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PHASE I STUDIES

Clinical outcomes of advanced stage cancer patients treated with sequential immunotherapy in phase 1 clinical trials

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Summary

Background Given the increasing number of available immunotherapeutic agents, more patients are presenting after failing immunotherapy in need of new treatment options. In this study, we investigated the clinical outcomes of patients treated with sequential immunotherapy. *Methods* We performed a retrospective review of 90 advanced stage cancer patients treated on immunotherapy-based phase 1 clinical trials at Winship Cancer Institute from 2009 to 2017. We included 49 patients with an immune checkpoint inhibitor (ICI)-indicated histology. Patients were analyzed based on whether they had received prior ICI. Clinical outcomes were overall survival (OS), progression-free survival (PFS), and clinical benefit (best response of complete response, partial response, or stable disease). Univariate analysis (UVA) and multivariate analysis (MVA) were performed using Cox proportional hazard or logistic regression model. Covariates included age, liver metastases, number of prior lines of therapy, histology, and Royal Marsden Hospital (RMH) risk group. *Results* The most common histologies were melanoma (61%) and lung/head and neck cancers (37%). More than half of patients ($n = 27$, 55%) received at least one ICI prior to trial enrollment: ten received anti-PD-1, two received anti-CTLA-4, five received anti-PD-1/CTLA-4 combination, and ten received multiple ICI. In MVA, ICI-naïve patients had significantly longer OS (HR: 0.22, CI: 0.07–0.70, $p = 0.010$) and trended towards higher chance of CB (HR: 2.52, CI: 0.49–12.97, $p = 0.268$). Patients who received prior ICI had substantially shorter median OS (10.9 vs 24.3 months, $p = 0.046$) and PFS (2.8 vs. 5.1 months, $p = 0.380$) than ICI-naïve patients per Kaplan-Meier estimation. Within the ICI-naïve group, 78% (7 of 9) of patients who received prior interleukin (IL-2) or interferon gamma (IFNγ) experienced disease control for at least 6 months, compared to a disease control rate of 15% (2 of 13) in patients who had received chemotherapy, targeted therapy, or no prior treatment. *Conclusions* ICI-naïve patients may experience improved clinical outcomes on immunotherapy-based phase 1 clinical trials than patients who have received prior ICI. This may be particularly true for patients who received prior IL-2 or IFNγ. Further development of immunotherapy combination therapies is needed to improve clinical outcomes of these patients. These results should be validated in a larger study.

Keywords Combination therapies · Immunotherapies · Immune checkpoint blockade · Immune response · T cell exhaustion

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Background

The immune checkpoint pathway is an important aspect of the cancer-immunity cycle that allows tumor cells to evade the host immune system [1]. Over the past decade, monoclonal antibodies targeting CTLA-4, PD-1, and PD-L1 have shown to be effective in treating several different solid tumor and hematologic malignancies [2–13]. A total of six immune checkpoint inhibitors (ICI) have gained FDA approval and several others are in development [14]. These agents have become an attractive treatment option for patients given their

tolerable toxicity profile and potential for durable response [15–17]. Given the increased availability of immunotherapeutic agents, more patients are presenting after failure of immunotherapy (IO) in need of new treatment options. This is particularly common in malignancies for which there are already FDA-approved ICI-based regimens in multiple settings such as melanoma, renal cell carcinoma (RCC), non-small cell lung cancer (NSCLC), and urothelial carcinoma. Unfortunately, limited data exists regarding the clinical outcomes of patients who receive multiple IO agents. Hence, this important clinical situation is an unmet need in the field of oncology.

Although ICI have targets in the same pathway, they exert differential effects on the host immune system. Interestingly, the immunologic effects induced in patients treated with ICI are distinct between patients treated with monotherapy, combination therapy, and sequential ICI treatment [18]. More specifically, PD-1 blockade upregulates genes involved in cytolysis and natural killer (NK) cell function, while anti-CTLA-4 stimulates proliferation of transitional memory T-cells and combination treatment with anti-PD-1/anti-CTLA-4 ICI induces upregulation of genes involved in chemokine signaling and proliferation [18]. There are even minor structural differences between drugs within the same class, such as the PD-1 inhibitors nivolumab and pembrolizumab, that may impact patients' response to treatment [19]. Furthermore, the clinical activity of sequential IO is not clear.

Characterizing the clinical outcomes of patients treated with sequential ICI is of significant clinical importance. Increasing numbers of patients are presenting as candidates for subsequent ICI-based regimens. Given the limited data available in this subset of oncology patients, we investigated the outcomes of advanced stage cancer patients who were treated with IO-based regimens as part of phase 1 clinical trials after previously receiving an ICI. The results of this study are hypothesis-generating and useful for practicing oncologists in the academic and community settings.

Patients and methods

Patient selection and data collection

We gathered clinical information for 90 patients treated on IO-based phase 1 clinical trials at Winship Cancer Institute of Emory University between 2009 and 2017. All patients ($n = 49$) with an ICI-indicated histology (melanoma, lung cancer, head and neck cancer, and bladder cancer) were included in the study. Patients were then characterized based on whether they had received an ICI prior to enrollment onto the trial. Data collected included demographic information, medication allergies, sites of metastases, prior lines of systemic therapy in the metastatic setting, start and stop date of IO on trial, date of death or hospice referral, best response to IO, Royal Marsden Hospital (RMH) risk group (albumin <3.5, lactate dehydrogenase (LDH) > upper normal limit, and > 2 sites of metastatic disease). Response to the IO-based trial regimen was determined by Response Evaluation Criteria in Solid Tumors v1.1 [20].

Statistical methods

Overall survival (OS) and progression-free survival (PFS) were calculated in months from IO initiation to date of death or hospice referral and clinical or radiographic progression, respectively. Clinical benefit (CB) was defined as complete response (CR), partial response (PR), or stable disease (SD). Univariate analysis (UVA) and multivariate analysis (MVA) were carried out using Cox proportional hazard or logistic regression model. Covariates included histology, RMH risk group, number of prior lines of systemic treatment, age, and presence of liver metastases. The significance level was set at 0.05 by two-sided test.

Results

Baseline disease characteristics and demographics

The baseline demographic and disease characteristics are shown in Table 1. The median age was 67 years and most patients (78%) were men. The most common histologies were melanoma (61%) and lung and head and neck cancers (37%). The majority of patients (81%) had good RMH risk status.

Prior systemic treatment

The majority of patients ($n = 29$, 59%) had received at least 2 lines of prior systemic treatment before receiving IO on trial. More than one-half of patients ($n = 27$, 55%) had received at least one ICI prior to trial enrollment. Of these 27 patients, ten received anti-PD-1, two received anti-CTLA-4, five received anti-PD-1/CTLA-4 combination therapy, and ten received multiple ICI. Of the 22 patients who had not received prior ICI, three were treatment-naïve, seven received high-dose interleukin-2 (IL-2), one received only interferon-gamma (IFNγ), one received IFNγ and IL-2 sequentially, and ten were previously treated with chemotherapy, targeted therapy, or both.

IO treatment regimens

Of the ten patients who had received a prior PD-1 inhibitor, three subsequently received anti-CTLA-4 combined with an experimental agent, four were subsequently treated with anti-

Table 1 Baseline patient information

Variable	Category		*n* (%)	
Gender				
	M		38 (77.6)	
	F		11 (22.4)	
Race				
	White		43 (87.8)	
	Black		4 (8.2)	
	Asian/unknown		2 (4.1)	
ECOG PS				
	0		19 (39.6)	
	1+		29 (60.4)	
	Missing		1 (2.0)	
Histology				
	Melanoma		30 (61.2)	
	Lung/Head & Neck Cancer		18 (36.7)	
	Urothelial Carcinoma		1 (2.0)	
RMH Risk Group				
	Good		39 (81.3)	
	Poor		9 (18.8)	
	Missing		1 (2.0)	
Liver Metastases				
	Yes		15 (30.6)	
	No		34 (69.4)	
Prior ICI				
	Yes		27 (55.1)	
		Anti-PD-1		10 (20.4)
		Anti-CTLA4		2 (4.0)
		Anti-PD-1/CTLA-4 Combination		5 (10.2)
		2+ prior ICI		10 (20.4)
	No		22 (44.9)	
IO Regimen on Trial				
	Experimental IO Monotherapy		10 (20.4)	
	Anti-PD-L1 Monotherapy		15 (30.6)	
	FDA-Approved ICI + Experimental IO Combination		24 (49.0)	

M Male, *F* Female, *ECOG PS* Eastern cooperative oncology group performance status, *RMH* Royal Marsden Hospital, *ICI* Immune checkpoint inhibitor, *PD-1* Programmed cell death protein-1, *CTLA4* Cytotoxic T lymphocyte-associated protein 4

PD-1 and an experimental agent, and three received experimental IO monotherapy on trial. Six of the ten patients who had received multiple prior ICI received experimental IO monotherapy on trial, while the other four received an experimental agent plus either anti-PD-1 ($n = 2$) or CTLA-4 ($n = 2$). Of the five patients who had received a prior PD-1/CTLA-4 combination, three received anti-PD-1 with an experimental agent, one received an experimental IO agent, and one anti-CTLA-4 combined with an experimental agent. Both patients who had received prior anti-CTLA-4 monotherapy received an experimental agent plus either anti-PD-1 ($n = 1$) or anti-CTLA-4 ($n = 1$).

All nine patients who had received high dose IL-2 or IFNγ received anti-PD-L1 monotherapy on trial. The three patients who had not received any prior treatment were treated with a regimen of anti-PD-1 plus an experimental agent. Of the ten patients who had received chemotherapy and/or targeted therapy, six received anti-PD-L1 monotherapy and four received anti-PD-1 with an experimental agent.

Prior ICI vs ICI-naïve clinical outcomes

In multivariate analysis, patients who had not received a prior ICI had significantly longer OS than patients who had (HR: 0.22, CI:

0.07–0.70, $p = 0.010$, Table 2). These patients also trended towards a higher chance of CB (HR: 2.52, CI: 0.49–12.97, $p = 0.268$). The CB rate was much lower for patients who had received prior ICI (52%), compared to patients who had not (77%). ICI-naïve patients also had substantially longer median OS (24.3 vs 10.9 months, $p = 0.046$, Fig. 1) and PFS (5.1 vs. 2.8 months, $p = 0.3796$, not shown) than patients who had received a prior ICI per Kaplan-Meier estimation. Within the ICI-naïve cohort, patients who had received prior IL-2 or IFNγ gamma had improved clinical outcomes (Table 3**)** compared to patients who had received chemotherapy or targeted therapy prior to treatment with IO on trial (Table 4**)**. Seven out of the nine (78%) patients who had received prior IL-2 or IFNγ experienced disease control for at least 6 months on treatment, compared to a disease control rate of 15% (2 of 13) for patients who had received chemotherapy, targeted therapy, both, or no treatment at all. The median OS ($p = 0.1728$) and PFS ($p = 0.0014$) were also substantially longer for patients who had received prior IL-2 or IFNγ (26.2 months and 8.2 months, respectively) compared to patients who had not (13.5 months and 3.6 months, respectively) per Kaplan-Meier estimation (Figs. 2-3).

Discussion

As of May 2018, six ICI agents are approved for the treatment of several malignancies including NSCLC, RCC, urothelial carcinoma, and melanoma. In addition to the approved regimens, several other immunotherapeutic agents are currently being investigated in ongoing clinical trials. The increasing number of available immunotherapeutic agents has led to an increasing population of patients who have failed previous ICI and are candidates for treatment with a subsequent IO regimen. Despite this common clinical situation, few studies have investigated the effectiveness of sequential IO. In this study, we showed that immunotherapy-naïve patients had improved clinical outcomes on immunotherapy-based phase 1 clinical trials compared to patients who had previously received at least one immune checkpoint inhibitor. The difference in clinical outcomes between these two subsets of patients was mainly due to our finding that patients who had received prior IL-2 or IFNγ had significantly longer OS and PFS than patients who had been treated with chemotherapy, targeted therapy, or no therapy at all.

These results build upon previous data investigating the clinical outcomes of cancer patients receiving sequential immunotherapy. A retrospective study that explored outcomes in 84 melanoma patients who failed anti-PD-1 therapy and subsequently received ipilimumab ($n = 47$) or ipilimumab and nivolumab ($n = 37$) found that patients in the ipilimumab group fared much better (disease control rate = 42%) than patients who received ipilimumab and nivolumab (disease control rate = 33%) [21]. A phase 1/2 study in melanoma ($n = 91$) also found that 62% of patients treated with nivolumab after

Table 2 UVA and MVA* of prior ICI and clinical outcomes

	OS				PFS				CB			
	UVA		MVA		UVA		MVA		UVA		MVA	
	HR (CI)	*p* value	HR (CI)	*p* value	HR (CI)	*p* value	HR (CI)	*p* value	OR (CI)	*p* value	OR (CI)	*p* value
No Prior ICI ($n = 22$)	0.44 (0.19–1.01)	0.052	0.22 (0.07–0.70)	**0.01****	0.76 (0.42–1.40)	0.385	0.86 (0.39–1.87)	0.699	3.16 (0.90–11.03)	0.072	2.52 (0.49–12.97)	0.268
	Median: 24.3 months				Median: 5.1 months				Rate: 77% (4 PR, 13 SD, 3 PD, 2 NE)			
Prior ICI ($n = 27$)	Median: 10.9 months				Median: 2.8 months				Rate: 52% (1 CR, 2 PR, 11 SD, 10 PD, 3 NE)			

*Controlled for cancer type, RMH risk group, number of prior lines of treatment, age, and presence of liver metastases

**statistically significant

UVA Univariate analysis, *MVA* Multivariate analysis, *OS* Overall survival, *PFS* Progression-free survival, *CB* Clinical benefit, *ICI* Immune checkpoint inhibitor, *PR* Partial response, *SD* Stable disease, *PD* Progressive disease, *NE* Not evaluable, *CR* Complete response

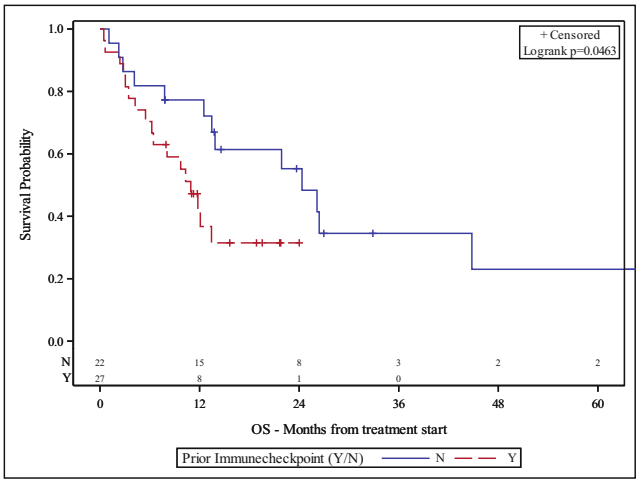

Fig. 1 Kaplan-Meier plot of prior ICI and overall survival

failed ipilimumab had maintained a best response of CR, PR, or SD 24 weeks after treatment initiation [22]. A case series of two RCC and one melanoma patient reported progressive disease as best response on nivolumab after failing anti-PD-L1 combination therapy (RCC) or pembrolizumab (melanoma) [23]. Taken together, the current literature suggests that patients who received prior anti-CTLA-4 may be more likely to have improved clinical outcomes than patients who received prior anti-PD-1 when treated with sequential immunotherapy.

Current data regarding the efficacy of sequential immunotherapy are likely explained by the differential effects that ICI regimens exert on the host immune system. Anti-CTLA-4 therapy targets priming and activation of T-cells, an earlier step in the cancer immunity cycle than that targeted by anti-PD-1 therapy, which aims to induce the final step of the cycle: killing of cancer cells [1]. Furthermore, treatment with anti-CTLA-4 monotherapy or anti-CTLA-4/PD-1 combination therapy upregulates coding genes involved in the cell cycle and proliferation of T-cells [18]. Anti-PD-1 treatment, on the other hand, induces a genomic signature involved in cytolysis and regulation of effector T and NK cells [18]. Thus, it is possible that prior treatment with anti-CTLA-4 causes proliferation and activation of T-cells, which increases the number of tumor infiltrating lymphocytes and leads to a "hot" phenotype. Moreover, the presence of intra-tumoral T-cells has been associated with a significantly higher likelihood of response to immunotherapy [24, 25].

Interestingly, all but one of the patients in this study who received IL-2 or IFNγ prior to IO experienced SD or better as their best response on trial and 78% of these patients experienced disease control for at least 6 months. This may be explained by the crucial immunoregulatory roles of IL-2, including the maintenance of $CD4^+$ regulatory T cells and differentiation of naïve $CD4^+$ T cells into T helper-1 (Th1) and T helper-2 (Th2) cells [26]. IL-2 also promotes differentiation of B and NK cells and clonal expansion after T cell exposure to antigen [27]. Pre-clinical data suggests that there may be a synergistic effect of IL-2 and anti-PD-L1 via exhausted T cell reactivation [28], which has led to the development of several ongoing clinical trials investigating the efficacy and safety of IL-2 and ICI combination therapy (NCT02983045, NCT02989714). Unfortunately, there is sparse data available regarding sequential IL-2 and ICI therapy. There is an ongoing clinical trial of 29 melanoma patients treated with sequential IL-2 and ipilimumab (NCT01856023). Early results from this trial indicate a tolerable toxicity profile as well as significantly prolonged 1-year OS (77%) and higher objective response rate (not reported) [29]. Our findings build upon the early results of this trial. Larger studies are needed to validate these preliminary findings.

Biomarkers are particularly useful in early phase clinical trials in which the investigational treatments may be effective only for a select population of patients. Biomarkers can be

Table 3 Clinical outcomes of ICI-naïve patients who received prior IL-2 or IFNγ

ID#	Histology	Prior Treatment	Trial Treatment	BR	PFS (months)	OS (months)
1	BRAF WT melanoma	IL-2	PD-L1	SD	6.90	7.82+
2	BRAF V600E mutant melanoma	IL-2	PD-L1	SD	9.82	21.88
3	BRAF V600E mutant melanoma	IFNγ, IL-2	PD-L1	PD	4.07	44.81
4	NRAS variant melanoma	IL-2	PD-L1	SD	5.26	24.35
5	BRAF V600E mutant melanoma	IL-2	PD-L1	SD	8.15	13.87
6	BRAF WT melanoma	IL-2	PD-L1	SD	8.21	14.59+
7	BRAF and NRAS WT melanoma	IL-2	PD-L1	SD	12.02	26.15
8	Melanoma	IL-2	PD-L1	PR	72.84+	72.84+
9	Melanoma	IFNγ	PD-L1	PR	19.25	101.13+

BR Best response, *PFS* Progression-free survival, *OS* Overall survival, *WT* Wild-type, *IL-2* Interleukin-2, *IFNγ* Interferon gamma, *PD-L1* Programmed death-ligand 1, *SD* Stable disease, +: Ongoing

Table 4 Clinical outcomes of ICI-naïve patients who did not receive prior IL-2 or IFNγ

ID#	Histology	Prior Treatment	Trial Treatment	BR	PFS (months)	OS (months)
1	NSCLC adenocarcinoma	Carboplatin/pemetrexed/bevacizumab	PD-L1	SD	3.65	26.42+
2	EGFR/ALK negative NSCLC	Carboplatin/paclitaxel//bevacizumab, experimental targeted therapy regimen	PD-L1	SD	5.36	32.89+
3	Squamous NSCLC	Carboplatin/paclitaxel/pemetrexed/ gemcitabine/erlotinib, experimental regimen	PD-L1	SD	2.27	2.27
4	Squamous NSCLC	Carboplatin/paclitaxel, gemcitabine, docetaxel	PD-L1	NE	0.46	1.08
5	NSCLC adenocarcinoma	Carboplatin/paclitaxel/ bevacizumab, pemetrexed, erlotinib	PD-L1	SD	4.11	12.52
6	SCC of lung	Unspecified chemotherapy regimen, carboplatin/paclitaxel, carboplatin/vinorelbine	PD-L1	SD	4.11	4.14
7	Melanoma	N/A	PD-1 + Experimental	PD	0.92	7.79
8	Rectal melanoma	N/A	PD-1 + Experimental	PR	4.90	13.77+
9	EGFR mutant NSCLC	Carboplatin/paclitaxel, gefitinib, pemetrexed, experimental targeted therapy regimen, gefitinib	PD-1 + Experimental	NE	0.62	23.69+
10	L858R and T790 M EGFR mutant NSCLC	Erlotinib, carboplatin/pemetrexed/erlotinib, pemetrexed/erlotinib, experimental targeted therapy regimen	PD-1 + Experimental	SD	6.80	13.47
11	Exon 19 deletion and T790 M mutant NSCLC	Erlotinib, carboplatin/paclitaxel/bevacizumab, experimental targeted therapy regimen, pemetrexed	PD-1 + Experimental	SD	2.53	26.97+
12	Urothelial	N/A	PD-1 + Experimental	PR	7.82+	7.82+
13	TSC-1 mutant NSCLC	Cisplatin/erlotinib, erlotinib, cisplatin/erlotinib, carboplatin/erlotinib, everolimus	PD-1 + Experimental	PD	1.38	2.76

BR Best response, *PFS* Progression-free survival, *OS* Overall survival, *NSCLC* Non-small cell lung cancer, *WT* Wild-type, *IL-2* Interleukin-2, *PD-L1* Programmed death-ligand 1, *SD* Stable disease, +: Ongoing, *NE* Non-evaluable, *SCC* Squamous cell carcinoma, *PR* Partial response, *N/A* Not applicable, *PD-1* Programmed cell death protein-1

used to identify patients who are more likely to derive clinical benefit from treatment. This may lead to personalized treatment regimens for patients, which is now recognized as the optimal way to treat oncology patients [30]. Several biomarkers have been implicated in predicting clinical responses to immunotherapy such as PBRM1, angiopoietin-2, PD-L1 staining, and inflammatory markers [31–34]. The best-studied biomarker of response to immunotherapy is PD-L1 [34]. The PD-1/ PD-L1 pathway is a critical junction in tumor evasion of immune attack, and thus increased expression of PD-L1 may indicate a poor prognosis. However, some studies report that PD-L1 negativity may not prevent tumor proliferation. Thus, while the biological framework implies that increased PD-L1 may correlate with poor prognosis, this implication is inconclusive at this point [34]. There are several challenges to using biomarkers for prediction of responses to immunotherapy. First, there is no standardized approach for PD-L1 testing, which limits the clinical utility of this biomarker [35]. Additionally, there are no widely accepted biomarkers that can be used across multiple malignancies. These challenges highlight the critical importance of identifying optimal biomarkers in patients treated with immunotherapy.

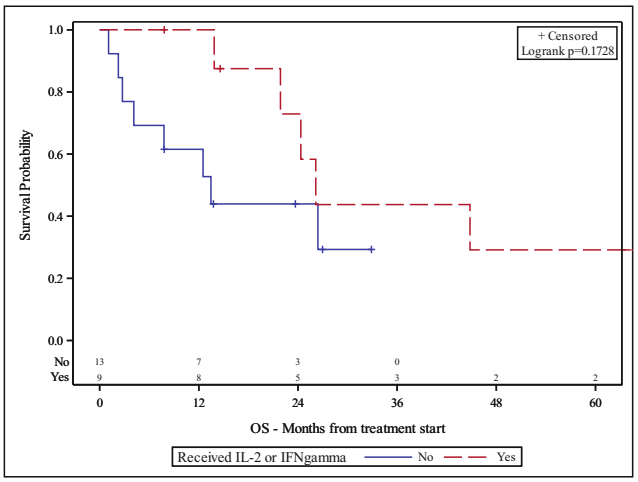

Fig. 2 Association of prior IL-2 or IFNγ with OS

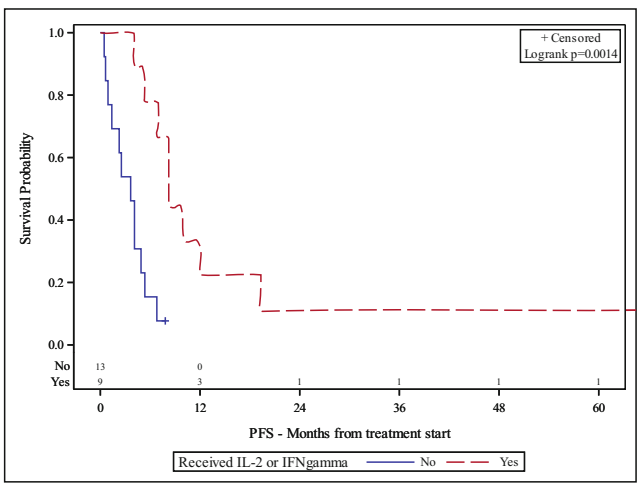

Fig. 3 Association of prior IL-2 or IFNγ with PFS

The findings of this study are hypothesis-generating and several limitations should be considered when evaluating the results of this study. First, this is a retrospective study and it is inherently subject to selection bias. We attempted to mitigate this by including all patients with an ICI-indicated histology, regardless of their disease characteristics or prior treatments. The study population was a small, heterogeneous patient population and types of IO that patients received on trial varied significantly, which limits our ability to generalize our findings to the larger population. Furthermore, it is possible that the worse clinical outcome observed in patients treated with prior ICI may be due to a poorer prognosis of these patients, given that we cannot control for all possible confounders. Additionally, the difference in clinical outcomes between patients who received prior IL-2 or IFNγ and those that did not may be at least partially explained by the differences in primary histology between the two groups, since most of the patients who received IL-2 or IFNγ had melanoma while the majority of the patients who did not receive IL-2 or IFNγ had NSCLC. Finally, we did not perform any tissue analysis and, therefore, were unable to explore the biological mechanisms underlying our clinical observations in these patients.

Conclusions

The results from this study suggest that patients who receive a second IO-based regimen may have worse clinical outcomes than patients who are ICI-naïve prior to receiving IO in a phase 1 clinical trial. We also present data that may suggest that treatment with IL-2 or IFNγ has a priming effect for subsequent treatment with IO by activating T-cells. Further development of immunotherapy combination therapies is needed to improve clinical outcomes for patients who have failed ICI previously. The results from this study should be validated in a larger, prospective study.

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Authors' contributions DJM was involved in study design and methodology, identification and selection of patients, construction of the database, data acquisition, interpretation and analysis of study results, writing the manuscript, and administrative support. MAB was involved in the identification and selection of patients, construction of the database, caring for the patients included in the study, study design and methodology, interpretation and analysis of study results, and writing the manuscript. YL was involved in the design and methodology of the study, all statistical analysis, interpretation and analysis of study results, and writing of the manuscript. JMS was involved in interpretation and analysis of study results, editing the manuscript, and administrative support. MRK was involved in editing the manuscript and administrative support. MAB and RDH supervised the study. All remaining authors were involved in the care of the patients in this study, interpretation and analysis of study results, and editing the manuscript. All authors reviewed and accepted the final version of the manuscript.

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Compliance with ethical standards

Conflict of interest Author M.A. Bilen has a consulting/advisory role with Exelixis, Nektar, and Sanofi and receives research funding from Bayer, Bristol-Myers Squibb, Genentech/Roche, Incyte, Nektar, AstraZeneca, Tricon Pharmaceuticals, Peleton, and Pfizer.

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Author R.D. Harvey declares that he has no conflict of interest.

Ethical approval All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and with the 1964 Helsinki declaration and its later amendments or comparable ethical standards.

Abbreviations ICI, immune checkpoint inhibitor; OS, overall survival; PFS, progression-free survival; UVA, univariate analysis; MVA, multivariate analysis; RMH, Royal Marsden Hospital; PD-1, programmed cell death protein-1; CTLA4, cytotoxic T lymphocyte-associated protein 4; CB, clinical benefit; IL-2, interleukin 2; IFNγ, interferon gamma; FDA, US Food and Drug Administration; IO, immunotherapy; RCC, renal cell carcinoma; NSCLC, non-small cell lung cancer; NK, natural killer; LDH, lactate dehydrogenase; HR, hazard ratio; CI, confidence interval; RCC, renal cell carcinoma; SD, stable disease; Th1, T helper-1; Th2, T helper-2

References

1. Chen DS, Mellman I (2013) Oncology meets immunology: the cancer-immunity cycle. Immunity 39(1):1–10
2. Balar AV, Galsky MD, Rosenberg JE, Powles T, Petrylak DP, Bellmunt J, Loriot Y, Necchi A, Hoffman-Censits J, Perez-Gracia JL, Dawson NA, van der Heijden M, Dreicer R, Srinivas S, Retz MM, Joseph RW, Drakaki A, Vaishampayan UN, Sridhar SS, Quinn DI, Durán I, Shaffer DR, Eigl BJ, Grivas PD, Yu EY, Li S, Kadel EE 3rd, Boyd Z, Bourgon R, Hegde PS, Mariathasan S, Thåström A, Abidoye OO, Fine GD, Bajorin DF (2017) IMvigor210 Study Group., Atezolizumab as first-line treatment in cisplatin-ineligible patients with locally advanced and metastatic urothelial carcinoma: a single-arm, multicentre, phase 2 trial. Lancet 389(10064):67–76
3. Bellmunt J, de Wit R, Vaughn DJ, Fradet Y, Lee JL, Fong L, Vogelzang NJ, Climent MA, Petrylak DP, Choueiri TK, Necchi A, Gerritsen W, Gurney H, Quinn DI, Culine S, Sternberg CN, Mai Y, Poehlein CH, Perini RF, Bajorin DF (2017) KEYNOTE-045 Investigators., Pembrolizumab as second-line therapy for advanced urothelial carcinoma. N Engl J Med 376(11):1015–1026
4. Bellmunt J, Bajorin DF (2017) Pembrolizumab for advanced urothelial carcinoma. N Engl J Med 376(23):2304
5. Motzer RJ, Escudier B, McDermott D, George S, Hammers HJ, Srinivas S, Tykodi SS, Sosman JA, Procopio G, Plimack ER, Castellano D, Choueiri TK, Gurney H, Donskov F, Bono P, Wagstaff J, Gauler TC, Ueda T, Tomita Y, Schutz FA, Kollmannsberger C, Larkin J, Ravaud A, Simon JS, Xu LA, Waxman IM, Sharma P, CheckMate 025 Investigators (2015) Nivolumab versus Everolimus in advanced renal-cell carcinoma. N Engl J Med 373(19):1803–1813
6. Borghaei H, Paz-Ares L, Horn L, Spigel DR, Steins M, Ready NE, Chow LQ, Vokes EE, Felip E, Holgado E, Barlesi F, Kohlhäufl M, Arrieta O, Burgio MA, Fayette J, Lena H, Poddubskaya E, Gerber DE, Gettinger SN, Rudin CM, Rizvi N, Crinò L, Blumenschein GR Jr, Antonia SJ, Dorange C, Harbison CT, Graf Finckenstein F, Brahmer JR (2015) Nivolumab versus docetaxel in advanced nonsquamous non-small-cell lung Cancer. N Engl J Med 373(17):1627–1639
7. Brahmer J, Reckamp KL, Baas P, Crinò L, Eberhardt WEE, Poddubskaya E, Antonia S, Pluzanski A, Vokes EE, Holgado E, Waterhouse D, Ready N, Gainor J, Arén Frontera O, Havel L, Steins M, Garassino MC, Aerts JG, Domine M, Paz-Ares L, Reck M, Baudelet C, Harbison CT, Lestini B, Spigel DR (2015) Nivolumab versus docetaxel in advanced squamous-cell non-small-cell lung Cancer. N Engl J Med 373(2):123–135
8. Ferris RL, Blumenschein G Jr, Fayette J, Guigay J, Colevas AD, Licitra L, Harrington K, Kasper S, Vokes EE, Even C, Worden F, Saba NF, Iglesias Docampo LC, Haddad R, Rordorf T, Kiyota N, Tahara M, Monga M, Lynch M, Geese WJ, Kopit J, Shaw JW, Gillison ML (2016) Nivolumab for recurrent squamous-cell carcinoma of the head and neck. N Engl J Med 375(19):1856–1867
9. Kaufman HL, Russell J, Hamid O, Bhatia S, Terheyden P, D'Angelo SP, Shih KC, Lebbé C, Linette GP, Milella M, Brownell I, Lewis KD, Lorch JH, Chin K, Mahnke L, von Heydebreck A, Cuillerot JM, Nghiem P (2016) Avelumab in patients with chemotherapy-refractory metastatic Merkel cell carcinoma: a multicentre, single-group, open-label, phase 2 trial. Lancet Oncol 17(10):1374–1385
10. Ribas A, Puzanov I, Dummer R, Schadendorf D, Hamid O, Robert C, Hodi FS, Schachter J, Pavlick AC, Lewis KD, Cranmer LD, Blank CU, O'Day SJ, Ascierto PA, Salama AKS, Margolin KA, Loquai C, Eigentler TK, Gangadhar TC, Carlino MS, Agarwala SS, Moschos SJ, Sosman JA, Goldinger SM, Shapira-Frommer R, Gonzalez R, Kirkwood JM, Wolchok JD, Eggermont A, Li XN, Zhou W, Zernhelt AM, Lis J, Ebbinghaus S, Kang SP, Daud A (2015) Pembrolizumab versus investigator-choice chemotherapy for ipilimumab-refractory melanoma (KEYNOTE-002): a randomised, controlled, phase 2 trial. Lancet Oncol 16(8):908–918
11. Younes A, Santoro A, Shipp M, Zinzani PL, Timmerman JM, Ansell S, Armand P, Fanale M, Ratanatharathorn V, Kuruvilla J, Cohen JB, Collins G, Savage KJ, Trneny M, Kato K, Farsaci B, Parker SM, Rodig S, Roemer MGM, Ligon AH, Engert A (2016) Nivolumab for classical Hodgkin's lymphoma after failure of both autologous stem-cell transplantation and brentuximab vedotin: a multicentre, multicohort, single-arm phase 2 trial. Lancet Oncol 17(9):1283–1294
12. Hodi FS, Chesney J, Pavlick AC, Robert C, Grossmann KF, McDermott DF, Linette GP, Meyer N, Giguere JK, Agarwala SS, Shaheen M, Ernstoff MS, Minor DR, Salama AK, Taylor MH, Ott PA, Horak C, Gagnier P, Jiang J, Wolchok JD, Postow MA (2016) Combined nivolumab and ipilimumab versus ipilimumab alone in patients with advanced melanoma: 2-year overall survival outcomes in a multicentre, randomised, controlled, phase 2 trial. Lancet Oncol 17(11):1558–1568
13. Hodi FS, O'Day SJ, McDermott DF, Weber RW, Sosman JA, Haanen JB, Gonzalez R, Robert C, Schadendorf D, Hassel JC, Akerley W, van den Eertwegh AJM, Lutzky J, Lorigan P, Vaubel JM, Linette GP, Hogg D, Ottensmeier CH, Lebbé C, Peschel C, Quirt I, Clark JI, Wolchok JD, Weber JS, Tian J, Yellin MJ, Nichol GM, Hoos A, Urba WJ (2010) Improved survival with ipilimumab in patients with metastatic melanoma. N Engl J Med 363(8):711–723
14. Golubchikov VA et al (2001) *Pathogenetic bases of combined use of physical factors in multimodal treatment of patients with chronic prostatitis*. Urologiia 4:15–21
15. McDermott D, Lebbé C, Hodi FS, Maio M, Weber JS, Wolchok JD, Thompson JA, Balch CM (2014) Durable benefit and the potential for long-term survival with immunotherapy in advanced melanoma. Cancer Treat Rev 40(9):1056–1064
16. Martini DJ, Hamieh L, McKay RR, Harshman LC, Brandao R, Norton CK, Steinharter JA, Krajewski KM, Gao X, Schutz FA, McGregor B, Bossé D, Lalani AKA, de Velasco G, Michaelson MD, McDermott DF, Choueiri TK (2018) Durable clinical benefit in metastatic renal cell carcinoma patients who discontinue PD-1/PD-L1 therapy for immune-related adverse events. Cancer Immunol Res 6(4):402–408

17. Puzanov I et al (2017) Managing toxicities associated with immune checkpoint inhibitors: consensus recommendations from the Society for Immunotherapy of Cancer (SITC) toxicity management working group. J Immunother Cancer 5(1):95
18. Das R, Verma R, Sznol M, Boddupalli CS, Gettinger SN, Kluger H, Callahan M, Wolchok JD, Halaban R, Dhodapkar MV, Dhodapkar KM (2015) Combination therapy with anti-CTLA-4 and anti-PD-1 leads to distinct immunologic changes in vivo. J Immunol 194(3): 950–959
19. Lee JY, Lee HT, Shin W, Chae J, Choi J, Kim SH, Lim H, Won Heo T, Park KY, Lee YJ, Ryu SE, Son JY, Lee JU, Heo YS (2016) Structural basis of checkpoint blockade by monoclonal antibodies in cancer immunotherapy. Nat Commun 7:13354
20. Nishino M, Jagannathan JP, Ramaiya NH, van den Abbeele AD (2010) Revised RECIST guideline version 1.1: what oncologists want to know and what radiologists need to know. AJR Am J Roentgenol 195(2):281–289
21. Zimmer L, Apuri S, Eroglu Z, Kottschade LA, Forschner A, Gutzmer R, Schlaak M, Heinzerling L, Krackhardt AM, Loquai C, Markovic SN, Joseph RW, Markey K, Utikal JS, Weishaupt C, Goldinger SM, Sondak VK, Zager JS, Schadendorf D, Khushalani NI (2017) Ipilimumab alone or in combination with nivolumab after progression on anti-PD-1 therapy in advanced melanoma. Eur J Cancer 75:47–55
22. Weber J, Gibney G, Kudchadkar R, Yu B, Cheng P, Martinez AJ, Kroeger J, Richards A, McCormick L, Moberg V, Cronin H, Zhao X, Schell M, Chen YA (2016) Phase I/II study of metastatic melanoma patients treated with Nivolumab who had progressed after Ipilimumab. Cancer Immunol Res 4(4):345–353
23. Martini DJ, Lalani AKA, Bossé D, Steinharter JA, Harshman LC, Hodi FS, Ott PA, Choueiri TK (2017) Response to single agent PD-1 inhibitor after progression on previous PD-1/PD-L1 inhibitors: a case series. J Immunother Cancer 5(1):66
24. Sharma P, Allison JP (2015) The future of immune checkpoint therapy. Science 348(6230):56–61
25. Teng MW et al (2015) Classifying cancers based on T-cell infiltration and PD-L1. Cancer Res 75(11):2139–2145
26. Jiang T, Zhou C, Ren S (2016) Role of IL-2 in cancer immunotherapy. Oncoimmunology 5(6):e1163462
27. Sim GC, Radvanyi L (2014) The IL-2 cytokine family in cancer immunotherapy. Cytokine Growth Factor Rev 25(4):377–390
28. West EE, Jin HT, Rasheed AU, Penaloza-MacMaster P, Ha SJ, Tan WG, Youngblood B, Freeman GJ, Smith KA, Ahmed R (2013) PD-L1 blockade synergizes with IL-2 therapy in reinvigorating exhausted T cells. J Clin Invest 123(6):2604–2615
29. Patel SP et al (2016) Sequential administration of high-dose interleukin-2 and ipilimumab in patients with metastatic melanoma. J Clin Oncol 34(15) suppl, e21041-e21041 2016
30. Naing A., Being realistic and optimistic in curing cancer. J Immunother Precis Oncol [serial online] 2018 [cited 2019 Jan 15];1:53–5. Available from: http://www.jipoonline.org/text.asp?2018/1/2/53/243749. Accessed 21 Jan 2019
31. Bilen MA, Martini DJ, Liu Y, Lewis C, Collins HH, Shabto JM, Akce M, Kissick HT, Carthon BC, Shaib WL, Alese OB, Pillai RN, Steuer CE, Wu CS, Lawson DH, Kudchadkar RR, el-Rayes BF, Master VA, Ramalingam SS, Owonikoko TK, Harvey RD (2019) The prognostic and predictive impact of inflammatory biomarkers in patients who have advanced-stage cancer treated with immunotherapy. Cancer 125(1):127–134
32. Wu X, Giobbie-Hurder A, Liao X, Connelly C, Connolly EM, Li J, Manos MP, Lawrence D, McDermott D, Severgnini M, Zhou J, Gjini E, Lako A, Lipschitz M, Pak CJ, Abdelrahman S, Rodig S, Hodi FS (2017) Angiopoietin-2 as a biomarker and target for immune checkpoint therapy. Cancer Immunol Res 5(1):17–28
33. Miao D, Margolis CA, Gao W, Voss MH, Li W, Martini DJ, Norton C, Bossé D, Wankowicz SM, Cullen D, Horak C, Wind-Rotolo M, Tracy A, Giannakis M, Hodi FS, Drake CG, Ball MW, Allaf ME, Snyder A, Hellmann MD, Ho T, Motzer RJ, Signoretti S, Kaelin WG Jr, Choueiri TK, van Allen EM (2018) Genomic correlates of response to immune checkpoint therapies in clear cell renal cell carcinoma. Science 359(6377):801–806
34. Fujii T, Naing A, Rolfo C, Hajjar J (2018) Biomarkers of response to immune checkpoint blockade in cancer treatment. Crit Rev Oncol Hematol 130:108–120
35. Udall M, Rizzo M, Kenny J, Doherty J, Dahm SA, Robbins P, Faulkner E (2018) PD-L1 diagnostic tests: a systematic literature review of scoring algorithms and test-validation metrics. Diagn Pathol 13(1):12