Exposure-Response Relationships of the Efficacy and Safety of Ipilimumab in Patients with Advanced Melanoma

Yan Feng, Amit Roy, Eric Masson, Tai-Tsang Chen, Rachel Humphrey, Jeffrey S. Weber

1Bristol-Myers Squibb, Princeton, New Jersey, USA.
2Bristol-Myers Squibb, Wallingford, Connecticut, USA.
3Moffit Cancer Center, Tampa, Florida, USA.

*Current affiliation: MethylGene Inc., Montreal, Quebec, Canada.

Running title: Ipilimumab exposure-response relationships in advanced melanoma

Keywords: advanced melanoma; cytotoxic T-lymphocyte antigen 4; exposure-response relationship; ipilimumab; steady-state trough concentration

Financial support: Research support for this study was provided by Bristol-Myers Squibb Co.

Corresponding author:
Jeffrey S. Weber, M.D., Ph.D.
Moffitt Cancer Center
12902 Magnolia Drive SRB-2
Tampa, FL 33612 USA
Phone: 813-745-2007
Fax: 813-449-8260
E-mail: jeffrey.weber@moffitt.org
Conflicts of interest: Yan Feng, Eric Masson, Amit Roy and Tai-Tsang Chen are currently employed by the study sponsor (Bristol-Myers Squibb Co). Rachel Humphrey is a paid consultant for Merck-Serono, Johnson & Johnson, and the University of Toronto. Jeffrey Weber has received honoraria for consulting (<$10,000 dollars annually) from GlaxoSmithKline, Genentech, Bristol-Myers Squibb, Abbott, and AstraZeneca. Jeffrey Weber also declares equity in Altor Bioscience, Celldex Therapeutics, and Genesis Biopharma.

Word count: 4559 (Max. 5000 words allowed)

Number of figures and tables: 2 tables, 4 figures

Number of references: 19
Abstract

**Purpose:** This retrospective analysis was conducted to characterize ipilimumab exposure-response relationships for measures of efficacy and safety in patients with advanced melanoma.

**Experimental design:** Data were pooled from 498 patients who received ipilimumab monotherapy at 0.3, 3 or 10 mg/kg in one of four completed phase II clinical trials. The relationships between steady-state ipilimumab trough concentration (Cminss), complete or partial tumor response (CR or PR), and safety (immune-related adverse events; irAEs) were described by logistic regression models. The relationship between exposure and overall survival was characterized using a Cox proportional-hazards model.

**Results:** The steady-state trough concentration of ipilimumab was found to be a significant predictor of a CR or PR (p < 0.001). Model-based estimates indicate that the probabilities of a CR or PR at median Cminss for the 0.3, 3, and 10 mg/kg groups were 0.6%, 4.9%, and 11.6%, respectively. Overall survival at the median Cminss for ipilimumab at 0.3 mg/kg was estimated to be 0.85- and 0.58-fold lower relative to that at the median Cminss for 3 and 10 mg/kg, respectively. Model-based estimates indicate that the probabilities of a grade ≥3 irAE at the median Cminss for the 0.3, 3, and 10 mg/kg doses were 3%, 13%, and 24%, respectively.

**Conclusions:** Higher doses of ipilimumab produce greater Cminss that may be associated with increased tumor responses, longer survival, and higher rates of irAEs. The efficacy and safety of ipilimumab at 3 vs. 10 mg/kg in patients with advanced melanoma is being evaluated in an ongoing phase III trial.
Translational Relevance

Ipilimumab monotherapy at 3 mg/kg is currently approved in several countries for the treatment of advanced (unresectable stage III or IV) melanoma, based on the results of a phase III, randomized controlled trial (MDX010-20) in which ipilimumab at 3 mg/kg demonstrated a statistically significant and clinically meaningful improvement in overall survival in previously treated patients. We conducted a retrospective analysis of data from four phase II studies, investigating the relationship between ipilimumab exposure and measures of efficacy (including overall survival) as well as safety. These analyses include survival data from a wide dose range (0.3 to 10 mg/kg) that were not available when study MDX010-20 was initiated, and suggest that higher ipilimumab exposure is associated with better survival, albeit at a greater risk of adverse events. A formal comparison of ipilimumab monotherapy at 3 vs. 10 mg/kg is currently being evaluated in a separate phase III trial.
Introduction

Cytotoxic T-lymphocyte antigen-4 (CTLA-4) is a key immune checkpoint molecule that down-regulates T-cell activation by binding to its ligands, B7-1 (CD80) and B7-2 (CD86), on antigen-presenting cells (1). Ipilimumab is a fully human IgG1 monoclonal antibody that blocks CTLA-4 from binding to its ligands, thereby augmenting antitumor immune responses (2). Consistent with its proposed mechanism of action, ipilimumab has been shown to increase the percentage of activated CD4+ and CD8+ T cells in peripheral circulation, with a concomitant decrease in naïve CD4+ and CD8+ T cells, in patients with advanced (unresectable stage III or IV) melanoma (3). Ipilimumab has demonstrated a statistically significant improvement in overall survival (OS) in two randomized controlled, phase III trials of patients with advanced melanoma; one with ipilimumab monotherapy at 3 mg/kg in previously treated patients (4) and the other with ipilimumab at 10 mg/kg in combination with dacarbazine in previously untreated patients (5).

Ipilimumab monotherapy has been extensively studied in phase II clinical trials of patients with advanced melanoma, most of whom had received prior therapy for metastatic disease (6–8). The results of one of these trials (CA184-022) suggested that ipilimumab elicits a dose-dependent effect on both efficacy and safety endpoints, where the best overall response (BOR) rates were 11.1%, 4.2%, and 0% for ipilimumab at doses of 10, 3, and 0.3 mg/kg, respectively (6). Across ipilimumab clinical trials, the most common treatment-related adverse events were immune-related (irAEs) (4–8), i.e., AEs that are inflammatory in nature and consistent with an immune-based mechanism of action (2, 9). The incidence of irAEs of any grade, and irAEs of grade 3-4, showed a dose-dependent increase with ipilimumab in study CA184-022 (6). These efficacy and safety outcomes correlated with the results of population pharmacokinetic analyses, which showed greater achievement of the target trough concentration during the induction dosing period (every 3 weeks for 4 doses) for ipilimumab at 10 vs. 3 mg/kg, and with the results of pharmacodynamic analyses, which showed a greater mean rate of increase in absolute lymphocyte count (ALC) at 10 mg/kg compared with lower doses (6).

Ipilimumab continues to be evaluated in melanoma and other tumor types, including castration resistant prostate cancer and non–small cell lung cancer (2, 10, 11). To better understand the risk-benefit profile of ipilimumab monotherapy across the dose range evaluated in clinical studies, we conducted a retrospective analysis of pooled data from phase II trials including all patients for whom pharmacokinetic samples were available. More than 450 patients with advanced melanoma were included in the analyses, which were done in order to characterize ipilimumab exposure-response (E-R) relationships for important measures of efficacy and safety. Specifically, the E-R relationships for tumor response and OS were analyzed to characterize efficacy and those for irAEs were analyzed to characterize safety.
Materials and Methods

Patients

The patients included in the current analyses had participated in one of the following four completed phase II clinical trials which evaluated ipilimumab monotherapy in advanced melanoma: CA184-004, bmyCA184-007, CA184-008, and CA184-022 (6–8, 12). Study CA184-004 investigated potential biomarkers of clinical activity in patients who received ipilimumab at 3 or 10 mg/kg (12), and study CA184-007 evaluated the impact of prophylactic oral budesonide on the rate of grade ≥2 diarrhea in patients who received ipilimumab at 10 mg/kg (7). Both of these studies included previously treated and previously untreated patients. In study CA184-022 (6), patients intolerant to or who progressed on prior therapy were randomized to ipilimumab at 0.3, 3, or 10 mg/kg, whereas in study CA184-008, only one dose of ipilimumab (10 mg/kg) was evaluated in previously treated patients (8). In all four of these studies, ipilimumab was given every 3 weeks for up to 4 doses (induction phase), followed by a maintenance phase (every 12 weeks beginning at week 24) in eligible patients. The first tumor assessment was performed at week 12 (end of induction dosing). All patients, or their legal representatives, gave written informed consent to participate in their respective trials. A summary of patient demographics and laboratory values for the E-R analyses is given in Supplementary Table 1.

Data analysis

The E-R analysis of efficacy was characterized by two measures of tumor response and by overall survival (OS), and the E-R analysis of safety included toxicities characterized as irAEs. The two measures of tumor response were: (1) BOR of partial response (PR) or complete response (CR) by mWHO criteria, and (2) clinical activity of PR and CR by immune-related Response Criteria (irRC), an exploratory endpoint defined to capture the delayed tumor responses that are sometimes observed with immunotherapy (13). The irRC are derived from mWHO criteria and include new lesions in the assessment of total tumor burden; in contrast to standard criteria, the irRC do not necessarily characterize patients with new lesions as having progressive disease (13).

The E-R analysis of BOR by mWHO criteria was conducted with data from patients in studies CA184-007, CA184-008, and CA184-022 for whom summary measures of ipilimumab exposure and BOR assessment were available (N=354; ~73% of the total treated). Data from study CA184-004 were not included in the E-R analyses of BOR as tumor assessments in this trial were not adjudicated by an independent review committee (IRC), and patients who underwent biopsy of the index lesions were censored for efficacy. Similarly, the E-R analysis of clinical activity by irRC was conducted with data from patients in studies CA184-007, CA184-008, and CA184-022 for whom summary
measures of ipilimumab exposure and the assessment of clinical activity were available [N=419; 86% of the total number of patients treated/randomized (487)].

The E-R relationships for efficacy (OS) and safety (irAE) were analyzed with data from patients in studies CA184-004, CA184-007, CA184-008, and CA184-022 for whom summary measures of ipilimumab exposure were available [N=498; 88% of the total number of patients treated/randomized (569)]. The safety endpoint reflected the worst reported grade of an irAE in any one of the categories: gastrointestinal, hepatic, skin, endocrine, and other.

The measure of ipilimumab exposure employed in the E-R analyses was ipilimumab steady-state trough concentration (Cminss) during the induction phase, as determined by a previously developed population pharmacokinetic model (6), using data from the four phase II studies. PK concentrations in samples from patients enrolled in the four phase II studies were analyzed by a validated ELISA assay that had a lower limit of quantification of 0.4 µg/mL (14). Model validation in a previously published linear two-compartment PPK model with zero-order IV infusion and first-order elimination have shown that it provided a good description of the entire ipilimumab serum concentration-time profile for the 3 and 10 mg/kg doses (15). The observed and model-predicted PK profile for patients who received ipilimumab at 3 or 10 mg/kg is presented in Supplementary Figure 1. Steady-state peak and time-averaged concentrations were not selected for these analyses, as they are highly correlated with Cminss (correlation coefficient >0.9). Moreover, the pharmacologic rationale for Cminss was considered to be stronger than the other two summary measures of exposure, based on the assumption that T-cell activation in the induction phase of treatment is maximized by blocking the interaction of CTLA-4 with its ligands over the entire dosing interval.

**E-R analysis: Tumor response**

The E-R analysis of tumor response was characterized by proportional-odds logistic regression models relating Cminss to the probability of achieving BOR of (CR or PR), as defined by mWHO criteria as well as clinical activity by irRC, and based on IRC adjudicated tumor size measurements. The logit (log-odds) are given by:

$$\text{logit Pr}_{l,BOR} = \log\left(\frac{P_{l,BOR}}{1 - P_{l,BOR}}\right) = \beta_0 + \beta^T X_i,$$

where $\beta_0$ and $\beta$ are scalar and vector parameters that represent baseline odds and the effect of $X_i$ on achieving BOR or clinical activity, respectively. The model parameters were estimated by maximum likelihood. This model formulation assumes that the predictor variables $X_i$ have proportional effects on the odds of Pr(BOR) or Pr(clinical activity).
Model development was conducted in 3 stages. First, a base model was developed to establish the existence and functional form of a relationship between ipilimumab Cmin ss and probability of a tumor response. Second, the covariate effects that may potentially modulate the E-R relationship were examined in a full model. Third, the final model was developed by backward elimination to retain only the covariates that were significant at a 1% level. The final model was evaluated by assessing the agreement between the observed and predicted proportion of tumor response and the associated 90% model prediction intervals (PI) in each dose group (0.3, 3, and 10 mg/kg). The model-predicted proportions (and associated 90% PI) in each dose group were obtained by simulation using the model-predicted probability of response of the patients in each dose group.

The full model examined the potential effects of the following covariates: body weight, age, gender, Eastern Cooperative Oncology Group performance status (ECOG PS), baseline serum lactate dehydrogenase (LDH) levels, concomitant budesonide, prior systemic anticancer therapy, metastatic stage (M-stage), prior immunotherapy, HLA-A*0201 status, and prior interleukin (IL)-2 therapy.

**E-R analysis: OS**

The E-R analysis of OS was characterized by a Cox proportional-hazards (CPH) model relating ipilimumab Cmin ss to the hazard of death. The hazard function is expressed as \( \dot{\lambda}(t) = \dot{\lambda}_0(t) \exp(\beta^T x_i) \), where \( \dot{\lambda}_0(t) \) is the baseline hazard function and \( x_i \) is a vector of predictor variables. The parameter vector \( \beta \) is estimated by maximum partial-likelihood.

We also assessed the potential effects of the following covariates on the E-R relationship: age, weight, gender, baseline ALC, HLA-A*0201 status, baseline LDH levels, prior systemic anticancer therapy, prior immunotherapy, prior IL-2 therapy, ECOG PS, and M-stage at study entry.

The CPH model was developed in 3 stages: (1) a base model was developed to establish the existence and functional form of the E-R relationship between OS and ipilimumab exposure (Cmin ss); (2) a full model was developed to assess the effect of all the covariates simultaneously; (3) the final model was developed by retaining the covariates that were significant at a 1% level, with appropriate functional forms of their relationships with OS. The CPH model was evaluated by comparing model-predicted cumulative probability of OS versus time with that obtained by Kaplan-Meier (KM) analyses.

**E-R analysis: irAEs**

The E-R of irAEs was characterized by a proportional-odds ordinal logistic regression model relating Cmin ss to the probability of experiencing grade \( \geq 2 \) or grade \( \geq 3 \) irAEs. The logit (log-odds) of grade \( \geq 2 \) and grade \( \geq 3 \) irAEs are given by: 
\[
\logit p_{1_{\text{Grade} \geq 2}} = \log\left(\frac{p_{1_{\text{Grade} \geq 2}}}{1 - p_{1_{\text{Grade} \geq 2}}}\right) = \beta_{\text{Grade} \geq 2} + \beta^T x_i, \quad \text{Eq. 1}
\]
\[
\logit p_{1_{\text{Grade} \geq 3}} = \log\left(\frac{p_{1_{\text{Grade} \geq 3}}}{1 - p_{1_{\text{Grade} \geq 3}}}\right) = \beta_{\text{Grade} \geq 2} + \beta_{\text{Grade} \geq 3} + \beta^T x_i, \quad \text{Eq. 2}
\]

where \(\beta_{\text{Grade} \geq 2}, \beta_{\text{Grade} \geq 3}\) and \(\beta\) are scalar and vector parameters that represent baseline odds of grade \(\geq 2\) and grade \(\geq 3\) irAEs and the effect of \(x\), [i.e., Cminss, log(Cminss)] on achieving irAEs, respectively. The model parameters were estimated by maximum likelihood. This model formulation assumes that the predictor variables \(x\) have proportional effects on the odds of probability of irAEs.

Model development was conducted in three stages, similar to that for the E-R analysis of tumor response. Covariate-parameter relationships were examined for the baseline covariates of body weight, age, gender, ECOG PS, baseline LDH levels, concomitant budesonide, prior systemic anticancer therapy, metastatic stage, prior immunotherapy, HLA status, and prior IL-2 therapy. The final model was evaluated by assessing the agreement between the observed proportion of irAEs by dose and the associated 90% model PI.

**Results**

**E-R analysis: BOR by mWHO**

Cminss was found to be a statistically significant predictor of the probability of a BOR (CR or PR) by mWHO criteria \((p < 0.001)\), and the probability was found to increase with an increase in log-transformed Cminss (Table 1 and Figure 1A). However, none of the covariates examined were retained in the final model as none had statistically significant effect on the E-R relationship for BOR by mWHO criteria.

The odds of a patient achieving a BOR by mWHO criteria increased by 2.41-fold for a 2.7-fold increase in Cminss [log(Cminss) increases by one unit for approximately 2.7-fold increase in Cminss]. The observed responders at 0.3, 3 and 10 mg/kg were 0%, 6% and 11.7%, respectively, consistent with model-estimated probabilities of a BOR at median Cminss for 0.3, 3, and 10 mg/kg groups of 0.6%, 4.9%, and 11.6%, respectively. Figure 1A shows the results of model evaluation that compares observed and predicted proportion of mWHO responders at each dose level. The results from a predictive check showed that there is good agreement between the model-predicted probability of BOR and the observed proportion of BOR responders. Estimates of covariate effects from the full E-R
model for BOR are presented in Supplementary Figure 2, and the results suggest that Cminss had a greater effect on achieving a tumor response than other covariates.

**E-R analysis: Clinical activity by irRC**

Cminss was found to be a statistically significant predictor of the probability of clinical activity by irRC (p < 0.001), and the probability increased with higher log-transformed Cminss (Table 1 and Figure 1B). However, as with BOR by mWHO criteria, none of the covariates examined were retained in the final model as none had statistically significant effect on the E-R relationship for clinical activity by irRC.

The odds of a patient achieving clinical activity by the irRC increased by 1.76-fold for a 2.7-fold increase in Cminss. The observed clinical activities by irRC at 0.3, 3, and 10 mg/kg were 6.3%, 15.0% and 25.1%, respectively, consistent with the model-estimated probabilities of irRC responses at median Cminss for the 3 dose groups of 4.5%, 15.1%, and 25.6%, respectively. These results suggest that the probability of achieving clinical activity by the irRC increased with dose of ipilimumab. Figure 1B shows the observed proportion and model-predicted median proportion/probability of an irRC response vs. Cminss. The results from a predictive check showed that there is good agreement between the model-predicted probability of irRC responses and the observed proportion of irRC responses. Estimates of covariate effects from the full E-R model for tumor response by irRC are presented in Supplementary Figure 3, and similar to BOR by mWHO criteria, Cminss appeared to have a greater effect on achieving a tumor response than other covariates.

**E-R analysis: OS**

From the KM analysis, OS improved with increasing ipilimumab exposure (Cminss) and dose as illustrated in Figure 2, although OS appears to be more closely associated with exposure than dose. Almost all the patients in the 2nd and 3rd tertiles were from the 10 mg/kg dose group (approximately 90% and 99%, respectively), whereas most patients in the 1st tertile were from the 0.3 and 3 mg/kg dose groups (approximately 29%, and 49% respectively). Patients in the highest tertile of Cminss appeared to have markedly better OS than patients in the lower tertiles of Cminss, suggesting the existence of an E-R relationship even within the 10 mg/kg dose group. However, it is important to note that the KM analysis for OS by Cminss does not account for the potential effect of confounding variables (such as LDH levels), and therefore model-based analyses were performed.

Estimates from the full E-R model for OS are presented in Supplementary Figure 4, and the results suggest that baseline LDH levels and Cminss had greater effects on the risk of death relative to other covariates. The results of the final model show that Cminss, baseline LDH, and ECOG PS were
significant predictors of OS (p < 0.001). Importantly, HLA-A*0201 genotype did not have a significant impact on E-R OS in this analysis. OS improved with increasing Cminss, decreased with increasing LDH, and was worse for patients with ECOG PS >0. Patients in the 5th percentile of Cminss (1.75 µg/mL) had an OS hazard ratio of 1.52 relative to patients with median Cminss of 43.9 µg/mL. Patients in the 95th percentile of LDH levels (846 IU/L) had an OS hazard ratio of 3.27 relative to those with median LDH levels (206 IU/L), which was less than the upper limit of normal (225 IU/L). Furthermore, the estimated OS hazard ratio for patients with an ECOG PS >0 was 1.72 relative to patients with an ECOG PS of 0. The hazard ratios at the 5th and 95th percentiles of the continuous predictors Cminss and LDH, relative to their median values, indicate that the magnitude of the effect was greater for LDH than for Cminss (Table 2).

At the median Cminss determined for the 0.3 mg/kg dose of ipilimumab, OS was estimated to be 0.85-fold and 0.58-fold lower than at the median Cminss for the 3 mg/kg and 10 mg/kg doses, respectively. The results of the final CPH model are presented in Figure 3. In the final model, the effects of the continuous predictors Cminss and LDH and the categorical predictor ECOG PS were evaluated by a 4×2 stratification of the data according to whether Cminss and LDH were above or below the median values in patients with ECOG PS of 0 or >0 for the dataset. For all of the stratified groups, there was generally good agreement between the model-predicted cumulative probability of OS and the corresponding KM curves (the latter lying within the 90% model PI) (Supplementary Fig. 5). The predicted distribution of OS in patients by LDH level, ECOG PS, and ipilimumab exposure (median Cminss at 0.3, 3 or 10 mg/kg) indicates that the effect of LDH on the risk of death is greater than that for ECOG PS and Cminss (Fig. 3). Patients with greater ipilimumab exposure appear to have lower risk of death at any given value of LDH and ECOG PS relative to patients with lower exposure at the same value for LDH and ECOG PS.

**E-R analysis: irAEs**

As depicted in Figure 4, the probability of experiencing grade ≥2 and grade ≥3 irAEs appeared to increase with increasing ipilimumab exposure; however, patients who did not experience an irAE during the induction period were not likely to experience an irAE during maintenance therapy with ipilimumab. The KM analysis of irAEs show that the majority of events occurred during the induction dosing period (Supplementary Figs. 6 and 7).

Estimates from the full E-R irAE model are graphically presented in Supplementary Figure 8. The results suggest that Cminss had the largest effect on the occurrence of an irAE among the covariates tested. The results of the final model (developed by backward elimination) suggest that, among the covariates tested, only ipilimumab Cminss was identified as a significant predictor for the occurrence...
of an irAE (p <0.001). HLA-A*0201 genotype was not found to have a significant impact on E-R irAEs in this analysis.

The E-R analysis indicated that the probability of experiencing a grade ≥2 and grade ≥3 irAE increased with increasing Cminss (Table 1). The point estimates of the odds ratio for 5th:median Cminss (1.75 μg/mL) and 95th (103 μg/mL):median Cminss (43.9 μg/mL) were 0.124 and 1.74, respectively; therefore, the odds of a patient experiencing a grade ≥3 irAE (versus no irAE, grade 1 or grade 2) are expected to increase by 1.74-fold for a Cminss increase from 43.9 (median) to 103 μg/mL (95th percentile). Model-predicted probabilities of experiencing a grade ≥2 irAE at the median Cminss (5th, 95th percentiles) for the 0.3, 3, and 10 mg/kg treatment groups are approximately 9.8% (5.6, 14), 33% (19, 43), and 51% (34, 62), respectively, and the corresponding probabilities of experiencing a grade ≥3 irAE are approximately 3.3% (1.8, 4.9), 13% (6.8, 19), and 24% (14, 33) (Fig. 4). The results from the predictive check indicate that there is good agreement between the model-predicted probability of grade ≥2 and grade ≥3 irAEs and the observed proportion of grade ≥2 and grade ≥3 irAEs.

**Discussion**

The current pooled analyses of data from phase II trials in advanced melanoma represent the first quantification of E-R relationships for ipilimumab efficacy and safety endpoints in any tumor type. Specifically, the purpose of this study was to determine if there is an association between pharmacokinetics (exposure) and key clinical outcomes with ipilimumab, and to determine baseline patient characteristics that may impact these relationships. While there are two completed phase III clinical trials of ipilimumab in patients with advanced melanoma (4, 5), data from the four completed phase II studies included herein provide the largest dataset across a consistent patient population and treatment regimen, and provide a range of evaluated ipilimumab doses. Exposure-response analyses for ipilimumab at 10 mg/kg in combination with dacarbazine in patients with previously untreated advanced melanoma (5) are the subject of separate publications.

One of the objectives of the E-R analyses for efficacy was to characterize the relationship between ipilimumab exposure and objective response. Using either mWHO criteria or the exploratory irRC, the E-R analyses indicated that Cminss during the induction phase was a statistically significant predictor of response. By mWHO criteria, the probability of BOR increased with a higher log-transformed Cminss, even though the median Cminss at 10 mg/kg was well above the target concentration (20 μg/mL) required for maximal inhibition of CTLA-4 binding to its ligands (6). While a similar association was observed using irRC, model-based estimates indicated that the probabilities
of a tumor response at the median Cminss for 0.3, 3, and 10 mg/kg doses were greater when evaluated by the irRC [4.3%, 15.0%, and 25.6% for irRC vs. 0.6%, 4.9%, and 11.6% for BOR by mWHO criteria, respectively]. This observation reinforces the potential value of the irRC as a tool for characterizing the efficacy of immunotherapies, where durable objective responses are sometimes seen in patients who are characterized as having disease progression by mWHO criteria. Collectively, regardless of which efficacy criteria are used, these results suggest that the probability of achieving an objective response is greater with higher ipilimumab exposure. None of the covariates tested, including baseline LDH levels, had a statistically significant effect on the E-R analyses for tumor response.

An E-R analysis was performed to characterize the relationship between ipilimumab exposure and OS using a Cox proportional-hazard model. The analysis showed that there was a statistically significant relationship between ipilimumab Cminss and the hazard ratio for OS. Patients at the 5th percentile of Cminss (1.75 μg/mL) had an OS hazard ratio of 1.52 relative to patients with median Cminss (43.9 μg/mL), and the OS of patients at the 95th percentile of Cminss (103 μg/mL) had an OS hazard ratio of 0.552 relative to patients at the median Cminss. These results suggest that OS improves with increasing Cminss and ipilimumab dose.

The OS of patients with advanced melanoma is known to be associated with several prognostic factors, including ECOG PS and baseline serum LDH (16, 17). LDH has been shown to be an independent prognostic factor for survival even after accounting for site and number of metastases, and has been incorporated into the current AJCC staging classification (16). The K-M analysis with respect to dose should be interpreted as being suggestive of a qualitative dose response rather than a definitive quantification of the dose response, as the pooled phase II data included in the analyses are not balanced with respect to prognostic factors across doses. Similarly, the differences in OS in K-M analyses by Cminss tertiles should be interpreted as being indicative of a qualitative exposure-response, because this univariate analysis does not account for potential imbalances in risk factors between the Cminss groups. However, the multivariate Cox-proportional hazards model does include the key prognostic factors, and this model provides an estimate of the effect of Cminss on reducing the risk of death after adjusting for the presence of these risk factors.

Among the covariates tested, baseline serum LDH levels and ECOG PS were found to be significant predictors of OS. Our results further show that HLA-A*0201 status does not significantly impact E-R relationships for OS or safety, consistent with the results of a retrospective analysis of ipilimumab clinical trial data showing that efficacy and safety outcomes were independent of HLA-A*0201 status (18). Thus, while the phase III trial of ipilimumab monotherapy at 3 mg/kg enrolled only patients that
were HLA-A*0201-positive (4), the results collectively suggest that ipilimumab efficacy and safety are independent of HLA-A*0201 status.

The risk of death increased with increasing LDH levels, and was higher for patients with ECOG PS>0 compared to those with an ECOG PS of 0. LDH appeared to have a greater effect on the risk of death than Cminss or ECOG PS in our E-R analyses. Although baseline LDH levels do not have a clinically meaningful impact on the systemic clearance of ipilimumab, patients with lower baseline LDH levels tended to have higher ipilimumab concentrations, which may have impacted OS. However, LDH levels and Cminss were accounted for in the E-R model and both were found to be independent covariates for OS. Interestingly, Cminss appeared to be more closely associated with OS than treatment group (dose). Based on the E-R OS analysis, a higher dose of ipilimumab may also provide a survival benefit for patients with poor prognostic features, e.g., elevated baseline LDH levels. This result is consistent with the improved OS in phase III trials of ipilimumab in advanced melanoma, where patients with normal and elevated LDH levels were enrolled (4, 5).

The E-R analysis for safety showed that the probability of experiencing an irAE of grade ≥2 or grade ≥3, and the probability of first occurrence of an irAE at a given time point, increased with increasing Cminss over the evaluated dose range. The model predicted that the probabilities of grade ≥3 irAEs were approximately 3%, 13%, and 24% at the median Cminss of the 0.3, 3, and 10 mg/kg doses. These data are consistent with the frequencies of grade 3-4 irAEs reported in the phase III trial of ipilimumab monotherapy at 3 mg/kg, which ranged from 10.2% to 14.5% (4). IrAEs may reflect ipilimumab’s immune-based mechanism of action and the majority occur during the induction dosing period. They can be severe and life-threatening, but most were reversible with treatment guidelines developed in ipilimumab clinical trials (4–8; 19). There were no study drug-related deaths in CA184-007 (7), one at 3 mg/kg in CA184-022 (6), and one at 10 mg/kg in CA184-008 (8). In the first phase III trial, 12 deaths in 511 treated patients (2.3%) were related to ipilimumab at 3 mg/kg (4). However, there were no study drug-related deaths in the subsequent phase III trial in 247 patients who received ipilimumab at 10 mg/kg plus dacarbazine (5). Thus, while greater ipilimumab exposure increases the frequency of grade ≥3 irAEs, the availability of treatment guidelines for the management of irAEs may prevent this increased incidence from causing a higher rate of treatment-related deaths.

The results of the current analyses demonstrate a positive association between ipilimumab exposure, and by extension dose, and key clinical outcomes. An ipilimumab dose of 10 mg/kg produced a greater Cminss with numerically higher tumor responses and an increased probability of improved OS, although these outcomes were associated with a greater likelihood of developing an irAE. Given the association of Cminss with both efficacy and safety measures, individualized dosing might be an effective approach for the ipilimumab treatment of patients with advanced melanoma in the future,
provided it is possible to determine a range of ipilimumab exposure that is optimal with respect to both efficacy and safety with data being collected in an ongoing phase III dose optimization trial (11). Both the 3 and 10 mg/kg doses of ipilimumab have been shown to improve OS in phase III clinical trials of patients with advanced melanoma (4, 5). While the current results provide evidence for the potential of better efficacy outcomes with greater ipilimumab exposure, further clinical studies of the risk-benefit profile are needed in order to determine the most effective dose of ipilimumab.

Acknowledgements

Editorial and writing assistance was provided by StemScientific, funded by Bristol-Myers Squibb Co.
References


Table 1. Parameter estimates from the logistic regression model for ipilimumab E-R relationships.

<table>
<thead>
<tr>
<th>E-R Model</th>
<th>Predictor</th>
<th>Odds Ratio Coefficient (95%CI)&lt;sup&gt;a&lt;/sup&gt;</th>
<th>p-value</th>
<th>Median Cminss (5th-95th Percentile) [μg/mL]</th>
<th>Odds Ratio: 5th Percentile :Median of Cminss (95%CI)</th>
<th>Odds Ratio: 95th Percentile :Median of Cminss (95%CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Log(Cminss)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>2.41 (1.66, 4.26)</td>
<td>&lt; 0.01</td>
<td>49.3 (1.75 - 110)</td>
<td>0.0531 (0.00742, 0.38)</td>
<td>2.03 (1.26, 3.26)</td>
</tr>
<tr>
<td>BOR</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Log(Cminss)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.76 (1.35, 2.54)</td>
<td>&lt; 0.001</td>
<td>47.7 (1.61 - 106)</td>
<td>0.146 (0.0551, 0.387)</td>
<td>1.58 (1.25, 1.99)</td>
</tr>
<tr>
<td>irRC</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td></td>
<td>Log(Cminss)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>1.91 (1.58, 2.38)</td>
<td>&lt; 0.001</td>
<td>43.9 (1.75, 103)</td>
<td>0.124 (0.0653, 0.237)</td>
<td>1.74 (1.46, 2.06)</td>
</tr>
<tr>
<td>irAEs</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<sup>a</sup>Cminss was log-transformed. Log(Cminss) increases by one unit for approximately 2.7-fold increase in Cminss.

<sup>b</sup>95% CI obtained from bootstrap (N=500).
Table 2. Parameter estimates from the CPH model for ipilimumab E-R relationships with OS.

<table>
<thead>
<tr>
<th>Predictor</th>
<th>Hazard Ratio Coefficient&lt;sup&gt;a&lt;/sup&gt; (95% CI)</th>
<th>Hazard Ratio P05: Median (95% CI)</th>
<th>Hazard Ratio P95: Median (95% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cminss [mg/mL]</td>
<td>0.990 (0.986, 0.994)</td>
<td>1.52 (1.29, 1.8)</td>
<td>0.552 (0.437, 0.697)</td>
</tr>
<tr>
<td>LDH [IU/L]&lt;sup&gt;b&lt;/sup&gt;</td>
<td>2.32 (1.91, 2.8)</td>
<td>0.684 (0.628, 0.745)</td>
<td>3.27</td>
</tr>
<tr>
<td>Comparator:</td>
<td>Hazard Ratio Coefficient&lt;sup&gt;c&lt;/sup&gt; (95% CI)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>ECOG PS &gt;0:0</td>
<td>1.72</td>
<td></td>
<td></td>
</tr>
<tr>
<td>(N=176:322)</td>
<td>1.72 (1.37, 2.15)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

<sup>a</sup>Hazard ratio coefficient represents the hazard ratio for one unit of change in the predictor variable.

<sup>b</sup>The hazard ratio coefficient for LDH corresponds to log transformed LDH (which was employed as the linear predictor, as the distribution of LDH is right-skewed).

<sup>c</sup>Hazard ratio coefficient represents the hazard ratio for comparator relative to reference predictor variable.

<sup>d</sup>P05: 5th percentile and P95: 95th percentile.
Figure Legends

Figure 1. Model-predicted probability of BOR by mWHO criteria (A) or ir-Clinical Activity by
the irRC (B) versus ipilimumab steady-state trough concentration. The solid line and shaded area
represent E-R model expected Pr(BOR) and 95% bootstrap confidence intervals (N=500). The
horizontal box plots represent the distributions of Cminss at each dose group as follows: boxes (25th,
50th, and 75th percentiles) and whiskers (5th and 95th percentiles). Open circles represent the observed
proportion of responders for each dose group, plotted at the median Cminss of each dose. The vertical
bars represent 90% prediction intervals of each observed proportion obtained from simulated trials.

Figure 2. Kaplan-Meier estimates of OS by Cminss tertiles (A) and by dose (B). In both (A) and
(B), 1T/2T/3T indicate tertiles of Cminss and numbers below the graphs represent the patients at risk
at 0, 6, 12, 18, 24, 30 and 36 months.

Figure 3: Model-predicted OS by baseline LDH, ECOG PS, and ipilimumab exposure (median
Cminss at 0.3, 3, and 10 mg/kg doses) in patients with advanced melanoma. Lines represent the
predicted survival by the Cox proportional-hazards model.

Figure 4. Model-predicted probability of grade ≥2 and grade ≥3 irAEs vs. ipilimumab steady-
state trough concentration. The solid line and dash line represent E-R model predicted probability of
grade ≥2 and grade ≥3 irAEs, respectively. The shaded areas represent 95% bootstrap confidence
intervals (N=500). The horizontal box plots represent the distributions of Cminss at each dose group
as follows: boxes (25th, 50th, and 75th percentiles) and whiskers (5th and 95th percentiles). Open
circles and open triangles represent the observed proportion of irAEs grade ≥2 and grade ≥3 for each
dose group, respectively. The vertical bars represent 90% prediction intervals of each observed
proportion obtained from simulated trials (N=500).
Figure 2A.

Survival Probability vs. Time [Months]

1T (0.6–26.2 mcg/mL)
2T (26.2–57.5 mcg/mL)
3T (57.5–155.3 mcg/mL)

Subjects at Risk

<table>
<thead>
<tr>
<th></th>
<th>1T</th>
<th>2T</th>
<th>3T</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>165</td>
<td>168</td>
<td>165</td>
</tr>
<tr>
<td>93</td>
<td>106</td>
<td>144</td>
<td></td>
</tr>
<tr>
<td>52</td>
<td>69</td>
<td>116</td>
<td></td>
</tr>
<tr>
<td>34</td>
<td>55</td>
<td>89</td>
<td></td>
</tr>
<tr>
<td>26</td>
<td>42</td>
<td>66</td>
<td></td>
</tr>
<tr>
<td>5</td>
<td>7</td>
<td>17</td>
<td></td>
</tr>
<tr>
<td>0</td>
<td>0</td>
<td>2</td>
<td></td>
</tr>
</tbody>
</table>
Clinical Cancer Research

Exposure-Response Relationships of the Efficacy and Safety of Ipilimumab in Patients with Advanced Melanoma


Clin Cancer Res Published OnlineFirst June 5, 2013.

Updated version
Access the most recent version of this article at:
doi:10.1158/1078-0432.CCR-12-3243

Supplementary Material
Access the most recent supplemental material at:
http://clincancerres.aacrjournals.org/content/suppl/2013/06/05/1078-0432.CCR-12-3243.DC1

Author Manuscript
Author manuscripts have been peer reviewed and accepted for publication but have not yet been edited.

E-mail alerts
Sign up to receive free email-alerts related to this article or journal.

Reprints and Subscriptions
To order reprints of this article or to subscribe to the journal, contact the AACR Publications Department at pubs@aacr.org.

Permissions
To request permission to re-use all or part of this article, contact the AACR Publications Department at permissions@aacr.org.