To: Cancer Therapeutic Evaluation Program

From: Jacqueline S. Garcia, MD

Date: June 30, 2020

Re: Response to Request for Amendment for Protocol #10026: "A Phase 1 Study of Ipilimumab in Combination with Decitabine in Relapsed or Refractory Myelodysplastic Syndrome/Acute Myeloid Leukemia"

I. Request for Amendment

#	Section	Comments
1.	Participating Organizations	Changes to Participating LAOs: Deleted: GA031 Northside Hospital LAO-OH007 / Ohio State University Comprehensive Cancer Center LAO RESTRICTED TO: OH029 Case Western Reserve University **PI Response**: We have made the requested changes.

NCI Protocol #: *10026*

Local Protocol #: *17-718*

ClinicalTrials.gov Identifier: *NCT02890329*

TITLE: A Phase 1 Study of Ipilimumab in Combination with Decitabine in Relapsed or Refractory Myelodysplastic Syndrome/Acute Myeloid Leukemia

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Participating Organizations

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	RESTRICTED TO:
CA043	City of Hope Comprehensive Cancer Center
LAO-CT018	**Yale University Cancer Center LAO**
	RESTRICTED TO:
CA249	University of California San Diego Moores Cancer Center
LAO-MD017 / JHU Sidney Kimmel Comprehensive Cancer Center LAO	
	RESTRICTED TO:
VA009	University of Virginia Cancer Center

**Disclaimer: Sites require approval from overall PI before opening the study.*

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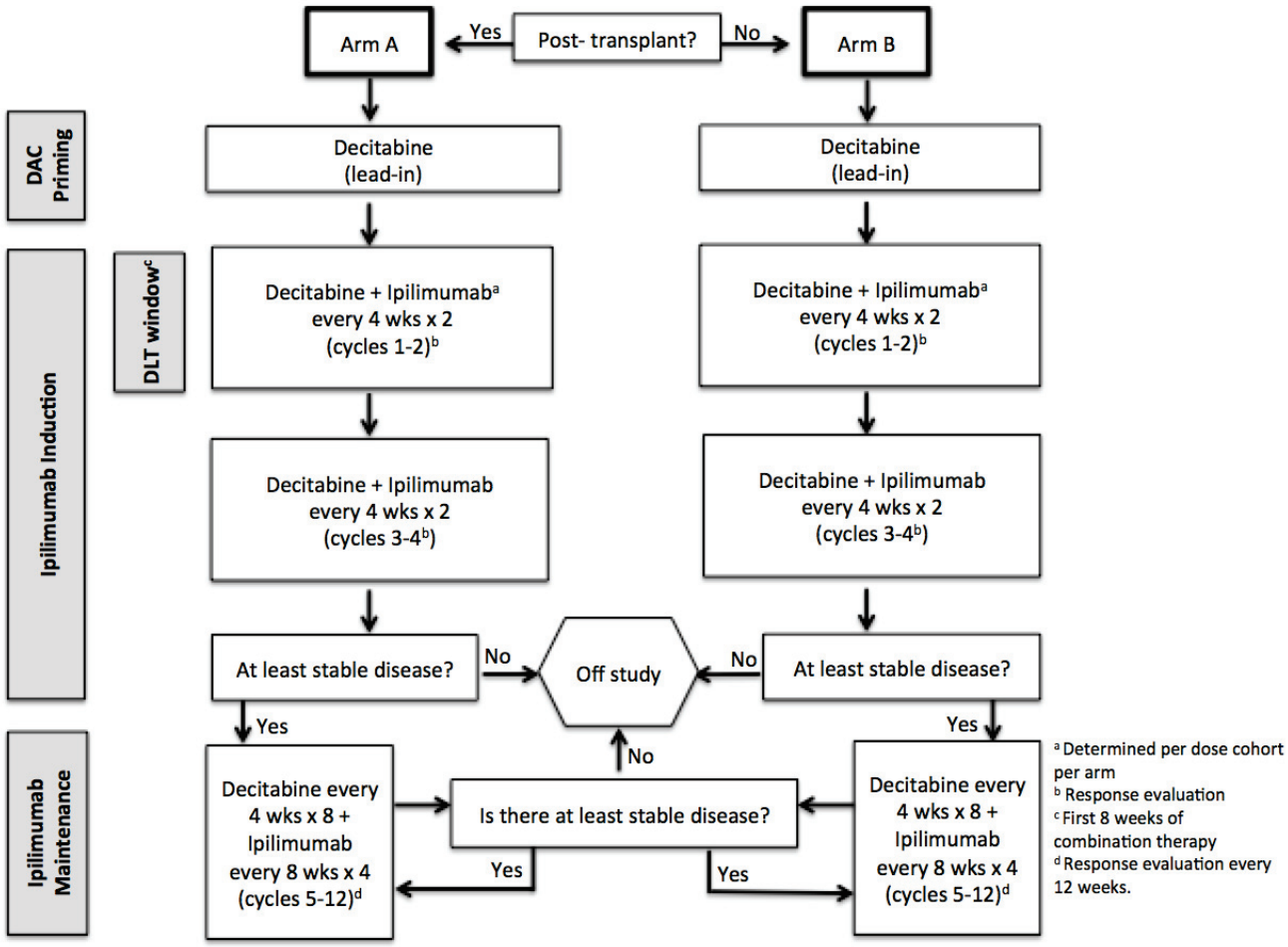
NCI-Supplied Agent: Ipilimumab (NSC 732442)
Other Agent: Decitabine (NSC 127716) - Commercial

IND #:
IND Sponsor: DCTD, NCI

Protocol Type / Version # / Version Date: Original/ Version #21 / 6/30/2020

SCHEMA



Dose Escalation Schedule		
Dose Level	**Dose**	
	Ipilimumab (mg/kg)	**Decitabine (mg/m^2)**
Level -1	1	20
Level 0 (starting dose)	3	20
Level 1	5	20
Level 2	10	20

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1. OBJECTIVES

A recognized feature of cancer is escape from immune surveillance. Therefore augmenting tumor immunity is a promising treatment approach. Immune checkpoint blockade with drugs such as ipilimumab (anti-cytotoxic T-lymphocyte antigen-4 (CTLA-4) therapy) [1] and nivolumab [2] or pembrolizumab (anti-PD-1 therapies) [3] have been successful in providing marked prolonged survival benefits among patients with metastatic melanoma. The role of CTLA-4 blockade has been preliminarily explored in hematologic malignancies in the post-transplant setting with demonstration of safety and activity primarily in patients with acute myeloid leukemia (AML) with low disease burden [4, 5].

The primary objective of this phase 1 study is to identify an MTD for ipilimumab in combination with standard dose decitabine therapy in patients with relapsed or refractory myelodysplastic syndrome (MDS)/AML. If there is demonstration of safety, tolerability and preliminary efficacy, further testing of ipilimumab in combination with decitabine will be explored in a future phase 2 trial.

This is an open-label phase 1 study using a 3+3 design with planned dose escalation of ipilimumab in combination with standard dose decitabine in patients with relapsed or refractory MDS/AML. The study will include three treatment phases, including epigenetic priming (lead in) with decitabine monotherapy x 1 cycle), induction (cycles 1-4 with combination decitabine and ipilimumab), and maintenance (cycles 5-12 with combination decitabine and ipilimumab).

1.1 Primary Objectives

1.1.1 To determine the maximum tolerated dose (MTD) or recommended phase 2 dose (RP2D) of combination decitabine and ipilimumab for relapsed or refractory MDS or relapsed or refractory AML in patients who are post allogeneic hematopoietic stem cell transplant (allo-HCT).

1.1.2 To determine the MTD or RP2D of combination decitabine and ipilimumab for relapsed or refractory MDS or relapsed or refractory AML in patients who are transplant naive.

1.2 Secondary Objectives

1.2.1 To observe and record anti-tumor activity. Although the clinical benefit of this novel combination has not yet been established, the intent of offering this treatment is to provide a possible therapeutic benefit, and thus the patient will be carefully monitored for tumor response and symptom relief in addition to safety and tolerability.

1.2.2 To determine the overall response rate (ORR) including complete remission (CR) and complete remission with incomplete count recovery (CRi) for AML following 2003 IWG response criteria [6].
1.2.3 To determine the ORR including CR, partial remission, marrow CR, hematologic improvement for MDS using 2006 IWG criteria [7].
1.2.4 To determine the overall survival and progression free survival at 1 year.
1.2.5 To determine the duration of remission.
1.2.6 To capture the incidence and severity of acute graft-versus-host disease (GVHD) in the post allo-HCT cohort.
1.2.7 To capture the incidence and severity of chronic graft-versus-host disease (GVHD) in the post allo-HCT Cohort

1.3 Exploratory Objectives

1.3.1 To measure the absolute lymphocyte count (ALC) prior to treatment and during treatment. We will compare changes in ALC between responders and non-responders given evidence that ALC after two cycles may be a predictor of clinical response in solid tumors.
1.3.2 To evaluate the genome for evidence of clonal evolution among longitudinal samples (prior to treatment, during treatment, and at relapse if relevant) from individual patients. We will compare leukemic genotypic patterns among responders and non-responders.
1.3.3 To evaluate the histopathologic findings of immune response using immunohistochemistry.
1.3.4 To determine the immune response in the AML tumor microenvironment by using flow cytometry and single cell mass cytometry to evaluate T cell subsets. We will evaluate cytokine and chemokine panels. We will further evaluate the gene expression patterns of immune infiltrates. We will evaluate for alterations in antigen presenting cells in response to decitabine and combination therapy. We will evaluate for resistance to immune response due to STAT3 activation.

2. BACKGROUND

2.1 Relapsed/Refractory MDS/AML

High-risk MDS and outcomes
MDS is predominantly a disease of the elderly. The International Prognostic Scoring System is a tool used to predict the likely survival of MDS patients at the time of diagnosis [8, 9]. Patients with higher-risk MDS account for 29% of the patient population, including 22% with intermediate-2 MDS and 7% with high-risk MDS [9]. In fact, for patients with intermediate-2 and high-risk MDS, the median survival without therapy was predicted to be 1.1 years and 0.4 years, respectively. Thus, for higher-risk MDS patients, the primary goals for treatment focus on extending overall survival and altering the natural course of disease by delaying time to leukemic transformation. First line agents to treat MDS include hypomethylating agents such as azacitidine and decitabine. These DNA methyltransferase inhibitors promote the synthesis of DNA that otherwise were silenced by methylation marks. Results from a multi-center study

demonstrated that treatment with 5 consecutive days of decitabine at 20 mg/m2 intravenously (IV) among 99 patients resulted in an improvement in approximately half the patients on study, including 17 patients with complete remission (CR), 15 patients with marrow CR, and 18 patient with hematologic improvement [10]. Among patients who benefited from treatment, responses were as early as the end of cycle 2.

AML and outcomes

AML is a disease of the elderly with a median age of diagnosis of 67 years (SEER 2008-2012)**.** Chemotherapy resistance and disease relapse are the primary challenges affecting the majority of patients in the treatment of acute leukemia. For patients less than 60 years of age, traditional induction chemotherapy with combination anthracycline and cytarabine results in CR rates of 50-70% [11]. Despite achieving remission, a significant number of younger patients with AML with poor risk features at diagnosis will be refractory to induction therapy or ultimately relapse after initial response to induction chemotherapy. Poor risk features include antecedent hematological disorder, adverse cytogenetic features, and adverse molecular mutations [12]. Disease relapse remains the primary cause of treatment failure. Elderly patients with AML have a poor prognosis due to increased disease resistance to standard intensive chemotherapy regimens and relatively decreased tolerance to these agents [13]. There is no identified optimal treatment strategy for elderly patients who are not candidates for allo-HCT, which remains the only curative option in this setting but is actually applied to only a small number of patients. Less intensive and therefore better-tolerated regimens include low-dose cytarabine and hypomethylating agents. Azacitidine is approved for the treatment of MDS and for patients with AML with low blast count (20-30% bone marrow blasts). Although approved in the United States for the treatment of MDS including patients with 20-30% bone marrow blasts and in Europe for the treatment of AML, decitabine continues to be used off-label given its anti-leukemic activity and tolerability. In a multicenter phase 2 study, elderly patients with untreated AML received decitabine 20 mg/m^2 intravenously for 5 consecutive days every 28 days [14]. Results from this study showed a CR rate of 24%, a median OS of 7.7 months and a 30-day mortality rate of 7% [14]. A large, randomized phase III study for patients 65 years and older with untreated AML comparing decitabine to supportive care and cytarabine demonstrated a CR plus CRp rate of 17.8% versus 7.8%, respectively [15]. Hence, hypomethylating agents such as decitabine are a reasonable anti-leukemic agent given its tolerability but its anti-leukemic activity and survival benefits are limited as a single agent. The development of novel and effective non-intensive therapeutic strategies for elderly AML patients remain an unmet need.

Relapsed MDS/AML after transplant

Patients with relapsed or refractory AML have a dismal prognosis. Allo-HCT is an established and potentially curative treatment option for patients with AML and high-risk MDS. Transplant or treatment related mortality has decreased significantly with the implementation of reduced-intensity conditioning regimens, enabling allo-HCT to become an option for patients with older age and with co-morbidities [16]. Despite these recent improvements in conditioning regimens and in supportive care, 30-40% of patients with AML [17] and 15-35% of patients MDS [18, 19] still relapse after allo-HCT. Disease recurrence after allo-HCT is a devastating event with a median time to relapse of approximately 4 months. These patients have a 5-year OS of about 5%. Available treatment options with variable outcomes include clinical trials with novel therapies, withdrawal of immunosuppressive therapy, donor lymphocyte infusion (DLI), chemotherapy,

hypomethylating agents, and a second allo-HCT.

Role of immunotherapies in MDS/AML
The role of immunotherapy, specifically checkpoint blockade, requires further exploration in AML. The graft versus tumor (or graft versus leukemia) effect observed in patients who undergo allo-HCT [20, 21] is evidence that modulation of the immune system can have an anti-leukemic effect. Tumors can avoid immune surveillance by stimulating immune inhibitory receptors that negatively regulate T-cell activation. CTLA-4 is expressed on both activated T cells and regulatory T cells and binds to ligands B7.1 (CD80) and B7.2 (CD86) on antigen presenting cells (APC) [22]. It functions as a negative regulator of T cell activation by triggering intracellular pathways that suppress proliferative and anti-apoptotic signals on effector T cells, enhancing activity of CD4+ regulatory T cells, and down-regulating T-helper cell activity [22]. Blockade of this immune inhibitory checkpoint provides a potential method to boost anti-leukemic immunity.

2.2 CTEP IND Agent: Ipilimumab

2.2.1 Ipilimumab

For complete information, please refer to the most recent Investigator's Brochure.

Ipilimumab (MDX-010, MDX-CTLA4, BMS-734016) is being developed by CTEP as an anticancer agent in collaboration with Bristol-Myers-Squibb (BMS). On March 25, 2011, the FDA approved ipilimumab injection (YERVOY, BMS) for the treatment of unresectable or metastatic melanoma. Ipilimumab is a human $IgG_1\kappa$ monoclonal antibody (mAb); it is specific for human cytotoxic T lymphocyte-associated antigen-4 (CTLA-4, CD152) expressed on activated T cells. Ipilimumab is now produced and formulated from transfected Chinese hamster ovary (CHO) cells.

CTLA-4 is a negative regulator of T-cell responses following T-cell stimulation [23, 24]. CTLA-4 knockout mice suffer from a fatal lymphoproliferative disorder, supporting the idea that CTLA-4 functions as a negative regulator of T-cell responses *in vivo* [25-27]. Disrupting CTLA-4 interaction with its ligands B7-1 (CD80) and B7-2 (CD86), which are expressed on antigen-presenting cells (APCs), with ipilimumab, augments immune responses (Investigator Brochure, 2011). *In vivo* blockade of CTLA-4, utilizing anti-CTLA-4 mAb, induced regression of established tumors and enhanced antitumor immune responses in several murine tumor models. Blockade of CTLA-4-mediated signals is effective in inducing rejection of immunogenic cancers in mice. Moreover, when anti-CTLA-4 mAb is used in conjunction with granulocyte macrophage-colony stimulating factor (GM-CSF)-secreting tumor vaccines, poorly immunogenic cancers in mice are rejected. These findings suggest that CTLA-4 blockade, alone or in combination with antigenic stimulation and other immune modulating agents can induce a potent antitumor response.

Pharmacology of Ipilimumab
In vitro studies were performed with ipilimumab to demonstrate that it is specific for CTLA-4, actively inhibits CTLA-4 interactions with B7.1 and B7.2, does not show any cross-reactivity

with human B7.1 or B7.2 negative cell lines, and stains the appropriate cells without non-specific cross-reactivity in normal human tissues. Ipilimumab does cross-react with CTLA-4 in non-human primates including cynomolgus monkeys. Blockade of CTLA-4/B7 interactions enhanced T-cell responses to CD3 / CD28, peptide antigens, or superantigens in mice [28-30]. CTLA-4 knockout mice appear to have spontaneously activated T cells evident at approximately 1 week after birth, followed by rampant lymphoproliferation and lymphadenopathy. These mice die at approximately 3 weeks of age, either as a result of polyclonal T-cell expansion and tissue destruction or as a result of toxic shock resulting from lymphokine production. Genetically engineered mice heterozygous for CTLA-4 (CTLA-4+/-), appeared healthy and gave birth to healthy CTLA-4+/- heterozygous offspring. Mated CTLA-4+/- heterozygous mice also produced offspring deficient in CTLA-4 (homozygous negative, CTLA-4-/-). Since thymocyte differentiation and selection proceed normally in CTLA-4-deficient mice, the rampant T-cell expansion that occurs in the mice indicates that CTLA-4 plays a critical role in down-regulating post- thymic T-cell responses in the periphery following stimulation of naïve, memory, and effector T cells [30].

Pharmacodynamic Effects
In clinical studies, ipilimumab increased absolute lymphocytes counts (ALC) in peripheral blood (Investigator Brochure, 2011). However, CD4+/CD8+ ratio did not appear to be affected. Across three phase 2 studies in 463 subjects with advanced melanoma, ipilimumab increased ALC in a dose-dependent manner, with the largest increase observed at 10 mg/kg dose. ALC continued to increase over time during the induction treatment at least until week 12 at the 3 mg/kg and 10 mg/kg dose, but not at the 0.3 mg/kg dose. The slope of ALC increase also suggested the 10 mg/kg dose is more biologically active than the 3.0 mg/kg or 0.3 mg/kg dose

Mechanism of Action
The proposed mechanism of action for ipilimumab is T-cell potentiating through interference of the interaction of CTLA-4 with B7 (CD80 or CD86) molecules on APCs, with subsequent blockade of the inhibitory function of CTLA-4 (Investigator Brochure, 2011). Ipilimumab impacts tumor cells indirectly, and measurable clinical effects emerge after the immunological effects. Tumor infiltration with lymphocytes and the associated inflammation is likely the cornerstone of the effect of ipilimumab and can manifest in various patterns of clinical activity leading to tumor control. These immunologic responses may take time to develop and so tumor responses may be delayed and tumor progression may occur during the initial period followed by responses. In some cases, tumor response based on tumor infiltration with immune cells may be preceded by an apparent increase in initial tumor volume and/or the appearance of new lesions, which may be taken for tumor progression on radiological evaluations. Delayed responses following increasing tumor size or appearance of new lesions have been seen in approximately 10-20% of patients with metastatic melanoma. For patients who are not experiencing rapid clinical deterioration, allowing sufficient time to observe responses including disease stabilization or confirmation of progression is recommended; as discussed in the section “Overall Risk/Benefit Assessment” may allow better assessment of clinical activity and avoid unnecessarily initiating additional therapies in subjects who might be benefitting from treatment. Immune-related (ir) response criteria were developed based on these observations in patients with melanoma to systematically categorize novel patterns of clinical activity and are currently being prospectively evaluated in clinical studies.

Pharmacokinetics
The pharmacokinetics of ipilimumab was studied in 785 patients with advanced melanoma who received induction doses ranging from 0.3 to 10 mg/kg administered once every 3 weeks for 4 doses. Cmax, Cmin and AUC of ipilimumab were found to be dose proportional within the dose range examined. Upon repeated dosing of ipilimumab administered every 3 weeks, clearance was found to be time-invariant, and minimal systemic accumulation was observed as evident by an accumulation index 1.5 fold or less. Ipilimumab steady-state was reached by the third dose. Based on population pharmacokinetic analysis, the following mean (percent coefficient of variation) parameters of ipilimumab were obtained: terminal half-life of 15.4 days (34.4%); systemic clearance of 16.8 ml/h (38.1%); and volume of distribution at steady-state of 7.47 l (10.1%). The mean (percent coefficient of variation) ipilimumab Cmin achieved at steady-state with a 3 mg/kg induction regimen was 19.4 μg/ml (74.6%). Ipilimumab clearance increased with increasing body weight and with increasing LDH at baseline; however, no dose adjustment is required for elevated LDH or body weight after administration on a mg/kg basis. Clearance was not affected by age (range 23-88 years), gender, concomitant use of budesonide or dacarbazine, performance status, HLA-A2*0201 status, mild hepatic impairment, renal impairment, immunogenicity, and previous anticancer therapy. The effect of race was not examined as there was insufficient data in non-Caucasian ethnic groups. No controlled studies have been conducted to evaluate the pharmacokinetics of ipilimumab in the paediatric population or in patients with hepatic or renal impairment. Based on an exposure-response analysis in 497 patients with advanced melanoma, OS was independent of prior systemic anti-cancer therapy and increased with higher ipilimumab Cminss plasma concentrations.

Renal impairment
In the population pharmacokinetic analysis of data from clinical studies in patients with metastatic melanoma, pre-existing mild and moderate renal impairment did not influence the clearance of ipilimumab. Clinical and pharmacokinetic data with pre-existing severe renal impairment are limited; the potential need for dose adjustment cannot be determined

Hepatic impairment
In the population pharmacokinetic analysis of data from clinical studies in patients with metastatic melanoma, pre-existing mild hepatic impairment did not influence the clearance of ipilimumab. Clinical and pharmacokinetic data with pre-existing moderate hepatic impairment are limited; the potential need for dose adjustment cannot be determined. No patients with pre-existing severe hepatic impairment were identified in clinical studies.

Clinical Pharmacokinetics of Ipilimumab Monotherapy
The PK of ipilimumab has been extensively studied in subjects with melanoma, at the 3 and 10-mg/kg doses administered as a 1.5-hour IV infusion. The PK of ipilimumab was characterized by population PK (PPK) analysis and determined to be linear and time invariant in the dose range of 0.3 to 10 mg/kg. The mean CL (+/-SD) value after IV administration of 10 mg/kg was 18.3 ± 5.88 mL/h, and the mean steady-state volume of distribution (Vss) [+/-SSD] value was 5.75 +/-S1.69 L. The CL and Vss from Studies CA184007 and CA184008 were similar to those reported from Study MDX010-15.

Population Pharmacokinetics
The PPK of ipilimumab was studied in 785 subjects (3200 serum concentrations) with advanced melanoma in 4 Phase 2 studies (CA184004, CA184007, CA184008, and CA184022), 1 Phase 3 study (CA184024), and 1 Phase 1 study (CA184078). The PPK analysis demonstrated that the PK of ipilimumab is linear, the exposures are dose proportional across the tested dose range of 0.3 to 10 mg/kg, and the model parameters are time-invariant, similar to that determined by noncompartmental analyses. Upon repeated dosing of ipilimumab, administered q3w, minimal systemic accumulation was observed by an accumulation index of 1.5-fold or less, and ipilimumab steady-state concentrations were achieved by the third dose. The ipilimumab CL of 16.8 mL/h from PPK analysis is consistent with that determined by noncompartmental PK analysis. The terminal T-HALF and Vss of ipilimumab calculated from the model were 15.4 days and 7.47 L, respectively, which are consistent with that determined by noncompartmental analysis. Volume of central compartment (Vc) and peripheral compartment were found to be 4.35 and 3.28 L, respectively, suggesting that ipilimumab first distributes into plasma volume and, subsequently, into extracellular fluid space. CL of ipilimumab and Vc were found to increase with increase in BW. However, there was no significant increase in exposure with increase in BW when dosed on a milligram/kilogram basis, supporting dosing of ipilimumab based on a weight normalized regimen. The PK of ipilimumab is not affected by age, gender, race, and immunogenicity (anti-drug antibody [ADA] status); concomitant use of chemotherapy; prior therapy; BW; performance status; or tumor type. Other covariates had effects that were either not statistically significant or were of minimal clinical relevance.

Nonclinical Toxicology

Please note relevant toxicity for single agent ipilimumab has been almost completely derived from clinical studies.

In a study using cynomolgus macaques, anti-melanocyte responses were observed in animals given up to four doses of 10 mg/kg ipilimumab after receiving a melanoma cell vaccine [31]. Depigmentation has been observed in other nonclinical immunotherapy studies that involve treatment with melanoma peptides [32-37]. The symptoms in animals appear to resemble vitiligo observed in clinical immunotherapy trials of melanoma patients and may be an unavoidable consequence of treatment [38].

Additional repeat-dose toxicity studies conducted using cynomolgus macaques demonstrated that the IV administration of ≤30 mg/kg every 3 days for three doses, 10 mg/kg weekly for 1 month, 1 mg/kg weekly for 10 weeks, or 10 mg/kg monthly for 6 months was generally well tolerated, without significant clinical, immunotoxicological, or histopathological findings (Investigator Brochure, 2011). However, when ipilimumab was administered in combination with another immunomodulatory antibody (BMS-663513, a fully human anti-CD137 mAb) and simian immunodeficiency virus (SIV) DNA, two immune-related adverse events (irAEs) were observed: severe colitis requiring euthanasia in one monkey and reversible dermatitis/rash in the inguinal area and peripheral lymphadenopathy in another monkey.

Complete information on the pre-clinical toxicology studies can be found in the Ipilimumab Investigator Brochure (IB). Non-clinical toxicity assessments included *in vitro* cynomolgus monkeys alone and in the presence of vaccines. Low to moderate ADCC activity was noted at concentrations up to 50 mcg/mL. These data are consistent with the requirement of high levels of antigen expression on the surface of target cells for efficient ADCC or CDCC. No mortality or signs of toxicity were observed in three independent 14-day intravenous (IV) toxicology studies in cynomolgus monkeys at multiple doses up to 30 mg/kg/dose. Furthermore, ipilimumab was evaluated in sub-chronic and chronic toxicology studies in cynomolgus monkeys with and without Hepatitis B (HepB) Vaccine and Melanoma Vaccine. Ipilimumab was well tolerated alone or in combination in all studies. There were no significant changes in clinical signs, body weight values, clinical pathology values or T-cell activation markers. In addition, there were no significant histopathology changes in the stomach or colon.

Clinical Development of Ipilimumab

Company-Sponsored Studies

BMS and Medarex (acquired by BMS in Sep-2009) have co-sponsored an extensive clinical development program for ipilimumab, encompassing more than 19,500 subjects (total number of subjects enrolled in ipilimumab studies) in several cancer types in completed and ongoing studies, including a compassionate use. The focus of the clinical program is in melanoma, prostate cancer, and lung cancer, with advanced melanoma being the most comprehensively studied indication. Ipilimumab is being investigated both as monotherapy and in combination with other modalities such as chemotherapy, radiation therapy, and other immunotherapies. Phase 3 programs are ongoing in melanoma, prostate cancer, and lung cancer. In melanoma, 2 completed Phase 3 studies (MDX010-20 and CA184024) have demonstrated a clinically meaningful and statistically significant survival benefit in pretreated advanced melanoma and previously untreated advanced melanoma with a manageable safety profile, respectively. An ongoing Phase 3 study (CA184029) in melanoma is investigating ipilimumab as adjuvant monotherapy for high-risk Stage III melanoma. In addition, a Phase 3 study (CA184169) comparing the safety and efficacy of 3 versus 10 mg/kg ipilimumab monotherapy in pretreated or treatment-naïve subjects with unresectable or metastatic melanoma is ongoing

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treatment-naïve subjects with unresectable or metastatic melanoma is ongoing

The completed Phase 3 study (CA184043) evaluated ipilimumab in subjects with mCRPC who had progressed during or following treatment with docetaxel. Eligible subjects were randomized to a single dose of bone-directed RT, followed by either ipilimumab 10 mg/kg or placebo (799 randomized: 399 ipilimumab and 400 placebo). This study did not meet its primary endpoint of OS. The HR of 0.85 (95% CI: 0.72, 1.00) for survival favored ipilimumab but did not reach statistical significance with a P value of 0.053. Planned sensitivity analyses favored ipilimumab, where the greatest benefit appeared to be in subgroups defined by good prognostic features and low burden of disease. Additional evidence of ipilimumab activity observed in the study included a reduced risk of disease progression relative to placebo (HR = 0.70), superior clinical outcomes compared to placebo in tumor regression, and declines in PSA. The safety profile in this study was consistent with the previously defined AE profile at the same dose.

A second Phase 3 study (CA184095) evaluated ipilimumab 10 mg/kg versus placebo in subjects with asymptomatic or minimally symptomatic, chemotherapy-naïve mCRPC with no visceral metastases.

Activity was also observed in a large Phase 2 study in lung cancer (NSCLC and SCLC; CA184041) in combination with chemotherapy. Two ongoing Phase 3 studies are evaluating ipilimumab in combination with chemotherapy in squamous NSCLC (CA184104) and SCLC (CA184156). In Study CA184104, the last patient, last visit was achieved in June-2015, and database lock occurred on 01-Sep-2015. No final data are currently available, but preliminary data indicate that no new safety concerns were identified in the course of standard clinical safety monitoring of the study. In Study CA184156, preliminary data indicate the primary endpoint of prolonging survival was not achieved, but no new safety signals were identified.

While the types of safety events observed in subjects receiving ipilimumab do not appear to change, even in combination with other anti-cancer agents, the proportion of subjects experiencing 1 type or another irAE may be impacted by the choice of combination partner. Skin and GI irAEs predominate in monotherapy studies. In combination with DTIC (melanoma), the incidence of skin and GI irAEs was lower than expected, and the incidence of hepatic irAEs was higher. In combination with paclitaxel and carboplatin (NSCLC), the incidence of all types of irAEs appeared to be numerically lower compared to the incidence observed for ipilimumab monotherapy in the Phase 2 program. In a Phase 1 study (CA184161), the concomitant administration of vemurafenib and ipilimumab in subjects with BRAF-mutated metastatic melanoma resulted in asymptomatic and reversible increases in aspartate aminotransferase (AST) and alanine aminotransferase (ALT), exceeding the incidence to be expected when either agent is administered as a single agent therapy, leading to discontinuation of this treatment. In a Phase 2 study (CA184240), sequential treatment with vemurafenib followed by 10 mg/kg ipilimumab in subjects with BRAF-mutated metastatic melanoma was tolerable with a manageable safety profile. No significant signals of hepatobiliary toxicity were reported. The benefit/risk of this sequence needs to be evaluated further based on individual subject characteristics and new treatment options.

Ipilimumab is also being evaluated in clinical studies conducted independently by the Cancer Therapy Evaluation Program of the US NCI, as well as in several additional externally-sponsored studies.

Other Clinical Studies with Ipilimumab

Renal cell carcinoma (RCC)
Yang and colleagues presented data on a phase 2 study of ipilimumab conducted in patients with metastatic RCC [39]. Sequential cohorts received either 3 mg/kg followed by 1 mg/kg or all doses at 3 mg/kg q3w. One of 21 patients receiving the lower dose had a PR. Five of 40 patients at the higher dose had PRs (95% CI, cohort response rate 4 to 27%) and responses were seen in patients who had previously not responded to IL-2. Thirty-three percent of patients experienced grade 3 or 4 irAEs. There was a highly significant association between autoimmune events and tumor regression (response rate = 30% with AE, 0% without AE). The authors concluded that CTLA-4 blockade with ipilimumab induced cancer regression in some patients with metastatic clear cell renal cancer, even if they had not responded to other immunotherapies.

Melanoma (ipilimumab plus bevacizumab)
At the 2011 ASCO meeting Hodi and colleagues presented results on 21 evaluable patients (22 patients enrolled) with unresectable stage III or stage IV melanoma treated with the combination of 10 mg/kg ipilimumab and 7.5 mg/kg bevacizumab on a phase 1 study [40]. AEs included giant cell arteritis (1), hypophysitis (3), thyroiditis (4), grade 3-4 hepatitis (2), bilateral uveitis (2), and grade 2 colitis (2); 5 patients required systemic steroids and stopped treatment. All toxicities were resolved. Eight PRs and 6 SDs were observed. All responses were durable (>6 months). Post-treatment biopsies in 12 patients revealed activated vessel endothelium with extensive T-cell trafficking non-productive central angiogenesis, and peripheral blood monitoring revealed a marked increase in CD4/CCR7/CD45RO central memory cells in the majority of patients, not seen with ipilimumab alone. The authors concluded that the combination of ipilimumab with bevacizumab can be safely administered with clinical activity and correlatives suggesting synergistic effects.

Bladder cancer
Carthon and colleaugues reported immunodulatory effects following a brief exposure of anti-CTLA-4 in patients with urothelial carcinoma of the bladder requiring surgery (BMS study CA184027) [41]. 12 patients were enrolled (6 patients received 3 mg/kg/dose of ipilimumab and another 6 patients received 10 mg/kg/dose for two doses prior to surgery). The treatment was found to be tolerable in the cohort of patients with 11 of 12 patients receiving both doses of antibody. Grade 1-2 diarrhea and rash were the most common drug-related AEs. The only noted grade 3 irAEs were ischemic papillopathy and diarrhea, which were both responsive to treatment with steroids.

Liakou and colleagues found that CD4 T cells from peripheral blood and tumor tissues of all bladder cancer patients treated with anti-CTLA-4 antibody had markedly increased expression of inducible costimulator (ICOS) [42]. These $CD4^{+}ICOS^{hi}$ T cells produced interferon-gamma (IFN-γ) and could recognize the tumor antigen NY-ESO-1. Increase in $CD4^{+}ICOS^{hi}$ cells led to an increase in the ratio of effector to regulatory T cells. The authors indicated that these

immunologic changes were reported in both tumor tissues and peripheral blood as a result of treatment with anti-CTLA-4 antibody, and they may be used to guide dosing and scheduling of this agent to improve clinical responses. A sustained increased frequency of $CD4^{+}ICOS^{hi}$ T cells may serve as a biomarker of anti-CTLA-4 activity and/or of clinical benefit for patients who are being treated with this novel agent [41].

Pancreatic cancer

Royal and colleagues presented the results on 27 patients (metastatic disease: 20 and locally advanced: 7) [43]. Three subjects experienced ≥ grade 3 irAEs (colitis:1, encephalitis:1, hypophysitis:1). One subject experienced a delayed response after initial progressive disease. In this subject, new metastases after 2 doses of ipilimumab established progressive disease. However, continued administration of the agent per protocol resulted in significant delayed regression of the primary lesion and 20 hepatic metastases with normalization of tumor markers and clinically significant improvement of performance status. The investigators concluded that single agent ipilimumab at 3.0 mg/kg/dose was ineffective for the treatment of advanced pancreatic cancer. However, a significant delayed response in one subject of this trial suggests that immunotherapeutic approaches to pancreatic cancer deserve further exploration.

Lung Cancer

Study CA184041 was a double-blind, randomized, parallel-group, 3-arm, multicenter, Phase 2 study in previously untreated subjects with NSCLC and SCLC that evaluated the efficacy and safety of 2 schedules of ipilimumab (10 mg/kg) in combination with paclitaxel (175 mg/m2) and carboplatin (AUC = 6) compared to subjects receiving paclitaxel/carboplatin alone at the same doses.

Control arm: patients received up to 6 cycles of paclitaxel and carboplatin.

Concurrent schedule: ipilimumab induction doses were given with the first 4 chemotherapy doses (Cycles 1 to 4).

Phased schedule: ipilimumab induction doses were given with the last 4 chemotherapy doses (Cycles 3 to 6). Patients in all 3 arms received maintenance with ipilimumab 10 mg/kg or placebo q12w after completion of induction therapy.The primary endpoint of the study was irPFS in the NSCLC cohort. Immune-related response criteria were developed to better capture novel response patterns observed with immunotherapy in melanoma (Wolchok J.D., Clin Cancer Res, 2009). Secondary endpoints included PFS by WHO criteria, BORR, and OS in the NSCLC and SCLC cohorts.

CTEP-Sponsored Studies

The DCTD, NCI, has sponsored nine studies with ipilimumab including one pilot study (NCI #5744, lymphoma), three phase 1 studies (5708 [ovarian], 6082 [solid tumors], and 7458 [solid tumors]), one phase 1/2 study (6359 [non-Hodgkin's lymphoma]) with single agent ipilimumab, two phase 1 combination studies in prostate cancer with GM-CSF (6032) and with prostate-specific antigen (PSA)-TRICOM vaccine (7207), one phase 2 combination study of ipilimumab with GM-CSF (E1608, melanoma) and one phase 3 study (E1609) of adjuvant ipilimumab therapy versus high-dose interferon alpha-2b in patients with resected high-risk melanoma.

Results from 11 patients (colon, n=3; non-Hodgkin's lymphoma, n=4; prostate, n=4) who

received ipilimumab on study 5744 included tumor regression in 2 patients with lymphoma; 1 of whom (follicular lymphoma patient) had a partial response (PR) of 14-month duration [44]. Ipilimumab was well tolerated with predominantly grade 1/2 toxicities. One drug-related grade 3 AE was observed. Tregs, as detected by expression of $CD4^+CD25^+CD62L^+$, declined at early time points but rebounded to levels at or above baseline values at the time of the next infusion. The investigators concluded that ipilimumab treatment depressed Treg numbers at early time points in the treatment cycle but was not accompanied by an increase in vaccine-specific CD8+ T-cell responses in these patients previously treated with a variety of investigational anticancer vaccines.

Hodi and colleagues reported preliminary results on 20 patients (11 metastatic melanoma patients and 9 metastatic ovarian carcinoma patients) on study 5708 [45]. None of the 11 patients from the metastatic melanoma cohort manifested grade 3 or 4 inflammatory toxicities; however, all subjects revealed mild inflammatory pathologies associated with low-level constitutional symptoms. The most common toxicity (10/11 subjects) was a grade 1-2 reticular and erythematous rash on the trunk and/or extremities that arose between 3 days and 3 weeks after antibody administration and then gradually resolved without specific intervention. Biopsies of involved skin revealed low-grade interface dermatitis, minor to moderate mononuclear infiltrates surrounding the superficial dermal vasculature, and increased mucin deposition in the papillary and reticular dermis. These pathologic features resembled those observed in mild cutaneous forms of systemic lupus erythematosis. Three PRs (range, 21-34+ months) and five events of stable disease (SD) (range, 4-25 months) were observed. One PR and three SDs (2, 4, and 6+ months) were observed in the ovarian carcinoma group. The investigators concluded that selective targeting of antitumor regulatory T cells (Treg) may constitute a complementary strategy for combination of ipilimumab and GM-CSF-based antigen tumor cell vaccine therapy.

Results from 29 patients with malignancies that were recurrent or progressive after allogeneic hematopoietic cell transplantation (allo-HCT) demonstrated that drug was well tolerated at single doses up to 3 mg/kg [5]. Four patients experienced organ-specific irAEs of reversible grade 3 arthritis, grade 2 hyperthyroidism, dyspnea, and grade 4 pneumonitis. Three patients had objective responses: one PR lasting for 2 months, and two durable complete responses (CRs). Two additional patients with Hodgkin's disease who had evidence of rapid disease progression prior to ipilimumab treatment achieved SD for 3 and 6 months, following infusion at the 3 mg/kg dose level. Median OS was 24.7 months. At a 3.0 mg/kg dose, active serum concentrations of ipilimumab were maintained for >30 days following a single infusion. Zhou and colleagues reported immunophenotypes of peripheral blood T cells, including T-cell reconstitution, activation, and Treg expression, in 29 patients before and after a single-dose infusion of ipilimumab [46]. CTLA-4 blockade by a single infusion of ipilimumab increased $CD4^+$ and $CD4^+HLA\text{-}DR^+$ T lymphocyte counts and intracellular CTLA-4 expression at the highest dose level (3.0 mg/kg). There was no significant change in Treg cell numbers after ipilimumab infusion. These data demonstrate that significant changes in T-cell populations occur on exposure to a single dose of ipilimumab.

Harzstark and colleagues reported results on 36 patients with hormone refractory metastatic prostate cancer [47]. Of six patients treated with ipilimumab at a dose of 3 mg/kg, three patients had confirmed PSA declines of ≥50%, with a time to progression (TTP) of 22, 26, and 103

weeks. One of these patients had a PR in hepatic metastases. Grade 3 IrAEs consisted of rash in five patients, panhypopituitarism in one patient, temporal arteritis in one patient, and diarrhea in three patients. Non-irAEs included grade 3 and 4 cerebrovascular events (one patient each), grade 3 angina (one patient), grade 3 atrial fibrillation (one patient), grade 3 fatigue (four patients), and grade 5 pulmonary embolism (one patient). One patient treated at 10 mg/kg had a PSA decline of ≥50% with a TTP of 39 weeks. Higher doses of treatment with MDX-010 + GM-CSF induced the expansion of activated circulating $CD25^+$, $CD69^+$, and $CD8^+$ T cells more frequently than was seen in patients who received the same doses of either MDX-010 or GM-CSF alone [48]. The sera screening with protein arrays showed that the treatment can induce antibody responses to the testicular antigen NY-ESO-1.

Patients with metastatic prostate cancer were treated with ProstVac vaccine and ipilimumab before chemotherapy. The median OS for all patients on study was 31.8 months with a 74% survival probability at 24 months [49]. The median Halabi predicted OS for all patients was 18.5 months. There was no significant difference in OS at different dose levels of antibody (range 1-10 mg). A unique effect of the vaccines on the rate of tumor growth may be a novel method to evaluate the anti-tumor effects of the vaccine [50]. The authors suggested that the addition of immune checkpoint inhibition may augment the clinical benefit of vaccines.

Ansell and colleagues reported data on 18 treated patients with NHL [51]. Two clinical responses were observed: one patient with diffuse large B-cell lymphoma (BCL) had an ongoing CR (>31 months), and one with follicular lymphoma had a PR lasting 19 months. In 5 of 16 cases tested, T-cell proliferation to recall antigens was >2 fold increased after ipilimumab therapy. The investigators have found that blockade of CTLA-4 signaling with the use of ipilimumab is well tolerated at the doses used. Ipilimumab has antitumor activity in patients with BCL, resulting in durable responses in a minority of patients. Ipilimumab at 3 mg/kg monthly for 4 months can be given safely and is the dose that recommended for future combination studies.

Clinical Safety

Safety Experience

The most common treatment-related AEs (those considered possibly, probably, or definitely related to study drug by the investigator) associated with the use of ipilimumab were immune related irAEs (Investigator Brochure, 2011). The irAEs primarily involved the gastrointestinal (GI) tract (*e.g.*, diarrhea and colitis) and skin (*e.g.*, pruritus and rash), and less frequently, the liver, endocrine glands (including the thyroid, pituitary, and adrenal glands) and nervous system. IrAEs were generally managed with either symptomatic therapy (grade 1-2 events), systemic corticosteroids (grade 3-4 events), or other immunosuppressants (*e.g.*, infliximab, mycophenolate mofetil) for steroid-unresponsive GI or hepatic irAEs, as appropriate. Management of irAEs was usually paired with omission of dosing for mild or moderate events and permanent discontinuation for severe irAEs. Ipilimumab can result in severe and fatal immune-mediated reactions due to T-cell activation and proliferation. Fatalities due to GI perforation, hepatic failure, toxic epidermal necrolysis, and Guillain-Barré syndrome have been reported in clinical trials of ipilimumab.

Clinical trials are conducted under widely varying conditions so that extrapolation to novel settings and combinations regarding rates and severity of events may be unreliable. Given the expected rate of toxicity which may require stopping study drug but may also be related to a therapeutic immunologic response alternative DLT criteria are discussed in section 6.

Min and colleagues reported three patients who received ipilimumab alone or combined with bevacizumab therapy and developed thyroiditis, and the first report of euthyroid Graves' ophthalmopathy [52]. They recommend that all patients on ipilimumab alone or combined with bevacizumab therapy have baseline thyroid function tests and careful monitoring for new onset of thyroid disease, particularly during the first 3 months of treatment. See specific events in section 5.

Safety Profile of Ipilimumab at a Dose of 10 mg/kg (Phase 2 data)
The safety profile of ipilimumab as monotherapy over multiple doses at a dose of 10 mg/kg in 325 subjects was determined from 4 completed melanoma studies. Overall, the incidence of grade 3/4 AEs attributable to study drug was 31%. The target organ system, the incidence, and the severity of the most commonly observed irAEs vary among studies and with drug combinations. Typically, the severity but not necessarily the overall incidence increases with dose. Additional information on specific events is provided in section 7.1 and the IB:

Summary of irAE Safety Data for 10 mg/kg in Melanoma

	Total	**Low-grade (Grade 1 - 2) (%)**	**High-grade (Grade 3 - 4) (%)**	**Median Time to Resolution of Grade 2 - 4 irAEs (weeks)**
All irAEs	72.3	46.2	25.2	-
Skin (*e.g.*, rash, pruritus)	52.0	49.2	2.8	6.14
GI (*e.g.*, colitis, diarrhea)	37.2	24.9	12.3	2.29
Liver (*e.g.*, LFT elevations)	8.0	0.9	6.8	4.0
Endocrine (*e.g.*, hypophysitis, hypothyroid)	6.2	3.7	2.5	20.1

Pregnancy
Preliminary results are available in cynomolgus monkeys. Pregnant monkeys received ipilimumab every 21 days from the onset of organogenesis in the first trimester through delivery, at dose levels either 2.6 or 7.2 times higher than the clinical dose of 3 mg/kg of ipilimumab (by AUC). No treatment-related adverse effects on reproduction were detected during the first two trimesters of pregnancy. Beginning in the third trimester, the ipilimumab groups experienced higher incidences of abortion, stillbirth, premature delivery (with corresponding lower birth weight), and higher incidences of infant mortality in a dose-related manner compared to controls. Based on animal data, ipilimumab may cause fetal harm. The use of ipilimumab during human pregnancy has not been formally studied in clinical trials. There have been 7 known pregnancies during ipilimumab treatment: in 3 female subjects and in the partners of 4 male study subjects. Two (2) of the 3 female pregnancies ended with elected terminations. The third female subject had a history of seizures and delivered the baby at 36 weeks gestation. The baby had respiratory

complications that resolved by birth week 16. Three (3) of the 4 partners of male study subjects had full term, normal babies. The fourth baby had small ureters, which are expected to grow as the baby matures. Although these outcomes do not indicate that stillbirths or other severe abnormalities will occur, pregnancy should be avoided during treatment with ipilimumab.

Immunogenicity

In clinical studies, 1.1% of 1024 evaluable patients tested positive for binding antibodies against ipilimumab in an electrochemiluminescent (ECL) based assay. This assay has substantial limitations in detecting anti-ipilimumab antibodies in the presence of ipilimumab. Infusion-related or peri-infusional reactions consistent with hypersensitivity or anaphylaxis were not reported in these 11 patients nor were neutralizing antibodies against ipilimumab detected. Because trough levels of ipilimumab interfere with the ECL assay results, a subset analysis was performed in the dose cohort with the lowest trough levels. In this analysis, 6.9% of 58 evaluable patients, who were treated with 0.3 mg/kg dose, tested positive for binding antibodies against ipilimumab. These results are highly dependent on methodology, and comparison of incidence of antibodies to ipilimumab with the incidences of antibodies to other products may be misleading.

Study Results and Clinical Efficacy

The clinical efficacy of ipilimumab as a single agent at a dose of 3 mg/kg administered q3w for 4 doses has been established in MDX010-20 (a randomized, controlled study in second line, locally advanced/metastatic melanoma), which led to approval of ipilimumab by the FDA. In study CA184024, the addition of 10 mg/kg ipilimumab to dacarbazine led to a prolongation of overall survival in patients with previously untreated melanoma.

In melanoma studies, disease stabilization in subjects receiving ipilimumab is characteristic of anti-tumor activity. Stable disease, sometimes of long duration, or slow steady decline of tumor lesion size over long periods of time, has been observed. Some subjects demonstrate initial tumor volume increase before response, possibly due to T-cell infiltration as shown by biopsies or to the time required for immunologic activation. Consequently, an initial determination of progressive disease and consequently PFS may not capture all patterns response and may underestimate the clinical activity of ipilimumab. Please see section “Considerations for Using Immune-Related Tumor Assessment Criteria (irRC).”

MDX010-20 (Phase 3, 3 mg/kg, previously treated melanoma)

A Phase 3, double-blind study enrolled patients with advanced (unresectable or metastatic) melanoma who had previously been treated with regimens containing one or more of the following: IL-2, dacarbazine, temozolomide, fotemustine, or carboplatin. Patients were randomized in a 3:1:1 ratio to receive ipilimumab 3 mg/kg + an investigational gp100 peptide vaccine (gp100), ipilimumab 3 mg/kg monotherapy, or gp100 alone. All patients were HLA-A2*0201 type; this HLA type supports the immune presentation of gp100. Patients were enrolled regardless of their baseline BRAF mutation status. Patients received ipilimumab every 3 weeks for 4 doses as tolerated (induction therapy). Patients with apparent tumour burden increase before completion of the induction period were continued on induction therapy as tolerated if they had adequate performance status. Assessment of tumour response to ipilimumab was conducted at approximately Week 12, after completion of induction therapy. Additional

treatment with ipilimumab (re-treatment) was offered to those who developed PD after initial clinical response (PR or CR) or after SD (per the modified WHO criteria) > 3 months from the first tumour assessment. The primary endpoint was OS in the ipilimumab+ gp100 group vs. the gp100 group. Key secondary endpoints were OS in the ipilimumab+ gp100 group vs. the ipilimumab monotherapy group and in the ipilimumab monotherapy group vs. the gp100 group. A total of 676 patients were randomized: 137 to the ipilimumab monotherapy group, 403 to the ipilimumab + gp100 group, and 136 to the gp100 alone group. The majority had received all 4 doses during induction. Thirty-two patients received re-treatment: 8 in the ipilimumab monotherapy group, 23 in the ipilimumab + gp100 group, and 1 in the gp100 group. Duration of follow-up ranged up to 55 months. Baseline characteristics were well balanced across groups. The median age was 57 years. The majority (71-73%) of patients had M1c stage disease and 37-40% of patients had an elevated lactate dehydrogenase (LDH) at baseline. A total of 77 patients had a history of previously treated brain metastases. The ipilimumab-containing regimens demonstrated a statistically significant advantage over the gp100 control group in OS. The hazard ratio (HR) for comparison of OS between ipilimumab monotherapy and gp100 was 0.66 (95% CI: 0.51, 0.87; p = 0.0026).

By subgroup analysis, the observed OS benefit was consistent within most of the subgroups of patients (M [metastases]-stage, prior interleukin-2, baseline LDH, age, sex, and the type and number of prior therapy). However, for women above 50 years of age, the data supporting an OS benefit of ipilimumab treatment were limited. The efficacy of ipilimumab for women above 50 years of age is therefore uncertain. As the subgroups analysis includes only small numbers of patients, no definitive conclusions can be drawn from these data

In the ipilimumab 3 mg/kg monotherapy group, median OS was 22 months and 8 months for patients with SD and those with PD, respectively. At the time of this analysis, medians were not reached for patients with CR or PR.

For patients who required re-treatment, the BORR was 38% (3/8 patients) in the ipilimumab monotherapy group, and 0% in the gp100 group. The disease control rate (DCR) (defined as CR+PR+SD) was 75% (6/8 patients) and 0%, respectively. Because of the limited number of patients in these analyses, no definitive conclusion regarding the efficacy of ipilimumab re-treatment can be drawn. The development or maintenance of clinical activity following ipilimumab treatment was similar with or without the use of systemic corticosteroids

CA184024 (Phase 3, previously untreated melanoma, 10 mg/kg)
The primary objective of this study was the comparison of OS in subjects administered ipilimumab (10 mg/kg) + DTIC versus DTIC monotherapy in previously untreated patients. The HR for comparison of OS between the groups was 0.72 (95% CI: 0.59, 0.87; P = 0.0009) at the time of the primary analysis.66 The improvement in OS for ipilimumab + DTIC versus placebo + DTIC is evident in the clear separation of treatment groups in the Kaplan-Meier plot and in an increased median OS (95% CI) of 11.17 (9.40, 13.60) months and 9.07 (7.75, 10.51) months, respectively. The Kaplan-Meier survival curve for the ipilimumab plus DTIC group revealed that a plateau begins at approximately Year 3, which extends to Year 5 and beyond (although heavy subject censoring may limit interpretation of survival data beyond 5 years). One of the hallmarks of ipilimumab efficacy is long-term survival. Survival updates from subjects enrolled in clinical

trials continue to show a durable long-term benefit with ipilimumab. Five-year survival rates from Study CA184024 (DTIC with or without ipilimumab) continue to support a long-term benefit of ipilimumab in treatment-naïve subjects with advanced melanoma. The 5-year OS rates in the current analyses were 18.2% for ipilimumab + DTIC and 8.8% for placebo + DTIC. The rates are similar to the previously reported 3- and 4-year OS rates and suggest that OS plateaus at the 3-year mark.

Overall Adverse Events (CA184024)
The most common treatment-related AEs in the ipilimumab + DTIC group were skin and GI irAEs. The most common (≥20%) treatment-related AEs of any grade in the ipilimumab plus DTIC group were nausea (40.1%), fatigue (33.6%), diarrhea (32.8%), pyrexia (31.2%), ALT increased (29.1%), AST increased (26.7%), pruritus (26.7%), vomiting (26.3%), and rash (22.3%). The most common events in the DTIC group were nausea (42.6%), fatigue (27.1%), and vomiting (20.7%). The most common

10 mg/kg Dosing with Ipilimumab
Several additional trials studied the efficacy and safety of 10 mg/kg dosing, and additional information gained from these trials is listed below:

A dose of 10 mg/kg may be necessary to ensure blockade of the CTLA-4 pathway; in vitro, a concentration of 20 mcg/mL of ipilimumab was the minimal concentration able to fully abrogate the binding of CTLA-4 to B7.1 and B7.2. With a dose of 3 mg/kg q3w, 30% achieved a trough concentration of ipilimumab greater than 20 μg/mL, compared to 95% of subjects treated at 10 mg/kg q3w.

In addition, in all ipilimumab trials examined to date, mean Absolute Lymphocyte Count (ALC) increased after ipilimumab treatment throughout the 12-week induction-dosing period, in a dose-dependent manner. In an analysis of ipilimumab at 0.3, 3, or 10 mg/kg in melanoma studies CA184007, CA184008, and CA184022 combined, the rate of change in ALC after ipilimumab treatment was significantly associated with dose ($p = 0.0003$), with the largest rate at 10 mg/kg ipilimumab. Moreover, the rate of change in ALC over the first half of the induction-dosing period was significantly associated with clinical activity in these studies ($p = 0.009$), where clinical activity was defined as CR, PR, or prolonged SD (i.e., SD lasting at least 6 months from first dose). Although these analyses alone could not determine whether the rate of change in ALC was specifically associated with clinical activity in response to ipilimumab treatment, as opposed to being generally prognostic, these results do suggest a potential benefit to higher rates of ALC increase after ipilimumab treatment. Among the 3 doses evaluated, 10 mg/kg ipilimumab led to the greatest such rates.

In the 3 primary studies conducted in advanced melanoma (CA184007, CA184008, and CA184022), subjects treated with 10 mg/kg single agent ipilimumab had the highest response, disease control rates, median OS as well as 1-year and 2-year survival rates compared to lower doses. The CA184022 data are summarized in the table below.

Summary of Phase 2 Response Data in Melanoma (CA184022 see IB)

	10 mg/kg (n = 72)	3 mg/kg (n = 72)	0.3 mg/kg (n = 73)
BORR (mWHO) – %	11.1	4.2	0
(95% CI)	(4.9 - 20.7)	(0.9 - 11.7)	(0.0 - 4.9)
DCR (mWHO) – %	29.2	26.4	13.7
(95% CI)	(19.0 - 41.1)	(16.7 - 38.1)	(6.8 - 23.8)
Survival rate at 1 year - %	48.64	39.32	39.58
%, 95% CI	(36.84, 60.36)	(27.97, 50.87)	(28.20, 51.19)
Survival rate at 2 year - %	29.81	24.20	18.43
%, 95% CI	(19.13, 41.14)	(14.42, 34.75)	(9.62, 28.22)
Overall median survival	11.43	8.74	8.57
95%CI (months)	(6.90, 16.10)	(6.87, 12.12)	(7.69, 12.71)

Dose, Schedule, and Regimen

While optimal doses and schedules for ipilimumab have not yet been determined, in proposed proof of principle studies demonstration of efficacy at 10 mg/kg would allow future studies to explore biologic and clinical efficacy at lower doses with reduced toxicity. For most studies in new combinations or settings, a short phase 1 component at 3 mg would be appropriate with a 5 or 6 mg/kg dose added as an additional cohort if needed.

A recommended dose of 10 mg/kg is proposed by the manufacturer for most studies of ipilimumab. In melanoma, a similar survival benefit was demonstrated in phase 3 trials at the 3 mg/kg and at 10 mg/kg with DTIC. However, the incidence of grade $\geq$3 toxicity was 15 and 25% respectively.

Based on Phase 2 studies, response rates of ipilimumab appear to be dose dependent up to 10 mg/kg. Exposure-response analyses [C_{min}ss Analysis of PK data from patients treated with ipilimumab at 0.3 mg/kg (N=47), 3 mg/kg (N=60) and 10 mg/kg (N=311)], showed that the target C_{min}ss target threshold of 20 mcg/ml was exceeded in 0%, 30% and 95% subjects respectively. The slope of change in absolute lymphocyte count (ALC) correlated with clinical benefit and T-cell activation markers such as HLA-class II expression may also be dose dependent. Responses have not been compared systematically in randomized phase 2 or phase 3 studies in patients with tumor types other than melanoma.

Regarding schedule, the typical schedule for advanced melanoma at present is once q3w for four doses followed by a maintenance phase of four doses every 12 weeks. Of interest, ipilimumab was evaluated in NSCLC and SCLC using a dose of 10 mg/kg given concomitantly or following initial paclitaxel/cisplatin. When used in the phased schedule, 10 mg significantly improved irPFS and mWHO defined responses but not PFS determined by Response Evaluation Criteria in Solid Tumors (RECIST). There also was a trend for an improvement in OS in both indications. Doses less than 10 mg/kg have not been evaluated in either NSCLC or SCLC.

Studies comparing doses in non-melanoma and combinations have not been widely done. There are also no clear data that peak levels, Cmin, AUC, exposure and number of doses given, or the occurrence of autoimmune events, predict responses in individual patients. We note that the incidence of specific events such as hypophysitis may vary from study to study and with different combinations of agents. The severity and possibly time to onset but not necessarily the frequency of events increases with dose. In addition, there are rare but serious events such as toxic epidermal necrolysis (TEN) for which a dose relationship has not been established. Case report forms should include data on the prior treatment, timing, number of doses, duration of event, response to treatment, and complications to allow comparisons among studies.

Prescribing Information
Ipilimumab is indicated for the treatment of (1) unresectable or metastatic melanoma and (2) adjuvant treatment of patients with cutaneous melanoma with pathologic involvement of regional lymph nodes of more than 1 mm who have undergone complete resection, including total lymphadenectomy. For (1) unresectable or metastatic melanoma, ipilimumab is dosed at 3 mg/kg intravenously over 90 minutes every 3 weeks for a total of 4 doses. For (2) adjuvant melanoma treatment, ipilimumab is dosed at 10 mg/kg intravenously over 90 minutes every 3 weeks for 4 doses, followed by 10 mg/kg every 12 weeks for up to 3 years or until documented disease recurrence or unacceptable toxicity.

Considerations in Using Immune-Related Tumor Assessment Criteria (irRC)
New end point definitions for trials of immunologic agents have been proposed based on novel patterns of clinical activity in malignant melanoma [53, 54]. These alternative definitions allow time for immunologically mediated effectors to develop that may result in late tumor responses even after initial progression by RECIST. Also, in some patients, tumors necrosis and inflammation may increase tumor size radiographically prior to response. Changing the definitions of OR and PD may alter (increase) the number patients achieving responses and the duration of PFS.

On a protocol by protocol basis, we would consider allowing study treatment to continue during initial progression up to the 12-16 week assessment to allow time for responses to be observed, if the patient is clinically stable, there is no deterioration in PS, and there is no need for immediate additional treatment. While maintaining standard definitions of progression and response, we would allow new lesions and some progression beyond 20% increases in tumor measurements during the initial treatment period to allow time for responses to develop (these delayed tumor responses may be seen in 10-20% of melanoma patients who initially progress during the initial treatment cycles and evaluation). We do not have experience with response patterns with combination therapy nor in diseases other than melanoma. Please use standard response definitions as the primary end point in these studies.

Note that the proposed irRC may be incorporated as secondary end points to compare to standard criteria and evaluate alternative patterns of response in various disease setting and treatment regimens.

Patients who demonstrate mixed responses, stable disease, or objective responses by standard RECIST following initial progression may be identified separately as "delayed SD, PR, or CR".

Overall Risk/Benefit Assessment

The unique immune-based mechanism of action is reflected in the clinical patterns of anti-cancer activity in some patients. Ipilimumab affects tumor cells indirectly, and measurable clinical effects emerge after the immunological effects. Tumor infiltration with lymphocytes and the associated inflammation (documented by biopsy in some subjects) is likely the cornerstone of the effect of ipilimumab and can manifest in various patterns of clinical activity leading to tumor control. In some cases, response may be preceded by an apparent increase in initial tumor volume and/or the appearance of new lesions, which may be mistaken for tumor progression on radiological evaluations. Therefore, in subjects who are not experiencing rapid clinical deterioration, confirmation of progression is recommended, at the investigator's discretion, to better understand the prognosis as well as to avoid unnecessarily initiating potentially toxic alternative therapies in subjects who might be benefitting from treatment. Immune-related (ir) response criteria were developed based on these observations to systematically categorize novel patterns of clinical activity and are currently being prospectively evaluated in clinical studies.

In metastatic diseases, stabilization is more common than response, and in some instances is associated with slow, steady decline in tumor burden over many months, sometimes improving to partial and/or complete responses. Thus, the immune-based mechanism of action of ipilimumab results in durable disease control, sometimes with novel patterns of response, which contribute to its improvement in OS.

The immune-based mechanism of action is also reflected in the safety profile. The most common drug-related AEs are immune-mediated, consistent with the mechanism of action of the drug and generally medically manageable with topical and/or systemic immunosuppressants. As previously discussed, the immune-mediated adverse reactions primarily involve the GI tract, skin, liver, endocrine glands, and nervous system.

The early diagnosis of immune-mediated adverse reactions is important to initiate therapy and minimize complications. Immune-mediated adverse reactions are generally manageable using symptomatic or immunosuppressive therapy as recommended through detailed diagnosis and management guidelines, as described fully in the current IB. The management guidelines for general immune-mediated adverse reactions and ipilimumab-related GI toxicities, hepatotoxicity, endocrinopathy, and neuropathy are provided in the appendices of the current IB.

For this clinical trial (CTEP 10026), ipilimumab has already demonstrated a signal of clinical efficacy in patients with relapsed/refractory MDS/AML [4]. There is no accepted standard of care for patients with relapsed/refractory MDS/AML including for those that relapse post-HCT. Thus treatment for this population remains an unmet need. There has been significant clinical data demonstrating safety with hypomethylating agents in relapse both in the post-transplant setting and in the transplant naive. The combination of checkpoint blockade with hypomethylating agent is still investigational.

Rationale for Ipilimumab in this Study

There is no clear standard of care for patients with hematologic malignances who relapse after allo-HCT. Ipilimumab has been evaluated in two separate studies for patients with relapsed hematologic malignancies after allo-HCT with evidence of safety at low doses and preliminary efficacy when tested at doses currently approved in advanced melanoma [4, 5]. These two clinical studies suggest that the role for checkpoint blockade requires further exploration in the relapsed setting.

2.3 Decitabine

Decitabine (Dacogen) is an FDA-approved drug for the treatment of MDS. The information below is gathered from the FDA package label. Please see package insert for more details. Dacogen is indicated for treatment of patients with myelodysplastic syndromes (MDS) including previously treated and untreated, *de novo* and secondary MDS of all French-American-British subtypes (refractory anemia, refractory anemia with ringed sideroblasts, refractory anemia with excess blasts, refractory anemia with excess blasts in transformation, and chronic myelomonocytic leukemia) and intermediate-1, intermediate-2, and high-risk International Prognostic Scoring System groups.

Mechanism of Action
Decitabine is believed to exert its antineoplastic effects after phosphorylation and direct incorporation into DNA and inhibition of DNA methyltransferase, causing hypomethylation of DNA and cellular differentiation or apoptosis. Decitabine inhibits DNA methylation *in vitro,* which is achieved at concentrations that do not cause major suppression of DNA synthesis. Decitabine-induced hypomethylation in neoplastic cells may restore normal function to genes that are critical for the control of cellular differentiation and proliferation. In rapidly dividing cells, the cytotoxicity of decitabine may also be attributed to the formation of covalent adducts between DNA methyltransferase and decitabine incorporated into DNA. Nonproliferating cells are relatively insensitive to decitabine.

Pharmacokinetics
Pharmacokinetic parameters were evaluated in patients. Eleven patients received 20 mg/m2 infused over 1 hour intravenously (treatment Option 2), fourteen patients received 15 mg/m2 infused over 3 hours (treatment Option 1). Plasma concentration-time profiles after discontinuation of infusion showed a biexponential decline. The CL of decitabine was higher following treatment Option 2. Upon repeat doses there was no systemic accumulation of decitabine or any changes in PK parameters. Population PK analysis (N=35) showed that the cumulative AUC per cycle for treatment Option 2 was 2.3-fold lower than the cumulative AUC per cycle following treatment Option 1.

Pharmacodynamics
Decitabine has been shown to induce hypomethylation both in vitro and in vivo. However, there have been no studies of decitabine induced hypomethylation and pharmacokinetic parameters.

Major route of elimination

The exact route of elimination and metabolic fate of decitabine is not known in humans. One of the pathways of elimination of decitabine appears to be deamination by cytidine deaminase found principally in the liver but also in granulocytes, intestinal epithelium and whole blood.

Metabolism
Drug interaction studies with decitabine have not been conducted. *In vitro* studies in human liver microsomes suggest that decitabine is unlikely to inhibit or induce cytochrome P450 enzymes. *In vitro* metabolism studies have suggested that decitabine is not a substrate for human liver cytochrome P450 enzymes. As plasma protein binding of decitabine is negligible (<1%), interactions due to displacement of more highly protein bound drugs from plasma proteins are not expected.

NONCLINICAL TOXICOLOGY
Carcinogenesis, Mutagenesis and Impairment of Fertility
Carcinogenicity studies with decitabine have not been conducted. The mutagenic potential of decitabine was tested in several *in vitro* and *in vivo* systems. Decitabine increased mutation frequency in L5178Y mouse lymphoma cells, and mutations were produced in an *Escherichia coli lac-I* transgene in colonic DNA of decitabine-treated mice. Decitabine caused chromosomal rearrangements in larvae of fruit flies. The effect of decitabine on postnatal development and reproductive capacity was evaluated in mice administered a single 3 mg/m2 IP injection (approximately 7% the recommended daily clinical dose) on day 10 of gestation. Body weights of males and females exposed *in utero* to decitabine were significantly reduced relative to controls at all postnatal time points. No consistent effect on fertility was seen when female mice exposed *in utero* were mated to untreated males. Untreated females mated to males exposed *in utero* showed decreased fertility at 3 and 5 months of age (36% and 0% pregnancy rate, respectively). In male mice given IP injections of 0.15, 0.3 or 0.45 mg/m2 decitabine (approximately 0.3% to 1% the recommended clinical dose) 3 times a week for 7 weeks, decitabine did not affect survival, body weight gain or hematological measures (hemoglobin and WBC counts). Testes weights were reduced, abnormal histology was observed and significant decreases in sperm number were found at doses ≥ 0.3 mg/m2. In females mated to males dosed with ≥ 0.3 mg/m2 decitabine, pregnancy rate was reduced and pre-implantation loss was significantly increased.

Clinical activity in AML
Decitabine is a well-tolerated DNA-hypomethylating agent that induces differentiation and cytotoxicity of leukemic cells. CR rates from phase 2 and 3 AML clinical trials with front-line decitabine range from 18-25% [15].

Rationale for the Proposed Starting Dose and Dose Escalation Scheme in this Trial
The optimal dosing schedule for ipilimumab in hematologic malignancies is not entirely clear. Studies thus far have mimicked scheduled based on solid tumors (primarily melanoma studies). Ipilimumab is currently approved in unresectable or metastatic melanoma on a 3 week schedule for a total of 4 doses and in the adjuvant melanoma setting on a 3 week schedule for 4 doses (for induction phase) followed by an every 12 week schedule for up to 3 years (for maintenance phase) until disease recurrence or unacceptable toxicity. Ipilimumab has a prolonged half life of ~ 15 days. Dosing during the induction phase on a four week schedule has been previously

evaluated in melanoma in combination with dacarbazine [55] and in B-cell non-Hodgkin lymphoma as monotherapy with evidence of safety and anti-tumor activity [51]. In the Checkmate-012 lung cancer study, multiple schedules of combined nivolumab and ipilimumab were evaluated with similar safety profiles when ipilimumab was dosed on a 3 week versus 6 week versus 12 week schedule. Ipilimumab has been further evaluated in combination with peptide vaccination with a 6 to 8 week schedule with a similar safety profile [56]. Given the highly proliferative nature of AML and the known activity with single agent decitabine (median time to best response 2-4 cycles and median duration of response from 9-14 months) [14, 15], we will shorten the interval between doses in the maintenance phase for ipilimumab from 12 weeks to 8 weeks.

This study will have two arms (cohorts) to the study separated by transplant status. Arm A will include patients who are post allo-HCT. These include patients who are previously had an allo-HCT of any donor source at any time. Arm B will include patients who are transplant naïve. Both arms A and B will be evaluated at the same three dose escalation levels (and same starting dose level) but as the MTD/RP2D may differ based on transplant status, these two patient cohorts will be evaluated separately for safety and toxicity. There will be three intended phases of this clinical trial (for both Arms A and B): priming, induction phase, and maintenance phase.

- **Priming Phase** (Lead- In): The priming phase includes decitabine alone at 20 mg/m2 given daily for 5 consecutive doses starting on days 1-5 on a 28 day cycle to provide priming and disease control. Hypomethylating agents, such as decitabine have been tested in the post allo-HCT setting with evidence of safety and tolerability [57-59] including at the same dose proposed in this study.
- **Induction Phase** (cycles 1-4): Cycles 1 through 4 will include treatment with decitabine 20 mg/m2 daily for five consecutive doses on days 1-5 plus ipilimumab (given per dose level) on day 1 on a 28 day cycle. There will be three dose escalation levels tested: 3.0 mg/kg (starting dose), 5.0 mg/kg, and 10.0 mg/kg. There will be a dose de-escalation level provided (1.0 mg/kg). Pharmacokinetic data generated from the initial pilot study demonstrated that higher doses of ipilimumab correlated with increased ipilimumab exposure and that those doses may be sufficient to saturate CTLA-4 binding sites [5]. The starting dose level of 3.0 mg/kg was tested in the phase 1b study for relapsed hematologic malignancies after allo-HCT with demonstration of safety and tolerability [4]. There was evidence of preliminary clinical activity (discussed in Section 2.4 in detail) in this study at 10 mg/kg but with evidence of immune related adverse events and GVHD in some cases. Thus we will also test the dose of 5.0 mg/kg.
- **Maintenance Phase** (cycles 5-12): Cycles 5 through 12 will include treatment with decitabine 20 mg/m2 daily for five consecutive doses on days 1-5 on a 28 day cycle. Ipilimumab will continue on day 1 every 8 weeks for a total of 4 doses.

2.4 Rationale

Disease relapse remains the predominant problem for patients with AML/MDS. Data from solid tumor studies suggest that treatment including CTLA-4 blockade may provide durable remissions. In pooled analysis across advanced melanoma studies, treatment with ipilimumab regardless of dose resulted in survival curves that clearly plateau around year 3 (with an OS rate of ~20%) [60]. Responses in metastatic melanoma have been associated with tumor reactive T

cells expanding from a tumor site. In vitro evaluation with CTLA-4 blockade has shown induction of a proliferative signature predominantly in a subset of transitional memory T cells [61]. PD-1 blockade, alternatively, showed changes in genes involved in cytolysis and NK cell function. CTLA-4, thus, is a reasonable target for therapy.

Clinical activity with CTLA-4 blockade in relapsed hematologic malignancies in the post-transplant setting

Evidence for anti-leukemic activity in the absence of graft versus host reactivity with CTLA-4 blockade was previously studied in a mouse model of minor histocompatibility-mismatched bone marrow transplantation [62]. In a mouse model of minor histocompatibility mismatched bone marrow transplantation, delayed CTLA-4 blockade induced anti-leukemic activity in the absence of graft versus host reactivity [62]. A pilot study next demonstrated safety with low-dose ipilimumab in the treatment of patients with relapsed hematologic malignancies post alloHCT [5]. Results from this study suggested that titration of ipilimumab might be possible in order to generate a graft-versus-tumor response. Pharmacokinetic data generated from this pilot study demonstrated that higher doses of ipilimumab correlated with increased ipilimumab exposure and that those doses may be sufficient to saturate CTLA-4 binding sites [5]. A Dana-Farber Cancer Institute led Phase I/Ib multi-center study (NCT01822509) of dose-escalated ipilimumab in the post-transplant setting for relapsed hematologic malignancies demonstrated substantial anti-tumor activity [4, 63]. There was no formal clinical response for patients treated in the 3 mg/kg cohort. Out of 6 evaluable patients in the 3 mg/kg cohort, one patient had chronic GVHD of the liver, which was the only DLT in the cohort. There was evidence of immune-related adverse events in two patients (1 patient with grade 2 pneumonitis and 1 patient with grade 2 diarrhea), which rapidly reversed with glucocorticoids and did not prevent further administration of ipilimumab. Twenty-two patients including an expansion cohort went on to receive 10 mg/kg dosing of ipilimumab. The overall response rate (ORR) in the 10 mg/kg dose cohort was 32% (7/22) although 59% (13/22) had disease reduction. Responders included 5 with AML, 1 partial response (PR) with classical Hodgkin's, and 1 PR with multiple myeloma with highly refractory lung plasmacytoma. Of the AML patients treated on study, 42% (5 of 12) achieved complete remission (CR), including 3 with extramedullary AML, 1 with myeloid sarcoma and 1 with smoldering MDS/AML with marrow involvement (low blast burden). Importantly, responses have been durable in three of the AML responders, ranging from 12-16 months. The median duration of response has not yet been reached. The 1 year OS was 49% at 15 months [4]. Exploratory correlative studies revealed a decrease in the ratio of T_{reg}/T_{conv} ratio, suggesting possible augmentation of the graft versus tumor effect. Results from this phase I/Ib study suggest that ipilimumab is feasible for relapsed disease after allo-HCT although at the dose of 10 mg/kg there was evidence of both immune related adverse events and GVHD, which were distinguished by biopsy. In the 10 mg/kg cohort, there was evidence of dose-limiting toxic effects including 2 patients with chronic GVHD of the liver and 1 patient with grade II acute GVHD of the gut. These GVHD cases resolved with steroids but precluded further administration of ipilimumab. Immune-related adverse events were observed in 3 patients (1 patient with grade 2 immune thrombocytopenia; 1 patient with grade 3 colitis; 1 patient with grade 2 pneumonitis and 1 patient with grade 4 pneumonitis). With steroid treatment, ipilimumab was resumed ~ 3 weeks in 2 patients. One of these patients died 42 days after receiving initial ipilimumab (grade 3 colitis and grade 4 pneumonitis). Notably, no infectious complications were seen that were grade 3 or higher. Consequently, the trial is now investigating the safety and preliminary efficacy

at an intermediate dose arm of 5 mg/kg.

Epigenetic priming prior to checkpoint blockade

Epigenetic silencing of immune-protective signature genes may affect clinical responses to immunotherapy. Preclinical data support the combination of epigenetic therapy with checkpoint blockade to induce the expression of immune regulatory genes in cancer. Treatment with epigenetic therapies in epithelial cancer models led to (1) the induction of regulators of interferon signaling through the hypomethylation of endogenous retroviral sequences [64] and (2) the removal of repression of Th1 type-chemokine expression (ie CXCL9 and CXCL10) which correlated with anti-tumor activity [65]. In AML, treatment with decitabine in cultured cells and in an established mouse EL4 tumor model resulted in upregulated expression of the CTLA-4 ligand CD80 [66]. Further, in leukemic blasts isolated from AML patients treated with hypomethylating agents (HMA), there was evidence for CTLA-4 expression upregulation [67]. We hypothesize that decitabine priming prior to ipilimumab treatment may lead to further activation of the immune system to stimulate anti-leukemic activity. HMA use post-HCT has been studied as maintenance [58] or in the setting of relapse with DLI [68]. HMA, such as azacitidine, has been shown to increase the number of regulatory T cells (which inhibit the development of graft-versus-host disease) and cytotoxic CD8+ T cells (which may enhance the graft-versus-leukemia effect) in the post-HCT setting [69]. HMA therapy has been shown to be safely administered to heavily pretreated post-HCT patients with evidence of prolonged event free survival and overall survival (OS) [58, 59]. Decitabine is more potent inhibitor of DNMT-1 than azacitidine. The Washington University group has demonstrated that decitabine enhances FOXP3 expression and converts CD4+CD25-FOXP3- T cells into CD4+CD25+FOXP3+ T regulatory cells, the latter are cells critical to modulating GVHD without compromising the GVL effect [70]. For this study, thus, decitabine is an attractive agent, given its mild toxicity and proven anti-leukemic activity, for combination therapy.

Preclinical data support the combination of epigenetic therapy with immunotherapy. Studies from AML cell lines and murine models demonstrate up-regulation of CD80 expression with decitabine treatment [66]. In primary AML samples, CTLA-4 up-regulation was also detected among patients who were treated with hypomethylating agents [71]. Recently, an Italian group demonstrated enhanced anti-tumor activity with CTLA-4 blockade in a syngenic mouse model bearing palpable tumor grafts of murine mammary carcinoma cells following epigenetic priming with the second generation investigational SGI-110, a dinucleotide of decitabine [72]. In this study, mice were treated with a CTLA-4 monoclonal antibody either concurrent (on days 2, 5, and 8) with or subsequent (on days 8, 11, and 14) to SGI-110 therapy. Tumor growth inhibition of 84% was appreciated when treatment was scheduled sequentially. Epigenetic silencing of immune-protective signature genes may affect clinical responses to immunotherapy. Peng D et al., found that 5-aza-2'-deoxycytidine reprogrammed epigenetic pathways by removing repression of T helper 1 (TH1) type-chemokine expression, including that of CXCL9 and CXCL10, and increased effector T-cell tumor infiltration leading to slowed tumor growth and improved therapeutic efficacy of checkpoint blockade in tumor-bearing mice [65]. Therefore, we hypothesize that sequential decitabine and ipilimumab treatments may lead to further activation of the immune system to stimulate anti-leukemic activity.

Hypothesis

Given the evidence of single-agent ipilimumab activity among AML patients with low-disease burden from the on-going Phase 1/1b study [4], we aim to improve the overall response by combining ipilimumab with a therapy that not only cytoreduces, but has the potential to alter the expression of immune regulatory genes to render leukemic cells more sensitive to checkpoint blockade. We hypothesize that epigenetic modifying agents may enhance the immune response by improving the recognition and destruction of leukemia cells by cytotoxic T lymphocytes in combination with anti-CTLA-4 therapy. We thus propose a phase 1 clinical study of sequential decitabine and ipilimumab to evaluate for safety and toxicity in patients with relapsed/refractory MDS and AML. We hypothesize that patients that relapse post-HCT may be at risk for GVHD at higher doses of ipilimumab compared to patients that have no prior transplant history. If this occurs, patients post-transplant may have a different maximum tolerated dose (MTD) or recommended phase 2 dose (RP2D) compared to the non-transplant population.

We have thus designed a dose-escalation study separating the patient cohorts by transplantation status. Biologically, it is also unknown whether alloreactivity in the post-HCT setting is required for clinical activity. This clinical study will evaluate the role of CTLA-4 blockade in AML and hopefully will add to the limited armamentarium available for the relapsed and refractory AML population. Current therapies available for elderly and relapsed AML are not curative without allo-HCT. Immunotherapy, alternatively, may generate long-lasting durable responses through the generation of cancer specific central memory T cells. If there is demonstration of preliminary efficacy and safety with sequential epigenetic and ipilimumab therapy, then this combination can serve as a new therapeutic strategy for AML and may potentially enhance graft-versus-leukemia effects for those who relapse post-transplant. Positive findings from this study would support further exploration of the role of checkpoint blockade to modulate anti-leukemic immune responses.

2.5 Correlative Studies Background

The goal of this study is to evaluate if decitabine (priming) sensitizes patients with MDS/AML to checkpoint blockade in the transplant naïve and post allo-HCT setting. We seek to evaluate if epigenetic therapy combined with blockade of immune checkpoints might augment response in leukemia by shifting the balance between immune activation and immune inhibition. This trial will be biopsy driven and offer the opportunity to examine whether decitabine, a nucleotide analog DNA demethylating agent will sensitize malignant cells to anti-leukemic immune responses. We predict that targeting of DNA methyltransferases with decitabine may "reprogram" tumor cells (as seen in solid tumor *in vitro* studies [73, 74]) by up-regulating the tumor immune stimulating profile. Additional work is still needed to understand how these mechanisms may differ between CTLA-4 blockade in the transplant naive and post-HCT setting and what effect epigenetic priming may have on the mechanism of checkpoint blockade. We will collaborate with several investigators, including Drs. Jerry Ritz, Catherine Wu, Scott Rodig, and Stephen Hodi (Center for Immuno-Oncology) to perform the immunologic correlative studies proposed in this clinical study. The correlative studies proposed in this clinical study will evaluate the role of epigenetic priming prior to CTLA-4 blockade in the transplant naive and post-HCT setting. We have designed a comprehensive set of phenotypic and genotypic assays to interrogate the immune and leukemic population dynamics both to identify novel predictors of response to checkpoint blockade as well as illuminate the mechanism of

action of CTLA-4 blockade in myeloid leukemias in the pre- and post allo-HCT setting.

Pharmacodynamic study: Measuring absolute lymphocyte count as a potential biomarker for response to ipilimumab
We will capture absolute lymphocyte counts (ALC) at baseline and on treatment with each cycle. It has been previously demonstrated that a rise in ALC during ipilimumab treatment may be associated with clinical response and OS in patients with cutaneous and uveal melanoma [75, 76]. We will compare measured levels at baseline (defined as prior to dosing on cycle 1 day 1) and at pre-determined time points (end of cycles 1, 2, and 4) and we will also compare changes in ALC from baseline (defined as prior to dosing on cycle 1 day 1) to the pre-determined time point (end of cycle 2) for survival differences given the evidence that ALC after two cycles may be a predictor of clinical response. As decitabine may cause myelosuppression, ALC elevation may not be reliable.

Correlative study: Clonal evaluation of patients on treatment
AML is commonly characterized by genetic alterations including gene mutations, copy number alterations and translocations. Genomic analysis of AML samples has expanded the number of known molecular mutations present in this disease [77]. Clonal evolution is a feature of the natural history of the disease that may also be influenced by the selective pressure of chemotherapy [78]. MDS is also a recognized clonal disorder with evidence of evolution or dynamic shaping of complex oligoclonal architectures upon some treatments [79]. Understanding the dynamic shaping of the clonal architectures including the outgrowths of founder-, sub- or even fully independent clones will provide insight on the dynamic rate of clonal turnover and potential guide clinical treatment decisions. In addition to their prognostic value, mutations present at diagnosis may also affect the response to initial treatment. To understand the genotypic alterations that may be associated with response, we will perform evaluations at baseline, after four cycles, and at time of relapse if relevant. Specifically, we will compare changes in variant allele fraction of pathogenic lesions found at baseline to response. Following clonal patterns may provide an alternative measurement of antitumor activity and help to identify the persistence of emergence of resistant clones during treatment, which mediate disease relapse [80].

Correlative study: Determining histopathologic patterns of response
We hypothesize that infiltrating T cells at the site of tumor microenvironment is critical to tumor control. Samples to be tested include marrow and extramedullary site (if amenable to biopsy) including skin in leukemia cutis. Using immunohistochemistry, we will evaluate for activated T-cells with CD8 and,granzyme B double IHC at baseline and post-treatment. We will also evaluate CD4, PD-1, and PD-L1. We will collaborate with Dr. Scott Rodig and his core laboratory at the Brigham and Women's Hospital to perform these evaluations. We will evaluate samples from time of screening and at the end of 2 cycles of treatment with the combination. As biopsies will otherwise be required per protocol at additional time points, we will capture IHC responses at later time points if we see evidence for delayed biological activity.

Correlative study: Evaluation of the immune phenotypic response to combination therapy
CTLA-4 expression is induced upon T cell activation. We hypothesize that epigenetic therapy may modulate T cell activation and that the addition of immunotherapy will give rise to an

increased number of CD4 and CD8 T cells infiltrating the tumor sites. Further, there is limited data available on the immune response to epigenetic therapy. Based on the observations in the single-agent ipilimumab post allo-HCT study [4] , we hypothesize that checkpoint blockade increases the number of effector memory T cells (TEM), which likely helps to augment GVT in responders. From whole blood, we will perform serial immunophenotypic analysis of immune cells utilizing flow cytometry to evaluate lymphocyte subsets and ELISA to assess chemokine/cytokine profiles (CXCL2, CXCL5, MICA, and others) to assess whether we observe similar results in patients treated with combination ipilimumab/ decitabine. Lymphocytes have an expansive repertoire and heterogeneity that is difficult to fully capture by conventional flow cytometry techniques. If we detect T cell activation using conventional flow cytometry, we then propose to further interrogate the lymphocyte repertoire using mass cytometry. Fluorescent dye-based flow cytometry can analyze up to 8-10 parameters of a single cell, but due to overlapping excitation and emission spectra of the different fluorescent dyes, the parameters cannot be expanded. We will use single cell mass cytometry (cytometry by time-of-flight, CyTOF), which enables analysis to 34 or more simultaneous parameters, to do a detailed analysis of both phenotypic and functional markers of all peripheral blood lymphocytes for patients on trial. We will use a panel of surface membrane and intracellular markers to assess the number and activation status of Treg, T_{EM}, T_{CM}, T_{naive}, and NK cells, including but not limited to CTLA-4, ICOS, PDL-1, TIM-3. We will collaborate with Dr. Jerome Ritz and Dr. Jacalyn Rosenblatt at the Dana-Farber Cancer Institute and Center for Immuno-Oncology. We will evaluate samples collected at baseline (prior to dosing on study), after the lead in period (epigenetic priming), and on treatment (at the end of cycles 2 and 4).

Correlative study: Evaluation of the transcriptional response to therapy
We will evaluate the gene expression pattern of immune infiltrates to understand differences in the tumor microenvironment in situ. We will utilize whole exome sequencing or bulk versus single cell transcriptomics to elucidate the heterogeneity of immune and cancer cell populations before and after therapy to understand how leukemic and immune populations co-evolve during response or resistance. Immune cell gene expression patterns and networks that correlate with clinical outcome will be inferred computationally and confirmed by multiplexed immunofluorescence for associated protein markers. Leukemic neoantigens that attract and drive anti-tumor immune responses will be identified and evaluated for correlation with clinical outcome. We will collaborate with Drs. Pavan Bachireddy/Catherine Wu at the Dana-Farber Cancer Institute. We use samples collected at baseline (i.e. aspirate if marrow disease, at time of screening), end of the lead in period (end of epigenetic priming), and end of cycle 2.

Potential Future Correlative study: Evaluation for resistance to immune response
Inadequate co-stimulation of T cells may be one mechanism underlying failure of adoptive immunity after alloHCT. T cell immune responses result from a balance between stimulatory and inhibitory signals. STAT3/5 activation can increase the expression of immunosuppressive proteins and decrease expression of co-stimulatory proteins. STAT3 is an oncogenic transcription factor that modulates immune checkpoint activity in cancer cells and its activation has been detected in leukemic blasts and is associated with chemoresistance [81]. Chemical inhibition of STAT3 activity using atovaquone and pyrimethamine in cultured AML cell lines up regulates the expression of immune co-stimulatory molecules and down regulates the expression of potential immunosuppressive proteins (*unpublished data,* see Figure 1 below, laboratory of

Dr. David Frank at the Dana-Farber Cancer Institute).

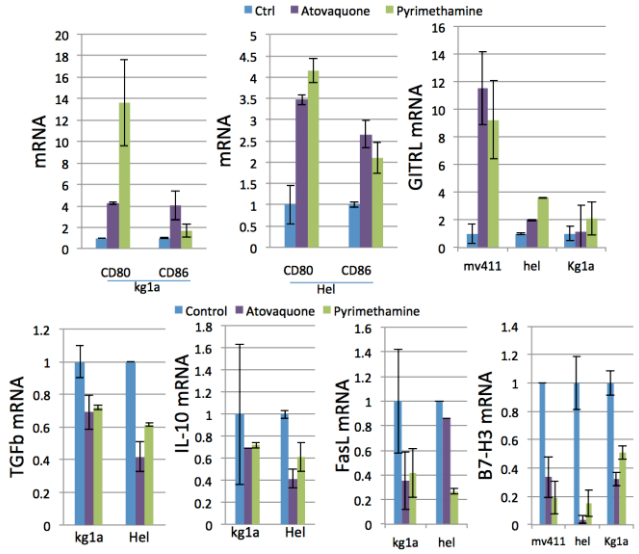

Figure 1. STAT3 inhibition with atovaquone or pyrimethamine (a) up regulates co-stimulatory molecules and (b) down regulates expression of immunosuppressive cytokines and molecules

We hypothesize that non-responders may show evidence of STAT3/5 activation in the tumor microenvironment, which may dampen the immunogenicity of AML cells. To evaluate the role of STAT3/5 activation as a mechanism of resistance, STAT3, STAT5, p-STAT3 and p-STAT5 will be captured by CyTOF among T cell subsets. If there is evidence of STAT3 activation among non-responders to treatment, we will further explore whether or not STAT3 target genes may be associated with treatment response [82].

Future studies: As more data becomes available during the study and as we analyze the transcriptome data, we may find candidate genes of interest to pursue further (i.e. are they subject to methylation) from the tumor microenvironment. In anticipation of this, we will save an aliquot of the samples to evaluate the methylome (one potential strategy is to perform whole-genome bisulfite sequencing to assess intra-tumor DNA methylation heterogeneity before and after epigenetic priming and after combination therapy). We will also consider TCR repertoire sequencing studies comparing peripheral blood and bone marrow samples. This technique will identify expanded T cell clones that likely mediate any clinical response. Samples from baseline (aspirate at screening), at the end of cycle 1, and on treatment will be saved for future exploration.

3. PATIENT SELECTION

3.1 Eligibility Criteria

3.1.1 Age 18 and older.

3.1.2 Subjects with evidence of AML or MDS that meet at least one of the following criteria:

- Relapsed AML: evidence of ≥ 5% blasts in the bone marrow; or reappearance of blasts in the peripheral blood; or development of

extramedullary disease (according to 2003 IWG criteria [6]) who relapse after:
    - Allogeneic hematopoietic stem cell transplant, or
    - After one cycle of standard cytotoxic chemotherapy or two cycles of any hypomethylating agent-based therapy
- Refractory AML: ≤ 2 prior induction regimens (example: patients who receive 7+3 followed by 5+2 would count as one induction regimen) or a minimum of two cycles of any hypomethylating agent-based therapy.
- Treatment-naive AML: must be 75 years and older with *de novo* or *secondary* AML to be considered eligible
- Relapsed MDS: disease recurrence after CR, PR or hematologic improvement with bone marrow blasts ≥ 5% who relapse after:
    - Allogeneic hematopoietic stem cell transplant, or
    - After four cycles of any hypomethylating agent-based therapy
- Refractory MDS: disease progression at any time after initiation of hypomethylating agent treatment or persistent bone marrow blasts ≥ 5% despite a minimum of four cycles of hypomethylating agent therapy.
- Untreated or previously treated therapy- related or secondary MDS

3.1.3 Allowed prior allogeneic hematopoietic stem cell transplantation (allo-HCT) regardless of stem cell source. Patients must be at least 3 months post allo-HCT (at time of treatment start). Mismatched transplantations would be allowed.

3.1.4 Patients must be off systemic immunosuppressive medications >2 weeks prior to treatment start. If patients are in systemic corticosteroids and must be on a dose of prednisone 5 mg/day or less (or equivalent), then patients must be on this reduced dose for >1 week prior to treatment start. Topical steroids are allowed

3.1.5 If post allo-HCT, then patient must have baseline donor T cell chimerism of ≥ 20% (from peripheral blood). Evaluation can be made within 4 weeks of treatment start.

3.1.6 No limitations on prior therapies.

3.1.7 ECOG performance status ≤ 2 (see **Appendix A**)

3.1.8 Participants must have normal organ as defined below:
- Total bilirubin ≤ 1.5 × local institutional upper limit of normal (ULN)
    - If elevated total bilirubin is due Gilbert's disease or disease-related hemolysis then total bilirubin ≤ 3.0 x local institutional ULN
- Aspartate aminotransferase (AST) or SGOT ≤ 3.0 × local institutional ULN
- Alanine aminotransferase (ALT) or SGPT ≤ 3.0 × local institutional ULN
- Serum creatinine ≤ 2.0 x local institutional ULN

3.1.9 Negative serum pregnancy test for women who are of child bearing potential (test must be repeated if performed> 72 hours from treatment start). The effects of ipilimumab on the developing human fetus are unknown. For this reason and because immunotherapy agents as well as decitabine are known to be teratogenic, women of child-bearing potential and men must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) prior to study entry and for the duration of study participation. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately. Men treated or enrolled on this protocol must also agree to use adequate contraception prior to the study, for the duration of study participation, and 4 months after study drug administration.

3.1.10 Patients with known active HIV infection. Patients with chronic HIV with a CD4 > 250, undetectable viral load by PCR, without opportunistic infection, and on a stable regimen of HAART therapy would be eligible

3.1.11 Ability to understand and the willingness to sign a written informed consent document.

3.2 Exclusion Criteria

3.2.1 Participants who have had chemotherapy or radiotherapy within 2 weeks prior to treatment start or those who have not recovered from adverse events due to agents administered more than 2 weeks prior to treatment start.

- Hydroxyurea is allowed for symptomatic leukocytosis if clinically necessary. A total white blood cell (WBC) count < 25 x 10^9/L prior to first dose of decitabine on trial is required. Prior leukopheresis and/or prior or concurrent treatment with hydroxyurea to achieve this level are allowed.
- Ongoing concurrent hormonal therapy is allowed

3.2.2 Participants with known CNS involvement with leukemia or who are receiving intrathecal chemotherapy for active CNS leukemia

- Those with a history of CNS involvement that has been completely treated and those who require intrathecal chemotherapy prophylaxis are eligible in the expansion cohorts

3.2.3 Prior HMA therapy is allowed, however this study *excludes* patients with progression or relapse that occur while receiving HMA-based therapy within 12 weeks prior to treatment start on study.

Disease progression is defined as either: (1) patients with prior MDS who progress to AML (defined by the presence of ≥ 20% blasts in peripheral blood or bone marrow) on HMA-based therapy; OR (2) patients with AML with evidence of progressive disease according to ELN 2017 criteria (Appendix H for details) (e.g.

>50% increase in marrow blasts over baseline or >50% increase in peripheral blasts to > 25 x10^9/L (>25,000/uL) (in absence of differentiation syndrome)).

(Note: Patients who relapse post-transplant who received HMA treatment prior to transplant are eligible for study.)

3.2.4 Donor lymphocyte infusion within 8 weeks prior to treatment start if post-transplant.

3.2.5 For patients that are post-transplant, ineligible patients include those with a history of overall grade III or IV (severe) acute GVHD at any time even if resolved.

3.2.6 Patients with a history of prior treatment with anti-CTLA-4 antibody, anti-PD 1 antibody, or anti- PDL1 antibody.

3.2.7 Participants who are receiving any other investigational agents.

3.2.8 Participants with known CNS involvement with leukemia or who are receiving intrathecal chemotherapy that is either prophylactic or therapeutic. History of CNS involvement that has been completely treated (no longer receiving intrathecal chemotherapy) will be allowed.

3.2.9 Uncontrolled intercurrent illness including, but not limited to, ongoing or active infection, symptomatic congestive heart failure, unstable angina pectoris, cardiac arrhythmia, or psychiatric illness/social situations that would limit compliance with study requirements. Any other prior or ongoing condition, in the opinion of the investigator, that could adversely affect the safety of the patient or impair the assessment of study results. As patients with AML and MDS are prone to infections, if patients are actively being treated with appropriate antibiotics or antifungal therapy with clinical evidence of infection control, then they will be considered eligible for study.

3.2.10 **Autoimmune disease**: Patients who are not eligible include those with a history of inflammatory bowel disease, including ulcerative colitis and Crohn's Disease, are excluded from this study, as are patients with a history of symptomatic disease (e.g., rheumatoid arthritis, systemic progressive sclerosis [scleroderma], systemic lupus erythematosus, autoimmune vasculitis [e.g., Wegener's Granulomatosis]); CNS or motor neuropathy considered of autoimmune origin (e.g., Guillain-Barre Syndrome and Myasthenia Gravis, multiple sclerosis).Patients with a history of autoimmune disease (specifically including: diabetes mellitus, vitiligo, hashimoto's thyroiditis) who are asymptomatic, do not require immune suppression or steroids, and do not have threatened vital organ function from these conditions may be considered after discussion with the PI

3.2.11 No concurrent active malignancies are allowed on study for ≥ 2 years prior to treatment start with the exception of currently treated basal cell or squamous cell carcinoma of the skin, or carcinoma in-situ of the cervix or breast.

3.2.12 Patients with known active hepatitis B virus (HBV) infection should be excluded because of potential effects on immune function and/or drug interactions. However, if a patient has HBV history with an undetectable HBV load by polymerase chain reaction (PCR), no liver-related complications, and is on definitive HBV therapy, then he/she would be eligible for study.

3.2.13 Patients with known active hepatitis C virus (HCV) infection. Patients with a history of HCV infection who received definitive therapy and has an undetectable viral load by PCR would be eligible.

3.2.14 Pregnant women are excluded from this study because ipilimumab has the potential for teratogenic or abortifacient effects. Because there is an unknown but potential risk for adverse events in nursing infants secondary to treatment of the mother with ipilimumab, breastfeeding should be discontinued if the mother is treated with ipilimumab. These potential risks may also apply to decitabine.

3.3 Pregnancy

The effects of ipilimumab on the developing human fetus are unknown. For this reason and because immunotherapy agents as well as decitabine are known to be teratogenic, women of child-bearing potential and men must agree to use adequate contraception (hormonal or barrier method of birth control; abstinence) prior to study entry and for the duration of study participation. Should a woman become pregnant or suspect she is pregnant while she or her partner is participating in this study, she should inform her treating physician immediately. Men treated or enrolled on this protocol must also agree to use adequate contraception prior to the study, for the duration of study participation, and 4 months after study drug administration.

Sexually active women of child-bearing potential (WOCBP) must use an effective method of birth control during the course of the study, in a manner such that risk of failure is minimized. Before enrolling WOCBP in this clinical study, the investigator must review the guideline about study participation for WOCBP which can be found in the GCP Manual for Investigators. The topics include the following:

- General Information
- Informed Consent Form
- Pregnancy Prevention Information Sheet
- Drug Interactions with Hormonal Contraceptives
- Contraceptives in Current Use
- Guidelines for the Follow-up of a Reported Pregnancy.

Before study enrollment, WOCBP must be advised of the importance of avoiding pregnancy during study participation and the potential risk factors for an unintentional pregnancy. The subject must sign an informed consent form documenting this discussion.

All WOCBP MUST have a negative serum quantitative pregnancy test within 72 hours of receiving treatment on study. The minimum sensitivity of the pregnancy test must be 25 IU/L or equivalent units of HCG. If the pregnancy test is positive, the subject must not receive ipilimumab and must not be enrolled in the study.

In addition, all WOCBP should be instructed to contact the investigator immediately if they suspect they might be pregnant (e.g., missed or late menstrual period) at any time during study participation.

If, following initiation of the investigational product, it is subsequently discovered that a study subject is pregnant or may have been pregnant at the time of investigational product exposure, including during at least 6 half-lives after product administration, the investigational product will be permanently discontinued in an appropriate manner (*e.g.*, dose tapering if necessary for subject safety). Pregnancies should be reported directly to CTEP through the Adverse Event Reporting System (AERS) and must also be accompanied by the Pregnancy Information Form

Protocol-required procedures for study discontinuation and follow-up must be performed on the subject unless contraindicated by pregnancy (*e.g.*, X-ray studies). Other appropriate pregnancy follow-up procedures should be considered if indicated. In addition, the investigator must report and follow-up on information regarding the course of the pregnancy, including perinatal and neonatal outcome. Infants should be followed for a minimum of 8 weeks.

3.4 Inclusion of Women and Minorities

NIH policy requires that women and members of minority groups and their subpopulations be included in all NIH-supported biomedical and behavioral research projects involving NIH-defined clinical research unless a clear and compelling rationale and justification establishes to the satisfaction of the funding Institute & Center (IC) Director that inclusion is inappropriate with respect to the health of the subjects or the purpose of the research. Exclusion under other circumstances must be designated by the Director, NIH, upon the recommendation of an IC Director based on a compelling rationale and justification. Cost is not an acceptable reason for exclusion except when the study would duplicate data from other sources. Women of childbearing potential should not be routinely excluded from participation in clinical research. Please see http://grants.nih.gov/grants/funding/phs398/phs398.pdf.

Both men and women of all races and ethnic groups are eligible for this trial. Please see table in section 13.2 on planned distribution of subjects by sex/gender, race, and ethnicity.

4. REGISTRATION PROCEDURES

4.1 Investigator and Research Associate Registration with CTEP

Food and Drug Administration (FDA) regulations and National Cancer Institute (NCI) policy require all individuals contributing to NCI-sponsored trials to register and to renew their registration annually. To register, all individuals must obtain a Cancer Therapy Evaluation Program (CTEP) Identity and Access Management (IAM) account

(https://ctepcore.nci.nih.gov/iam). In addition, persons with a registration type of Investigator (IVR), Non-Physician Investigator (NPIVR), or Associate Plus (AP) (i.e., clinical site staff requiring write access to OPEN or RAVE or acting as a primary site contact) must complete their annual registration using CTEP's web-based Registration and Credential Repository (RCR) (https://ctepcore.nci.nih.gov/rcr). Documentation requirements per registration type are outlined in the table below.

Documentation Required	**IVR**	**NPIVR**	**AP**	**A**
FDA Form 1572	✔	✔		
Financial Disclosure Form	✔	✔	✔	
NCI Biosketch (education, training, employment, license, and certification)	✔	✔	✔	
HSP/GCP training	✔	✔	✔	
Agent Shipment Form (if applicable)	✔			
CV (optional)	✔	✔	✔	

An active CTEP-IAM user account and appropriate RCR registration is required to access all CTEP and CTSU (Cancer Trials Support Unit) websites and applications. In addition, IVRs and NPIVRs must list all clinical practice sites and IRBs covering their practice sites on the FDA Form 1572 in RCR to allow the following:

- Added to a site roster
- Assigned the treating, credit, consenting, or drug shipment (IVR only) tasks in OPEN
- Act as the site-protocol PI on the IRB approval
- Assigned the Clinical Investigator (CI) role on the Delegation of Tasks Log (DTL).

Additional information can be found on the CTEP website at https://ctep.cancer.gov/investigatorResources/default.htm. For questions, please contact the RCR ***Help Desk*** by email at < RCRHelpDesk@nih.gov >.

4.2 Site Registration

Each investigator or group of investigators at a clinical site must obtain IRB approval for this protocol and submit IRB approval and supporting documentation to the CTSU Regulatory Office before they can be approved to enroll patients. Assignment of site registration status in the CTSU Regulatory Support System (RSS) uses extensive data to make a determination of whether a site has fulfilled all regulatory criteria including but not limited to the following:

- An active Federal Wide Assurance (FWA) number
- An active roster affiliation with the Lead Network or a participating organization
- A valid IRB approval
- Compliance with all protocol specific requirements.

In addition, the site-protocol Principal Investigator (PI) must meet the following criteria:

- Active registration status
- The IRB number of the site IRB of record listed on their Form FDA 1572
- An active status on a participating roster at the registering site

Sites participating on the NCI CIRB initiative that are approved by the CIRB for this study are not required to submit IRB approval documentation to the CTSU Regulatory Office. For sites using the CIRB, IRB approval information is received from the CIRB and applied to the RSS in an automated process. Signatory Institutions must submit a Study Specific Worksheet for Local Context (SSW) to the CIRB via IRBManager to indicate their intent to open the study locally. The CIRB's approval of the SSW is then communicated to the CTSU Regulatory Office. In order for the SSW approval to be processed, the Signatory Institution must inform the CTSU which CIRB-approved institutions aligned with the Signatory Institution are participating in the study.

4.2.1 Downloading Regulatory Documents

Site registration forms may be downloaded from the *10026* protocol page located on the CTSU Web site. Permission to view and download this protocol is restricted and is based on person and site roster data housed in the CTSU RSS. To participate, Investigators and Associates must be associated with the Corresponding or Participating protocol organization in the RSS.

- Go to https://www.ctsu.org and log in using your CTEP-IAM username and password.
- Click on the Protocols tab in the upper left of your screen.
- Either enter the protocol # in the search field at the top of the protocol tree, or

- Click on the By Lead Organization folder to expand, then select LAO-MA036 and protocol *#10026.*
- Click on LPO Documents, select the Site Registration documents link, and download and complete the forms provided. (Note: For sites under the CIRB initiative, IRB data will load to RSS as described above)

4.2.2 Requirements for 10026 Site Registration

- A screening request form must be completed by the site and sent to the lead site for approval and acknowledgement prior to consenting a patient.
- IRB approval (For sites not participating via the NCI CIRB; local IRB documentation, an IRB-signed CTSU IRB Certification Form, Protocol of Human Subjects Assurance Identification/IRB Certification/Declaration of Exemption Form, or combination is accepted)
- Site Initiation Visit (SIV Report) must be uploaded to the CTSU Regulatory Office prior to enrolling patients

4.2.3 Submitting Regulatory Documents

Submit required forms and documents to the CTSU Regulatory Office, where they will be entered and tracked in the CTSU RSS.

Regulatory Submission Portal: www.ctsu.org (members' area) → Regulatory Tab →Regulatory Submission

When applicable, original documents should be mailed to:

CTSU Regulatory Office

1818 Market Street, Suite 3000

Philadelphia, PA 19103

Institutions with patients waiting that are unable to use the Portal should alert the CTSU Regulatory Office immediately at 1-866-651-2878 in order to receive further instruction and support.

4.2.4 Checking Site Registration Status

You can verify your site registration status on the members' section of the CTSU website.

- Go to https://www.ctsu.org and log in to the members' area using your CTEP-IAM

username and password

- Click on the Regulatory tab at the top of your screen
- Click on the Site Registration tab
- Enter your 5-character CTEP Institution Code and click on Go

Note: The status given only reflects compliance with IRB documentation and institutional compliance with protocol-specific requirements as outlined by the Lead Network. It does not reflect compliance with protocol requirements for individuals participating on the protocol or the enrolling investigator's status with the NCI or their affiliated networks.

4.3 Patient Registration

4.3.1 Obtaining Lead Site/Coordinating Center Approval Prior to Registration

To obtain approval to enroll a participant, the following documents should be completed by the external site research nurse or data manager and e-mailed to the Research Project Manager of the lead site (Dana Farber Cancer Institute)

□ Copy of labs and clinical information that satisfy inclusion criteria

□ Signed participant consent form

□ HIPAA authorization form (if applicable)

□ Registration Form

□ Completed eligibility checklist

To complete the registration process, the Research Project Manager at the lead site will follow DF/HCC Standard Operating Procedure for Human Subject Research titled

Subject Protocol Registration (SOP# REGIST-101) and send an email to the external site

confirming that the patient is cleared to be enrolled into OPEN/IWRS for formal registration.

4.3.2

- OPEN / IWRS

Patient enrollment will be facilitated using the Oncology Patient Enrollment Network (OPEN). OPEN is a web-based registration system available to users on a 24/7 basis. It is integrated with the CTSU Enterprise System for regulatory and roster data interchange and with the Theradex Interactive Web Response System (IWRS) for retrieval of patient registration/randomization assignment. Patient enrollment data entered by Registrars in OPEN / IWRS will automatically transfer to the NCI's clinical data management system, Medidata Rave.

For trials with slot reservation requirements, OPEN will connect to IWRS at enrollment

initiation to check slot availability. Registration staff should ensure that a slot is available and secured for the patient before completing an enrollment.

The OPEN system will provide the site with a printable confirmation of registration and treatment information. Please print this confirmation for your records.

- OPEN/IWRS User Requirements

OPEN/IWRS users must meet the following requirements:

- Have a valid CTEP-IAM account (*i.e.*, CTEP username and password).
- To enroll patients or request slot reservations: Be on an ETCTN Corresponding or Participating Organization roster with the role of Registrar. Registrars must hold a minimum of an AP registration type
- To approve slot reservations or access cohort management: Be identified to Theradex as the "Client Admin" for the study.
- Have regulatory approval for the conduct of the study at their site.

Prior to accessing OPEN/IWRS, site staff should verify the following:

- All eligibility criteria have been met within the protocol stated timeframes.
- If applicable, all patients have signed an appropriate consent form and HIPAA authorization form.

- OPEN/IWRS Questions?

Further instructional information on OPEN is provided on the OPEN tab of the CTSU website at https://www.ctsu.org or at https://open.ctsu.org. For any additional questions contact the CTSU Help Desk at 1-888-823-5923 or ctsucontact@westat.com.

Theradex has developed a Slot Reservations and Cohort Management User Guide, which is available on the Theradex website:
http://www.theradex.com/clinicalTechnologies/?National-Cancer-Institute-NCI-11
This link to the Theradex website is also on the CTSU website OPEN tab. For questions about the use of IWRS for slot reservations, contact the Theradex Helpdesk: 609-619-7802 or Theradex main number 609-799-7580; CTMSSupport@theradex.com.

4.4 General Guidelines

Following registration, patients should begin protocol treatment as soon as possible (within 5 days of enrollment). Issues that would cause treatment delays should be discussed with the Principal Investigator. If a patient does not receive protocol therapy following registration, the patient's registration on the study may be canceled. The Study Coordinator should be notified of cancellations as soon as possible.

5. TREATMENT PLAN

5.1 Agent Administration

Treatment will be administered on an outpatient basis. Reported adverse events and potential risks are described in Section 7. Appropriate dose modifications are described in Section 6. No investigational or commercial agents or therapies other than those described below may be administered with the intent to treat the patient's malignancy, except for hydroxyurea. Prophylactic intrathecal chemotherapy is allowed if determined to be clinically necessary. Please see section 6.5 for details on IT chemotherapy prophylaxis.

This is a standard 3+3 dose escalation study with two patient cohorts, separated by transplant status (Arm A = post allo-HSCT; Arm B = transplant naive). Given the concern for potential toxicity in the post allo-HSCT arm (Arm A), we will first demonstrate safety at the first dose level (e.g. **Dose Level 0**; starting dose, see table in Section 5.1 below) with patients on Arm B (transplant naive cohort) prior to Arm A enrollment. If there is no DLT in the first 3 patients, then we will proceed to enrollment on Arm A at the first dose level. However, if there is a DLT event in the first 3 patients assessed in Arm B, then following 3+3 rules, an additional 3 patients (a total of 6) will be tested at the same dose level. If there are no additional DLTs within those 6 patients, then we will allow enrollment on Arm A.

After the first dose level is assessed for safety in Arm B, then subjects can be simultaneously enrolled to the appropriate arm with no further observation period or delay between the start of therapy for each enrolled patient in either arm. Overall, patients in Arm A and Arm B will be tested at the same three dose levels, as described in the table below (**section 5.1**).

In the event that 0 of 3 patients in a dose level experience a DLT, the dose level may still be expanded to 6 evaluable patients based on observed toxicity at the current dose level at the discretion of the overall PI to capture patient safety prior to expanding.

Dose Escalation Schedule for both Arm A and Arm B		
Dose Level	**Dose**	
	Ipilimumab (mg/kg) on day 1	Decitabine* (mg/m^2) on days 1-5
Level -1 (dose de-escalation)	1	20
Level 0 (starting dose)	3	20
Level 1	5	20
Level 2	10	20

*Decitabine is NOT dose-escalated and will be given at standard dosing of 20 mg/m2 IV daily x 5 consecutive doses on days 1-5 of a 28-day cycle

Treatment Overview

- Each cycle is 28 days in length

- There will be three phases: priming, induction and maintenance
- Hydroxyurea may be used to control leukocytosis

- **Lead In: Priming phase with decitabine alone**
  - **Treatment**: Decitabine 20 mg/m2 IV daily over 60 minutes (+/- 15 minutes) x 5 consecutive doses on **days 1-5** given per institutional guidelines
    - **Pre-medication prior to chemotherapy:** per institutional standards
    - Supportive medication such as allopurinol or equivalent should be considered in patients with white blood cell counts > 10 x 10^9 /L to reduce risk of tumor lysis syndrome.
  - Clinic visits on day 1 and day 15 (+/- 3 days)
  - Labs on day 1 and day 15 (+/- 3 days)
  - Research blood and aspirate collection at the end of the lead in period on a day between days 24-28 (see correlatives calendar in section 10.2).
  - Adverse event evaluation:
    - **Intolerability during priming phase**: If a patient experiences unexpected intolerability to decitabine during the lead in that cannot be ameliorated by supportive care measures, then that patient will not continue on trial and will be replaced.
    - **Graft versus host disease** (GVHD): If there is evidence of acute GVHD that requires the use of systemic corticosteroids agents during the lead in, then the patient will be taken off study due to concern for safety with checkpoint blockade and replaced.
- **Cycles 1-4: Induction phase with combination decitabine and ipilimumab**
  - **Pre-Treatment Requirements**
    - Hematologic criteria to start cycle 1: None required
    - Hematologic criteria to start cycle 2:
      - If there is persistent disease (e.g. peripheral blasts, any marrow disease, persistent leukemia cutis or other form of extramedullary disease) at the time of disease response evaluation at the end of cycle 1, then there are no hematologic requirements to start cycle.
      - In the absence of disease, patients must have an absolute neutrophil count ≥ 500 x 10^9/L or platelet count to ≥ 25,000 x 10^9/L to proceed; otherwise administration of treatment will be delayed.
    - Hematologic criteria to start cycle 3:
      - If there is persistent disease (e.g. peripheral blasts, any marrow disease, persistent leukemia cutis or other form of extramedullary disease) at the time of disease response evaluation at the end of cycle 2, then there are no hematologic requirements to start cycle.
      - In the absence of disease, patients must have an absolute neutrophil count ≥ 500 x 10^9/L or platelet count to ≥ 25,000 x 10^9/L to proceed.
    - Hematologic criteria to start cycle 4:
      - If there is persistent disease (e.g. peripheral blasts, any marrow disease, persistent leukemia cutis or other form of extramedullary

disease), then there are no hematologic requirements to start cycle .
- In the absence of disease, patients must have an absolute neutrophil count ≥ 500 x 10^9/L or platelet count to ≥ 25,000 x 10^9/L to proceed; otherwise administration of treatment will be delayed.

- **Treatment**
  - Decitabine 20 mg/m2 IV daily over 60 minutes (+/- 15 minutes) x 5 consecutive doses on **days 1-5** every 4 weeks given per institutional guidelines
    - **Pre-medication prior to chemotherapy:** per institutional standards
  - Ipilimumab IV (per dose level as in table above) on **day 1** over 90 minutes (+/- 15 minutes) every 4 weeks for a total of 4 doses during induction
    - Dose escalation levels: 3 mg/kg (**starting dose**, dose level 0), 5 mg/kg (dose level 1), and 10 mg/kg (dose level 2)
    - Dose de-escalation level: 1 mg/kg
  - Decitabine will be dosed prior to ipilimumab on day 1
  - Vitals signs will be repeated hourly from time of ipilimumab infusion for up to two hours after infusion ends (+/- 30 minutes) on day 1 after first dose and up to one hour after infusion ends (+/- 30 minutes) on day 1 for subsequent doses.
- Clinic visits (including physical exam, medication review, ECOG and AE review)
  - Cycle 1: on day 1 and day 15 (+/- 3 days)
  - Cycles 2-4: on day 1 (+/- 3 days).
- **Procedures:**
  - Labs
    - Cycle 1: on day 1, day 15 (+/- 3 days).
    - Cycles 2-4: on day 1 (+/- 3 days).
    - End of cycles 1, 2 and 4 on day of bone marrow biopsy (on a day between days 24-28).
  - Bone marrow biopsy (including aspirate (if aspirable), core, flow cytometry, cytogenetics) will be at the end of cycles 1, 2 and 4 (on a day between days 24-28) for response assessment.
  - If sole extramedullary disease (unless leukemia cutis), routine evaluation may include PET imaging (if FDG avid at time of diagnosis) at the end of cycles 2 and 4 (on a day between days 24-28) for response assessment.
- **Correlative studies:** peripheral blood and aspirate collection at the end of cycles 1, 2 and 4 on a day between days 24-28 (see correlatives calendar in section 10.2).
- The DLT window will be the first 8 weeks of combination treatment with decitabine and ipilimumab (starting day 1 of cycle 1).
- At the time of response assessments, patients with evidence of disease progression will come off study unless they have evidence of treatment tolerability and clinical benefit (i.e. improvement in transfusion frequency or peripheral blast reduction or symptom improvement). Patients with evidence of disease progression may be allowed to stay on study if deriving clinical benefit with approval from the overall PI as there may be a delayed response with

immunotherapy.
- Patients with stable disease or better at the end of cycle 4 will move forward with treatment in the maintenance phase.

- **Cycles 5-12: Maintenance phase with combination decitabine and ipilimumab**
  - Pre-treatment Criteria
    - In the absence of disease, patients should have an absolute neutrophil count ≥ 500 x 10^9/L or platelet count to ≥ 25,000 x 10^9/L to proceed with each cycle. Dose delay recommendations are provided in section 6.
    - If there is still disease (partial or stable) then no hematologic requirements are required to start cycle
  - **Treatment**
    - Decitabine 20 mg/m2 IV daily over 60 minutes (+/- 15 minutes) x 5 consecutive doses on **days 1-5** every 4 weeks given per institutional guidelines
      - **Pre-medication prior to chemotherapy:** per institutional standards
    - Ipilimumab IV (per dose level as in table above) on **day 1** over 90 minutes (+/- 15 minutes) every **8 weeks** for a total of 4 doses during maintenance
    - Decitabine will be dosed prior to ipilimumab on day 1
    - Vitals signs will be repeated hourly from time of ipilimumab infusion for up to one hour after infusion ends (+/- 30 minutes) on day 1.

  - Clinic visits (including physical exam, medication review, ECOG and AE review)
    - Cycles 5-12: on day 1 (+/- 3 days).
  - **Procedures:**
    - Labs
      - Cycles 5-12: on day 1 (+/- 3 days).
      - At the end of cycles 7 and 10 on a day between days 24-28 and at end of study on a day between days 24-28.
    - Bone marrow biopsy (including aspirate (if aspirable), core, flow cytometry, cytogenetics) will be at the end of cycles 7, 10 and at end of study (on a day between days 24-28) for response assessment.
  - **Correlative studies:** peripheral blood and aspirate collection at the end of cycles 7 and 10 on a day between days 24-28 and at the end of study (see correlatives calendar in section 10.2).
  - At the time of response assessments, patients with evidence of disease progression will come off study unless they have evidence of clinical benefit (i.e. improvement in transfusion frequency or peripheral blast reduction). Patients with evidence of disease progression may be allowed to stay on study if deriving clinical benefit with approval from the overall PI as there may be a delayed response with immunotherapy.
  - Additional maintenance dosing (following the same schedule of ipilimumab dosed on day 1 every other cycle with monthly decitabine) for patients with clinical benefit at completion of maintenance therapy will be considered with Overall PI until progression or unacceptable toxicity.

Regimen Description					
Agent	**Premedications; Precautions**	**Dose**	**Route**	**Schedule**	**Cycle Length**
Decitabine	Anti-emetic agent per institutional guidelines (i.e. compazine 10 mg injection or equivalent as slow push at a rate not to exceed 5 mg/min per institutional standards given 30 minutes [+/- 15 minutes] prior to chemotherapy)	20 mg/m^2 (reconstituted and mixed per institutional guidelines)	Recommend giving decitabine first prior to ipilimumab. IV over 60 minutes [+/- 15 minutes].	Days 1-5 every 4 weeks	28 days (4 weeks)
Ipilimumab	None unless history of prior reaction Please see section 6.3 for recommendations for hypersensitivity precautions	1, 3 (starting dose level), 5, or 10 mg/kg	IV over 90 minutes (not bolus or IV push) [+/- 15 minutes]. Administer after decitabine infusion is completed (any time within four hours after infusion completion).	Day 1* every 4 weeks during induction and every 8 weeks during maintenance	
*Ipilimumab does not start until cycle 1					

Please also refer to section 6 for dose delay guidelines.

5.1.1 CTEP IND Agent: Ipilimumab

Unit: Ipilimumab 5 mg/mL, in 40-mL vials

Route: Intravenous infusion

Dosing: Treatment doses of both decitabine and ipilimumab will be based on the participant's weight obtained on day 1 of each cycle. Dose levels include: 1 mg/kg (dose level -1, dose de-escalation), 3 mg/kg (dose level 0, starting dose, FDA-approved dose), 5 mg/kg (dose level 1), and 10 mg/kg (dose level 2, FDA-approved dose).

Appearance: Clear, colorless solution

5.1.2 Decitabine

Administration: Per institutional guidelines.. Patients will be dosed for 5 consecutive days unless there is a clinical reason to hold the drug as described in section 6 of the protocol

Dosing: On days when both drugs are required, decitabine should be administered prior to ipilimumab. Treatment doses of both decitabine and ipilimumab will be based on the participant's weight obtained on day 1 of each cycle.

Prophylaxis: No prophylaxis is required however anti-viral therapy (i.e. acyclovir 400 mg po three times a day or equivalent) is common practice.

Supportive care regimens: Prior to administration of chemotherapy (decitabine) recommend an anti-emetic agent to be given 30 minutes (+/- 15 minutes) prior to chemotherapy to increase tolerability. Anti-emetic agent should be per institutional guidelines.

5.2 Definition of Dose-Limiting Toxicity

DLT for dose-escalation purposes will be determined during the first 8 weeks of combination therapy (56 days from start of combination, starting cycle 1 day 1 of ipilimumab and decitabine) during the induction phase only. A DLT will be defined as any of the following toxicities that are determined to be *at least possibly* related to treatment with combination ipilimumab and decitabine and not related to underlying disease or intercurrent illness or to decitabine alone. Adverse events that occur **after** the first 8 weeks will also be evaluated by the investigator and may be considered dose limiting depending on the clinical scenario. Events will be graded by CTCAE v5.0 criteria.

DLTs include:

- Any study treatment related death (not due to disease or intercurrent illness).
- Acute GVHD that is an overall grade III or higher (see Appendix B)
    - Exception: For skin GVHD alone, must be an overall grade II with at least stage 3 involvement or higher to be considered a DLT (skin GVHD that is stage 1-2 will not be considered a DLT).
- Grade 3 or higher non-hematologic toxicity
    - Exceptions:
        - Fatigue, nausea, weight loss, electrolyte abnormalities managed by replacement or correction, endocrine abnormalities managed by replacement therapy, skin rash, and those attributable to GVHD (as above)
        - Infections will be considered a DLT if grade 4 or higher or grade 3 that does not recover to grade 1 within 4 weeks
- Admission and IV antibiotics for neutropenic fever without sepsis or severe complication will not be considered a DLT (this is a frequent complication of underlying disease and of decitabine therapy)
- Grade 3 immune-related adverse events (e.g. diarrhea, pruritus, rash, endocrinopathies) that respond to corticosteroids and improve to grade 1 or less within 4 weeks will NOT count as DLTs. Optional biopsy of affected organs will be encouraged to help determine whether the immune-related condition is due to direct effects of ipilimumab versus GVHD.
- Grade 3 ipilimumab infusion reactions; a second consecutive occurrence or grade 4 or high infusion reaction will be considered a DLT.
- Grade 4 or higher hematologic toxicity will be considered a DLT if patients do not recover their absolute neutrophil count to $\geq 500 \times 10^9$/L or platelet count to $\geq 25{,}000 \times 10^9$/L, at the end of the DLT window, with a hypocellular marrow and absence of

underlying disease.

Management and dose delays associated with the above adverse events are outlined in Section 6.

Dose escalation will proceed within each cohort according to the following scheme. Dose-limiting toxicity (DLT) is defined above.

For patients who are removed from treatment because of a > 4 week delay in receiving the next dose of ipilimumab: this will be classified as a DLT only if AE resulting in the treatment delay qualifies as a DLT.

Number of Patients with DLT at a Given Dose Level	Escalation Decision Rule
0 out of 3	Enter 3 patients at the next dose level.
1 out of 3	Enter at least 3 more patients at this dose level. • If 0 of these 3 patients experience DLT, proceed to the next dose level. • If 1 or more of this group experience DLT, then dose escalation is stopped, and this dose is declared the maximally administered dose. Three (3) additional patients will be entered at the next lowest dose level if only 3 patients were treated previously at that dose.
≥2	Dose escalation will be stopped. This dose level will be declared the maximally administered dose (highest dose administered). Three (3) additional patients will be entered at the next lowest dose level if only 3 patients were treated previously at that dose.
≤1 out of 6 at highest dose level below the maximally administered dose	This is generally the recommended phase 2 dose. At least 6 patients must be entered at the recommended phase 2 dose.

Delayed toxicities (toxicities after 8 weeks of combination therapy): As delayed toxicity may occur (immune related AE) at any point after checkpoint blockade therapy, we recommend that any event beyond the first 8 weeks of combination therapy that is considered to be an immune related AE or concerning for GVHD be reported to the Overall PI. These events will be captured and reported, but will not determine dose-escalation. These patients would also be removed from study if they have a toxicity event described in section 5.2.

5.3 Dose Expansion Cohorts

Once the RP2D is reached, an additional 6-12 patients will be treated at this dose per arm. For a given arm, if all 3 dose cohorts are filled to 6 patients, then a 6 patient expansion will be added. If 2 or fewer dose cohorts are filled to 6 patients, then a 12 patient expansion will be added so that no more than 24 patients (dose escalation + expansion) will be treated on a given arm.

For the expansion cohort, patients will continue to be monitored for occurrence of DLT. If ≥2 of 6 patients experience DLT, the Principal Investigator will discuss with all study investigators and with CTEP whether further addition of patients is needed to re-assess the RP2D. Monitoring of all safety and toxicity data is done by the Principal Investigator and the Corresponding Organization on a real-time basis as data are entered into Medidata Rave using the Web Reporting Module. All participating sites are expected to notify the Principal Investigator when a DLT has occurred.

5.4 General Concomitant Medication and Supportive Care Guidelines

Growth factor support is not routinely recommended for low blood counts unless there is a concern for severe sepsis, septic shock or persistent infection (e.g. fungal infection) per institutional standards. Transfusion support is allowed, and can be used at the discretion of the investigators. Prophylactic antibiotics can be utilized as appropriate for disease or as needed in the post allo-HCT setting at the discretion of the investigators.

Suggested hypersensitivity precautions with ipilimumab: In the event of a hypersensitivity reaction, stop infusion, notify MD/NP/respiratory and pharmacy, administer oxygen as needed, monitor vital signs and proceed with administering hypersensitivity medications as needed. Medications to consider having available for hypersensitivity precautions:

Suggested medications for hypersensitivity precautions:

- Diphenhydramine 25-50mg IV as needed (begin with 25 mg, if continued reaction then administer additional 25 mg)
- Methylprednisolone or equivalent 40mg IV once as needed,
- Meperidine injection 12.5-25mg IV as needed for rigor/fever and/or chills during hypersensitivity reaction (start with 12.5 mg). If patient has continued reaction, administer additional 12.5 mg.
- Acetaminophen tablet 650 mg (oral) as needed for fever; hold if given already as a premedication
- Famotidine 20mg/50mL in NS IVPB or equivalent as needed for hypersensitivity; hold if already given as premedication
- Epinephrine IM injection 0.3mg as needed for anaphylaxis; may administer every 5 minutes up to 3 doses)
- Please refer to institution specific hypersensitivity reaction treatment algorithm for more clinical support

5.5 Duration of Therapy

Duration of therapy will depend on individual response, evidence of progression, and tolerance. In the absence of treatment delays due to adverse events, treatment (including priming, induction and maintenance) may continue for up to 12 cycles of combination therapy or until one of the following criteria applies:

- Disease progression (as defined by ELN 2017 criteria for AML and by IWG criteria for MDS). As T-cell infiltration of the tumor can look like progressive disease, please

discuss the results of radiographic assessments (for extramedullary disease) showing progressive disease with the overall PI before removing a patient from the study for disease progression.

- Intercurrent illness that prevents further administration of treatment,
- Unacceptable adverse event(s) requiring permanent stopping of both ipilimumab and decitabine
- Patient decides to withdraw from the study
- Pregnancy
    - All women of child bearing potential should be instructed to contact the investigator immediately if they suspect they might be pregnant (e.g., missed or late menstrual period) at any time during study participation.
    - The investigator must immediately notify CTEP in the event of a confirmed pregnancy in a patient participating in the study.
- General or specific changes in the patient's condition render the patient unacceptable for further treatment in the judgment of the investigator.
- Termination of the study by sponsor.

Ipilimumab is expected to trigger immune-mediated responses, which require activation of the immune system prior to the observation of clinical responses. Such immune activation may take weeks to months to be evident. For patients with a mass, some patients may have objective volume increase of tumor lesions or other disease parameters (based on study indication, i.e., hematologic malignancies) within (specific 12-24) weeks following the start of ipilimumab dosing. Such patients may not have had sufficient time to develop the required immune activation or, in some patients, tumor volume or other disease parameter increases may represent infiltration of lymphocytes into the original tumor or blood. In conventional studies, such tumor volume or relevant laboratory parameter increases during the first 12 weeks of the study would constitute disease progression and lead to discontinuation of imaging to detect response, thus disregarding the potential for subsequent immune-mediated clinical response. For the purposes of this clinical trial with AML and MDS, patients with sole extramedullary disease with evidence of tumor progression by PET imaging at the end of cycle 2 but without rapid clinical deterioration or change in performance status who do not require additional immediate therapy, may continue to be treated with ipilimumab and clinically observed until the next response evaluation (in this scenario, would then consider repeating a PET scan at the end of cycle 2) to allow detection of a subsequent tumor response.

The reason(s) for protocol therapy discontinuation, the reason(s) for study removal, and the corresponding dates must be documented in the Case Report Form (CRF).

5.6 Duration of Follow Up

Patients will be followed for up to 52 weeks (1 year) after removal from study or until death, whichever occurs first. Patients removed from study for unacceptable adverse event(s) will be followed until resolution or stabilization of the adverse event. Patients will be followed every 3 months by phone after removal from study.

6. DOSING DELAYS/DOSE MODIFICATIONS

Summary of dose delays and modifications allowed on study (please see sections below for detail):

- Dose delay or modification of decitabine is allowed for patients diagnosed with MDS (per institutional guidelines) as described in section 6.1. If a patient with MDS is in a CR, due for both decitabine and ipilimumab and a dose delay of decitabine is recommended, then decitabine and ipilimumab can be delayed so that they can be dosed together.
- Dose delay or modification of decitabine is allowed for patients diagnosed with AML in absence of disease if there are severe cytopenias after discussion with overall PI. Please see pre-dose requirements in section 5.1.
- Holding ipilimumab is allowed based on recommendations in section 6.2.
- If ipilimumab is held per recommendations in section 6.2, decitabine can continue alone. Ipilimumab may be re-introduced upon ipilimumab-AE resolution after discussion with overall PI. Delayed benefit from checkpoint blockade may occur.
- Participants may delay ipilimumab dosing beyond 8 weeks after discussion and approval by the overall PI (each clinical situation will be reviewed on case by case basis).
- Missed doses of ipilimumab are not given later.

6.1 Dose Delay Recommendations for Patients with Suspected Decitabine Related Toxicity

If the toxicity is thought to be due to decitabine:

If toxicity is thought to be due to decitabine alone, then ipilimumab dosing can be continued as monotherapy. This decision should be made with the overall PI.

- **AML:** Dose modification is not recommended for patients with AML. For cycle 4 on-ward, if there is evidence of marrow hypocellularity with <5% blasts, then can consider delaying treatment cycle for up to 14 days, in absence of disease.

- **MDS:** Dose delay can be considered (not mandatory) in patients with MDS per FDA package label after evidence of marrow remission or better. Any other treatment delays must be discussed with the Overall PI.

  Patients that have grade 3 or higher thrombocytopenia (50,000/mm3 or less) OR grade 3 or higher neutropenia (1000/mm3 or less) at the time of screening are not recommended for dose reduction until after response evaluation at the end of cycle 2. If there is evidence of marrow remission or better for patients with MDS on study, then dose delay may be considered for subsequent cycles per institutional guidelines. If there is evidence of persistent disease, then would not recommend delaying treatment unless thought to be

due to decitabine (and not disease).

6.2 Dose Delay Recommendations for Patients with Suspected Ipilimumab Toxicity or Combination Therapy

Intra-patient dose modification (reductions or increases) of ipilimumab is not allowed in this study.

Decisions to delay an ipilimumab dose must be made on specified safety criteria. Treatment with ipilimumab will be delayed or discontinued if the subject experiences at least one adverse event, specified below, considered by the investigator to be **"possibly," probably," or "definitely" related to ipilimumab treatment.**

Any adverse event that will prompt a delayed dose or discontinuation of ipilimumab must be reported. All AEs that require dose holding (for ipilimumab) will be reviewed during regularly scheduled conference call with the PI and co-investigators. Calls will be planned at a minimum of every 4-6 weeks. A representative from the site (co-investigator, research nurse, or clinical trial coordinator) will be required to be present on the call. **The investigator should contact the study PI or drug monitor to discuss any questions that may arise.**

Doses of ipilimumab will be referred to as dose #1, dose #2, dose #3, or dose #4 during the induction phase and dose #5, dose #6, dose #7 or dose #8 during the maintenance phase. The induction phase will include the first four doses of ipilimumab and the maintenance phase will include the last four doses of ipilimumab. If there are any ipilimumab dose delays, the cycle and day will not change. The "study cycle" number is determined by the cycle of decitabine that the patient is on (with "cycle 1" starting as the first cycle of COMBINATION with ipilimumab; the priming phase of decitabine monotherapy could be considered "cycle 0" for purposes of this study). If there is a delay in ipilimumab dosing, decitabine may be continued as scheduled. Disease response assessments will be as scheduled regardless of how many doses of ipilimumab is administered. As an example, if dose #2 of ipilimumab (which would be due on cycle 2 day 1) is delayed for four weeks due to an ipilimumab related toxicity, decitabine can still be administered on cycle 2 days 1-5 and the scheduled disease response assessment will still occur at the end of cycle 2. If the ipilimumab related toxicity has resolved by cycle 3 day 1, then dose #2 of ipilimumab will be given then concurrent with decitabine. Continuing decitabine will help to maintain disease control given the proliferative nature of AML. We will capture how many doses of ipilimumab that patients are able to receive in this study.

Delay window: If ipilimumab dosing must be delayed for > 4 weeks +/- 3 days for holiday or scheduling logistics (more than 8 weeks [+/- 3 days] since the prior dose administration), then patients must come off treatment. For example, if patient cannot receive ipilimumab on cycle 2 day 1 due to development of grade 2 colitis, then the patient's dose will be delayed to cycle 3 day 1. If the patient cannot be dosed on cycle 3 day 1 (and it has now been more than 8 weeks since last dose), then the patient must come off treatment.

Exceptions may be made to hold ipilimumab for > 8 weeks after discussion with overall PI for grade 2 or lower ipi-induced immune related AEs and GVHD events that are less than grade III.

Patients may remain on decitabine alone until ipilimumab can be re-introduced if patients demonstrate continued benefit and resolution of suspected ipilimumab-induced AE to grade 1 or baseline (skin GVHD can be controlled with topical steroids).

Suspicion for graft versus host disease (GVHD): If there is a concern for GVHD, then we recommend consideration of a biopsy for diagnostic work-up if clinically appropriate. Evidence of grade III or higher acute GVHD would be considered a DLT (except for skin GVHD, must have at least stage 3 involvement to be a DLT) as this often triggers systemic corticosteroids. These patients would stop receiving study treatment (both ipilimumab and decitabine), and followed for up to 1 year after treatment for AE resolution, response and survival. Those with skin GVHD that is overall grade II with stage 1-2 involvement or less will be able to stay on study. Topical corticosteroids are allowed. For GVHD events that are less than grade III, ipilimumab dose can be skipped (decitabine can continue) until event resolution or when the event is controlled without requiring systemic corticosteroids or immunosuppressive agents (exception: topical steroids) (missed ipilimumab doses will not be made up); these patients may remain on study. Please see Appendix B for resources to be used for acute and chronic GVHD grading.

It is necessary to delay ipilimumab dosing for the following related adverse events:

- Any ≥ Grade 2 non-skin related adverse event (including immune related AE (irAEs)), except for asymptomatic laboratory abnormalities, grade 2-3 fatigue, and grade 2 anorexia (as the latter two events can be caused by disease alone). Treatment does not need to be delayed for controlled (on treatment) or recently treated infections.
- Any ≥ Grade 3 non-hematologic laboratory abnormality except for grade 3 fatigue (as can be caused by disease). Treatment does not need to be delayed for controlled (on treatment) or recently treated infections.
- Any adverse event, laboratory abnormality or intercurrent illness that, in the judgment of the investigator, presents a substantial clinical risk to the subject with continued dosing
- Any ≥ Grade 3 skin related adverse event (unless considered to be related to underlying disease, leukemia cutis, or a drug-allergy that is not due to study drug (ie antibiotics))
- An event that is not considered to be necessarily related to ipilimumab but requires a brief course of corticosteroid therapy.
    - An example, if a patient develops Sweet's syndrome (febrile neutrophilic dermatosis, confirmed by skin biopsy), then prednisone would be warranted. Patients would be allowed to continue decitabine to maintain disease control.
- An event that is considered to be due to ipilimumab, such as immune related AEs that are grade 2 or less can receive corticosteroids if deemed clinically necessary so as long as they are tapered off by next dose. If ipilimumab dosing must be delayed for > 4 weeks (+/- 3 days for holiday or scheduling logistics) (more than 8 weeks since the prior dose administration) for any of the above reasons, patients must come off treatment.
- Decitabine may continue if AE considered to be due to ipilimumab

If there is a dose delay in ipilimumab:

For any dose delay, patients must re-meet laboratory criteria (including complete blood count with differential and platelets, comprehensive metabolic profile including liver enzymes) to be treated again on day of therapy (prior to dosing).

Resuming ipilimumab:

- Should coincide with day 1 of treatment cycle.
- If the adverse event is suspected to be due to ipilimumab, then the AE must resolve to ≤ grade 1 severity or returns to baseline within 4 weeks of initiating the dose delay (no more than 8 weeks since the most recent prior dose administration). This includes immune-related AE. Patients must be tapered off prednisone prior to resuming ipilimumab.
- If the *adverse event has resolved to* **≤ grade 1***,* ipilimumab dosing may be restarted at the next scheduled time point per protocol. Please see suggested guidelines for management of specific immune related adverse events (see Appendix C). Please note that re-initiating treatment may be associated with recurrence or exacerbation of autoimmune or inflammatory events. In some instances clinical resolution of events such as colitis may be associated with residual pathologic changes and should require evaluation of complete resolution prior to restarting therapy.
- If ipilimumab dosing must be delayed for > 4 weeks (+/- 3 days for holiday or scheduling logistics) (more than 8 weeks [+/- 3 days) since the prior dose administration) for any of the above reasons, patients must come off treatment.
- Autoimmune/inflammatory events are presumably related to the mechanisms of action of ipilimumab and potentially to a therapeutic effect. The incidence and severity of these events may be dose related, but once initiated, there is no evidence that lowered doses can be administered without continued autoimmune activity and there has so far been no demonstrable benefit to continuing ipilimumab after an autoimmune event during the initial treatment. The significance and benefit of toxicity or continued treatment in the maintenance phase has not been determined. Typically no dose modification is used for ipilimumab.

Permanent Discontinuation of Ipilimumab for Related Adverse Events

If discontinuing ipilimumab, participants will have the option to continue decitabine alone on study if they are determined to be clinically benefitting from treatment at the investigator's discretion.

Permanently discontinue ipilimumab for any of the following:

- Grade III or higher acute GVHD.
- Persistent adverse reactions that requires holding more than 2 ipilimumab doses (exception: patients with a GVH event(s) that is determined to be less than a grade III who are benefitting from study treatment can continue on study after approval from overall PI).
- Any persistent grade 3 or 4 event (see exceptions below in section 6.2.1).
- Any immune related AE that is grade 3 or higher requiring systemic corticosteroids at a dose of 1 to 2 mg/kg/day of prednisone or equivalent. Immune related AEs that were

grade 2 or less are allowed to stay on study and receive corticosteroids if necessary so as long as they are tapered off by next scheduled dose of ipilimumab. They cannot be delayed by more than 4 weeks (meaning 8 weeks between doses), or they have to come off study.

- Severe or life-threatening adverse reactions, including any of the following:
  - Colitis with abdominal pain, fever, ileus, or peritoneal signs; increase in stool frequency (7 or more over baseline), stool incontinence, need for IV hydration for more than 24 hours, gastrointestinal hemorrhage, and gastrointestinal perforation
  - Aspartate aminotransferase (AST) or alanine aminotransferase (ALT) >5 times the upper limit of normal or total bilirubin >3 times the upper limit of normal
  - Stevens-Johnson syndrome, toxic epidermal necrolysis, or rash complicated by full thickness dermal ulceration, or necrotic, bullous, or hemorrhagic manifestations
  - Severe motor or sensory neuropathy, Guillain-Barré syndrome, or myasthenia gravis
  - Severe immune-mediated reactions involving any organ system (*e.g.*, nephritis, pneumonitis, pancreatitis, non-infectious myocarditis)
  - Immune-mediated ocular disease that is unresponsive to topical immunosuppressive therapy
  - Any adverse event, laboratory abnormality or intercurrent illness which, in the judgment of the investigator, presents organ specific injury and/or a substantial clinical risk to the patient with continued dosing.

The following neurological adverse event requires permanent discontinuation of ipilimumab and defines unacceptable neurotoxicity:

- Any motor neurologic toxicity >/= grade 3 regardless of causality
- Any >/= grade 3 treatment related sensory neurologic toxicity

Please refer to Appendix C and the Investigator Brochure for suggested specific treatment algorithms.

6.2.1 Exceptions to Permanent Discontinuation

- Potentially reversible inflammation (< grade 4), attributable to a local anti-tumor reaction and a potential therapeutic response. This includes inflammatory reactions at sites of tumor resections or in draining lymph nodes, or at sites suspicious for, but not diagnostic of metastasis.
- Hospitalization for neutropenic fever evaluation (this is a common adverse event of underlying disease and of decitabine).
- Hospitalization for ≤ grade 2 adverse events where the primary reason for hospitalization is to expedite the clinical work-up.
- Patients with the following conditions where in the investigator's opinion continuing study drug administration is justified based on the potential for continued clinical benefit:

- Patients treated with systemic steroids for less than 4 weeks without evidence of autoimmune disease requiring steroids treatment
- Grade 2 skin rash treated with topical steroids for less than 4 weeks
- Grade 2 Ocular toxicity that has completely responded to topical therapy .within 4 weeks
- Endocrinopathies where clinical symptoms are controlled with appropriate hormone replacement therapy. **Note:** Ipilimumab may not be restarted while the patient is being treated with systemic corticosteroids except for patients on stable doses of hormone replacement therapy such as hydrocortisone.

6.2.2 Immune-Related Adverse Events (irAEs)Reactions and Immune-mediated Adverse Reactions: Definition, Monitoring, and Treatment

These are suggested guidelines for the management of immune-related adverse events. For the purposes of this study, an immune-related adverse reaction is defined as an adverse reaction of unknown etiology associated with drug exposure and consistent with an immune phenomenon. Efforts should be made to rule out neoplastic, infectious, metabolic, toxin or other etiologic causes prior to labeling an event an irAEs. Serolo gic, immunologic, and histologic (biopsy) data should be used to support the diagnosis of an immune-related toxicity. Suspected immune-related adverse reactions must be documented on an AE or SAE form. Another term for an irAE is an immune-mediate adverse reaction, as it is termed in the Ipilimumab US Prescribing Information. Both terms may be used in this protocol document.

Patients should be informed of and carefully monitored for evidence of clinically significant systemic immune-mediated adverse reactions (e.g., systemic lupus erythematosus-like diseases) or organ-specific immune-mediated adverse reaction (e.g., rash, colitis, uveitis, hepatitis or thyroid disease). If an immune-mediated adverse reaction is noted, appropriate work-up (including biopsy if possible) should be performed, and steroid therapy may be considered if clinically necessary.

It is unknown if systemic corticosteroid therapy has an attenuating effect on ipilimumab activity. However, clinical anti-tumor responses have been maintained in patients treated with corticosteroids and discontinued from ipilimumab. If utilized, corticosteroid therapy should be individualized for each patient. Prior experience suggests that colitis manifested as ≥ grade 3 diarrhea requires corticosteroid treatment.

Suggested guidelines for specific immune-mediated adverse reactions are included in section 6.4 below, in Appendix C, and the package insert. These suggested guidelines should be utilized as clinically appropriate for the treatment of individual patients. Please contact the overall PI or drug monitor for any questions.

Overview of Management of Immune Related AE

Immune related AE include dermatologic, gastrointestinal, hepatic, endocrine and other less common inflammatory events that resolve from general immunologic enhancement. Temporary

immunosuppression with corticosteroids, tumor necrosis factor-alpha antagonists, mycophenolate mofetil, or other agents can be effect treatment in most cases.

For patients with moderate or severe immune related AE, this requires interruption of the ipilimumab and the use of corticosteroid immunosuppression. Treatment is based upon the severity of the observed toxicity:

- For patients with grade 2 (moderate) immune-mediated toxicities, treatment with ipilimumab should be withheld and should not be resumed until symptoms or toxicity is grade 1 or less. Corticosteroids (prednisone 0.5 mg/kg/day or equivalent) should be started if symptoms do not resolve within a week.
- For patients experiencing grade 3 or 4 (severe or life-threatening) immune-mediated toxicities, treatment with ipilimumab should be permanently discontinued. High doses of corticosteroids (prednisone 1 to 2 mg/kg/day or equivalent) should be given. When symptoms subside to grade 1 or less, steroids can be gradually tapered over at least one month.

6.3 Other Guidance for the Treatment of Ipilimumab Associated Reactions

The following guidance is provided for the management of ipilimumab treatment related events. These suggested guidelines, treatment algorithms in Appendix C, and further information in the investigator's brochure, should be considered in the context of appropriate medical treatment for each patient.

6.3.1 Treatment of Infusion Reactions Associated with Ipilimumab

Since ipilimumab contains only human protein sequences, it is less likely that any allergic reaction will be seen in patients. However, it is possible that infusion of ipilimumab will induce a cytokine release syndrome that could be evidenced by fever, chills, rigors, rash, pruritus, hypotension, hypertension, bronchospasm, or other symptoms. No prophylactic pre-medication should be given unless indicated by previous experience in an individual patient. Reactions should be treated based upon the following recommendations.

- For mild symptoms (*e.g.*, localized cutaneous reactions such as mild pruritus, flushing, rash):
  - Decrease the rate of infusion until recovery from symptoms, remain at bedside and monitor patient.
  - Complete the ipilimumab infusion at the initial planned rate.
  - Diphenhydramine 50 mg IV may be administered at the discretion of the treating physician and patients may receive additional doses with close monitoring.
  - Premedication with diphenhydramine may be given at the discretion of the investigator for subsequent doses of ipilimumab.
- For moderate symptoms (any symptom not listed above [mild symptoms] or below [severe symptoms] such as generalized pruritus, flushing, rash, dyspnea, hypotension with systolic BP >80 mmHg):
  - Interrupt ipilimumab.
  - Administer diphenhydramine 50 mg IV.

- Monitor patient closely until resolution of symptoms.
- Corticosteroids may abrogate any beneficial immunologic effect, but may be administered at the discretion of the treating physician.
- Resume ipilimumab infusion after recovery of symptoms.
- At the discretion of the treating physician, ipilimumab infusion may be resumed at *one half the initial infusion rate, then increased incrementally to the initial infusion rate.*
- If symptoms develop after resumption of the infusion, the infusion should be discontinued and no additional ipilimumab should be administered that day.
- The next dose of ipilimumab will be administered at its next scheduled time and may be given with pre-medication (diphenhydramine and acetaminophen) and careful monitoring, following the same treatment guidelines outlined above.
- At the discretion of the treating physician additional oral or IV antihistamine may be administered prior to dosing with ipilimumab.

- For severe symptoms (e.g., any reaction such as bronchospasm, generalized urticaria, systolic blood pressure <80 mm Hg, or angioedema):
  - Immediately discontinue infusion of ipilimumab, and disconnect infusion tubing from the subject.
  - Consider bronchodilators, epinephrine 1 mg IV or subcutaneously, and/or diphenhydramine 50 mg IV, with solumedrol 100 mg IV, as needed.
  - Patients should be monitored until the investigator is comfortable that the symptoms will not recur.
  - No further ipilimumab will be administered.
- In case of late-occurring hypersensitivity symptoms (e.g., appearance within one week after treatment of a localized or generalized pruritus), symptomatic treatment may be given (e.g., oral antihistamine, or corticosteroids).

6.3.2 Treatment of Ipilimumab-Related Isolated Drug Fever

In the event of isolated drug fever, the investigator must use clinical judgment to determine if the fever is related to the ipilimumab or to an infectious etiology. If a patient experiences isolated drug fever, for the next dose, pre-treatment with acetaminophen or non-steroidal anti-inflammatory agent (investigator discretion) should be instituted and a repeated antipyretic dose at 6 and 12 hours after ipilimumab infusion, should be administered. The infusion rate will remain unchanged for future doses. If a patient experiences recurrent isolated drug fever following premedication and post dosing with an appropriate antipyretic, the infusion rate for subsequent dosing should be decreased to 50% of the previous rate. If fever recurs following infusion rate change, the investigator should assess the patient's level of discomfort with the event and use clinical judgment to determine if the patient should receive further ipilimumab.

6.4 Monitoring and Management of Immune-mediated Adverse Reactions

6.4.1 Immune-mediated Enterocolitis

The clinical presentation of GI immune-related AEs included diarrhea, increase in the frequency of bowel movements, abdominal pain, or hematochezia, with or without fever. However inflammation may occur in any part of the GI tract including esophagitis and gastritis. Fatalities due to GI perforation have been reported in clinical trials of ipilimumab. Patients should be carefully monitored for GI symptoms that may be indicative of immune-related colitis, diarrhea, or GI perforation. Diarrhea or colitis occurring after initiation of ipilimumab therapy should be evaluated to exclude infectious or alternate etiologies. In clinical trials, immune-related colitis was associated with evidence of mucosal inflammation, with or without ulcerations, and lymphocytic infiltration.

Monitor patients for signs and symptoms of enterocolitis (such as diarrhea, abdominal pain, mucus or blood in stool, with or without fever) and bowel perforation (such as peritoneal signs and ileus). In symptomatic patients, rule out infectious etiologies and consider endoscopic evaluation to establish etiology and for persistent or severe symptoms. *C.difficile* toxin has been detected in several patients with colitis and may be an independent entity or may co-exist with ipilimumab induced inflammatory colitis.

Withhold ipilimumab dosing for any patients with enterocolitis pending evaluation; administer anti-diarrheal treatment and, if persistent evaluate with colonoscopy and initiate systemic corticosteroids at a dose of 0.5 mg/kg/day prednisone or equivalent.

Permanently discontinue ipilimumab in patients with severe enterocolitis and initiate systemic corticosteroids at a dose of 1 to 2 mg/kg/day of prednisone or equivalent. Upon improvement to grade 1 or less, initiate corticosteroid taper and continue to taper over at least one month. In clinical trials, rapid corticosteroid tapering has resulted in recurrence or worsening symptoms of enterocolitis in some patients.

Patients have been treated with anti-TNF agents for persistent colitis not responding to steroids.

Please note autoimmune pancreatitis may cause abdominal pain and should be included in all evaluations. Enteritis may occur occasionally with other autoimmune events including hepatitis, pancreatitis, and endocrine insufficiency, which should be evaluated as clinically indicated.

6.4.2 Immune-mediated Hepatitis and Pancreatitis

Hepatic immune-related AEs were mostly clinically silent and manifested as transaminase or bilirubin laboratory abnormalities. Fatal hepatic failure has been reported in clinical trials of ipilimumab. **Serum transaminase and bilirubin and lipase levels must be evaluated before each dose of ipilimumab as early laboratory changes may be indicative of emerging immune-related hepatitis/ pancreatitis and elevations in liver function tests (LFTs) may develop in the absence of clinical symptoms.** Increase in LFT or total bilirubin should be evaluated to exclude other causes of hepatic injury, including infections, disease progression, or other medications, and monitored until resolution. Liver biopsies from patients who had immune-related hepatotoxicity showed evidence of acute inflammation (neutrophils, lymphocytes, and macrophages).

Monitor liver function tests (hepatic transaminase and bilirubin levels, lipase) and assess patients for signs and symptoms of hepatotoxicity/ pancreatitis before each dose of ipilimumab. In patients with hepatotoxicity, rule out infectious or malignant causes and increase frequency of liver function test monitoring until resolution. Withhold ipilimumab in patients with grade 2 hepatotoxicity.

Permanently discontinue ipilimumab in patients with grade 3–5 hepatotoxicity/pancreatitis and administer systemic corticosteroids at a dose of 1 to 2 mg/kg/day of prednisone or equivalent. When liver function tests show sustained improvement or return to baseline, initiate corticosteroid tapering and continue to taper over 1 month. Across the clinical development program for ipilimumab, mycophenolate treatment has been administered in patients who have persistent severe hepatitis despite high-dose corticosteroids.

6.4.3 Immune-mediated Dermatitis

Skin immune-related AEs presented mostly frequently as a rash and/or pruritus. Some subjects reported vitiligo associated with ipilimumab administration. Fatal toxic epidermal necrolysis has been reported in clinical trials of ipilimumab.

Monitor patients for signs and symptoms of dermatitis such as rash and pruritus. Unless an alternate etiology has been identified, signs or symptoms of dermatitis should be considered immune-mediated.

Permanently discontinue ipilimumab in patients with Stevens-Johnson syndrome, toxic epidermal necrolysis, or rash complicated by full thickness dermal ulceration, or necrotic, bullous, or hemorrhagic manifestations. Administer systemic corticosteroids at a dose of 1 to 2 mg/kg/day of prednisone or equivalent. When dermatitis is controlled, corticosteroid tapering should occur over a period of at least 1 month. Withhold ipilimumab dosing in patients with moderate to severe signs and symptoms.

For mild to moderate dermatitis, such as grade 2 localized rash and pruritus, treat symptomatically. For persistent grade 2, grade 3, or greater, topical steroids may be administered. Administer topical or systemic corticosteroids as indicated if there is no improvement of symptoms within 1 week.

6.4.4 Immune-related Neurological Events

Fatal Guillain-Barré syndrome has been reported in clinical trials of ipilimumab. Patients may present with muscle weakness and myasthenia gravis, cranial nerve palsy (n VII Bell's palsy), and aseptic meninigits, encephalopathy. Unexplained motor neuropathy, muscle weakness, or sensory neuropathy lasting more than 4 days should be evaluated and non-inflammatory causes such as disease progression, infections, metabolic syndromes, nerve entrapment, and medications should be excluded as causes.

Withhold ipilimumab dosing in patients with any evidence of neuropathy pending evaluation.

Monitor for symptoms of motor or sensory neuropathy such as unilateral or bilateral weakness, sensory alterations, or paresthesia. Permanently discontinue ipilimumab in patients with severe

neuropathy (interfering with daily activities) such as Guillain-Barré-like syndromes. Institute medical intervention as appropriate for management of neuropathy and other neurologic events. Consider initiation of systemic corticosteroids at a dose of 1 to 2 mg/kg/day prednisone or equivalent for severe neuropathies.

There have also been reports of encephalitis and myositis.

6.4.5 Immune-mediated Endocrinopathies

Ipilimumab can cause inflammation of endocrine organs including thyroid (Hashimoto's thryroiditis with positive antibodies) and adrenal glands, hypophysitis, hypopituitarism, and resulting thyroid and adrenal insufficiency, low ADH, prolactin, FSH, LH. Hyperthyroid with Graves' disease and positive antibody has been reported. Patients may present with subtle and nonspecific symptoms. The most common clinical presentation includes headache and fatigue. Symptoms may also include visual field defects, behavioral changes, and electrolyte disturbances including hyponatremia and hypotension. Adrenal crisis as a cause of the patient's symptoms should be excluded. Based on the available data with known outcome, most of the subjects symptomatically improved with hormone replacement therapy. Long-term hormone replacement therapy with HC and synthroid will typically be required for subjects developing hypophysitis/hypopituitarism after treatment with ipilimumab. Some patients have regained partial function following steroid treatment.

Monitor patients for clinical signs and symptoms of hypophysitis, adrenal insufficiency (including adrenal crisis), and hyper- or hypothyroidism. Headache is often the first symptoms of hypophysitis. Patients may present with fatigue, headache, mental status changes, loss of libido, abdominal pain, unusual bowel habits, and hypotension, or nonspecific symptoms which may resemble other causes such as brain metastasis or underlying disease. Unless an alternate etiology has been identified, signs or symptoms of endocrinopathies should be considered immune-mediated and drug withheld pending evaluation. Patients may demonstrate both central (hypophysitis) and peripheral adrenal and thyroid insufficiency. Evaluation of hypophysitis should include pituitary MRI.

Endocrine evaluation, including TSH, should be performed at baseline prior to initial treatment. Monitor thyroid function tests and clinical chemistries at the start of treatment and hold blood for possible evaluation should clinical events require determining baseline function and anti-thyroid antibodies. A plan for evaluating endocrine function at each visit either by history or monitoring TSH should be included in the protocol with further evaluation as clinically indicated. Endocrine evaluation, including TSH, should be performed at baseline prior to treatment, and the protocol must include a plan for continued monitoring of endocrine and pituitary function. The package insert for ipilimumab includes a recommendation for monitoring TSH prior to each infusion; as an early indication for pituitary dysfunction and hypophysitis, clinical monitoring of symptoms may be equally or more sensitive as an initial presentation. Clinical monitoring is required for all protocols as above, and should include any requirements per protocol for laboratory evaluation, both periodically or as clinically indicated, consistent with good medical practice. In a limited number of patients, hypophysitis was diagnosed by imaging studies through enlargement of the pituitary gland.

Withhold ipilimumab dosing in patients symptomatic for hypophysitis. Initiate systemic corticosteroids at a dose of 1 to 2 mg/kg/day of prednisone or equivalent, and initiate appropriate hormone replacement therapy.

6.4.6 Other Immune-mediated Adverse Reactions, Including Ocular Manifestations

Ocular inflammation, manifested as grade 2 or grade 3 episcleritis or uveitis, was associated with concomitant diarrhea in a few subjects (<1%) and occasionally occurred in the absence of clinically apparent GI symptoms. Other presumed immune-related AEs reported include, but were not limited to, arthritis/arthralgias, pneumonitis, pancreatitis, autoimmune (aseptic) meningitis, autoimmune nephritis, pure red cell aplasia, noninfective myocarditis, polymyositis, and myasthenia gravis, of which were individually reported for <1% of subjects.

The following clinically significant immune-mediated adverse reactions were seen in less than 1% of ipilimumab-treated patients in Study 1: nephritis, pneumonitis, pulmonary granuloma resembling sarcoidosis, meningitis, pericarditis, uveitis, iritis, ITP, neutropenia and hemolytic anemia.

Across the clinical development program for ipilimumab, the following likely immune-mediated adverse reactions were also reported with less than 1% incidence: myocarditis, angiopathy, temporal arteritis, vasculitis, polymyalgia rheumatica, conjunctivitis, blepharitis, episcleritis, scleritis, leukocytoclastic vasculitis, erythema multiforme, psoriasis, pancreatitis, arthritis, and autoimmune thyroiditis.

Permanently discontinue ipilimumab for clinically significant or severe immune-mediated adverse reactions. Initiate systemic corticosteroids at a dose of 1 to 2 mg/kg/day prednisone or equivalent for severe immune-mediated adverse reactions.

Administer corticosteroid eye drops to patients who develop uveitis, iritis, or episcleritis. Permanently discontinue ipilimumab for immune-mediated ocular disease that is unresponsive to local immunosuppressive therapy.

Overall, immune-related AEs commonly started within 3 to 10 weeks from first dose, were successfully managed in most instances by omitting doses, discontinuing dosing, and/or through administering symptomatic or immunosuppressive therapy, including corticosteroids, as mentioned above and detailed in Section 7. Immune-related AEs generally resolved within days to weeks in the majority of subjects.

There have also been reports of encephalitis.

6.4.7 Cardiac Toxicities Associated with Ipilimumab

If there is any concern for ipilimumab induced cardiac toxicity, alert the Overall PI immediately, and consider following the management algorithm below:

Cardiac *	**Management for Ipilimumab Cardiac Toxicities**

≤ Grade 1	Hold dose pending evaluation and observation.** Evaluate for signs and symptoms of CHF, ischemia, arrhythmia or myositis. Obtain history EKG, CK (for concomitant myositis), CK-MB. Repeat troponin, CK and EKG 2-3 days. If troponin and labs normalize may resume therapy. If labs worsen or symptoms develop then treat as below. Hold pending evaluation.
Grade ≥2 with suspected myocarditis	Hold dose.** Admit to hospital. Cardiology consult. Rule out MI and other causes of cardiac disease. Cardiac Monitoring. Cardiac Echo. Consider cardiac MRI and cardiac biopsy. Initiate high dose methylprednisolone. If no improvement within 24 hours, add either infliximab, ATG or tacrolimus. Consult algorithm for more details. Resume therapy if there is a return to baseline and myocarditis is excluded or considered unlikely.
Grade ≥2 with confirmed myocarditis	Off protocol therapy. Admit to CCU (consider transfer to nearest Cardiac Transplant Unit). Treat as above. Consider high dose methylprednisolone. Add ATG or tacrolimus if no improvement. Off treatment.
**Including CHF, LV systolic dysfunction, Myocarditis, CPK, and troponin* ***Patients with evidence of myositis without myocarditis may be treated according as "other event"* Note: The optimal treatment regimen for immune mediated myocarditis has not been established. Since this toxicity has caused patient deaths, an aggressive approach is recommended.	

6.5 Prohibited and Restricted Therapies

Patients in this study may use standard vaccines. Where possible, routine vaccination for influenza, pneumococcal pneumonia should be given prior to the start of therapy but may be administered during treatment when clinically indicated. Vaccination should be given when there is enough separation to distinguish any vaccine reactions from drug toxicity. There is no experience using live attenuated vaccination during ipilimumab therapy, so that live vaccine should be used cautiously during treatment.

Concomitant systemic anti-cancer medications or treatments are prohibited in this study while receiving ipilimumab treatments. However, IT chemotherapy prophylaxis is allowed and can be considered after discussion with the overall PI and further these cases must be discussed on the safety calls. If IT chemotherapy prophylaxis is recommended, it can be given at any time during decitabine only lead-in cycle for patients in either dose-escalation or dose-expansion. If IT chemotherapy prophylaxis is recommended during decitabine plus ipilimumab cycles, it will be considered and allowed in patients who enter dose-expansion and we recommend:

(1) To provide a 2 week window before Ipilimumab dosing (thus Ipilimumab may be delayed),

(2) consideration for IT hydrocortisone 50 mg with the IT chemotherapy of choice (methotrexate or cytarabine per provider),

(3) additional clinic visit for neurologic exam 3-5 days after IT chemo administration to ensure no additional toxicities are seen. Common toxicities experienced during IT chemotherapy include spinal headache and nausea; rarely arachnoiditis. Events in the CNS that are considered excessive beyond what is seen will be counted as an AE related to study drug. Platelets must be > 50 for IT chemotherapy or per institutional guidelines.

Patients who remain on hydroxyurea and/or IT chemotherapy prophylaxis will be discussed on the safety calls to ensure new adverse events are appropriately captured if relevant.

In select cases of extensive disease involvement, palliative radiation may be administered to a localized problematic site during the study if there is evidence of disease involvement outside of the bone marrow with permission from the Protocol Chair. A delay of both decitabine and ipilimumab of 2 weeks is required starting the day that radiation is completed. Example: vertebral mass encroaching on spine with evidence of disease elsewhere in the body.

Patients may not use any of the following therapies during the study:

- Any non-study anti-cancer agent (investigational or non-investigational), except for hydroxyurea or prophylactic intrathecal chemotherapy as clinically indicated
- Any other investigational agents
- Any other CTLA-4 inhibitors or agonists
- CD137 or other immunologic activation agonists
- Immunosuppressive agents
- Chronic systemic corticosteroids (short courses of corticosteroids may be allowed while ipilimumab is delayed as discussed in section 6.2)

6.5.1 Precautions

Combination therapy may result in unexpected toxicity especially in novel combinations with other immune modifying agents. A striking example in macaques is presented in Vaccari*, et al.* 2012.

Please note that while unproven, there is a suggestion that autoimmune events, including hepatitis, may occur more frequently at sites of metastases or prior injury.

Caution is advised when considering treatment with high-dose IL-2 in patients who have previously been administered ipilimumab, particularly in patients who experienced ipilimumab-related diarrhea/colitis. Colonoscopy or sigmoidoscopy with biopsy may be advisable prior to IL-2 administration once the patient is no longer receiving ipilimumab. The management guidelines for general inflammatory AEs and ipilimumab-related GI toxicities, hepatotoxicity, endocrinopathy, and neuropathy (Investigator Brochure, 2011) are provided in Appendix C.

Patients who have received ipilimumab may potentially develop autoimmune disease with subsequent therapy including the appearance of colitis, hypophysitis or adrenal insufficiency.

7. ADVERSE EVENTS: LIST AND REPORTING REQUIREMENTS

Adverse event (AE) monitoring and reporting is a routine part of every clinical trial. The following list of AEs (Section 7.1) and the characteristics of an observed AE (Section 7.2) will determine whether the event requires expedited reporting via the CTEP Adverse Event Reporting System (CTEP-AERS) **in addition** to routine reporting.

7.1 Comprehensive Adverse Events and Potential Risks List (CAEPR)

The Comprehensive Adverse Event and Potential Risks list (CAEPR) provides a single list of reported and/or potential adverse events (AE) associated with an agent using a uniform presentation of events by body system. In addition to the comprehensive list, a subset of AEs, the Specific Protocol Exceptions to Expedited Reporting (SPEER), appears in a separate column and is identified with ***bold*** and ***italicized*** text. The SPEER is a list of events that are protocol-specific exceptions to expedited reporting to NCI (except as noted below). Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' http://ctep.cancer.gov/protocolDevelopment/adverse_effects.htm for further clarification.

NOTE: The highest grade currently reported is noted in parentheses next to the AE in the SPEER. Report **ONLY** AEs higher than this grade expeditiously. If this CAEPR is part of a combination protocol using multiple investigational agents and has an AE listed on different SPEERs, use the lower of the grades to determine if expedited reporting is required.

7.1.1 Adverse Event List for Ipilimumab

Comprehensive Adverse Events and Potential Risks list (CAEPR)
for
Ipilimumab (MDX-010, NSCs 732442 and 720801)

The Comprehensive Adverse Events and Potential Risks list (CAEPR) provides a single list of reported and/or potential adverse events (AE) associated with an agent using a uniform presentation of events by body system. In addition to the comprehensive list, a subset, the Specific Protocol Exceptions to Expedited Reporting (SPEER), appears in a separate column and is identified with bold and italicized text. This subset of AEs (SPEER) is a list of events that are protocol specific exceptions to expedited reporting to NCI (except as noted below). Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf for further clarification. *Frequency is provided based on 2678 patients.* Below is the CAEPR for Ipilimumab (MDX-010).

NOTE: Report AEs on the SPEER **ONLY IF** they exceed the grade noted in parentheses next to the AE in the SPEER. If this CAEPR is part of a combination protocol using multiple investigational agents and has an AE listed on different SPEERs, use the lower of the grades to determine if expedited reporting is required.

Version 2.10, March 29, 2019[1]

Adverse Events with Possible Relationship to Ipilimumab (MDX-010) (CTCAE 5.0 Term) [n= 2678]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)**
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
BLOOD AND LYMPHATIC SYSTEM DISORDERS			
		Blood and lymphatic system disorders - Other (acquired hemophilia)	
CARDIAC DISORDERS			
	Atrial fibrillation		
		Myocarditis[2]	
		Pericardial effusion	
EAR AND LABYRINTH DISORDERS			
	Hearing impaired		
ENDOCRINE DISORDERS			
	Adrenal insufficiency[2]		
	Hyperthyroidism[2]		
	Hypophysitis[2]		
	Hypopituitarism[2]		
	Hypothyroidism[2]		
	Testosterone deficiency[2]		
EYE DISORDERS			
	Eye disorders - Other (episcleritis)[2]		
	Uveitis[2]		
GASTROINTESTINAL DISORDERS			
	Abdominal pain		
	Colitis[2]		***Colitis[2] (Gr 3)***
		Colonic perforation[3]	
	Constipation		
Diarrhea			***Diarrhea (Gr 3)***
	Enterocolitis		
	Esophagitis		
		Ileus	
Nausea			***Nausea (Gr 3)***
	Pancreatitis[2]		

Adverse Events with Possible Relationship to Ipilimumab (MDX-010) (CTCAE 5.0 Term) [n= 2678]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)**
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Vomiting		
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
	Chills		
Fatigue			***Fatigue (Gr 3)***
	Fever		***Fever (Gr 2)***
		General disorders and administration site conditions - Other (Systemic inflammatory response syndrome [SIRS])	
		Multi-organ failure	
HEPATOBILIARY DISORDERS			
	Hepatobiliary disorders - Other (hepatitis)[2]		
IMMUNE SYSTEM DISORDERS			
	Autoimmune disorder[2]		
		Immune system disorders - Other (GVHD in the setting of allotransplant)[4]	
INFECTIONS AND INFESTATIONS			
		Infections and infestations - Other (aseptic meningitis)[2]	
INJURY, POISONING AND PROCEDURAL COMPLICATIONS			
	Infusion related reaction		
INVESTIGATIONS			
	Alanine aminotransferase increased		
	Aspartate aminotransferase increased		
		Lymphocyte count decreased	
	Neutrophil count decreased		
	Weight loss		
METABOLISM AND NUTRITION DISORDERS			
	Anorexia		
	Dehydration		

Adverse Events with Possible Relationship to Ipilimumab (MDX-010) (CTCAE 5.0 Term) [n= 2678]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Hyperglycemia		
		Metabolism and nutrition disorders - Other (exacerbation of pre-existing diabetes mellitus)	
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS			
	Arthralgia		
	Arthritis		
		Generalized muscle weakness	
	Musculoskeletal and connective tissue disorder - Other (polymyositis)[2]		
NERVOUS SYSTEM DISORDERS			
		Ataxia	
	Facial nerve disorder[2]		
	Guillain-Barre syndrome[2]		
	Headache		
	Myasthenia gravis[2]		
		Nervous system disorders - Other (immune-mediated encephalitis)[2]	
		Peripheral motor neuropathy	
		Peripheral sensory neuropathy	
	Trigeminal nerve disorder		
PSYCHIATRIC DISORDERS			
		Psychiatric disorders - Other (mental status changes)	
RENAL AND URINARY DISORDERS			
	Acute kidney injury		
	Renal and urinary disorders - Other (granulomatous tubulointerstitial nephritis)		
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS			
	Pneumonitis		

Adverse Events with Possible Relationship to Ipilimumab (MDX-010) (CTCAE 5.0 Term) [n= 2678]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
		Respiratory failure	
		Respiratory, thoracic and mediastinal disorders - Other (bronchiolitis obliterans with organizing pneumonia)	
		Respiratory, thoracic and mediastinal disorders - Other (lung infiltration)	
SKIN AND SUBCUTANEOUS TISSUE DISORDERS			
		Erythema multiforme	
	Pruritus		***Pruritus (Gr 3)***
Rash maculo-papular			***Rash maculo-papular (Gr 3)***
	Skin and subcutaneous tissue disorders - Other (Sweet's Syndrome)		
		Stevens-Johnson syndrome	
		Toxic epidermal necrolysis	
	Urticaria		
VASCULAR DISORDERS			
	Hypotension		

[1]This table will be updated as the toxicity profile of the agent is revised. Updates will be distributed to all Principal Investigators at the time of revision. The current version can be obtained by contacting PIO@CTEP.NCI.NIH.GOV. Your name, the name of the investigator, the protocol and the agent should be included in the e-mail.

[2]Ipilimumab can result in severe and fatal immune-mediated adverse events probably due to T-cell activation and proliferation. These can include (but are not limited to) autoimmune hemolytic anemia, acquired anti-factor VIII immune response, autoimmune aseptic meningitis, autoimmune hepatitis, autoimmune thyroiditis, hepatic failure, pure red cell aplasia, pancreatitis, ulcerative and hemorrhagic colitis, endocrine disorders (e.g., autoimmune thyroiditis, hyperthyroidism, hypothyroidism, autoimmune hypophysitis/hypopituitarism, and adrenal insufficiency), ocular manifestations (e.g., uveitis, iritis, conjunctivitis, blepharitis, and episcleritis), sarcoid granuloma, myasthenia gravis, polymyositis, and Guillain-Barre syndrome. The majority of these reactions manifested early during treatment; however, a minority occurred weeks to months after discontinuation of ipilimumab especially with the initiation of additional treatments.

[3]Late bowel perforations have been noted in patients receiving MDX-010 (ipilimumab) in

association with subsequent IL-2 therapy.

[4]Complications including hyperacute graft-versus-host disease (GVHD), may occur in patients receving allo stem cell transplant (SCT) after receiving Ipilimumab (MDX-010). These complications may occur despite intervening therapy between receiving Ipilimumab (MDX-010) and allo-SCT.

[5]In rare cases diplopia (double vision) has occurred as a result of muscle weakness (Myasthenia gravis).

[6]Gastrointestinal hemorrhage includes Anal hemorrhage, Cecal hemorrhage, Colonic hemorrhage, Duodenal hemorrhage, Esophageal hemorrhage, Esophageal varices hemorrhage, Gastric hemorrhage, Hemorrhoidal hemorrhage, Ileal hemorrhage, Intra-abdominal hemorrhage, Jejunal hemorrhage, Lower gastrointestinal hemorrhage, Oral hemorrhage, Pancreatic hemorrhage, Rectal hemorrhage, Retroperitoneal hemorrhage, and Upper gastrointestinal hemorrhage under the GASTROINTESTINAL DISORDERS SOC.

[7]Infection includes all 75 sites of infection under the INFECTIONS AND INFESTATIONS SOC.

Adverse events reported on Ipilimumab (MDX-010) trials, but for which there is insufficient evidence to suggest that there was a reasonable possibility that Ipilimumab (MDX-010) caused the adverse event:

BLOOD AND LYMPHATIC SYSTEM DISORDERS - Anemia; Blood and lymphatic system disorders - Other (pure red cell aplasia)[2]; Febrile neutropenia
CARDIAC DISORDERS - Conduction disorder; Restrictive cardiomyopathy
EYE DISORDERS - Extraocular muscle paresis[5]; Eye disorders - Other (retinal pigment changes)
GASTROINTESTINAL DISORDERS - Colonic ulcer; Dyspepsia; Dysphagia; Gastrointestinal disorders - Other (gastroenteritis); Gastrointestinal hemorrhage[6]; Proctitis
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Flu like symptoms; Non-cardiac chest pain
HEPATOBILIARY DISORDERS - Hepatic failure[2]
IMMUNE SYSTEM DISORDERS - Allergic reaction
INFECTIONS AND INFESTATIONS - Infection[7]
INVESTIGATIONS - Creatinine increased; Investigations - Other (rheumatoid factor); Lipase increased; Platelet count decreased; Serum amylase increased; White blood cell decreased
METABOLISM AND NUTRITION DISORDERS - Tumor lysis syndrome
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Back pain; Joint range of motion decreased; Myalgia; Pain in extremity
NEOPLASMS BENIGN, MALIGNANT AND UNSPECIFIED (INCL CYSTS AND POLYPS) - Tumor pain
NERVOUS SYSTEM DISORDERS - Dizziness; Dysphasia; Ischemia cerebrovascular; Seizure
PSYCHIATRIC DISORDERS - Anxiety; Confusion; Depression; Insomnia

RENAL AND URINARY DISORDERS - Proteinuria
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Allergic rhinitis; Cough; Dyspnea; Laryngospasm
SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Alopecia; Dry skin; Hyperhidrosis; Skin hypopigmentation
VASCULAR DISORDERS - Flushing; Hypertension; Vascular disorders - Other (temporal arteritis)

Note: Ipilimumab (MDX-010) in combination with other agents could cause an exacerbation of any adverse event currently known to be caused by the other agent, or the combination may result in events never previously associated with either agent.

7.1.2 Adverse Event List for Decitabine

Comprehensive Adverse Events and Potential Risks list (CAEPR)
for
Decitabine (5-aza-2'-deoxycytidine, NSC 127716)

The Comprehensive Adverse Events and Potential Risks list (CAEPR) provides a single list of reported and/or potential adverse events (AE) associated with an agent using a uniform presentation of events by body system. In addition to the comprehensive list, a subset, the Specific Protocol Exceptions to Expedited Reporting (SPEER), appears in a separate column and is identified with bold and italicized text. This subset of AEs (SPEER) is a list of events that are protocol specific exceptions to expedited reporting to NCI (except as noted below). Refer to the 'CTEP, NCI Guidelines: Adverse Event Reporting Requirements' http://ctep.cancer.gov/protocolDevelopment/electronic_applications/docs/aeguidelines.pdf for further clarification. *Frequency is provided based on 1832 patients.* Below is the CAEPR for Decitabine (5-aza-2'-deoxycytidine).

NOTE: Report AEs on the SPEER **ONLY IF** they exceed the grade noted in parentheses next to the AE in the SPEER. If this CAEPR is part of a combination protocol using multiple investigational agents and has an AE listed on different SPEERs, use the lower of the grades to determine if expedited reporting is required.

Version 2.5, March 28, 2019[1]

Adverse Events with Possible Relationship to Decitabine (5-aza-2'-deoxycytidine) (CTCAE 5.0 Term) [n= 1832]			**Specific Protocol Exceptions to Expedited Reporting (SPEER)**
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
BLOOD AND LYMPHATIC SYSTEM DISORDERS			
Anemia			***Anemia (Gr 4)***
	Febrile neutropenia		***Febrile neutropenia (Gr 3)***
GASTROINTESTINAL DISORDERS			
	Abdominal pain		***Abdominal pain (Gr 2)***
	Anal mucositis		***Anal mucositis (Gr 2)***

Adverse Events with Possible Relationship to Decitabine (5-aza-2'-deoxycytidine) (CTCAE 5.0 Term) [n= 1832]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Constipation		***Constipation (Gr 2)***
	Diarrhea		***Diarrhea (Gr 2)***
	Mucositis oral		***Mucositis oral (Gr 2)***
Nausea			***Nausea (Gr 2)***
	Rectal mucositis		***Rectal mucositis (Gr 2)***
	Small intestinal mucositis		***Small intestinal mucositis (Gr 2)***
	Vomiting		***Vomiting (Gr 2)***
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS			
	Chills		
	Edema limbs		***Edema limbs (Gr 2)***
Fatigue			***Fatigue (Gr 3)***
Fever			***Fever (Gr 2)***
	Non-cardiac chest pain		
	Pain		
IMMUNE SYSTEM DISORDERS			
	Autoimmune disorder		***Autoimmune disorder (Gr 2)***
INFECTIONS AND INFESTATIONS			
Infection[2]			***Infection[2] (Gr 3)***
INJURY, POISONING AND PROCEDURAL COMPLICATIONS			
	Bruising		
INVESTIGATIONS			
	Alanine aminotransferase increased		***Alanine aminotransferase increased (Gr 2)***
	Aspartate aminotransferase increased		***Aspartate aminotransferase increased (Gr 2)***
	Blood bilirubin increased		***Blood bilirubin increased (Gr 2)***
	Creatinine increased		***Creatinine increased (Gr 2)***
	Lymphocyte count decreased		
Neutrophil count decreased			***Neutrophil count decreased (Gr 4)***
Platelet count decreased			***Platelet count decreased (Gr 4)***
White blood cell decreased			***White blood cell decreased (Gr 4)***
METABOLISM AND NUTRITION DISORDERS			
	Anorexia		***Anorexia (Gr 2)***
	Hyperglycemia		
	Hyperuricemia		
	Hypoalbuminemia		
	Hypocalcemia		
	Hypokalemia		
	Hypomagnesemia		
	Hyponatremia		
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS			
	Arthralgia		
	Back pain		
	Bone pain		
	Pain in extremity		
NERVOUS SYSTEM DISORDERS			

Adverse Events with Possible Relationship to Decitabine (5-aza-2'-deoxycytidine) (CTCAE 5.0 Term) [n= 1832]			Specific Protocol Exceptions to Expedited Reporting (SPEER)
Likely (>20%)	**Less Likely (<=20%)**	**Rare but Serious (<3%)**	
	Dizziness		
	Headache		***Headache (Gr 2)***
		Intracranial hemorrhage	
	Somnolence		***Somnolence (Gr 2)***
PSYCHIATRIC DISORDERS			
	Anxiety		
	Insomnia		
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS			
	Cough		***Cough (Gr 2)***
	Dyspnea		***Dyspnea (Gr 2)***
	Laryngeal mucositis		***Laryngeal mucositis (Gr 2)***
	Pharyngeal mucositis		***Pharyngeal mucositis (Gr 2)***
	Pharyngolaryngeal pain		
	Respiratory hemorrhage[3]		
	Tracheal mucositis		***Tracheal mucositis (Gr 2)***
SKIN AND SUBCUTANEOUS TISSUE DISORDERS			
	Alopecia		***Alopecia (Gr 2)***
	Pruritus		
	Purpura		***Purpura (Gr 2)***
	Rash maculo-papular		***Rash maculo-papular (Gr 2)***
VASCULAR DISORDERS			
	Hematoma		
	Phlebitis		***Phlebitis (Gr 2)***
	Vascular disorders - Other (hemorrhage with decreased platelets)		***Vascular disorders - Other (hemorrhage with decreased platelets) (Gr 2)***

[1]This table will be updated as the toxicity profile of the agent is revised. Updates will be distributed to all Principal Investigators at the time of revision. The current version can be obtained by contacting PIO@CTEP.NCI.NIH.GOV. Your name, the name of the investigator, the protocol and the agent should be included in the e-mail.

[2]Infection includes all 75 sites of infection under the INFECTIONS AND INFESTATIONS SOC.

[3]Respiratory hemorrhage includes Bronchopulmonary hemorrhage, Epistaxis, Laryngeal hemorrhage, Mediastinal hemorrhage, Pharyngeal hemorrhage, and Pleural hemorrhage under the RESPIRATORY, THORACIC, AND MEDIASTINAL DISORDERS SOC.

[4]Gastrointestinal hemorrhage includes Anal hemorrhage, Cecal hemorrhage, Colonic hemorrhage, Duodenal hemorrhage, Esophageal hemorrhage, Esophageal varices hemorrhage, Gastric hemorrhage, Hemorrhoidal hemorrhage, Ileal hemorrhage, Intra-abdominal hemorrhage, Jejunal hemorrhage, Lower gastrointestinal hemorrhage, Oral hemorrhage, Pancreatic hemorrhage, Rectal hemorrhage, Retroperitoneal hemorrhage, and Upper gastrointestinal

hemorrhage under the GASTROINTESTINAL DISORDERS SOC.

[5]Gastrointestinal obstruction includes Colonic obstruction, Duodenal obstruction, Esophageal obstruction, Ileal obstruction, Jejunal obstruction, Obstruction gastric, Rectal obstruction, and Small intestinal obstruction under the GASTROINTESTINAL DISORDERS SOC.

Adverse events reported on decitabine (5-aza-2'-deoxycytidine) trials, but for which there is insufficient evidence to suggest that there was a reasonable possibility that decitabine (5-aza-2'-deoxycytidine) caused the adverse event:

BLOOD AND LYMPHATIC SYSTEM DISORDERS - Blood and lymphatic system disorders - Other (coagulopathy); Blood and lymphatic system disorders - Other (lymphadenopathy); Blood and lymphatic system disorders - Other (pancytopenia); Blood and lymphatic system disorders - Other (spleen disorder); Bone marrow hypocellular; Eosinophilia; Hemolysis; Leukocytosis
CARDIAC DISORDERS - Atrial fibrillation; Atrial flutter; Atrioventricular block complete; Cardiac arrest; Cardiac disorders - Other (cardiac murmur); Cardiac disorders - Other (dilation atrial); Chest pain - cardiac; Heart failure; Myocardial infarction; Restrictive cardiomyopathy; Sinus bradycardia; Sinus tachycardia; Supraventricular tachycardia; Ventricular arrhythmia
EAR AND LABYRINTH DISORDERS - Ear pain; Vertigo
EYE DISORDERS - Blurred vision; Eye disorders - Other (eye hemorrhage); Eye disorders - Other (eye swelling); Eye pain
GASTROINTESTINAL DISORDERS - Abdominal distension; Anal fissure; Ascites; Dyspepsia; Dysphagia; Enterocolitis; Esophagitis; Flatulence; Gastritis; Gastroesophageal reflux disease; Gastrointestinal disorders - Other (diverticulitis); Gastrointestinal disorders - Other (mouth ulceration); Gastrointestinal disorders - Other (oral mucosal blistering); Gastrointestinal hemorrhage[4]; Gastrointestinal obstruction[5]; Gastrointestinal pain; Hemorrhoids; Ileus; Oral pain; Periodontal disease; Proctitis; Rectal pain; Toothache; Typhlitis
GENERAL DISORDERS AND ADMINISTRATION SITE CONDITIONS - Edema face; Gait disturbance; Injection site reaction; Localized edema; Malaise; Multi-organ failure
HEPATOBILIARY DISORDERS - Cholecystitis; Hepatic failure; Hepatobiliary disorders - Other (cholestasis); Hepatobiliary disorders - Other (hepatomegaly)
IMMUNE SYSTEM DISORDERS - Allergic reaction
INJURY, POISONING AND PROCEDURAL COMPLICATIONS - Fall; Fracture; Infusion related reaction; Injury, poisoning and procedural complications - Other (catheter site pain); Injury, poisoning and procedural complications - Other (hernia); Injury, poisoning and procedural complications - Other (procedural pain); Injury, poisoning and procedural complications - Other (stent occlusion)
INVESTIGATIONS - Activated partial thromboplastin time prolonged; Alkaline phosphatase increased; Blood bicarbonate decreased; Blood lactate dehydrogenase increased; CPK increased; Cardiac troponin I increased; Ejection fraction decreased; Electrocardiogram QT corrected interval prolonged; Fibrinogen decreased; GGT increased; INR increased; Investigations - Other (blood bicarbonate increased); Investigations - Other (blood bilirubin decreased); Investigations - Other (blood chloride decreased); Investigations - Other (blood chloride increased); Investigations - Other (blood urea increased); Investigations - Other (elevated ammonia);

Investigations - Other (platelet count increased); Investigations - Other (protein total decreased); Lipase increased; Serum amylase increased; Weight loss
METABOLISM AND NUTRITION DISORDERS - Acidosis; Dehydration; Hypercalcemia; Hyperkalemia; Hypermagnesemia; Hyperphosphatemia; Hypoglycemia; Hypophosphatemia; Metabolism and nutrition disorders - Other (malnutrition)
MUSCULOSKELETAL AND CONNECTIVE TISSUE DISORDERS - Chest wall pain; Generalized muscle weakness; Muscle cramp; Myalgia
NEOPLASMS BENIGN, MALIGNANT AND UNSPECIFIED (INCL CYSTS AND POLYPS) - Leukemia secondary to oncology chemotherapy; Tumor pain
NERVOUS SYSTEM DISORDERS - Amnesia; Aphonia; Ataxia; Cognitive disturbance; Dysesthesia; Dysgeusia; Ischemia cerebrovascular; Lethargy; Paresthesia; Peripheral sensory neuropathy; Seizure; Stroke; Syncope; Transient ischemic attacks
PSYCHIATRIC DISORDERS - Confusion; Delirium; Depression; Personality change
RENAL AND URINARY DISORDERS - Acute kidney injury; Cystitis noninfective; Hematuria; Urinary fistula; Urinary frequency; Urinary retention; Urinary tract pain; Urinary urgency
REPRODUCTIVE SYSTEM AND BREAST disorders - Breast pain; Uterine hemorrhage; Vaginal hemorrhage
RESPIRATORY, THORACIC AND MEDIASTINAL DISORDERS - Adult respiratory distress syndrome; Allergic rhinitis; Atelectasis; Bronchospasm; Hypoxia; Nasal congestion; Pleural effusion; Pneumonitis; Postnasal drip; Pulmonary edema; Respiratory failure; Respiratory, thoracic and mediastinal disorders - Other (breath sounds abnormal /decreased); Respiratory, thoracic and mediastinal disorders - Other (crepitations); Respiratory, thoracic and mediastinal disorders - Other (pulmonary congestion); Sinus disorder; Wheezing
SKIN AND SUBCUTANEOUS TISSUE DISORDERS - Bullous dermatitis; Dry skin; Erythema multiforme; Hyperhidrosis; Hyperkeratosis; Skin and subcutaneous tissue disorders - Other (Sweet's syndrome); Skin hyperpigmentation; Skin hypopigmentation; Skin ulceration; Stevens-Johnson syndrome; Urticaria
VASCULAR DISORDERS - Flushing; Hypertension; Hypotension; Thromboembolic event; Vascular disorders - Other (aortic aneurysm); Vascular disorders - Other (catheter site hemorrhage); Vascular disorders - Other (circulatory collapse); Vascular disorders - Other (hemorrhage); Vascular disorders - Other (splenic infarct vs hemorrhage/rupture); Vascular disorders - Other (veno-occlusive disease); Vasculitis

Note: Decitabine (5-aza-2'-deoxycytidine) in combination with other agents could cause an exacerbation of any adverse event currently known to be caused by the other agent, or the combination may result in events never previously associated with either agent

7.2 Adverse Event Characteristics

- **CTCAE term (AE description) and grade:** The descriptions and grading scales found in the revised NCI Common Terminology Criteria for Adverse Events (CTCAE) version 5.0 will be utilized for AE reporting. All appropriate treatment areas should have access to a copy of the CTCAE version 5.0. A copy of the CTCAE version 5.0 can be downloaded from the CTEP web site

http://ctep.cancer.gov/protocolDevelopment/electronic_applications/ctc.htm.

- **For expedited reporting purposes only:**
  - AEs for the agent that are ***bold and italicized*** in the CAEPR (*i.e.*, those listed in the SPEER column, Section 7.1.1) should be reported through CTEP-AERS only if the grade is above the grade provided in the SPEER.
  - Other AEs for the protocol that do not require expedited reporting are outlined in section 7.3.4.

- **Attribution** of the AE:
  - Definite – The AE *is clearly related* to the study treatment.
  - Probable – The AE *is likely related* to the study treatment.
  - Possible – The AE *may be related* to the study treatment.
  - Unlikely – The AE *is doubtfully related* to the study treatment.
  - Unrelated – The AE *is clearly NOT related* to the study treatment.

7.3 Expedited Adverse Event Reporting

7.3.1 Expedited AE reporting for this study must use CTEP-AERS (CTEP Adverse Event Reporting System), accessed via the CTEP Web site (https://eapps-ctep.nci.nih.gov/ctepaers). The reporting procedures to be followed are presented in the "NCI Guidelines for Investigators: Adverse Event Reporting Requirements for DCTD (CTEP and CIP) and DCP INDs and IDEs" which can be downloaded from the CTEP Web site (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/adverse_events.htm). These requirements are briefly outlined in the tables below (Section 7.3.3).

In the rare occurrence when Internet connectivity is lost, a 24-hour notification is to be made to CTEP by telephone at 301-897-7497. Once Internet connectivity is restored, the 24-hour notification phoned in must be entered electronically into CTEP-AERS by the original submitter at the site.

7.3.2 Distribution of Adverse Event Reports

CTEP-AERS is programmed for automatic electronic distribution of reports to the following individuals: Principal Investigator and Adverse Event Coordinator(s) (if applicable) of the Corresponding Organization or Lead Organization, the local treating physician, and the Reporter and Submitter. CTEP-AERS provides a copy feature for other e-mail recipients.

The Coordinating Center of the Corresponding Organization is responsible for submitting to the CTSU documentation of AEs that they deem reportable for posting on the CTSU protocol web page and inclusion on the CTSU bi-monthly broadcast.

7.3.3 Expedited Reporting Guidelines

In addition to reporting events to NCI/ETCTN, it is mandatory for all participating sites to report (informally via email) sites should notify the overall PI and the lead site study manager of any grade 3 or above ipilimumab related event informally via email. Any grade 5 event regardless of attribution should be reported to the Overall Study Chair and project manager within 24 hours of knowledge of the event.

Use the NCI protocol number and the protocol-specific patient ID assigned during trial registration on all reports.

Note: A death on study requires both routine and expedited reporting, regardless of causality. Attribution to treatment or other cause must be provided.

Death due to progressive disease should be reported as Grade 5 "Disease progression" in the system organ class (SOC) "General disorders and administration site conditions."

Phase 1 and Early Phase 2 Studies: Expedited Reporting Requirements for Adverse Events that Occur on Studies under an IND/IDE within 30 Days of the Last Administration of the Investigational Agent/Intervention [1, 2]

FDA REPORTING REQUIREMENTS FOR SERIOUS ADVERSE EVENTS (21 CFR Part 312)
NOTE: Investigators **MUST** immediately report to the sponsor (NCI) **ANY** Serious Adverse Events, whether or not they are considered related to the investigational agent(s)/intervention (21 CFR 312.64)
An adverse event is considered serious if it results in **ANY** of the following outcomes:

1) Death
2) A life-threatening adverse event
3) An adverse event that results in inpatient hospitalization or prolongation of existing hospitalization for ≥ 24 hours
4) A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions
5) A congenital anomaly/birth defect.
6) Important Medical Events (IME) that may not result in death, be life threatening, or require hospitalization may be considered serious when, based upon medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent one of the outcomes listed in this definition. (FDA, 21 CFR 312.32; ICH E2A and ICH E6).

ALL SERIOUS adverse events that meet the above criteria MUST be immediately reported to the NCI via electronic submission within the timeframes detailed in the table below.

Hospitalization	Grade 1 and Grade 2 Timeframes	Grade 3-5 Timeframes
Resulting in Hospitalization ≥ 24 hrs	10 Calendar Days	24-Hour 5 Calendar Days
Not resulting in Hospitalization ≥ 24 hrs	Not required	

NOTE: Protocol specific exceptions to expedited reporting of serious adverse events are found in the Specific Protocol Exceptions to Expedited Reporting (SPEER) portion of the CAEPR.

Expedited AE reporting timelines are defined as:

- "24-Hour; 5 Calendar Days" - The AE must initially be submitted electronically within 24 hours of learning of the AE, followed by a complete expedited report within 5 calendar days of the initial 24-hour report.
- "10 Calendar Days" - A complete expedited report on the AE must be submitted electronically within 10 calendar days of learning of the AE.

[1]Serious adverse events that occur more than 30 days after the last administration of investigational agent/intervention and have an attribution of possible, probable, or definite require reporting as follows:
Expedited 24-hour notification followed by complete report within 5 calendar days for:

- All Grade 3, 4, and Grade 5 AEs

Expedited 10 calendar day reports for:

- Grade 2 AEs resulting in hospitalization or prolongation of hospitalization

[2]For studies using PET or SPECT IND agents, the AE reporting period is limited to 10 radioactive half-lives, rounded UP to the nearest whole day, after the agent/intervention was last administered. Footnote "1" above applies after this reporting period.

Effective Date: May 5, 2011

7.3.4 Additional Protocol-Specific Expedited Adverse Event Reporting Exclusions

For this protocol only, the AEs/grades listed below do not require expedited reporting via CTEP-AERS. However, they still must be reported through the routine reporting mechanism (Section 7.4). All events except those specifically mentioned the SPEER and listed below must be reported via CTEP AERS:

- Grade 2 or 3 (<3000/mm3-1000/mm3) White Blood Cell Count Decrease
- Grade 2 or 3 (<1500/mm3-500/mm3) Absolute Neutrophil Count Decrease
- Grade 2 or 3 (<75,000mm/3-25,000/mm3) Platelet Count Decrease

7.3.5 Central Review of Suspected or Confirmed Adverse Events of Interest

In addition to reporting requirements above, for adverse events that are suspicious for GVHD, confirmed to be consistent with GVHD, suspicious for an immune related AE, or confirmed to be consistent with immune related AE, we ask that investigators send pathology reports and relevant primary data including but not limited to surgical or imaging reports (e.g. colonoscopy report for colitis due to GVHD) for central review by the overall PI. If there is a biopsy, we also ask that slides be sent for central review (will be returned). Please also include data on GVHD event description, grading and staging of GVHD (e.g. clinic note). Address to send these items: Attn: Alexandra Savell, Research Project Manager for CTEP 10026; Dana-Farber Cancer Institute; 450 Brookline Avenue; LG-GC; Boston, MA 02215. If there are reports only, please fax to 617-632-5152 or email to: alexandrae_savell@dfci.harvard.edu. PHI must be redacted on reports and imaging when possible. Please see Appendix B for resource that should be used for acute and chronic GVHD grading.

7.4 Routine Adverse Event Reporting

All Adverse Events **must** be reported in routine study data submissions. **AEs reported expeditiously through CTEP-AERS must also be reported in routine study data**

submissions.

Adverse event data collection and reporting, which are required as part of every clinical trial, are done to ensure the safety of patients enrolled in the studies as well as those who will enroll in future studies using similar agents. AEs are reported in a routine manner at scheduled times during the trial using Medidata Rave. For this trial the Adverse Event CRF is used for routine AE reporting in Rave.

7.5 Secondary Malignancy

A *secondary malignancy* is a cancer caused by treatment for a previous malignancy (*e.g.*, treatment with investigational agent/intervention, radiation or chemotherapy). A secondary malignancy is not considered a metastasis of the initial neoplasm.

CTEP requires all secondary malignancies that occur following treatment with an agent under an NCI IND/IDE be reported expeditiously via CTEP-AERS. Three options are available to describe the event:

- Leukemia secondary to oncology chemotherapy (*e.g.*, acute myelocytic leukemia [AML])
- Myelodysplastic syndrome (MDS)
- Treatment-related secondary malignancy

Any malignancy possibly related to cancer treatment (including AML/MDS) should also be reported via the routine reporting mechanisms outlined in each protocol.

7.6 Second Malignancy

A second malignancy is one unrelated to the treatment of a prior malignancy (and is **NOT** a metastasis from the initial malignancy). Second malignancies require **ONLY** routine AE reporting unless otherwise specified.

8. PHARMACEUTICAL INFORMATION

A list of the adverse events and potential risks associated with the investigational or commercial agents administered in this study can be found in Section 7.1.

8.1 CTEP IND Agent: Ipilimumab (NSC 732442 IND #10200)

8.1.1 **Chemical Name or Amino Acid Sequence:** 4 polypeptide chains, 2 identical heavy chains with 447 amino acids and 2 identical light chains consisting of 215 amino acids.

8.1.2 **Other Names:** Anti-CTLA-4 monoclonal antibody, MDX-010, Yervoy™

8.1.3 **Classification:** Human monoclonal antibody

8.1.4 **M.W**.: 147,991 Daltons

8.1.5 **Mode of Action:** Ipilimumab is specific for the CTLA4 antigen expressed on a subset of activated T-cells. CTLA4 interaction with the B7 molecule, one of its ligands expressed on professional antigen presenting cells, can down-regulate T-cell response. Ipilimumab is, thought to act by blocking the interaction of CTLA4 with the B7 ligand, resulting in a blockade of the inhibitory effect of T-cell activation. The CTLA4/B7 creates the interaction.

8.1.6 **Description:** Ipilimumab is a fully human immunoglobulin ($IgG_1\kappa$) with two manufacturing processes – ongoing trials have been using substances manufactured using Process B. New clinical trials will be using ipilimumab that is manufactured by Process C. The Process C has been developed using a higher producing sub-clone of the current Master Cell Bank, and modified cell culture and purification steps.

8.1.7 **How Supplied:** Bristol-Myers-Squibb (BMS) supplies Ipilimumab to the DCTD/NCI. Ipilimumab injection, 200 mg/40 mL (5 mg/mL), is formulated as a clear to slightly opalescent, colorless to pale yellow, sterile, nonpyrogenic, single-use, isotonic aqueous solution that may contain particles.

Each vial is a Type I flint glass vial with gray butyl stoppers and sealed with aluminum seals.

	Process C
Component	**200 mg/ vial**[a]
Ipilimumab	213 mg
Sodium Chloride, USP	249 mg
TRIS-hydrochloride	134.3 mg
Diethylenetriamine pentacetic acid	1.67 mg
Mannitol, USP	426 mg
Polysorbate 80 (plant-derived)	4.69 mg
Sodium Hydroxide	QS to pH 7
Hydrochloric acid	QS to pH 7
Water for Injection	QS: 42.6 mL
Nitrogen[b]	Processing agent

[a]Includes 2.6 mL overfill.
[b]Nitrogen is used to transfer the bulk solution through the pre-filled and sterilizing filters into the aseptic area.

8.1.8 **Preparation:** Ipilimumab is given undiluted or further diluted in 0.9% NaCl Injection, USP or 5% Dextrose Injection, USP in concentrations between 1 mg/mL and 4 mg/mL. Ipilimumab is stable in a polyvinyl chloride (PVC), non-PVC/non DEHP (di-(2-ethylhexyl) phthalate) IV bag or glass container up to 24 hours refrigerated at (2^0 to 8^0 C) or at room temperature/ room light.

Recommended safety measures for preparation and handling include protective clothing, gloves, and safety cabinets.

8.1.9 **Storage:** Store intact vials refrigerated at (2^0 to 8^0 C), protected from light. Do not freeze. If a storage temperature excursion is identified, promptly return ipilimumab to 2°C-8°C and quarantine the supplies. Provide a detailed report of the excursion (including documentation of temperature monitoring and duration of the excursion) to PMBAfterHours@mail.nih.gov for determination of suitability.

8.1.10 **Stability:** Shelf-life surveillance of the intact vials is ongoing. Solution as described above is stable up to 24 hours refrigerated at (2^0 to 8^0 C) or at room temperature/ room light.

CAUTION: Ipilimumab does not contain antibacterial preservatives. Use prepared IV solution immediately. Discard partially used vials.

8.1.11 **Route***(s)* **of Administration:** Intravenous infusion. Do not administer ipilimumab as an IV push or bolus injection.

8.1.12 **Method of Administration:** Can use a volumetric pump to infuse ipilimumab at the protocol-specific dose(s) and rate(s) via a PVC IV infusion set with an in-line, sterile, non-pyrogenic, low-protein-binding filter (0.2 micron to 1.2 micron).

8.1.13 **Patient Care Implications:** Monitor patients for immune-related adverse events, e.g., rash/vitiligo, diarrhea/colitis, uveitis/episcleritis, hepatitis and hypothyroidism. If you suspect toxicity, refer to the protocol guidelines for ruling out other causes.

Post-marketing surveillance identified a fatal toxic epidermal necrolysis (TEN) event in a patient who received ipilimumab after experiencing a severe or life-threatening skin adverse reaction on a prior cancer immune stimulating therapy. Caution should be used when considering the use of ipilimumab in a patient who has previously experienced a severe or life-threatening skin adverse reaction on a prior cancer immune stimulating therapy.

8.1.14 Availability

Ipilimumab is an investigational agent supplied to investigators by the Division of Cancer Treatment and Diagnosis (DCTD), NCI.

Ipilimumab is provided to the NCI under a Collaborative Agreement between the Pharmaceutical Collaborator and the DCTD, NCI (see Section 12.4).

8.1.15 Agent Ordering and Agent Accountability

NCI -supplied agents may be requested by eligible participating investigators (or their authorized designee) at each participating institution. The CTEP-assigned protocol number must be used for ordering all CTEP-supplied investigational agents. The eligible participating investigators at each participating institution must be registered with CTEP, DCTD through an annual submission of FDA Form 1572 (Statement of Investigator), NCI Biosketch, Agent Shipment Form (IDF), and Financial Disclosure Form (FDF). If there are several participating investigators at one institution, CTEP-supplied investigational agents for the study should be ordered under the name of one lead participating investigator at that institution.

In general, sites may order initial agent supplies when a subject is being screened for enrollment onto the study.

Submit agent requests through the PMB Online Agent Order Processing (OAOP) application. Access to OAOP requires the establishment of a CTEP Identity and Access Management (IAM) account, the maintenance of an "active" account status, a "current" password, and active person registration status. For questions about drug orders, transfers, returns, or accountability, call or email PMB any time. Refer to the PMB's website for specific policies and guidelines related to agent management.

8.1.16 Agent Inventory Records – The investigator, or a responsible party designated by the investigator, must maintain a careful record of the receipt, dispensing and final disposition of all agents received from the PMB using the appropriate NCI Investigational Agent (Drug) Accountability Record (DARF) available on the CTEP forms page. Store and maintain separate NCI Investigational Agent Accountability Records for each agent, strength, formulation and ordering investigator on this protocol.

8.1.17 Useful Links and Contacts

- CTEP Forms, Templates, Documents: http://ctep.cancer.gov/forms/
- NCI CTEP Investigator Registration: RCRHelpDesk@nih.gov
- PMB policies and guidelines: http://ctep.cancer.gov/branches/pmb/agent_management.htm
- PMB Online Agent Order Processing (OAOP) application: https://eapps-ctep.nci.nih.gov/OAOP/pages/login.jspx
- CTEP Identity and Access Management (IAM) account: https://ctepcore.nci.nih.gov/iam/
- CTEP Associate Registration and IAM account help: ctepreghelp@ctep.nci.nih.gov
- PMB email: PMBAfterHours@mail.nih.gov

- PMB phone and hours of service: (240) 276-6575 Monday through Friday between 8:30 am and 4:30 pm (ET)
- IB Coordinator: IBCoordinator@mail.nih.gov

8.1.18 Investigator Brochure Availability

The current versions of the IBs for Ipilimumab will be accessible to site investigators and research staff through the PMB OAOP application. Access to OAOP requires the establishment of a CTEP IAM account and the maintenance of an "active" account status, a "current" password and active person registration status. Questions about IB access may be directed to the PMB IB Coordinator via email.

8.2 Commercial Agent: Decitabine

8.2.1 **Product description**: Decitabine for Injection contains decitabine (5-aza-2'-deoxycitidine), an analogue of the natural nucleoside 2'-deoxycytidine. Decitabine is a fine, white to almost white powder with the molecular formula of $C_8H_{12}N_4O_4$ and a molecular weight of 228.21. Its chemical name is 4-amino-1-(2-deoxy-β-D-erythro-pentofuranosyl)-1,3,5-triazin- 2(1*H*)-one. Decitabine is slightly soluble in ethanol/water (50/50), methanol/water (50/50) and methanol; sparingly soluble in water and soluble in dimethylsulfoxide (DMSO). Dacogen (decitabine) for Injection is a white to almost white sterile lyophilized powder supplied in a clear colorless glass vial. Each 20 mL, single dose, glass vial contains 50 mg decitabine, 68 mg monobasic potassium phosphate (potassium dihydrogen phosphate) and 11.6 mg sodium hydroxide.

8.2.2 **Solution preparation** (how the dose is to be prepared): Please refer the reader to the package insert for 'standard' preparation instructions in detail. Highlights include the following: Decitabine is a cytotoxic drug and caution should be exercised when handling and preparing Decitabine. Procedures for proper handling and disposal of antineoplastic drugs should be applied. Several guidances on this subject have been published. Decitabine should be aseptically reconstituted with 10 mL of Sterile Water for Injection (USP); upon reconstitution, each mL contains approximately 5.0 mg of decitabine at pH 6.7-7.3. Immediately after reconstitution, the solution should be further diluted with 0.9% Sodium Chloride Injection, 5% Dextrose Injection, or Lactated Ringer's Injection to a final drug concentration of 0.1 - 1.0 mg/mL. Unless used within 15 minutes of reconstitution, the diluted solution must be prepared using cold (2℃ - 8℃) infusion fluids and stored at 2℃ - 8℃ (36℉ - 46℉) for up to a maximum of 7 hours until administration. Parenteral drug products should be inspected visually for particulate matter and discoloration prior to administration, whenever solution and container permit. Do not use if there is evidence of particulate matter or discoloration. Decitabine (decitabine) for Injection is supplied as a sterile, lyophilized white to almost white powder, in a single-dose vial, packaged in cartons of 1 vial. Each vial contains 50 mg of decitabine.

8.2.3 **Route of administration:** Decitabine is administered at a dose of 20 mg/m^2 by continuous intravenous infusion over 1 hour repeated daily for 5 days. This cycle should be repeated every 4 weeks. Patients may be premedicated with standard anti-emetic therapy.

8.2.4 **Agent Ordering:** The agent is commercially available.

9. BIOMARKER, CORRELATIVE, AND SPECIAL STUDIES

9.1 Pharmacokinetics:

Not required for this present study as the PK of ipilimumab has been well established for the doses being used in the study.

9.2 Correlative studies:

All sites will be encouraged to provide the correlative samples for analysis when possible. While the goal of the biomarkers is to provide supportive data for the clinical study, there may be circumstances when a decision is made to stop a collection, not perform, or discontinue an analysis due to either practical or strategic reasons (e.g., inadequate sample number, issues related to the quality of the sample or issues related to the assay that preclude analysis, impossibility to perform correlative analyses, etc). Therefore, depending on the results obtained during the study, sample collection/analysis may be omitted or added at the discretion of the PI. **We highly encourage all sites to meet the collection requirements to the best of their ability so we can best characterize the biological activity of combination therapy in relapsed/refractory MDS/AML**.

To assist with collection of correlative samples, kits will be prepared and distributed to sites upon activation.

Summary of time points for collection: ***We cannot receive samples over the weekend. Please plan to collect samples Monday-Thursday for delivery Monday-Friday if this is done off-site (not Dana-Farber Cancer Institute). For patients being treated at DFCI, samples can be collected and delivered Monday-Friday.***

Peripheral blood, aspirate (bone marrow), FFPE tissue blocks will be collected for correlative studies following the schedule provided below and in table format in Section 10:

Samples will be collected and cryopreserved at one central site (Dana Farber Cancer Institute, Pasquarello Laboratory) for all assays described. Samples will be processed according to the accompanying laboratory study manual. Samples will be otherwise stored and labeled (PHI removed, sample # assigned for HIPAA) in the Pasquarello Laboratory at temperature as required per sample (FFPE, at room temperature; aspirate in liquid nitrogen and plasma in -80C). Samples for specific assays will be appropriately delivered in batches for analysis (including for IHC). For the samples below, prioritization will be made for genotyping, RNA sequencing of immune infiltrates, flow cytometry, CYTOF, and other serial immune studies (including expression of checkpoint inhibitors, cytokine/chemokine patterns). All other samples will be saved for future studies including examination of tumor driven immune response and evaluation of methylation patterns underlying response. Samples will be collected and sent from all participating institutions to the Pasquarello Laboratory (Hematologic Malignancies Tissue Banking Laboratory) at the Dana-Farber Cancer Institute, under the direction of Dr. Jerome Ritz.

Address and phone number are provided:
Dana-Farber Cancer Institute
Pasquarello Tissue Bank-J604
450 Brookline Ave.
Boston, MA 02215
Phone Number: 617-582-8214
For more information, the Overall PI can be contacted (Jacqueline Garcia, MD) at 617-632-6349 during business hours, and afterhours or weekends can be paged at 617-632-3352 (pager #44038).

9.3 Cancer Immune Monitoring and Analysis Centers (CIMAC) Plan for Correlative Samples

The following is a summary of assays and scientific objectives that will be performed on the samples collected from this study as approved by the CIMAC leadership. Assays will be performed subject to availability of samples:

Priority	Biomarker Name	Assay- CLIA: Y/N	Use in protocol (integral, integrated, or exploratory?) Purpose	Sample Types and Time points of sample collection	M/ O	Sample Usage	Investigator/ Assay Lab
1	Multiplex IF panel[4, 5]	mIF (N) with custom-built panel to identify leukemic myeloid blasts and immune marker CD8, CD4 and PD-1.	•Exploratory •To identify biomarkers of immune response if there is evidence of T Cell Activation •This is dependent on findings from CD4 and CD8 IHC studies. •If cannot create custom built then will use: FoxP3, PD-1, DAPI, either CD8/CD4/CD3 and either SOX10/PAX5/Cytokeratin	•FFPE -Tumor •Pre Tx (baseline) •End of lead In •End of Cycle 2 •End of Cycle 4	M	5 unstained FFPE slides at each timepoint	DFCI CIMAC Scott Rodig, MD
6	T cell Receptor[1,4]	TCR Sequencing CLIA: N	•Integrated •To characterize clonality of T cell repertoire before/after therapy and to identify if T cell receptor diversity impacts clinical response. We hypothesize that high TCR diversity will associate with durable response.	•PBMCs from EDTA Tubes •Pre Tx (baseline) •End of lead In •End of Cycle 2 •	M	Minimum 1 million PBMCs (1 vial)	DFCI CIMAC: Catherine Wu, MD
7	Somatic myeloid mutation analysis	NGS CLIA:Y	•Integrated •To evaluate for changes in mutational abundance and clonal presence and associate with response. We hypothesize that mutational clearance will associate with durable response.	•BMMCs from EDTA Tubes •Pre Tx (baseline) •End of Cycle 4 •End of Treatment / Progression	M	2cc marrow (1 vial)	Non-CIMAC: BWH CAMD (Kim/M Baltay)
8	Immune profiling – T cell immunogenicity[1]	ELISA CLIA: N	•Exploratory •To evaluate for leukemia specific T cell response in responders versus non-responders. We hypothesize that decitabine will make blasts more visible to T cells.	•PBMCs from EDTA Tubes •Pre Tx (baseline) •End of lead In •End of Cycle 1 •End of Cycle 4	M	Minimum 3 million PBMCs (1 vial)	Non-CIMAC: Catherine Wu, MD
9	Methylation of immune infiltrates & leukemic blasts[1,4]	RRBS CLIA: N	•Exploratory •To evaluate for hypomethylation of immune gene sets in response to therapy. •Separate assessment of blasts and tumor immune infiltrates. We hypothesize that decitabine increases checkpoint inhibitor expression in blasts and immune gene set expression in tumor immune infiltrates.	•BMMCs from EDTA Tubes •Pre Tx (baseline) •End of Lead In •End of Cycle 2	M	Minimum 1 million BMMCs (1 vial)	Non-CIMAC: Catherine Wu, MD

10	Open chromatin at immune gene sets[1,4]	ATAC-Seq CLIA: N	•Exploratory •To evaluate for open chromatic at immune gene sets in response to therapy using tumor immune infiltrates We hypothesize that open chromatin at immune gene sets will inform responsiveness.	•BMMCs from EDTA Tubes •Pre Tx (baseline) •End of Lead In •End of Cycle 2	M	Minimum 1 million BMMCs (1 vial)	Non-CIMAC: Catherine Wu, MD
11	Sequencing of immune infiltrates & leukemic blasts[1,4]	RNA-seq CLIA: N	•Exploratory •To evaluate for biomarkers of responsiveness in tumor versus immune infiltrates as it relates to alloreactive tumor environment (transplant naïve vs post-transplant) •Separate assessment of blasts and tumor immune infiltrates We hypothesize that there will be genetic biomarkers of responsiveness that are unique to post-transplant environment.	•BMMCs from EDTA Tubes •Pre Tx (baseline) •End of Lead In •End of Cycle 2	M	Minimum 1 million BMMCs (1 vial)	DFCI CIMAC Catherine Wu, MD
12	Immune profiling[1,4]	CyTOF CLIA: N	•Exploratory •To quantify and determine the functional activation of immune and myeloid markers as it relates to response. •Using a custom myeloid/immune panel (developed by laboratory of DFCI Dr. Jerome Ritz lab). We hypothesize there will be a relative increased T cell presence and decreased T regulatory cells in responders compared to non-responders.	•PBMCs from EDTA Tubes •Pre Tx (baseline) •End of lead In •End of Cycle 4	M	3-5 million PBMCs (1 vial)	DFCI CIMAC Steve Hodi, MD
13	Sequencin[1, 3]	WES CLIA: N	•Exploratory •To identify for expression of novel antigens after combination decitabine/ipilimumab that correlates with response. We hypothesized that responders to treatment will express novel neoantigens as a consequence of combination decitabine and ipilimumab exposure.	•Germline Skin and Tumor and BMMC •At any time collected for disease assessment, or for evaluation of toxicity (paired)	O	Minimum 1 million BMMCs (1 vial)	DFCI CIMAC Catherine Wu, MD
14	Cytokine profile	Olink CLIA: N	•Exploratory •To evaluate cytokine profiles as a marker of T cell activation in responders versus non-responders. We hypothesize that there will be increased interferon gamma and chemokines that enable leukocyte trafficking in patients with increased T cell infiltrates.	•Plasma from EDTA Tubes •Pre Tx (baseline) •End of Cycle 1 •End of Cycle 2	M	1 vial (plasma)	Mt Sinai CIMAC

15	MDSC[2] and tumor specific immunity (cytolytic capacity)	Flow CLIA: N	•Exploratory •To evaluate presence of MDSCs as it relates to T cell response We hypothesize that decitabine will decrease MDSC presence resulting in increased T cells in presence of ipilimumab.	•PBMCs from EDTA Tubes •Pre Tx (baseline) •End of lead In •End of Cycle 2	M	5-10 million PBMCs	Avigan Lab – BIDMC

[1] Provided there will be specimen available

[2]*MDSC and leukemia specific immunity (cytolytic capacity) will be performed by the laboratory of Dr. David Avigan at Beth Israel Deaconess.* PBMCs from AML patients will be isolated by Ficoll density-gradient centrifugation and stained with antibodies for CD11b, HLA-DR, CD14, CD15, and CD33 expression. The cells will then be analyzed with flow cytometry. If present, tumor cells will be gated out on the basis of forward scatter/side scatter and known blast phenotype, and total MDSCs ($CD33^+CD15^-$ or $CD33^+CD15^+$) will be quantified as a percentage of total cells and as a percentage of gated immature $CD11b^+/HLA\text{-}DR^-$ myeloid cells. MDSCs will be further characterized as granulocytic, by the presence of $CD15^+$, or monocytic, by $CD15^-$ and side scatter

[3] Tumor and germline controls for WES will be patient-specific. PBMCs will be used as a tumor specimen if there is a leukemic burden in the blood. Otherwise, FFPE tumor tissue will be used if available. Germline DNA samples (used for WES) may be bone-marrow-derived mesenchymal stromal cells, or, if not available, tissue/PBMCs from archival pre-transplant remission timepoint. From the latter source of germline DNA, computational tools may be used to estimate the amount of tumor contamination of normal tissue (e.g. DeTiN from Taylor-Weiner A, et al. Nat Methods 2018) to improve mutation detection.

[4] Additional timepoints may be analyzed for blood-based assays if specimens are available and the CIMAC and trial PI agree.

[5] Leukemia based assays will be performed on leukemia cells in the peripheral blood, bone marrow aspirates, or in FFPE-preserved tissue biopsies (bone marrow, lymph node, skin, etc) depending on the assay type and location of the leukemia burden in the participant

[6] Mandatory or Optional

9.3.1 **Bone Marrow Aspirates**

From each bone marrow aspirate, ~10 x 10^7 mononuclear cells, divided into aliquots of 1 x 10^7 cells, will be RBC lysed and then snap frozen as cell pellets. Pellets will be stored in liquid nitrogen at -80C. We anticipate that following will be isolated from the cell pellets:

1. RNA, including mRNA, to be used for (in order of priority):
    a. RNA-sequencing of immune cells (CD3+), tumor (CD34+), and TCR-sequencing
2. Genomic DNA to be used for (in order or priority):
    a. Through CIMAC-independent process: NGS to determine somatic mutations (will use Rapid Heme Panel at the Brigham & Women Hospital with the Center for Advanced Molecular Diagnostics (CAMD) that evaluates 95 genes (includes most common myeloid mutations) and hot spot codons for oncogenes and entire coding regions for tumor suppressors using an NGS platform)
    b. Through CIMAC-independent process: DNA methylation sequencing
    c. Through CIMAC-independent process: Cytolytic capacity of tumor immune cells in collaboration with the Avigan Laboratory at the Beth Isreal Deaconness Medicla Center
    d. Whole Exome DNA sequencing, for serial tracking of mutation load and development of new mutations which may be associated with resistance

Remaining aspirate may be used for additional assays of potentially relevant biomarkers, depending upon reagent and/or tissue availability, and on responses seen in patients.

9.3.2 **Whole Blood - Peripheral blood mononuclear cells**

Peripheral blood mononuclear cells will be purified using the standard Ficoll gradient method and if not used immediately, cryopreserved in standard freezing media and stored in liquid nitrogen.

1. Flow cytometry to examine markers related to myeloid differentiation and T cell subsets including isolating specific cell populations of interest (CD34+ blasts, T cells)
2. Mass cytometry (CyTOF) analysis
3. Through CIMAC-independent process: Cytolytic capacity of tumor immune cells and phenotyping of MDSCs in collaboration with the Avigan Laboratory at the Beth Isreal Deaconness Medicla Center

Remaining peripheral blood may be used for additional assays of potentially relevant biomarkers, depending upon reagent and/or tissue availability, and on responses seen in patients.

9.3.3 **FFPE tissue blocks**

In collaboration with the Brigham and Women Pathology Core Facility and DFCI CIMAC Tissue Biomarker laboratory of Dr. Scott Rodig, minimum IHC staining for CD34, CD3, CD8, granzymeB, CD4. A second panel of IHC staining will be recommended following initial IHC panel analysis, which may include but is not limited to pending findings: PD-1, PD-L1, CTLA-4 or markers of CD4 subsets. Formalin Fixed Paraffin Embedded (FFPE) samples of bone marrow core (or extramedullary tumor where it applies) or unstained slides will also be obtained from the Brigham and Women's pathology department that have been archived as part of standard tissue banking and tandem sample collection protocols. The FFPE samples collected for the purpose of the clinical trial and as part of parallel tissue banking are stored at the Brigham and Women's Pathology department.

9.3.4 **Plasma**

Plasma will be stored at -80C. Plasma samples will be used to measure cytokines serially using the multiplex protein assays and ELISAs on Luminex platforms and Olink platform.

9.3.5 **Skin biopsies**

Skin biopsies for (1) leukemia cutis and (2) germline DNA assessment will be requested from exceptional responders (from any part of the study) and from some non-responders (at dose-expansion) to allow for paired analysis with tumor DNA after approval from overall PI. This is optional and individual cases that will be requested will be determined by the overall PI.

9.4 Pharmacodynamic Study

Objective #1: Measuring absolute lymphocyte count as a potential biomarker for response to ipilimumab. We will compare measured levels at baseline (defined as prior to dosing on cycle 1 day 1) and at pre-determined time points (end of cycles 1, 2, and 4) and we will also compare changes in ALC from baseline (defined as prior to dosing on cycle 1 day 1) to the pre-determined time point (end of cycle 2) for survival differences given the evidence that ALC after two cycles may be a predictor of clinical response. As decitabine may cause myelosuppression, ALC elevation may not be reliable. We will compare measured levels at baseline (defined as prior to dosing on cycle 1 day 1) and at pre-determined time points (end of cycles 1, 2, and 4) and we will also compare changes in ALC from baseline (defined as prior to dosing on cycle 1 day 1) to the pre-determined time point (end of cycle 2) for survival differences given the evidence that ALC after two cycles may be a predictor of clinical response.
-**Time-points**: baseline, end of cycles 1, 2, and 4

9.5 Exploratory/Ancillary Correlative Studies

Please see Rationale for correlative studies provided in section 2.

Correlative study objective
Objective 1: To determine clonal patterns of patients on treatment
Whole exome sequencing will be performed on the baseline bone marrow aspirate samples and

bone marrow-derived fibroblasts for germline tissue to determine 1) the presence of known recurrent mutations in AML and 2) presence of novel mutations not previously associated with AML. The response to therapy will be correlated with presence of known recurrent mutations, with particular attention paid to mutations associated with baseline treatment resistance or exceptional response. If no correlation is found between treatment response and known recurrent mutations, we will extend our analysis to include any novel mutations found in either patients with resistant disease or those with exceptional response. We will also identify neoantigens to determine if neoantigen load correlates with clinical and/or immunologic response. If we are unable to perform WES, we will use the Rapid Heme Panel as a way to capture common myeloid mutations and variant allele frequency. This assay is routinely used as standard of care testing for newly diagnosed or relapsed patients with hematologic malignancies. Mutation variant allele frequency for both those mutations present at screening and those arising during treatment will be followed serially, given the known role of mutation frequency with treatment response in other contexts. This will be performed on baseline samples, after four cycles of combination therapy and at time of relapse if relevant.
-**Time-points for collection**: baseline (screening), end of cycle 4, and time of relapse

Objective 2: To determine histopathologic patterns of response
We hypothesize that infiltrating T cells at the site of tumor microenvironment is critical to tumor control. Samples to be tested include marrow and extramedullary site (if amenable to biopsy) including skin in leukemia cutis. Using immunohistochemistry, we will evaluate for activated T-cells by CD8/granzyme B double IHC at baseline and post-treatment. We will also evaluate for CD4, PD-1, and PD-L1 expression. We will collaborate with Dr. Scott Rodig and his core laboratory at the Brigham and Women's Hospital to perform these evaluations. As biopsies will otherwise be required per protocol at additional time points, we will capture IHC responses at later time points if we see evidence for delayed biological activity. Archival tissue from patients on study may be utilized or requested as necessary.
-**Sample type:** formalin-fixed, paraffin-embedded (FFPE) samples for analysis including core biopsies, skin biopsies and tumor biopsies (extramedullary)
-**Time-points for collection**: baseline (screening), on treatment (proposed: end of cycle 2 and end of cycle 4), time of relapse if relevant
-Correlative to be performed in: Laboratory of Dr. Scott Rodig (Brigham and Women's Hospital, Attn: Rodig Lab, Thorn Building #603B, 20 Shattuck Street, Boston, MA 02115).
-**Total anticipated # samples:** ~ up to 4 per patient

Objective 3: To evaluate the phenotype of immunologic consequences of combination treatment
CTLA-4 expression is induced upon T cell activation. We hypothesize that epigenetic therapy may modulate T cell activation and that the addition of immunotherapy will give rise to an increased number of CD4 and CD8 T cells infiltrating the tumor sites. Further, there is limited data available on the immune response to epigenetic therapy. Exploratory studies from the recent ipilimumab study in the post allo-HCT setting showed that CD4 Tregs were decreased and CD4 Tcons (effector cells) were increased in responders 8 weeks after ipilimumab. Further characterization of both dcreased Treg and increased Tcon at 8 weeks showed that they were compromised predominantly by the most mature subsets (effective memory). Naive and central memory (CM) subsets of Treg and Tcon were decreased in responders at 8 weeks. Further

studies are thus needed to understand these changes in T cell subsets and to correlate these changes with response.

(A) Flow cytometry: Proliferation, cytokine, and immune checkpoint inhibitor induction by T-cells will be serially examined. Fluorochrome conjugated antibodies will be added to each reaction tube followed by the addition of 100ul of whole blood. After incubation, BD FACSLyse will be added to each reaction tube, and the sample will be vortexed and incubated. Antibody-labeled cells will then be fixed using formaldehyde and analyzed using a BD FACSCanto II plus with BD FACSDiva software. If by conventional flow cytometry we see evidence of T cell activation, we will then move those samples forward for further interrogation using CyTOF to explore subpopulations.

(B) CyTOF: IWe hypothesize that epigenetic therapy and checkpoint inhibition may result in at least partial restoration of normal hematopoietic developmental programs and subsequent reversal of the aberrant immunophenotype. AML cells also have altered intracellular signaling which can be measured by mass cytometry. The DFCI CIMAC Immune Assessment Lab offers CyTOF; a single-cell proteomic technology that allows for the analysis of 40+ cellular parameters using antibodies tagged with heavy metal isotopes to elucidate complex phenotypic and functional characteristics of heterogeneous immune populations. We will use a custom designed panel of surface membrane and intracellular markers to assess the number and activation status of Treg, T_{EM}, T_{CM}, T_{naive}, and NK cells, including but not limited to CTLA-4, ICOS, PDL-1, TIM-3. The CyTOF panel that is available is included below:

CyTOF Panel – Focus on Treg and cytokine signaling pathways

	TARGET	SPECIES	CLONE	ISOTOPE	Manufacturer
	Surface				
1	CD45	Human	HI30	154Sm	Fluidigm
2	HLA-DR	Cross	L243	141Pr	Biolegend
3	CD95	Human	DX2	164Dy	Fluidigm
4	CD3	Human	UCHT1	170Er	Fluidigm
5	CD4	Human	SK3	174Yb	Fluidigm
6	CD8	Human	SK1	168Er	Fluidigm
7	CD25	Human	2A3	149Sm	Fluidigm
8	CD127	Human	A019D5	176Yb	Fluidigm
9	CD45RA	Human	HI100	169Tm	Fluidigm
10	CD122 (IL2Rβ)	Human	Tu27	143Nd	BioLegend
11	CD132 (IL2Rγ)	Human	TUG h 4	166Er	BioLegend
12	CCR7	Human	G043H7	173Tb	BioLegend
13	CD62L	Human	DREG-56	153Eu	Fluidigm
14	CD31	Human	WM59	145Nd	Fluidigm
15	CD279(PD1)	Human	EH12.2H7	175Lu	Fluidigm
16	CD19	Human	HIB19	142Nd	Fluidigm
17	IgD	Human	IA6-2	146Nd	Fluidigm
18	CD38	Human	HIT2	144Nd	Fluidigm

19	CD27	Human	L128	155Gd	Fluidigm
20	BAFF-R(CD268)	Human	11C1	160Gd	Biolegend
21	CD56	Human	HCD56	162Dy	Biolegend
22	CD16	Human	3G8	148Nd	Fluidigm
23	NKG2D (CD314)	Human	1D11	161Dy	Fluidigm
24	ICOS (CD278)	Human	DX29	147sm	BD
	Intracellular				
25	Foxp3	Human	PCH101	165Ho	eBioscience
26	BCL-2	Human	Bcl-2/100	171Yb	BD Bioscience
27	Ki67	Cross	B56	151Eu	BD Bioscience
28	BIM	Cross	Polyclonal	159Tb	BD Bioscience
29	Helios	Cross	22F6	156Gd	Biolegend
30	CD152(CTLA4)	Human	14D3	163Dy	eBioscience
	Signaling				
31	pStat5 [Y694]	Cross	47	150Nd	Fluidigm
32	pStat3 [Y705]	Cross	4/P-Stat3	158Gd	Fluidigm
33	pERK 1/2 [T202/Y204]	Cross	D13.14.4E	167Er	Fluidigm
34	pS6 Ribo(S235/236)	Cross	N7-548	172Yb	Fluidigm
35	p-AKT	Cross	S473	152Sm	Fluidigm

(C) Chemokine and cytokine profiling: Levels of cytokines involved in inflammation, tolerance and Th1, Th2 and Th17 responses will be measured in plasma samples using Luminex and Olink platforms. From whole blood, we will perform serial immunophenotypic analysis of immune cells using ELISA to assess chemokine/cytokine profiles (CXCL2, CXCL5, MICA, and others) to assess whether we observe similar results in patients treated with combination ipilimumab/ decitabine. The Proseek Olink Proteomics platform operating on a Fluidigm Biomark HD microfluidic PCR system is a quantitative and reproducible assay to measure levels cytokines, chemokines, growth factors, in addition to detection of circulating immune co-stimulatory and inhibitory molecules, and other relevant immune-oncology markers, in a 92-plex format known as the Olink Immuno-Onology panel, which only requires 1 micro-liter of sample.

-**Sample type:** whole blood or aspirate (flow cytometry and CYTOF), plasma (Luminex)

-**Time-points for collection**: baseline (screening), end of the lead in, end of cycle 1, end of cycle 2 and end of cycle 4

-Correlative to be performed in:

- Flow cytometry and CyTOF: Laboratory of Dr. Jerome Ritz (Dana-Farber Cancer Institute Pasquarello Tissue Bank-J604, 450 Brookline Ave., Boston, MA 02215)
- Chemokine and cytokine multiplex assays: Center for Immuno-Oncology (Dana-Farber Cancer Institute, D2-131, 450 Brookline Avenue, Boston, MA 02215)
- Olink Immuno-Onology panel: Mount Sinai CIMAC, Icahn School of Medicine at Mount Sinai;Hess Center for Science and Medicine, 5th floor, rooms 310/313 Human Immune Monitoring Center (HIMC), 1470 Madison Avenue, New York, NY 10029

-**Total anticipated # samples:** ~ up to 4-5 per patient

Objective 4: To characterize the genotypic immunologic consequences of combination treatment

Epigenetic priming and checkpoint blockade have been shown to have a variety of immunologic effects as described above. We hypothesize that any observed therapeutic effect may be partially due to immunomodulatory effects on lymphocytes in addition to effects on the AML cells. We will evaluate these changes by evaluation of the leukemic transcriptome to infer changes to immune response. We will utilize whole exome sequencing or bulk versus single cell transcriptomics to elucidate the heterogeneity of immune and cancer cell populations before and after therapy to understand how leukemic and immune populations co-evolve during response or resistance. We will evaluate the gene expression pattern of immune infiltrates to understand differences in the tumor microenvironment in situ. We will evaluate samples collected at baseline (prior to dosing on study) and on treatment (at the end of cycles 2).

-**Sample type:** aspirate

-**Time-points for collection**: baseline (screening), end of cycle 1 and end of cycle 2 (will collect and hold samples from cycle 4 for analysis depending on findings)

-**Correlative to be performed in:** The DFCI CIMAC translational immuno-genomics laboratory of Dr. Catherine Wu (Dana-Farber Cancer Institute, Dana 540B, 450 Brookline Avenue, Boston, MA 02215) will prepare the respective DNA/RNA, and sequencing will be performed at the following external lab:

Broad Institute Genomics Services
320 Charles Street
Cambridge, MA 02141

Sequencing data will then be provided back to the Wu Lab and in the form of BAM files. The samples that are sent for analysis are exhausted during the process, and thus not able to be returned.

-**Total anticipated # samples:** ~ up to 4 per patient

Objective 5: To quantitate regulatory T cell subsets and to evaluate for tumor reactive lymphocytes activity and for tetramer binding.

In collaboration with Drs. David Avigan and Jacalyn Rosenblatt, we will perform flow cytometry-based analysis of peripheral-blood mononuclear cells to evaluate for (1) T cell subsets and expression of inhibitory immune receptors on different immune cells, (2) quantification of circulating CD4/CD8 populations, and (3) levels of circulating regulatory T cells and tumor reactive lymphocytes (collaboration with Dr. Ritz and Drs. Avigan/Rosenblatt). Peripheral blood samples will be analyzed by FACS-gating to determine the percentage of activated effector cells (using CD4/CD25+ fraction and CD69 expression) and regulatory T cells (measuring expression of Foxp3, GITR, CTLA-4, CD127, and CD62L on the CD4/CD25+ population). We will then stain for CD45RA to evaluate for the naive memory cell prevalence and CD45RO, CCR7 and CD28 for memory cell response. We will evaluate if *ex vivo* immunologic response correlates with levels of memory effector and regulatory T cells. We will evaluate the immunomodulatory effect of decitabine alone and in conjunction with checkpoint blockade in AML by evaluating for (1) transcriptional induction of and (2) binding to leukemia-associated antigens as suggestion of

enhanced presentation of leukemia antigens to T cells. We will evaluate if decitabine enhances antigen presentation. There is preclinical evidence that HMA therapy enhances MHC class I and co-stimulatory molecule expression which may increase recognition of leukemia-associated antigens (LAA). We will use real-time quantitative polymerase chain reaction to measure leukemia-associated antigens such as WT1, MUC1, and PR3, which may be targets for AML immunotherapy. In collaboration with Drs. Avigan/Rosenblatt, we will evaluate the percentage of CD8+ T cells binding tetramers constructed from these antigens and for functional activity in the tetramer-positive population by measuring corresponding levels of IFN-gamma, granzyme, IL-4, and IL-10 expression by FACS staining.
-Sample type: aspirate and peripheral blood
-Time-points for collection: baseline (screening), end of the lead in, and end of cycle 2.
-Correlative to be performed in:
Dr. David Avigan's laboratory at Beth Israel Deaconess Medical Center
Address: 3 Blackfan Circle CLS 724 Boston, MA 02215
-Total anticipated # samples: up to 3 per patient

Similar tests will also be run on tissue samples for any patient requiring a lymph node or bone marrow biopsy for clinical reasons. No extra procedures will be done for the purpose of this study, but if these tests are being done for standard of care evaluations, an extra sample may be taken to use for the above research evaluations. Extra pull of aspirate (~5cc) at any non-scheduled bone marrow evaluation will be accepted for future studies.

Objective 6: To evaluate the genomic and epigenomic state of tumor immune infiltrates and leukemic blasts before and after decitabine priming and after ipilimumab treatment.

From paired samples, we will perform reduced representation bisulfite sequencing to evaluate DNA methylation changes and ATAC-sequencing to evaluate for chromatin accessibility. These assays will be performed in Dr. Catherine Wu's laboratory.
-Sample Type: aspirate
-Time Points for collection: baseline (screening), end of decitabine monotherapy lead in month, end of cycles 1 or 2.
-Correlative to be performed in: Dr. Wu's laboratory at Dana-Farber Cancer Institute will prepare the DNA library, and sequencing will be performed at the following external lab:

Broad Institute Genomics Services
320 Charles Street
Cambridge, MA 02141

Sequencing data will then be provided back to the Wu Lab in the form of BAM files. The samples that are sent for analysis are exhausted during the process, and thus not able to be returned.

-Total anticipated # samples: up to 4 per patient

10. STUDY CALENDAR

10.1 Study Schedule

Baseline evaluations and screening labs/procedures performed are to be conducted within 2 weeks prior to start of protocol therapy except for serum pregnancy in women of childbearing potential which must be repeated if resulted > 72 hours prior to treatment start. Baseline evaluation can be used as screening for eligibility if performed within 1 week of starting protocol therapy. If treatment start occurs within 48 hours of registration, then baseline evaluation does NOT need to be repeated to confirm eligibility. Scans, EKG and echocardiogram (or MUGA scan) are allowed within 4 weeks of treatment start (day 1 of the lead in period). In the event that the patient's condition is deteriorating, laboratory evaluations should be repeated within 48 hours prior to initiation of the next cycle of therapy. Please see screening table below for specific windows for diagnostic tests including bone marrow and PET.

Screening, Priming and Induction Period												
	Pre-Study	**Lead In D1**	**Lead In D15[b]**	**Lead In D24[a]**	**C1 D1[b]**	**C1 D15[b]**	**C1 D24[a]**	**C2 D1[b]**	**C2 D24[a]**	**C3 D1[b]**	**C4 D1[b]**	**C4 D24[a]**
Ipilimumab[A]					X			X		X	X	
Decitabine[B]		X			X			X		X	X	
Informed consent	X											
Demographics	X											
Medical history	X											
Concurrent meds	X	X	X		X	X		X		X	X	
Physical Exam	X	X	X		X	X		X		X	X	
Vital signs	X	X	X		X	X		X		X	X	
Height	X											
Weight	X	X			X			X		X	X	
Performance status	X				X			X			X	
CBC w/diff, plts	X	X	X	X	X	X	X	X	X	X	X	X
Serum chemistry[c]	X	X	X		X	X		X		X	X	
Immune-related labs[d]	X				X						X	
Adverse event evaluation[e,q,r]	X		X		X	X		X		X	X	
GVHD Assessment	X				X			X		X	X	
PET[f]	X[f]								X[f]			X[g]
B-HCG[h]	X[h]											
Bone marrow[i]	X			X[j]			X[i]		X[i]			X[i]
Skin biopsy[k]	X[k]						X[k]		X[k]			X[k]
T cell chimerism[l]	X			X			X		X			X
Hepatitis Panel[m]	X											
HIV testing[n]	X											
Echocardiogram[o,q,s]	X											
EKG[o,q]	X											
Correlative studies[p]	X			X			X		X			X

A: Ipilimumab: Dose as assigned by dose level. Administered every 4 weeks. Dosed per weight on day 1 of a cycle.
B: Decitabine: 20 mg/m2 daily IV for 5 consecutive days on days 1-5. Dosed per weight on day 1 of the lead in period
a: Can be done any time between day 24-28
b: +/- 3 days. If there is evidence of persistent disease or blood counts meet criteria to treat, final bone marrow results are not required prior to dosing on day 1 of next cycle.
c: Albumin, alkaline phosphatase, total bilirubin, bicarbonate, BUN, calcium, chloride, creatinine, glucose, potassium, total protein, SGOT [AST], SGPT [ALT], sodium, uric acid, magnesium, phosphorus.
d: Every 3 months unless clinically indicated: gamma-GT, direct bilirubin, LDH, TSH, and lipase
e: Please see Appendix F for helpful study check list including immune related AE evaluation
f: PET scans are required only for those with sole extramedullary disease (no marrow involvement by morphology) and optional for those with extramedullary and marrow involved AML. PET scans are standard imaging tests for those with extramedullary disease involvement. Study can be performed any time between day 24-28 at the end of cycles 2 and 4 (suggested time frame). However as clinical scenario may vary with patients with sole extramedullary disease, we will allow imaging window of +/- 4 weeks based on clinical scenario or scheduling logistics. Please send report and imaging (on CD) with redacted PHI for central review of extramedullary AML events to Attn: Alexandra Savell, Research Project Manager for CTEP 10026; Dana-Farber Cancer Institute; 450 Brookline Avenue; LG-GC; Boston, MA 02215. If there are reports only, please fax to 617-632-5152 or email to: alexandrae_savell@dfci.harvard.edu
g: If patient has sole extramedullary disease and there was no resolution of FDG-avid disease by the end of cycle 2, then patients should have a repeat PET at the end of cycle 4 per standard of care- Study can be performed any time between day 24-28 at the end of cycles 2 and 4 (suggested time frame). However as clinical scenario may vary with patients with sole extramedullary disease, we will allow imaging window of +/- 4 weeks based on clinical scenario or scheduling logistics. Please send report and imaging (on CD) with redacted PHI for central review of extramedullary AML events to Attn: Alexandra Savell, Research Project Manager for

CTEP 10026; Dana-Farber Cancer Institute; 450 Brookline Avenue; LG-GC; Boston, MA 02215. If there are reports only, please fax to 617-632-5152 or email to: alexandrae_savell@dfci.harvard.edu

h: Serum pregnancy test (women of childbearing potential).- Negative results required within 72 hours of treatment start

i: For bone marrow biopsies, the minimum requested includes (1) aspirate (if aspirable), (2) core, (3) flow cytometry (if sufficient blasts; may not be possible in setting of low blasts or remission; if MRD flow cytometry is an option we will accept this data in lieu of standard flow cytometry if this test is available at the time of suspected remission), (4) cytogenetics if possible, (5) molecular testing per institutional guidelines (suggested at minimum at time of screening and relapse but not required; correlatives will include genotyping leukemic blasts). Core biopsy is mandatory at screening, at the end of cycles 2 and 4- See sample collection table in section 10.2 for details. If a diagnostic bone marrow biopsy was performed within 4 weeks of treatment start and no anti-leukemic therapy has been given (except for hydrea and radiation), then flow cytometry and cytogenetics do not need to be repeated. If a diagnostic bone marrow was performed within 2 weeks of treatment start, then only a research aspirate and research core are required to be collected during screening. In cases of persistent leukemic blood, bone marrow biopsy can be performed on day 1 of subsequent cycle prior to treatment.

j: Aspirate ONLY for correlative study (unless inaspirable). See Correlative studies.

k: For those with leukemia cutis only - repeat evaluation at end of cycles 1, 2 and/or 4 (pending response)- If there is suspected resolution or response based on skin exam, biopsy may occur sooner as clinically indicated. After confirmation of resolution of lesion, there does not need to be a repeat biopsy at the same site. If there is concern for persistence of disease based on skin examination, then repeat biopsy is recommended at the end of cycles 1, 2 and/or 4 if it can be safely performed.

l: Routine response evaluation for patients in Arm A who are status post allo-HCT only. Peripheral blood source only. Can be performed locally and if not available locally then should be analyzed at site where patient's transplant was performed.

m: For those with known HBV they should get an HBV viral load (PCR). For those with known HCV they should get an HCV viral load (PCR). It is recommended that patients with known HBV get monthly (+/- 14 days) HBV viral load assessments after ipilimumab is initiated. Please update overall PI if there is significant or clinical concern for HBV re-activation despite anti-viral therapy.

n: For those with known HIV they must get a CD4 count and an HIV viral load (PCR).

o: Baseline echocardiogram and EKG required for screening (if done within 4 weeks of treatment start does not need to repeat unless there is a clinical concern) and at any other time that is clinically indicated during the trial as it relates to footnote q

p: Peripheral blood, aspirate, core biopsy, or FFPE to be collected per schedule described correlatives table below.

q: Alert PI if there is concern for cardiac toxicity (minimum assessment: cardiology consult, EKG, echocardiogram, and labs CPK and troponin; refer to section 6.4.7)

r: Please see section 7.3.5 of the protocol for reporting requirements for GVHD and suspected immune related AEs to enable central review

s: MUGA scan is a suitable alternative to an echocardiogram

t: For participants enrolled on Arm A (post transplant), a formal GVHD assessment is required at minimum on Day 1 of each cycle utilizing the GVHD assessment worksheet provided in Appendix B

Maintenance cycles and end of study											
	C5 D1[b]	**C6 D1[b]**	**C7 D1[b]**	**C7 D24[a]**	**C8 D1[b]**	**C9 D1[b]**	**C10 D1[b]**	**C10 D24[a]**	**C11 D1[b]**	**C12 D1[b]**	**End of study[i,j]**
Ipilimumab[A]	X		X			X			X		
Decitabine[B]	X	X	X		X	X	X		X	X	
Concurrent meds	X	X	X		X	X	X		X	X	
Physical Exam	X	X	X		X	X	X		X	X	X[k]
Vital signs	X	X	X		X	X	X		X	X	
Height	X										
Weight	X	X	X		X	X	X		X	X	
Performance status	X	X	X		X	X	X		X	X	
CBC w/diff, plts	X	X	X	X	X	X	X	X	X	X	X
Serum chemistry[c]	X	X	X		X	X	X		X	X	X
Immune-related labs[d]			X				X				X
Adverse event evaluation[e,l,m]	X	X	X		X	X	X		X	X	X[k]
GVHD Assessment	X	X	X		X	X	X		X	X	X
Bone marrow[f]				X[f]				X			X[f]
T cell chimerism[g]											X
Correlative studies[h]				X				X			X

A: Ipilimumab: Dose as assigned by dose level. Administered every 8 weeks. Dosed per weight on day 1 of a cycle.
B: Decitabine: 20 mg/m2 daily IV for 5 consecutive days on days 1-5. Dosed per weight on day 1 of the lead in
a: Can be done any time between day 24-28
b: +/- 3 days
c: Albumin, alkaline phosphatase, total bilirubin, bicarbonate, BUN, calcium, chloride, creatinine, glucose, potassium, total protein, SGOT [AST], SGPT [ALT], sodium, uric acid, magnesium, phosphorus.
d: At baseline, C1D1, and then every 3 cycles thereafter or more frequently when clinically indicated: gamma-GT, direct bilirubin, LDH, TSH, and lipase
e: Please see Appendix F for helpful study check list including immune related AE evaluation
f: For bone marrow biopsies, the minimum requested includes (1) aspirate (if aspirable), (2) core, (3) flow cytometry (if sufficient blasts; may not be possible in setting of low blasts or remission; if MRD flow cytometry is an option we will accept this data in lieu of standard flow cytometry if this test is available at the time of suspected remission), (4) cytogenetics if possible, (5) molecular testing per institutional guidelines (suggested at minimum at time of screening and relapse; but not required correlatives will include genotyping leukemic blasts). If concern for relapse after a response is achieved, patient should get marrow per routine clinical standards. At the end of treatment (either completion of therapy or at time of suspected relapse), encourage marrow sampling unless clinical condition precludes this. In cases of persistent leukemic blood, bone marrow biopsy can be performed on day 1 of subsequent cycle prior to treatment.
g: Routine response evaluation for patients in Arm A who are status post allo-HCT only. Peripheral blood source. Can be performed locally and if not available locally then should be analyzed at site where patient's transplant was performed.
h: Peripheral blood, aspirate, core biopsy, or FFPE to be collected per schedule described correlatives table below.
i: End of study includes patients who: complete 12 combination cycles, come off study for toxicity, or have disease progression.
j: Quarterly phone call for survival follow-up for up to 1 year after treatment is complete. All efforts to contact the patient should be made.
k: As there can be a delay in the presentation of an immune-related adverse event, patients will be asked to come back for a clinical exam 3 months (+/-1 month) after their last treatment on study.
l: Alert PI if there is concern for cardiac toxicity (minimum assessment: cardiology consult, EKG, echocardiogram, and labs CPK and troponin; refer to section 6.4.7)
m: Please see section 7.3.5 of the protocol for reporting requirements for GVHD and suspected immune related AEs to enable central review
n: For participants enrolled on Arm A (post transplant), a formal GVHD assessment is required at minimum on Day 1 of each cycle utilizing the GVHD assessment worksheet provided in Appendix B

10.2 Correlative Sample Schedule

All sites will send samples to Dana-Farber Cancer Institute at the following address:

Dana-Farber Cancer Institute
Smith Receiving
Pasquarello Tissue Bank- CMCF
1 Jimmy Fund Way- DA-L181A
Boston, MA 02115

Time Point[1]	Sample Type To Collect[1]	Sample Tube/Container[2], Amount Requested (Proposed Processing Site[3])	Shipping Method[4,5]	Shipping Address
Screening or pre-treatment (T0)	Marrow Aspirate	1 x 10 mL green top (P) 1 x 5 mL green top (A)	FedEx Priority overnight with cool pack[4]	Dana-Farber Cancer Institute
	Marrow Aspirate	2 mL lavender top (C)		
	Peripheral blood[10]	4 x 10 mL lavender top (P) 1x 10mL green top (A)		
	Extramedullary FFPE Tissue Block[5]	No specific size; standard paraffin blocks or unstained glass slides (R)	FedEx Priority overnight or same day RT	
	(Mandatory) Bone Marrow Core[9]	No specific size; 10% buffered formalin		
End[8] of Lead-In (T1)	Marrow Aspirate[7]	1 x 10 mL green top (P)	FedEx Priority overnight with cool pack[4]	Dana-Farber Cancer Institute
	Peripheral blood	3 x 10 mL lavender top (P) 1x 10 mL green top (A)		
End[8] of Cycle 1[12] (T2)	Marrow Aspirate	1 x 10 mL green top (P)	FedEx Priority overnight with cool pack[4]	Dana-Farber Cancer Institute
	Marrow Core	5-8 Unstained Slides		
	Peripheral Blood	4 x 10 mL lavender top (P)		
	Extramedullary FFPE Tissue Block[5]	No specific size; standard paraffin blocks or unstained glass slides (R)	FedEx Priority overnight or same day RT	
End[8] of Cycle 2[12] (T3)	Marrow Aspirate	1 x 10 mL green top (P) 1 x 5 mL green top (A)	FedEx Priority overnight with cool pack[4]	Dana-Farber Cancer Institute
	Peripheral Blood[10]	4 x 10 mL lavender top (P) 1x 10 mL green top (A)		
	Extramedullary FFPE Tissue Block[5]	No specific size; standard paraffin blocks or unstained glass slides (R)	FedEx Priority overnight or same day RT	
	(Mandatory) Bone Marrow Core[9]	No specific size; 10% buffered formalin (R)		
End[8] of Cycle 4[12]	Marrow Aspirate	1 x 10 mL green top (P)	FedEx Priority overnight with	Dana-Farber Cancer Institute

(T4)	Marrow Aspirate[6]	2 mL lavender top (C)	cool pack[4]	
	Peripheral Blood	3 x10 mL lavender top (P)		
	Extramedullary FFPE Tissue Block[5]	No specific size; standard paraffin blocks or unstained glass slides (R)	FedEx Priority overnight or same day RT	
	(Mandatory) Bone Marrow Core[9]	No specific size; 10% buffered formalin		
End[8] of Cycle 7 **(T5)**	Marrow Aspirate	1 x 5 mL green top (P)	FedEx Priority overnight with cool pack[4]	Dana-Farber Cancer Institute
	Peripheral Blood	2 x 10 mL lavender top (P)		
End[8] of Cycle 10 **(T6)**	Marrow Aspirate	1 x 5 mL green top (P)	FedEx Priority overnight with cool pack[4]	Dana-Farber Cancer Institute
	Peripheral Blood	2 x 10 mL lavender top (P)		
End of Treatment or Suspected Relapse **(T7)[12]**	Marrow Aspirate	1 x 5 mL green top (P)	FedEx Priority overnight with cool pack[4]	Dana-Farber Cancer Institute
	Marrow Aspirate[6]	2 mL lavender top (C)		
	Peripheral Blood	2 x 10 mL lavender top (P)		
	Extramedullary FFPE Tissue Block[5]	No specific size; standard paraffin blocks or unstained glass slides (R)	FedEx Priority overnight or same day RT	
	(Optional) Bone Marrow Core[9]	No specific size; 10% buffered formalin		
Clinically indicated (unscheduled) **(Tx)**	Marrow Aspirate	1 x 5 mL green top (P)	FedEx Priority overnight with cool pack[4]	Dana-Farber Cancer Institute
	Peripheral Blood	2 x 10 mL lavender top (P)		
One-time for germline DNA assessment[11] (Optiona)	Skin biopsy	Punch 4 mm in sterile container with saline.	Fed Ex Priority overnight Frozen	Dana-Farber Cancer Institute
Suspected Ipi-induced Toxicity (Optional)[13]	Skin biopsy	Punch 4 mm in sterile container with saline.	Fed Ex Priority overnight Frozen	Dana-Farber Cancer Institute

[1]All efforts should be made to collect correlative samples unless there are unforeseen clinical circumstances, or patient refusal.
[2]**Containers/Tubes**: Green top= **sodium heparin** tube; Lavender top = EDTA tube. Any tube size combination is acceptable to collect the approximate requested volume of blood or aspirate, however please use what is included in kits if possible to reduce missed samples.
[3]**Proposed Processing +/- Storage Sites for samples**: **(P) Pasquarello Heme Tissue Bank Lab (**Dana-Farber Cancer Institute, Pasquarello Tissue Bank - J614, Smith Receiving, 1 Jimmy Fund Way, Boston, MA 02215); **(R) Rodig Laboratory,** Attn: Evisa Gjini/Sara Abdelrahman, Thorn Building, Room 603B, Brigham & Women's Hospital, 20 Shattuck Street, Boston, MA 02115; **(A) Avigan Laboratory (Beth Israel Deaconess Medical Center),** 3 Blackfan Circle CLS 724 Boston, MA 02215; **(C) Center for Advanced Molecular Diagnostics,** 75 Francis Street, Boston, MA 02115
[4]**Shipping:** Aspirate and peripheral blood should be shipped with a cool pack if coming overnight (FedEx) and received Monday-Thursday ONLY for processing. Samples coming from participants at the corresponding organization (site: Dana-Farber Cancer Institute) must be submitted before 4 pm Monday-Friday.
[5]**FFPE Tissue Block** (including extramedullary disease biopsies including but not limited to leukemia cutis): At the screening time point, FFPE of extramedullary sites are required for patients enrolling with extramedullary AML only and optional for those enrolling with extramedullary plus marrow involved AML. FFPE samples at screening may be archival (if performed within 12 weeks, then biopsy does not need to be repeated unless a patient received anti-leukemic treatment (such as chemotherapy or radiation) after the biopsy with the exception of hydrea). These extramedullary FFPE tissue block samples can be shipped Monday-Friday at room temperature. These samples do NOT need to be sent with the kit (peripheral blood and aspirate) unless immediately ready; may be sent separately.
[6]**Rapid Heme Panel Sample:** This 1cc aspirate sample will only be collected for all patients in both Arm A and Arm B to perform research-based molecular testing with a Rapid Heme Panel (RHP) (performed in Center for Advanced Molecular

Diagnostics at the Brigham & Women's Hospital, Boston, MA). If a patient has a dry aspirate, then RHP may be drawn from peripheral blood if there are circulating blasts in setting of AML (circulating blasts are not required for a peripheral blood draw in setting of MDS). However, prefer sample to come from aspirate to allow for proper intra-patient and inter-patient comparison. In the event the "end of treatment" time point falls within the first 3 months on study treatment (+/- 2 weeks), this sample is optional

[7]**End of lead-in marrow aspirate sample:** This is a research aspirate only; no formal submission for pathology review is required.

[8]**End of cycle samples:** can be collected any time between Days 24-28.

[9]**Bone Marrow Core**: At the screening time point, submission of bone marrow biopsy core will be mandatory (to be fixed in 10% buffered formalin prior to sending using materials provided in kit) (this extra core should be collected at time of screening marrow for the purpose of IHC correlative studies). Mandatory submission of bone marrow biopsy core or 6-8 unstained slides will be collected at the end of cycles1, 2 and 4. For the end of treatment/suspected relapse timepoint, a core is requested, but not mandatory. At the EOT/suspected relapse timepoint, 6-8 unstained slides may be sent in leiu of a fresh core sample. Cores arriving in formalin will be transferred to 70% ethanol and decalcified in the Brigham & Women's Hospital Pathology Core laboratory in preparation for staining with IHC in the Rodig Laboratory. These bone marrow core samples should be shipped Monday-Thursday at room temperature. Archival tissue (FFPE) from participants who are consented on the clinical trial including bone marrow biopsies (tissue blocks or slides) performed at other time points where a bone marrow is clinically indicated (while on study) may be requested at a later time for IHC analysis in the Rodig laboratory

[10] **PB sample for PD studies:** It is preferred that the PB is drawn at one visit. However, depending on scenario, the peripheral blood PD sample can be collected over two visits if clinically necessary (e.g. for the "screening/pre-treatment" collection, PB can be collected at screening up until day 1 of lead-in cycle as long as it's prior to decitabine dosing; for the "end of cycle 2" collection, PB can be collected on days 24-28 of cycle 2 up until day 1 of cycle 3 as long as it's prior to decitabine and ipilimumab dosing). If this is the case, please update the study team to anticipate and plan on two sample deliveries.

[11]**Germline assessment**: Skin biopsies will be performed on patients who are identified to be exceptional responders (at any point of the trial) and some non-responders (in dose expansion cohort) after discussion with the overall PI. Samples are to be sent in normal saline in appropriate leakproof container to BWH CAMD by way of Dana-Farber. If sent from outside of DFCI, the sample should be sent frozen (both saline and tissue) and shipped to the Pasquarello Tissue Bank at Dana-Farber Cancer Institute. DNA will be extracted in batches in Brigham and Women's Hospital CAMD. This is optional.

[12]**FFPE bone marrow core slide or tissue block** may be requested if sample available for IHC comparison at this time point.

[13] Optional, if sample is being taken for clinical reasons, and it is safe to do so, we are requesting a small sample be sent to Dana-Farber Cancer Institute for analysis.

11. MEASUREMENT OF EFFECT

Although the clinical benefit of combination ipilimumab and decitabine has not yet been established, the intent of offering this treatment is to provide a possible therapeutic benefit, and thus the patient will be carefully monitored for tumor response and symptom relief in addition to safety and tolerability. Patients with MDS/AML will be assessed by standard criteria. For the purposes of this study, patients should be re-evaluated at the end of cycles 1, 2 and 4, and then every 3 cycles on treatment, and then at time of suspected relapse if relevant. The aspirate sample collected at the end of cycle 1 is for correlative research only. In addition to a baseline PET scan (for those with extramedullary disease, not required for those with leukemia cutis only), follow-up response scans will also be obtained at the end of four cycles of combination therapy following initial documentation of an objective response.

11.1 Antitumor Effect – Hematologic Tumors

Please see Appendix D for IWG Response Criteria for AML [6]. Disease progression will be determined by European LeukemiaNet 2017 criteria (Appendix H).

Please see Appendix E for IWG Response Criteria for MDS [7].

Bone marrow biopsy and aspirate will be performed at screening (will serve as baseline) and end of cycles 1, 2 and 4, and every 3 cycles thereafter (end of cycles 7 and 10). If there is evidence or clinical concern for disease progression, then a bone marrow biopsy and aspirate will be

collected. At each evaluation the patient should have aspirate (if aspirable), cytogenetics and flow cytometry performed. Genotyping will be performed as a part of study correlates.

Hematologic evaluations will be performed with CBC with differential at a minimum of once per cycle. Response assessments may be determined by labs (CBC with differential, particularly platelet count and absolute neutrophil count) within 2 weeks of a bone marrow biopsy as long as it is obtained prior to day 1 of the next cycle.

The best overall response is the best response recorded from the start of the treatment until disease progression/recurrence.

- For those with AML: Best overall response including CR and CRi will be recorded. Patients may remain on study if they have evidence of clinical benefit, stable disease, partial remission, complete remission or complete remission with incomplete count recovery.
- For those with MDS: Best overall response rate will include CR, marrow CR, PR, and hematologic improvement.

11.1.1 Duration of Response

Duration of overall response: The duration of overall response is measured from the time measurement criteria are met for CR or PR or HI (for those with MDS) (whichever is first recorded) until the first date that recurrent or progressive disease is objectively documented.

The duration of overall CR is measured from the time measurement criteria are first met for CR until the first date that progressive disease is objectively documented.

Duration of stable disease: Stable disease is measured from the start of the treatment until the criteria for progression are met, taking as reference the smallest measurements recorded since the treatment started, including the baseline measurements.

11.1.2 Progression-Free Survival

Overall Survival: Overall Survival (OS) is defined as the time from time of registration to death due to any cause, or censored at date last known alive. We will evaluate for survival differences among patients that respond based on transplant status.

Progression-Free Survival: Progression-Free Survival (PFS) is defined as the time from time of registration to the earlier of progression or death due to any cause. Participants alive without disease progression are censored at date of last disease evaluation.

12. STUDY OVERSIGHT AND DATA REPORTING / REGULATORY REQUIREMENTS

Adverse event lists, guidelines, and instructions for AE reporting can be found in Section 7.0 (Adverse Events: List and Reporting Requirements).

12.1 Study Oversight

This protocol is monitored at several levels, as described in this section. The Protocol Principal Investigator is responsible for monitoring the conduct and progress of the clinical trial, including the ongoing review of accrual, patient-specific clinical and laboratory data, and routine and serious adverse events; reporting of expedited adverse events; and accumulation of reported adverse events from other trials testing the same drug(s). The Protocol Principal Investigator and statistician have access to the data at all times through the CTMS web-based reporting portal.

For Phase 1 studies, all decisions regarding dose escalation/expansion/de-escalation require sign-off by the Protocol Principal Investigator through the CTMS/IWRS. In addition, for the Phase 1 portion, the Protocol Principal Investigator will have at least monthly, or more frequently, conference calls with the Study Investigators and the CTEP Medical Officer(s) to review accrual, progress, and adverse events and unanticipated problems.

All Study Investigators at participating sites who register/enroll patients on a given protocol are responsible for timely submission of data via Medidata Rave and timely reporting of adverse events for that particular study. This includes timely review of data collected on the electronic CRFs submitted via Medidata Rave.

All studies are also reviewed in accordance with the enrolling institution's data safety monitoring plan.

In addition to reporting to NCI/ETCN, it is mandatory for all participating sites to report all grade 4 events within 24 hours if possible, no more than 72 hours, to the Overall Study Chair except for those mentioned in section 7.3.4. All grade 5 events should be reported to the Overall Study Chair within 24 hours.

12.2 Data Reporting

Data collection for this study will be done exclusively through Medidata Rave. Access to the trial in Rave is granted through the iMedidata application to all persons with the appropriate roles assigned in the Regulatory Support System (RSS). To access Rave via iMedidata, the site user must have an active CTEP IAM account (check at < https://ctepcore.nci.nih.gov/iam >) and the appropriate Rave role (Rave CRA, Read-Only, Rave CRA (Lab Admin), Rave SLA, or Rave Investigator) on either the LPO or participating organization roster at the enrolling site. To hold the Rave CRA role or Rave CRA (Lab Admin) role, the user must hold a minimum of an AP registration type. To hold the Rave Investigator role, the individual must be registered as an NPIVR or IVR. Associates can hold read-only roles in Rave.

Upon initial site registration approval for the study in RSS, all persons with Rave roles assigned on the appropriate roster will be sent a study invitation e-mail from iMedidata. To accept the invitation, site users must log into the Select Login (https://login.imedidata.com/selectlogin) using their CTEP-IAM user name and password, and click on the "accept" link in the upper right-corner of the iMedidata page. Please note, site users will not be able to access the study in Rave until all required Medidata and study specific trainings are completed. Trainings will be in

the form of electronic learnings (eLearnings), and can be accessed by clicking on the link in the upper right pane of the iMedidata screen.

Users that have not previously activated their iMedidata/Rave account at the time of initial site registration approval for the study in RSS will also receive a separate invitation from iMedidata to activate their account. Account activation instructions are located on the CTSU website, Rave tab under the Rave resource materials (Medidata Account Activation and Study Invitation Acceptance). Additional information on iMedidata/Rave is available on the CTSU members' website under the Rave tab or by contacting the CTSU Help Desk at 1-888-823-5923 or by e-mail at ctsucontact@westat.com.

12.2.1 Method

This study will be monitored by the Clinical Trials Monitoring Service (CTMS). Data will be submitted to CTMS at least once every two weeks via Medidata Rave (or other modality if approved by CTEP). Information on CTMS reporting is available at http://www.theradex.com/clinicalTechnologies/?National-Cancer-Institute-NCI-11. On-site audits will be conducted three times annually (one annual site visit and two data audits). For CTMS monitored studies, after users have activated their accounts, please contact the Theradex Help Desk at (609) 799-7580 or by email at CTMSSupport@theradex.com for additional support with Rave and completion of CRFs.

12.2.2 Responsibility for Data Submission

For ETCTN trials, it is the responsibility of the PI(s) at the site to ensure that all investigators at the ETCTN Sites understand the procedures for data submission for each ETCTN protocol and that protocol specified data are submitted accurately and in a timely manner to the CTMS via the electronic data capture system, Medidata Rave.

Data are to be submitted via Medidata Rave to CTMS on a real-time basis, but no less than once every 2 weeks. The timeliness of data submissions and timeliness in resolving data queries will be tracked by CTMS. Metrics for timeliness will be followed and assessed on a quarterly basis. For the purpose of Institutional Performance Monitoring, data will be considered delinquent if it is greater than 4 weeks past due.

Data from Medidata Rave and CTEP-AERS is reviewed by the CTMS on an ongoing basis as data is received. Queries will be issued by CTMS directly within Rave. The queries will appear on the Task Summary Tab within Rave for the CRA at the ETCTN to resolve. Monthly web-based reports are posted for review by the Drug Monitors in the IDB, CTEP. Onsite audits will be conducted by the CTMS to ensure compliance with regulatory requirements, GCP, and NCI policies and procedures with the overarching goal of ensuring the integrity of data generated from NCI-sponsored clinical trials, as described in the ETCTN Program Guidelines, which may be found on the CTEP (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/adverse_events.htm) and CTSU websites.

An End of Study CRF is to be completed by the PI, and is to include a summary of study endpoints not otherwise captured in the database, such as (for phase 1 trials) the recommended phase 2 dose (RP2D), and a description of any dose-limiting toxicities (DLTs). CTMS will utilize a core set of eCRFs that are Cancer Data Standards Registry and Repository (caDSR) compliant (http://cbiit.nci.nih.gov/ncip/biomedical-informatics-resources/interoperability-and-semantics/metadata-and-models). Customized eCRFs will be included when appropriate to meet unique study requirements. The PI is encouraged to review the eCRFs, working closely with CTMS to ensure prospectively that all required items are appropriately captured in the eCRFs prior to study activation. CTMS will prepare the eCRFs with built-in edit checks to the extent possible to promote data integrity.

CDUS data submissions for ETCTN trials activated after March 1, 2014, will be carried out by the CTMS contractor, Theradex. CDUS submissions are performed by Theradex on a monthly basis. The trial's lead institution is responsible for timely submission to CTMS via Rave, as above.

Further information on data submission procedures can be found in the ETCTN Program Guidelines (http://ctep.cancer.gov/protocolDevelopment/electronic_applications/adverse_events.htm).

12.3 CTEP Multicenter Guidelines

N/A

12.4 Collaborative Agreements Language

The agent(s) supplied by CTEP, DCTD, NCI used in this protocol is/are provided to the NCI under a Collaborative Agreement (CRADA, CTA, CSA) between the Pharmaceutical Company(ies) (hereinafter referred to as "Collaborator(s)") and the NCI Division of Cancer Treatment and Diagnosis. Therefore, the following obligations/guidelines, in addition to the provisions in the "Intellectual Property Option to Collaborator" (http://ctep.cancer.gov/industryCollaborations2/intellectual_property.htm) contained within the terms of award, apply to the use of the Agent(s) in this study:

1. Agent(s) may not be used for any purpose outside the scope of this protocol, nor can Agent(s) be transferred or licensed to any party not participating in the clinical study. Collaborator(s) data for Agent(s) are confidential and proprietary to Collaborator(s) and shall be maintained as such by the investigators. The protocol documents for studies utilizing Agents contain confidential information and should not be shared or distributed without the permission of the NCI. If a copy of this protocol is requested by a patient or patient's family member participating on the study, the individual should sign a confidentiality agreement. A suitable model agreement can be downloaded from: http://ctep.cancer.gov.

2. For a clinical protocol where there is an investigational Agent used in combination with (an)other Agent(s), each the subject of different Collaborative Agreements, the access to and use of data by each Collaborator shall be as follows (data pertaining to such combination use shall hereinafter be referred to as "Multi-Party Data"):

    a. NCI will provide all Collaborators with prior written notice regarding the existence and nature of any agreements governing their collaboration with NCI, the design of the proposed combination protocol, and the existence of any obligations that would tend to restrict NCI's participation in the proposed combination protocol.

    b. Each Collaborator shall agree to permit use of the Multi-Party Data from the clinical trial by any other Collaborator solely to the extent necessary to allow said other Collaborator to develop, obtain regulatory approval or commercialize its own Agent.

    c. Any Collaborator having the right to use the Multi-Party Data from these trials must agree in writing prior to the commencement of the trials that it will use the Multi-Party Data solely for development, regulatory approval, and commercialization of its own Agent.

3. Clinical Trial Data and Results and Raw Data developed under a Collaborative Agreement will be made available to Collaborator(s), the NCI, and the FDA, as appropriate and unless additional disclosure is required by law or court order as described in the IP Option to Collaborator (http://ctep.cancer.gov/industryCollaborations2/intellectual_property.htm). Additionally, all Clinical Data and Results and Raw Data will be collected, used and disclosed consistent with all applicable federal statutes and regulations for the protection of human subjects, including, if applicable, the *Standards for Privacy of Individually Identifiable Health Information* set forth in 45 C.F.R. Part 164.

4. When a Collaborator wishes to initiate a data request, the request should first be sent to the NCI, who will then notify the appropriate investigators (Group Chair for Cooperative Group studies, or PI for other studies) of Collaborator's wish to contact them.

5. Any data provided to Collaborator(s) for Phase 3 studies must be in accordance with the guidelines and policies of the responsible Data Monitoring Committee (DMC), if there is a DMC for this clinical trial.

6. Any manuscripts reporting the results of this clinical trial must be provided to CTEP by the Group office for Cooperative Group studies or by the principal investigator for non-Cooperative Group studies for immediate delivery to Collaborator(s) for advisory review and comment prior to submission for publication. Collaborator(s) will have 30 days from the date of receipt for review. Collaborator shall have the right to request that publication be delayed for up to an additional 30 days in order to ensure that Collaborator's confidential and proprietary data, in addition to Collaborator(s)'s intellectual property rights, are protected. Copies of abstracts must be provided to CTEP for forwarding to Collaborator(s) for courtesy review as soon as possible and preferably at least three (3) days prior to submission, but in any case, prior to presentation at the meeting or publication in the proceedings. Press

releases and other media presentations must also be forwarded to CTEP prior to release. Copies of any manuscript, abstract and/or press release/ media presentation should be sent to:

Email: ncicteppubs@mail.nih.gov

The Regulatory Affairs Branch will then distribute them to Collaborator(s). No publication, manuscript or other form of public disclosure shall contain any of Collaborator's confidential/ proprietary information.

12.5 Genomic Data Sharing Plan

Correlative studies proposed will aim to include genotyping patient leukemic blasts. If genomic data will be studied, analyzed, and collected, then we will plan to share and store this data in an NIH/NCI Genomic Data Biorepository (e.g., dbGaP, Cancer Genomic Database, other) following the genomic data sharing policy.

13. STATISTICAL CONSIDERATIONS

13.1 Study Design/Endpoints

This is an open-label phase 1 study using a standard 3+3 dose-escalation design with planned dose escalation of ipilimumab in combination with decitabine, to define the safety profile and MTD (RP2D) in patients with R/R MDSAML with and without previous allogeneic HSCT. Two arms are planned. Arm A will include a cohort of patients who are post-transplant (patients who were transplanted by allogeneic HSCT at any time). Arm B will include a cohort of patients who are transplant naive (patients who never received allo-HSCT). Both cohorts will be tested at three different ipilimumab dose levels (dose level 0 (starting dose; first dose level): 3 mg/kg, dose level 1: 5 mg/kg, and dose level 2: 10 mg/kg). Three to six patients will be evaluated at each dose level per arm.

Please see section 5.1 for details on patient enrollment. We will demonstrate safety in the first dose level in patients on Arm B prior to enrolling patients onto Arm A. After safety is demonstrated at this first dose level, enrollment can then occur simultaneously without further delay or observation period. A dose de-escalation level (-1) will be allowed with ipilimumab at 1 mg/kg. Arm A does not need to complete dose escalation before Arm B if Arm B is able to move to dose-expansion; and vice versa. In the event that 0 of 3 patients in a dose level experience a DLT, the dose level may still be expanded to 6 evaluable patients based on observed toxicity at the current dose level at the discretion of the overall PI to capture patient safety prior to expanding.

Dose escalation will follow a standard 3+3 design exploring 3 dose levels of ipilimumab (starting at dose level 0, at 3 mg/kg) with decitabine (20 mg/m2 daily x 5 consecutive doses). We will escalate to next dose cohort if 0/3 or 1/6 participants have a DLT during the first two cycles of combination therapy (cycles 1+2; first 8 weeks of combination therapy). If the rate of dose limiting toxicity (DLT) exceeds 30%, it is less likely that the dose of ipilimumab will be

dose-escalated. The table below provides the probability that the dose will be escalated, i.e., that 0 in 3 or 1 in 6 patients experience a DLT, given the true but unknown rate of DLT.

	True but unknown probability of DLT				
	0.10	0.20	0.30	0.40	0.50
P(escalate)	0.91	0.71	0.49	0.31	0.17

The MTD will be the highest dose at which 1 or fewer of 6 patients experience a DLT. If no MTD has been established, then dose level 2 will be used in the dose expansion cohort as the recommended phase 2 dose (RP2D). Once an MTD (or RP2D) has been established an expansion cohort will be enrolled for each arm. For a given arm, if all 3 dose cohorts are filled to 6 patients, then a 6 patient expansion will be added. If 2 or fewer dose cohorts are filled to 6 patients, then a 12 patient expansion will be added so that no more than 24 patients (dose escalation + expansion) will be treated on a given arm. Patients will be monitored for DLTs during the first 8 weeks of combination therapy (56 days from start of combination) during the induction phase only. If a patient experiences disease progression in cycle 1, the patient will be evaluable for DLT if the patient completes a minimum of 28 days. Any patient who does not complete cycle 1 for any reason other than DLT will be considered non-evaluable for dose escalation decisions and will be replaced by an additional patient at that same dose level. Patients in cycle 2 will be assessed for DLT.

Safety and tolerability of decitabine plus ipilimumab will be evaluated by means of drug related DLT (defined as first 8 weeks of combination treatment with decitabine and ipilimumab), AE reports (following CTCAE V.5 criteria), physical examinations, and laboratory safety evaluations. These results will be tabulated overall and by dose level to examine their frequency, organ systems affected, severity, and relationship to study drugs.

The goal of the expansion cohort will be to further assess the tolerability of decitabine plus ipilimumab. Frequency of grade 3 or higher non-hematologic toxicities will be reported by arm and frequency of all grade immune-related adverse events will be reported by arm. Additionally, GVHD for the patients on the post-transplant arm will be estimated and reported with a 90% exact binomial confidence interval.

Additionally, efficacy of decitabine plus ipilimumab will be assessed for the total number of patients enrolled at the MTD/RP2D for each arm. Anti-leukemic activity will be described in terms of best overall response rate (for AML, to include CR+CRi; for MDS, to include CR+marrow CR+PR+HI). Assuming 6 patients are enrolled at the MTD/RP2D from the dose escalation phase and an additional 6 patients for the cohort expansion, the 90% exact binomial confidence interval will be within +/- 25%. Assuming 6 patients are enrolled at the MTD/RP2D from the dose escalation phase and an additional 12 patients from the cohort expansion, the 90% exact binomial confidence interval will be within +/- 21%.

Overall survival (OS) is defined as the time from registration to death due to any cause or censored at the date last known alive. Progression-free survival (PFS) is defined as the time from registration to the earlier of progression or death due to any cause. Survival outcomes will be

estimated by the method of Kaplan and Meier. OS and PFS will be evaluated within each arm.

13.2 Sample Size/Accrual Rate

For each arm (Arm A and Arm B), assuming all dose cohorts are filled, the estimated sample will be at most 24. Hence the total estimated total sample size is at most 48 patients. The expansion cohort will be set at the MTD/RP2D. After the first dose level is assessed in Arm B (transplant naive), then we will then allow enrollment of patients onto Arm A. Further enrollment will occur in parallel with no observation period or delay between the start of therapy for each enrolled patient in either arm. We anticipate completing accrual within 24 months. 1-2 patients per month will be accrued.

Up to an additional 12 months of follow-up will be required on the last participant to observe the participant's survival; for total study duration of 3 years.

PLANNED ENROLLMENT REPORT

Racial Categories	Ethnic Categories				Total
	Not Hispanic or Latino		Hispanic or Latino		
	Female	Male	Female	Male	
American Indian/ Alaska Native	0	0	0	0	**0**
Asian	2	2	0	0	**4**
Native Hawaiian or Other Pacific Islander	0	0	0	0	**0**
Black or African American	4	4	0	0	**8**
White	11	11	4	4	**30**
More Than One Race	2	2	1	1	**6**
Total	**19**	**19**	**5**	**5**	**48**

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13.3 Stratification Factors

N/A

13.4 Analysis of Primary Endpoints

We will determine the MTD/RP2D of dose escalated ipilimumab in combination with decitabine in patients who are post allo-HCT and in patients who are transplant naive.

13.5 Analysis of Secondary Endpoints

Safety analyses will be based on the safety population, defined as all enrolled patients receiving at least 1 dose of ipilimumab. Categorical data will be presented as frequencies and percentages. All tests of treatment effects will be conducted at a 2-sided alpha level of 0.05, unless otherwise stated. All tests of interactions will be conducted at a 2-sided alpha level of 0.1, and all CIs will be given at a 2-sided 95% level, unless otherwise stated. Patients who are registered to the study and have received at least one dose of ipilimumab on study will be evaluable for response. Clinical response to treatment will be assessed following the 2003 IWG criteria for AML [6] and the 2006 IWG criteria for MDS [7]. Anti-leukemic activity will be described in terms of best overall response rate (for AML, to include CR+CRi; for MDS, to include CR+marrow CR+PR+HI), PFS, OS, and remission duration. Time to event summaries will use the Kaplan-Meier method. We will separately evaluate the incidence of acute GVHD and chronic GVHD in patients in the post-allo-HCT cohort, and compare these events with response to treatment.

The focus of this trial is to establish the safety and RP2D of the combination; thus the biomarker studies should be considered exploratory. We will evaluate whether or not absolute lymphocyte count can predict for response for patients who receive combination treatment. ALC levels will be divided into: low (<1000 cells/ul) and normal/high (greater than or equal to 1000 cells/ul). We will compare the response and overall survival of patients with low ALC versus normal/high ALC using Kaplan-Meier estimates and differences will be assessed using log-rank test. We will further capture the absolute change in ALC level measured at baseline to end of cycle 2 (collected prior to dosing on cycle 3 day 1) for response and survival differences. For correlative laboratory tests, we will compare nonrandom measurements in an individual patient either across time (pre-treatment versus on-treatment) with a paired Student *t* test. OS will be censored at the date of last contacts.

REFERENCES

1. Hodi FS, O'Day SJ, McDermott DF, Weber RW, Sosman JA, Haanen JB, et al. Improved survival with ipilimumab in patients with metastatic melanoma. The New England journal of medicine. 2010;363(8):711-23.
2. Wolchok JD, Kluger H, Callahan MK, Postow MA, Rizvi NA, Lesokhin AM, et al. Nivolumab plus ipilimumab in advanced melanoma. The New England journal of medicine. 2013;369(2):122-33.
3. 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(26):2521-32.
4. Davids MS, Kim HT, Bachireddy P, Costello C, Liguori R, Savell A, et al. Ipilimumab for Patients with Relapse after Allogeneic Transplantation. The New England journal of medicine. 2016;375(2):143-53.
5. Bashey A, Medina B, Corringham S, Pasek M, Carrier E, Vrooman L, et al. CTLA4 blockade with ipilimumab to treat relapse of malignancy after allogeneic hematopoietic cell transplantation. Blood. 2009;113(7):1581-8.
6. Cheson BD, Bennett JM, Kopecky KJ, Buchner T, Willman CL, Estey EH, et al. Revised recommendations of the International Working Group for Diagnosis, Standardization of Response Criteria, Treatment Outcomes, and Reporting Standards for Therapeutic Trials in Acute Myeloid Leukemia. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2003;21(24):4642-9.
7. Cheson BD, Greenberg PL, Bennett JM, Lowenberg B, Wijermans PW, Nimer SD, et al. Clinical application and proposal for modification of the International Working Group (IWG) response criteria in myelodysplasia. Blood. 2006;108(2):419-25.
8. Greenberg PL, Tuechler H, Schanz J, Sanz G, Garcia-Manero G, Sole F, et al. Revised international prognostic scoring system for myelodysplastic syndromes. Blood. 2012;120(12):2454-65.
9. Greenberg P, Cox C, LeBeau MM, Fenaux P, Morel P, Sanz G, et al. International scoring system for evaluating prognosis in myelodysplastic syndromes. Blood. 1997;89(6):2079-88.
10. Steensma DP, Baer MR, Slack JL, Buckstein R, Godley LA, Garcia-Manero G, et al. Multicenter study of decitabine administered daily for 5 days every 4 weeks to adults with myelodysplastic syndromes: the alternative dosing for outpatient treatment (ADOPT) trial. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2009;27(23):3842-8.
11. Fernandez HF, Sun Z, Yao X, Litzow MR, Luger SM, Paietta EM, et al. Anthracycline dose intensification in acute myeloid leukemia. The New England journal of medicine. 2009;361(13):1249-59.
12. Dohner H, Estey EH, Amadori S, Appelbaum FR, Buchner T, Burnett AK, et al. Diagnosis and management of acute myeloid leukemia in adults: recommendations from an international expert panel, on behalf of the European LeukemiaNet. Blood. 2010;115(3):453-74.
13. Kantarjian H, O'Brien S, Cortes J, Giles F, Faderl S, Jabbour E, et al. Results of intensive chemotherapy in 998 patients age 65 years or older with acute myeloid leukemia or high-risk myelodysplastic syndrome: predictive prognostic models for outcome. Cancer.

2006;106(5):1090-8.
14. Cashen AF, Schiller GJ, O'Donnell MR, DiPersio JF. Multicenter, phase II study of decitabine for the first-line treatment of older patients with acute myeloid leukemia. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2010;28(4):556-61.
15. Kantarjian HM, Thomas XG, Dmoszynska A, Wierzbowska A, Mazur G, Mayer J, et al. Multicenter, randomized, open-label, phase III trial of decitabine versus patient choice, with physician advice, of either supportive care or low-dose cytarabine for the treatment of older patients with newly diagnosed acute myeloid leukemia. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2012;30(21):2670-7.
16. Bornhauser M, Kienast J, Trenschel R, Burchert A, Hegenbart U, Stadler M, et al. Reduced-intensity conditioning versus standard conditioning before allogeneic haemopoietic cell transplantation in patients with acute myeloid leukaemia in first complete remission: a prospective, open-label randomised phase 3 trial. The Lancet Oncology. 2012;13(10):1035-44.
17. Barrett AJ, Battiwalla M. Relapse after allogeneic stem cell transplantation. Expert Rev Hematol. 2010;3(4):429-41.
18. Ruutu T, Volin L, Beelen DW, Trenschel R, Finke J, Schnitzler M, et al. Reduced-toxicity conditioning with treosulfan and fludarabine in allogeneic hematopoietic stem cell transplantation for myelodysplastic syndromes: final results of an international prospective phase II trial. Haematologica. 2011;96(9):1344-50.
19. Damaj G, Mohty M, Robin M, Michallet M, Chevallier P, Beguin Y, et al. Upfront allogeneic stem cell transplantation after reduced-intensity/nonmyeloablative conditioning for patients with myelodysplastic syndrome: a study by the Societe Francaise de Greffe de Moelle et de Therapie Cellulaire. Biology of blood and marrow transplantation : journal of the American Society for Blood and Marrow Transplantation. 2014;20(9):1349-55.
20. Gale RP, Horowitz MM. Graft-versus-leukemia in bone marrow transplantation. The Advisory Committee of the International Bone Marrow Transplant Registry. Bone marrow transplantation. 1990;6 Suppl 1:94-7.
21. Horowitz MM, Gale RP, Sondel PM, Goldman JM, Kersey J, Kolb HJ, et al. Graft-versus-leukemia reactions after bone marrow transplantation. Blood. 1990;75(3):555-62.
22. Pardoll DM. The blockade of immune checkpoints in cancer immunotherapy. Nature reviews Cancer. 2012;12(4):252-64.
23. Thompson CB, Allison JP. The emerging role of CTLA-4 as an immune attenuator. Immunity. 1997;7(4):445-50.
24. Kuhns MS, Epshteyn V, Sobel RA, Allison JP. Cytotoxic T lymphocyte antigen-4 (CTLA-4) regulates the size, reactivity, and function of a primed pool of CD4+ T cells. Proceedings of the National Academy of Sciences of the United States of America. 2000;97(23):12711-6.
25. Tivol EA, Borriello F, Schweitzer AN, Lynch WP, Bluestone JA, Sharpe AH. Loss of CTLA-4 leads to massive lymphoproliferation and fatal multiorgan tissue destruction, revealing a critical negative regulatory role of CTLA-4. Immunity. 1995;3(5):541-7.
26. Waterhouse P, Penninger JM, Timms E, Wakeham A, Shahinian A, Lee KP, et al. Lymphoproliferative disorders with early lethality in mice deficient in Ctla-4. Science. 1995;270(5238):985-8.
27. Chambers CA, Sullivan TJ, Allison JP. Lymphoproliferation in CTLA-4-deficient mice is mediated by costimulation-dependent activation of CD4+ T cells. Immunity.

1997;7(6):885-95.
28. Walunas TL, Lenschow DJ, Bakker CY, Linsley PS, Freeman GJ, Green JM, et al. CTLA-4 can function as a negative regulator of T cell activation. Immunity. 1994;1(5):405-13.
29. Kearney ER, Walunas TL, Karr RW, Morton PA, Loh DY, Bluestone JA, et al. Antigen-dependent clonal expansion of a trace population of antigen-specific CD4+ T cells in vivo is dependent on CD28 costimulation and inhibited by CTLA-4. Journal of immunology. 1995;155(3):1032-6.
30. Krummel MF, Sullivan TJ, Allison JP. Superantigen responses and co-stimulation: CD28 and CTLA-4 have opposing effects on T cell expansion in vitro and in vivo. Int Immunol. 1996;8(4):519-23.
31. Keler T, Halk E, Vitale L, O'Neill T, Blanset D, Lee S, et al. Activity and safety of CTLA-4 blockade combined with vaccines in cynomolgus macaques. Journal of immunology. 2003;171(11):6251-9.
32. Hara I, Takechi Y, Houghton AN. Implicating a role for immune recognition of self in tumor rejection: passive immunization against the brown locus protein. The Journal of experimental medicine. 1995;182(5):1609-14.
33. Naftzger C, Takechi Y, Kohda H, Hara I, Vijayasaradhi S, Houghton AN. Immune response to a differentiation antigen induced by altered antigen: a study of tumor rejection and autoimmunity. Proceedings of the National Academy of Sciences of the United States of America. 1996;93(25):14809-14.
34. Bloom MB, Perry-Lalley D, Robbins PF, Li Y, el-Gamil M, Rosenberg SA, et al. Identification of tyrosinase-related protein 2 as a tumor rejection antigen for the B16 melanoma. The Journal of experimental medicine. 1997;185(3):453-9.
35. Overwijk WW, Tsung A, Irvine KR, Parkhurst MR, Goletz TJ, Tsung K, et al. gp100/pmel 17 is a murine tumor rejection antigen: induction of "self"-reactive, tumoricidal T cells using high-affinity, altered peptide ligand. The Journal of experimental medicine. 1998;188(2):277-86.
36. Overwijk WW, Lee DS, Surman DR, Irvine KR, Touloukian CE, Chan CC, et al. Vaccination with a recombinant vaccinia virus encoding a "self" antigen induces autoimmune vitiligo and tumor cell destruction in mice: requirement for CD4(+) T lymphocytes. Proceedings of the National Academy of Sciences of the United States of America. 1999;96(6):2982-7.
37. Weber LW, Bowne WB, Wolchok JD, Srinivasan R, Qin J, Moroi Y, et al. Tumor immunity and autoimmunity induced by immunization with homologous DNA. J Clin Invest. 1998;102(6):1258-64.
38. Rosenberg SA, White DE. Vitiligo in patients with melanoma: normal tissue antigens can be targets for cancer immunotherapy. J Immunother Emphasis Tumor Immunol. 1996;19(1):81-4.
39. Yang JC, Hughes M, Kammula U, Royal R, Sherry RM, Topalian SL, et al. Ipilimumab (anti-CTLA4 antibody) causes regression of metastatic renal cell cancer associated with enteritis and hypophysitis. Journal of immunotherapy. 2007;30(8):825-30.
40. Hodi FS, Friedlander PA, Atkins MB, McDermott DF, Lawrence DP, Ibrahim N, et al., editors. A phase I trial of ipilimumab plus bevacizumab in patients with unresectable stage III or stage IV melanoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology; 2011.
41. Carthon BC, Wolchok JD, Yuan J, Kamat A, Ng Tang DS, Sun J, et al. Preoperative CTLA-4 blockade: tolerability and immune monitoring in the setting of a presurgical clinical

trial. Clinical cancer research : an official journal of the American Association for Cancer Research. 2010;16(10):2861-71.
42. Liakou CI, Kamat A, Tang DN, Chen H, Sun J, Troncoso P, et al. CTLA-4 blockade increases IFNgamma-producing CD4+ICOShi cells to shift the ratio of effector to regulatory T cells in cancer patients. Proceedings of the National Academy of Sciences of the United States of America. 2008;105(39):14987-92.
43. Royal RE, Levy C, Turner K, Mathur A, Hughes M, Kammula US, et al. Phase 2 trial of single agent Ipilimumab (anti-CTLA-4) for locally advanced or metastatic pancreatic adenocarcinoma. Journal of immunotherapy. 2010;33(8):828-33.
44. O'Mahony D, Morris JC, Quinn C, Gao W, Wilson WH, Gause B, et al. A pilot study of CTLA-4 blockade after cancer vaccine failure in patients with advanced malignancy. Clinical cancer research : an official journal of the American Association for Cancer Research. 2007;13(3):958-64.
45. 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. Proceedings of the National Academy of Sciences of the United States of America. 2008;105(8):3005-10.
46. Zhou J, Bashey A, Zhong R, Corringham S, Messer K, Pu M, et al. CTLA-4 blockade following relapse of malignancy after allogeneic stem cell transplantation is associated with T cell activation but not with increased levels of T regulatory cells. Biology of blood and marrow transplantation : journal of the American Society for Blood and Marrow Transplantation. 2011;17(5):682-92.
47. Harzstark AL, Fong L, Weinberg VK, Ryan CJ, Lin AM, Sun J, et al., editors. Final results of a phase I study of CTLA-4 blockade in combination with GM-CSF for metastatic castration resistant prostate cancer (mCRPC). Journal of clinical oncology : official journal of the American Society of Clinical Oncology; 2010.
48. Fong L, Kwek SS, O'Brien S, Kavanagh B, McNeel DG, Weinberg V, et al. Potentiating endogenous antitumor immunity to prostate cancer through combination immunotherapy with CTLA4 blockade and GM-CSF. Cancer research. 2009;69(2):609-15.
49. Madan RA, Mohebtash M, Arlen PM, al. E, editors. Overall survival (OS) analysis of a phase I trial of a vector-based vaccine (PSA-TRICOM) and ipilimumab (Ipi) in the treatment of metastatic castration-resistant prostate cancer (mCRPC). Genitourinary Cancers Symposium; 2010.
50. Stein WD, Gulley JL, Schlom J, Madan RA, Dahut W, Figg WD, et al. Tumor regression and growth rates determined in five intramural NCI prostate cancer trials: the growth rate constant as an indicator of therapeutic efficacy. Clinical cancer research : an official journal of the American Association for Cancer Research. 2011;17(4):907-17.
51. Ansell SM, Hurvitz SA, Koenig PA, LaPlant BR, Kabat BF, Fernando D, et al. Phase I study of ipilimumab, an anti-CTLA-4 monoclonal antibody, in patients with relapsed and refractory B-cell non-Hodgkin lymphoma. Clinical cancer research : an official journal of the American Association for Cancer Research. 2009;15(20):6446-53.
52. Min L, Vaidya A, Becker C. Thyroid autoimmunity and ophthalmopathy related to melanoma biological therapy. European journal of endocrinology / European Federation of Endocrine Societies. 2011;164(2):303-7.
53. Wolchok JD, Hoos A, O'Day S, Weber JS, Hamid O, Lebbe C, et al. Guidelines for the evaluation of immune therapy activity in solid tumors: immune-related response criteria.

Clinical cancer research : an official journal of the American Association for Cancer Research. 2009;15(23):7412-20.
54. Hoos A, Eggermont AM, Janetzki S, Hodi FS, Ibrahim R, Anderson A, et al. Improved endpoints for cancer immunotherapy trials. J Natl Cancer Inst. 2010;102(18):1388-97.
55. Hersh EM, O'Day SJ, Powderly J, Khan KD, Pavlick AC, Cranmer LD, et al. A phase II multicenter study of ipilimumab with or without dacarbazine in chemotherapy-naive patients with advanced melanoma. Investigational new drugs. 2011;29(3):489-98.
56. Sarnaik AA, Yu B, Yu D, Morelli D, Hall M, Bogle D, et al. Extended dose ipilimumab with a peptide vaccine: immune correlates associated with clinical benefit in patients with resected high-risk stage IIIc/IV melanoma. Clinical cancer research : an official journal of the American Association for Cancer Research. 2011;17(4):896-906.
57. Ravandi F, Kantarjian H, Cohen A, Davis M, O'Brien S, Anderlini P, et al. Decitabine with allogeneic peripheral blood stem cell transplantation in the therapy of leukemia relapse following a prior transplant: results of a phase I study. Bone marrow transplantation. 2001;27(12):1221-5.
58. Pusic I, Choi J, Fiala MA, Gao F, Holt M, Cashen AF, et al. Maintenance Therapy with Decitabine after Allogeneic Stem Cell Transplantation for Acute Myelogenous Leukemia and Myelodysplastic Syndrome. Biology of blood and marrow transplantation : journal of the American Society for Blood and Marrow Transplantation. 2015;21(10):1761-9.
59. Jabbour E, Giralt S, Kantarjian H, Garcia-Manero G, Jagasia M, Kebriaei P, et al. Low-dose azacitidine after allogeneic stem cell transplantation for acute leukemia. Cancer. 2009;115(9):1899-905.
60. Schadendorf D, Hodi FS, Robert C, Weber JS, Margolin K, Hamid O, et al. Pooled Analysis of Long-Term Survival Data From Phase II and Phase III Trials of Ipilimumab in Unresectable or Metastatic Melanoma. Journal of clinical oncology : official journal of the American Society of Clinical Oncology. 2015;33(17):1889-94.
61. Das R, Verma R, Sznol M, Boddupalli CS, Gettinger SN, Kluger H, et al. Combination therapy with anti-CTLA-4 and anti-PD-1 leads to distinct immunologic changes in vivo. Journal of immunology. 2015;194(3):950-9.
62. Fevery S, Billiau AD, Sprangers B, Rutgeerts O, Lenaerts C, Goebels J, et al. CTLA-4 blockade in murine bone marrow chimeras induces a host-derived antileukemic effect without graft-versus-host disease. Leukemia. 2007;21(7):1451-9.
63. Davids MS, Kim HT, Costello CL, McSweeney PA, Luiguori R, Lukez A, et al., editors. A Multicenter Phase I/Ib Study of Ipilimumab for Relapsed Hematologic Malignancies after Allogeneic Hematopoietic Stem Cell Transplantation. American Society of Hematology; 2015 December 7, 2015; Orlando, FL.
64. Chiappinelli KB, Strissel PL, Desrichard A, Li H, Henke C, Akman B, et al. Inhibiting DNA Methylation Causes an Interferon Response in Cancer via dsRNA Including Endogenous Retroviruses. Cell. 2015;162(5):974-86.
65. Peng D, Kryczek I, Nagarsheth N, Zhao L, Wei S, Wang W, et al. Epigenetic silencing of TH1-type chemokines shapes tumour immunity and immunotherapy. Nature. 2015;527(7577):249-53.
66. Wang LX, Mei ZY, Zhou JH, Yao YS, Li YH, Xu YH, et al. Low dose decitabine treatment induces CD80 expression in cancer cells and stimulates tumor specific cytotoxic T lymphocyte responses. PloS one. 2013;8(5):e62924.
67. Yang ZZ, Grote DM, Ziesmer SC, Xiu B, Novak AJ, Ansell SM. PD-1 expression

defines two distinct T-cell sub-populations in follicular lymphoma that differentially impact patient survival. Blood cancer journal. 2015;5:e281.
68. Steinmann J, Bertz H, Wasch R, Marks R, Zeiser R, Bogatyreva L, et al. 5-Azacytidine and DLI can induce long-term remissions in AML patients relapsed after allograft. Bone marrow transplantation. 2015;50(5):690-5.
69. Goodyear OC, Dennis M, Jilani NY, Loke J, Siddique S, Ryan G, et al. Azacitidine augments expansion of regulatory T cells after allogeneic stem cell transplantation in patients with acute myeloid leukemia (AML). Blood. 2012;119(14):3361-9.
70. Magenau JM, Qin X, Tawara I, Rogers CE, Kitko C, Schlough M, et al. Frequency of CD4(+)CD25(hi)FOXP3(+) regulatory T cells has diagnostic and prognostic value as a biomarker for acute graft-versus-host-disease. Biology of blood and marrow transplantation : journal of the American Society for Blood and Marrow Transplantation. 2010;16(7):907-14.
71. Yang H, Bueso-Ramos C, DiNardo C, Estecio MR, Davanlou M, Geng QR, et al. Expression of PD-L1, PD-L2, PD-1 and CTLA4 in myelodysplastic syndromes is enhanced by treatment with hypomethylating agents. Leukemia. 2014;28(6):1280-8.
72. Covre A, Fazio C, Nicolay H, Parisi G, Taverna P, Azab M, et al., editors. Epigenetic priming with novel hypomethylating agent SGI-110 improved antitumor activity of CTLA-4 blockade in a syngenic mouse model. . Molecular cancer therapeutics; 2013 November 2013.
73. Tsai HC, Li H, Van Neste L, Cai Y, Robert C, Rassool FV, et al. Transient low doses of DNA-demethylating agents exert durable antitumor effects on hematological and epithelial tumor cells. Cancer cell. 2012;21(3):430-46.
74. Wrangle J, Wang W, Koch A, Easwaran H, Mohammad HP, Vendetti F, et al. Alterations of immune response of Non-Small Cell Lung Cancer with Azacytidine. Oncotarget. 2013;4(11):2067-79.
75. Delyon J, Mateus C, Lefeuvre D, Lanoy E, Zitvogel L, Chaput N, et al. Experience in daily practice with ipilimumab for the treatment of patients with metastatic melanoma: an early increase in lymphocyte and eosinophil counts is associated with improved survival. Ann Oncol. 2013;24(6):1697-703.
76. Luke JJ, Callahan MK, Postow MA, Romano E, Ramaiya N, Bluth M, et al. Clinical activity of ipilimumab for metastatic uveal melanoma: a retrospective review of the Dana-Farber Cancer Institute, Massachusetts General Hospital, Memorial Sloan-Kettering Cancer Center, and University Hospital of Lausanne experience. Cancer. 2013;119(20):3687-95.
77. Cancer Genome Atlas Research N. Genomic and epigenomic landscapes of adult de novo acute myeloid leukemia. The New England journal of medicine. 2013;368(22):2059-74.
78. Corces-Zimmerman MR, Majeti R. Pre-leukemic evolution of hematopoietic stem cells: the importance of early mutations in leukemogenesis. Leukemia. 2014;28(12):2276-82.
79. Mossner M, Jann JC, Wittig J, Nolte F, Fey S, Nowak V, et al. Mutational hierarchies in myelodysplastic syndromes dynamically adapt and evolve upon therapy response and failure. Blood. 2016.
80. Lindsley RC, Mar BG, Mazzola E, Grauman PV, Shareef S, Allen SL, et al. Acute myeloid leukemia ontogeny is defined by distinct somatic mutations. Blood. 2015;125(9):1367-76.
81. Benekli M, Xia Z, Donohue KA, Ford LA, Pixley LA, Baer MR, et al. Constitutive activity of signal transducer and activator of transcription 3 protein in acute myeloid leukemia blasts is associated with short disease-free survival. Blood. 2002;99(1):252-7.
82. Alvarez JV, Febbo PG, Ramaswamy S, Loda M, Richardson A, Frank DA.

Identification of a genetic signature of activated signal transducer and activator of transcription 3 in human tumors. Cancer research. 2005;65(12):5054-62.
83. Chen J, Deangelo DJ, Kutok JL, Williams IR, Lee BH, Wadleigh M, et al. PKC412 inhibits the zinc finger 198-fibroblast growth factor receptor 1 fusion tyrosine kinase and is active in treatment of stem cell myeloproliferative disorder. Proceedings of the National Academy of Sciences of the United States of America. 2004;101(40):14479-84.
84. Pikman Y, Lee BH, Mercher T, McDowell E, Ebert BL, Gozo M, et al. MPLW515L is a novel somatic activating mutation in myelofibrosis with myeloid metaplasia. PLoS Med. 2006;3(7):e270.
85. Eboigbodin K, Filen S, Ojalehto T, Brummer M, Elf S, Pousi K, et al. Reverse transcription strand invasion based amplification (RT-SIBA): a method for rapid detection of influenza A and B. Appl Microbiol Biotechnol. 2016.
86. Harrison C, Kiladjian JJ, Al-Ali HK, Gisslinger H, Waltzman R, Stalbovskaya V, et al. JAK inhibition with ruxolitinib versus best available therapy for myelofibrosis. The New England journal of medicine. 2012;366(9):787-98.
87. Wilkins BS, Radia D, Woodley C, Farhi SE, Keohane C, Harrison CN. Resolution of bone marrow fibrosis in a patient receiving JAK1/JAK2 inhibitor treatment with ruxolitinib. Haematologica. 2013;98(12):1872-6.

APPENDIX A PERFORMANCE STATUS CRITERIA

ECOG Performance Status Scale	
Grade	Descriptions
0	Normal activity. Fully active, able to carry on all pre-disease performance without restriction.
1	Symptoms, but ambulatory. Restricted in physically strenuous activity, but ambulatory and able to carry out work of a light or sedentary nature (*e.g.*, light housework, office work).
2	In bed <50% of the time. Ambulatory and capable of all self-care, but unable to carry out any work activities. Up and about more than 50% of waking hours.
3	In bed >50% of the time. Capable of only limited self-care, confined to bed or chair more than 50% of waking hours.
4	100% bedridden. Completely disabled. Cannot carry on any self-care. Totally confined to bed or chair.
5	Dead.

APPENDIX B1 ACUTE GRAFT VERSUS HOST DISEASE WORKSHEET

Acute GVHD Worksheet
Consensus Conference on Acute GVHD Grading
Reference: Przepiorka, DD et al., *Bone Marrow Transplant,* 825-828.

Instructions:

1. Is acute GVHD present (circle): YES or NO
2. If yes, please circle below the Overall Grade (and the Individual Stages as Relevant)

GVHD Classification

Overall Grade (please circle)	Skin	Liver	Gut
I	Stage 1-2	None	None
II	Stage 3 or	Stage 1 or	Stage 1
III		Stage 2-3 or	Stage 2-4
IV	Stage 4 or	Stage 4	

For each overall grade of acute GVHD, an assessment of skin disease plus liver and/or gut is required.

Individual organ staging
Clinical manifestations and staging of acute GVHD

Organ	Clinical Manifestation	Staging	Tips to Investigator
Skin	Erythematous, maculopapular rash involving palms and soles; may become confluence; severe disease: bullae	Stage 1: < 25% rash Stage 2: 25-50% rash Stage 3: generalized erythroderma Stage 4: bullae	Use rule of 9s to determine extent of rash
Liver	Painless jaundice with conjugated hyperbilirubinemia and increased alkaline phosphatase	Stage 1: bilirubin 2-3 mg/dL Stage 2: bilirubin 3.1-6 mg/dL Stage 3: bilirubin 6.1-15 mg/dL Stage 4: bilirubin > 15 mg/dL	Range given is for total bilirubin; downgrade one stage if a cause of elevated bilirubin other than GvHD has been documented
GI	Upper: nausea, vomiting, anorexia Loewr: diarrhea, abdominal cramps, distention, ileus, bleeding	Stage 1: diarrhea > 500 mL/day or persistent nausea or anorexia Stage 2: diarrhea > 1000 mL/day Stage 3: diarrhea > 1500 mL/day Stage 4: large volume diarrhea and severe abdominal pain +/- ileus	Downgrade one stage if a cause of diarrhea other than GvHD has been documented; downgrade upper GI one stage if biopsy result is negative; or if no biopsy is done and gVHD is not etiology or if biops is equivocal and GVHD is not an etiology

Subject ID: Site:
Signature of Treating Investigator: Date of evaluation:
Cycle/Day:

APPENDIX B2 CHRONIC GRAFT VERSUS HOST DISEASE WORKSHEET

Chronic GVHD Worksheet
Consensus Conference on Acute GVHD Grading
Reference: Jagasia MH et al., Biol Blood Marrow Transplant, 2015, 389-401.

Instructions:

1. Is chronic GVHD present (circle): YES or NO
2. If yes, please circle global severity below (mild, moderate or severe). See sheets on Scoring per organ

Mild	• 1 or 2 organs or sites (except lung) with score 1 • Mild oral symptoms, no decrease in oral intake • Mild dry eyes, lubricant eyedrops ≤ 3x/day
Moderate	• 3 or more organs with score 1 • At least 1 organ or site with score 2 • 19-50% body surface area involved or superficial sclerosis • Moderate dry eyes, eyedrops > 3x/day or punctal plugs • Lung score 1 (FEV1 60-79% or dyspnea with stairs)
Severe	• At least 1 organ or site with score 3 • > 50% body surface area involved • Deep sclerosis, impaired mobility or ulceration • Severe oral symptoms with major limitation in oral intake • Severe dry eyes affecting ADL • Lung score 2 (FEV1 40-59% or dyspnea walking on flat ground)

Figure 4. Calculation of mild, moderate, and severe global severity, with examples. If the entire abnormality in an organ is noted to be unequivocally explained by a non-GVHD documented cause, that organ is not included for calculation of the global severity. If the abnormality in an organ is attributed to multifactorial causes (GVHD plus other causes) the scored organ will be used for calculation of the global severity regardless of the contributing causes (no downgrading of organ severity score). ADL, activities of daily living; FEV1, forced expiratory volume in 1 second.

Subject ID:
Site:
Signature of Treating Investigator:
Date of evaluation:
Cycle/Day:

	SCORE 0	SCORE 1	SCORE 2	SCORE 3
PERFORMANCE SCORE: ☐ **KPS ECOG LPS**	ϒ Asymptomatic and fully active (ECOG 0; KPS or LPS 100%)	ϒ Symptomatic, fully ambulatory, restricted only in physically strenuous activity (ECOG 1, KPS or LPS 80-90%)	ϒ Symptomatic, ambulatory, capabl of self-care, >50% of waking hours ou of bed (ECOG 2, KPS or LPS 60-70%)	ϒ Symptomatic, limited self-care, >50% of waking hours in bed (ECOG 3-4, KPS or LPS <60%)
SKIN† **SCORE % BSA** *GVHD features to be scored by BSA:* **Check all that apply:** ϒ Maculopapular rash/erythema ϒ Lichen planus-like features ϒ Sclerotic features ϒ Papulosquamous lesions ofichthyosis ϒ Keratosis pilaris-like GVHD	ϒ No BSA involve d	ϒ 1-18% BSA	ϒ 19-50% BSA	ϒ >50% BSA
SKIN FEATURES SCORE:	ϒ No sclerotic features		ϒ **S**uperficial sclerotic features "not hidebound" (able to pinch)	**Check all that apply:** ϒ Deep sclerotic features ϒ "Hidebound" (unable to pinch) ϒ Impaired mobility ϒ Ulceration _

Other skin GVHD features (NOT scored by BSA)
Check all that apply:
ϒ Hyperpigmentation
ϒ Hypopigmentation
ϒ Poikiloderma
ϒ Severe or generalized pruritus
ϒ Hair involvement
ϒ Nail involvement
ϒ *Abnormality present but explained entirely by non-GVHD documented cause (specify):*

MOUTH *Lichen planus-like features present:* ϒ **Yes** ϒ **No**	ϒ No symptoms	ϒ Mild symptoms **with** disease signs but not limiting oral intake significantl y	ϒ Moderate symptoms with disease signs **with** partial limitation of oral intake	ϒ Severe symptoms with disease signs on examination **with** major limitation of oral intake

ϒ *Abnormality present but explained entirely by non-GVHD documented cause (specify):*

	SCORE 0	SCORE 1	SCORE 2	SCORE 3
EYES *Keratoconjunctivitis sicca (KCS) confirmed by ophthalmologist:* ϒ **Yes** ϒ **No** ϒ **Not examined**	ϒ No symptoms	ϒ Mild dry eye symptoms not affecting ADL (requirement of lubricant eye drops ≤ 3 x per day)	ϒ Moderate dry eye symptoms partially affecting ADL (requiring lubricant eye drops > 3 x per day or punctal plugs), **WITHOUT** new vision impairment due to KCS	ϒ Severe dry eye symptoms significantly affecting ADL (special eyeware to relieve pain) **OR** unable to work because of ocular symptoms **OR** loss of vision due to KCS
ϒ *Abnormality present but explained entirely by non-GVHD documented cause (specify):*				
GI Tract ***Check all that apply:*** ϒ Esophageal web/ proximal stricture or ring ϒ Dysphagia ϒ Anorexia ϒ Nausea ϒ Vomiting ϒ Diarrhea ϒ Weight loss ≥5%* ϒ Failure to thrive	ϒ No symptoms	ϒ Symptoms without significant weight loss* (<5%)	ϒ Symptoms associated with mild to moderate weight loss* (5-15%) **OR** moderate diarrhea without significant interference with daily living	ϒ Symptoms associated with significant weight loss* >15%, requires nutritional supplement for most calorie needs **OR** esophageal dilation **OR** severe diarrhea with significant interference with daily living
ϒ *Abnormality present but explained entirely by non-GVHD documented cause (specify):*				
LIVER	ϒ Normal total bilirubin and ALT or AP < 3 x ULN	ϒ Normal total bilirubin with ALT ≥3 to 5 x ULN or AP ≥ 3 x ULN	ϒ Elevated total bilirubin but ≤3 mg/dL or ALT > 5 ULN	ϒ Elevated total bilirubin > 3 mg/dL
ϒ *Abnormality present but explained entirely by non-GVHD documented cause (specify):*				
LUNGS**				
Symptom score:	ϒ No symptoms	ϒ Mild symptoms (shortness of breath after climbing one flight of steps)	ϒ Moderate symptoms (shortness of breath after walking on flat ground)	ϒ Severe symptoms (shortness of breath at rest; requiring 0_2)
Lung score: **%** FEV1 *Pulmonary function tests* ϒ Not performed	ϒ FEV1≥80%	ϒ FEV1 60-79%	ϒ FEV1 40-59%	ϒ FEV1 ≤39%
ϒ *Abnormality present but explained entirely by non-GVHD documented cause (specify):*				

JOINTS AND FASCIA P-ROM score *(see below)* Shoulder (1-7):___ Elbow (1-7): ____ Wrist/finger (1-7): Ankle (1-4):_	ϒ No symptoms	ϒ Mild tightness of arms or legs, normal or mild decreased range of motion (ROM) **AND** not affecting ADL	ϒ Tightness of arms or legs **OR** joint contractures, erythema thought due to fasciitis, moderate decrease ROM **AND** mild to moderate limitation of ADL	ϒ Contractures **WITH** significant decrease of ROM ***AND*** significant limitation of ADL (unable to tie shoes, button shirts, dress self etc.)

ϒ *Abnormality present but explained entirely by non-GVHD documented cause (specify):*

GENITAL TRACT (*See Supplemental figure*‡) ϒ Not examined *Currently sexually active* ϒ Yes ϒ No	ϒ No signs	ϒ Mild signs‡ and females with or without discomfort on exam	ϒ Moderate signs‡ andmay have symptoms with discomfort on exam	ϒ Severe signs‡ with or without symptoms

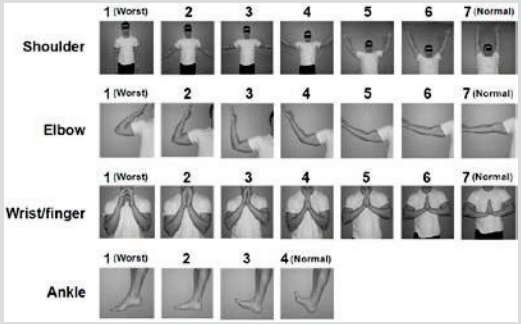
ϒ *Abnormality present but explained entirely by non-GVHD documented cause (specify):*

Other indicators, clinical features or complications related to chronic GVHD (check all that apply and assign a score to severity (0-3) based on functional impact where applicable none – 0,mild -1, moderate -2, severe – 3)

ϒ Ascites (serositis) _____	ϒ Myasthenia Gravis___	
ϒ Pericardial Effusion _____	ϒ Peripheral Neuropathy _____	ϒ Eosinophilia > 500/µl____
ϒ Pleural Effusion(s) _____	ϒ Polymyositis___	ϒ Platelets <100,000/µl ___
ϒ Nephrotic syndrome _____	ϒ Weight loss>5%* without GI symptoms _____	ϒ Others (specify): _____

Overall GVHD Severity *(Opinion of the evaluator)*	❑ **No GVHD**	❑ **Mild**	❑ **Moderate**	❑ **Severe**

Photographic Range of Motion (P-ROM)



† Skin scoring should use both percentage of BSA involved by disease signs and the cutaneous features scales. When a discrepancy exists between the percentage of total body surface (BSA) score and the skin feature score, OR if superficial sclerotic features are present (Score 2), but there is impaired mobility or ulceration (Score 3), the higher level should be used for the final skin scoring.

* Weight loss within 3 months.

**Lung scoring should be performed using both the symptoms and FEV1 scores whenever possible. FEV1 should be used in the final lung scoring where there is discrepancy between symptoms and FEV1 scores.

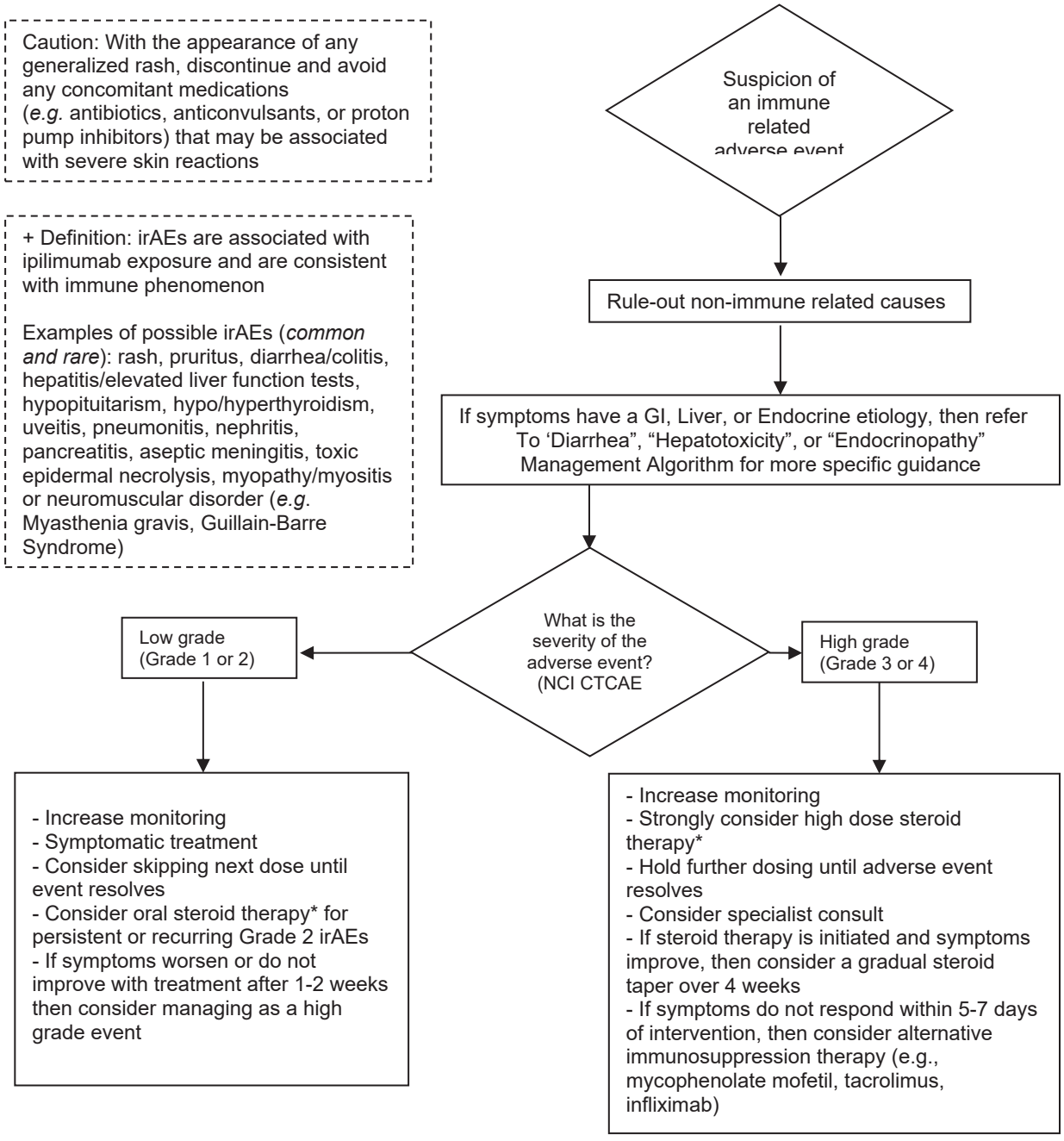
Abbreviations: ECOG (Eastern Cooperative Oncology Group), KPS (Karnofsky Performance Status), LPS (Lansky Performance Status); BSA (body surface area); ADL (activities of daily living); LFTs (liver function tests); AP (alkaline phosphatase); ALT (alanine aminotransferase); ULN (normal upper limit).

‡ To be completed by specialist or trained medical providers (see Supplemental Figure).

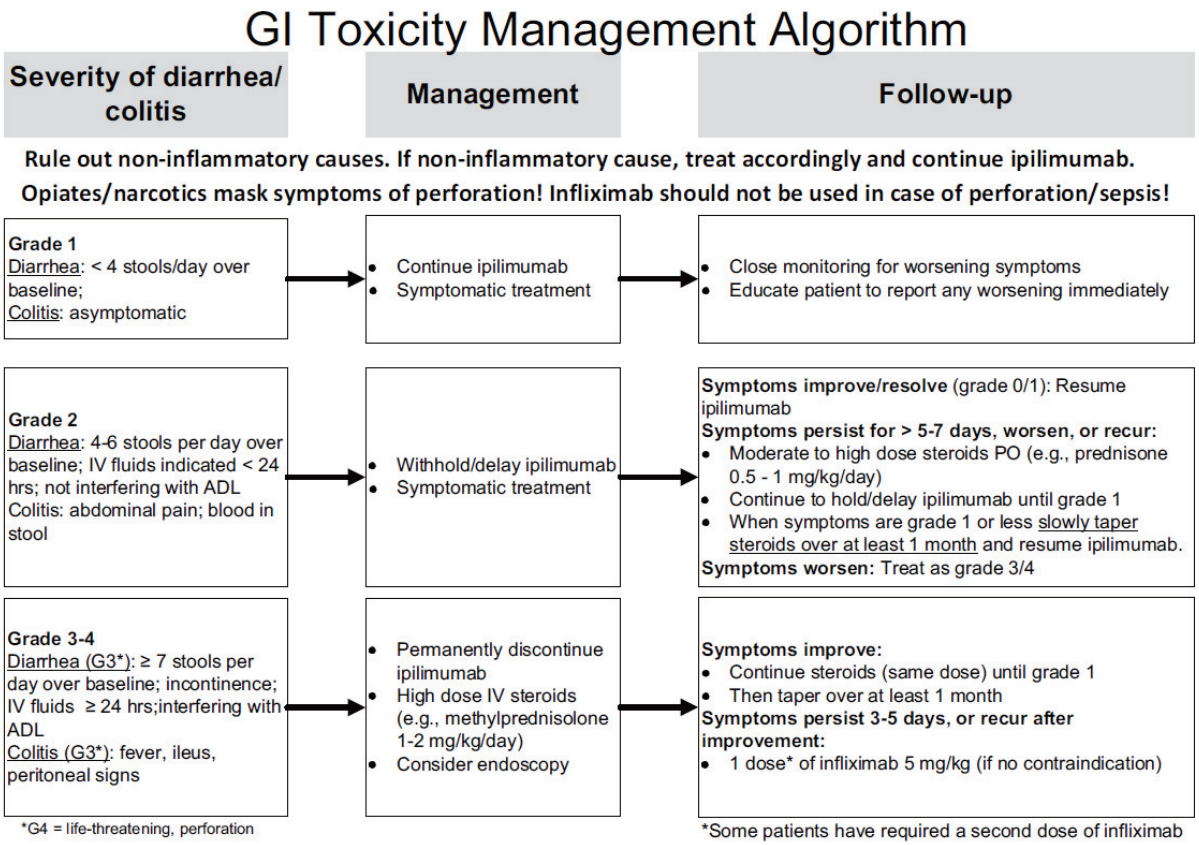
APPENDIX C MANAGEMENT OF IMMUNE-RELATED ADVERSE EVENTS, DIARRHEA, HEPATOTOXICITY, ENDOCRINOPATHY, SKIN AND NEUROPATHY *

*Investigator's Brochure (March 2016)

General Recommendations for Management of Suspected Inflammatory Events



Diarrhea Management Algorithm



Patients on IV steroids may be switched to oral corticosteroid (e.g., prednisone) at an equivalent dose at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of PO corticosteroids.

Diarrhea				
GRADE 1 Increase of <4 stools per day over baseline; mild increase in ostomy output compared with baseline	**GRADE 2** Increase of 4-6 stools per day over baseline; IV fluids indicated < 24 hrs; moderate increase in ostomy output compared to baseline; not interfering with activities of daily living (ADL)	**GRADE 3** Increase of ≥7 stools per day over baseline; incontinence; IV fluids ≥ 24 hrs; hospitalization; severe increase in ostomy output compared to baseline; interfering with ADL	**GRADE 4** Life-threatening consequences (*e.g.*, hemodynamic collapse)	**GRADE 5** Death

Hepatoxicity Management Algorithm

Situation: rising liver function tests (LFTs) >8X ULN or suspected immune-mediated hepatitis

1) Admit subject to hospital for evaluation and close monitoring
2) Stop further ipilimumab dosing until hepatotoxicity is resolved. Consider permanent discontinuation of ipilimumab per protocol
3) Start at least 120 mg methylprednisolone sodium succinate per day, given IV as a single or divided dose
4) Check liver laboratory test values (LFTs, T-bilirubin) daily until stable or showing signs of improvement for at least 3 consecutive days
5) If no decrease in LFTs after 3 days or rebound hepatitis occurs despite treatment with corticosteroids, then add mycophenolate mofetil 1g BID per institutional guidelines for immunosuppression of liver transplants (supportive treatment as required, including prophylaxis for opportunistic infections per institutional guidelines)
6) If no improvement after 5 to 7 days, consider adding 0.10 to 0.15 mg/kg/day of tacrolimus (trough level 5-20 ng/mL)
7) If target trough level is achieved with tacrolimus but no improvement is observed after 5 to 7 days, consider infliximab, 5 mg/kg, once
8) Continue to check LFTs daily for at least 2 weeks to monitor sustained response to treatment

A flow chart of the algorithm is depicted on the following page.

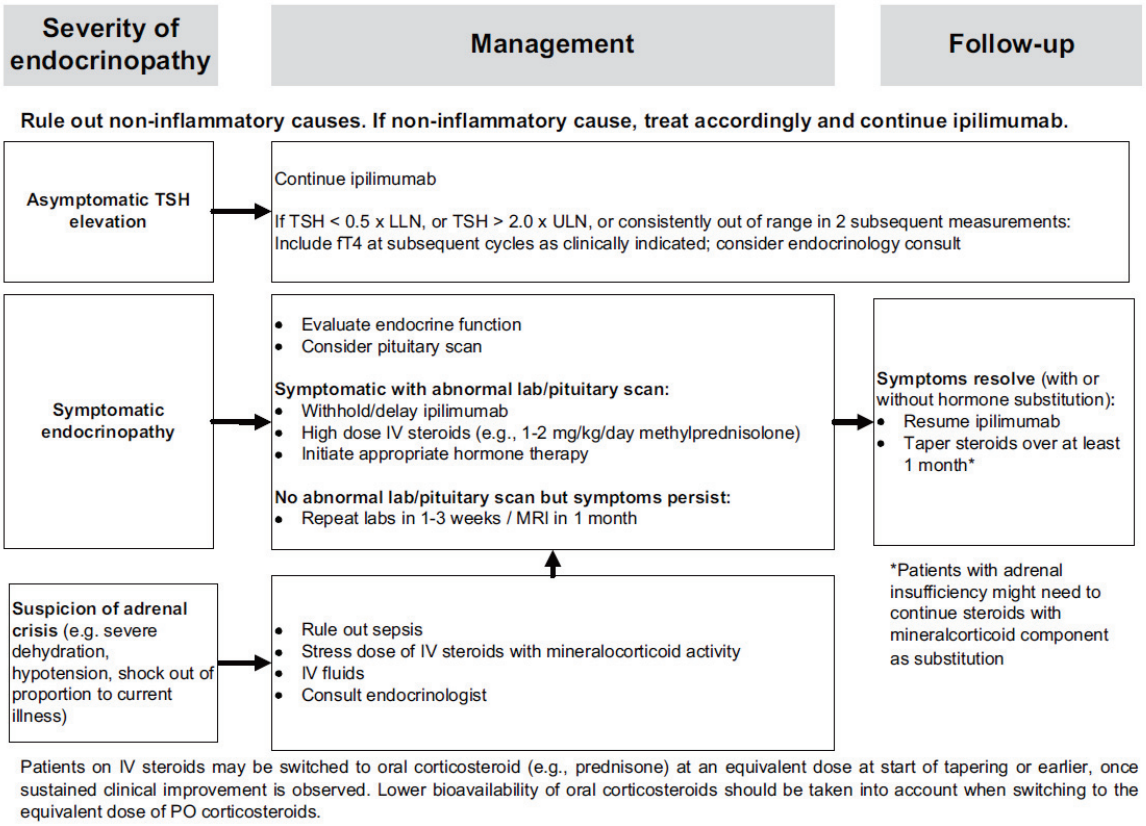
Hepatotoxicity Management Algorithm

Severity of hepatitis	Management	Follow-up
Rule out non-inflammatory causes. If non-inflammatory cause, treat accordingly and continue ipilimumab.		
Caution: Infliximab is contraindicated in hepatic insufficiency!		
Grade 1 AST or ALT ≤ 2.5 x ULN or Total bilirubin ≤ 1.5 x ULN	• Continue ipilimumab	• Continue monitoring LFT prior to each dose
Grade 2 AST or ALT > 2.5 to ≤ 5.0 x ULN **or** Total bilirubin > 1.5 to ≤ 3.0 x ULN	• Increase frequency of monitoring (q3d) • Withhold/delay ipilimumab while investigating alternative etiology	**Return to baseline** • Resume q3w monitoring • Resume ipilimumab **Elevations persist for > 5-7 days or worsen, and no alternative etiologies are identified** • Consider moderate to high dose steroids PO (e.g., 0.5- 1 mg/kg/day prednisone or equivalent)
Grade 3-4 AST or ALT > 5.0 x ULN **or** Total bilirubin > 3.0 ULN	• Discontinue ipilimumab* • Increase frequency of monitoring (q1-3d) • High dose IV steroids (e.g., methylprednisolone 1-2 mg/kg/day**)	**AST/ALT/Tbili return to grade 2** • Taper steroids over at least 1 month **Labs do not decrease over 3-5 days, worsen or rebound:** • Add MMF*** 1 g BID • If no response within 3-5 days, consider other immunosuppressants per local guidelines

*Ipilimumab may be held/delayed rather than discontinued if AST/ALT ≤ 8 x ULN and Tbili ≤ 5 x ULN. Resume ipilimumab when AST/ALT/ Tbili return to grade 2 and meet protocol specific retreatment criteria.
**The recommended starting dose for grade 4 hepatitis is 2 mg/kg/day methylprednisolone IV.
*** MMF, mycophenolate mofetil

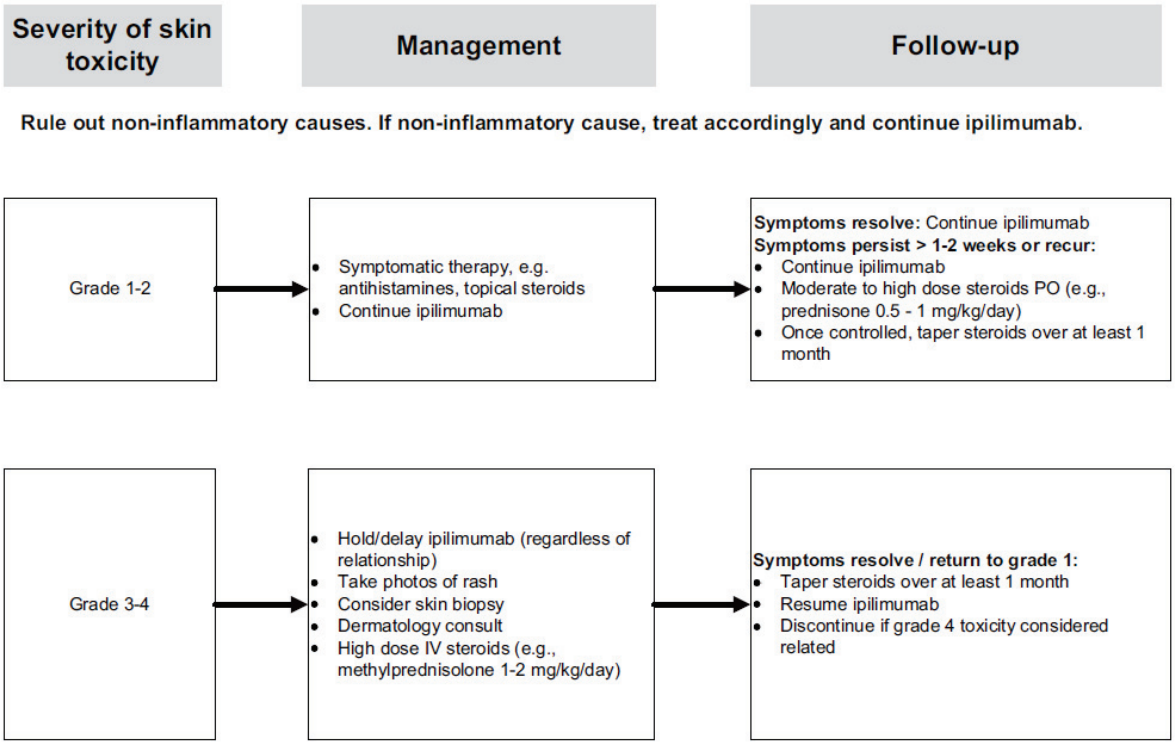
Patients on IV steroids may be switched to oral corticosteroid (e.g., prednisone) at an equivalent dose at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of PO corticosteroids.

Endocrinopathy Management Algorithm



Patients on IV steroids may be switched to oral corticosteroid (e.g., prednisone) at an equivalent dose at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of PO corticosteroids.

Skin Toxicity Management Algorithm



Patients on IV steroids may be switched to oral corticosteroid (e.g., prednisone) at an equivalent dose at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of PO corticosteroids.

Neurological Toxicity Management Algorithm

Rule out non-inflammatory causes. If non-inflammatory cause, treat accordingly and continue ipilimumab. Discontinue ipilimumab for any grade 3-4 motor neuropathy, regardless of relationship.

Severity of neurological toxicity	Management	Follow-up
Grade 1	• Continue ipilimumab	Continue monitoring the patient. If symptoms worsen, treat as below.
Grade 2	• Hold/delay ipilimumab • Treat symptoms per local guidelines	Resume ipilimumab when resolved / grade 1 If symptoms worsen, treat as below.
Grade 3-4 sensory	• Discontinue ipilmumab if considered related • Obtain neuro consult • Treat symptoms per local guidelines • High dose IV steroids (e.g., 1-2 mg/kg/ day methylprednisolone)	**Symptoms resolve / return to grade 2:** • Taper steroids over at least 1 month
Grade 3-4 motor	• Discontinue ipilimumab **regardless of relationship** • Obtain neuro consult • Treat symptoms per local guidelines • High dose IV steroids (e.g., 1-2 mg/kg/ day methylprednisolone)	**Symptoms resolve / return to grade 2:** • Taper steroids over at least 1 month **Symptoms do not resolve or progress; atypical presention:** • Consider IVIg or other immunosuppressive therapies per local guidelines

Patients on IV steroids may be switched to oral corticosteroid (e.g., prednisone) at an equivalent dose at start of tapering or earlier, once sustained clinical improvement is observed. Lower bioavailability of oral corticosteroids should be taken into account when switching to the equivalent dose of PO corticosteroids.

APPENDIX D ACUTE MYELOID LEUKEMIA RESPONSE CRITERIA

Cheson, B et al., J Clin Oncol, 2003, 4642-9.

Morphological Complete Remission (CR):
-Normalization of the peripheral blood absolute neutrophil count > $1.0x10^9$/L, platelets > than $100x10^9$/L no residual evidence of extramedullary disease and bone marrow aspirate with ≤ 5% blasts, no blasts with Auer rods

Morphological Complete Remission with incomplete blood count recovery (CRi):
-Same as CR but without normalization of the peripheral blood absolute neutrophil and platelet count

Relapse from CR or CRi:
-Reappearance of leukemic blasts in the peripheral blood; or> 5% blasts in the bone marrow not attributable to another cause (e.g. recovery of normal cells following treatment –induced aplasia or use of growth factors) OR
-Appearance or reappearance of extramedullary disease.
-If there are no circulating blasts and no extramedullary disease and the bone marrow blast percentage is >5% but <20%, then a repeat bone marrow performed at least 7 days after the first marrow examination and documenting bone marrow blast percentage >5% is necessary to establish relapse.

Partial Remission (PR):
-Normalization of the peripheral blood absolute neutrophil count $1.0x10^9$/L, platelets > than $100x10^9$/L, and at least a 50% decrease in the percentage of marrow aspirate blasts to 5-25% , or marrow blasts <5% with Auer rods.

Cytogenetic Responses
Complete cytogenetic response: An abnormal clone is detected in all metaphases prior to treatment and only normal metaphases are observed in all metaphases following treatment.

Partial cytogenetic response: An abnormal clone is detected in all metaphases prior to treatment and the post-treatment sample has 50% or fewer abnormal metaphases compared to the pretreatment value.

Cytogenetic non-response: An abnormal clone is detected in all metaphases prior to treatment and neither complete nor partial cytogenetic response is observed in post-treatment specimens Specimens with normal cytogenetic results before treatment will not be evaluated for cytogenetic response. The method used to determine cytogenetic response will be standard metaphase cytogenetics.

Stable disease:
-Subjects who fail to achieve CR, Cri, or PR and who do not have criteria for PD will be defined as having the stable disease. If the subject dies prior to response assessment at the end of Cycle 1, then they will be classified as “indeterminate”.

Progressive disease (PD): may be defined as ONE of the following:
->50% increase in peripheral blood or bone marrow blasts from best assessment with minimum threshold of 20% blasts in the marrow or $1.0x10^9$/L blasts in peripheral blood.
-Development of biopsy proven extramedullary leukemia (if the subject has extramedullary disease at baseline, then PD will be defined by blood and marrow criteria or if new sites of extramedullary disease appear).
-If subject who present with an initial marrow blast percentage sufficiently high to preclude the ability to base disease progression on a >50% increase in marrow blast percentage, disease progression should be based on peripheral blood criteria or development of extramedullary leukemia as above.

Persistent Disease
Hematologic Persistence – Bone marrow or peripheral blasts >5% after d+30 following alloSCT

APPENDIX E MDS RESPONSE CRITERIA

Table below from IWG 2006 criteria (Cheson B, Blood, 2006, 419-425)

Category	Response criteria (responses must last at least 4 wk)
Complete remission	Bone marrow: ≤ 5% myeloblasts with normal maturation of all cell lines*
	Persistent dysplasia will be noted*†
	Peripheral blood‡
	Hgb ≥ 11 g/dL
	Platelets ≥ 100 × 10^9/L
	Neutrophils ≥ 1.0 × 10^9/L†
	Blasts 0%
Partial remission	All CR criteria if abnormal before treatment except:
	Bone marrow blasts decreased by ≥ 50% over pretreatment but still > 5%
	Cellularity and morphology not relevant
Marrow CR†	Bone marrow: ≤ 5% myeloblasts and decrease by ≥ 50% over pretreatment†
	Peripheral blood: if HI responses, they will be noted in addition to marrow CR†
Stable disease	Failure to achieve at least PR, but no evidence of progression for > 8 wks
Failure	Death during treatment or disease progression characterized by worsening of cytopenias, increase in percentage of bone marrow blasts, or progression to a more advanced MDS FAB subtype than pretreatment
Relapse after CR or PR	At least 1 of the following:
	Return to pretreatment bone marrow blast percentage
	Decrement of ≥ 50% from maximum remission/response levels in granulocytes or platelets
	Reduction in Hgb concentration by ≥ 1.5 g/dL or transfusion dependence
Cytogenetic response	Complete
	Disappearance of the chromosomal abnormality without appearance of new ones
	Partial
	At least 50% reduction of the chromosomal abnormality
Disease progression	For patients with:
	Less than 5% blasts: ≥ 50% increase in blasts to > 5% blasts
	5%-10% blasts: ≥ 50% increase to > 10% blasts
	10%-20% blasts: ≥ 50% increase to > 20% blasts
	20%-30% blasts: ≥ 50% increase to > 30% blasts
	Any of the following:
	At least 50% decrement from maximum remission/response in granulocytes or platelets
	Reduction in Hgb by ≥ 2 g/dL
	Transfusion dependence

APPENDIX F ADVERSE EVENT GUIDANCEAND PRE TREATMENT CRITERIA CHECKLIST

(Note: this is not all inclusive for all visits, but for general study visit only)

- Clinic Visit
- Physical exam
- ECOG score
- Medication review
- Adverse event evaluation and grading if appropriate
- Immune-related adverse event evaluation:
    - Immediately alert Overall PI (required) if there is concern for cardiac toxicity at any time during treatment including clinical signs or concerns for heart failure, myocardial infarction, cardiomyopathy, or myositis. If there is concern for this, obtain at minimum CPK and troponin, cardiology consult, and EKG and echocardiogram, if it is clinically relevant. Please refer to section 6.4.7 for guidance on cardiac toxicity management for ipilimumab.
    - Prior to initiating treatment, always assess stool frequency (bowel movements per 24 hours).
    - Change in bowel habits (colitis)?
    - Blood in the stool, abdominal pain (colitis)?
    - Headache, especially frontal (hypophysitis)?
    - Unusual fatigue, impaired libido (hypophysitis)?
    - Cough, dyspnea (pneumonitis)?
- Labs including minimum of complete blood counts with differential and comprehensive metabolic panel
- Additional immune related labs every 3 months including: gamma GT, direct bilirubin, LDH, TSH and lipase
- If due for ipilimumab: LFTs must be reviewed and must be:
    - ≤ 3.0 x institutional ULN for AST and ALT
    - ≤1.5 x institutional ULN for total bilirubin
    - If elevated total bilirubin is due Gilbert's disease or disease-related hemolysis then total bilirubin ≤ 3.0 x institutional ULN
    - If, during the course of treatment abnormal LFT values are detected, ipilimumab should not be administered. The subject should be evaluated and recommendations for management are in section 6 of the protocol with helpful algorithms in Appendix C, as clinically appropriate.

APPENDIX G PATIENT SAFETY WALLET MEDICATION CARD

How To Fill Out Your Wallet Medication Card

The Card in Your Wallet That Could Save Your Life!

This wallet medicine card was made to help you and your family remember all the medicines you are taking. Giving your doctor, hospital, or other healthcare workers a complete list of medicines helps them take better care of you.

1. **Always keep this card with you.** Fold it and keep it in your wallet, so it will be handy in case of an emergency.

2. **Fill out the information at the top of the form:**

- Last Adult Immunizations: Write the month and year of your most recent vaccinations (for example a flu or tetanus shot).
- Doctor and Pharmacy or Drug Store Information: Write the name and phone number of each of your doctors and each pharmacy where you get your prescriptions filled. *Since you are enrolled in a clinical trial, be sure to include your experimental or investigational prescription, and your research team's contact information.* This information will make it easier for your doctor or other healthcare workers to figure out who to call with questions about your medicines if you can't answer questions.
- Emergency Contact: Write the name and phone number of the person that you would want to be called in case of an emergency. It is important to list this person in case you are too ill to provide emergency medical workers with information.
- Allergies: List all allergies that you have, including allergies to medicines and to food.
- Other Important Information: List any other information you think a doctor may need to take care of you, including any conditions you have (such as cancer, diabetes or high blood pressure) or if you have a pacemaker or have had a knee or hip replaced.

3. **Fill out the information at the bottom of the form:**

- Write down **ALL** medicines you take (a list of the kinds of medicines to include is provided at the top of the form).
- Start date: Write the date you began taking each medicine. If you don't know the date, list the month that you began taking the medicine (or the year if you have been taking the medicine for a long time).
- Drug name and (amount): For each of your medicines, copy the name of the medicine and amount from the label on the medicine bottle or other container (for example, aspirin 40 mg).
- Dose: Write how much of the medicine you take each time (for example, 2 pills, 3 drops, 2 puffs).
- When do you take it: Write how many times a day you take the medicine, what time of day you take it, and if you take it before or after meals.
- Reason you are taking: Write the reason your doctor said you need the medicine (for example, for your heart).

4. **Update this form when you change any medicine:**
Take this form to all doctor visits, when you go for any medical tests, and all hospital visits. Write down any changes made to your medicines; cross out any medicines that you have stopped taking, add new medicines, or change the dose.

Date of Most Recent Adult Immunizations:

Pneumonia:

Tetanus:

Hepatitis:

Flu:

What medications should I include?
Prescription medicines
Over-The-Counter medicines
Vitamins
Herbal remedies
Nutrition pills
Respiratory therapy medicines (such as inhalers)
Blood factors (such as Factor VIII)
IV solutions
IV nutrition
Investigational or experimental drug

Other Important Information:

I am participating on a clinical trial to treat my MDS or AML ***cancer. The trial number is*** 10026 ***, and I am taking the investigational drug***s
Ipilimumab and Decitabine ,

INVESTIGATIONAL DRUG STUDY CONTACTS

Doctor:
Name:
Phone:

Study Nurse:
Name:
Phone:

Pharmacy:
Name:
Phone:

Doctors:
Name:
Phone:

Name:
Phone:

Pharmacies:
Name:
Phone:

Name:
Phone:

Name:
Phone:

Fold here first

Wallet Medication Card

Name:

Address:

(TIP: Use a return address label)

Phone:

Emergency Contact Name:

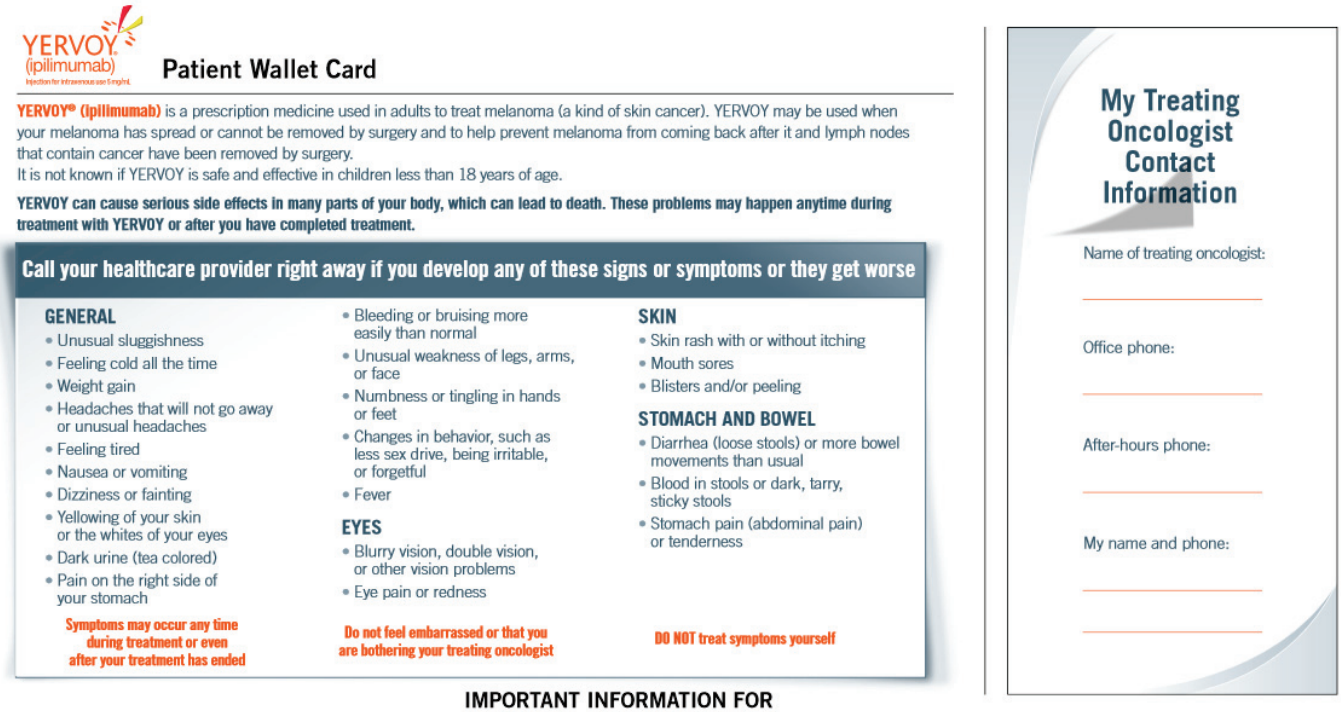
Emergency Contact Phone:

Allergies:

Fold here first

Start date	**Drug Name & (Strength)**	**Dose** (pills, units, puffs, drops)	**When do you take it?** How many times a day? Morning & night? After meals?	**Reason** Why do you take it?
1/1/06	Medicine (40 mg) **(Example)**	2 pills	Once a day with dinner	Heart

Start date	**Drug Name & (Strength)**	**Dose** (pills, units, puffs, drops)	**When do you take it?** How many times a day? Morning & night? After meals?	**Reason** Why do you take it?



YERVOY (ipilimumab)

Patient Wallet Card

YERVOY® (ipilimumab) is a prescription medicine used in adults to treat melanoma (a kind of skin cancer). YERVOY may be used when your melanoma has spread or cannot be removed by surgery and to help prevent melanoma from coming back after it and lymph nodes that contain cancer have been removed by surgery.
It is not known if YERVOY is safe and effective in children less than 18 years of age.

YERVOY can cause serious side effects in many parts of your body, which can lead to death. These problems may happen anytime during treatment with YERVOY or after you have completed treatment.

Call your healthcare provider right away if you develop any of these signs or symptoms or they get worse

GENERAL
- Unusual sluggishness
- Feeling cold all the time
- Weight gain
- Headaches that will not go away or unusual headaches
- Feeling tired
- Nausea or vomiting
- Dizziness or fainting
- Yellowing of your skin or the whites of your eyes
- Dark urine (tea colored)
- Pain on the right side of your stomach

Symptoms may occur any time during treatment or even after your treatment has ended

- Bleeding or bruising more easily than normal
- Unusual weakness of legs, arms, or face
- Numbness or tingling in hands or feet
- Changes in behavior, such as less sex drive, being irritable, or forgetful
- Fever

EYES
- Blurry vision, double vision, or other vision problems
- Eye pain or redness

Do not feel embarrassed or that you are bothering your treating oncologist

SKIN
- Skin rash with or without itching
- Mouth sores
- Blisters and/or peeling

STOMACH AND BOWEL
- Diarrhea (loose stools) or more bowel movements than usual
- Blood in stools or dark, tarry, sticky stools
- Stomach pain (abdominal pain) or tenderness

DO NOT treat symptoms yourself

My Treating Oncologist Contact Information

Name of treating oncologist: _____

Office phone: _____

After-hours phone: _____

My name and phone: _____

_____

IMPORTANT REMINDERS FOR PATIENTS

If you experience any signs or symptoms listed on this card or they get worse, please notify your treating healthcare provider immediately. Getting medical treatment right away may keep the problem from becoming more serious.
- Your healthcare provider will check you for these problems during treatment with YERVOY. Your healthcare provider may treat you with corticosteroid medicines. Your healthcare provider may need to delay or completely stop treatment with YERVOY if you have serious side effects

Be sure to tell all healthcare providers you see that you are being treated with YERVOY and SHOW THEM THIS CARD.
- Take this card with you if you go to the emergency room

For more information, visit **www.YERVOY.com** or call **1-855-YERVOY-1 (1-855-937-8691)**.

IMPORTANT INFORMATION FOR HEALTHCARE PROVIDERS

WARNING: IMMUNE-MEDIATED ADVERSE REACTIONS

PLEASE SEE ATTACHED FULL PRESCRIBING INFORMATION FOR BOXED WARNING AND COMPLETE IMPORTANT SAFETY INFORMATION.

YERVOY (ipilimumab) can result in severe and fatal immune-mediated adverse reactions. These immune-mediated reactions may involve any organ system; however, the most common severe immune-mediated adverse reactions are enterocolitis, hepatitis, dermatitis (including toxic epidermal necrolysis), neuropathy, and endocrinopathy. The majority of these immune-mediated reactions initially manifested during treatment; however, a minority occurred weeks to months after discontinuation of YERVOY.

Permanently discontinue YERVOY and initiate systemic high-dose corticosteroid therapy for severe immune-mediated reactions.

Assess patients for signs and symptoms of enterocolitis, dermatitis, neuropathy, and endocrinopathy and evaluate clinical chemistries including liver function tests (LFTs), adrenocorticotropic hormone (ACTH) level, and thyroid function tests, at baseline and before each dose.

Please see U.S. Full Prescribing Information, **including Boxed WARNING regarding immune-mediated adverse reactions**, and the Medication Guide at the end of this document.

APPENDIX H EUROPEAN LEUKEMIA NET CRITERIA

Table 6. Response criteria in AML

Category	Definition	Comment
Response		
CR without minimal residual disease (CR_{MRD-})	If studied pretreatment, CR with negativity for a genetic marker by RT-qPCR, or CR with negativity by MFC	Sensitivities vary by marker tested, and by method used; therefore, test used and sensitivity of the assay should be reported; analyses should be done in experienced laboratories (centralized diagnostics)
Complete remission (CR)	Bone marrow blasts <5%; absence of circulating blasts and blasts with Auer rods; absence of extramedullary disease; ANC ≥1.0 × 10^9/L (1000/μL); platelet count ≥100 × 10^9/L (100 000/μL)	MRD^+ or unknown
CR with incomplete hematologic recovery (CR_i)	All CR criteria except for residual neutropenia (<1.0 × 10^9/L [1000/μL]) or thrombocytopenia (<100 × 10^9/L [100 000/μL])	
Morphologic leukemia-free state (MLFS)	Bone marrow blasts <5%; absence of blasts with Auer rods; absence of extramedullary disease; no hematologic recovery required	Marrow should not merely be "aplastic"; at least 200 cells should be enumerated or cellularity should be at least 10%
Partial remission (PR)	All hematologic criteria of CR; decrease of bone marrow blast percentage to 5% to 25%; and decrease of pretreatment bone marrow blast percentage by at least 50%	Especially important in the context of phase 1-2 clinical trials
Treatment failure		
Primary refractory disease	No CR or CR_i after 2 courses of intensive induction treatment; excluding patients with death in aplasia or death due to indeterminate cause	Regimens containing higher doses of cytarabine (see Table 8) are generally considered as the best option for patients not responding to a first cycle of 7+3; the likelihood of responding to such regimens is lower after failure of a first
Death in aplasia	Deaths occurring ≥7 d following completion of initial treatment while cytopenic; with an aplastic or hypoplastic bone marrow obtained within 7 d of death, without evidence of persistent leukemia	
Death from indeterminate cause	Deaths occurring before completion of therapy, or <7 d following its completion; or deaths occurring ≥7 d following completion of initial therapy with no blasts in the blood, but no bone marrow examination available	
Response criteria for clinical trials only		
Stable disease	Absence of CR_{MRD-}, CR, CR_i, PR, MLFS; and criteria for PD not met	Period of stable disease should last at least 3 mo
Progressive disease (PD)*,†	Evidence for an increase in bone marrow blast percentage and/or increase of absolute blast counts in the blood: • >50% increase in marrow blasts over baseline (a minimum 15% point increase is required in cases with <30% blasts at baseline; or persistent marrow blast percentage of >70% over at least 3 mo; without at least a 100% improvement in ANC to an absolute level (>0.5 × 10^9/L [500/μL], and/or platelet count to >50 × 10^9/L [50 000/μL] nontransfused); or • >50% increase in peripheral blasts (WBC × % blasts) to >25 × 10^9/L (>25 000/μL) (in the absence of differentiation syndrome)†; or • New extramedullary disease	Category mainly applies for older patient given low-intensity or single-agent "targeted therapies" in clinical trials In general, at least 2 cycles of a novel agent should be administered Some protocols may require blast increase in 2 consecutive marrow assessments at least 4 wk apart; the date of progression should then be defined as of the first observation date Some protocols may allow transient addition of hydroxyurea to lower blast counts "Progressive disease" is usually accompanied by a decline in ANC and platelets and increased transfusion requirement and decline in performance status or increase in symptoms
Relapse		
Hematologic relapse (after CR_{MRD-}, CR, CR_i)	Bone marrow blasts ≥5%; or reappearance of blasts in the blood; or development of extramedullary disease	
Molecular relapse (after CR_{MRD-})	If studied pretreatment, reoccurrence of MRD as assessed by RT-qPCR or by MFC	Test applied, sensitivity of the assay, and cutoff values used must be reported; analyses should be done in experienced laboratories (centralized diagnostics)

ANC, absolute neutrophil count; IDH, isocitrate dehydrogenase; MLFS, morphologic leukemia-free state; WBC, white blood cell.

*The authors acknowledge that this new provisional category is arbitrarily defined; the category aims at harmonizing the various definitions used in different clinical trials.

†Certain targeted therapies, for example, those inhibiting mutant IDH proteins, may cause a differentiation syndrome, that is, a transient increase in the percentage of bone marrow blasts and an absolute increase in blood blasts; in the setting of therapy with such compounds, an increase in blasts may not necessarily indicate PD.

APPENDIX I: IPSS AND IPSS-R

International Prognostic Scoring System -Revised (Greenberg PL et al., Blood, 2012, 2454-2465).

System	Factors	Prognostic Factors Scored	Risk Groups Based on Total Risk Score
IPSS-R	Blast cells in bone marrow (percent)	• Less than or equal to 2 = 0 • Greater than 2 to less than 5 = 1 • 5 to 10 = 2 • Greater than 10 = 3	
	Cytogenetics (chromosome changes)	• –Y, del(11q) = 0 • Normal, del(5q), del(12p), del(20q), double including del(5q) = 1 • del(7q), +8, +19, i(17q), any other single or double independent clone = 2 • -7, inv(3)/+(3q)/del(3q), double including -7/ del(7q), complex: 3 abnormalities = 3 • Greater than 3 abnormalities = 4	• Very Low Total IPSS-R Score = 1.5 or lower • Low Total IPSS-R Score = 2 to 3 • Intermediate Total IPSS-R Score = 3.5 to 4.5
	Cytopenias	Hemoglobin level (g/dL) • Equal to or greater than 10 = 0 • 8 to less than 10 = 1 • Less than 8 = 1.5 Platelet count (x 10^9/L of blood) • Equal to or greater than 100 = 0 • 50 to less than 100 = 0.5 • Less than 50 = 1 Neutrophil count [(ANC) x 10^9/L of blood] • Equal to or greater than 0.8 = 0 • Less than 0.8 = 0.5	• High Total IPSS-R Score = 5 to 6 • Very High Total IPSS-R Score = 6.5 or higher

International Prognostic Scoring System- (Greenberg P et al., Blood, 1997, 2079-2088)

System	Factors	Prognostic Factors Scored	Risk Groups Based on Total Risk Score
IPSS	Blast cells in bone marrow (percent)	• Less than 5 = 0 • 5 to 10 = 0.5 • 11 to 20 = 1.5 • 21 to 30 = 2.0	• Low Total IPSS Risk score = 0
	Cytogenetics (chromosome changes)	• None, del(5q), del(20q) = 0 • 3 or more abnormalities, abnormal chromosome 7 = 1.0 • Other abnormalities = 0.5	• Intermediate –1 Total IPSS Risk Score = 0.5 to 1.0 • Intermediate –2 Total IPSS Risk Score = 1.5 to 2.0
	Cytopenias	Number of cytopenias (anemia, neutropenia or thrombocytopenia) • None or 1 = 0 • 2 or 3 = 0.5	• High Total IPSS Risk Score = 2.5 or higher