RESEARCH ARTICLE

Bempegaldesleukin (NKTR-214) plus Nivolumab in Patients with Advanced Solid Tumors: Phase I Dose-Escalation Study of Safety, Efficacy, and Immune Activation (PIVOT-02)

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ABSTRACT

This single-arm, phase I dose-escalation trial (NCT02983045) evaluated bempegaldesleukin (NKTR-214/BEMPEG), a CD122-preferential IL2 pathway agonist, plus nivolumab in 38 patients with selected immunotherapy-naïve advanced solid tumors (melanoma, renal cell carcinoma, and non–small cell lung cancer). Three dose-limiting toxicities were reported in 2 of 17 patients during dose escalation [hypotension ($n = 1$), hyperglycemia ($n = 1$), metabolic acidosis ($n = 1$)]. The most common treatment-related adverse events (TRAE) were flu-like symptoms (86.8%), rash (78.9%), fatigue (73.7%), and pruritus (52.6%). Eight patients (21.1%) experienced grade 3/4 TRAEs; there were no treatment-related deaths. Total objective response rate across tumor types and dose cohorts was 59.5% (22/37), with 7 complete responses (18.9%). Cellular and gene expression analysis of longitudinal tumor biopsies revealed increased infiltration, activation, and cytotoxicity of $CD8^+$ T cells, without regulatory T-cell enhancement. At the recommended phase II dose, BEMPEG 0.006 mg/kg plus nivolumab 360 mg every 3 weeks, the combination was well tolerated and demonstrated encouraging clinical activity irrespective of baseline PD-L1 status.

SIGNIFICANCE: These data show that BEMPEG can be successfully combined with a checkpoint inhibitor as dual immunotherapy for a range of advanced solid tumors. Efficacy was observed regardless of baseline PD-L1 status and baseline levels of tumor-infiltrating lymphocytes, suggesting therapeutic potential for patients with poor prognostic risk factors for response to PD-1/PD-L1 blockade.

See related commentary by Rouanne et al., p. 1097.

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INTRODUCTION

Immunotherapy, especially checkpoint inhibition, has emerged as an effective treatment option for a range of solid tumors (1–3). Checkpoint inhibitors (CPI), such as nivolumab, are designed to harness the body's immune system to help restore antitumor immune responses. However, only a subset of patients experience deep and durable responses with CPI, highlighting a need for novel treatment approaches (4). Combinations of CPI, such as nivolumab plus ipilimumab, have incrementally improved outcomes for the treatment of melanoma, but durable responses in most patients are still limited, and the combination is associated with a greater incidence of adverse events (AE; refs. 5, 6). As low tumor PD-L1 expression, low levels of baseline tumor-infiltrating lymphocytes (TIL), and absence of a T-cell inflamed tumor microenvironment can be associated with a poor response to CPI (7–10), novel therapeutic approaches that stimulate T cells or overcome T-cell exhaustion may complement or synergize with checkpoint inhibition to achieve durable responses for more patients.

High-dose IL2 therapy, such as with aldesleukin, confers durable clinical benefit in select patients, partly through lymphoid expansion (11, 12). However, it is associated with severe toxicity necessitating in-patient administration at specialist centers, thereby limiting its use (13). Bempegaldesleukin (BEMPEG or NKTR-214) is a CD122-preferential IL2 pathway agonist conjugated with multiple releasable chains of polyethylene glycol and is designed to provide sustained signaling through the heterodimeric IL2βγ (CD122/132) receptor pathway (IL2βγR), with limited binding to the IL2αR subunit. Preferential IL2 pathway signaling through the IL2βγR drives proliferation and activation of $CD8^+$ T and natural killer (NK) cells without unwanted expansion of T regulatory cells (Treg) in the tumor microenvironment (14–17).

BEMPEG monotherapy has been shown to induce proliferation and activation of $CD4^+$ and $CD8^+$ T cells and NK cells in the blood and tumor microenvironment, including increased expression of PD-1 on these cells, in patients with advanced solid tumors (17). These immunologic changes do not appear to wane after repeated cycles of BEMPEG administration. BEMPEG monotherapy was well tolerated at the recommended phase II dose (RP2D) of 0.006 mg/kg every 3 weeks. On the basis of the biological activity, tolerability, and nonoverlapping toxicities with approved CPI, we hypothesized that the combination of BEMPEG and nivolumab could result in increased tumor response rates compared

with those achieved with CPI monotherapy. This phase I dose-escalation study was initiated to evaluate the safety, dose, and activity of BEMPEG administered in combination with nivolumab.

RESULTS

Patient Disposition and Baseline Characteristics

Between December 19, 2016, and August 21, 2017, 38 patients were enrolled at five centers in the United States across the following tumor cohorts: first-line (1L) melanoma (n = 11), 1L renal cell carcinoma (RCC; n = 14), second-line (2L) immunotherapy (I-O)-naïve RCC (n = 8), and I-O-naïve non–small cell lung cancer (NSCLC; n = 5). Patient baseline characteristics are shown in Table 1.

As of the January 18, 2019, data cutoff, all 38 patients had received study treatment and were included in the safety population. One patient (2L I-O-naïve RCC receiving BEMPEG 0.006 mg/kg every 3 weeks; nivolumab 360 mg every 3 weeks) died from progressive disease (PD) on study at day 20 before the first postbaseline scan and therefore was excluded from the efficacy analysis (n = 37 in the efficacy evaluable population). Patient disposition is shown in Supplementary Fig. S1. Median duration of follow-up was 18.0 months [interquartile range (IQR), 14.8–20.2]: 1L melanoma 18.5 months (IQR, 14.8–21.7), 1L RCC 17.2 months (IQR, 16.0–18.3), 2L I-O-naïve RCC 18.5 months (IQR, 12.1–21.7), and I-O-naïve NSCLC 18.0 months (IQR, 11.5–19.0).

The median duration of exposure to either study drug was 13.3 months (IQR, 3.7–16.9; Supplementary Table S1). Patients received a median of 15 cycles (IQR, 6.0–22.0) of BEMPEG plus nivolumab across all dose cohorts. Thirty of 38 patients (79%) received six or more cycles of study treatment. At the time of data cutoff, 9 of 38 patients (23.7%) were still receiving study treatment [six partial responses (PR); three complete responses (CR); range of treatment duration, 18–22 months). Reasons for discontinuation in 29 patients were: PD (14 patients, 36.8%), AEs (7 patients, 18.4%), clinical progression (3 patients, 7.9%), achievement of maximal response (2 patients, 5.3%), patient decision (2 patients, 5.3%), and physician decision (1 patient, 2.6%). Eight patients died while on study, all from disease progression.

Safety and Tolerability

Five treatment schedules were explored during the dose escalation in a total of 17 patients: (i) BEMPEG 0.006 mg/kg every 3 weeks plus nivolumab 240 mg every 2 weeks (n = 4); (ii) BEMPEG 0.003 mg/kg every 2 weeks plus nivolumab 240 mg every 2 weeks (n = 3); (iii) BEMPEG 0.006 mg/kg every 2 weeks plus nivolumab 240 mg every 2 weeks (n = 3); (iv) BEMPEG 0.006 mg/kg every 3 weeks plus nivolumab 360 mg every 3 weeks (n = 4); and (v) BEMPEG 0.009 mg/kg every 3 weeks plus nivolumab 360 mg every 3 weeks (n = 3). No dose-limiting toxicities (DLT) were observed in dose cohorts 1–4. The maximum tolerated dose (MTD) was exceeded at BEMPEG 0.009 mg/kg every 3 weeks plus nivolumab 360 mg every 3 weeks, with 2 of 3 patients experiencing a DLT [grade 3 hypotension (n = 1); grade 4 hyperglycemia and metabolic acidosis (n = 1)]. All DLT events resolved within 5 days, and the 2 patients continued on treatment with a lower dose of BEMPEG combined with nivolumab. The MTD of the combination was defined as BEMPEG 0.006 mg/kg every 3 weeks plus nivolumab 360 mg every 3 weeks, and this dose was selected as the RP2D. In total, 25 patients received BEMPEG plus nivolumab at the RP2D. Hypotension management guidelines, including hydration instructions (Supplementary Materials and Methods: Hydration Guidelines) were implemented at this stage to mitigate the potential development of hypotension associated with BEMPEG administration.

All 38 patients enrolled in the study experienced a treatment emergent adverse event (TEAE; Supplementary Table S2), and all patients had TEAEs that were considered related to the study combination [treatment-related adverse event (TRAE); Table 2]. The most common TRAEs (occurring in ≥40% of patients) were flu-like symptoms (86.8%), rash (78.9%), fatigue (73.7%), pruritus (52.6%), arthralgia (47.4%), decreased appetite (42.1%), and headache (42.1%). Grade ≥3 TRAEs occurred in 8 patients (21.1%; Table 2), with the majority being grade 3. One patient experienced grade 4 hyperglycemia (BEMPEG 0.006 mg/kg every 3 weeks/nivolumab 360 mg every 3 weeks) and 1 patient experienced grade 4 hyperglycemia and grade 4 acidosis (BEMPEG 0.009 mg/kg every 3 weeks plus nivolumab 360 mg every 3 weeks). Generally, grade ≥3 TRAEs were manageable using standard guidelines. Immune-mediated adverse events (imAE) related to the study drug were observed in 12 of 38 patients (31.6%): hypothyroidism in 11 patients, hyperthyroidism in 2 patients, hyperglycemia in 2 patients, and skin adverse reaction and colitis/diarrhea in 1 patient each (some patients may have experienced more than one imAE). Elevated eosinophil counts, which are a marker of IL2 activity (18), were observed in most patients and were more marked with extended duration of therapy (data not shown). Cytokine-related symptoms (such as flu-like symptoms, rash, pruritus, or hypotension) were observed primarily in cycles 1 and 2 and became significantly reduced with additional cycles (Supplementary Fig. S2). There were no treatment-related deaths.

Antitumor Activity

The investigator-assessed objective response rate (ORR) was 22 of 37 [59.5%; 95% confidence interval (CI), 42.1%–75.2%], and the disease control rate (DCR) was 31 of 37 (83.8%; Table 3). Among the 22 patients with confirmed objective responses, median time to treatment response (TTR) was 1.9 months (range 1.3–7.8) and median duration of response (DOR) was not reached. Among the 24 patients eligible for the efficacy analysis who received the RP2D of BEMPEG plus nivolumab, the ORR was 66.7% (95% CI, 44.7%–84.4%; 16/24), DCR was 83.3% (20/24), and patients had a median 67.6% reduction in target lesions from baseline.

For the 1L melanoma cohort (n = 11), the investigator-assessed ORR was 63.6%, with four CRs (36.4%) and three (27.3%) PRs. The median DOR was not reached. Of note, there were 4 patients with liver metastasis at baseline, of which 2 (50.0%) had a CR and 1 (25.0%) had a PR. There were 8 patients with high levels of lactate dehydrogenase (>1× the upper limit of normal) at baseline, of which 4 (50.0%) had a CR and 1 (12.5%) had a PR.

ORR, TTR, DOR, and median progression-free survival for all tumor cohorts are shown in Table 3. Best percentage

Table 1. Patient baseline characteristics

	1L melanoma (*N* = 11)	1L RCC (*N* = 14)	2L RCC (I-O naïve) (*N* = 8)	NSCLC (I-O naïve) (*N* = 5)
Age (years)				
Mean (standard deviation)	56.1 (13.85)	59.4 (6.93)	59.5 (7.62)	59.6 (7.64)
Median	62.0	59.5	61.0	58.0
Min, max	22, 70	48, 72	45, 70	53, 72
Sex				
Female	4 (36.4%)	1 (7.1%)	2 (25.0%)	1 (20.0%)
Male	7 (63.6%)	13 (92.9%)	6 (75.0%)	4 (80.0%)
ECOG performance status				
0	8 (72.7%)	10 (71.4%)	5 (62.5%)	2 (40.0%)
1	3 (27.3%)	3 (21.4%)	3 (37.5%)	3 (60.0%)
Unknown	0	1 (7.1%)	0	0
PD-L1 status				
Negative <1%	4 (36.4%)	8 (57.1%)	4 (50.0%)	3 (60.0%)
Positive ≥1%	7 (63.6%)	4 (28.6%)	4 (50.0%)	2 (40.0%)
Missing	0	2 (14.3%)	0	0
Brain metastases				
Yes	0	0	0	2 (40.0%)
No	11 (100%)	14 (100%)	8 (100%)	3 (60.0%)
Number of prior systemic therapies				
0	11 (100%)	14 (100%)	0	1 (20.0%)
1	0	0	8 (100%)	3 (60.0%)
2	0	0	0	1 (20.0%)
LDH baseline				
Normal	3 (27.3%)	6 (42.9%)	2 (25.0%)	1 (20.0%)
>1–<2 ULN	2 (18.2%)	2 (14.3%)	3 (37.5%)	0
≥2–<3 ULN	3 (27.3%)	6 (42.9%)	3 (37.5%)	3 (60.0%)
≥3 ULN	3 (27.3%)	0	0	1 (20.0%)
Stage (AJCC V7)				
M1a	0	N/A	N/A	N/A
M1b	1 (9.1%)	N/A	N/A	N/A
M1c	10 (90.9%)	N/A	N/A	N/A
BRAF mutation status				
V600E or V600K	7 (63.6%)	N/A	N/A	N/A
Negative	4 (36.4%)	N/A	N/A	N/A
Liver metastases at baseline				
Yes	4 (36.4%)	2 (14.3%)	2 (25.0%)	2 (40.0%)
No	7 (63.6%)	12 (85.7%)	6 (75.0%)	3 (60.0%)
Nephrectomy				
Yes	N/A	14 (100%)	7 (87.5%)	N/A
No	N/A	0	1 (12.5%)	N/A
IMD risk group				
Favorable	N/A	8 (57.1%)	1 (12.5%)	N/A
Intermediate	N/A	5 (35.7%)	6 (75.0%)	N/A
Poor	N/A	0	1 (12.5%)	N/A
Unknown	N/A	1 (7.1%)	0	N/A
Histology subtype				
Squamous	N/A	N/A	N/A	0
Adenocarcinoma	N/A	N/A	N/A	4 (80.0%)
Other	N/A	N/A	N/A	1 (20.0%)
Smoker				
Never	N/A	N/A	N/A	1 (20.0%)
Current	N/A	N/A	N/A	1 (20.0%)
Former	N/A	N/A	N/A	3 (60.0%)
EGFR mutation status				
Positive	N/A	N/A	N/A	0
Negative	N/A	N/A	N/A	5 (100.00%)
ALK translocation status				
Negative	N/A	N/A	N/A	5 (100.0%)

Abbreviations: AJCC, American Joint Committee on Cancer; ALK, anaplastic lymphoma kinase; ECOG, Eastern Cooperative Oncology Group; IMD, International Metastatic Renal Cell Carcinoma Database; LDH, lactate dehydrogenase; N/A, not applicable; ULN, upper limit of normal.

Table 2. TRAEs (grade ≥3, and all-grade in ≥10% of patients)

Preferred term[a]	All patients ($N = 38$)	Patients treated at the RP2D ($n = 25$)
Grade ≥3 (all patients)	8 (21.1)	4 (16.0)
Hyperglycemia	2 (5.3)	1 (4.0)
Lipase increased	2 (5.3)	1 (4.0)
Rash[b]	2 (5.3)	1 (4.0)
Acidosis	1 (2.6)	..
Arthralgia	1 (2.6)	..
Cerebrovascular accident	1 (2.6)	1 (4.0)
Colitis	1 (2.6)	..
Diarrhea	1 (2.6)	..
Hyperthyroidism	1 (2.6)	..
Hyponatremia	1 (2.6)	1 (4.0)
Hypotension	1 (2.6)	..
Infectious pleural effusion	1 (2.6)	1 (4.0)
Myalgia	1 (2.6)	..
Syncope	1 (2.6)	1 (4.0)
Vomiting	1 (2.6)	..
All grade (occurring in ≥10% of patients)	38 (100.0)	25 (100.0)
Flu-like symptoms[c]	33 (86.8)	20 (80.0)
Rash[b]	30 (78.9)	20 (80.0)
Fatigue	28 (73.7)	19 (76.0)
Pruritus	20 (52.6)	12 (48.0)
Arthralgia	18 (47.4)	11 (44.0)
Decreased appetite	16 (42.1)	9 (36.0)
Headache	16 (42.1)	10 (40.0)
Diarrhea	15 (39.5)	10 (40.0)
Nausea	15 (39.5)	10 (40.0)
Edema peripheral	14 (36.8)	9 (36.0)
Hypotension	12 (31.6)	7 (28.0)
Nasal congestion	12 (31.6)	8 (32.0)
Cough	11 (28.9)	7 (28.0)
Dry skin	11 (28.9)	6 (24.0)
Myalgia	11 (28.9)	8 (32.0)
Hypothyroidism	10 (26.3)	7 (28.0)
Peripheral sensory neuropathy	10 (26.3)	6 (24.0)
Vomiting	10 (26.3)	5 (20.0)
Face edema	8 (21.1)	4 (16.0)
Flushing	8 (21.1)	2 (8.0)
Dyspepsia	6 (15.8)	4 (16.0)
Malaise	6 (15.8)	3 (12.0)
Abdominal pain	5 (13.2)	3 (12.0)
Constipation	5 (13.2)	3 (12.0)
Dizziness	5 (13.2)	4 (16.0)
Dysgeusia	5 (13.2)	4 (16.0)
Infusion-related reaction	5 (13.2)	4 (16.0)
Musculoskeletal pain	5 (13.2)	3 (12.0)
Dry mouth	4 (10.5)	3 (12.0)
Dyspnoea	4 (10.5)	3 (12.0)
Stomatitis	4 (10.5)	2 (8.0)
Vision blurred	4 (10.5)	4 (16.0)
Weight decreased	4 (10.5)	3 (12.0)

Note: All data are *n* (%).

[a]Patients are counted only once under each preferred term.

[b]Includes the following preferred terms: erythema, rash, rash erythematous, rash generalized, rash macular, rash maculopapular, rash maculovesicular, rash papular, rash pruritic, rash pustular, rash vesicular, and exfoliative rash.

[c]Includes the following preferred terms: chills, influenza-like illness, and pyrexia.

Table 3. Overall response (investigator assessed)

	1L melanoma (*n* = 11)	1L RCC (*n* = 14)	2L RCC (I-O naïve) (*n* = 7)	NSCLC (I-O naïve) (*n* = 5)	Total (*N* = 37)
ORR[a], *n* (%)	7 (63.6)	10 (71.4)	2 (28.6)	3 (60.0)	22 (59.5)
CR	4 (36.4)	1 (7.1)	0	2 (40.0)	7 (18.9)
PR	3 (27.3)	9 (64.3)	2 (28.6)	1 (20.0)	15 (40.5)
SD	3 (27.3)	1 (7.1)	4 (57.1)	1 (20.0)	9 (24.3)
DCR[b]	10 (90.9)	11 (78.6)	6 (85.7)	4 (80.0)	31 (83.8)
PD	1 (9.1)	3 (21.4)	1 (14.3)	1 (20.0)	6 (16.2)
Median TTR, months (range)	1.7 (1.4–3.3)	3.5 (1.3–7.8)	2.7 (1.9–3.5)	1.7 (1.3–3.7)	1.9 (1.3–7.8)
Median DOR, months (95% CI)	NE (NE–NE)	NE (3.5–NE)	10.8 (NE–NE)	16.7 (NE–NE)	NE (13.0–NE)
Median PFS, months (95% CI)	NE (6.0–NE)	14.2 (1.8–NE)	14.3 (0.7–NE)	18.0 (1.7–NE)	14.8 (11.2–NE)
Median % reduction from baseline	−89.1	−67.6	−8.7	−56.4	−56.4
Ongoing responses[c]	7 (63.6%)	6 (42.9%)	1 (14.3%)	2 (40.0%)	16 (43.2%)
ORR[a] in PD-L1^{+}[d]	5/7 (71.4)	3/4 (75.0)	1/4 (25.0)	2/2 (100)	11/17 (64.7)
ORR[a] in PD-L1^{-}[d]	2/4 (50.0)	5/8 (62.5)	1/3 (33.3)	1/3 (33.0)	9/18 (50.0)
ORR[a] in PD-L1 unknown[d]	..	2/2 (100)	..	..	2/2 (100)

Note: All data *n* (%), unless otherwise stated. Patients with at least one postbaseline tumor assessment were evaluable for efficacy per RECIST 1.1.

[a]ORR is CR + PR.

[b]DCR is CR + PR + SD.

[c]Includes patients with CR or PR who had neither progressed nor died at the time of data cutoff.

[d]1L melanoma: 11 patients were evaluable for tumor PD-L1 expression (7 PD-L1^{+}, 4 PD-L1^{-}); 1L RCC: 12 of 14 patients were evaluable for tumor PD-L1 expression (4 PD-L1^{+}; 8 PD-L1^{-}); 2L RCC: 7 patients were evaluable for tumor PD-L1 expression (4 PD-L1^{+}, 3 PD-L1^{-}); 1–2 NSCLC: 5 patients were evaluable for tumor PD-L1 expression (2 PD-L1^{+}; 3 PD-L1^{-}).

Abbreviations: NE, not estimable; SD, stable disease.

change in target lesion by tumor cohort is shown in Fig. 1A. The observed responses with the BEMPEG and nivolumab combination continued to deepen over time with additional cycles of study treatment (Fig. 1B; Supplementary Fig. S3). Best overall response by tumor cohort, including timing of response and maximal percentage reduction in target lesions from baseline per patient, and according to baseline PD-L1 status, is shown in Table 3.

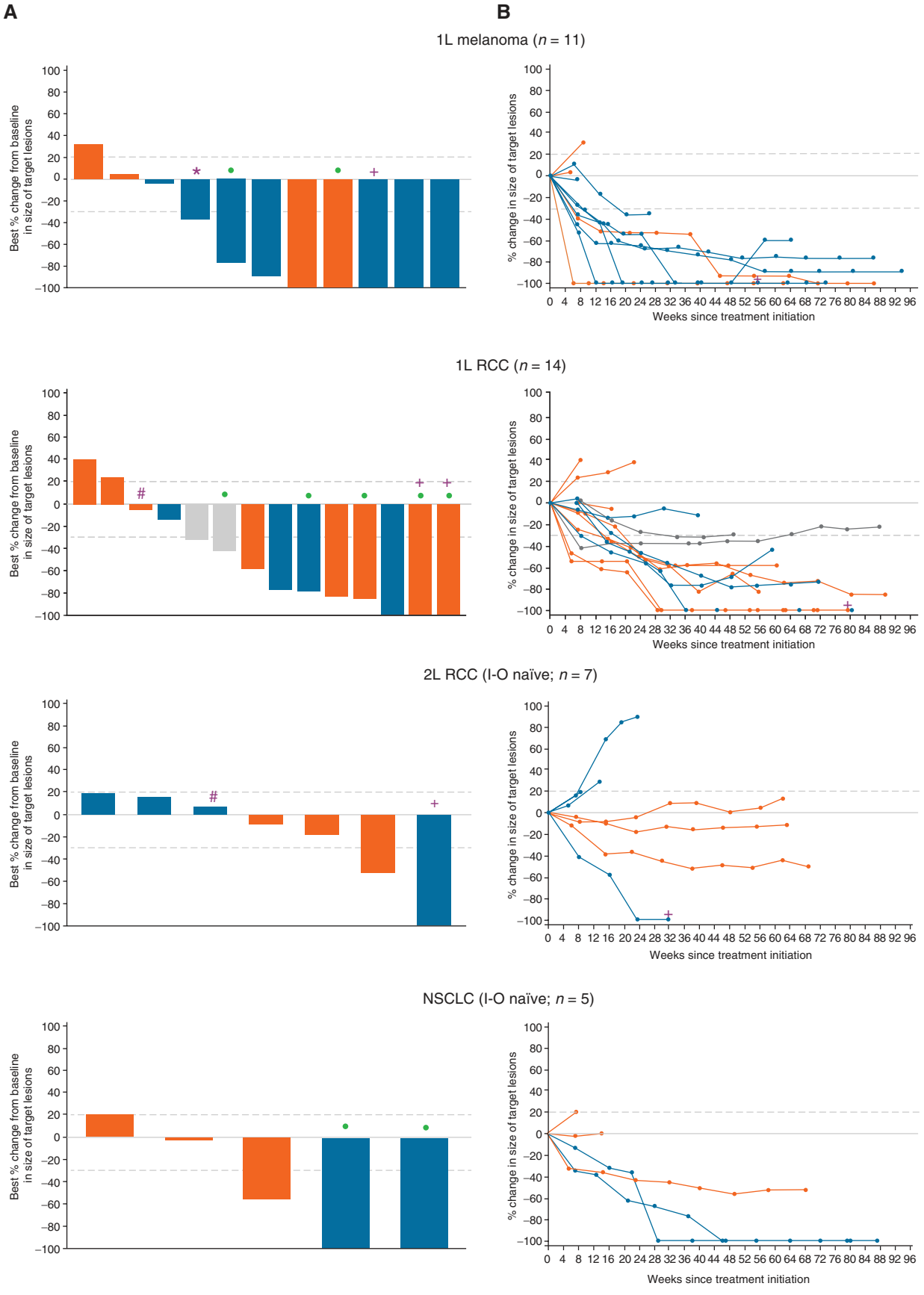
Tumor responses were observed regardless of baseline PD-L1 expression across all cohorts (Table 3; Fig. 1A). Eleven of 17 patients (64.7%) with positive baseline tumor PD-L1 expression (≥1% PD-L1 expression) and 9 of 18 patients (50.0%) with negative baseline PD-L1 expression (<1% PD-L1 expression) responded to therapy (Table 3). CD8^{+} T-cell analyses showed that 5 of 16 patients (31.3%) with lower CD8^{+} tumor infiltration (<150 cells/mm^{2} pretreatment) and 13 of 15 patients (86.7%) with higher CD8^{+} tumor infiltration (≥150 cells/mm^{2} pretreatment) responded to therapy (data not shown).

Investigator-assessed efficacy evaluations were consistent with those determined by independent central review (data not shown).

Immunologic Activity

Peripheral blood lymphocytes were mobilized at each dose level, with similar magnitude of mobilization in the BEMPEG 0.006 mg/kg and 0.009 mg/kg dose cohorts regardless of dosing schedule (i.e., every 2 weeks or every 3 weeks). Lymphocyte mobilization was observed with each treatment cycle (Fig. 2A), similar to the lymphocyte kinetics observed with single-agent BEMPEG in a separate phase I trial (Fig. 2B; ref. 17). Immune cell profiling confirmed statistically significant increases in the absolute numbers of CD4^{+}, CD8^{+}, and NK cells at time of maximal lymphocyte expansion (Fig. 2C), with 3.7-, 2.0-, and 4.4-fold average expansion, respectively, in matched C1D1 and C1D8 samples. Immunophenotypic characterization indicated a significantly higher median percentage of proliferating T and NK cells (40%–50% and 35%, respectively; Fig. 2D) at day 8 than day 1. BEMPEG plus nivolumab induced significantly higher expression of CTLA4 on proliferating (Ki67^{+}) compared with nonproliferating (Ki67^{-}) CD4^{+} T cells (Fig. 2E). The percentage of Tregs in the blood was significantly increased at day 8 of treatment compared with baseline (Fig. 2F). Peripheral blood neutrophil to lymphocyte ratio was consistent by cycle and did not appear to change according to best overall clinical response (Supplementary Fig. S4).

To assess immune activation, as well as cytotoxic and effector function in tumors, biopsies (baseline: 19; on treatment: nine) were analyzed for changes in gene expression across 760 genes. BEMPEG plus nivolumab treatment significantly ($P < 0.05$, unadjusted for multiplicity) changed expression levels in 218 genes, leading to upregulation in 197 genes and downregulation in 21 genes (Fig. 3A; Supplementary Table S3).



Upregulated genes included those associated with T-cell signaling (Fig. 3B), T-cell activation, and coinhibitory molecules, including *PDCD1*, the gene encoding PD-1 (Fig. 3C), and cytotoxic effector genes (Fig. 3D). Several immune NK cell genes were also significantly upregulated (Fig. 3E). Notably, $CD8^+$- and Th1-associated genes were increased after treatment (*CD8A, CD8B, EOMES, TBX21*, *IFNG*, and *CXCR3*; $P \leq 0.05$; Fig. 3F), whereas the majority of genes associated with inflammatory cellular pathways (Th2/Th17) remained unmeasurable (*IL17A, IL4*, and *GATA3*) or did not increase (*RORC*) in the on-treatment tumor biopsy (Fig. 3G). Treatment with BEMPEG plus nivolumab lead to upregulation of various cellular pathways (Fig. 3H), notably including those relating to regulation and T-cell functions (Supplementary Fig. S5A). Cell type analysis showed that the combination led to enhanced expression of genes associated with $CD45^+$ lymphocytes, $CD8^+$ T cells, macrophages, and cytotoxic cells, but not B cells or neutrophils (Supplementary Fig. S5B). Overall, our data show that BEMPEG plus nivolumab induced a gene signature reflecting cytotoxicity and effector functions, which is consistent with its mechanism of action.

Complementing the increase in activated T cells in the tumor and periphery, multiplex immunofluorescence demonstrated enhanced T-cell infiltration within the same lesion after one cycle of treatment. Pre- and on-treatment biopsies (in 1 patient each with melanoma and RCC) showed increases in $CD3^+$ T cells and $CD8^+$ T cells, colocalization of PD-L1 expression with T cells, and a concomitant decrease in malignant cells (Fig. 4A and B). No clear association between baseline PD-L1 expression, baseline $CD8^+$ T-cell level, and response was observed (Fig. 4C). IHC of 18 matched baseline and on-treatment tumor samples showed an increase in $CD8^+$ T cells and an increase in PD-L1 expression on treatment (Supplementary Fig. S6A and S6B). A trend was observed between the increase in $CD8^+$ infiltration and response using IHC, but these findings were not statistically significant (Supplementary Fig. S6C).

To determine whether BEMPEG plus nivolumab promoted a preferential increase in TILs, 12 matched pretreatment and on-treatment tumor biopsies were analyzed by flow cytometry for the presence of $CD8^+$ TILs and Tregs. $CD8^+$ TILs significantly increased on treatment (average 3.9-fold; Fig. 4D). Unlike Tregs in the periphery, which increased on treatment, Tregs in the tumor remained low (Fig. 4D). Without an increase in intratumoral Tregs, the $CD8^+$/Treg ratio increased significantly on treatment (average 4.2-fold; Fig. 4D).

T-cell receptor (TCR) repertoire sequencing of 12 pretreatment and on-treatment samples showed an increase in T-cell infiltration within the tumor with combination treatment (Fig. 4E). Despite the small sample size, these changes in the T-cell fraction were significant in patients who responded to combination treatment ($n = 5$; $P = 0.01$) but not in patients who did not ultimately respond ($n = 7$; Fig. 4E). Furthermore, there was a trend toward increased clonality in the tumor microenvironment in responders (CR or PR) versus nonresponders [stable disease (SD) or PD; Fig. 4E].

Recent attention has focused on the importance of tumor-associated B cells to sustain tumor-associated inflammation. This research also demonstrated how tumor-associated B cells can predict survival and response to immune CPI (19). We evaluated the effect of BEMPEG plus nivolumab on B cells in 14 matched tumor samples. Consistent with our other findings, BEMPEG plus nivolumab did not induce an increase of intratumoral B cells (in either responders or nonresponders), and B-cell percentages at baseline did not predict clinical response in this small sample size (Supplementary Fig. S7A and S7B).

DISCUSSION

This phase I dose-escalation study demonstrated that the CD122-preferential IL2 pathway agonist BEMPEG and nivolumab can be safely combined with a RP2D of BEMPEG 0.006 mg/kg every 3 weeks plus nivolumab 360 mg every 3 weeks in patients with selected advanced or metastatic solid tumors. The R2PD was one dose level below the maximum administered dose of BEMPEG 0.009 mg/kg every 3 weeks plus nivolumab 360 mg every 3 weeks and was selected on the basis of improved tolerability and equivalent immunologic activity.

AEs experienced with the combination of BEMPEG plus nivolumab were consistent with those reported with either BEMPEG (17) or nivolumab monotherapy (3, 5, 20–22). The most frequent TRAEs were grade 1 or 2 severity and presented as predictable and transient flu-like symptoms, rash, fatigue, or pruritus. Most TRAEs resolved spontaneously without intervention, or were mitigated by over-the-counter oral or topical treatments (e.g., acetaminophen and ibuprofen, antihistamines, or corticosteroids), similar to previous experiences with BEMPEG monotherapy (17). Cytokine-related symptoms (such as flu-like symptoms, rash, pruritus, or hypotension) were observed primarily in cycles one and two, and became significantly reduced with additional cycles. Of note, there was no grade ≥3 hypotension observed at the RP2D following implementation of hydration guidelines (Supplementary Materials and Methods: Hydration Guidelines). Elevated eosinophil counts (reported since the earliest use of aldesleukin; ref. 23) were seen in nearly all patients and were more marked with extended duration of therapy. Increased eosinophils occur secondary to IL2 signaling, resulting in elevated plasma concentrations of IL5, a growth factor for eosinophils. Although this can be associated with

Figure 1. Waterfall and spider plots by tumor type for the response evaluable population (n = 37). **A,** Waterfall plot showing the best percentage change in target lesion at data cutoff (January 18, 2019) according to tumor PD-L1 status at baseline. **B,** Spider plots showing the deepening of responses over time according to tumor type and PD-L1 status at baseline. End of treatment is based on date and reason for discontinuing both BEMPEG and nivolumab, whichever is later. Patients may discontinue treatment due to nontarget lesion progression or appearance of new lesion. Response-evaluable population includes eligible patients with at least one postbaseline assessment. Best overall response per investigator assessment. Orange rectangle, PD-L1 negative (<1%); blue rectangle, PD-L1 positive (≥1%); gray rectangle, PD-L1 unknown; #, best overall response is progressive disease due to progression of a nontarget lesion or presence of a new lesion; *, best overall response is stable disease; +, best overall response is PR; CR for target lesion; nontarget lesions are still present; green circle, treatment ongoing at data cutoff.

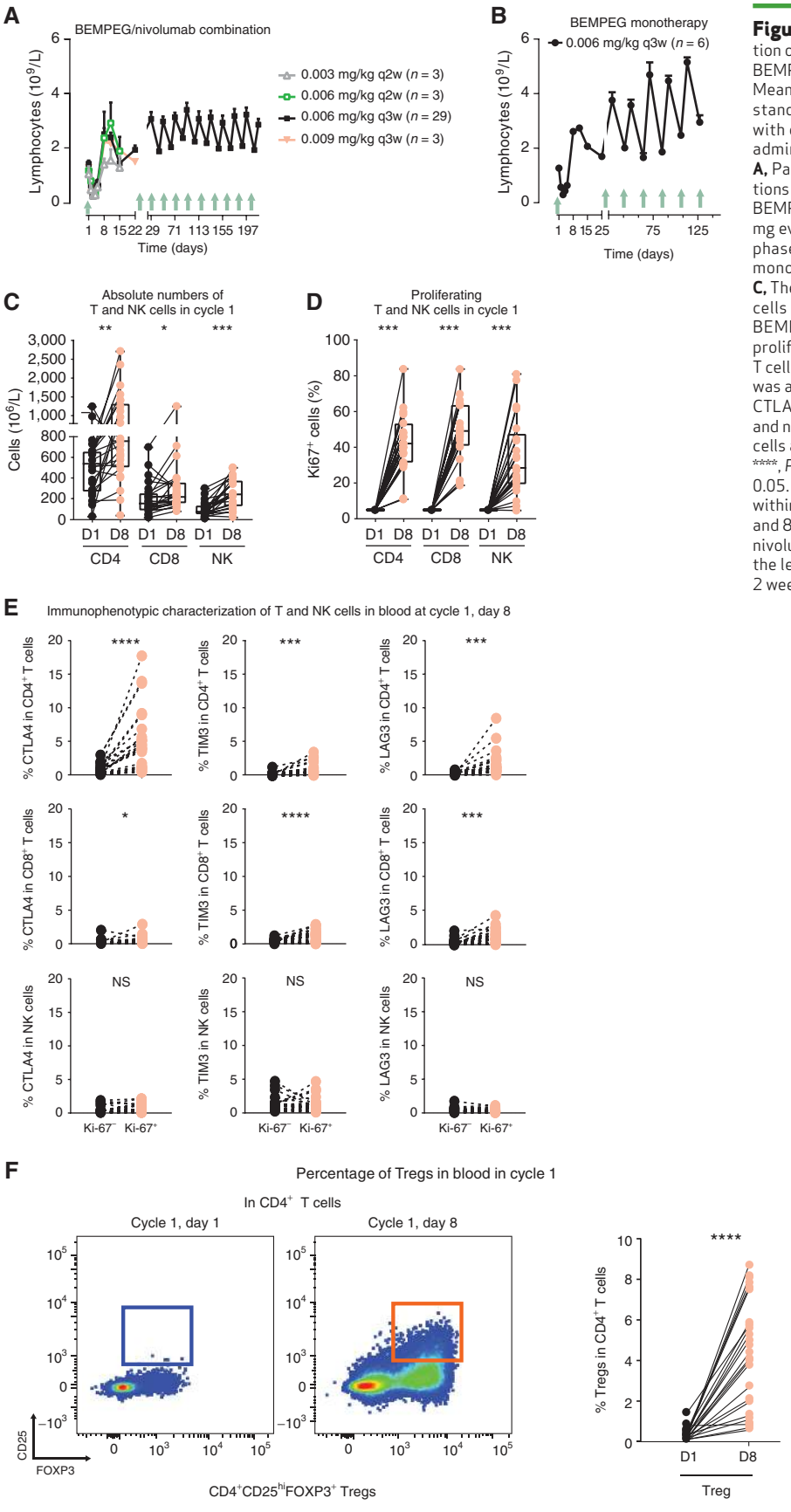


Figure 2. Increase in proliferation and activation of immune cells in peripheral blood after BEMPEG plus nivolumab treatment. **A** and **B,** Mean (SE) lymphocyte concentrations using standard hematology (complete blood count with differential). Arrows indicate treatment administration of BEMPEG plus nivolumab. **A,** Patients received various dose combinations of BEMPEG plus nivolumab in cycle 1, and BEMPEG at 0.006 mg/kg plus nivolumab 360 mg every 3 weeks in cycle 1–11. **B,** In a separate phase I trial (17) patients received BEMPEG monotherapy at 0.006 mg/kg in cycles 1–7. **C,** The absolute number of CD4+, CD8+, and NK cells before and 8 days after treatment with BEMPEG plus nivolumab. **D,** Percentage of proliferating (Ki67+) cells within CD4+ and CD8+ T cells and NK cells before and after treatment was analyzed by flow cytometry. **E,** Expression of CTLA4, TIM3, and LAG3 by proliferating (Ki67+) and nonproliferating (Ki67−) CD8+, CD4+, and NK cells at day 8 on treatment ($N = 24$). Paired t test: ****, $P < 0.0001$; ***, $P < 0.001$; **, $P < 0.01$; *, $P < 0.05$. **F,** Percentage of CD4+CD25hiFOXP3+ Tregs within total CD4+ T cells in the blood before and 8 days after treatment with BEMPEG plus nivolumab. A representative result is shown on the left. D, day; NS, not significant; q2w, every 2 weeks; q3w, every 3 weeks.

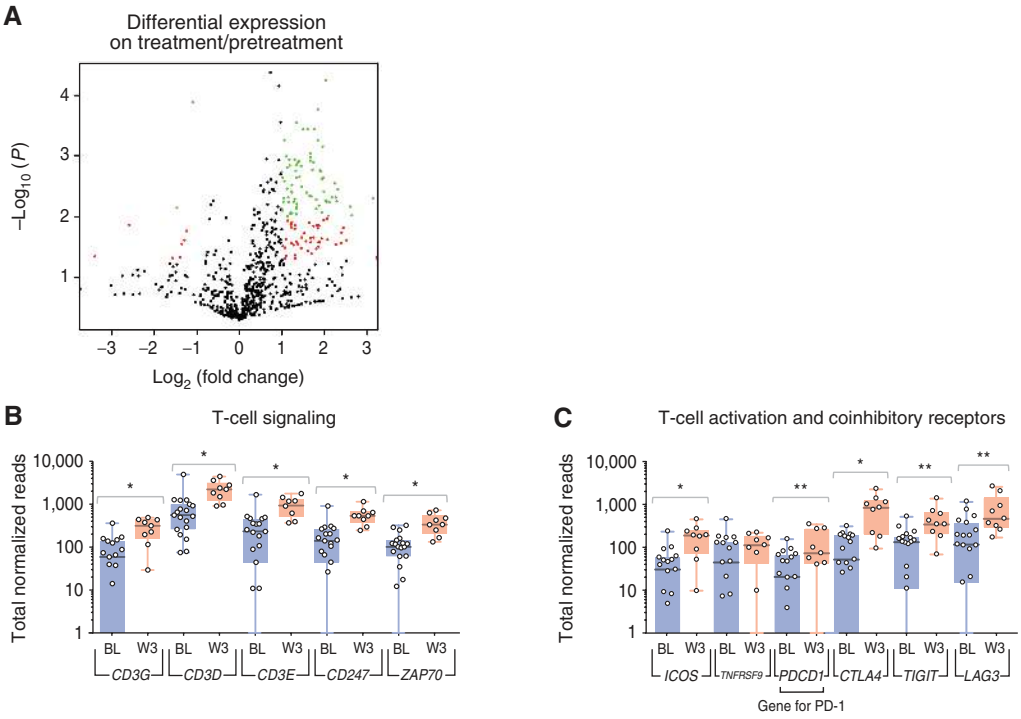


Figure 3. Combination therapy enhances intratumoral expression of immune-based gene sets. NanoString gene analysis using the nCounter PanCancer Immune Profiling Panel to quantify transcript levels of 784 genes was performed on 19 baseline (BL) and nine on-treatment tumor biopsies. **A,** Volcano plot displaying each gene's $-\log_{10}$ (*P* value) and log 2-fold change with the selected covariate. Highly statistically significant genes fall at the top of the plot, and highly differentially expressed genes fall to either side. Red and green dots in the volcano plot are genes ≥2-fold change in linear space at a significant level of $P \leq 0.05$ and $P \leq 0.01$, respectively, when applying a one-tailed *t* test. **B**–**G,** Transcriptional analysis of the tumor biopsies at baseline (blue) and after BEMPEG plus nivolumab treatment (orange); *P* values were obtained using unpaired *t* test (one-tailed), *, $P \leq 0.05$; **, $P \leq 0.01$. **B,** Selected genes related to T-cell signaling. **C,** Selected genes reflecting T-cell activation and coinhibitory receptors. (*continued on next page*)

hypereosinophilic syndrome, this AE was not observed with the BEMPEG/nivolumab combination. As we continue to learn about the safety profile of BEMPEG, improved AE management and updated guidelines may allow for higher doses of BEMPEG to be used in future studies.

ImAEs were defined in this trial by the investigator's judgment, considering the need for steroid treatment and excluding other possible etiologies. Although not formally tested and limited by the small cohort treated with the combination, the incidence of imAEs appeared to be lower with the BEMPEG/nivolumab combination (31.6%) than with nivolumab monotherapy (50.3%; ref. 24). One possible explanation for this is that BEMPEG limits systemic Th17 induction while promoting an increase in Th1/CD8$^+$ T cells in the tumor (25, 26). BEMPEG limits upregulation of inflammatory markers of Th17 and Th2 in tumors (17), which are associated with anti–PD-1– and anti-CTLA4 therapy–induced imAEs (27, 28). Thus, the BEMPEG-induced Th1–CD8$^+$ T cell/Th17 balance may help limit imAEs when combined with nivolumab, contrary to what is observed in regimens combining two CPIs (6). Assessment of this hypothesis can be shown only in randomized controlled clinical trials.

BEMPEG is an engineered IL2 receptor agonist with an average of six releasable polyethylene glycol molecules, which bias binding toward CD122 and interfere with binding to CD25 (IL2Rα). It was developed with the rationale that preferentially targeting the CD122/CD132 intermediate-affinity receptor over the CD122/CD132/CD25 high-affinity receptor would expand effector T cells over Tregs. We show that BEMPEG plus nivolumab induces proliferation and activation of peripheral CD4$^+$ and CD8$^+$ T cells and NK cells in the blood. This finding is underscored by the significant increases observed in the absolute numbers of CD4$^+$ and CD8$^+$ T cells and NK cells. Similar to previous reports (15, 17), and aligned with the proposed mechanism of action of BEMPEG, we show systemic proliferation and activation of lymphoid cells, accompanied by an increase in TILs during treatment, with no expansion of Tregs in the tumor. A lower neutrophil to lymphocyte ratio has been identified as a potential biomarker for response to CPIs (29). However, given the small sample size, we were not able to draw any conclusions on whether neutrophil to lymphocyte ratio was predictive of clinical response to BEMPEG plus nivolumab. Nevertheless, this is a point of interest that could be considered in larger clinical trials of this combination.

Importantly, the BEMPEG plus nivolumab combination also induced meaningful immunologic changes in the tumor. At week 3, TCR repertoire sequencing showed an increase in T-cell infiltration within the tumor with combination treatment, and a trend toward increased clonality. Low CD8$^+$ tumor infiltration has been shown to be a negative prognostic factor for response to immunotherapies, including single-agent PD-1 blockade (8, 30). High CD8$^+$ tumor density is reportedly predictive for response to PD-1 blockade (9), with

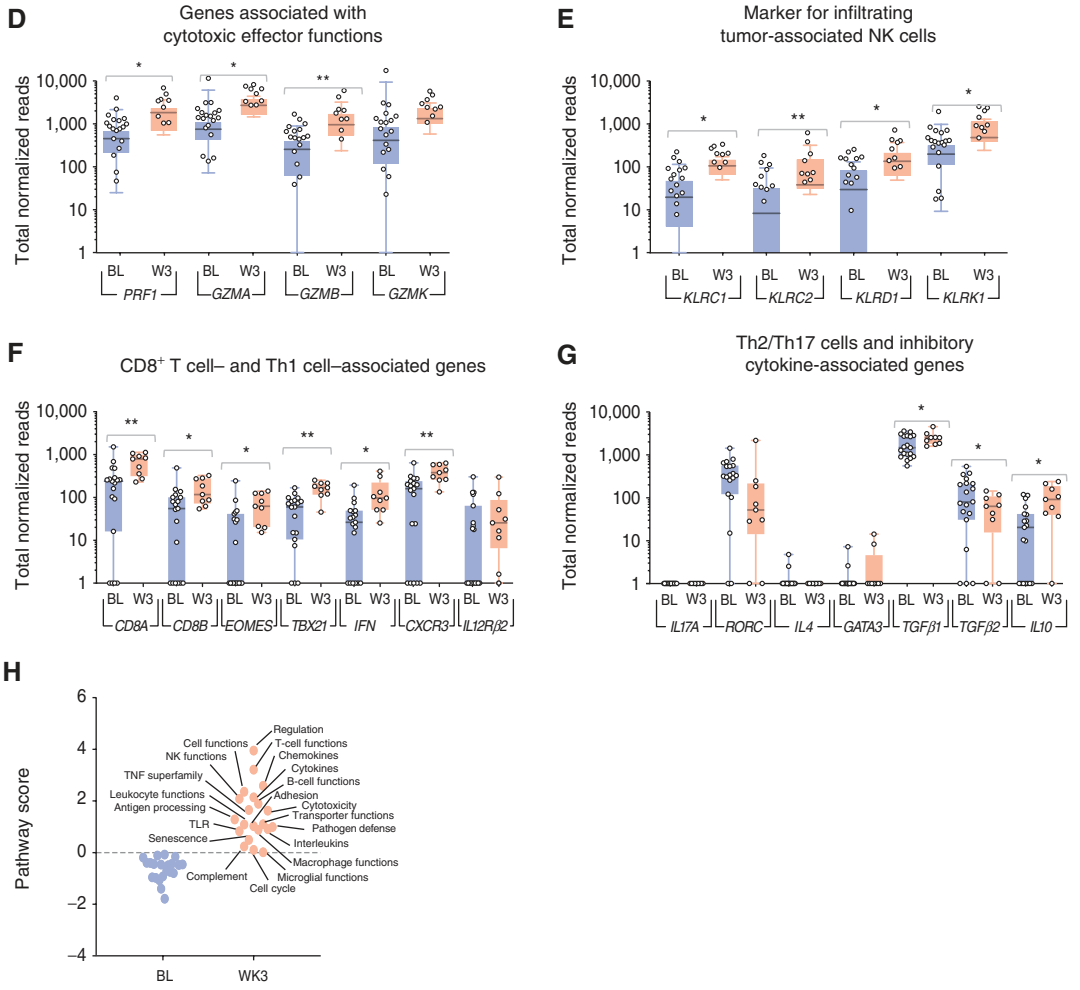
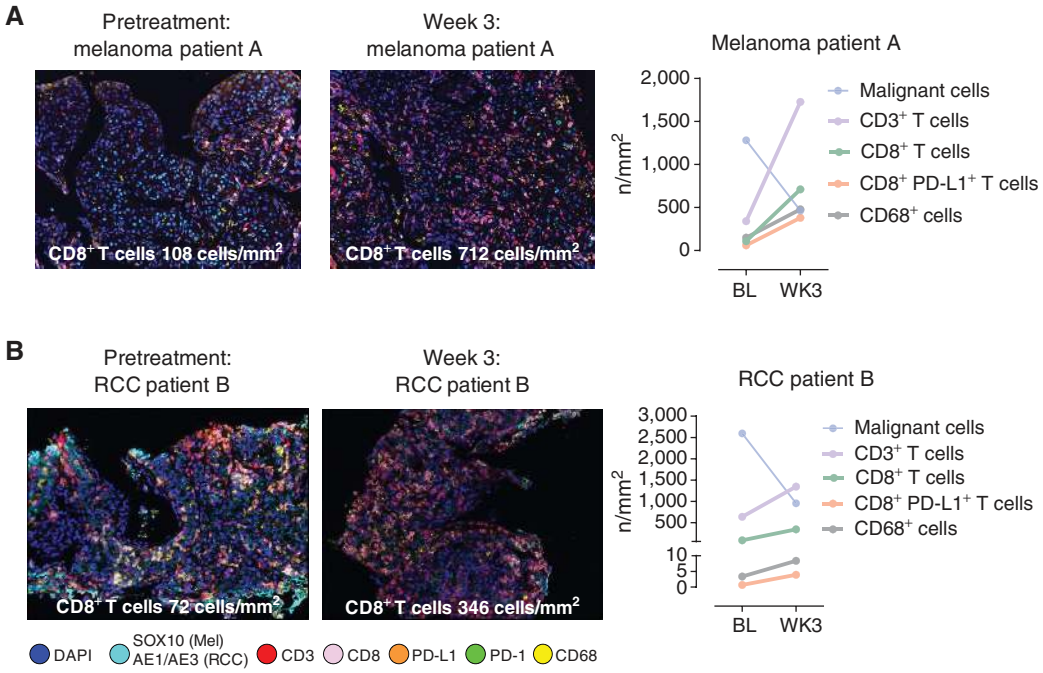


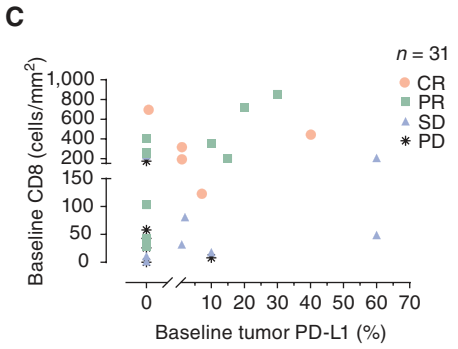
Figure 3. ***(Continued)*** **D,** Selected genes associated with cytotoxic effector functions. **E,** Selected markers found on infiltrating tumor-associated NK cells. **F,** Selected genes associated with CD8+ T cells and Th1 cells. **G,** Th2/Th17 cells and inhibitory cytokine–associated genes. **H,** Pathway scores pre-treatment and on treatment (week 3, WK3) with BEMPEG plus nivolumab for all patients with available tumor samples (19 baseline and nine on-treatment tumor biopsies) using the nSolver platform. Genes shown in **B**–**G** remained significant with multiple testing correction (Benjamini–Hochberg correction).

low CD8+ tumor density predicting poor outcome (30, 31). In this study, we observed that T-cell infiltration and clonality were significantly increased on treatment in tumor samples from patients who responded to BEMPEG plus nivolumab, but not in those from patients without a response. These findings suggest that BEMPEG plus nivolumab treatment induced clonal TIL expansion in the tumor tissue. These findings warrant exploration in a larger clinical trial to determine whether immunologic changes in the tumor microenvironment at week 3 are predictive for ultimate clinical response.

Transcriptional analysis of on-treatment tumor biopsies provides further evidence that BEMPEG plus nivolumab stimulates immune-related gene expression, including expression relating to immune activation, as well as cytotoxic and effector functions. The combination led to enhanced expression of genes associated with CD45+ lymphocytes, CD8+ T cells, macrophages, and cytotoxic cells, but not B cells or neutrophils. BEMPEG plus nivolumab increased mRNA expression of *PDCD1*, the gene encoding PD-L1. IHC data also demonstrated increased PD-L1 protein. This phenomenon is of great interest because low tumor PD-L1 expression has been shown to be a negative prognostic factor for response to single-agent PD-1 blockade (7). Together, these findings suggest that BEMPEG has an immunologic role in driving the rapid and continuous activation of the immune system (15–17).

The BEMPEG and nivolumab combination was tested in this trial based on the hypothesis that IL2-induced immune activation would enhance the antitumor response of nivolumab monotherapy. The total investigator-assessed ORR with BEMPEG plus nivolumab was 59.5%, with tumor-specific ORRs of 63.6% in 1L melanoma (including four CRs), 71.4% in 1L RCC, 28.6% in 2L I-O-naïve RCC, and 60.0% in I-O-naïve NSCLC. At the RP2D, the ORR across tumor types was 66.7%. Five of the 7 patients who achieved a CR did so on or after the fourth postbaseline scan (≥ eight treatment cycles). This deepening of response over time could





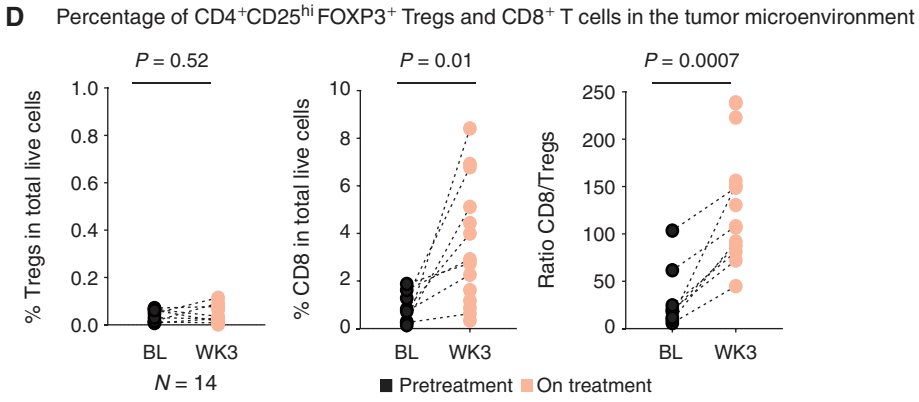


Figure 4. BEMPEG combined with nivolumab increases immune infiltration in treated tumors. Formalin-fixed, paraffin-embedded tumor biopsies collected pretreatment and on treatment were stained for immune cell markers by multispectral immunofluorescence: 1 patient with melanoma (**A**); and 1 patient with RCC (**B**). **C,** Thirty-one baseline (BL) tumor samples were evaluable for CD8+ T cells, PD-L1 expression, and response. **D,** Percentage of CD4+CD25hiFOXP3+ Tregs (left) and CD8+ T cells (center) within total live cells in tumor pretreatment and on-treatment samples, and the ratio of CD8 to Tregs (right). On-treatment tumor biopsies were analyzed from 14 patients (paired *t* test with a significance value of $P < 0.05$). (*continued on next page*)

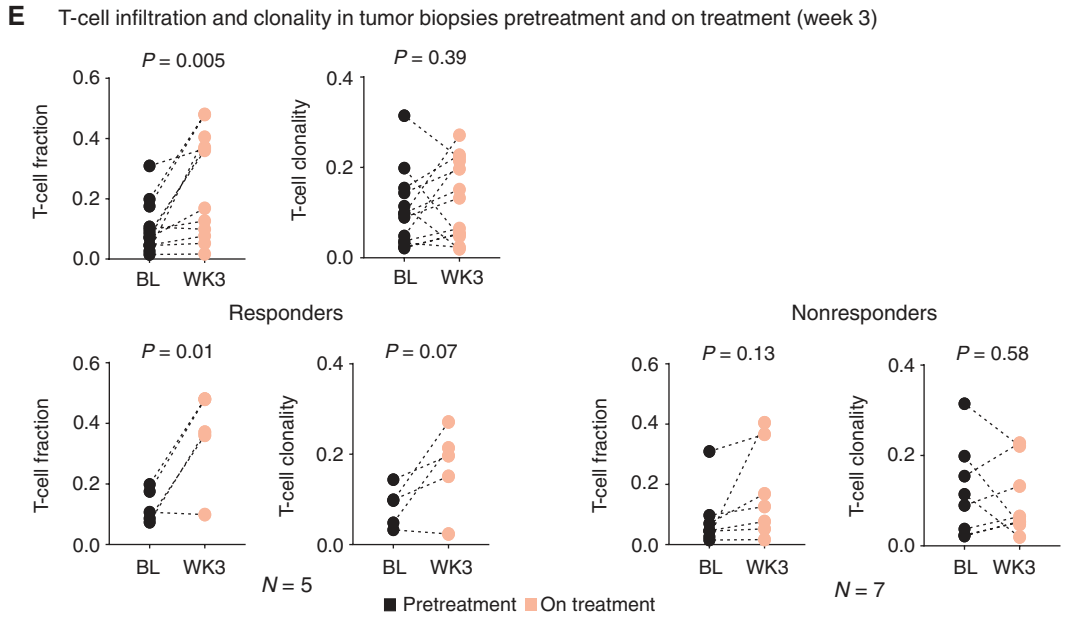


Figure 4. ***(Continued)*** **E,** TCR repertoire sequencing examining the percentage of T-cell infiltration (the fraction productive of cells mass estimate values obtained through the immuSeq analyzer software) and clonality in pretreatment and on-treatment tumor samples, $n = 12$ [responders ($n = 5$) and nonresponders ($n = 7$), paired t test]. Best overall response (RECIST v1.1) was CR or PR for responders or SD or PD for nonresponders. SD, stable disease; WK, week.

possibly be due to BEMPEG's ability to continuously mobilize antigen-specific lymphocytes. Although comparisons cannot be formally made, especially with the low number of patients in this trial, it is of interest to note the rates of objective response with nivolumab monotherapy in clinical trials: 43.7% in previously untreated metastatic melanoma (22), 25.1% in previously treated advanced RCC (20), and 20.0% in previously treated advanced NSCLC (21).

As is common with all phase I noncomparator studies, there are limitations that must be taken into consideration when interpreting results of this study, including the small number of patients and the lack of comparison with a randomized nivolumab monotherapy arm.

In conclusion, this study has established the RP2D of BEMPEG in combination with nivolumab to take forward into future studies. AEs were manageable and generally reversible using local treatment standards and preventative guidelines. The addition of BEMPEG did not appear to exacerbate the incidence of AEs typically associated with anti–PD-1 blockade, suggesting a lack of overlapping toxicities. BEMPEG plus nivolumab achieved encouraging ORRs compared with historic data for nivolumab alone, independent of baseline PD-L1 expression. These responses continue to deepen over time, and thus we anticipate more patients may develop CRs during phase II of the study. Importantly, we show that BEMPEG plus nivolumab increased systemic and intratumoral $CD8^+$ T-cell responses with T-cell infiltration into the tumor, and increased expression of the gene encoding PD-L1. These findings support the hypothesized mechanism of action of BEMPEG, suggesting that this compound may alter the blood and tumor microenvironment to enhance the $CD8^+$ T cell–mediated tumor destruction induced by PD-1 blockade, independently of established predictors of response to PD-1 blockade. This report also reinforces the importance of cytokines for productive antitumor T-cell immunity and the potential for combining optimized cytokine therapies with complementary agents, such as CPIs, to enable more patients to experience deep and durable responses. On the basis of our phase I findings, the PIVOT-02 clinical trial has opened additional cohorts in other tumor types including breast, urothelial, and colorectal cancers. Furthermore, BEMPEG plus nivolumab is also being evaluated in separate phase II and III pivotal studies [PIVOT-09 (RCC; NCT03729245); PIVOT-10 (urothelial cancer; NCT03785925), PIVOT IO 009 (muscle-invasive bladder cancer; NCT04209114); and PIVOT-IO 001 (melanoma; NCT03635983)].

METHODS

Study Design and Patients

This multicenter, nonrandomized, open-label study (NCT02983045; PIVOT-02) of BEMPEG in combination with nivolumab in selected advanced or metastatic solid tumors comprises a phase I dose-escalation portion, presented herein, and an ongoing phase II dose-expansion portion, which will be reported separately.

Eligible patients had confirmed locally advanced or metastatic solid tumors: treatment-naïve metastatic melanoma and a known *BRAF* status (1L melanoma); metastatic RCC with either no prior treatment for metastatic disease (1L RCC) or only one prior line of VEGF-tyrosine kinase inhibitor therapy (2L I-O naïve RCC); or advanced or metastatic NSCLC which lacks *EGFR* inhibitor–sensitizing mutation/deletion and *ALK* translocation by local testing, with no prior treatment for advanced disease or no prior immunotherapy treatment and up to two prior lines of systemic therapy for advanced disease (I-O-naïve NSCLC). Patients with a history

of autoimmune disease, active brain metastases, or who had received prior immunotherapies, including IL2 therapy, were excluded. Other eligibility criteria included: age 18 years or older, measurable disease per RECIST v1.1, life expectancy of >12 weeks, Eastern Cooperative Oncology Group performance status of 0 or 1, and adequate organ function. All patients were required to have baseline (archival and fresh) and on-treatment tumor biopsies unless the biopsy could not be safely performed.

The study was conducted in accordance with Good Clinical Practice guidelines and the Declaration of Helsinki. All patients provided written informed consent. Approvals for the study protocol (and any modifications thereof) were obtained from independent ethics committees and the institutional review board at each participating center.

Outcomes

The primary objectives were to evaluate safety and tolerability, establish the MTD and/or RP2D, and evaluate efficacy by assessing the ORR at the RP2D per RECIST v1.1. Secondary objectives included further evaluation of efficacy, including clinical benefit rate (also known as DCR) and DOR. Exploratory objectives included evaluation of the immunologic effects in blood and tumor, and efficacy as it relates to tumor PD-L1 expression.

Procedures

On day 1 of each cycle, BEMPEG was administered intravenously (i.v.) over 30 (±5) minutes at a starting dose of 0.006 mg/kg every 3 weeks, followed by nivolumab at doses of 360 mg i.v. every 3 weeks or 240 mg every 2 weeks in an outpatient setting. Each treatment cycle lasted 21 or 14 days for the every 3 weeks and every 2 weeks schedules, respectively.

At least 3 patients were enrolled per dose cohort. If no DLT was observed in the first 3 patients treated, doses were escalated to the next cohort or treatment schedule. If one DLT was observed in the first 3 patients, then 3 more patients were enrolled into the same dose cohort. If 2 or more patients within a cohort experienced DLTs, then that dose level exceeded the MTD. The benefit/risk profile of lower doses of BEMPEG could be evaluated within the protocol upon joint agreement by the sponsor and an investigator. Patients were observed for DLTs during the DLT observation period, defined as one cycle for every 3 weeks schedules and two cycles for every 2 weeks schedules. A DLT was defined as a grade 3 or higher AE [NCI Common Terminology Criteria for Adverse Events (NCI CTCAE) version 4.03], deemed related or possibly related to study drug, that does not resolve to grade 1 or baseline within 7 days. Other DLTs were defined as per the Supplementary Materials and Methods: Definition of a DLT.

MTD evaluation was based on the DLT population (all patients who completed at least the DLT observation period or discontinued from study treatment due to DLT). The MTD/RP2D was declared as the dose below the maximum-administered dose where at least 6 patients were enrolled and 1 or fewer patients had experienced a DLT. Additional patients could be treated at the RP2D to further evaluate the safety and tolerability of the BEMPEG/nivolumab combination.

Patients were treated until disease progression, death, unacceptable toxicity, symptomatic deterioration, achievement of maximal response, investigator decision to discontinue treatment, patient withdrawal of consent, pregnancy, loss to follow-up, or study termination by the sponsor. Patients with a confirmed CR could be treated on study for a maximum period of 2 years at the discretion of the investigator.

Biomarker Analysis

Tumor tissue samples (archival and fresh) were obtained at baseline and in cycle 1 between days 15 and 21 for: flow cytometry analysis of Tregs, $CD8^+$ and $CD4^+$ T cells, and NK cells (fresh only); IHC analysis of immune markers, including PD-L1 and CD8 expression; gene expression profiling using nCounter Human PanCancer Immune Profiling Panel (NanoString); and TCR sequencing (Adaptive Biotechnologies; ref. 32). The expression levels of each gene were normalized to those of control genes using a customary software (Precision for Medicine) and corrected for false discovery using the Benjamini–Hochberg method. Pathway and cell type gene analysis were performed and normalized using NanoString's software nSolver v3.0.22 with the Advanced Analysis Module v2.0.

PD-L1–positive tumors were defined by central testing using the Dako 28-8 PharmDx assay as staining on ≥1% of tumor cells (minimum of 100 evaluable tumor cells in the sample). In the case of insufficient tumor tissue, local pathology data were used to assess baseline PD-L1 status. Lower baseline tumor T-cell infiltration was defined as containing <150 cells/mm^2, the median number of $CD8^+$ T cells observed at baseline. To characterize $CD4^+$ and $CD8^+$ T cells and NK cells in the blood, peripheral blood mononuclear cells and fresh tumor tissue homogenates were stained and analyzed using flow cytometry as described previously (33). Further details on biomarker analyses are provided in the Supplementary Materials and Methods: Biomarker Methodology.

Statistical Analysis

Safety was evaluated in all patients who received at least one treatment dose and was summarized according to NCI CTCAE v4.03. ImAEs, such as those characteristic of CPIs, were recorded per the clinical judgment of the investigator, including the requirement for steroid treatment and ruling out neoplastic, infectious, metabolic, toxic, or other etiologies to the extent possible.

Tumors were evaluated at baseline and every 8 weeks by the local investigator and by blinded independent central radiology. Per protocol, the primary efficacy measurement was ORR per RECIST v1.1, based on antitumor activity data provided by the investigator's assessment using the response-evaluable population, defined as all patients who had at least one postbaseline scan. Definitions were: ORR = confirmed CR or PR; DCR = confirmed CR, PR, or SD; DOR = date from the first documented CR or PR to the date of the first objectively documented disease progression or death due to any cause; TTR = the time from the date of the first dose to the date of first documented CR or PR.

Summary statistics were provided for continuous variables; categorical variables were summarized by frequency counts and percentages. All statistical calculations were performed using the Statistical Analysis System (SAS) version 9.4 (SAS Institute). Statistical analyses for immune profiling were performed with GraphPad Prism 8.0 (GraphPad Software), used a two-tailed paired and unpaired t test with a significance value of $P < 0.05$.

Disclosure of Potential Conflicts of Interest

A. Diab reports receiving a commercial research grant from, has received speakers bureau honoraria from, and has an advisory board relationship with Nektar Therapeutics. N.M. Tannir reports receiving commercial research grants from and has an advisory board relationship with Bristol-Myers Squibb and Nektar Therapeutics. P. Hwu is a scientific advisory committee member at Dragonfly and Sanofi and is a consultant at Immatics and GlaxoSmithKline. V. Papadimitrakopoulou is vice president, clinical development leader, at Pfizer, Inc., is an advisory board member at Clovis Oncology, Merck, Lilly, Janssen, AstraZeneca, ARIAD, Nektar Therapeutics, LOXO, Araxes Pharma, AbbVie, Bristol-Myers Squibb, Exelixis, Pfizer, Novartis, Takeda, Tesaro, Arrys Therapeutics, Gritstone Oncology, Genentech/Roche, and Leeds Biolabs, is a consultant at Bolt Therapeutics, G2 Innovation, and ACEA Biosciences, and has received speakers bureau honoraria from Roche-Genentech. H.M. Kluger is a consultant at Corvus, Nektar Therapeutics, Biodesix, Iovance, Immunocore, Celldex, Array Biopharma, Merck, Instil Bio, and Elevate Bio, and reports receiving commercial research grants from Merck, Apexigen, and Bristol-Myers Squibb. S.N. Gettinger is an advisory board member at

Nektar Therapeutics and BMS, and has an advisory board relationship with NextCure. M. Sznol is a consultant at Nektar Therapeutics, Adaptive Biotechnologies, Immunocore, Molecular Partners, Verastem, Boehringer Ingelheim, Torque, Agenus, Innate, Pieris, Numab, AbbVie, Evolveimmune, Seattle Genetics, Biontech, Eli Lilly, AstraZeneca, Nextcure, Adaptimmune, Dragonfly, Bristol-Myers, Genentech-Roche, Servier, and Alligator, and has ownership interest (including patents) in Nextcure, Adaptive Biotechnologies, and Evolveimmune. S.S. Tykodi reports receiving commercial research support from Merck, Nektar Therapeutics, Clinigen, Calithera Biosciences, Pfizer, Bristol-Myers Squibb, Exelixis, and Jounce Therapeutics. B.D. Curti is a data safety monitoring board member at Merck and reports receiving commercial research grants from AstraZeneca and Clinigen. M.A. Tagliaferri is chief medical officer at Nektar Therapeutics and has ownership interest (including patents) in Nektar Therapeutics. J. Zalevsky is chief research and development officer at Nektar Therapeutics and has ownership interest (including patents) in Nektar Therapeutics. A.L. Hannah is a consultant at Nektar Therapeutics. U. Hoch is VP clinical pharmacology at Nektar Therapeutics. S. Aung has ownership interest (including patents) in Nektar Therapeutics and was senior director, oncology clinical development at Nektar Therapeutics at the time the work was conducted. C. Fanton is senior director, translational research at and has ownership interest (including patents) in Nektar Therapeutics. A. Rizwan was director, clinical pharmacology at Nektar Therapeutics at the time the work was conducted. E. Iacucci is an employed scientist at Nektar Therapeutics. Y. Liao is a biostatistician at Nektar Therapeutics. C. Bernatchez reports receiving commercial research support from Nektar Therapeutics. M.E. Hurwitz reports receiving commercial research support from and has an advisory board relationship with Nektar Therapeutics. D.C. Cho is a consultant at Nektar Therapeutics, Pfizer, HUYA, Puretech, and Torque. No potential conflicts of interest were disclosed by the other authors.

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