The combinatorial benefit of BRAF inhibitors and immunotherapy was demonstrated in another syngeneic melanoma model harboring oncogenic *BRAF* mutation (as well as CDKN2A −/−, *PTEN* −/−) (81), where blocking mutated BRAF increased CD8+ T cell infiltration, and the antitumor effects was enhanced when BRAF inhibitor was combined with PD-1 or PD-L1 inhibitors by enhanced T cell activity and improved survival of the treated mice.

More recently, inhibition of Pi3K/Akt/mTOR pathway was shown to induce the expansion of TILs promoting a memory T cell phenotype (82), which provides a rationale for enhancing the persistence of transferred tumor specific T cells in the adoptive cell transfer (82) immunotherapy approach. Therefore, a phase I trial investigating the persistence of adoptively transferred TILs cultured with an AKT inhibitor in patients with metastatic melanoma is currently ongoing by Steven Rosenberg’s groups at NCI (NCT02489266).

A newer class of drugs targeting focal adhesion kinase (83) has attracted interest as inhibition of FAK is associated with decrease of pluripotent cells that are considered cancer stem/progenitor cells (CSCs). This was initially described in breast cancer where ablation of FAK reduced the pool of CSCs in primary tumors of FAK-targeted mice and impaired their self-renewal and migration in vitro. In addition, CSCs isolated from FAK-targeted mice have compromised tumorigenicity and impaired maintenance *in vivo* (84). Recently FAK inhibitor has been showed to reduce CSCs and delay tumor growth following cisplatin plus pemetrexed treatment in a patient-derived xenograft model of malignant mesothelioma and other tumor types (85, 86). Interestingly, FAK inhibitors have been recently reported to help proliferation of CD8 cytotoxic T cells and inhibit tumor associated macrophages (87), and combination of FAK inhibitor (VS-4718) and anti-PD-1 agent extended survival of mice bearing colon cancer tumors, providing the rationale for translation into clinical trials.

2.3. Effects on the tumor microenvironment

In melanoma, besides the direct effects on cytotoxic T cells, BRAF and MEK inhibition has also demonstrated immunomodulatory effects in the melanoma tumor microenvironment. The work referenced above combining BRAF and MEK inhibition with immunotherapy (75), showed that the triple combination therapy resulted in increased melanosomal antigen and MHC expression and global immune-related gene up-regulation. Single-agent dabrafenib increased tumor-associated macrophages and T regulatory cells (Tregs) in tumors, which decreased with the addition of trametinib. MEK inhibitors have been shown to decrease immunosuppressive cytokines, including IL-1, IL-6, and IL-8, IL-10 as well as decrease angiogenic factors such as vascular endothelial growth factor (VEGF) (88). Inhibition of BRAF reduced expression of IL-1 in cell lines and tumor biopsies, and because the immune inhibitory activity of tumor-associated fibroblasts (89) is enhanced by IL-1, treatment with BRAF inhibition can potentially decrease the number of tumor-associated TAF in the stroma, as suggested by *Khalili et al* (90). When a xenograft model of BRAF-mutated human melanoma cell line transduced with gp100 and H-2D was used to assess melanocyte differentiation antigen-independent enhancement of immune responses by BRAF inhibitor, it was found that administration of vemurafenib significantly increased the tumor infiltration and function of adoptively transferred gp100-specific pmel-1 T cells in vivo (91), primarily mediated by the ability of vemurafenib to inhibit melanoma tumor cell production of VEGF. Analysis of human melanoma biopsies showed down-regulated VEGF before and during BRAF inhibitor treatment.

Imatinib, a selective inhibitor of the c-kit receptor tyrosine kinase that has shown great success in treating chronic myelogenous leukemia (CML) and gastrointestinal stromal tumors (92) was found to be associated with activation of CD8 T cells and reduction of Tregs, through reduction of indoleamine 2,3-dioxygenase (IDO) in the tumor microenvironment (93). Sunitinib, a multi-targeted receptor tyrosine kinase (RTK) inhibitor against platelet-derived growth factor receptors (PDGF-Rs), vascular endothelial growth factor receptors (VEGFRs) as well as c-kit and approved by the FDA for the treatment of renal cell carcinoma (RCC) and imatinib-resistant gastrointestinal stromal tumor (92), has been shown to decrease the amount of immune-suppressive MDSCs and Tregs (94) in the tumor microenvironment.

3. Clinical trials testing the combination of immunotherapy and targeted therapy

Based on the rationale detailed above, the concept of combining targeted therapy and immunotherapy are being tested in clinical trials. The first reported combination therapy with ipilimumab and vemurafenib had been terminated early due to grade 3 hepatotoxicities (73). The etiology of this hepatotoxicity was unclear, but could be related to the paradoxical activation of the MAPK pathway in BRAF wild type cells. However, a separate trial (NCT01767454) involving ipilimumab and dabrafenib did not encounter hepatotoxicity, suggesting a drug-specific process. Interestingly, a second treatment arm that examined the triple combination of dabrafenib, trametinib and ipilimumab had to be discontinued because of two out of seven serious adverse events involving colon perforations (95). Currently at least three clinical trials have been currently ongoing to test the combination of BRAF plus MEK inhibitors with PD1/L1 blockade and have shown encouraging results.

These unexpected toxicities highlighted the complexity of the translational clinical scenario, and a pressing need for judicious evaluation both in the clinic and in a wide-range of potentially clinically predictive animal models to better guide the clinical adaptability of these two promising modalities. For that reason, development of more genetically relevant animal models that can closely resemble the human circumstance across all tumor histologies is warranted. The current tumor models, which are frequently derived from genetically engineered mice (GEM) with constitutively active oncogenic signaling, fail to recapitulate the antigenic complexity derived from skin UV damage in human melanoma. New models are being developed and hopefully they will succeed in better translating the biology behind each tumor-type. On the other hand, the biology and immune response in mouse models does not correlate well with activity in the corresponding human cancer, exemplified by the success of anti-PD1 therapies in human cancers in contrast to the lack of response to these antibodies in majority of tumor models. Therefore, if strong rationale stands, testing judiciously in the clinical setting should be warranted, even wirh lack of activities in the preclinical setting.

Tables 1 to 5 summarize ongoing clinical trials involving the combination of targeted therapy and immune checkpoint blockade. The number of clinical trials has increased exponentially in the last few years, and the involved tumor types have become more diverse and are no longer limited to the traditionally “immunotherapy-sensitive” melanoma or kidney cancers (Table 1). Some studies are assessing increased tumor cell killing and antigen expression/presentation that could enhance T cell activation, whereas other studies are attempting to overcome the immune suppressive environment within the tumors. These tables also included clinical trials exploring the combination of targeted therapy with ACT, majority of which involve combination with TIL therapy conducted by NCI. The rationale behind the combination of targeted therapy and immunotherapy is strongly supported by the above mentioned preclinical data, however, translation into a clinical setting will require carefully selection of the targets in a case by case setting, and optimization of the schedule and sequence of the involved drugs.

Monoclonal antibodies targeting the human epidermal growth factor receptor (HER) family members can enhance dendritic cell mediated T cell priming and antibody dependent cellular cytotoxicity (ADCC), therefore is another potential candidate to combine with PD-1 checkpoint inhibitors (96, 97). Table 2 summarized ongoing clinical trials designed to investigate such combinations involving HER2 inhibitors such as trastuzumab (NCT02318901) and HER1 (27) inhibitor cetuximab (NCT02105636, NCT02252042).

Similar to the melanoma setting, both checkpoint inhibitors (immunotherapy) and anti-angiogenic agents (targeted therapy) are successful in the treatment of renal cell carcinoma, therefore there is great interest in combining these two classes of agents for better disease control and potential synergy in RCC. Preliminary results from trials involving the combination of nivolumab with sunitinib or pazopanib revealed response rates as high as 50%, but increased toxicity, especially hepatotoxicity was observed(98) (NCT02014636, NCT 01472081). Similarly, a phase I trial combining CTLA-4 monoclonal antibody tremelimumab with sunitinib resulted in unexpected renal toxicity(99). Several other trials are subsequently open to evaluate alternative anti-angiogenic drugs (Table 3).

As described earlier, the role of epigenetic modulation to improve the tumor immune microenvironment using HDAC inhibitors is also being investigated. Table 4 summarizes the current clinical trials involving HDAC inhibitors and immune checkpoint blockade. A phase I trial testing priming with azacytidine (a hypomethylation agent) plus entinostat (a HDAC inhibitor) prior to nivolumab in advanced NSCLC, has induced responses in six patients (100). Three of these patients experienced durable responses and two had stable diseases for 9 months (101). Based on these results, a phase II trial (NCT01928576) is similarly designed and currently recruiting. Previous clinical data combining another HDAC inhibitor vorinostat and tamoxifen has provided 19% response rate in hormone therapy resistant breast cancer (102). More recently, the combination of vorinostat, tamoxifen and PD-1 inhibition is being investigated (NCT02395627) with both concurrent and sequential schedules.

Finally, Table 5 summarizes trials that combine drugs that target cancer stem cells (CSC) with immune checkpoint blockade, including a FAK inhibitor defactinib in combination with pembrolizumab and gemcitabine for advanced pancreatic cancer patients (NCT02546531), and a cancer stem cell inhibitor BBI608 combined with ipilimumab or nivolumab or pembrolizumab (at the investigator’s discretion) in advanced solid tumors (NCT02467361).

4. Conclusions

The advance in cancer immunotherapy has resulted in a paradigm shift in the management of patients across several tumor types, including the traditionally non-immunotherapy responsive histologies, with the promise of long-term disease control. The immediate challenge facing the field is how to improve the response towards majority of patients and tumor types. The readily available targeted therapies that are already approved in many tumor types with an association with high response rate for the indicated patient population provide an attractive combination strategy. The potential synergy of targeted therapy and immunotherapy has been shown in both preclinical models and patient derived samples.

However, critical questions have to be answered before translation of this approach into clinical applications. Particular concepts need to be explored and confirmed in relevant animal models and optimized in clinical trials, and toxicity needs to be evaluated. In all of the ongoing trials, tumor biopsies and translational studies need to be incorporated into the study design.

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Highlights

- Targeted therapy has the potential to enhance immunotherapy by inducing immune effects in tumor cells, modulating T cell homing and function, as well as the tumor immune microenvironment.
- The potential benefits of combined targeted therapy and immunotherapy are not limited to tyrosine kinase inhibitors (TKIs) or melanoma, but extend to other molecular targets and tumors histologies.
- Many clinical trials are currently underway, but optimization of the dosing regimen and schedule is needed to confirm benefits and avoid toxicity associated with these combinations.

Table 1

Clinical trials involving the combination of tyrosine kinase inhibitors (TKIs) and immunotherapy.

Immune checkpoint blockade +TKIs			
Clinical trial	**Condition**	**Phase**	**Intervention**
NCT01738139	Advanced tumors	Phase I	Ipilimumab+Imatinib Mesylate
NCT02133742	CCmRC	Phase I	Pembrolizumab+Axitinib
NCT02420912	CLL	Phase II	Nivolumab + Ibrutinib
NCT02011945	CML	Phase I	Nivolumab+Dasatinib
NCT02329847	Hematologic	Phase I/II	Nivolumab + Ibrutinib
NCT02446457	Lymphoma	Phase II	Pembrolizumab+Rituximab
NCT01656642	Melanoma	PhaseI	PD-L1inhibitor+vemurafenib PD-L1inhibitor+vemurafenib+cobimetinib
NCT01659151	Melanoma	Phase II	ACT with TIL+High Dose IL-2+Lymphodepletion+Vemurafenib
NCT01940809	Melanoma	PhaseI	Ipilimumab Ipilimumab+trametinib Nivolumab+ipilimumab Ipilimumab+dabrafenib Ipilimumab+dabrafenib+trametinib Nivolumab+ipilimumab+trametinib Nivolumab+ipilimumab+Dabrafenib Nivolumab+Ipilimumab+Dabrafenib+Trametinib
NCT02027961	Melanoma	PhaseI/II	PD-L1inhibitor+Trametinib PD-L1 inhibitor+ Trametinib+Dabrafenib
NCT02489266	Melanoma	Phase I	Lymphodepletion+AKTi-treated TIL+IL-2
NCT02354690	Melanoma	Phase I/II	Vemurafenib –> TIL+lymphodepletion+IL2
NCT02357732	Melanoma	Phase I	Nivolumab+Dabrafenib Nivolumab+Trametinib Nivolumab+Dabrafenib+Trametinib
NCT02400385	Melanoma	Phase II	Sunitinib+Nivolumab
NCT02130466	Melanoma	Phase I/II	Dabrafenib+Trametinib Pembrolizumab+Dabrafenib Pembrlizumab+Trametinib Pembrolizumab+Dabrafenib+Trametinib
NCT01454102	NSCLC	Phase I	(19 arms at different dose combination) Nivolumab + Gemcitabine + Cisplatin Nivolumab + Pemetrexed + Cisplatin Nivolumab + Paclitaxel + Carboplatin Nivolumab + Bevacizumab maintenance Nivolumab + Erlotinib Nivolumab Nivolumab + Ipilimumab Nivolumab Nivolumab + Ipilimumab
NCT02323126	NSCLC	Phase II	Nivolumab + EGF816 (EGFRinhibitor) Nivolumab + INC280 (cMET inhibitor)
NCT02039674	NSCLC	Phase I/II	Pembrolizumab+Paclitaxel+Carboplatin Pembrolizumab+Paclitaxel+Carboplatin+Bevacizumab Pembrolizumab+Pemetrexed+Carboplatin Pembrolizumab+Ipilimumab Pembro+Erlotinib Pembrolizumab+Gefitinib Carboplatin+Pemetrexed+/− Pembrolizumab) Pembrolizumab + ipilimumab
NCT01998126	NSCLC	Phase I	Ipilimumab+Erlotinib Ipilimumab+Crizotinib

Immune checkpoint blockade +TKIs			
Clinical trial	**Condition**	**Phase**	**Intervention**
NCT02364609	NSCLC	Phase I	Pembrolizumab+Afatinib
NCT02448303	NSCLC	Phase II	Pembrolizumab Pembrolizumab+ACP-196 (BTKi)
NCT02511184	NSCLC	Phase I	Pembrolizumab+Crizotinib
NCT01767454	Solid tumors	Phase I	Ipilimumab+Dabrafenib Ipilimumab+Dabrafenib+Trametinib
NCT02423343	Solid tumors	Phase I/II	Nivolumab+Galunisertib

TKI = tyrosine kinase inhibitors; CLL = chronic lymphocytic leukemia; NSCLC = Non-small cell lung cancer; CCmRC = Clear cell metastatic renal cancer; CML = chronic myelogenous leukemia; IL-2 – interleukin-2; TIL – tumor infiltrating lymphocytes;

Table 2

Clinical trials involving the combination of target-specific monoclonal antibodies and immunotherapy.

Immune Checkpoint blockade + Monoclonal antibodies			
Clinical trial	**Condition**	**Phase**	**Intervention**
NCT00182650	HL	Phase I	IL-2+rituximab+lymphodepletion+therapeutic autologous lymphocytes
NCT02318901	Solid tumors	Phase I/II	Pembrolizumab+Trastuzumab Pembrolizumab+T-DM1 Pembrolizumab+Cetuximab

SSC = Squamous cell carcinoma; T-DM1 – trastuzumab-DM1; IL-2 – interleukine-2

Table 3

Clinical trials involving the combination of anti-angiogenic agents and immunotherapy.

Immune Checkpoint blockade + anti-angiogenic agents			
Clinical trial	**Condition**	**Phase**	**Intervention**
NCT02348008	CCmRC	Phase I/II	Pembrolizumab+Bevacizumab
NCT02014636	CCmRC	Phase I	Pazopanib Pembrolizumab Pembrolizumab+Pazopanib
NCT01472081	CCmRC	Phase I	Nivolumab+Ipilimumab Nivolumab+Pazopanib Nivolumab+ Sunitinib
NCT02337491	Glioblastoma	Phase II	Pembrolizumab Pembrolizumab+Bevecizumab
NCT02501096	Solid tumors	Phase I/II	Pembrolizumab+Lenvatinib

CCmRC = clear cell metastatic renal cancer;

Table 4

Clinical trials involving the combination with epigenetic modulators and immunotherapy.

Immune Checkpoint blockade + Epigenetic modulator			
Clinical trial	**Condition**	**Phase**	**Intervention**
NCT02395627	Breast	Phase II	Pembrolizumab+Tamoxifen+Vorinostat
NCT02538510	Head&neck	Phase I/II	Pembrolizumab+Vorinostat
NCT02437136	NSCLC/Melanoma	Phase I/II	Pembrolizumab+Entinostat
NCT01928576	NSCLC	Phase II	Azacitidine –> Nivolumab Azacitidine+Entinostat –> Nivolumab

NSCLC = non-small cell lung cancer;

Table 5

Clinical trials involving the combination of CSC inhibitors and immunotherapy.

Immune Checkpoint blockade + CSC inhibitor or others			
Clinical trial	**Condition**	**Phase**	**Intervention**
NCT02467361	Solid tumors	Phase I/II	Ipilimumab+BBI608 Nivolumab+BBI608 Pembrolizumab+BBI608
NCT02546531	Solid tumors	Phase I	Pembrolizumab+Defactinib+Gemcitabine

CSC = Cancer stem cell;