Clinical Cancer Research

Pembrolizumab Exposure–Response Assessments Challenged by Association of Cancer Cachexia and Catabolic Clearance

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Abstract

Purpose: To investigate the relationship of pembrolizumab pharmacokinetics (PK) and overall survival (OS) in patients with advanced melanoma and non–small cell lung cancer (NSCLC).

Patients and Methods: PK dependencies in OS were evaluated across three pembrolizumab studies of either 200 mg or 2 to 10 mg/kg every 3 weeks (Q3W). Kaplan–Meier plots of OS, stratified by dose, exposure, and baseline clearance (CL_0), were assessed per indication and study. A Cox proportional hazards model was implemented to explore imbalances of typical prognostic factors in high/low NSCLC CL_0 subgroups.

Results: A total of 1,453 subjects were included: 340 with pembrolizumab-treated melanoma, 804 with pembrolizumab-treated NSCLC, and 309 with docetaxel-treated NSCLC. OS was dose independent from 2 to 10 mg/kg for pembrolizumab-treated melanoma [HR = 0.98; 95% confidence interval (CI), 0.94–1.02] and NSCLC (HR = 0.98; 95% CI, 0.95–1.01); however, a strong CL_0–OS association was identified for both cancer types (unadjusted melanoma HR = 2.56; 95% CI, 1.72–3.80 and NSCLC HR = 2.64; 95% CI, 1.94–3.57). Decreased OS in subjects with higher pembrolizumab CL_0 paralleled disease severity markers associated with end-stage cancer anorexia-cachexia syndrome. Correction for baseline prognostic factors did not fully attenuate the CL_0–OS association (multivariate-adjusted CL_0 HR = 1.64; 95% CI, 1.06–2.52 for melanoma and HR = 1.88; 95% CI, 1.22–2.89 for NSCLC).

Conclusions: These data support the lack of dose or exposure dependency in pembrolizumab OS for melanoma and NSCLC between 2 and 10 mg/kg. An association of pembrolizumab CL_0 with OS potentially reflects catabolic activity as a marker of disease severity versus a direct PK-related impact of pembrolizumab on efficacy. Similar data from other trials suggest such patterns of exposure–response confounding may be a broader phenomenon generalizable to antineoplastic mAbs. *Clin Cancer Res; 24(23); 5841–9. ©2018 AACR.*

See related commentary by Coss et al., p. 5787

Introduction

Exposure–response (E–R) assessments in oncology have played an increasingly important role toward understanding the impact of dose selection on patient outcomes (1). To date, several notable E–R analyses in different cancer types have considered a range of clinical endpoints, including overall survival (OS), progression-free survival (PFS; refs. 2–6), and overall response rate (ORR)/tumor kinetics (7–16). The registration of pembrolizumab for the treatment of melanoma and non–small cell lung cancer (NSCLC) at the dose of 2 mg/kg administered once every 3 weeks (Q3W) was supported by dose–response analyses and model-based E–R analyses of longitudinal tumor size (10, 17) using techniques adapted from earlier work (7, 18).

Pembrolizumab (KEYTRUDA; Merck) is a humanized IgG4 mAb that directly binds programmed death 1 (PD-1) expressed on T cells, blocking the interaction between PD-1 and its ligands, PD-L1 and PD-L2. The PD-1 pathway represents a major immune checkpoint that tumor cells exploit to evade antitumor T-cell activity. When pembrolizumab binds PD-1, this restores antitumor immunity and T-cell activity against cancerous cells. The randomized comparisons of pembrolizumab doses of 2 and 10 mg/kg have corroborated all E–R simulations to date showing similarity in efficacy outcomes for pembrolizumab-treated melanoma and NSCLC across this 5-fold dose range. In melanoma, three randomized comparisons demonstrated similar efficacy for pembrolizumab at 2 versus 10 mg/kg Q3W regimens (11, 12). In a phase II/III study of advanced NSCLC, pembrolizumab doses of 2 and 10 mg/kg Q3W provided superior OS compared with docetaxel, with similar ORR and PFS outcomes at each dose (19).

Outside of pembrolizumab development, relatively few E–R analyses involve data from mAb dose-ranging studies. Therefore, the present pembrolizumab evaluations from two large randomized trials of pembrolizumab at 2 and 10 mg/kg Q3W have provided an important opportunity to gain insight into the

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Note: Supplementary data for this article are available at Clinical Cancer Research Online (http://clincancerres.aacrjournals.org/).

Clinical trial information: ClinicalTrials.gov: NCT01704287, NCT01905657, and NCT02142738.

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Translational Relevance

These retrospective analyses of data from two large randomized trials of pembrolizumab, KEYNOTE-002 in patients with melanoma and KEYNOTE-010 in patients with non–small cell lung cancer (NSCLC), demonstrate a >∼15-month overall survival (OS) advantage in patients with slower baseline catabolic clearance. This trend was confirmed prospectively with data from another large pivotal study of pembrolizumab in first-line NSCLC, KEYNOTE-024. To our knowledge, no other baseline factor provides this significant magnitude of OS differentiation. Of note, data from trials of other antineoplastic mAbs report similar patterns of exposure–response confounding, suggesting that this is a broader phenomenon that may be generalizable to this class of oncology biologics. These results from the largest set of OS analyses for pembrolizumab to date further highlight the potential importance of metabolic wasting disorders and survival in the immunotherapy setting.

challenges and potential complications for E–R of large molecules in oncology. To our knowledge, this is the largest analysis of OS to date for pembrolizumab-treated patients and the first published exposure–survival analysis of pembrolizumab.

Patients and Methods

Study design

The primary analyses include data from two large randomized trials, KEYNOTE-002 in patients with melanoma (ClinicalTrials.gov, NCT01704287; ref. 20) and KEYNOTE-010 in patients with NSCLC (ClinicalTrials.gov, NCT01905657; ref. 21). Additional data from the pivotal study of pembrolizumab in first-line NSCLC (KEYNOTE-024; ClinicalTrials.gov, NCT02142738) served as a separate validation cohort (22). The KEYNOTE-002 and KEYNOTE-010 studies were expected to provide a robust, well-balanced data set for investigation of pembrolizumab exposure–survival relationships for each indication.

All studies were conducted in accordance with Good Clinical Practice guidelines and the Declaration of Helsinki. All protocols and subsequent amendments were approved by the appropriate institutional review boards or ethics committees at each participating institution. All patients provided voluntary written informed consent. Brief details on study treatment, enrollment criteria, and design are included in Supplementary Methods. Additional complete details regarding the designs for each trial have been published (19, 22, 23).

Data analyses

Pharmacokinetics. Clearance (CL) is the typical variable derived in pharmacokinetic (PK) analyses to reflect the kinetics of the elimination process. It relates concentration of drug measured in the body to a dose or amount administered. In this report, CL is specifically defined as the volume of serum from which pembrolizumab is completely removed per unit time. Results from a recently developed, time-dependent pharmacokinetic (TDPK) model provided *post hoc* CL estimates for E–R assessments in this report (sensitivity analyses conducted to confirm results were independent of the choice of model structure/exposure metric). A population PK approach was applied to determine typical pembrolizumab PK parameter values as well as associations of covariates and parameter values. In this type of analysis, data from every individual are considered simultaneously in a unified model. Further details on PK sampling and brief background information on the methods of this approach are provided in Supplementary Materials. Full details of PK model methods are published separately (24).

Serum exposure, i.e., area under the serum concentration–time curve (AUC), was calculated using the first-dose CL as $Dose/CL_0$. As in prior E–R analyses, AUC was normalized to a 6-week interval [$AUC_{6weeks,CL0} = Dose/CL_0 \times (6/\text{dosing frequency in weeks})$] to provide an exposure value over an integer number of dosing intervals when considering every 2 weeks (Q2W) and Q3W.

OS analyses and patient covariates. Patient demographics and laboratory values were explored to account for survival variation using the Cox model methods described in Supplementary Methods. Eastern Cooperative Oncology Group (ECOG) status (0 vs. 1) and region (East Asian vs. not East Asian) were two of the stratification variables for the primary KEYNOTE-010 efficacy analyses and therefore considered for the NSCLC modeling. Other potential baseline variables of interest included baseline sum of the longest diameter of the target lesions (BSLD), lactate dehydrogenase (LDH), serum albumin (ALB), platelet count (PLT), age, gender, cancer stage, presence of EGFR-sensitizing mutation, and histology (squamous vs. nonsquamous). For melanoma, potential covariates also included a variable indicating tumor PD-L1 expression positivity (PD-L1) and presence/absence of BRAF mutation. For evaluating the possible connection of on-study cancer cachexia progression and OS, postbaseline rates of body weight change (WTRATE) and serum albumin change (ALBRATE) were incorporated into the analyses as time-dependent covariates. These variables reflect an instantaneous rate of change (% change/day) from baseline and thus normalize for potential differences in follow-up. Using the time-varying values of WTRATE and ALBRATE, the Cox model compares risk of an event across updated values at each event/time interval when patient measurements are taken, re-evaluating which risk group each person belongs based only on values occurring up to and not beyond the considered time interval.

Investigating alternate clinical factors explaining survival differences in patients with high and low catabolism melanoma and NSCLC. Separate Cox models for pembrolizumab-treated melanoma and NSCLC were implemented to determine if another factor or combination of factors unrelated to pembrolizumab exposure could explain a perceived survival gap between rapid and slow pembrolizumab elimination, i.e., CL_0 subgroups. Such a substitution, if possible, would facilitate an unbiased assessment of pembrolizumab exposure–related influence on patient outcome (as exposure is proportional to dose and inversely proportional to CL_0). Univariate OS models were established considering CL_0 alone. Final multivariate models were developed both with and without CL_0 considering all patient factors selected during model construction (Akaike information criterion used in selection). The ability of the multivariate models to account for the CL_0-associated survival gap was assessed by comparisons of the CL_0 HR with the univariate model and change in log-likelihood from adding CL_0 after entering other factors in each model.

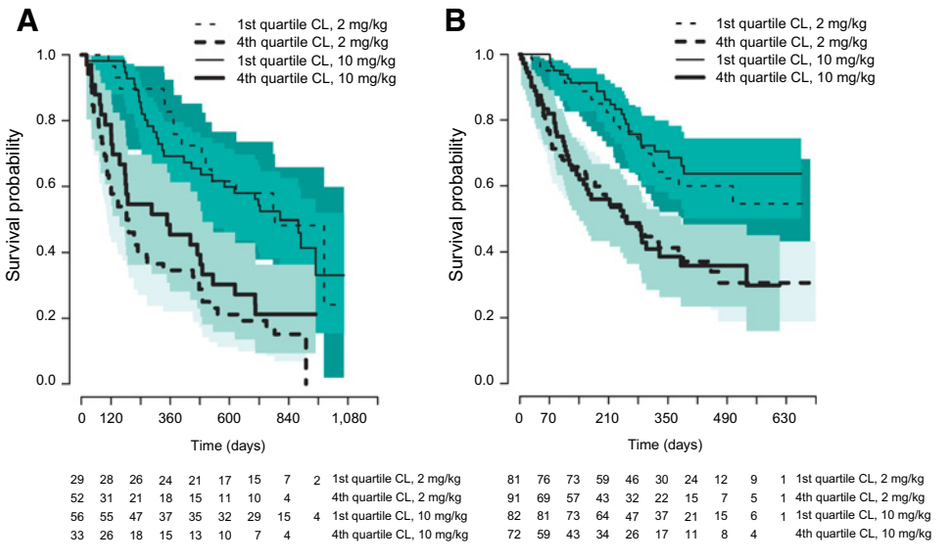

Figure 1.
Kaplan–Meier plots of OS from 2 and 10 mg/kg doses by within-dose baseline CL (CL_0) quartiles demonstrate a strong association of CL_0 and OS in both (**A**) intradose 1st and 4th quartiles in advanced ipilimumab-refractory melanoma (KEYNOTE-002) and (**B**) intradose 1st and 4th quartiles in previously treated PD-L1–positive NSCLC (KEYNOTE-010).

To explore alternative E–R methodology proposed in earlier publications, a matched, case–control E–R analysis was also performed using the NSCLC KEYNOTE-010 data. Similar to methods from the prior analyses (4), patients of each pembrolizumab dose arm and respective exposure quartiles were included in case groups. Control patients receiving docetaxel alone were matched 1:1 by the Mahalanobis metric method to the case groups based on AIC-selected risk factors described above for the multivariate Cox models (25). The relative pembrolizumab treatment effect versus chemotherapy for the case–control matched datasets was summarized by HR. The intent of this method was to explain some or all of the confounded CL_0–OS association by alternative factors to permit unbiased E–R assessments.

Results

Patient demographics

Details regarding study centers and investigators have been previously reported (19, 22, 23). Only pembrolizumab-treated subjects with at least one PK measurement were included ($n = 340$ for melanoma, $n = 652$ for NSCLC). The subsets of these patients with no missing covariates of interest were included in multivariate Cox regressions and case–control analyses (complete-case dataset $n = 211$ melanoma; $n = 537$ NSCLC). The source of analysis datasets, number of subjects, and baseline characteristics are detailed in Supplementary Tables S1 and S2.

Separate comparisons of OS across a 5-fold dose range in melanoma and NSCLC

Supplementary Fig. S1 shows the OS curves for the two dose groups in melanoma and NSCLC (based only on subjects with available PK data from KEYNOTE-002/-010). The overlap in the confidence intervals and Cox HRs reflects comparability of OS across a 5-fold dose and exposure range from 2 to 10 mg/kg in both tumor types [melanoma Cox HR = 0.98; 95% confidence interval (CI), 0.94–1.02 and NSCLC Cox HR = 0.98; 95% CI, 0.95–1.01].

Association of pembrolizumab AUC or CL_0 with OS in melanoma and NSCLC

OS stratified by exposure and dose is presented in Supplementary Figs. S2 and S3, and subjects in the 1st quartile of exposure within the 2 mg/kg arms of KEYNOTE-002/-010 have similar OS compared with the 1st quartile within the 10 mg/kg arms, despite a 5-fold dose and exposure range across these subgroups. However, subjects in the 4th quartile of exposure for 2 mg/kg demonstrate substantially better OS than the 1st quartile exposure of 10 mg/kg, despite having a lower exposure [4th quartile of exposure for 2 mg/kg (~2,000 μg/mL x day); 1st quartile exposure of 10 mg/kg (~4,000 μg/mL x day)]. This unusual pattern of improved survival in subjects with higher exposure within each dose is incongruent with the similarity in OS across the 5-fold dose/exposure range, suggesting a confounding of PK and OS independent of direct pharmacologic effects on patient outcome. Thus, E–R trends were further explored by comparing the relationships of OS and CL_0 both within and across 2 and 10 mg/kg (Fig. 1 focusing on the outer quartiles). These data reveal a considerable difference in median OS for pembrolizumab-treated NSCLC between subjects with rapid (4th quartile) and slow (1st quartile) CL_0, i.e., 8.4 months (95% CI, 6.4–11.0) versus >~23 months (lower 95% CI not reached in subjects with slow CL). Additionally, Supplementary Fig. S4 demonstrates 2nd and 3rd quartiles showing a pattern of graded response between the 1st and 4th quartiles. These same CL_0–OS trends are observed in melanoma (Fig. 1; Supplementary Fig. S4) and previously untreated (first-line) NSCLC (Supplementary Fig. S5; KEYNOTE-024, $n = 152$). Taken together, the dose–response analyses reinforce a lack of exposure-dependency in outcome, with CL_0–OS trends highlighting an underlying correlation of OS with pembrolizumab elimination.

Multivariate Cox proportional hazards regression analyses

Table 1 describes distributions of demographic and other baseline/on-study characteristics for subjects treated in KEYNOTE-002 and -010, stratified per CL_0 quartile within each indication; Fig. 2 shows results of the multivariate Cox analyses for melanoma and NSCLC. The multivariate Cox model in pembrolizumab-treated melanoma indicated BSLD, PD-L1, PLT, WTRATE, ALB, BRAF mutation status, and ECOG score were independently ($P < 0.05$) associated with OS in advanced melanoma (Fig. 2A). ALBRATE, ALB, WTRATE, baseline LDH, histology, gender, BSLD, and ECOG status were associated with OS in

Table 1. Demographics and clinical characteristics for pembrolizumab-treated subjects in complete-case modeling analysis datasets, stratified per baseline CL value

	KEYNOTE-002; advanced melanoma (complete-case dataset $n = 211$)				KEYNOTE-010; advanced, previously treated NSCLC (complete-case dataset $n = 537$)			
Characteristic	**1st quartile CL_0 ($n = 53$)**	**2nd quartile CL_0 ($n = 53$)**	**3rd quartile CL_0 ($n = 52$)**	**4th quartile CL_0 ($n = 53$)**	**1st quartile CL_0 ($n = 135$)**	**2nd quartile CL_0 ($n = 134$)**	**3rd quartile CL_0 ($n = 134$)**	**4th quartile CL_0 ($n = 134$)**
Weight change, week 9, %/day[a]								
Median	0.09%	0.09%	0.07%	−0.52%	0.14%	0.05%	−0.19%	−0.38%
Range	(−3.08–3.10)	(−3.57–2.34)	(−5.01–3.70)	(−5.32–6.90)	(−2.51–3.92)	(−3.29–3.13)	(−4.60–2.47)	(−4.49–3.84)
Albumin change, week 9, %/day[a]								
Median	−0.04%	−0.04%	−0.04%	−0.08%	−0.04%	−0.01%	−0.12%	−0.15%
Range	(−0.32–0.22)	(−0.42–0.33)	(−0.29–0.22)	(−0.74–0.61)	(−0.92–0.74)	(−1.41–0.66)	(−1.56–1.10)	(−2.52–1.90)
Age, year								
Median	61	62	67	57	64	63	62	62
Range	(23–85)	(23–84)	(24–89)	(27–78)	(31–82)	(38–86)	(29–84)	(20–88)
Gender								
Male	13 (25%)	32 (60%)	36 (69%)	40 (75%)	99 (73%)	56 (42%)	43 (32%)	23 (17%)
Female	40 (75%)	21 (40%)	16 (31%)	13 (25%)	36 (27%)	78 (58%)	91 (68%)	111 (83%)
Stage								
I	0 (0%)	0 (0%)	0 (0%)	0 (0%)	1 (<1%)	0 (0%)	0 (0%)	0 (0%)
II	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)
III	0 (0%)	1 (2%)	0 (0%)	1 (2%)	9 (7%)	13 (10%)	14 (10%)	6 (4%)
IV	53 (100%)	52 (98%)	52 (100%)	52 (98%)	125 (93%)	121 (90%)	120 (90%)	128 (96%)
ECOG								
0	35 (66%)	29 (55%)	30 (58%)	24 (45%)	60 (44%)	45 (34%)	54 (40%)	28 (21%)
1	18 (34%)	24 (45%)	22 (42%)	29 (55%)	75 (56%)	89 (66%)	80 (60%)	106 (79%)
Histology								
Squamous	N/A	N/A	N/A	N/A	12 (9%)	23 (17%)	27 (20%)	35 (26%)
Nonsquamous					123 (91%)	111 (83%)	107 (80%)	99 (74%)
EGFR status								
Mutant	N/A	N/A	N/A	N/A	19 (14%)	15 (11%)	11 (8%)	10 (7%)
Wild type					116 (86%)	119 (89%)	123 (92%)	124 (93%)
BRAF mutation								
Yes	16 (30%)	12 (23%)	16 (31%)	13 (25%)	N/A	N/A	N/A	N/A
No	37 (70%)	41 (77%)	36 (69%)	40 (75%)				
Region								
East Asia	0 (0%)	1 (2%)	1 (2%)	1 (2%)	51 (38%)	28 (21%)	27 (20%)	13 (10%)
Not East Asia	53 (100%)	52 (98%)	51 (98%)	52 (98%)	84 (62%)	106 (79%)	107 (80%)	121 (90%)
Albumin, g/L								
Median	41	41	38	37	42	41	39	36
Range	(32–49)	(27–49)	(25–47)	(19–79)	(29–50)	(29–52)	(26–48)	(19–48)
Platelet, billion/L								
Median	247	240	267	291	254	253	275	318
Range	(110–687)	(117–708)	(115–506)	(101–735)	(123–538)	(123–585)	(101–561)	(86–636)
PD-L1 expression								
Positive (>1%)	17 (32%)	17 (32%)	16 (31%)	18 (34%)	135 (100%)	134 (100%)	134 (100%)	134 (100%)
Negative (<1%)	36 (68%)	36 (68%)	36 (69%)	35 (66%)	0 (0%)	0 (0%)	0 (0%)	0 (0%)

Abbreviation: N/A, not applicable.

[a]Reflects instantaneous rate of on-study change determined at week 9 relative to baseline measure; actual Cox model implemented using time-varying values (WTRATE/ALBRATE).

pembrolizumab-treated NSCLC (Fig. 2B). Notably, the cachexia-related factor associated with change in body weight (WTRATE) was found to account for a portion of survival variability in both populations. Similar OS trends for rate of on-study weight change were also observed in docetaxel-treated NSCLC (Supplementary Fig. S6), suggesting a disease-level involvement of weight loss and OS. Generally, the 4th quartile of CL_0 in both melanoma and NSCLC corresponds to a point estimate for 9-week weight loss exceeding the consensus 5% cutoff commonly understood as the diagnostic criterion for cachexia (Table 1). This binary cutoff was initially considered in the Cox OS models, but ultimately it was not found to be significant as it did not afford the level of granularity that a continuous measure of weight loss provides. This is an oft-cited critique of dichotomizing continuous variables in regression analyses (26, 27).

Table 2 summarizes the relationship of CL_0 and OS for melanoma and NSCLC, both in the unadjusted, univariate Cox models and the multivariate models, where the relative risk is adjusted for potential confounders such as known clinical risk factors and other derived variables which serve as a proxy for on-study progression of cancer cachexia (WTRATE/ALBRATE; defined above). Overall, the univariate, unadjusted CL_0 HR for KEYNOTE-010 was estimated to be 2.64 (95% CI, 1.94–3.57, $P \leq 0.001$), representing the incremental risk of death per one unit increase of log-transformed CL. After entering the other baseline patient factors and on-study cachexia-related factors, WTRATE and ALBRATE, the adjusted point estimate of CL_0 HR was 1.53, but the 95% CI overlapped unity (0.97–2.41). The version of this model only adjusting for risk factors known at the start of treatment, i.e., excluding WTRATE/ALBRATE factors, showed CL_0

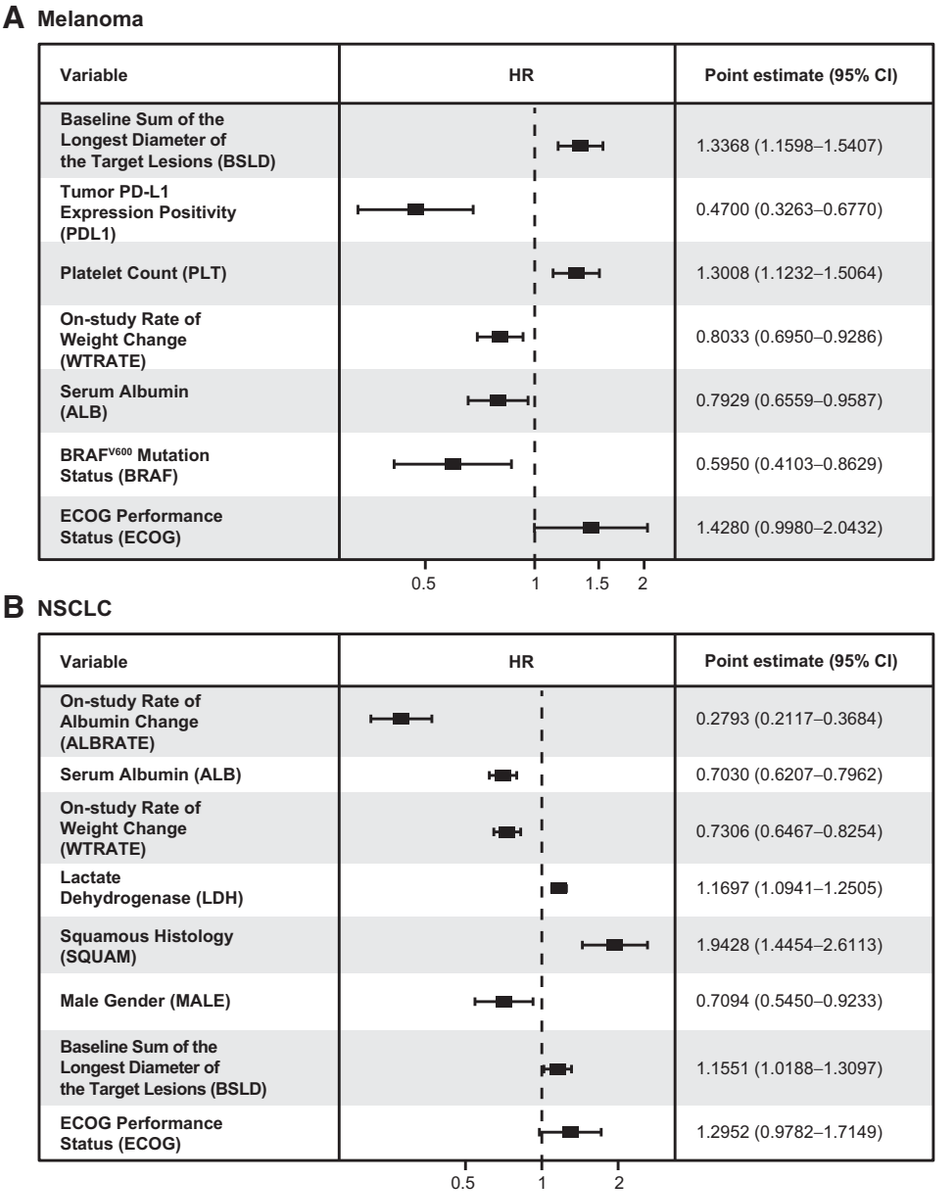

Figure 2.
Forest plot of pembrolizumab-treated melanoma Cox proportional hazards model–estimated OS HRs (**A**, $n = 211$) and pembrolizumab-treated NSCLC Cox proportional hazards model–estimated OS HRs (**B**, $n = 537$). HR estimates on continuous variables indicate the incremental risk per one SD unit increase of covariate. Continuous variables with HR larger than dashed unity line (HR >1) represent factors associated with additional risk of death per increasing SD unit value, whereas those with HR from 0 to 1 represent decreasing risk with increasing values (e.g., ALB). ALB, serum albumin level; ALBRATE, postbaseline rates of serum albumin change; ASIAN, region (East Asian = 1, non–East Asian = 0); SQUAM, histology (nonsquamous = 0, squamous = 1); WTRATE, postbaseline rates of body weight change.

HR to be 1.88 (95% CI, 1.22–2.89). The resultant $-2 \times$ log likelihood increased by 4.9 units in the version of this baseline-risk-factor model without CL_0, confirming an inability to fully explain a prominent CL_0–OS association without adjusting for the postbaseline cachectic markers, again despite the presence of 2 to 10 mg/kg dose–OS similarity. Similarly, the presence of a hidden E–R confounder is also demonstrated in the pembrolizumab-treated melanoma population (KEYNOTE-002; Table 2), where the univariate (unadjusted model) CL_0 HR was 2.56 (95% CI, 1.72–3.80), and the multivariate-adjusted (Fig. 2) CL_0 HR was found to be 1.60 (95% CI, 1.04–2.47), and in the baseline-factor-only version of this same multivariate model (without WTRATE), the CL_0 HR was 1.64 (95% CI, 1.06–2.52).

Figure 3, illustrating NSCLC case–control analyses, further substantiates these findings. The trends for slope of HR versus exposure appear markedly steeper within-dose than the overall across-dose exposure comparisons, including the case–control analyses in which pembrolizumab exposure subgroups and the docetaxel control arm were matched on similar AIC-selected risk factors (methods as described above). E–R analyses using early/late exposure metrics from the TDPK model or from the previously described, simpler, static, two-compartment model yield similar conclusions (Supplementary Table S3).

Discussion

This report describes the first E–R assessment of survival for the PD-1–targeted mAb, pembrolizumab. Consistent with prior dose–response assessments and E–R results showing a flat relationship by tumor size response (19, 23, 28), similar survival outcomes were observed here at both 2 and 10 mg/kg. A lack of clinically relevant exposure-dependency in OS is demonstrated

Table 2. Univariate/multivariate Cox regression model results with associated relative risk (HR) of one unit increase of $AUC_{6weeks,CL0}$/log-transformed CL_0 in melanoma (KEYNOTE-002) and NSCLC (KEYNOTE-010)

Model	KEYNOTE-002; advanced melanoma (complete-case dataset $n = 211$)			KEYNOTE-010; advanced, previously treated NSCLC (complete-case dataset $n = 537$)		
Univariate pooled $AUC_{6weeks,CL0}$ HR across 2 and 10 mg/kg	**0.71 (0.59–0.87)**			**0.77 (0.67–0.88)**		
	CL_0 HR (95% CI for HR)	**CL_0 *P* value**	**Δ*OBJV* with CL_0 included[a]**	**CL_0 HR (95% CI for HR)**	**CL_0 *P* value**	**Δ*OBJV* with CL_0 included[a]**
Unadjusted univariate or "crude" Cox model	2.56 (1.72–3.80)	<0.001	−20.67	2.64 (1.94–3.57)	<0.001	−33.21
Adjusted for time-varying on-study proxy factors of cancer cachexia and baseline clinical risk factors[b]	1.60 (1.04–2.47)	0.031	−4.49	1.53 (0.97–2.41)	0.068	−3.18
Adjusted for baseline clinical risk factors only[c]	1.64 (1.06–2.52)	0.025	−4.89	1.88 (1.22–2.89)	0.004	−7.58

[a]ΔOBJV: change in objective function value contrasting models before and after inclusion of CL_0. Significant association of CL_0 and OS indicated by reduction in OBJV of $\geq$3.84 ($P < 0.05$, based on the $\chi 2$ test for the difference in the −2 log-likelihood between two hierarchical models that differ by 1 degree of freedom).
[b]Feature selection conducted using forward selection with AIC penalty to avoid overparameterization/maintain model parsimony. NSCLC Cox regression model adjusted for baseline albumin (ALB), LDH, histology, gender, BSLD, ECOG, status, and on-study time-varying rate of weight change (WTRATE) and rate of albumin change (ALBRATE). Melanoma Cox regression model adjusted for BSLD, PD-L1 expression positivity, PLT, ALB, BRAF mutation status, ECOG, and time-dependent WTRATE.
[c]CL_0 HR adjusted for baseline factors, as in row above, but excluding the time-varying on-study factors related to cachexia (WTRATE and ALBRATE).

both by exploratory Kaplan–Meier plots, stratified per AUC quartiles, and in the Cox model–derived HRs (Fig. 3), reflecting similar values comparing the same quartile of AUC for 2 versus 10 mg/kg (~5-fold exposure differences). Apparent within-dose differences in response are therefore driven by a relationship between CL and survival rather than reflecting a true causal exposure effect on survival. These results confirm 2 mg/kg (or a comparable fixed dose) as appropriate for treatment of melanoma and NSCLC, providing near-maximal efficacy such that dosing 5-fold higher does not meaningfully increase OS (29, 30). Of note, recent pembrolizumab trials in NSCLC and melanoma have begun to evaluate the 200 mg Q3W regimen in lieu of weight-based dosing, based on doses of 200 mg and 2 mg/kg, providing similar exposure distributions with no advantage to either dosing approach with respect to controlling PK variability (30).

It is noteworthy that despite the confirmation of flat dose/E–R relationship, the analyses described here also reveal a prominent association of pembrolizumab CL_0 at baseline and OS, whereby subjects with slower CL_0 have a more than doubled life expectancy. Pembrolizumab CL_0 combined with dosing regimen determines exposure (e.g., AUC = Dose/CL); thus, the overall lack of influence of pembrolizumab dose and considerable within-dose CL_0/exposure–OS trends signify latent confounding between pembrolizumab elimination and disease status. Pembrolizumab has demonstrated target-mediated drug disposition (TMDD) at doses up to 0.1 mg/kg—a 20-fold lower dose compared with 2 mg/kg. Therefore, the impact of TMDD on total pembrolizumab CL is considered negligible at clinically relevant doses and is not thought to be a driver of the CL_0–OS associations observed here. A more plausible hypothesis for this relationship is due to an altered catabolic state among subjects with end-stage disease, independent of any causal relationship of exposure and OS.

Cancer anorexia-cachexia, with an estimated prevalence of 15% to 40% in the general cancer population (31), is known to portend poorer prognosis in advanced malignancy (32, 33). Affected individuals can suffer dramatic loss of body weight and muscle strength (34–36), and those same catabolic drivers accompanying skeletal muscle loss also constitute a primary elimination pathway of the humanized IgG4/κ mAb, pembrolizumab (and similar biologics). In recently published findings, Flint and colleagues reported patients receiving immunotherapy may be particularly susceptible to cancer-associated cachexia (37). Their results revealed that cancer cachexia, triggered by tumor-induced changes in liver metabolism, produces an upregulation of stress hormone production which can ultimately lead to systemic immunosuppression and a loss of immunotherapy efficacy in some patients. A correlation of rapid mAb catabolism (the mechanism of pembrolizumab CL) with poorer OS is demonstrated in the present analyses. Moreover, numerous recent analyses suggest these patterns may not be unique to pembrolizumab, but rather, a broader phenomenon generalizable to other antineoplastic mAbs with similar catabolic CL mechanisms. For example, the initial 2010 biologic license application for the IgG1 anti–CTLA-4 mAb, ipilimumab, contained E–R results describing a pronounced exposure–OS relationship in melanoma (38). This prompted a postmarketing commitment for a large comparative trial, prospectively evaluating OS with randomized doses of 3 and 10 mg/kg (clinicaltrials.gov: NCT01515189); yet, a recent integrated analysis of ipilimumab phase II and III data showed little OS difference across this dose range (39). The early trastuzumab (anti-HER2/neu IgG1 mAb) population PK and E–R analyses similarly suggested a strong PK (C_{min}) association with OS that was later acknowledged as capturing, at least partially, imbalances in general disease risk factors (4, 40). More recent examples of this same phenomenon have been described for nivolumab (anti–PD-1 IgG4 mAb) and atezolizumab (anti–PD-L1 IgG1 mAb; ref. 41). These repeated findings across biologics involving differing therapeutic targets imply a likely common source of confounding which has been postulated in other specific instances to involve a correlation of cachexia and increased mAb catabolism secondary to generalized protein turnover. (42, 43) Their shared metabolic pathways and the well-established link of anorexia/cachexia-related metabolic wasting and patient outcome thus present a credible explanation that should be further explored.

To further investigate the CL_0–OS association and determine which clinical factors could possibly be linked to hypercatabolism and contribute to PK–OS convolution, a multivariate Cox regression model was explored. Similar to techniques implemented by the FDA in a prior case–control E–R assessment for trastuzumab, this survival model sought a potential replacement for CL_0 as a correlate of survival among various relevant prognostic factors (4). Although these analyses demonstrated that some survival variation linked to pembrolizumab CL_0 can alternately be ascribed to other risk factors, some

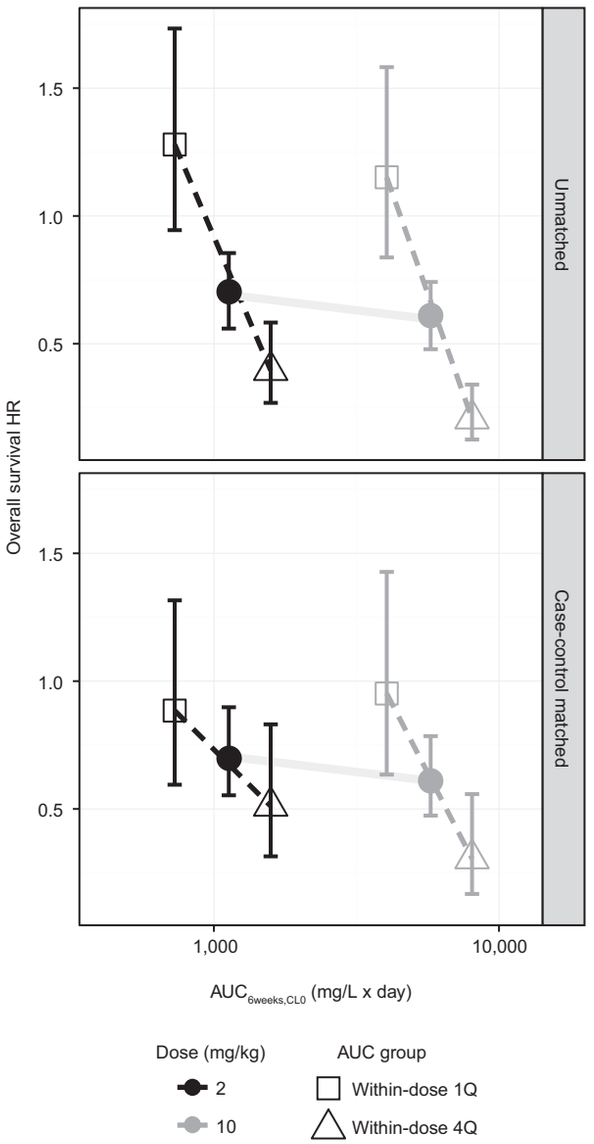

Figure 3.

KEYNOTE-010 univariate pembrolizumab treatment HR versus exposure shows stark inconsistency between E–R across dose (slope of solid gray line) and within-dose E–R (slope of dashed lines) both without (top) and with (bottom) matched case–control corrections, confirming that an imbalance in readily available prognostic factors cannot account entirely for the survival variability linked to CL; hence, case–control methodology is inadequate for E–R deconvolution. Illustrated are the HR and 95% CI; *x*-axis positioning reflects median exposure per group, and dashed lines connect within-dose 1st (open square) and 4th (open triangle) quartiles (Q) of exposure for 2 (black) and 10 mg/kg (gray).

correlation remained unexplained. The change in log-likelihood from the multivariate survival model of KEYNOTE-010 with and without CL_0 further confirms this finding. This highlights a recognized limitation of the E–R case–control approach, i.e., that one cannot discern whether any remaining unexplained CL_0–OS association is attributable to other hidden confounders or to a true E–R relationship (4, 44, 45). This investigation suggests that simply matching case controls on standard baseline disease factors may be inadequate to distinguish on-treatment cause and effect and delineate the influence of biologic drug exposure in this causal sequence. One could speculate that the association between pembrolizumab CL_0 and survival is difficult to displace by traditional risk factors such as ECOG, cancer stage, etc. because the measurement of CL_0 here provides a more precise estimate of catabolic rate, reflecting overall health and degree of cachexia.

In summary, a lack of clinically relevant exposure-dependency in OS with pembrolizumab across the dose range of 2 to 10 mg/kg was demonstrated for both melanoma and NSCLC. Consistent with prior randomized comparisons, these data support that increasing exposures above those attained at 2 mg/kg do not meaningfully improve response. In addition, we have shown rapid CL was strongly linked to decreased OS likely due to it being a proxy of disease severity and overall patient health. Given that the confounded association of longitudinal disease burden and PK has been observed across a class of oncology therapies, caution is warranted in interpreting E–R relationships, especially in the context of oncology trials evaluating a single dose level of biologic/mAb. Though challenging, randomized dose-ranging studies appear to be the only viable approach for decoupling PK of mAb and other latent confounders to delineate the role of exposure, and thus the impact of dose selection, on patient outcome. This ultimately underscores a broader need for better predictive clinical biomarkers of cachexia to understand the role of confounders of CL and survival in patients with cancer (46). Validation of a clear and standardized biomarker capturing the spectrum of cachexia in cancer could significantly aid in disease staging, refining trial inclusion/exclusion criteria, and, as evidenced here, E–R deconvolution.

Disclosure of Potential Conflicts of Interest

A.G. Robinson reports receiving speakers bureau honoraria from Merck & Co., Inc., and is a consultant/advisory board member for AstraZeneca, Bristol-Myers Squibb, and Merck & Co., Inc. E.B. Garon reports receiving other commercial research support from AstraZeneca, Bristol-Myers Squibb, and Merck & Co., Inc. S.W. Ebbinghaus and J.A. Stone hold ownership interest (including patents) in Merck & Co., Inc. No potential conflicts of interest were disclosed by the other authors.

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