DRUG EVALUATION

Melanoma Management

Pembrolizumab in the management of metastatic melanoma

Lavinia Spain[1], Eugenie Younger[1], Emine Hatipoglu[1] & James Larkin*[,1]

Practice points

- The incidence of malignant melanoma is rising.
- Immune checkpoint inhibitors have significantly improved overall survival for patients with metastatic melanoma.
- Pembrolizumab is an anti-PD1-humanized monoclonal IgG4-kappa isotype antibody, which binds to the PD-1 protein expressed on the surface of immune cells.
- Inhibition of this checkpoint increases effector T-cell activity in the tumor microenvironment with substantial anti-cancer effects.
- Pembrolizumab is licensed in the USA, Europe and Australia for treatment of patients with metastatic melanoma.
- Early-phase trials of pembrolizumab demonstrate encouraging and durable responses at all dose-levels and schedules tested.
- Pembrolizumab is superior to cytotoxic chemotherapy in patients refractory to ipilimumab and BRAF-targeted therapies.
- Recent Phase III data have confirmed a significant progression-free and overall survival advantage over ipilimumab in the first-line setting.
- Pembrolizumab is a safe, well-tolerated drug with a lower incidence of serious adverse effects compared with ipilimumab.
- Common side effects include fatigue, pruritus, rash and diarrhea.
- Specific immune-related adverse events include hypothyroidism, hyperthyroidism, colitis and hepatitis.
- No drug-related deaths have been reported.
- Benefit of pembrolizumab in combination with other immunotherapies and targeted therapies is yet to be established.
- Efficacy in the adjuvant setting is under investigation.

Pembrolizumab is a humanized IgG4 anti-PD-1 antibody that plays a major role in the treatment of advanced melanoma. Through blockade of PD-1, it leads to an increase in effector T-cell activity in the tumor microenvironment. Clinical trial outcomes for pembrolizumab in addition to pharmacokinetics, pharmacodynamics and safety of the compound are discussed in this article. Phase I trials have demonstrated safety and efficacy of pembrolizumab in advanced, pretreated melanoma patients. When compared with chemotherapy in a Phase II trial of ipilimumab-refractory patients, those treated with pembrolizumab showed superior progression-free survival. In addition, in the pivotal Phase III trial pembrolizumab improved overall survival compared with ipilimumab in patients naive to immune checkpoint inhibition. Pembrolizumab is well tolerated and has a favorable safety profile. Common adverse events are fatigue, rash, itching and diarrhea. Less frequent immune-related adverse events include hypothyroidism, colitis, hepatitis and pneumonitis.

[1]The Royal Marsden Hospital, Fulham Road, London, SW3 6JJ, UK
*Author for correspondence: Tel.: +44 20 7811 8576; Fax: +44 20 7811 8103; james.larkin@rmh.nhs.uk

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KEYWORDS
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Background

The incidence of malignant melanoma in the UK has been rising over the last 40 years [1]. Lifetime risk for men is one in 55 and for women is one in 56 [1]. The incidence of melanoma in children and young adults has more than doubled in this time frame but much of this is due to earlier detection of primary tumors [2]. A greater proportion of young patients suffer from melanoma relative to other malignancies such as lung and colorectal cancer which tend to occur later in life. Risk of metastatic disease is increased in those with ulcerated, thick primary tumors and lymph node involvement. Fortunately, for those who develop metastatic disease, effective therapies that prolong survival have become available in the last few years.

The management of metastatic melanoma has changed substantially with the development of targeted therapies that inhibit oncogenic drivers and immunotherapies that augment cell-mediated defense. Previously, no survival benefit had been demonstrated with chemotherapy over best supportive care [3]. High-dose Interleukin 2, while effective in inducing remission in a small number, was associated with considerable toxicity [4]. Agents such as vemurafenib and dabrafenib improve survival relative to dacarbazine for patients whose tumors harbor a *BRAF* mutation [5–7]. Combination of these agents with MEK inhibitors further improves response rate and survival compared with BRAF inhibition alone [8–10]. The average duration of benefit is 9–12 months with the combination [8–11] and patients need to be maintained on these agents for ongoing disease control. In the majority, acquired resistance to BRAF inhibitors eventually develops [12,13].

There are two main classes of effective immune checkpoint inhibitors in advanced melanoma. The first includes a monoclonal antibody directed against CTLA4 called ipilimumab. The second are monoclonal antibodies directed against the PD-1 such as pembrolizumab and nivolumab. A third class of antibody directed against the PD-L1 has also been developed.

Immune checkpoint inhibitors enable augmented antitumor immunity by blocking signals that inhibit an activated immune response. Around 20% of patients treated with ipilimumab are alive at 3 years according to a pooled analysis of several trials [14]. This is balanced against a risk of moderate to severe but manageable toxicity in 20–27% [15,16]. The PD-1 inhibitors have a more tolerable side effect profile but are given for a longer duration. The reported survival rate of 41% for nivolumab at 3 years is superior to ipilimumab, although this figure is currently derived from a single Phase I study [17].

Pembrolizumab, also known as MK3475 and originally called lambrolizumab, has been shown to improve survival in patients naive to immune checkpoint inhibition when compared with ipilimumab [16]. It is also active in patients whose melanoma has progressed on ipilimumab and BRAF inhibitors [18,19]. This article will detail its development, safety profile and current place in the dynamic field of melanoma treatment.

Overview of the market

The last 4 years have seen three immune checkpoint agents developed and licensed for the treatment of advanced melanoma: ipilimumab, nivolumab and pembrolizumab. Others, such as anti-PD-L1 antibodies, are only available in clinical trials. BRAF inhibitors such as vemurafenib and dabrafenib also remain important licensed treatment options.

Nivolumab was the first anti-PD-1 therapy to be developed and is the main market competitor for pembrolizumab. Compared with dacarbazine, in untreated patients it is superior in its overall response rate (ORR; 40 vs 13.9%) and improves overall survival (73 vs 42% at 1 year) [20]. In ipilimumab-treated patients, nivolumab has superior efficacy, progression-free survival (PFS) and fewer side effects than chemotherapy [21]. Long-term follow-up has confirmed its safety [22]. It is also superior to ipilimumab in the first-line setting with regards to median PFS (6.9 vs 2.9 months) [15]. The administration schedule differs from pembrolizumab in that it is given 2 weekly rather than 3 weekly. In patients who have an objective tumor response, these responses are durable and there is a low rate of serious side effects [20–22]. No head-to-head comparison of nivolumab with pembrolizumab has been undertaken.

Until recently, ipilimumab was the only immune checkpoint inhibitor licensed for treatment of advanced melanoma. Improved overall survival was demonstrated in both treated and untreated melanoma patients in two key studies representing a breakthrough in this field [23,24]. It has now been established that combination immunotherapy with ipilimumab and nivolumab results in superior response rates and

improved PFS but more toxicity than with either agent alone [15,25]. A study combining pembrolizumab with ipilimumab is currently recruiting (NCT02089685).

In a Phase I study across multiple tumor types, anti-PD-L1 inhibition was associated with objective responses in 17% of patients with melanoma and had a favorable toxicity profile [26]. It continues to be evaluated in clinical trials but no product is licensed for use in the open market.

The BRAF inhibitors dabrafenib and vemurafenib remain therapeutic options with associated survival benefit compared with chemotherapy in patients with *BRAF*-mutant melanoma, with or without the addition of a MEK inhibitor [5,7–10,27]. Due to rapid and profound responses in the majority of patients [8–10], their use is favored as first-line treatment in patients with a critical burden of disease. Vemurafenib in conjunction with ipilimumab has also been evaluated [28] but was noted to cause profound hepatotoxicity. Other trials involving dabrafenib and trametinib with an anti-PD-L1 antibody as well as vemurafenib in combination with a different anti-PD-L1 compound are ongoing (NCT02027961 and NCT01656642). Although there is emerging evidence that BRAF inhibitors may favorably alter the tumor microenvironment and antigen expression [29], thereby enhancing efficacy of the checkpoint inhibitors, retrospective reviews suggest patients who receive ipilimumab subsequent to BRAF inhibition may have a worse outcome [30,31].

Both BRAF and immune checkpoint inhibitors have been evaluated in the adjuvant setting. Ipilimumab has demonstrated an improvement in recurrence-free survival in resected stage III patients compared with placebo [32]. It is not considered standard of care however due to the selection of a higher dose (10 mg/kg as opposed to 3 mg/kg), considerable toxicity including treatment-related deaths and lack of a demonstrated overall survival advantage to date. We await the results of the studies evaluating BRAF inhibitors. A randomized controlled trial evaluating pembrolizumab versus placebo in the same population is currently recruiting (NCT02362594).

Introduction to the compound

Pembrolizumab is a humanized anti-PD-1 monoclonal IgG4-kappa isotype antibody [19]. It targets PD-1, a cell surface membrane protein that functions as an inhibitory immune checkpoint receptor, helping to regulate the T-cell response and prevent autoimmunity [33,34]. PD-1 is expressed on immune response cells such as dendritic cells, B cells, T cells and monocytes. T-cell receptor activation results in upregulation of PD-1 expression [35,36], helping to counterbalance a stimulated immune system. The PD-1 protein has two ligands, PD-L1 and PD-L2. PD-L1 is expressed by antigen presenting cells, activated T cells, other immune cells, nonhematopoietic cells as well as tumor cells [34,37–38]. PD-L2 is found on macrophages, dendritic cells and B cells [37]. Although PD-L2 is expressed in some malignancies [39–41], its role in this setting is less defined than PD-L1.

Pharmacodynamics

A high level of PD-L1 expression is seen in different tumor types, including melanoma [42]. When PD-1 binds to PD-L1, the activity of primed effector T cells is inhibited and they are driven to apoptosis [42,43]. This assists tumor cells evade immune attack. By contrast, interrupting this binding enables greater interaction between T cells and dendritic cells, promotes T-cell activation and results in increased secretion of cytokines that enhance antitumor immune activity [44,45]. Therefore, pembrolizumab, by blocking the interaction of PD-1 with its ligands, can lead to enhanced effector T-cell activity in the tumor microenvironment.

Pharmacokinetics & metabolism

Pembrolizumab is administered intravenously and has a distribution volume of 7.7 l at steady state. It is eventually degraded into amino acids which are then recycled into different proteins in the body [46].

From the Phase I studies, population pharmacokinetics showed a mean clearance of 0.22 l/day and elimination half-life ($t_{1/2}$) of 26 days [18,19]. Patients who received the agent every 3 weeks had steady concentrations by week 18. In addition, administration of 2 mg/kg up to 10 mg/kg of pembrolizumab every 3 weeks caused a proportional increase in maximum concentration (C_{max}), through concentration (C_{min}) and area under the curve plasma concentration versus time (i.e., 0.643 gl/day in patients given 2 mg/kg pembrolizumab and 3.77 gl/day in the cohort given 10 mg/kg pembrolizumab). Renal impairment and mild hepatic impairment do

not affect the clearance of pembrolizumab [18,47]. Pharmacokinetic characteristics are summarized in Box 1.

Clinical efficacy

• Phase I

In 2012, the first in-human study evaluating the safety of pembrolizumab in the treatment of advanced solid malignancies was published in abstract form [48]. In this open-label, dose-escalation study, cohorts of 3–6 patients (3 + 3 design) were enrolled at doses of 1, 3 and 10 mg/kg. After an initial 28-day cycle, patients continued treatment every 2 weeks with Response Evaluation Criteria in Solid Tumors (RECIST) evaluation every 8 weeks. Nine patients, three at each dose level, completed the dose-limiting toxicity period. Patients had NSCLC (n = 3), rectal cancer (n = 2), melanoma (n = 1), carcinoid (n = 1) and sarcoma (n = 1). Antitumor effects were observed. Final data, which included seven additional patients, demonstrated particularly encouraging results for those with melanoma [49]. One patient had a complete response at 57 weeks and three patients had partial response. Stable disease was observed in 15 patients with various malignancies. Ongoing responses were observed after discontinuation of pembrolizumab.

This initial study led to a large Phase I study evaluating the safety and antitumor activity of pembrolizumab in the treatment of advanced melanoma (KEYNOTE-001) [19]. The initial cohort of patients received pembrolizumab 10 mg/kg every 2 weeks and two further cohorts were nonrandomly assigned to pembrolizumab 2 or 10 mg/kg every 3 weeks. 135 patients were treated and response assessed every 12 weeks using RECIST and investigator immune-related response criteria.

The ORR across all groups (by RECIST) was 38% (95% CI: 25–44) with the greatest response rate (52%) seen in patients treated with pembrolizumab 10 mg/kg every 2 weeks. In this study, no difference in response was observed between patients who had previously received treatment with Ipilimumab (38%) and those who were Ipilimumab naive (37%; 95% CI: 26–49). Durable responses were observed; 81% of patients who responded to MK-3475 still receiving treatment at the time of analysis. The median duration of response was not reached at a median follow-up time of 11 months. The estimated median PFS using Kaplan–Meier analysis was greater than 7 months at the time of analysis.

Following this study, an international expansion cohort of KEYNOTE-001 was designed to evaluate pembrolizumab in patients with advanced melanoma refractory to Ipilimumab and, if *BRAF* V600 mutation positive, targeted-BRAF therapy [18]. One hundred and seventy three patients with progressive disease after at least two doses of Ipilimumab were randomly assigned (1:1 final ratio) to receive pembrolizumab 2 or 10 mg/kg every 3 weeks. The primary end point was ORR and secondary end points were duration of response, PFS and overall survival. Eighty nine patients received pembrolizumab 2 mg/kg and 84 patients pembrolizumab 10 mg/kg every 3 weeks until disease progression, withdrawal of consent or intolerable toxicity. ORR in both groups was 26% after a median follow-up time of 8 months. Groups were well matched for baseline characteristics such as ECOG performance status and brain-metastases. Again durable effects were observed with 88% of responses ongoing at the time of analysis. Estimated overall survival at 1 year using Kaplan–Meier analysis was 58% (95% CI: 47–68) in the pembrolizumab 2 mg/kg group and 63% (95% CI: 51–72) in the pembrolizumab 10 mg/kg group; hazard ratio (HR) between groups 1.09 (95% CI: 0.68–1.75). Notably at 16 months, 19% of patients with progressive disease according to RECIST had stable disease when evaluated by immune response criteria, which the authors propose may underestimate the therapeutic benefit of pembrolizumab.

In 2015, Daud *et al.* reported an updated pooled analysis of 655 patients with advanced melanoma who were treated with pembrolizumab as part of the KEYNOTE-001 study [50]. This included results from the cohorts of patients discussed above [18,19]. The ORR was 34% and the median response duration had not been reached, with 80% of responses ongoing at the time of analysis. Overall survival at 1 and 2 years was 67 and 50%, respectively.

Box 1. Pharmacokinetic characteristics of pembrolizumab.

- Absorption: intravenous administration
- Distribution: 7.7 l at steady state
- Metabolism: catabolized into amino acids
- Excretion: 0.22 l/day

• Phase II

The initial success of the Phase I studies led to a Phase II study (KEYNOTE-002) to evaluate superiority in PFS of pembrolizumab 2 or 10 mg/kg versus investigator-choice chemotherapy [51]. Patients with advanced melanoma refractory to ipilimumab and if BRAF mutant, BRAF-targeted therapy, were randomly assigned 1:1:1 to receive pembrolizumab 2 or 10 mg/kg every 3 weeks or investigator choice chemotherapy. At 12 weeks, patients on chemotherapy with progressive disease were allowed to cross-over to pembrolizumab. This international study enrolled 540 patients. The independently assessed median PFS by RECIST 1.1 was 2.9 months for both pembrolizumab 2 and 10 mg/kg groups and 2.7 months for the chemotherapy arm, indicating that many progressed around the time of the first scan in each group. Nonetheless, the HR for progression were 0.57 for 2 mg/kg and 0.50 for 10 mg/kg of pembrolizumab, demonstrating its superiority compared with chemotherapy ($p < 0.00001$). At 6 months, 34% receiving pembrolizumab 2 mg/kg and 38% receiving 10 mg/kg had not progressed compared with only 16% receiving chemotherapy. Response rates for the pembrolizumab groups were also superior to chemotherapy (21% [2 mg/kg] and 25% [10 mg/kg] vs 4%) according to independent review. Median duration of response was not reached in either pembrolizumab arm and was 37 weeks for those on chemotherapy. KEYNOTE-002 was not powered to detect a difference in the efficacy of the two pembrolizumab doses but overall they appear similar in objective response rates and PFS. These Phase I and II studies are summarized in Table 1.

• Phase III

Pembrolizumab has shown superior PFS and overall survival compared with ipilimumab when given to patients naive to immune checkpoint inhibition [16]. This large Phase III trial (KEYNOTE-006) involved 834 patients with either unresectable stage III or stage IV melanoma randomized to either pembrolizumab 10 mg/kg 2 weekly or 3 weekly or ipilimumab 3 mg/kg every 3 weeks. Pembrolizumab was administered for a maximum of 24 months, whereas ipilimumab was given for four cycles only. Patients receiving pembrolizumab who had a confirmed complete response were able to cease treatment after a minimum of 6 months of treatment. RECIST was used to determine PFS, whereas clinical decisions regarding treatment were made on the basis of immune-related response criteria. A third of patients had a *BRAF* mutation and half of these had prior treatment with a BRAF inhibitor. In this subgroup, enrollment was only permitted if lactate dehydrogenase levels were normal, there were no clinically significant symptoms of disease and the patient was not rapidly progressing.

Compared with ipilimumab, both schedules of pembrolizumab significantly prolonged time to progression (2 weekly pembrolizumab 5.5 months, 3-weekly pembrolizumab 4.1 months vs ipilimumab 2.8 months; HR: 0.58 and

Table 1. Summary of Phase I and II results of pembrolizumab in advanced melanoma.

Phase	Response rate (%)†	Median PFS† (months)	Median OS	Toxicity	Ref.
Phase I IpiN and IpiT n = 135	38 (RECIST) 37 (irRC)	7	Not reached	Fatigue, rash, pruritus, diarrhea; 12% grade 3/4 adverse events	[19]
Phase I IpiT n = 173	26 (RECIST) 30 (irRC)	4.5 (RECIST) 8.0 (irRC)	Not reached 12 month OS 61%	Fatigue, rash, pruritus; 13% grade 3/4 adverse events	[18]
Phase I IpiN and IpiT (pooled) n = 665 (abstract)	34	5.2	Not reached. 12 month OS 67%	14% grade 3/4 adverse events	[50]
Phase II IpiT n = 540 (abstract)	24	2.9 (independent review RECIST) 4.6 (investigator review RECIST)	Not reached	14% grade 3/4 adverse events	[51]

†Response rates and median PFS given as average of dose (2 or 10 mg/kg) and schedules tested (2 and 3 weekly).
IpiN: Ipilimumab naive; IpiT: Ipilimumab treated; irRC: Immune-related response criteria; OS: Overall survival; PFS: Progression-free survival; RECIST: Response evaluation criteria in solid tumors.

$p < 0.001$ for both groups). While median overall survival was not reached in any group, 12-month survival was 74.1% in patients receiving 2-weekly pembrolizumab, 68.4% for 3-weekly pembrolizumab and 58.2% for those receiving ipilimumab (HR for death compared with ipilimumab 0.63 [$p < 0.0005$] and 0.69 [$p < 0.0036$], respectively). On the basis of these survival results, the study was unblinded at the second interim analysis and pembrolizumab was made available to those who had progressed on ipilimumab.

Response rates were greater with pembrolizumab (33.7% 2 weekly and 32.9% 3 weekly) than ipilimumab (11.9%). Median time to response was around 12 weeks in all groups, at the time point of the first disease assessment scan. Responses were durable with around 90% of patients still responding at the time of analysis (median 7.9 months of follow-up) and no group reaching their median duration of response.

Although the benefit of pembrolizumab over ipilimumab was evident in nearly all subgroups analyzed, there were a couple of exceptions. PD-L1 overexpression, defined as at least 1%, was noted in 82% of patients. Those deemed PD-L1 negative did not have an OS benefit with pembrolizumab compared with ipilimumab. With regards to the BRAF mutant subgroup, pembrolizumab demonstrated better PFS regardless of prior anti-BRAF therapy but there was only a trend toward improved overall survival.

Table 2 highlights the results of KEYNOTE-006 in relation to the Phase III results of its main competitor nivolumab as well as those of ipilimumab.

Postmarketing surveillance

When available, results from expanded access programs (EAPs) will provide important data on the real-world efficacy and toxicity profile of pembrolizumab. The dose of 2 mg/kg every 3 weeks was selected for the UK EAP in patients who have progressed on ipilimumab, consistent with the US label.

Safety & tolerability

Pembrolizumab is a safe and well-tolerated drug. It is associated with grade 3 and 4 adverse events (AEs) at a rate of 12–14% [16,18–19]. Serious immune-related AEs (irAEs) are rare and reversible in the majority of cases with appropriate treatment. The most common general AEs include fatigue, pruritus, rash and diarrhea. No treatment-related deaths have been attributed to pembrolizumab.

In the first report of the Phase I trial looking at pembrolizumab safety in solid tumors, no mean-tolerated dose was established [49]. The nonrandomized dose expansion cohort of 135 patients with metastatic melanoma discussed above [19] looked at safety and efficacy of pembrolizumab administered in three different schedules. This demonstrated a higher rate of AEs in the 10 mg/kg every 2-week arm (23%) compared with 10 mg/kg every 3 weeks (4%) and 2 mg/kg every 3 weeks (9%). Importantly, prior exposure to ipilimumab did not appear to increase immune-related or other side effects from pembrolizumab [19].

To evaluate dosing in a randomized setting, 10 mg/kg every 3 weeks was compared with 2 mg/kg every 3 weeks in a further 173 patients [18]. In this study by Robert *et al.*, the ORR was 26% at both doses and safety profiles were similar with drug-related AEs of any grade occurring in 82% in both arms. Drug-related grade 3 or 4 AEs occurred more frequently at 2 mg/kg (15%) than 10 mg/kg (8%) but the small number of patients limits its interpretation. Common general AEs were fatigue (35%), pruritus (23%), rash (18%), diarrhea (13%) and arthralgias (12%). The only grade 3 or 4 AE reported by more than one patient was fatigue and this occurred in five patients in the 2 mg/kg cohort. Three patients had grade 3 or 4 immune-related AEs: autoimmune hepatitis, maculopapular rash and pancreatitis. Pembrolizumab was discontinued in 9% of patients due to drug-related side effects.

Overall analysis of the large cohort of patients included in the Phase I assessment of pembrolizumab, which includes the studies detailed above, confirmed similar rates of grade 3 and 4 events (12%) [52]. No deaths were attributed to the drug and only 4% of patients discontinued treatment due to toxicity. Results from the recently published KEYNOTE-002 Phase II study also confirm the rates of AEs are similar between 2 and 10 mg/kg of pembrolizumab. Grade 3 or 4 events occurred in 11 and 14%, respectively [51]. Fatigue, pruritis and rash were the three most common general AEs at both doses. Immune-related AEs were infrequent.

In KEYNOTE-006, the higher dose of pembrolizumab (10 mg/kg) given 2 or 3 weekly had a more favorable safety profile than ipilimumab [16]. The rate of grade 3 or 4 events

Table 2. Phase III trial efficacy and safety results of pembrolizumab, nivolumab and ipilimumab.

Drug(s)	Response rate (RECIST)	Median PFS (months)	Median OS (months)	Toxicity	Ref.
Pembrolizumab (ipilimumab-naive)	33%†	4.8†	Not reached; 12-month OS 71%	Fatigue, diarrhea, rash, pruritus	[16]
Nivolumab (ipilimumab-treated)	32%	4.7	Not reported	Fatigue, pruritus, diarrhea, nausea	[21]
Nivolumab (first-line)	40%	5.1	Not reached; 12-month OS 73%	Fatigue, pruritus, nausea, diarrhea	[20]
Ipilimumab + dacarbazine	15%	3.0	11.2	Pyrexia, diarrhea, increased hepatic transaminases, pruritus	[24]
Ipilimumab + Gp100 vaccine	11%	2.8	10	Diarrhea, nausea, fatigue, skin toxicity	[23]
Ipilimumab + nivolumab	58%	11.5	Not reported	Diarrhea, fatigue, pruritus, rash	[15]

†Mean value of the two dose cohorts of pembrolizumab (2 mg/kg 2 and 3 weekly).
OS: Overall survival; PFS: Progression-free survival; RECIST: Response evaluation criteria in solid tumors.

was 11.7% for pembrolizumab overall and in the ipilimumab cohort was 19.9% (including one patient death). Treatment discontinuation rates due to drug-related AEs were lower in the pembrolizumab groups compared with ipilimumab (4.0 and 6.9 versus 9.4%). Similar to the Phase I trials, in both 2- and 3-weekly pembrolizumab groups fatigue (20.9 and 19.1%, respectively), diarrhea (16.9 and 14.4%), rash (14.7 and 14.4%) and pruritus (14.4 and 14.1%) were the most common side effects of any grade. Grade 3 or 4 diarrhea occurred in 2.5% of those in the 2-weekly group and 1.2% in 3-weekly group. Hypothyroidism (10.1 and 8.7%) and hyperthyroidism (6.5 and 3.2%) were the most frequently observed irAEs. Presumed immune-related colitis (1.8 and 3.6%), hepatitis (1.1 and 1.8%), hypophysitis (0.4 and 0.7%) and pneumonitis (0.4 and 1.8%) occurred infrequently. It is also important to consider these safety results in the context of a median duration of exposure to treatment that was much longer in the pembrolizumab cohorts (164 and 151 days) compared with ipilimumab (50 days).

Some rare irAEs have been observed with pembrolizumab. These include Type 1 diabetes mellitus, myositis, nephritis, uveitis, conjunctivitis, myocarditis and a myasthenic syndrome [16,53–54]. Awareness of the diverse array of potential irAEs is very important for clinicians assessing patients during pembrolizumab therapy.

Table 3 highlights differences in the irAE profile of pembrolizumab, nivolumab and ipilimumab and Table 4 outlines the rates of grade 3 and 4 toxicity associated with each agent.

Regulatory affairs

Pembrolizumab was approved for use in the USA by the US FDA in 2014 for patients with metastatic melanoma after treatment with ipilimumab and BRAF inhibitors (if BRAF mutation positive). Recent evidence of superiority over ipilimumab in the first-line setting may see approval of pembrolizumab as a first-line agent as well. It was licensed in Australia by the Therapeutic Goods Administration (TGA) in April 2015 and by the EMA in May 2015 for use in patients with advanced (unresectable or metastatic) melanoma, regardless of the line of therapy.

Discussion & conclusion

In the management of metastatic melanoma, pembrolizumab is an active and safe immunotherapeutic agent that improves survival. The large number of patients recruited to the Phase I and II studies, as well as the relatively short-time interval from early trials to licensing on the open market, are representative of the need that existed in this field for effective and tolerable treatments.

The recent results of KEYNOTE-006 confirm the superiority of pembrolizumab as the first-choice immune checkpoint agent in advanced melanoma as it demonstrates an overall survival advantage compared with

Table 3. Immune-related adverse events (any grade) in Phase III trials of pembrolizumab, nivolumab and ipilimumab.

Immune-related adverse event	Pembrolizumab [16]	Nivolumab [15,20,21]	Ipilimumab [15,16]	Ipilimumab + nivolumab [15]
Diarrhea (%)	14.4–16.9	11.2–19.2	22.7–33.1	44.1
Colitis (%)	1.8–3.6	1.0–1.3	8.2–11.6	11.8
Hepatitis (%)	1.1–1.8	3.4–6.4	1.2–7.1	30.0
Hypothyroidism (%)	8.7–10.1	4.4–8.6	2.0–4.2	15.0
Hyperthyroidism (%)	3.2–6.5	1.9–4.2	1.0–2.3	9.9
Hypophysitis (%)	<1	<1	2.3–3.9	7.7
Pneumonitis (%)	<1	1.9–1.3	0.4–1.6	6.4
Uveitis (%)	<1	Not reported	0	Not reported
Pruritus (%)	14.1–14.4	16.0–18.8	25.4–35.4	33.2
Rash (%)	13.4–14.7	9.3–21.7	14.5–20.9	28.4
Vitiligo (%)	9.0–11.2	5.2–10.7	1.6–3.9	6.7

ipilimumab. One of the main challenges for the future lies in determining the optimal combination and sequencing of immunotherapy with BRAF inhibitors. KEYNOTE-006 did not include patients with rapidly progressive disease who were candidates for BRAF inhibitors. Given that the first scan was undertaken at 12 weeks, we cannot be conclusive about the number of responses prior to this. There is some retrospective evidence suggesting that patients with *BRAF* mutations have a better outcome if given immunotherapy first [30,31], but this is subject to bias as those with a greater volume of disease will usually be given a BRAF inhibitor as first-line therapy. Considering the potential for enhanced antigen presentation with BRAF inhibitors [29], there remains good rationale for a combination approach. In the context of superior response rates and PFS with combination immunotherapy (ipilimumab and nivolumab) [15,25], the major question in untreated patients is likely to be deciding whether combination or single agent immunotherapy should be first-line. However, we do not yet have confirmation that this combination improves overall survival.

Further research into biomarkers is paramount in helping us triage our therapeutic decisions. While PD-L1 expression is predictive for response in pembrolizumab, responses are also seen in PD-L1-negative tumors [16,21]. At the present time, this cannot be wholly relied upon as a discriminating biomarker and variability in testing and cut-offs make standardization challenging. Ideally, we would be able to use biomarkers to help determine the type and intensity of therapy required in the first-line setting. There is evidence that combination nivolumab and ipilimumab over nivolumab alone is of particular advantage in the PD-L1-negative subgroup [15]. Genetic sequencing to determine mutational signatures of tumors that impact on antigen expression and therefore response to immunotherapy also holds promise as a future therapeutic guide [55].

One of the key advantages of pembrolizumab is its safety profile. Only one in 20 people discontinues treatment due to adverse effects of the drug [16]. Common side effects such as diarrhea and rash are often amenable to simple supportive therapies. The rate of moderate to severe toxicity is comparable with nivolumab [15,20–21] and lower than ipilimumab [15,16]. With available guidelines to assist clinicians in prompt recognition and early management of the potentially more serious irAEs, fewer patients will be compromised [56,57]. In several years, we will discover

Table 4. Grade 3 and 4 adverse events in Phase III trials of pembrolizumab, nivolumab and ipilimumab.

Immune-related adverse event	Pembrolizumab [16]	Nivolumab [15,20–21]	Ipilimumab [15,16]	Ipilimumab + nivolumab [15]
G3/4 adverse events (%)	10.1–13.3	5.0–16.3	19.9–27.3	55.0
Most common (%)	Diarrhea (1.1–2.5) Colitis (1.4–2.5) Hepatitis (1.1–1.8) Hypophysitis (0.4)	Increased hepatic transaminases (2.3) Diarrhea (1.0–2.2) Fatigue (1.0–1.3) Rash (0.6)	Colitis (7.0–8.7) Diarrhea (3.1–6.1) Rash (1.9) Hypophysitis (1.6)	Increased hepatic transaminases (14.4) Diarrhea (9.3) Colitis (7.7) Rash (4.8)

whether these irAEs, readily managed in the acute setting with steroids and other immunesuppressants, have late effects. As the prognosis of advanced melanoma improves and we begin to cure people, survivorship issues may arise, especially in younger patients.

A substantial issue with response assessment in immune checkpoint inhibitor therapy is the application of traditional criteria to determine progression, such as RECIST, that were developed for chemotherapy. The large PFS and OS differences in the Phase I studies of pembrolizumab highlight the inadequacy of RECIST criteria to accurately define clinically significant progression. Many trial protocols allow treatment beyond progression in patients who may be clinically benefiting and remain asymptomatic from their disease. Registration studies, however, still rely on use of RECIST. It is important that clinicians become familiar with alternative evaluation criteria such as those developed by Wolchok *et al.* [58] to guide their clinical practice within and outside of trials. The ideal scheduling and duration of therapy remains another area of contention. In KEYNOTE-006, patients were able to continue on therapy for up to 24 months, including beyond RECIST progression, and at a median follow-up of 7.9 months around 90% had ongoing benefit. Whether such a long duration of continuous therapy is warranted, especially in responders, is an important question to evaluate in prospective studies.

Looking to the future, we await the results of studies evaluating pembrolizumab in the adjuvant setting, especially given the more serious toxicity profile and deaths seen with ipilimumab [32]. Whether pembrolizumab may be useful in reducing the incidence of brain metastases, which autopsy series suggest are present in up to 75% of melanoma patients [59], is also of interest. Its efficacy as a primary treatment modality for CNS disease is yet to be determined. Given the efficacy of ipilimumab in treating asymptomatic brain metastases [60–62], it is an area worthy of exploration. Furthermore, with scope to combine pembrolizumab with other checkpoint inhibitors, targeted therapies and even vaccines, it is an exciting time in the field of advanced melanoma treatment.

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References

Papers of special note have been highlighted as:
• of interest; •• of considerable interest

1 Cancer Research UK. Lifetime risk for skin cancer (2012). www.cancerresearchuk.org

2 Mitsis DKL, Groman A, Beaupin LM *et al.* Trends in demographics, incidence, and survival in children, adolescents and young adults (AYA) with melanoma: a surveillance, epidemiology and end results (SEER) population-based analysis. Presented at: *American Society of Clinical Oncology,* Chicago, IL, USA, 29 May–2 June 2015.

3 Crosby T, Fish R, Coles B, Mason MD. Systemic treatments for metastatic cutaneous melanoma. *Cochrane Database Syst. Rev.* (2), CD001215 (2000).

4 Atkins MB, Kunkel L, Sznol M, Rosenberg SA. High-dose recombinant interleukin-2 therapy in patients with metastatic melanoma: long-term survival update. *Cancer J. Sci. Am.* 6(Suppl. 1), S11–S14 (2000).

5 Ascierto PA, Minor D, Ribas A *et al.* Phase II trial (BREAK-2) of the BRAF inhibitor dabrafenib (GSK2118436) in patients with metastatic melanoma. *J. Clin. Oncol.* 31(26), 3205–3211 (2013).

6 Chapman PB, Hauschild A, Robert C *et al.* Improved survival with vemurafenib in melanoma with *BRAF* V600E mutation. *N. Engl. J. Med.* 364(26), 2507–2516 (2011).

7 McArthur GA, Chapman PB, Robert C *et al.* Safety and efficacy of vemurafenib in *BRAF*(V600E) and *BRAF*(V600k) mutation-positive melanoma (BRIM-3): extended follow-up of a Phase III, randomised, open-label study. *Lancet Oncol.* 15(3), 323–332 (2014).

8 Larkin J, Ascierto PA, Dreno B *et al.* Combined vemurafenib and cobimetinib in *BRAF*-mutated melanoma. *N. Engl. J. Med.* 371(20), 1867–1876 (2014).

9 Robert C, Karaszewska B, Schachter J *et al.* Improved overall survival in melanoma with combined dabrafenib and trametinib. *N. Engl. J. Med.* 372(1), 30–39 (2015).

10 Long GV, Stroyakovskiy D, Gogas H *et al.* Dabrafenib and trametinib versus dabrafenib and placebo for VAL600 *BRAF*-mutant melanoma: a multicentre, double-blind, Phase III randomised controlled trial. *Lancet* 386(9992), 444–451 (2015).

11 Larkin JMG, Yan Y, McArthur GA *et al.* Update of progression-free survival (pfs) and correlative biomarker analysis from cobrim: Phase III study of cobimetinib (COBI) plus vemurafenib (VEM) in advanced BRAF-mutated melanoma. Presented at: *American Society of Clinical Oncology,* Chicago, IL, USA, 29 May–2 June 2015.

12 Sun C, Wang L, Huang S *et al.* Reversible and adaptive resistance to *BRAF*(V600E) inhibition in melanoma. *Nature* 508(7494), 118–122 (2014).

13 Trunzer K, Pavlick AC, Schuchter L *et al.* Pharmacodynamic effects and mechanisms of resistance to vemurafenib in patients with metastatic melanoma. *J. Clin. Oncol.* 31(14), 1767–1774 (2013).

14 Schadendorf D, Hodi FS, Robert C *et al.* Pooled analysis of long-term survival data from Phase II and Phase III trials of ipilimumab in unresectable or metastatic melanoma. *J. Clin. Oncol.* doi:10.1200/JCO.2014.56.2736 (2015) (Epub ahead of print).

15 Larkin J, Chiarion-Sileni V, Gonzalez R *et al.* Combined nivolumab and ipilimumab or monotherapy in untreated melanoma. *N. Engl. J. Med.* 373(1), 23–34 (2015).

•• **Phase III comparison of ipilimumab versus nivolumab versus the combination – key article for nivolumab in the first-line setting and for combination immunotherapy.**

16 Robert C, Schachter J, Long GV *et al.* Pembrolizumab versus ipilimumab in advanced melanoma. *N. Engl. J. Med.* 372(26), 2521–2532 (2015).

•• **Phase III comparison of pembrolizumab versus ipilimumab in patients naive to immunotherapy demonstrating superiority of pembrolizumab for survival.**

17 Hodi FS, Sznol M, Kluger H *et al.* Long-term survival of ipilimumab-naive patients (pts) with advanced melanoma (MEL) treated with nivolumab (anti-PD-1, BMS-936558, ONO-4538) in a Phase I trial. Presented at: *American Society of Clinical Oncology,* Chicago, IL, USA, 30 May–3 June 2014.

18 Robert C, Ribas A, Wolchok JD *et al.* Anti-programmed-death-receptor-1 treatment with pembrolizumab in ipilimumab-refractory advanced melanoma: a randomised dose-comparison cohort of a Phase I trial. *Lancet* 384(9948), 1109–1117 (2014).

• **Large randomised dose comparison of pembrolizumab 2 versus 10 mg/kg.**

19 Hamid O, Robert C, Daud A *et al.* Safety and tumor responses with lambrolizumab (anti-PD-1) in melanoma. *N. Engl. J. Med.* 369(2), 134–144 (2013).

• **One of the first key papers demonstrating safety and efficacy of pembrolizumab.**

20 Robert C, Long GV, Brady B *et al.* Nivolumab in previously untreated melanoma without *BRAF* mutation. *N. Engl. J. Med.* 372(4), 320–330 (2015).

21 Weber JS, D'angelo SP, Minor D *et al.* Nivolumab versus chemotherapy in patients with advanced melanoma who progressed after anti-CTLA-4 treatment (CheckMate 037): a randomised, controlled, open-label, Phase III trial. *Lancet Oncol.* 16(4), 375–384 (2015).

22 Topalian SL, Sznol M, McDermott DF *et al.* Survival, durable tumor remission, and long-term safety in patients with advanced melanoma receiving nivolumab. *J. Clin. Oncol.* 32(10), 1020–1030 (2014).

23 Hodi FS, O'day SJ, Mcdermott DF *et al.* Improved survival with ipilimumab in patients with metastatic melanoma. *N. Engl. J. Med.* 363(8), 711–723 (2010).

24 Robert C, Thomas L, Bondarenko I *et al.* Ipilimumab plus dacarbazine for previously untreated metastatic melanoma. *N. Engl. J. Med.* 364(26), 2517–2526 (2011).

25 Postow MA, Chesney J, Pavlick AC *et al.* Nivolumab and ipilimumab versus ipilimumab in untreated melanoma. *N. Engl. J. Med.* 372(21), 2006–2017 (2015).

26 Brahmer JR, Tykodi SS, Chow LQ *et al.* Safety and activity of anti-PD-L1 antibody in patients with advanced cancer. *N. Engl. J. Med.* 366(26), 2455–2465 (2012).

27 Ribas A, Gonzalez R, Pavlick A *et al.* Combination of vemurafenib and cobimetinib in patients with advanced *BRAF* (V600)-mutated melanoma: a Phase IB study. *Lancet Oncol.* 15(9), 954–965 (2014).

28 Ribas A, Hodi FS, Callahan M, Konto C, Wolchok J. Hepatotoxicity with combination of vemurafenib and ipilimumab. *N. Engl. J. Med.* 368(14), 1365–1366 (2013).

29 Frederick DT, Piris A, Cogdill AP *et al.* BRAF inhibition is associated with enhanced melanoma antigen expression and a more favorable tumor microenvironment in patients with metastatic melanoma. *Clin. Cancer Res.* 19(5), 1225–1231 (2013).

30 Ascierto PA, Simeone E, Sileni VC *et al.* Sequential treatment with ipilimumab and BRAF inhibitors in patients with metastatic melanoma: data from the Italian cohort of the ipilimumab expanded access program. *Cancer Invest.* 32(4), 144–149 (2014).

31 Ackerman A, Klein O, Mcdermott DF *et al.* Outcomes of patients with metastatic melanoma treated with immunotherapy prior to or after BRAF inhibitors. *Cancer* 120(11), 1695–1701 (2014).

32 Eggermont AM, Chiarion-Sileni V, Grob JJ *et al.* Adjuvant ipilimumab versus placebo after complete resection of high-risk stage III melanoma (EORTC 18071): a randomised, double-blind, Phase III trial. *Lancet Oncol.* 16(5), 522–530 (2015).

33 Okazaki T, Honjo T. PD-1 and PD-1 ligands: from discovery to clinical application. *Int. Immunol.* 19(7), 813–824 (2007).

34 Dong H, Zhu G, Tamada K, Chen L. B7-H1, a third member of the B7 family, co-stimulates T-cell proliferation and interleukin-10 secretion. *Nat. Med.* 5(12), 1365–1369 (1999).

35 Agata Y, Kawasaki A, Nishimura H *et al.* Expression of the PD-1 antigen on the surface of stimulated mouse T and B lymphocytes. *Int. Immunol.* 8(5), 765–772 (1996).

36 Austin JW, Lu P, Majumder P, Ahmed R, Boss JM. STAT3, STAT4, NFATC1, and CTCF regulate PD-1 through multiple novel regulatory regions in murine T cells. *J. Immunol.* 192(10), 4876–4886 (2014).

37 Latchman Y, Wood CR, Chernova T *et al.* PD-l2 is a second ligand for PD-1 and inhibits T cell activation. *Nat. Immunol.* 2(3), 261–268 (2001).

38 Freeman GJ, Long AJ, Iwai Y *et al.* Engagement of the PD-1 immunoinhibitory receptor by a novel B7 family member leads to negative regulation of lymphocyte activation. *J. Exp. Med.* 192(7), 1027–1034 (2000).

39 Rosenwald A, Wright G, Leroy K *et al.* Molecular diagnosis of primary mediastinal B cell lymphoma identifies a clinically favorable subgroup of diffuse large B cell lymphoma related to hodgkin lymphoma. *J. Exp. Med.* 198(6), 851–862 (2003).

40 Hamanishi J, Mandai M, Iwasaki M *et al.* Programmed cell death 1 ligand 1 and tumor-infiltrating $CD8^+$ T lymphocytes are prognostic factors of human ovarian cancer. *Proc. Natl Acad. Sci. USA* 104(9), 3360–3365 (2007).

41 Ohigashi Y, Sho M, Yamada Y *et al.* Clinical significance of programmed death-1 ligand-1 and programmed death-1 ligand-2 expression in human esophageal cancer. *Clin. Cancer Res.* 11(8), 2947–2953 (2005).

42 Dong H, Strome SE, Salomao DR *et al.* Tumor-associated B7-H1 promotes T-cell apoptosis: a potential mechanism of immune evasion. *Nat. Med.* 8(8), 793–800 (2002).

43 Blank C, Brown I, Peterson AC *et al.* PD-L1/B7H-1 inhibits the effector phase of tumor rejection by T cell receptor (TCR) transgenic $CD8^+$ T cells. *Cancer Res.* 64(3), 1140–1145 (2004).

44 Brown JA, Dorfman DM, Ma FR *et al.* Blockade of programmed death-1 ligands on dendritic cells enhances T cell activation and cytokine production. *J. Immunol.* 170(3), 1257–1266 (2003).

45 Fife BT, Pauken KE, Eagar TN *et al*. Interactions between PD-1 and PD-L1 promote tolerance by blocking the TCR-induced stop signal. *Nat. Immunol.* 10(11), 1185–1192 (2009).

46 FDA. Clinical pharmacology BLA review (for pembrolizumab) (2014). www.accessdata.fda.gov

47 Merck Sharp & Dohme Corp, Keytruda Product Information (2015). www.merck.com

48 Patnaik A, Kang SP, Tolcher AW *et al*. Phase I study of MK-3475 (anti-PD-1 monoclonal antibody) in patients with advanced solid tumours. Presented at: *American Society of Clinical Oncology,* Chicago, IL, USA, 1 June–5 June 2012.

49 Patnaik A, Kang SP, Rasco D *et al*. Phase I study of pembrolizumab (MK-3475; anti-PD-1 monoclonal antibody) in patients with advanced solid tumors. *Clin. Cancer Res.* doi:10.1158/1078-0432.CCR-14-2607 (2015) (Epub ahead of print).

50 Daud A, Ribas A, Robert C *et al*. Long-term efficacy of pembrolizumab (pembro; MK-3475) in a pooled analysis of 655 patients (pts) with advanced melanoma (MEL) enrolled in keynote-001. Presented at: *American Society of Clinical Oncology,* Chicago, IL, USA, 29 May–2 June 2015.

51 Ribas A, Puzanov I, Dummer R *et al*. Pembrolizumab versus investigator-choice chemotherapy for ipilimumab-refractory melanoma (KEYNOTE-002): a randomised, controlled, Phase II trial. *Lancet Oncol.* 16(8), 908–918 (2015).

52 Ribas A, Wolchok JD, Robert C. Updated clinical efficacy of the anti-PD-1 monoclonal antibody pembrolizumab (pembro, MK-3475) in 411 patients (pts) with melanoma (MEL). *Pigment Cell Melanoma Res.* 27, 1222 (2014).

53 Mandel JJ, Olar A, Aldape KD, Tremont-Lukats IW. Lambrolizumab induced central nervous system (CNS) toxicity. *J. Neurol. Sci.* 344(1–2), 229–231 (2014).

54 Laubli H, Balmelli C, Bossard M, Pfister O, Glatz K, Zippelius A. Acute heart failure due to autoimmune myocarditis under pembrolizumab treatment for metastatic melanoma. *J. Immunother. Cancer* 3, 11 (2015).

55 Snyder A, Wolchok JD, Chan TA. Genetic basis for clinical response to CTLA-4 blockade. *N. Engl. J. Med.* 372(8), 783 (2015).

56 Weber JS, Kahler KC, Hauschild A. Management of immune-related adverse events and kinetics of response with ipilimumab. *J. Clin. Oncol.* 30(21), 2691–2697 (2012).

57 Howell M, Lee R, Bowyer S, Fusi A, Lorigan P. Optimal management of immune-related toxicities associated with checkpoint inhibitors in lung cancer. *Lung Cancer* 88(2), 117–123 (2015).

58 Wolchok JD, Hoos A, O'day S *et al*. Guidelines for the evaluation of immune therapy activity in solid tumors: immune-related response criteria. *Clin. Cancer Res.* 15(23), 7412–7420 (2009).

59 Hong A, Fogarty G, Izard MA. The role of radiation therapy in the management of metastatic melanoma in the brain. *Int. J. Surg. Oncol.* 2012, 294735 (2012).

60 Danielli R, Ridolfi R, Chiarion-Sileni V *et al*. Ipilimumab in pretreated patients with metastatic uveal melanoma: safety and clinical efficacy. *Cancer Immunol. Immunother.* 61(1), 41–48 (2012).

61 Di Giacomo AM, Ascierto PA, Pilla L *et al*. Ipilimumab and fotemustine in patients with advanced melanoma (NIBIT-M1): an open-label, single-arm Phase II trial. *Lancet Oncol.* 13(9), 879–886 (2012).

62 Margolin K, Ernstoff MS, Hamid O *et al*. Ipilimumab in patients with melanoma and brain metastases: an open-label, Phase II trial. *Lancet Oncol.* 13(5), 459–465 (2012).