Efficacy and safety of anti-PD1 monotherapy or in combination with ipilimumab after BRAF/MEK inhibitors in patients with BRAF mutant metastatic melanoma

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Abstract: Background: Patients with V600BRAF mutant metastatic melanoma have higher rates of progression-free survival (PFS) and overall survival (OS) with first-line anti-PD1 (PD1]+anti-CTLA-4 (IPI) versus PD1. Whether this is also true after BRAF/MEKi therapy is unknown. We aimed to determine the efficacy and safety of PD1 versus IPI +PD1 after BRAF/MEK inhibitors (BRAF/MEKi). Methods: Patients with V600BRAF mutant metastatic melanoma treated with BRAF/MEKi who had subsequent PD1 versus IPI+PD1 at eight centers were included. The endpoints were objective response rate (ORR), PFS, OS and safety in each group. Results: Of 200 patients with V600E (75%) or non-V600E (25%) mutant metastatic melanoma treated with BRAF/MEKi (median time of treatment 7.6 months; treatment cessation due to progressive disease in 77%), 115 (57.5%) had subsequent PD1 and 85 (42.5%) had IPI+PD1. Differences in patient characteristics between PD1 and IPI+PD1 groups included, age (med. 63 vs 54 years), time between BRAF/MEKi and PD1±IPI (16 vs 4 days), Eastern Cooperative Oncology Group Performance Status (ECOG PS) of ≥1 (62% vs 44%), AJCC M1C/M1D stage (72% vs 94%) and progressing brain metastases at the start of PD1±IPI (34% vs 57%). Median follow-up from PD1±IPI start was 37.8 months (95% CI, 33.9 to 52.9). ORR was 36%; 34% with PD1 vs 39% with IPI+PD1 (p=0.5713). Median PFS was 3.4 months; 3.4 with PD1 vs 3.6 months with IPI+PD1 (p=0.6951). Median OS was 15.4 months; 14.4 for PD1 vs 20.5 months with IPI+PD1 (p=0.2603). The rate of grade 3 or 4 toxicities was higher with IPI+PD1 (31%) vs PD1 (7%). ORR, PFS and OS were numerically higher with IPI+PD1 vs PD1 across most subgroups except for females, those with <10 days between BRAF/MEKi and PD1±IPI, and those with stage III/M1A/M1B melanoma. The combination of ECOG PS=0 and absence of liver metastases identified patients with >3 years OS (area under the curve, AUC=0.74), while ECOG PS ≥1, progressing brain metastases and presence of bone metastases predicted primary progression (AUC=0.67). Conclusions: IPI+PD1 and PD1 after BRAF/MEKi have similar outcomes despite worse baseline prognostic features in the IPI+PD1 group, however, IPI+PD1 is more toxic. A combination of clinical factors can identify long-term survivors, but less accurately those with primary resistance to immunotherapy after targeted therapy. Keywords: immunotherapy; melanoma.

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Efficacy and safety of anti-PD1 monotherapy or in combination with ipilimumab after BRAF/MEK inhibitors in patients with BRAF mutant metastatic melanoma

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ABSTRACT

Background Patients with V600BRAF mutant metastatic melanoma have higher rates of progression-free survival (PFS) and overall survival (OS) with first-line anti-PD1 (PD1]+anti-CTLA-4 (IPI) versus PD1. Whether this is also true after BRAF/MEKi therapy is unknown. We aimed to determine the efficacy and safety of PD1 versus IPI +PD1 after BRAF/MEK inhibitors (BRAF/MEKi).

Methods Patients with V600BRAF mutant metastatic melanoma treated with BRAF/MEKi who had subsequent PD1 versus IPI+PD1 at eight centers were included. The endpoints were objective response rate (ORR), PFS, OS and safety in each group.

Results Of 200 patients with V600E (75%) or non-V600E (25%) mutant metastatic melanoma treated with BRAF/MEKi (median time of treatment 7.6 months; treatment cessation due to progressive disease in 77%), 115 (57.5%) had subsequent PD1 and 85 (42.5%) had IPI+PD1. Differences in patient characteristics between PD1 and IPI+PD1 groups included, age (med. 63 vs 54 years), time between BRAF/MEKi and PD1±IPI (16 vs 4 days), Eastern Cooperative Oncology Group Performance Status (ECOG PS) of ≥1 (62% vs 44%), AJCC M1C/M1D stage (72% vs 94%) and progressing brain metastases at the start of PD1±IPI (34% vs 57%). Median follow-up from PD1±IPI start was 37.8 months (95% CI, 33.9 to 52.9). ORR was 36%; 34% with PD1 vs 39% with IPI+PD1 (p=0.5713). Median PFS was 3.4 months; 3.4 with PD1 vs 3.6 months with IPI+PD1 (p=0.6951). Median OS was 15.4 months; 14.4 for PD1 vs 20.5 months with IPI+PD1 (p=0.2603). The rate of grade 3 or 4 toxicities was higher with IPI+PD1 (31%) vs PD1 (7%). ORR, PFS and OS were numerically higher with IPI+PD1 vs PD1 across most subgroups except for females, those with <10 days between BRAF/MEKi and PD1±IPI, and those with stage III/M1A/M1B melanoma. The combination of ECOG PS=0 and absence of liver metastases identified patients with >3 years OS (area under the curve, AUC=0.74), while ECOG PS ≥1, progressing brain metastases and presence of bone metastases predicted primary progression (AUC=0.67).

Conclusions IPI+PD1 and PD1 after BRAF/MEKi have similar outcomes despite worse baseline prognostic features in the IPI+PD1 group, however, IPI+PD1 is more toxic. A combination of clinical factors can identify long-term survivors, but less accurately those with primary resistance to immunotherapy after targeted therapy.

KEY MESSAGES

- ⇒ In first-line setting, the benefit of combination immunotherapy (anti-PD1 + anti-CTLA-4) over anti-PD1 monotherapy appears significantly greater in patients with BRAF mutant than BRAF WT melanoma.
- ⇒ After BRAF/MEKi, combination anti-PD1 + anti-CTLA-4 and monotherapy anti-PD-1 have similar objective response rate, progression-free survival and overall survival (OS), although combination therapy has a higher rate of toxicity.
- ⇒ A combination of clinical variables can accurately identify patients with long-term survival (>3 years OS), and is less accurate at predicting progression with anti-PD1 ± anti-CTLA-4 after BRAF/MEKi.
- ⇒ This study will help us to better identify those who do not benefit from immune checkpoint inhibitors after BRAF/MEKi, in order to offer novel treatment strategies as part of clinical trials.

INTRODUCTION

Immune checkpoint inhibitors, anti-PD-(L)1 monotherapy or in combination with anti-CTLA-4, have significantly improved the outcome of patients with advanced melanoma,[1–3] non-small cell lung cancer (NSCLC),[4–7] renal cancer,[8] head and neck cancer[9 10] and hepatocellular carcinoma,[11] among others. This significant clinical benefit has been observed not only in the metastatic

setting, but also in earlier stages, including in the adjuvant[12–14] and neoadjuvant[15 16] settings.

In advanced melanoma, first-line therapy with the combination of ipilimumab (anti-CTLA-4; IPI) and nivolumab (anti-PD-1; PD1) has shown a numerically superior objective response rate (ORR), progression-free survival (PFS) and overall survival (OS) compared with nivolumab monotherapy.[3] Notably, in patients with BRAF mutant melanoma, the benefit of combination immunotherapy over nivolumab monotherapy appears significantly greater than those with BRAF WT melanoma.[3] Nevertheless, whether the significant superiority of combination versus PD1 monotherapy in patients with BRAF mutant melanoma is also seen after BRAF-targeted therapy (BRAF/MEKi) is yet to be determined.

Previous studies have reported lower efficacy with immune checkpoint therapy when given as second-line treatment or later,[17–20] likely due to the poorer performance status of the patient and larger volume of disease, as well as biological changes induced by previous treatments.[21] This was confirmed by two randomized studies, SECOMBIT and DREAMseq, which have shown better PFS and OS with IPI+PD1 as first-line treatment, followed by BRAF/MEKi compared with the inverse order,[22 23] even though in the latter study 24 of the 44 (55%) patients who progressed on first-line IPI+PD1 did not receive targeted therapy second line.

In this study, we sought to determine the efficacy and safety of PD1 monotherapy or in combination with IPI after BRAF/MEKi. We also aimed to identify the clinical factors associated with extremes of outcomes with treatment (progressive disease as best response vs survival ≥3 years) with PD1 alone or in combination with IPI after BRAF/MEKi.

METHODS

Study design and participants

We conducted a multicenter retrospective study which included patients ≥18 years of age with metastatic melanoma (unresectable stage III and IV) treated with BRAF and MEK inhibitors who were subsequently treated with PD1 monotherapy or in combination with IPI at eight major melanoma centers (Australia, Europe and USA).

Procedures

All patients included in this study had treatment with combination BRAF and MEK inhibitors (vemurafenib and cobimetinib; dabrafenib and trametinib; encorafenib and binimetinib), and were then treated with either PD1 monotherapy (nivolumab or pembrolizumab) or PD1 combined with IPI. The choice between PD1 and IPI+PD1 was determined by their treating physician, based on the availability of the therapies and clinical factors.

Patient demographics (age, gender), disease characteristics (primary melanoma site, histological subtype, thickness, presence/absence of ulceration, mitosis, mutational status, sites of metastases, Eastern Cooperative Oncology Group (ECOG) performance status (PS), all prior systemic therapie(s)), and baseline blood parameters (full blood count and lactate dehydrogenase (LDH)) at the time of commencing BRAF/MEKi, and at the time of commencing PD1±IPI, were collected and analyzed.

Efficacy and safety assessments

Tumor response to BRAF/MEKi treatment and to PD1±IPI therapy was assessed with regular scans as per standard of care and according to each institution's protocols (in general 3-monthly CT or CT/PET imaging), and was determined based on Response Evaluation Criteria in Solid Tumors (RECIST) V.1.1.[24]

Safety assessments were made continuously and for all systemic therapies from the commencement of BRAF/MEKi treatment until the data cut-off. The severity of treatment-related adverse events was graded according to the Common Terminology Criteria for Adverse Events V.4.03.[25]

Endpoints

The endpoints of this study were: ORR, defined as the proportion of patients who have a partial or complete response to treatment; progression-free survival (PFS), defined as time from starting PD1±IPI to disease progression or death or last follow-up; OS, defined as time from starting PD1±IPI to death or last follow-up; and safety, defined as proportion of patients with immune-related adverse events with PD1 monotherapy versus PD1 combined with IPI therapy groups.

Statistical methods

The primary characteristics of the patients were summarized through frequencies and proportions for categorical variables, and median, IQR and range for continuous variables. Differences in baseline characteristics between the two treatment groups was assessed using Wilcoxon rank-sum test for continuous variables and Pearson's χ^2 test for categorical variables. Survival curves were estimated using Kaplan-Meier method. Log rank test was used to test the difference between groups. Multivariable analysis was performed, including all the significant ($p<0.05$) variables from the univariable analysis, to identify clinical factors associated with ORR, PFS and OS.

A subgroup analysis was performed to compare treatment differences within clinically relevant prespecified subgroups. OR for ORR and HR for OS and PFS were calculated using univariable logistic and Cox regression, respectively. All subgroup results displayed in forest plots.

Clinical predictive models were generated to identify: (1) the subgroup of patients with progressive disease as best response to PD1±IPI; (2) the subgroup of patient with long-term survival (>3 years OS). Predictors in the multivariable models were automatically selected using the Backward elimination method from an initial full model that contained variables with a $p\leq0.20$ in the univariate analysis. The predictive performance of the clinical model was assessed using discrimination index

which was estimated using Harrell's C-statistic, also known as the area under the curve (AUC), reflecting how well the model identified patients with an outcome event.

All the statistical analyses were carried out in SAS V.9.4 (SAS Institute) and R V.3.6.3 (R Foundation for Statistical Computing, Vienna, Austria).

RESULTS

Patient characteristics

Two hundred patients with advanced melanoma treated with BRAF/MEKi who had subsequent anti-PD-1 monotherapy (n=115; 57.5%) or in combination with anti-CTLA-4 (n=85; 42.5%) were included in this study. Differences in patient characteristics for patients treated with PD1 monotherapy or in combination with IPI included ECOG PS of ≥1 (47% vs 29%: p=0.0181) and AJCC M1C/M1D stage at the start of BRAF/MEKi (63% vs 86%; p=0.0008), time between BRAF/MEKi and PD1±IPI (16 vs 4 days; p=0.0002), and age (med. 63 vs 54 years (years); p=0.0002), ECOG PS of ≥1 (62% vs 44%, p=0.0232), AJCC M1C/M1D stage (72% vs 94%; p=0.0002) and progressing brain metastases at the start of PD1±IPI (34% vs 57%; p=0.0014) (table 1). Sites of metastases at the start of PD1±IPI are presented in online supplemental table 1.

Efficacy

With a median follow-up from commencement of PD1±IPI of 37.8 months (95% CI 33.9 to 52.9), there was no significant difference in any of the efficacy endpoints between both treatment groups. The ORR was 36% in the entire cohort; 34% with PD1 compared with 39% with PD1 +IPI (p=0.5713) (figure 1A). PFS rate at 1 year and 2 years for the entire cohort was 32% (95% CI, 26% to 39%) and 24% (95% CI, 19% to 31%) (32% [95% CI, 25% to 42%] and 24% [95% CI, 17% to 33%] with PD1 vs 32% [95% CI, 24% to 44%] and 25% [95% CI, 17% to 37%] with PD1 +IPI), with a median PFS of 3.4 months (med PFS 3.4 for PD1 vs 3.6 mo for PD1 +IPI, p=0.6951) (figure 1B). OS rate at 1 year and 2 years for the entire cohort was 55% (95% CI, 48% to 70%) and 45% (95% CI, 38% to 53%) (53% [95% CI, 44% to 63%] and 43% [95% CI, 35% to 54%] with PD1 vs 58% [95% CI, 48% to 70%] and 47% [95% CI, 37% to 60%] with PD1 +IPI), with a median OS of 15.4 months (med OS 14.4 for anti-PD-1 vs 20.5 mo for PD1 +IPI, p=0.2603) (figure 1C).

On multivariable analysis, patients with ECOG PS 0 at the start of PD1±IPI had a higher objective response to PD1±IPI after BRAF/MEKi (online supplemental table 2). Features associated with longer PFS were ECOG PS 0 and absence of liver metastases at the start of PD1±IPI (online supplemental table 3), and these two variables in addition to elective BRAF/MEKi cessation were associated with longer OS (online supplemental table 4). Duration of prior BRAF/MEKi treatment, best RECIST response to BRAF/MEKi, interval between BRAF/MEKi and PD1±IPI, LDH level, and type of treatment (PD1 vs IPI +PD1), were not associated with ORR, PFS or OS.

On subgroup analysis, ORR, PFS and OS were numerically higher with anti-PD-1+anti-CTLA-4 compared with anti-PD1 monotherapy across most of the subgroups except for females (for ORR, PFS and OS), interval between BRAF/MEKi and PD1±IPI ≤ 10 days (for ORR and PFS) and stage III/M1A/M1B (for ORR and OS) (figure 2 and online supplemental figure 1). OS was significantly longer with PD1 +IPI compared with PD1 for patients with non-V600E mutations (1 year OS rate 80% vs 52%), but no difference was observed for V600E mutations (1 year OS rate 53% vs 53%) (online supplemental figure 2). In contrast, PFS was similar for V600E and non-V600E mutant with both treatment types (online supplemental figure 3). Of note, a higher proportion of patients with non-BRAF V600E mutant melanoma had subsequent systemic treatment (32%) after PD1±IPI compared with those with BRAF V600E mutant melanoma (22%). Within the subgroup of patients who had subsequent treatment, there was a higher proportion of patients rechallenged with BRAF-targeted therapy in the patients with non-BRAF V600E mutant melanoma (69%) than in patients with BRAF V600E mutant melanoma (56%).

In our cohort, 99 patients (50%) had brain metastases at the start of PD1 (n=44, 38%; of those, 37 patients (84%) had progressing brain metastases) or IPI +PD1 (n=55, 65%; of those, 44 patients (80%) had progressing brain metastases). Sixteen patients had corticosteroids for symptomatic brain metastases; 8 (22% of the patients with progressing brain metastases) in the PD1 treatment group and 8 (18% of the patients with progressing brain metastases) in the IPI +PD1 group. Similarly to the entire cohort, ORR (35% vs 27%; p=0.5157), PFS (med PFS, 2.8 vs 2.1 mo; p=0.2100) and OS (med OS; 14.1 vs 9.7 mo; p=0.2200) were numerically but insignificantly superior with IPI +PD1 compared with PD1 monotherapy (online supplemental figures 4 and 5).

Clinical predictors of non-responders and long-term survivors with PD1+/-IPI after BRAF/MEKi

We then built clinical models to predict outcome to PD1±IPI in two distinct and clinically relevant subgroups of patients: (1) patients with progressive disease as best response with PD1±IPI after BRAF/MEKi (116/200, 58%); (2) patients with long-term OS from the date of commencement of PD1±IPI (OS ≥3 years; 22%). The clinical model identified ECOG PS ≥1 at PD1±IPI start, progressing brain metastases, and the presence of bone metastases to predict progressors (AUC=0.67 (95% CI, 0.59 to 0.76)) (figure 3A; online supplemental table 5). In contrast, the model to predict long-term survivors identified patients with ECOG PS of 0 and absence of liver metastases at PD1±IPI start (AUC=0.74 (95% CI 0.66 to 0.82)) (figure 3B; online supplemental table 6).

Table 1 Baseline patient characteristics, by treatment type

Characteristics	Anti-PD1 (n=115)	Anti-PD1+anti-CTLA-4 (n=85)	P value
Sex (n, %)			0.9325
Male	78 (68)	59 (69)	
Female	37 (32)	26 (31)	
BRAF mutation (n, %)			0.0575
V600E	80 (70)	70 (82)	
Non-V600E	35 (30)	15 (18)	
ECOG PS at BRAF/MEKi start (n, %)			**0.0181**
0	61 (53)	60 (71)	
≥1	54 (47)	25 (29)	
AJCC staging v8 at BRAF/MEKi start (n, %)			**0.0008**
III/M1a/M1b	42 (37)	12 (14)	
M1c/M1d	73 (63)	73 (86)	
LDH at BRAF/MEKi start (n, %)*			0.5654
Normal	72 (65)	49 (60)	
Elevated	39 (35)	33 (40)	
Duration of BRAF/MEKi treatment (months)			0.5286
Median (range)	8.2 (0.0, 67.1)	6.0 (0.23, 80.3)	
Q1 – Q3	3.9–13.8	3.5–13.8	
Reason for BRAF/MEKi cessation (n, %)			0.7468
Progressive disease	90 (78)	64 (75)	
Other	25 (22)	21 (25)	
Interval between BRAF/MEKi and PD1±IPI (days)			**0.0002**
Median (range)	16 (-5, 861)	4 (0, 2038)	
Q1 - Q3	4–35	1–22	
No of progressive lesions while/after BRAF/MEKi (n, %)			0.1834
1	26 (26)	23 (33)	
2	24 (24%)	9 (13%)	
≥3	51 (50%)	38 (54%)	
Progressing brain metastases while/after BRAF/MEKi† (n,%)			**0.0014**
No	71 (66)	33 (43)	
Yes	37 (34)	44 (57)	
Progressing liver metastases while/after BRAF/MEKi‡ (n, %)			0.5183
No	95 (88)	65 (84)	
Yes	13 (12)	12 (16)	
Any steroids to control symptoms due to progression‡ (n, %)			0.5973
No	100 (93)	69 (90)	
Yes	8 (7)	8 (10)	
Age at PD1±IPI start, years			**0.0002**
Median (range)	63.4 (22.0, 91.1)	54.0 (19.8, 80.5)	
Q1 – Q3	54.0–71.8	44.0–67.1	
ECOG PS‡ at PD1±IPI start (n, %)			**0.0232**

Continued

Table 1 Continued

Characteristics	Anti-PD1 (n=115)	Anti-PD1+anti-CTLA-4 (n=85)	P value
0	43 (38)	43 (56)	
≥1	70 (62)	34 (44)	
AJCC staging v8 at PD1±IPI start (n, %)			**0.0002**
III/M1a/M1b	32 (28)	5 (6)	
M1c/M1d	83 (72)	80 (94)	
LDH§ at PD1±IPI start (n, %)			0.4322
Normal	59 (53)	39 (46)	
Elevated	52 (47)	45 (54)	

*LDH, missing values in the anti-PD1 cohort (n=4), missing in the anti-PD1+anti-CTLA-4 cohort(n=3).
†A subset of patients did not have response to BRAF/MEKi assessed due to a rapid switch to PD1 +/- IPI:7 patients in the PD1 treatment group and 8 patients in the IPI+PD1 treatment group.
‡ECOG PS, missing values in the anti-PD1 cohort (n=2), missing values in the anti-PD1+antiCTLA-4 cohort (n=8).
§LDH, missing values in the anti-PD1 cohort (n=4), missing in the anti-PD1+anti-CTLA-4 cohort (n=1).
AJCC, American Joint Committee on Cancer; ECOG PS, Eastern Cooperative Oncology Group performance status; IPI, ipilimumab; LDH, lactate dehydrogenase.

Safety

As expected, the combination of PD1 and IPI was associated with a significantly higher frequency of ≥grade 3 adverse events compared with PD1 monotherapy (31% vs 7%) (table 2). Diarrhea/colitis was the most common (15% with PD1+IPI vs 0% with anti-PD-1), followed by hepatitis (11% with PD1+IPI vs 3% with PD1). When considering immune-related adverse events of any grade, skin toxicity (except vitiligo) was the most common (24% with PD1+IPI vs 14% with PD1), followed by diarrhea/colitis (26% with PD1+IPI vs 8% with PD1) (online supplemental table 7). Fever, which can be associated with both targeted therapy and immunotherapy, was only reported in 4% of the patients (4% with PD1 and 4% with IPI+PD1). There were no unexpected toxicities and there were no deaths due to treatment-related toxicities in this cohort of patients.

DISCUSSION

In this large multicenter retrospective study, the patients treated with PD1+IPI had a similar ORR, PFS and OS to those treated with PD1 monotherapy after BRAF/MEKi. Notably, those who received PD1 +IPI had poorer prognostic features baseline features. There was a trend favoring IPI+PD1 over PD1 for all outcomes across most subgroups of patients, except females, those with an interval between BRAF/MEKi and PD1±IPI ≤10 days, and those with stage III/M1A/M1B melanoma. The combination of ECOG PS≥1 or worse, progressing brain metastases and presence of bone metastases identified the progressors (AUC=0.67), while the combination of ECOG PS 0 and absence of liver metastases identified the long-term survivors (AUC=0.74) to PD1±IPI after BRAF/MEKi. As expected, PD1+IPI had a significantly higher rate of adverse events compared with PD1 monotherapy; nearly a third of patients treated with PD1 +IPI had high-grade toxicity, most commonly colitis and hepatitis.

As shown in previous studies[17–20] and more recently in DREAMseq,[23] immune checkpoint inhibitors lose efficacy if given in the second line setting or later. When given after BRAF/MEKi, immune checkpoints' lower efficacy can be explained by clinical factors, such as larger volume of disease and the poorer performance status of the patient, but also due to changes in the microenvironment induced by BRAF/MEKi[26–29] including a lack of functional CD103+dendritic cells, and consequently an ineffective T cell response.[21] Moreover, SECOMBIT and DREAMseq trials have shown that the combination of IPI+PD1 followed by targeted therapy was associated with a significantly improved survival when compared with the reverse order, that is, targeted therapy followed by IPI +PD1[22 23], and therefore, BRAF/MEKi as initial therapy is likely to become less frequent in the metastatic setting, although it may still be used in the adjuvant setting. Whether melanoma that recurs on/after adjuvant BRAF/MEKi is biologically different to that which progresses (or received) 'palliative' BRAF/MEKi is unknown, as is the response to subsequent immunotherapy. In our study, PD1+IPI was associated with numerically superior ORR, PFS and OS compared PD1, however, this difference was not statistically significant and is much smaller than what was reported for BRAF mutant patients treated with IPI+PD1 vs PD1 in the first line.[3] Similarly, even though we saw a numerical superiority in all efficacy endpoints with IPI+PD1 versus PD1 within the subgroup of patients with brain metastases, this was smaller compared with what was reported in the ABC trial.[30] Of note, our cohort has a higher proportion of patients with brain metastases compared with what is commonly seen in a cohort of treatment naïve advanced melanoma patients, suggesting

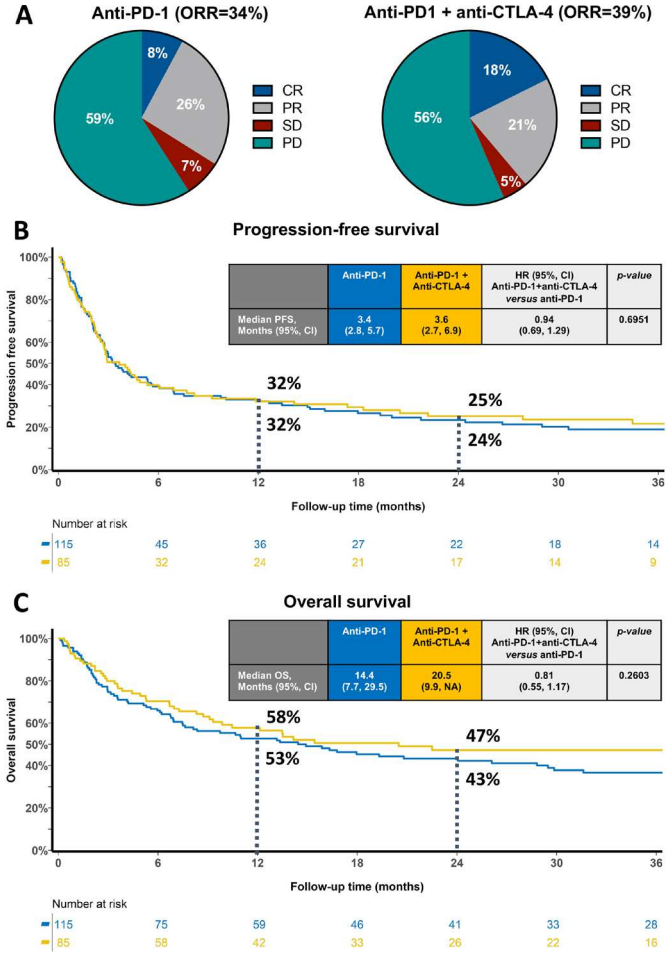


Figure 1 Efficacy of PD1 monotherapy or PD1+IPI after BRAF/MEKi. (A) Best objective response (CR, complete response; PR, partial response; SD, stable disease; PD, progressive disease) with PD1 monotherapy (left plot) or PD1+IPI (right plot). Kaplan-Meier curves showing (B) progression-free survival and (C) overall survival with PD1 monotherapy (blue line) or PD1+IPI (yellow line). Patients at risk at baseline, 6, 12, 18, 24, 30 and 36 months are presented in (B) and (C). IPI, ipilimumab.

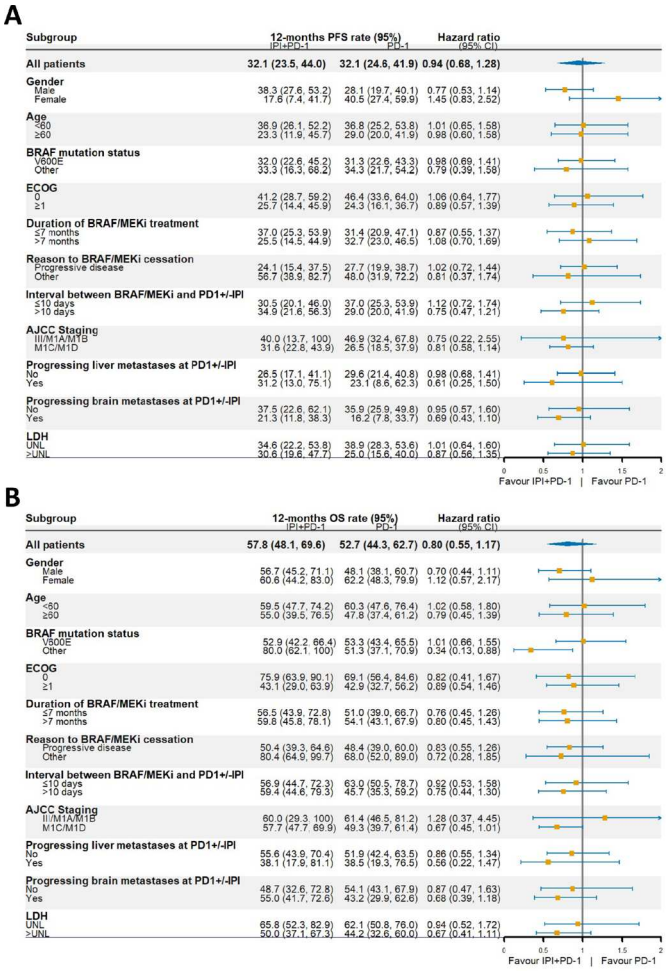


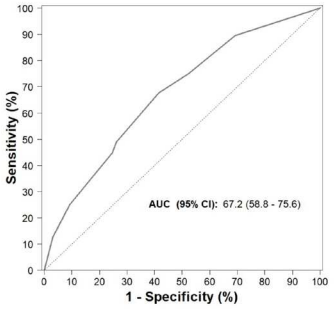
Figure 2 Subgroup analysis of progression-free survival and overall survival with PD1±IPI after BRAF/MEKi. Forest plot showing the subgroup analysis for the 12 months (A) PFS and (B) OS. AJCC, American Joint Committee on Cancer; ECOG, Eastern Cooperative Oncology Group; IPI, ipilimumab; LDH, lactate dehydrogenase; OS, overall survival; PFS, progression-free survival.

a different cohort of patients who receive BRAF/MEKi first line and a different progression pattern on BRAF/MEKi. In addition, the patients treated with PD1+IPI in our study had poorer baseline prognostic features compared with those treated with PD-1 monotherapy, including more progressing brain metastases and higher M substage. Such treatment selection bias, small numbers and retrospective nature of the data likely influenced these results; however, it is unlikely that a randomized trial in this setting will ever be conducted.

Patient characteristics at BRAF/MEKi start, duration and best response with BRAF/MEKi treatment were not significantly associated to clinical outcomes on multivariable analysis. The only factors significantly associated with clinical outcomes were well-known prognostic features, including ECOG PS[31 32] and presence/absence liver metastases,[33 34] as well as reason for ceasing BRAF/MEKi (progressive disease vs elective). These data have implications in clinical practice, suggesting a rapid switch to immunotherapy, rather than waiting until disease progression on BRAF/MEKi. The LDH level, which have been associated with poor clinical outcomes in treatment naïve melanoma patients, was not associated with ORR, PFS or OS in this study. Previous treatment with BRAF/MEKi might account for this finding, but the biology behind it is yet to be clarified. In three subgroups of patients, including females, those with melanoma stage III/M1A/M1B and those with an interval between BRAF/MEKi and PD1±IPI ≤10 days there was no trend favoring combination PD1+IPI, such that PD1 monotherapy may be more appropriate for these patients. From these three variables, only staging was included in the subgroup analysis of the CheckMate-067, however an opposite trend was seen in the first-line setting.[35] The only subgroup associated with significant benefit in OS from PD1 +IPI over PD1 monotherapy in this study was those patients with BRAF non-V600E mutation, while there was no statistical difference between both treatments in patients with a BRAF

A

	OR (95% CI)	p-value
Progressing brain metastases		0.0787
No	1	
Yes	1.83 (0.93, 3.60)	
ECOG at PD1+/- IPI start		0.0086
0	1	
>=1	2.42 (1.25, 4.70)	
Bone metastases		0.0494
No	1	
Yes	2.13 (1.00, 4.54)	



B

	OR (95% CI)	p-value
ECOG at PD1+/- IPI start		<0.0001
0	1	
>=1	0.19 (0.09, 0.44)	
Liver metastases		0.0005
No	1	
Yes	0.16 (0.06, 0.45)	

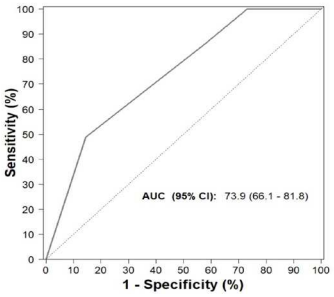


Figure 3 Clinical predictive model and respective ROC curves of non-responders (A) and long-term survivors (B) with PD1±IPI after BRAF/MEKi. AUC, area under the curve; ECOG, Eastern Cooperative Oncology Group; IPI, ipilimumab. ROC, receiver operating characteristic.

V600E mutation. No statistical difference was observed in PFS between the two treatments in patient with BRAF V600E versus non-V600E mutations. The higher proportion of patients with subsequent treatments, in particular the higher percentage of patients rechallenged with BRAF-targeted therapy within those with BRAF non-V600E mutant melanomas compared with those with BRAF V600E mutant melanomas, might contribute to this difference in OS. One could also hypothesize that treatment with BRAF/MEKi may alter the tumor microenvironment, such that the advantage IPI has shown in first-line studies is lost. Nevertheless, due to the retrospective nature of these data and small numbers in some of the subgroups, these findings should be interpreted with caution and require prospective validation.

Table 2 Proportion of ≥grade 3 (G3) immune-related adverse events, by treatment type

High-grade (≥G3) immune-related adverse events	Anti-PD1 (n=115)	Anti-PD1+anti-CTLA-4 (n=85)
Any (n, %)	8 (7)	26 (31)*
Diarrhea/colitis (n, %)	0	13 (15)
Hepatitis (n, %)	3 (3)	9 (11)
Skin (n, %)	0	1 (1)
Hypophysitis, thyroiditis (n, %)	0	1 (1)
Pneumonitis	1 (1)	0
Nephritis (n, %)	0	1 (1)
Fever (n, %)	0	0
Elevated amylase/lipase (n, %)	2 (2)	0
Others (n, %)	2 (2)†	5 (6)‡

*Four patients had >1 G3 immune-related adverse events: 1 patient had G3 colitis and G3 hepatitis; one patient had G3 hepatitis and G3 thyroiditis; one patient had G3 hepatitis and T1MD; and one patient had hepatitis and pericarditis.
†Inflammatory syndrome (cytokine release) and encephalitis.
‡T1MD, Myasthenia Gravis, peripheral neuropathy, pericarditis and immune thrombocytopenic purpura.

Our data indicate that more than one-third of patients will respond to PD1±anti IPI after BRAF/MEKi. Therefore, a key step toward improving outcomes in patients with advanced melanoma is to identify and separate the long-term survivors from those who progress with PD1±IPI after BRAF/MEKi are in urgent need of a novel therapeutic approaches, for example, clinical trials. Predictive clinical models perform better at identifying long-term survivors than progressors to anti-PD1±IPI after BRAF/MEKi, thus selecting those in most need of a novel therapy remains a challenge. This challenge is likely due to the heterogeneous nature of resistance, with multiple and distinct mechanisms.[36 37] Nevertheless, similar to previous studies across different cancers,[31–34] ECOG PS 0 and the absence of liver metastases, predicted long-term survivors to PD1±IPI in our cohort. In contrast, the presence of bone metastases, progressing brain metastases and ECOG PS ≥1, were the best predictors of progressors with PD1±IPI after BRAF/MEKi. The presence of bone metastases has been previously shown to be associated with poor outcome in patients treated with immunotherapy in NSCLC,[38 39] but to our knowledge, this is the first study showing bone metastases is associated with poor response to immune checkpoint inhibitors, after BRAF/MEKi, in patients with melanoma. Further research is needed to better identify those who do not benefit from immune checkpoint inhibitors after BRAF/MEKi, in order to offer novel treatment strategies as part of clinical trials.

As expected, the combination of PD1 and IPI was more toxic compared with PD1 monotherapy, with the most common adverse events being colitis/diarrhea and hepatitis, and with no unexpected toxicities. However, the proportion of patients experiencing immune-related adverse events, any grade or high grade, was lower in both treatment groups in our cohort compared with first-line treatment,[35] probably due to patient selection for second-line treatment.

In this retrospective analysis, the combination of PD1+IPI appears similarly active to PD1 after BRAF/MEKi, however, is more toxic. A combination of clinical variables can accurately identify patients with long-term survival (>3 years OS), and is less accurate at predicting progression with PD1±IPI after BRAF/MEKi.

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