Anti-programmed-death-receptor-1 treatment with pembrolizumab in ipilimumab-refractory advanced melanoma: a randomised dose-comparison cohort of a phase 1 trial

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Summary

Background **The anti-programmed-death-receptor-1 (PD-1) antibody pembrolizumab has shown potent antitumour activity at different doses and schedules in patients with melanoma. We compared the efficacy and safety of pembrolizumab at doses of 2 mg/kg and 10 mg/kg every 3 weeks in patients with ipilimumab-refractory advanced melanoma.**

Methods **In an open-label, international, multicentre expansion cohort of a phase 1 trial, patients (aged ≥18 years) with advanced melanoma whose disease had progressed after at least two ipilimumab doses were randomly assigned with a computer-generated allocation schedule (1:1 final ratio) to intravenous pembrolizumab at 2 mg/kg every 3 weeks or 10 mg/kg every 3 weeks until disease progression, intolerable toxicity, or consent withdrawal. Primary endpoint was overall response rate (ORR) assessed with the Response Evaluation Criteria In Solid Tumors (RECIST, version 1.1) by independent central review. Analysis was done on the full-analysis set (all treated patients with measurable disease at baseline). This study is registered with ClinicalTrials.gov, number NCT01295827.**

Findings **173 patients received pembrolizumab 2 mg/kg (n=89) or 10 mg/kg (n=84). Median follow-up duration was 8 months. ORR was 26% at both doses—21 of 81 patients in the 2 mg/kg group and 20 of 76 in the 10 mg/kg group (difference 0%, 95% CI –14 to 13; p=0·96). Treatment was well tolerated, with similar safety profiles in the 2 mg/kg and 10 mg/kg groups and no drug-related deaths. The most common drug-related adverse events of any grade in the 2 mg/kg and 10 mg/kg groups were fatigue (29 [33%] *vs* 31 [37%]), pruritus (23 [26%] *vs* 16 [19%]), and rash (16 [18%] *vs* 15 [18%]). Grade 3 fatigue, reported in five (3%) patients in the 2 mg/kg pembrolizumab group, was the only drug-related grade 3 to 4 adverse event reported in more than one patient.**

Interpretation **The results suggest that pembrolizumab at a dose of 2 mg/kg or 10 mg/kg every 3 weeks might be an effective treatment in patients for whom there are few effective treatment options.**

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Introduction

Despite recent advances such as the anti-cytotoxic T-lymphocyte-associated-antigen-4 (CTLA-4) antibody ipilimumab and the mitogen-activated protein kinase pathway inhibitors vemurafenib, dabrafenib, and trametinib, melanoma treatment remains a challenge because there are few effective treatment options for patients who relapse or do not respond to these therapies. Response has been reported in about 25% and 50% of patients treated with MEK and BRAF inhibitors, respectively, and these agents are associated with a survival advantage compared with chemotherapy.[1–5] However, their use is restricted to the roughly 50% of patients with *BRAF* V600-mutant melanoma,[6] and the median duration of response is about 6–7 months.[2,3] Combination therapy with BRAF and MEK inhibitors results in an objective response rate of 76% and extends progression-free survival (PFS), but most patients develop resistance to these inhibitors.[7] Thus, there is an urgent need to develop effective treatment options for patients who progress on these agents. The distinct mechanism of action of anti-programmed-death-receptor-1 (PD-1) antibodies, which increase tumour cell killing peripherally by cytotoxic T lymphocytes,[8] might have activity in patients with ipilimumab-refractory melanoma.

PD-1 is expressed on antigen-stimulated T cells and induces downstream signalling that inhibits T-cell proliferation, cytokine release, and cytotoxicity.[8–10] Many tumours, including melanoma, suppress cytotoxic T-cell activity[11,12] by expressing PD-1 ligand (PD-L1) on the cell surface. Anti-PD-1 and PD-L1 antibodies can reverse this T-cell suppression and induce long-lasting antitumour responses in patients with advanced solid tumours, including advanced melanoma.[13–20]

Pembrolizumab (MK-3475, previously known as lambrolizumab) is a highly selective, humanised

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monoclonal IgG4-kappa isotype antibody against PD-1 that has shown robust clinical activity with an acceptable safety profile. In 135 patients with advanced melanoma who were enrolled in non-randomised cohorts of the large, phase 1 study KEYNOTE-001 (ClinicalTrials.gov, number NCT01295827), pembrolizumab resulted in long-lasting objective responses as assessed per Response Evaluation Criteria In Solid Tumors (RECIST, version 1.1) in 31–51% of patients treated with doses ranging from 2 mg/kg every 3 weeks to 10 mg/kg every 2 weeks, and 81% of patients survived for at least 1 year after starting treatment.[15,16] Of note, promising antitumour activity and acceptable safety were noted in the 48 patients previously treated with ipilimumab; however, the sample size was insufficient to assess clinical benefit accurately, and lack of randomisation between doses and schedules restricted the ability to assess a dose-response relation.

In this study, an expansion cohort of KEYNOTE-001, we assessed further the clinical benefit of pembrolizumab in patients whose advanced melanoma progressed after ipilimumab and who were previously treated with a BRAF or MEK inhibitor, or both, a clinical scenario for which there is no effective treatment.

Methods

Study design and patients

This trial is a multicentre, international (Australia, Canada, France, and the USA), randomised expansion cohort of the phase 1 KEYNOTE-001 study. Eligible patients were aged 18 years or older, had progressive, measurable, unresectable melanoma that was previously treated with at least two doses of ipilimumab 3 mg/kg or higher administered every 3 weeks; had confirmed disease progression using immune-related response criteria[21] within 24 weeks of the last dose of ipilimumab; and had adequate performance status and organ function. Resolution of all ipilimumab-related adverse events to grade 0 to 1 was required. Patients with a history of grade 4 immune-related adverse events requiring steroid treatment or grade 3 immune-related adverse events requiring steroid treatment with prednisone at doses greater than 10 mg/day or equivalent for more than 12 weeks were excluded. Previous treatment with approved BRAF or MEK inhibitors, or both, was required for patients with *BRAF*-mutant melanoma. There were no limitations on the number of previous treatments. Patients were not screened for brain metastases at baseline, and those with previously treated brain metastases were eligible if there was no evidence of CNS progression for 8 weeks. Major exclusion criteria were previous treatment with a PD-1 or PD-L1 blocking agent, current systemic immunosuppressive therapy, and active infection or autoimmune disease.

This study was done in accordance with the protocol, good clinical practice standards, and the Declaration of Helsinki. The protocol and its amendments were approved by the relevant institutional review boards or ethics committees of the participating institutions. All patients provided written informed consent.

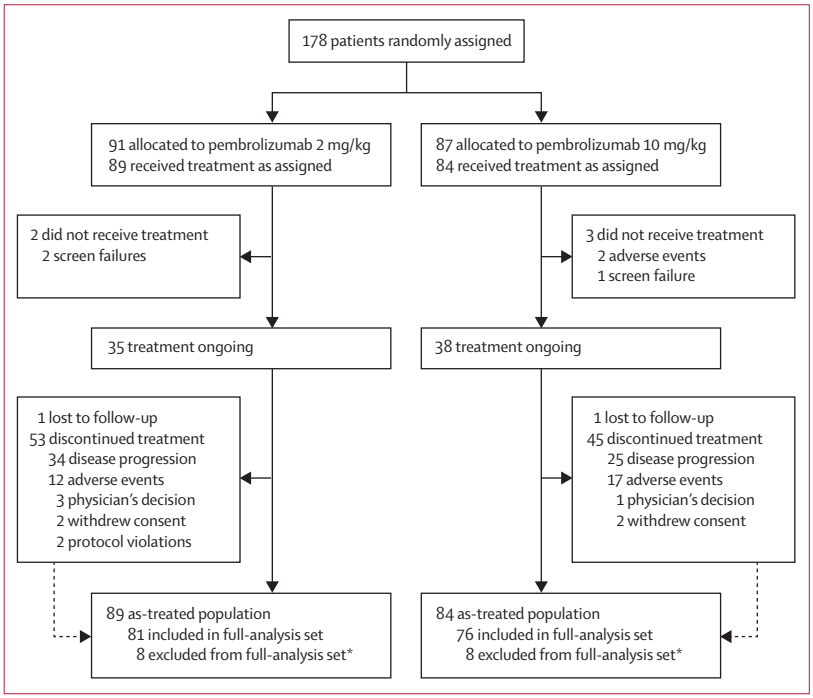

Figure 1: **Trial profile**
*Did not have measurable disease at baseline as per independent central review.

Randomisation and masking

This study was open label. The first 60 patients were randomly assigned in a 2:1 ratio to treatment with pembrolizumab at 2 mg/kg every 3 weeks or 10 mg/kg every 3 weeks administered intravenously over 30 min. The sample size was later increased by 100 patients. To achieve a final randomisation ratio of 1:1, the subsequent 100 patients were randomly assigned in a 2:3 ratio to pembrolizumab 2 mg/kg or 10 mg/kg. Any additional patients were randomly assigned in a 1:1 ratio. Treatment assignment was done by the study funder based on a computer-generated allocation schedule. After the patient completed all baseline procedures and met all eligibility requirements, the treating centre completed a registration form for the patient that was faxed or e-mailed to the study funder. The funder then assigned a unique allocation number to the patient and returned this information to the centre.

Procedures

The first dose of pembrolizumab was given within 7 days of assignment of the allocation number and continued until confirmed disease progression, intolerable toxicity, or consent withdrawal.

The primary study endpoint was the overall response rate (ORR) according to RECIST (version 1.1[22]) as assessed

by independent central review. ORR was also assessed according to immune-related response criteria[21] by the investigator (appendix). The definition of ORR was the percentage of patients who achieved a best overall response of confirmed complete or partial response.

Key secondary endpoints included response duration (ie, time from best overall response of partial or complete response to time of first documented disease progression), PFS (ie, time from treatment initiation to time of first documented disease progression or death due to any cause), and overall survival (ie, time from treatment initiation to death due to any cause). Patients without an event were censored at the time of the last tumour assessment of non-progressive disease or, for survival, date they were last known to be alive. Tumour response was assessed every 12 weeks after pembrolizumab initiation, with patients managed according to immune-related response criteria per investigator (appendix).

Adverse events were assessed continuously and were graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events (version 4.0). Investigators indicated whether an adverse event was potentially immune-related. The funder provided investigators with a list of preferred terms with specified grade criteria called adverse events of special interest (appendix). The list of adverse events of special interest was intentionally broad to gather all informative data about potentially immune-related adverse events, irrespective of attribution determined by investigator.

A mandatory baseline biopsy specimen was obtained from each patient for biomarker assessment. Peak and trough blood samples were obtained from patients at treatment initiation for pharmacokinetic analysis. Additional trough samples were gathered roughly every 12 weeks for the first 12 months on the study and every 6 months thereafter. Pembrolizumab serum concentrations were quantified with a validated electrochemiluminescent assay (lower limit of quantification 10 ng/mL).

Statistical analysis

Per protocol, with 80 ipilimumab-refractory patients at each dose, the study had 85% power to detect a 15% difference in ORR between the two doses at the 10% type 1 error (one-sided) when the ORR in the inferior group was 10%. Analyses of ORR and disease control rates were done on the full-analysis set, defined as all treated patients with measurable disease at baseline. PFS, overall survival, and safety were analysed in the as-treated population (n=173). ORRs with 95% CIs were estimated with the Clopper-Pearson method.[23] The Miettinen and Nurminen[24] method was used to compare ORRs between the pembrolizumab 2 mg/kg and 10 mg/kg groups. Time-to-event endpoints—response duration, PFS, overall survival, and response over time—were estimated with the Kaplan-Meier method. Median PFS and overall survival and the accompanying 95% CIs, PFS at 24 weeks, and overall survival at 1 year were estimated with the Kaplan-Meier method for censored data. The 95% CIs for the 24-week PFS and 1-year

See Online for appendix

	Pembrolizumab 2 mg/kg group (n=89)	Pembrolizumab 10 mg/kg group (n=84)	Total (n=173)
Sex			
Male	48 (54%)	57 (68%)	105 (61%)
Female	41 (46%)	27 (32%)	68 (39%)
Age (years; mean, range)	57·0 (18·0–88·0)	60·7 (27·0–86·0)	58·8 (18·0–88·0)
Ethnic origin			
Asian	1 (1%)	2 (2%)	3 (2%)
Black or African-American	1 (1%)	0	1 (<1%)
Mixed	0	1 (1%)	1 (<1%)
White	87 (98%)	81 (96%)	168 (97%)
ECOG performance status			
0	59 (66%)	57 (68%)	116 (67%)
1	30 (34%)	27 (32%)	57 (33%)
BRAF mutation			
Yes	12 (13%)	19 (23%)	31 (18%)
No	77 (87%)	65 (77%)	142 (82%)
Brain metastasis			
Yes	7 (8%)	8 (10%)	15 (9%)
No	81 (91%)	75 (89%)	156 (90%)
Unknown	1 (1%)	1 (1%)	2 (1%)
Lactate dehydrogenase concentration			
Normal	49 (55%)	55 (66%)	104 (60%)
Elevated	39 (44%)	29 (35%)	68 (39%)
Unknown	1 (1%)	0	1 (<1%)
Baseline tumour size* (mm; mean, range)	171 (15–895)	149 (14–535)	160 (14–895)
M staging of extent of metastasis†			
M0	1 (1%)	1 (1%)	2 (1%)
M1a	10 (11%)	17 (20%)	27 (16%)
M1b	20 (22%)	14 (17%)	34 (20%)
M1c	58 (65%)	52 (62%)	110 (64%)
Previous systemic therapies			
1	29 (33%)	19 (23%)	48 (28%)
2	31 (35%)	34 (40%)	65 (38%)
≥3	29 (33%)	31 (37%)	60 (35%)
Previous treatments			
Ipilimumab	89 (100%)	84 (100%)	173 (100%)
Immunotherapy, excluding ipilimumab	27 (30%)	26 (31%)	53 (31%)
Chemotherapy	39 (44%)	41 (49%)	80 (46%)
BRAF or MEK inhibitor, or both‡	14 (16%)	20 (24%)	34 (20%)

Data are number (%), unless otherwise indicated. ECOG=Eastern Cooperative Oncology Group. M0=no distant metastasis. M1a=metastasis to skin, subcutaneous tissues, or distant lymph nodes. M1b=metastasis to lung. M1c=metastasis to all other visceral sites or distant metastases at any site associated with elevated serum concentrations of lactate dehydrogenase. *Baseline tumour size was calculated as the sum of the longest diameters of all target lesions for patients with measurable disease by Response Evaluation Criteria In Solid Tumors (version 1.1) by independent central review at baseline (n=81 for 2 mg/kg group and n=76 for 10 mg/kg group). †Updated in June, 2014 to reflect data corrections that were necessary to account for incorrect data entry at the site level. ‡The number of patients with previous treatment with a BRAF or MEK inhibitor, or both, is greater than the number of patients with *BRAF*-mutant melanoma because those with a *BRAF* wild-type melanoma could have received a MEK inhibitor in a clinical trial.

Table 1: **Baseline characteristics of patients**

	RECIST, independent central review			Immune-related response criteria, investigator review		
	Pembrolizumab 2 mg/kg	Pembrolizumab 10 mg/kg	Estimated difference (95% CI)	Pembrolizumab 2 mg/kg	Pembrolizumab 10 mg/kg	Estimated difference (95% CI)
Best overall response*	n=81	n=76		n=89	n=84	
Complete response	1 (1%)	1 (1%)	..	3 (3%)	0 (0%)	..
Partial response	20 (25%)	19 (25%)	..	21 (24%)	27 (32%)	..
Stable disease	20 (25%)	18 (24%)	..	31 (35%)	27 (32%)	..
Progressive disease	27 (33%)	31 (41%)	..	24 (27%)	19 (23%)	..
Not evaluable†	13 (16%)	7 (9%)	..	..	..	..
No assessment‡	..	..	..	10 (11%)	11 (13%)	..
Overall response rate (95% CI)	26% (17 to 37)	26% (17 to 38)	0%§ (–14 to 13); p=0·96	27% (18 to 37)	32% (22 to 43)	–5%§ (–19 to 8); p=0·46
Disease control rate (95% CI)	51% (39 to 62)	50% (38 to 62)	1%§ (–15 to 16); p=0·94	62% (51 to 72)	64% (53 to 74)	–2%§ (–17 to 12); p=0·75
Patients with response	n=21	n=20	..	n=24	n=27	..
Time to response (weeks; median, range)	12 (11 to 36)	12 (7 to 17)	..	12 (11 to 24)	12 (7 to 39)	..
Response duration (weeks; median, range)	NR (6–37¶)	NR (8–37¶)	..	NR (12–42¶)	NR (4–37¶)	..
Progression-free survival	n=89	n=84	..	n=89	n=84	..
Median (weeks; 95% CI)	22 (12 to 36)	14 (12 to 24)	0·84‖ (0·57–1·23)	31 (22 to 48)	35 (24 to NR)	1·12‖ (0·73–1·72)
At 24 weeks (95% CI)	45% (34 to 55)	37% (27 to 48)	..	57% (46 to 67)	57% (45 to 67)	..

RECIST=Response Evaluation Criteria In Solid Tumors (version 1.1). NR=not reached. *Eight patients in each treatment group did not have measurable disease as per independent central review at baseline and were excluded from the analysis of best overall response as per RECIST by independent central review. †Patients who had no scans for response assessment or who had radiological images of non-diagnostic quality. ‡Patients who exited the study without post-baseline response assessment by the investigator. §Difference. ¶Non-progressive disease or ongoing response at the last assessment. ‖Hazard ratio. Two-sided p values are provided for testing the null hypothesis that there is no difference in the response between groups versus there is a response difference.

***Table 2*: Antitumour activity of pembrolizumab at 2mg/kg and 10 mg/kg**

overall survival were calculated in accordance with Greenwood's formula.[25] The hazard ratio (HR) and its 95% CI were calculated for between-dose comparisons of PFS with the Cox proportional hazards model with Efron's tie handling.[26] A population pharmacokinetic analysis was done with NONMEM.[27] Individual post-hoc estimates of the pembrolizumab area under the curve at steady state were summarised descriptively. Analysis was done on the population of patients who provided at least one post-baseline blood sample.

Role of the funding source

This study was funded by Merck Sharp and Dohme, Whitehouse Station, NJ, a subsidiary of Merck, which provided the study drug and worked jointly with the senior academic authors to design the study and gather, analyse, and interpret the results. CR, AR, and AD had full access to the study data and worked with employees of the funder to analyse the results. All authors approved the decision to submit the manuscript for publication, affirm the accuracy and completeness of the data, and attest that the study was done in accordance with the protocol and all amendments. All drafts of the manuscript were written by the corresponding author and AD with input from the other authors. The funder provided assistance with manuscript preparation and funded medical writing support. Aside from the authors and those listed in the acknowledgments, no one else contributed to the preparation of the manuscript.

Results

Between Aug 28, 2012, and April 5, 2013, 178 patients with ipilimumab-refractory advanced melanoma who were enrolled at 15 sites in four countries were randomly assigned to the pembrolizumab 2 mg/kg (n=91) or pembrolizumab 10 mg/kg group (n=87), and 173 received treatment (89 and 84 patients, respectively; figure 1). At baseline, 110 (64%) of 173 patients were stage M1c, 68 (39%) had elevated lactate dehydrogenase concentrations, 31 (18%) had *BRAF* mutations, and 15 (9%) had a history of brain metastases (table 1). The patients were heavily pretreated, with 125 (72%) receiving at least two and 60 (35%) receiving at least three previous treatments including ipilimumab (table 1). Baseline characteristics were generally well balanced between treatment groups, although there were some minor imbalances (eg, a higher percentage of patients with *BRAF*-mutant melanoma and a lower percentage with increases in lactate dehydrogenase concentrations in the pembrolizumab 10 mg/kg group; table 1). Ten (6%) of 170 patients with available data had an objective response and 35 (21%) had stable disease as their best overall response to previous ipilimumab treatment. The mean interval between the last dose of ipilimumab and the first dose of pembrolizumab was 33·4 weeks (range 4·0–173·0) in the pembrolizumab 2 mg/kg group and 34·1 weeks (6·0–248·0) in the pembrolizumab 10 mg/kg group.

At the time of analysis (Oct 18, 2013), median follow-up was 8 months (IQR 7–10), with all patients having at

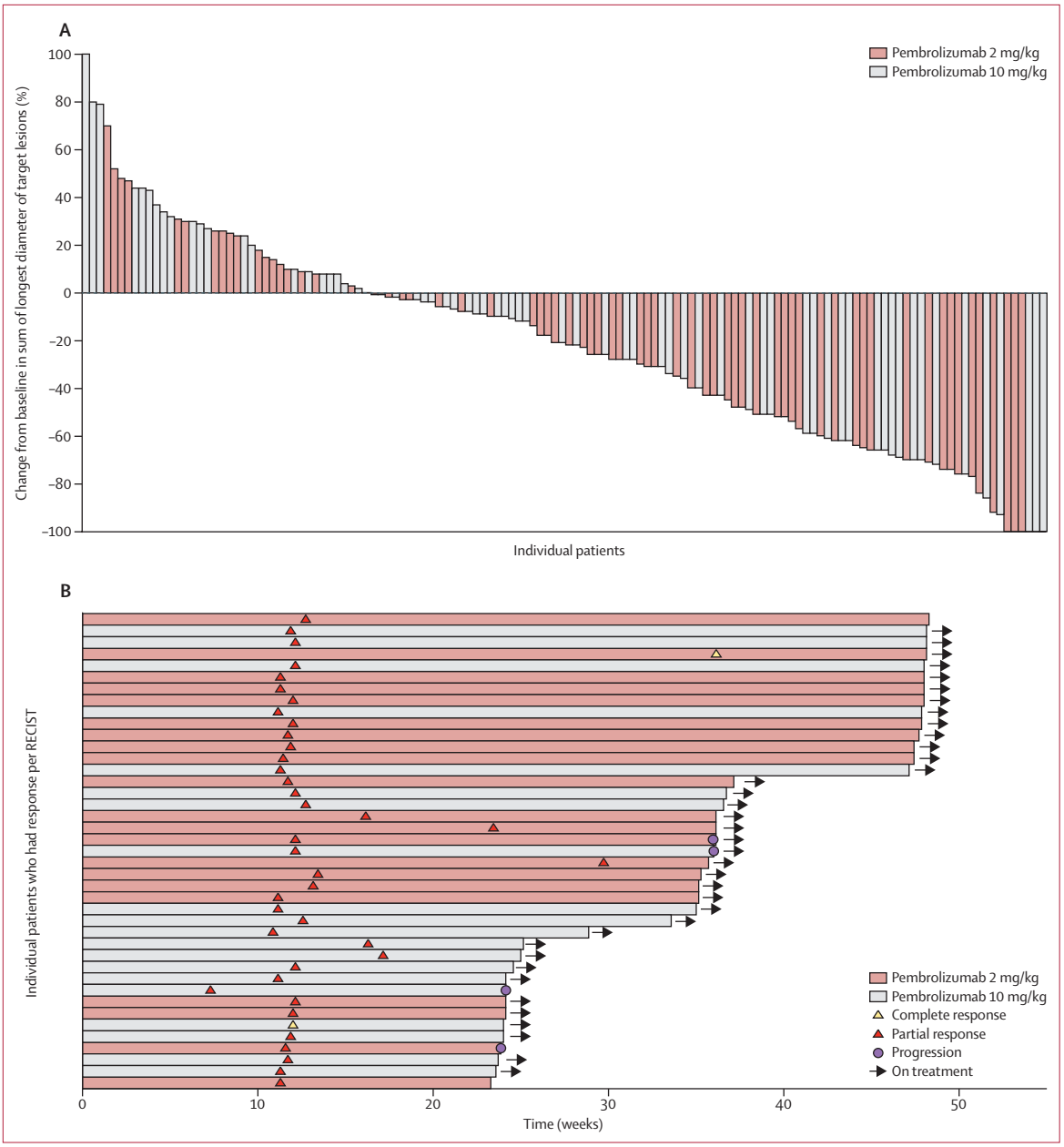

Figure 2: **Efficacy of pembrolizumab at 2 mg/kg and 10 mg/kg**

(A) Waterfall plot of maximum percentage change from baseline in the sum of the longest diameter of each target lesion as assessed per RECIST by independent central review for patients with measurable disease by independent central review at baseline who had at least one post-baseline tumour assessment. (B) Time to and duration of response in responders as assessed per RECIST by independent central review. RECIST=Response Evaluation Criteria In Solid Tumors (version 1.1).

least 6 months of follow-up. Median number of days from first to last pembrolizumab dose was 188·0 days in the pembrolizumab 2 mg/kg group and 185·5 days in the pembrolizumab 10 mg/kg group (appendix). At analysis, 73 (42%) of 173 patients were still on treatment (figure 1). The most common reason for discontinuation of treatment was disease progression (59 [34%] of 173).

The ORR was 26% in the pembrolizumab 2 mg/kg (21 of 81 patients) and 10 mg/kg groups (20 of 76 patients; p=0·96; table 2). 59 (73%) patients in the pembrolizumab 2 mg/kg group and 52 (68%) in the 10 mg/kg group had a reduction from baseline in target lesion size (figure 2A). Change from baseline in index lesion size by immune-related response criteria was also assessed (appendix). Median response duration was not reached in either dose group at the time of analysis (range >6 weeks to >37 weeks), with 36 (88%) of 41 responders alive with no other anticancer

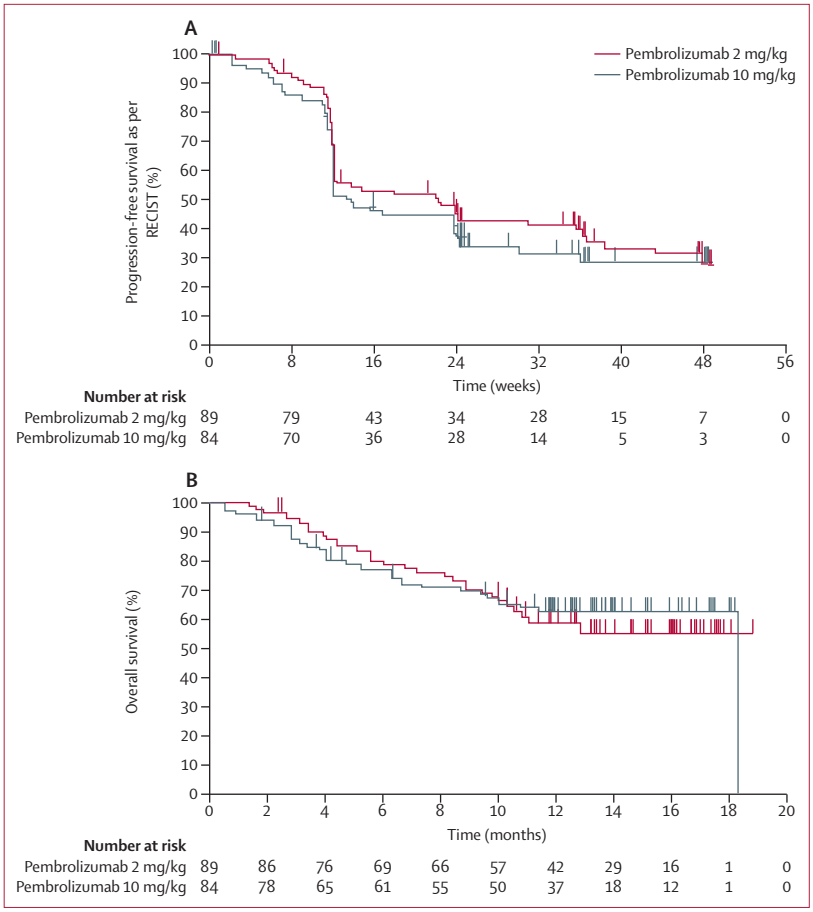

Figure 3: **Kaplan-Meier estimate of progression-free survival as assessed per RECIST by independent central review (A) and Kaplan-Meier estimates of overall survival (B)**
Analysis of progression-free survival was done in October, 2013. Analysis of overall survival was done in May, 2014. RECIST=Response Evaluation Criteria In Solid Tumors (version 1.1).

treatment and with non-progressive disease by independent central review (table 2, figure 2B). Analysis of time to response indicates that although most responses to pembrolizumab occurred by week 12, responses might also occur late in the course of treatment, with responses noted as late as 36 weeks after treatment initiation (table 2, figure 2B). Exploratory analysis showed that response rates were mostly similar in the major subgroups (appendix). ORR in the *BRAF*-wild-type subgroup of 131 patients (28%, 95% CI 20–36) was higher than in the *BRAF*-mutant subgroup of 26 patients (19%, 7–39). Of note, the 95% CIs for these subgroups overlapped.

Median PFS by independent central review was 22 weeks (95% CI 12–36) for the pembrolizumab 2 mg/kg group and 14 weeks (12–24) for the pembrolizumab 10 mg/kg group (HR 0·84, 95% CI 0·57–1·23; figure 3A). Kaplan-Meier estimates of PFS at 24 weeks were 45% in the pembrolizumab 2 mg/kg and 37% in the pembrolizumab 10 mg/kg groups. When assessed by the investigator using immune-related response criteria, median PFS was 31 weeks in the pembrolizumab 2 mg/kg group and 35 weeks in the 10 mg/kg group, and estimated 24-week PFS was 57% in each group (table 2; appendix). Of the 58 patients who had progressive disease according to RECIST, seven of 27 patients in the 2 mg/kg group and four of 31 patients in the 10 mg/kg group were progression free at 24 weeks, including three patients who had partial response using immune-related response criteria.

The survival analysis was updated in May, 2014. The HR for the difference in overall survival between the treatment groups was 1·09 (95% CI 0·68–1·75). The Kaplan-Meier estimated overall survival at 1 year (proportion of patients alive at 1 year) was 58% (95% CI 47–68) in the pembrolizumab 2 mg/kg group and 63% (51–72) in the pembrolizumab 10 mg/kg group (figure 3B).

Pembrolizumab was generally well tolerated in this population of patients with ipilimumab-refractory advanced melanoma. Overall, the safety profiles were similar between the pembrolizumab 2 mg/kg and 10 mg/kg groups (table 3). Drug-related adverse events occurred in 73 (82%) patients in the pembrolizumab 2 mg/kg group and 69 (82%) in the pembrolizumab 10 mg/kg group (table 3). However, drug-related grade 3 or 4 adverse events occurred in only 20 (12%) patients, and the only drug-related grade 3 to 4 adverse event that occurred in more than one patient was fatigue (five [3%]; table 3). Furthermore, only eight (5%) patients had drug-related serious adverse events and six (3%) discontinued treatment because of drug-related adverse events, and no drug-related deaths were reported. The most common drug-related adverse events of any grade were fatigue, pruritus, and rash (appendix). Adverse events of any grade occurring in at least 5% of patients and adverse events judged to be potentially immune-mediated by the investigators are reported in the appendix. Grade 3 or 4 immune-mediated adverse events occurred in only three patients: autoimmune hepatitis, maculopapular rash, and pancreatitis (appendix). Grade 3 or 4 adverse events of special interest occurred in 11 (6%) patients (appendix), and seven of these were classified as drug-related by investigators: autoimmune hepatitis, diarrhoea, hypophysitis, maculopapular rash, pancreatitis, pneumonitis, and rash. Potentially immune-mediated adverse events were generally manageable with treatment interruption and corticosteroid treatment, with only four patients discontinuing because of adverse events that were immune related or of special interest (table 3).

Pembrolizumab exposure as assessed with the area under the curve at steady state was 0·643 g·day/L with a coefficient of variation of 37% in the 2 mg/kg group and 3·77 g·day/L with a coefficient of variation of 33% in the 10 mg/kg group (appendix)—ie, exposure was 5·9 times higher in the higher-dose group.

Discussion

The anti-PD-1 antibody pembrolizumab at doses of 2 mg/kg and 10 mg/kg every 3 weeks had similar and substantial anticancer activity and an acceptable safety profile in patients with advanced melanoma whose disease had progressed on ipilimumab, and in those with *BRAF*-mutant disease, who were previously treated with BRAF or MEK inhibitors, or both. This study is the largest reported of anti-PD-1 therapy in patients with melanoma and the first reported randomised comparison of an anti-PD-1 agent (panel).

Similar to other immunotherapies,[14,28] the clinical benefit provided by pembrolizumab is long lasting. With a median follow-up of 8 months (minimum of 6 months), 88% of responses were ongoing at the time of analysis. This response duration is consistent with data previously reported for patients with pembrolizumab-treated melanoma, whereby after a median follow-up of 15 months, 88% of responses were ongoing and the median duration of response was not reached.[16] A high percentage of patients were progression free at 24 weeks. The finding that 19% of patients with progressive disease as per RECIST were progression free at 6 months as per immune-related response criteria suggests that conventional use of RECIST might underestimate the therapeutic benefit of pembrolizumab. The finding of delayed response also suggests that additional objective responses will occur with longer follow-up. According to previously reported data for a mixed population of ipilimumab-treated and ipilimumab-naive patients, initial responses occurred as late as 11 months after initiation of pembrolizumab, with complete responses occurring as late as 16 months.[16] This prolonged time to complete response might partly explain why the percentage of patients with complete response in this study after a median follow-up of 8 months was only 1%. With additional follow-up, it is possible that there will be more complete responses. Overall, and as was previously suggested for ipilimumab,[21,29] these data suggest that traditional response criteria might need to be revised for the overall therapeutic benefits of pembrolizumab to be fully appreciated.

Of note, there were a small number of patients with *BRAF*-mutant melanoma enrolled in this study relative to the frequency of *BRAF* mutation generally noted in patients with advanced melanoma. In our study, previous BRAF or MEK inhibitor therapy, or both, was required for patients with *BRAF*-mutant melanoma. Thus, it is possible that the small *BRAF*-mutant population enrolled is a result of a more aggressive disease that occurs after progression on a BRAF inhibitor,[30,31] which could have led to patients not having sufficient performance status to permit their enrolment in this study. Generally, the response rate associated with immunotherapy in patients with melanoma does not differ by *BRAF* mutation status.[32] In our study, there was a trend towards a lower response in the *BRAF*-mutant population. However, the small number of patients with *BRAF* mutation and BRAF or MEK inhibitor, or both, pretreatment makes it difficult to identify the various factors that might have contributed to response in this important patient group. In view of the small sample size, exploratory analysis, and overlapping 95% CIs compared with the overall and *BRAF*-wild type populations, our findings should be interpreted with caution. Data from the ongoing, randomised, controlled KEYNOTE-002 study (ClinicalTrials.gov, number NCT01704287), which is in progress in a similarly defined population but with a larger sample size (n=540), are expected to be more definitive about the activity of

	Pembrolizumab 2 mg/kg (n=89)	Pembrolizumab 10 mg/kg (n=84)	Total (n=173)
Drug-related adverse events			
Total	73 (82%)	69 (82%)	142 (82%)
Grade 3 or 4	13 (15%)	7 (8%)	20 (12%)
Serious	7 (8%)	1 (1%)	8 (5%)
Immune-related adverse events			
Grade 3 or 4	1 (1%)	2 (2%)	3 (2%)
Serious	3 (3%)	1 (1%)	4 (2%)
Adverse events of special interest			
Grade 3 or 4	4 (4%)	7 (8%)	11 (6%)
Drug-related, grade 3 or 4	3 (3%)	4 (5%)	7 (4%)
Serious	4 (4%)	4 (5%)	8 (5%)
Adverse events leading to discontinuation of drug*			
Total	6 (7%)	9 (11%)	15 (9%)
Drug-related, any grade	5 (6%)	1 (1%)	6 (3%)
Drug-related, grade 3 or 4	2 (2%)	1 (1%)	3 (2%)
Immune-related	3 (3%)	1 (1%)	4 (2%)
Of special interest	3 (3%)	1 (1%)	4 (2%)
Grade 3 or 4 drug-related adverse events occurring in one or more patients			
Fatigue	5 (6%)	0	5 (3%)
Amylase increased	1 (1%)	0	1 (<1%)
Anaemia	1 (1%)	0	1 (<1%)
Autoimmune hepatitis	1 (1%)	0	1 (<1%)
Confusion	1 (1%)	0	1 (<1%)
Diarrhoea	0	1 (1%)	1 (<1%)
Dyspnoea	0	1 (1%)	1 (<1%)
Encephalopathy	1 (1%)	0	1 (<1%)
Hypophysitis	1 (1%)	0	1 (<1%)
Hypoxia	0	1 (1%)	1 (<1%)
Muscular weakness	1 (1%)	0	1 (<1%)
Musculoskeletal pain	0	1 (1%)	1 (<1%)
Pancreatitis	0	1 (1%)	1 (<1%)
Peripheral motor neuropathy	1 (1%)	0	1 (<1%)
Pneumonitis	1 (1%)	0	1 (<1%)
Rash	0	1 (1%)	1 (<1%)
Rash maculopapular	0	1 (1%)	1 (<1%)

Data are number (%), unless otherwise indicated. *Medical Dictionary for Regulatory Activities preferred terms "progressive disease" and "malignant neoplasm progression" not related to study drug are excluded.

***Table 3*: Summary of adverse events in the pembrolizumab 2mg/kg and 10 mg/kg groups**

Panel: Research in context

Systematic review

To identify other studies of inhibitors of programmed death receptor 1 (PD-1) or PD-1-ligand (PD-L1) inhibitors in advanced cancers, including melanoma, we did an extensive search of PubMed and congress abstracts from the yearly meetings of the American Society of Clinical Oncology, European Society of Medical Oncology (included as part of the European Cancer Congress in odd years), and Society for Melanoma Research. The PubMed search was not limited by date. Congress abstracts were searched starting from 2010. Search terms were "PD-1", "PD-L1", "MK-3475", "lambrolizumab", "nivolumab", "BMS-936558", "MPDL3280A", and "BMS-936559". Our search identified several non-randomised phase 1 studies with promising results of antitumour response for PD-1 and PD-L1 inhibitors in patients with advanced solid tumours, including those with melanoma.[13–20] Although these data suggest activity for PD-1 inhibition in patients with melanoma after ipilimumab,[15–18] the sample sizes were too small (≤48 patients) to allow firm conclusions to be drawn about the efficacy and safety of PD-1 inhibition in patients with ipilimumab-refractory melanoma. Additionally, antitumour activity of anti-PD-1 agents was noted at a wide range of doses in several tumour types, but dose-response data from randomised studies were not available for any of the PD-1 or PD-L1 inhibitors.

Interpretation

The results of this study show the efficacy and safety of the anti-PD-1 monoclonal antibody pembrolizumab in patients with advanced melanoma who had confirmed disease progression on ipilimumab, and in those with *BRAF*-mutant disease who were previously treated with BRAF or MEK inhibitors, or both. This is a key finding in view of the lack of effective treatment options for patients whose disease progresses on ipilimumab and BRAF or MEK inhibitors. Results of this study will be invaluable for the future development of pembrolizumab and other monoclonal antibodies that block PD-1 or PD-L1 in different cancers.

pembrolizumab in patients with *BRAF*-mutant melanoma.

Previously reported data for pembrolizumab were from a heterogeneous, non-randomised cohort in which 31% of patients were treatment naive, 64% were ipilimumab naive, 36% received previous ipilimumab (confirmed progression not required), and BRAF or MEK inhibitor, or both, treatment was not required for *BRAF*-mutant melanoma.[15,16] By comparison, the current randomised cohort enrolled was a more homogeneous and heavily pretreated population because all patients had disease that progressed on previous ipilimumab (confirmed with two tumour assessments), 72% received at least two previous systemic therapies, and treatment with a BRAF or MEK inhibitor, or both, was required for all patients with *BRAF*-mutant melanoma. These factors might explain the lower percentages of patients with complete response (1% *vs* 9%), any response (26% *vs* 41%), 24-week PFS (37% and 45% *vs* 54%), and overall survival at 1 year (58% and 63% *vs* 81%) in the present cohort.[15,16] It is important to note that the pattern and duration of response are consistent with those reported previously.[15,16] The complete response rate and ORR with pembrolizumab are also generally consistent with those reported for nivolumab in patients with melanoma previously treated with at least three previous doses of ipilimumab (20% ORR, 3% complete response rate [as per modified WHO criteria]).[33]

The safety data corroborate previously published data showing that anti-PD-1 therapy is generally well tolerated and safe in patients previously treated with ipilimumab, with a safety profile similar to that reported for ipilimumab-naive patients.[13–16] Most drug-related adverse events in the current study were of grade 1 or 2 severity and were reversible. Although uncommon, severe adverse events of potential immune cause were successfully managed with treatment interruption or immunosuppressive therapy, or both. The overall safety profile was similar in the 2 mg/kg and the 10 mg/kg groups, and no deaths due to drug-related adverse events were reported.

Our findings suggest that pembrolizumab at a dose of 2 mg/kg or 10 mg/kg every 3 weeks could be an effective treatment option for patients with ipilimumab-refractory advanced melanoma, a population for whom there are few effective treatment options.

Contributors

CR, AR, and AD contributed equally to the development of this manuscript. AR, JDW, KG, JE-S, XNL, SPK, and AD contributed to the design and conduct of the study. CR, AR, FSH, OH, RK, JSW, AMJ, W-JH, TCG, AP, RD, HZ, BC, CM, KG, JE-S, SPK, and AD gathered data. AR, OH, JE-S, RI, SWE, and SPK did or supervised data analysis. CR, AR, FSH, OH, RK, AMJ, W-JH, TCG, RWJ, PB, BC, MAP, JE-S, XNL, RI, SWE, SPK, and AD interpreted the data. CR, JE-S, XNL, SPK, and AD wrote sections of the initial manuscript. CR, AR, JDW, OH, RK, JSW, AMJ, W-JH, TCG, AP, RD, RWJ, BC, CM, MAP, KG, and AD provided study materials or patients. OH, RK, and KG provided administrative, technical, or logistical support. XNL provided statistical expertise. All authors critically reviewed iterations of the manuscript and approved the final draft for submission.

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Declaration of interests

KG, JE-S, XNL, RI, SWE, and SPK are employees of Merck Sharp and Dohme, a subsidiary of Merck, Whitehouse Station, NJ, and might own stock or hold stock options in the company. CR has served on advisory boards for Merck, Roche, Bristol-Myers Squibb, GlaxoSmithKline, Novartis, and Amgen. AR has received research funding, served on advisory boards, and received honoraria from or for Merck. JDW has received research funding and non-financial support from Merck and Bristol-Myers Squibb and has served on advisory boards for Merck. FSH

has received funding from Merck for doing clinical research studies and has served as a non-paid consultant to Merck. OH has received research funding and has served as a consultant to Merck. RK has had travel paid by Merck, Bristol-Myers Squibb, Roche, and Novartis; has served on advisory boards for Merck, Bristol-Myers Squibb, GlaxoSmithKline, Roche, and Novartis; and has received honoraria from Merck. JSW has received research funding and personal fees from Merck. W-JH has received funding from Merck for clinical research, clinical trial support, and study drug. TCG has received personal fees from Merck. AP has received research funding from Merck. HZ has received research funding from Merck. PB has received research funding from Merck and speaker's fees from Bristol-Myers Squibb. BC has served on advisory boards for Merck, Genentech, Bristol-Myers Squibb, Prometheus, Morphotek, GlaxoSmithKline, and CytRx Corporation; has received research funding from Merck; and is a member of speaker's bureaus for Genentech, Bristol-Myers Squibb, and Prometheus. MAP has received research funding from Bristol-Myers Squibb. AD has received research funding from and served on advisory boards for Amgen, Genentech, GlaxoSmithKline, OncoSec, and Roche. The other authors declare no competing interests.

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