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CTLA-4 Blockade with Ipilimumab: Long-term Follow-up of 177 Patients with Metastatic Melanoma

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Abstract

Purpose: Treatment with ipilimumab can cause objective tumor responses in patients with metastatic melanoma. We have treated 177 evaluable patients in three clinical trials and have long-term follow-up to evaluate the durability of responses.

Experimental Design: Patients with metastatic melanoma were treated in three trials from 2002 to 2005. In protocol 1, 56 patients received ipilimumab with gp100 peptides. In protocol 2, 36 patients received ipilimumab with interleukin-2. In protocol 3, 85 patients received ipilimumab with intrapatient dose-escalation and were randomized to receive gp100 peptides. We have analyzed their long-term follow-up and survival data.

Results: With median follow-up for protocols 1, 2, and 3 being 92, 84, and 71 months, median survival was 14, 16, and 13 months with 5-year survival rates being 13%, 25%, and 23%, respectively. Patients in protocol 2 had a 17% complete response (CR) rate, compared with 7% in protocol 1 and 6% in protocol 3. These CR rates are higher than previously reported for the same trials because some patients who eventually became complete responders had continual tumor regression months to years after therapy. All but one of the 15 complete responders are ongoing at 54+ to 99+ months.

Conclusions: This report provides the longest follow-up of patients with melanoma treated with ipilimumab and shows that ipilimumab can induce durable, potentially curative tumor regression in a small percentage of patients with metastatic melanoma. The combination of ipilimumab and interleukin-2 seems to have an increased CR rate, but this needs to be tested in a randomized trial. *Clin Cancer Res; 18(7); 2039–47. ©2012 AACR.*

Introduction

Ipilimumab is a fully human IgG_1 monoclonal antibody that blocks cytotoxic T-lymphocyte–associated antigen 4 (CTLA-4), an immunosuppressive receptor on T cells (1, 2). This agent was approved by the U.S. Food and Drug Administration (FDA) for the treatment of metastatic melanoma in March 2011, the first drug to have received this indication since high-dose interleukin-2 (IL-2) received approval in 1998. We first published in 2003 a cohort of 14 patients with metastatic melanoma who received ipilimumab plus gp100 peptide vaccinations (3). Three patients achieved objective tumor responses; 6 patients developed grade III/IV immune-related adverse events (IRAE) including dermatitis, enterocolitis, and hypophysitis, among others. This initial report signified a critical role of CTLA-4 in regulating tolerance to self-antigens in humans and suggested that breaking this tolerance could lead to tumor regression. This preliminary study was expanded to 56 patients with an overall response rate of 13% (4). An intriguing association between the development of a serious IRAE and the development of an objective clinical response (OR) was seen: 5 (36%) of 14 patients who developed a grade III/IV IRAE had an OR, whereas only 2 (5%) of 42 patients without a grade III/IV IRAE developed an OR ($P = 0.008$; ref. 4).

Because of the possible correlation between the development of an IRAE and OR, we enrolled 85 patients with metastatic melanoma in an intrapatient dose-escalation protocol in which patients received an escalating dose of ipilimumab every 3 weeks from 3 to 5 to 9 mg/kg until the development of an OR or a grade III/IV IRAE (5). We have previously published the results for 46 patients who were HLA-A*0201–negative in this study; grade III/IV IRAEs developed in 35% of patients but an increase in OR rate (11%) was not seen despite the increased dose levels (5).

Because of the durability of complete responders from high-dose IL-2 (6, 7), we examined the safety and efficacy of combining ipilimumab with IL-2 in a phase I/II trial in

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Translational Relevance

The prognosis of patients with metastatic melanoma remains poor; new treatment modalities and agents are needed. Ipilimumab offers an important new therapeutic option and hope for these patients, as it is the first drug since 1998 to receive U.S. Food and Drug Administration approval for metastatic melanoma. With sustained objective complete responses lasting up to 99+ months, our long-term follow-up of 177 patients with metastatic melanoma documents the durability and potential curative outcome of antitumor responses to ipilimumab. The updated results of our exploratory study evaluating ipilimumab plus high-dose interleukin-2 show a higher-than-expected complete response rate (17%); thus, confirmatory trials testing these two agents in combination should be conducted.

which 36 patients received standard high-dose IL-2 along with ipilimumab every 3 weeks (8). Patients were enrolled into 5 cohorts with increasing doses of ipilimumab (0.1, 0.3, 1, 2, and 3 mg/kg). We reported the results in 2005 with 8 patients (22%) having ORs and 14% of patients experiencing grade III/IV IRAEs (8).

Since our initial publication showing ORs and IRAEs with ipilimumab, other investigators have shown similar findings (9–12). In 2010, Hodi and colleagues published a definitive phase III trial randomizing 676 patients with metastatic melanoma to receive either ipilimumab alone, ipilimumab plus gp100 peptides, or gp100 peptides alone (13). The OR rates for ipilimumab alone, ipilimumab plus gp100, and gp100 alone were 10.9%, 5.7%, and 1.5%, respectively. With median follow-up up to 27.8 months, patients receiving ipilimumab (either alone or with gp100) had improved median overall survival (10.1 and 10.0 months, respectively) compared with those receiving gp100 alone (6.4 months). This marked the first time that a drug for metastatic melanoma had been shown to improve overall survival in a randomized trial. Recently, ipilimumab combined with dacarbazine was reported to have increased median overall survival when compared with dacarbazine alone (11.2 vs. 9.1 months) in a phase III randomized trial (14).

Long-term follow-up of our ipilimumab patients since 2002 has revealed unique features associated with this treatment. We present here the longest follow-up for patients with melanoma treated with ipilimumab and show the durability of the responses and the distinctive characteristics of this agent.

Patients and Methods

Patients and protocol design

Patients were eligible if they were ≥18 years old and had measurable stage IV melanoma, Eastern Cooperative Oncology Group performance score of ≤2, no evidence or history of autoimmune or immunodeficiency disease, life expectancy of ≥3 months, and ≥3 weeks since any systemic cancer treatment. No patient had received prior therapy with ipilimumab. One treatment cycle is one dose of ipilimumab. Other specific details for each of the 3 protocols are discussed briefly below as all trials have been published in part (3–5, 8). All trials were approved by the Institutional Review Board of the NIH (Bethesda, MD); signed informed consent was obtained from every participant.

Protocol 1 enrolled 2 cohorts of HLA-A*0201–positive patients from 2002 to 2004 (3, 4). Cohort 1 consisted of 29 patients who received ipilimumab at 3 mg/kg every 3 weeks in conjunction with subcutaneous injections of 2 separate gp100 peptides [gp100:209–217(210M) and gp100:280–288(288V)] emulsified in Montanide ISA-51. Cohort 2 consisted of 27 patients receiving the identical gp100 peptides; however, after an initial dose of ipilimumab at 3 mg/kg, they received subsequent doses at 1 mg/kg every 3 weeks. No patient had received prior gp100 vaccination.

Protocol 2 was a phase I/II trial evaluating ipilimumab in combination with high-dose (720,000 IU/kg) intravenous IL-2 (given as tolerated every 8 hours up to a maximum of 15 doses). From 2003 to 2004, the trial enrolled 36 patients medically fit (15) to receive high-dose IL-2 (8). Patients were excluded if they had previously received high-dose IL-2 (defined as ≥600,000 IU/kg given intravenously). Patients received the first dose of ipilimumab without IL-2; in subsequent 3-week intervals, patients received ipilimumab at their designated dose level followed by IL-2 to start within 24 hours of ipilimumab. Three patients per dose level received ipilimumab at 0.1, 0.3, 1, and 2 mg/kg; the last 24 patients received ipilimumab at 3 mg/kg.

Protocol 3 was an intrapatient dose-escalating trial, which enrolled patients from 2004 to 2005 into 2 cohorts, HLA-A*0201–negative and HLA-A*0201–positive (5). HLA-A*0201–negative patients received ipilimumab alone starting at 3 mg/kg. After 2 cycles at 3 mg/kg, if an OR or a grade III/IV IRAE did not occur, the patient received the next 2 cycles at 5 mg/kg. If after 2 cycles at 5 mg/kg and an OR or a grade III/IV IRAE did not occur, the patient received the next 2 cycles at 9 mg/kg. HLA-A*0201–positive patients received ipilimumab in the same dose-escalating manner; however, they were also randomized to receive ipilimumab alone or in conjunction with the 2 gp100 peptides as given in protocol 1. Patients were allowed to have received prior gp100 vaccinations. After an initial enrollment of 38 patients, the rate of disease progression limited the number of patients able to reach the 9 mg/kg level; thus, the trial was amended to start at 5 mg/kg (instead of 3 mg/kg), and 50 additional patients were accrued. Of 88 patients enrolled, 3 were not evaluable; 2 patients' metastases were found not to be melanoma, and one patient refused treatment after being enrolled.

Clinical response evaluation and follow-up

Patients received computed axial tomography of the chest, abdomen, pelvis, and MRI of the brain within 4 weeks of beginning treatment and after every 2 treatment

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Table 1. Patient demographics

	Protocol 1: ipi + gp100 (N = 56)	Protocol 2: ipi + IL-2 (N = 36)	Protocol 3: ipi (DE) ± gp100 (N = 85)
Gender			
Female	19 (34)	14 (39)	29 (34)
Male	37 (66)	22 (61)	56 (66)
Age, y			
21–30	4 (7)	3 (8)	6 (7)
31–40	5 (9)	4 (11)	18 (21)
41–50	17 (30)	16 (44)	23 (27)
51–60	17 (30)	11 (31)	22 (26)
61–70	13 (23)	2 (6)	16 (19)
ECOG			
0	44 (79)	27 (75)	54 (64)
1	12 (21)	9 (25)	29 (34)
2	0 (0)	0 (0)	2 (2)
M1 stage			
M1a	11 (20)	10 (28)	13 (15)
M1b	12 (21)	8 (22)	21 (25)
M1c	33 (59)	18 (50)	51 (60)
Prior therapy			
Surgery	56 (100)	36 (100)	85 (100)
Chemotherapy	21 (38)	8 (22)	47 (55)
Radiotherapy	15 (27)	7 (19)	25 (29)
Immunotherapy	41 (73)	23 (64)	72 (85)
Hormonal	2 (4)	0 (0)	2 (2)
Prior systemic therapy[a]			
Any 1 or more	43 (76)	24 (66)	80 (94)
Any 2 or more	20 (36)	8 (22)	39 (46)
Any 3 or more	2 (4)	0 (0)	2 (2)

NOTE: All values are expressed as *n* (%).

Abbreviations: DE, intrapatient dose-escalation of ipilimumab; ECOG, Eastern Cooperative Oncology Group performance score; gp100, gp100:209-217(210M) and gp100:280-288(288V) peptides; ipi, ipilimumab.

[a]Systemic therapy includes chemotherapy, immunotherapy, and/or hormonal therapy.

cycles. Other imaging modalities were added as needed to evaluate specific disease sites. Tumor response was determined by standard Response Evaluation Criteria in Solid Tumors (RECIST; ref. 16): A partial response (PR) was defined as a decrease of ≥30% (but not 100%) of the sum of the longest diameters of predetermined target lesions with no new or enlarging lesions (target or nontarget). A complete response (CR) was defined as the disappearance of all lesions. All responses were confirmed by follow-up imaging ≥4 weeks after the initial response determination. Patients not experiencing PR or CR were deemed nonresponders. Patients whose tumors progressed by RECIST were taken off study. Provided they did not experience a dose-limiting toxicity, patients whose tumors did not progress received 2 additional cycles and were evaluated for response again after every 2 cycles. Responding patients not actively receiving treatment underwent follow-up physical examination, imaging, and screening for IRAEs at least every 3 months for 1 year, then at least every 6 months for 2 years, and then at least yearly thereafter. Survival updates were determined by contacting the patient or patient's local physician or by reviewing the medical records of those who have been seen in follow-up for less than 6 months prior.

Results

Patient demographics, tumor response, IRAEs, and lymphocyte counts

One hundred and eighty patients were enrolled into the 3 trials (56 patients in protocol 1, 36 patients in protocol 2, and 88 patients in protocol 3) from 2002 to 2005 (Table 1); 3 patients from protocol 3 were not evaluable and were excluded from analysis. The majority were heavily pretreated with other systemic agents. Although not statistically different, patients in protocol 2 had received less prior systemic therapy, and fewer patients had M1c disease than those in protocols 1 and 3. Median follow-up durations for protocols 1, 2, and 3 were 92, 84, and 71 months, respectively.

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Table 2. Frequency and duration of objective tumor responses

	Protocol 1: ipi + gp100 (N = 56)	Protocol 2: ipi + IL-2 (N = 36)	Protocol 3: ipi (DE) ± gp100 (N = 85)
Initial report			
PR	5 (9%)	5 (14%)	5 (of 46; 11%)
CR	2 (4%)	3 (8%)	0 (0%)
Total OR	7 (13%)	8 (22%)	5 (of 46; 11%)
Current status			
PR	3 (6%)	3 (8%)	12 (14%)
CR	4 (7%)	6 (17%)	5 (6%)
Total OR	7 (13%)	9 (25%)	17 (20%)
Response duration, mo			
PR	42, 5, 4	11, 11, 5	71+, 68, 66+, 56+, 25, 15, 11, 10, 9, 7, 6, 5
CR	99+, 94+, 94+, 88+	89+, 86+, 83+, 83+, 79+, 76+	76+, 74+, 62+ 54+, 42

Abbreviations: DE, intrapatient dose-escalation of ipilimumab; gp100, gp100:209-217(210M) and gp100:280-288(288V) peptides; ipi, ipilimumab.

As previously reported (4), protocol 1 had 7 responders (5 PRs and 2 CRs) of 56 patients for an overall response rate of 13% (Table 2). Over time, 2 PRs became CRs and are ongoing responders with the longest duration lasting 99+ months. Our initial publication in 2005 on the ipilimumab and IL-2 trial (protocol 2) reported 5 PRs and 3 CRs in the 36 patients treated (8). Continued follow-up revealed that 3 of the 5 initial PRs became CRs, and 1 previous mixed responder became a PR. This changes the OR rate to 25%, and the CR rate becomes 17%. The CRs for this protocol are all ongoing, with the longest duration lasting 89+ months. For the dose-escalation trial (protocol 3), an 11% (5 PRs, no CRs) OR rate was previously published for the 46 HLA-A*0201–negative patients who did not receive gp100 (5); HLA-A*0201–positive patients were not previously reported. Updated here to include all 85 evaluable patients, this trial achieved a 20% OR rate with 12 PRs and 5 CRs. Three PRs are still ongoing, with the longest duration lasting 71+ months. Four CRs are ongoing responders with the longest duration at 76+ months; one CR in this trial recurred at 42 months. Among those in protocol 3 who did not receive gp100 vaccines, there was no difference in the response rates for HLA-A*201–positive patients compared with HLA*A201–negative patients. No statistically significant association between M1 stage and tumor response was noted for all patients in the 3 protocols.

Among the 141 evaluable patients enrolled in protocols 1 and 3 (who did not receive IL-2 in conjunction with ipilimumab), 67 had been previously treated with IL-2 before receiving ipilimumab whereas 74 were IL-2–naive. The OR rate to ipilimumab among those who had received prior IL-2 was 12%, whereas the OR rate for IL-2–naive patients was 22%; this difference was not statistically significant ($P_2 = 0.18$; Fisher's exact test). The CR rate of those who had previously received IL-2 (4.5%; 3 of 67) was statistically the same as those who were IL-2–naive (8.1%; 6 of 74; $P_2 = 0.6$; Fisher's exact test).

The incidence of grade III/IV IRAEs was similar for protocols 1 and 3 (29% and 32%, respectively), whereas protocol 2 had an incidence of 17% (Table 3). Responders overall had a higher rate of grade III/IV IRAEs than nonresponders; 17 (51%) of the 33 responders developed grade III/IV IRAEs compared with 32 (22%) of 144 nonresponders ($P_2 = 0.002$; Fisher's exact test). When limited to just protocol 2, there was no statistical significance in the frequency of grade III/IV IRAEs between responders and nonresponders ($P_2 = 0.6$; Fisher's exact test). Gastrointestinal-related IRAEs (gastritis, duodenitis, enteritis, and colitis) were the most common grade III/IV IRAEs, consistent with other reports (9–13); one patient underwent emergent right colectomy and ileostomy for colonic perforation. As previously published in detail (3–5, 8, 17–19), patients with IRAEs were treated with supportive therapy and locally directed or high-dose systemic corticosteroids as indicated. In addition to systemic corticosteroids, patients with hypophysitis also received replacement hormones including thyroxine and testosterone (for males) as needed (18). No treatment-related death occurred in any of the 3 trials.

Pretherapy absolute lymphocyte count (ALC) was not found to be associated with the development of an OR or CR or survival (data not shown). Since it has been shown that IL-2 alone can cause lymphocytosis and that this lymphocytosis is associated with the development of an OR to IL-2 (20), we analyzed posttreatment ALC in patients in protocols 1 and 3 who did not receive IL-2 in conjunction with ipilimumab. For these patients, the change in ALC after one dose of ipilimumab (defined as ALC measured approximately 3 weeks after the first dose of ipilimumab minus pretherapy ALC) was associated the development of an OR. Responders had a higher mean increase in ALC (513 ± 73 counts/μL; range, −349 to 1,176 counts/μL) than nonresponders (313 ± 42 counts/μL; range, −612 to 2,816

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Table 3. Incidence of grade III/IV IRAEs

	Protocol 1: ipi + gp100 (N = 56)	Protocol 2: ipi + IL-2 (N = 36)	Protocol 3: ipi (DE) ± gp100 (N = 85)
Response status			
PR	1 (of 3 PRs; 33%)	1 (of 3 PRs; 33%)	7 (of 12 PRs; 58%)
CR	4 (of 4 CRs; 100%)	1 (of 6 CRs; 17%)	3 (of 5 CRs; 60%)
Any OR	5 (of 7 ORs; 71%)	2 (of 9 ORs; 22%)	10 (of 17 ORs; 59%)
Nonresponders	11 (of 49 NRs; 22%)	4 (of 27 NRs; 15%)	17 (of 68 NRs; 25%)
All patients	16 (29%)	6 (17%)	27 (32%)
Specific grade III/IV IRAE[a]			
Gastrointestinal	7	5	17[b]
Dermatitis	7	1	2
Hypophysitis	1	0	12
Uveitis	1	1	0[c]
Arthritis	0	1	1
Hepatitis	1	0	0
Nephritis	0	0	1
Mucositis	0	1	0

Abbreviations: DE, intrapatient dose-escalation of ipilimumab; gp100, gp100:209-217(210M) and gp100:280-288(288V) peptides; ipi, ipilimumab; NR, nonresponder.

[a]Number of IRAE events > number of patients experiencing IRAEs due to ≥1 IRAE per patient.

[b]One patient underwent emergency right colectomy and ileostomy for colonic perforation.

[c]One patient was previously reported (5) to have grade III/IV anterior uveitis in this protocol but on review actually had a grade II event.

counts/μL; $P_2 = 0.0052$; Mann–Whitney U test). This change in ALC was not associated with the development of CR or survival status.

Complete responders and surviving noncomplete responders

The characteristics of CR patients are presented in detail in Table 4. Overall the total dose of ipilimumab received varied widely. Except for patients on protocol 2, most CRs experienced a grade III/IV IRAE. Limited to protocols 1 and 3, 7 (78%) of 9 CR patients developed grade III/IV IRAEs, whereas 36 (27%) of 132 non-CR patients developed grade III/IV IRAEs (Table 3; $P_2 = 0.004$; Fisher's exact test). CR patients in protocol 2 tolerated the treatment well and were able to receive up to 6 cycles of IL-2 with ipilimumab. Most CRs showed evidence of OR by 2 months after starting treatment; however, considering all patients, it took an average of 30 months to reach an official CR. It took patient CR3 70 months to have one remaining lung lesion become undetectable on imaging. With the exception of patient CR15 who recurred after 42 months, all other declared CRs are ongoing.

Of 162 patients who did not achieve CR, 18 patients (11%) are alive with follow-up ranging from 56 to 101 months, including 9 nonresponders (Table 5). Three PRs are still ongoing with response durations up to 71+ months; they have had stable small lesions remaining visible on imaging in the liver and retroperitoneal lymph nodes (patient 6), subcutaneous areas (patient 7), and hilar and periportal lymph nodes (patient 8). Six patients became disease-free after undergoing metastasectomy or a locally directed procedure (patients 1, 2, 3, 10, 14, and 17) and have remained without evidence of recurrence; pathologic examination of the resected metastases showed viable tumor within the specimens. Four patients underwent adoptive cell therapy (ACT) with tumor-infiltrating lymphocytes after having progressive disease with ipilimumab and subsequently achieved ongoing CR with ACT (patients 4, 5, 12, and 13; ref. 21). Four patients have remained either disease-free or with stable PR after receiving various treatment combinations, including chemotherapy and targeted therapy (patients 9, 11, 16, and 18).

With median survival for protocols 1, 2, and 3 being 14, 16, and 13 months, respectively, survival analyses show that most patients succumbed to their disease within the first 2 years after starting treatment (Fig. 1). For the most part, the survival curves plateaued for those who survived beyond 4 years. The 5-year survival rates for protocols 1, 2, and 3 were 13%, 25%, and 23%, respectively.

Discussion

The prognosis of patients with stage IV melanoma remains poor with 1-year survival rates for those with M1a, M1b, and M1c being 62%, 53%, and 33%, respectively (22). In fact, malignant melanoma is one of the few cancers with an increasing mortality rate from 1990 to 2006, whereas others have decreased (23). Until the approval of ipilimumab in March 2011, the only FDA-approved therapies for stage IV disease were IL-2 and dacarbazine. In

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Table 4. Characteristics of complete responders

Patient	M1 stage	gp100	No. of ipi doses	Ipi dose level, mg/kg	Total ipi dose, mg/kg	Cycles of IL-2	Grade III/IV IRAE	Months until OR	Months until CR	CR duration	Alive
Protocol 1: ipi + gp100 (N = 56 patients)											
CR1	M1c	Yes	4	3	12	—	Yes	2	3	99+	Yes
CR2	M1b	Yes	4	3	12	—	Yes	2	3	94+	Yes
CR3	M1b	Yes	4	3 → 1	6	—	Yes	3	70	94+	Yes
CR4	M1c	Yes	11	3 → 1	13	—	Yes	1	42	88+	Yes
Protocol 2: ipi + IL-2 (N = 36 patients)											
CR5	M1a	—	9	0.3	2.7	6	No	2	45	89+	Yes
CR6	M1b	—	7	2	14	4	No	2	15	86+	Yes
CR7	M1c	—	3	3	9	2	Yes	2	4	83+	Yes
CR8	M1a	—	3	3	9	2	No	2	2	83+	Yes
CR9	M1a	—	9	3	27	6	No	2	25	79+	Yes
CR10	M1a	—	9	3	27	6	No	2	40	76+	Yes
Protocol 3: ipi (DE) ± gp100 (N = 85 patients)											
CR11	M1c	No	9	3	27	—	Yes	1	45	76+	Yes
CR12	M1c	No	7	3 → 5	31	—	Yes	3	21	74+	Yes
CR13	M1b	No	6	5 → 9	46	—	Yes	15	45	62+	Yes
CR14	M1a	No	8	5	40	—	No	2	55	54+	Yes
CR15	M1a	No	10	3 → 5 →9	70	—	No	4	30	42	No

Abbreviations: DE, intra-patient dose-escalation of ipilimumab; gp100, gp100:209-217(210M) and gp100:280-288(288V) peptides; ipi, ipilimumab.

August 2011, the FDA also approved vemurafenib, an inhibitor of mutated BRAF, for the treatment of metastatic melanoma (24). In a phase III trial randomizing 675 patients with BRAF-mutated metastatic melanoma to receive either vemurafenib or dacarbazine, vemurafenib therapy gave a 48% response rate and led to improved overall survival (25). Given that follow-up for this agent is limited (the longest patient follow-up in the phase III trial was <11 months), long-term response durability of vemurafenib has yet to be determined. Other developments are promising but are still in clinical investigation. ACT has shown OR rates up to 72% (26, 27) with long-term durability and potential cures in CRs (21, 28, 29); however, patients need to be medically fit for this rigorous inpatient treatment.

Although the initial overall response rate to ipilimumab is low in the phase III trial (OR = 7% of 540 patients who received either ipilimumab alone or ipilimumab plus gp100 peptides; ref. 13), with long-term follow-up, some of the nonresponders may become responders and some PRs may evolve into CRs. We have shown that it took an average of 30 months before our CRs achieved that status (Table 4). Most importantly, this agent is capable of inducing durable CRs which are potentially curative as shown by the plateauing of the survival curves (Fig. 1) with long-term follow-up; the level curves are reminiscent of patients with melanoma treated with high-dose IL-2 alone (6, 7). In addition, although IRAEs can occur, they are treatable provided the physician (and patient) is vigilant to screen for them and treat them early (9, 10, 17–19, 30); high-dose corticosteroids did not seem to abrogate the antitumor effects in those who experienced tumor regression. No initial survival difference was found in the phase III trial between those who received ipilimumab alone and those who received ipilimumab plus gp100 (13); we also did not see any difference in long-term results between those regimens (Tables 4 and 5).

It is surprising that 11% of patients who did not achieve CRs still seemed to derive long-term benefits (Table 5). For some patients, ipilimumab controlled most disease sites, allowing metastasectomies or other treatments to control the remaining limited sites. In other cases, some non-CR patients benefited from subsequent ACT which then mediated durable CRs (21). These interpretations must be tempered by the awareness that these are highly selected groups of patients whose long-term disease control status may be a reflection of their less aggressive tumor biology.

The 12% OR rate to ipilimumab in the 67 patients on protocols 1 and 3 who had received prior IL-2 (and progressed) before receiving ipilimumab is within the range of ORs seen in other ipilimumab protocols (9–11, 13). This suggests that the patients responding to ipilimumab are not the exact same ones who would have experienced a response to IL-2. Most striking is the long-term response update on the IL-2 plus ipilimumab trial (protocol 2) which resulted in a 17% rate of CRs (Table 2). Although the patients in

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Table 5. Characteristics of surviving noncomplete responders

Patient	M1 stage	Best ipi OR	Duration of OR, mo	gp100	No. of ipi doses	Ipi dose, mg/kg	Total ipi dose, mg/kg	Grade III/IV IRAE	Treatment(s) after ipilimumab	Current status	Follow-up, mo
Protocol 1: ipi + gp100 (N = 56 patients)											
1	M1b	PR	42	Yes	2	3	6	Yes	RFA of 1 recurrent lesion	NED	101
2	M1a	NR	—	Yes	8	3 → 1	10	No	IL-2 → PR → surgery	NED	91
3	M1c	NR	—	Yes	9	3 → 1	11	No	ACT with gp100-PBL → NR; chemotherapy; radiation; surgery	NED	84
Protocol 2: ipi + IL-2 (N = 36 patients)											
4	M1a	PR	5	—	3	3	9	Yes	ACT with TILs → CR	NED	80
5	M1c	NR	—	—	1	3	3	No	ACT with TILs → CR	NED	83
Protocol 3: ipi (DE) ± gp100 (N = 85 patients)											
6	M1c	PR	71+	Yes	10	5	50	Yes	None	Stable PR	71
7	M1c	PR	66+	Yes	4	5 → 9	28	No	None	Stable PR	66
8	M1a	PR	56+	No	6	5 → 9	46	No	None	Stable PR	56
9	M1a	PR	15	No	10	3 → 5 → 9	58	Yes	Surgery; chemotherapy	NED	82
10	M1c	PR	10	Yes	6	5	30	No	PD in brain treated with SRS; became NED 19 mos. post-ipi	NED	69
11	M1b	PR	7	No	6	5 → 9	46	Yes	Chemotherapy → CR	NED	66
12	M1b	PR	5	Yes	8	3	24	No	ACT with TILs → PR; repeat ACT → CR	NED	76
13	M1c	Mixed	—	No	10	3 → 5 → 9	70	No	ACT with TILs → CR	NED	73
14	M1c	Mixed	—	No	2	5	10	No	Surgery; radiation	NED	71
15	M1b	Mixed	—	No	4	3 → 5	16	No	PD in brain treated with SRS; PR at some sites; surgery	PD	69
16	M1a	NR	—	No	6	3 → 5 → 9	34	No	Bevacizumab; chemotherapy; BRAF inhibitor → PR	Stable PR	69
17	M1a	NR	—	No	2	3	6	Yes	IL-2 → stable → surgery	NED	67
18	M1c	NR	—	Yes	6	5 → 9	46	No	Biochemotherapy → PR	Stable PR	64

Abbreviations: DE, intrapatient dose-escalation of ipilimumab; gp100, gp100:209-217(210M) and gp100:280-288(288V) peptides; ipi, ipilimumab; NED, no evidence of disease; NR, no response; PBL, peripheral blood lymphocytes; PD, progressive disease; RFA, radiofrequency ablation; SRS, stereotactic radiosurgery; TILs, tumor-infiltrating lymphocytes.

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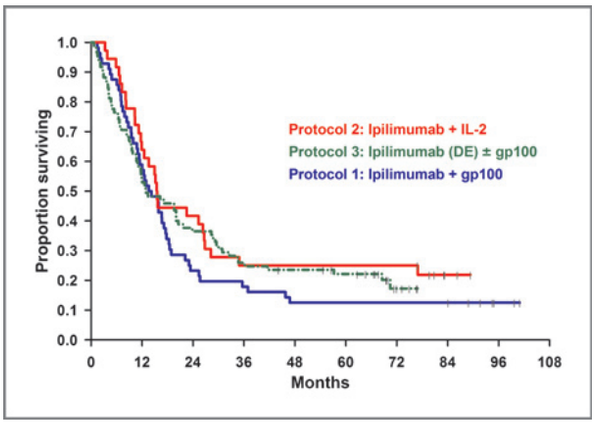

Figure 1. Overall survival for all patients, separated by protocol. DE, dose-escalation.

protocol 2 may theoretically be inclined to have a better response rate because they seem to be more treatment-naive and have less M1c disease (Table 1), the combination of IL-2 with ipilimumab may still have provided potentially an additive and conceivably a synergistic antitumor effect. Moreover, there was a trend toward a decreased rate of grade III/IV IRAEs in protocol 2 compared with the other 2 trials (17% vs. 29% and 32%; $P_2 = 0.14$, Fisher's exact test; Table 3). Only 1 (17%) of 6 CRs in protocol 2 developed a grade III/IV IRAE compared with 100% of CRs in protocol 1 and 60% of CRs in protocol 3 (Table 4). Although IL-2 was first described as a T-cell growth factor and has been shown to cause tumor regression in melanoma and renal cell cancer (6, 7), the exact mechanism by which it causes tumor regression is still not known. IL-2 causes T-cell expansion and augments T-cell cytolytic activity and the synthesis of other cytokines, including TNF-α, TNF-β, and IFN-γ, which may contribute to the antitumor activities and/or side effects of IL-2 administration (31, 32). However, IL-2 also increases the number of circulating $CD4^+CD25^{hi}FoxP3^+$ regulatory T cells (33, 34) which constitutively express CTLA-4 and are immunosuppressive (35). Thus, it is feasible that a timely and beneficial set of interactions may occur in patients receiving both ipilimumab and IL-2 in which IL-2 activates antitumor T cells but also increases the number of regulatory T cells which then decrease the activity of autoreactive T cells that may lead to IRAEs. Although this mechanism is hypothetical, the unusually high and durable CR rate in this group of patients who received ipilimumab concomitantly with IL-2 deserves further evaluation; a randomized trial comparing ipilimumab alone versus ipilimumab plus high-dose IL-2 needs to be conducted.

In summary, ipilimumab administration can lead to long-term and possibly curative regression of metastatic melanoma. The potential adverse events are serious and need to be proactively monitored and treated promptly. Given the relatively low tumor response rate of ipilimumab in comparison with other agents and modalities such as vemurafenib and ACT, further studies combining ipilimumab with other agents are needed to maximize its antitumor potential while decreasing its toxicities.

Disclosure of Potential Conflicts of Interest

No potential conflicts of interest were disclosed.

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References

1. Walunas TL, Bakker CY, Bluestone JA. CTLA-4 ligation blocks CD28-dependent T cell activation. J Exp Med 1996;183:2541–50.
2. Chambers CA, Krummel MF, Boitel B, Hurwitz A, Sullivan TJ, Fournier S, et al. The role of CTLA-4 in the regulation and initiation of T-cell responses. Immunol Rev 1996;153:27–46.
3. Phan GQ, Yang JC, Sherry RM, Hwu P, Topalian SL, Schwartzentruber DJ, et al. Cancer regression and autoimmunity induced by cytotoxic T lymphocyte-associated antigen 4 blockade in patients with metastatic melanoma. Proc Natl Acad Sci U S A 2003;100:8372–7.
4. Attia P, Phan GQ, Maker AV, Robinson MR, Quezado MM, Yang JC, et al. Autoimmunity correlates with tumor regression in patients with metastatic melanoma treated with anti-cytotoxic T-lymphocyte antigen-4. J Clin Oncol 2005;23:6043–53.
5. Maker AV, Yang JC, Sherry RM, Topalian SL, Kammula US, Royal RE, et al. Intrapatient dose escalation of anti-CTLA-4 antibody in patients with metastatic melanoma. J Immunother 2006;29:455–63.
6. Rosenberg SA, Yang JC, White DE, Steinberg SM. Durability of complete responses in patients with metastatic cancer treated with high-dose interleukin-2: identification of the antigens mediating response. Ann Surg 1998;228:307–19.
7. Atkins MB, Lotze MT, Dutcher JP, Fisher RI, Weiss G, Margolin K, et al. High-dose recombinant interleukin 2 therapy for patients with metastatic melanoma: analysis of 270 patients treated between 1985 and 1993. J Clin Oncol 1999;17:2105–16.
8. Maker AV, Phan GQ, Attia P, Yang JC, Sherry RM, Topalian SL, et al. Tumor regression and autoimmunity in patients treated with cytotoxic T lymphocyte-associated antigen 4 blockade and interleukin 2: a phase I/II study. Ann Surg Oncol 2005;12:1005–16.
9. Weber J, Thompson JA, Hamid O, Minor D, Amin A, Ron I, et al. A randomized, double-blind, placebo-controlled, phase II study comparing the tolerability and efficacy of ipilimumab administered with or without prophylactic budesonide in patients with unresectable stage III or IV melanoma. Clin Cancer Res 2009;15:5591–8.
10. O'Day SJ, Maio M, Chiarion-Sileni V, Gajewski TF, Pehamberger H, Bondarenko IN, et al. Efficacy and safety of ipilimumab monotherapy in patients with pretreated advanced melanoma: a multicenter single-arm phase II study. Ann Oncol 2010;21:1712–7.
11. Wolchok JD, Neyns B, Linette G, Negrier S, Lutzky J, Thomas L, et al. Ipilimumab monotherapy in patients with pretreated advanced melanoma: a randomised, double-blind, multicentre, phase 2, dose-ranging study. Lancet Oncol 2010;11:155–64.
12. Weber JS, O'Day S, Urba W, Powderly J, Nichol G, Yellin M, et al. Phase I/II study of ipilimumab for patients with metastatic melanoma. J Clin Oncol 2008;26:5950–6.

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13. Hodi FS, O'Day SJ, McDermott DF, Weber RW, Sosman JA, Haanen JB, et al. Improved survival with ipilimumab in patients with metastatic melanoma. N Engl J Med 2010;363:711–23.
14. Robert C, Thomas L, Bondarenko I, O'Day S, Weber J, Garbe C, et al. Ipilimumab plus dacarbazine for previously untreated metastatic melanoma. N Engl J Med 2011;364:2517–26.
15. Schwartzentruber DJ. Guidelines for the safe administration of high-dose interleukin-2. J Immunother 2001;24:287–93.
16. Therasse P, Arbuck SG, Eisenhauer EA, Wanders J, Kaplan RS, Rubinstein L, et al. New guidelines to evaluate the response to treatment in solid tumors. European Organization for Research and Treatment of Cancer, National Cancer Institute of the United States, National Cancer Institute of Canada. J Natl Cancer Inst 2000;92:205–16.
17. Robinson MR, Chan CC, Yang JC, Rubin BI, Gracia GJ, Sen HN, et al. Cytotoxic T lymphocyte-associated antigen 4 blockade in patients with metastatic melanoma: a new cause of uveitis. J Immunother 2004;27:478–9.
18. Blansfield JA, Beck KE, Tran K, Yang JC, Hughes MS, Kammula US, et al. Cytotoxic T-lymphocyte-associated antigen-4 blockage can induce autoimmune hypophysitis in patients with metastatic melanoma and renal cancer. J Immunother 2005;28:593–8.
19. Beck KE, Blansfield JA, Tran KQ, Feldman AL, Hughes MS, Royal RE, et al. Enterocolitis in patients with cancer after antibody blockade of cytotoxic T-lymphocyte-associated antigen 4. J Clin Oncol 2006;24:2283–9.
20. Phan GQ, Attia P, Steinberg SM, White DE, Rosenberg SA. Factors associated with response to high-dose interleukin-2 in patients with metastatic melanoma. J Clin Oncol 2001;19:3477–82.
21. Rosenberg SA, Yang JC, Sherry RM, Kammula US, Hughes MS, Phan GQ, et al. Durable complete responses in heavily pretreated patients with metastatic melanoma using T-cell transfer immunotherapy. Clin Cancer Res 2011;17:4550–7.
22. Balch CM, Gershenwald JE, Soong SJ, Thompson JF, Atkins MB, Byrd DR, et al. Final version of 2009 AJCC melanoma staging and classification. J Clin Oncol 2009;27:6199–206.
23. Jemal A, Siegel R, Ward E, Hao Y, Xu J, Thun MJ. Cancer statistics, 2009. CA Cancer J Clin 2009;59:225–49.
24. Flaherty KT, Puzanov I, Kim KB, Ribas A, McArthur GA, Sosman JA, et al. Inhibition of mutated, activated BRAF in metastatic melanoma. N Engl J Med 2010;363:809–19.
25. Chapman PB, Hauschild A, Robert C, Haanen JB, Ascierto P, Larkin J, et al. Improved survival with vemurafenib in melanoma with BRAF V600E mutation. N Engl J Med 2011;364:2507–16.
26. Dudley ME, Yang JC, Sherry R, Hughes MS, Royal R, Kammula U, et al. Adoptive cell therapy for patients with metastatic melanoma: evaluation of intensive myeloablative chemoradiation preparative regimens. J Clin Oncol 2008;26:5233–9.
27. Dudley ME, Wunderlich JR, Robbins PF, Yang JC, Hwu P, Schwartzentruber DJ, et al. Cancer regression and autoimmunity in patients after clonal repopulation with antitumor lymphocytes. Science 2002;298:850–4.
28. Rosenberg SA, Dudley ME. Adoptive cell therapy for the treatment of patients with metastatic melanoma. Curr Opin Immunol 2009;21:233–40.
29. Rosenberg SA, Restifo NP, Yang JC, Morgan RA, Dudley ME. Adoptive cell transfer: a clinical path to effective cancer immunotherapy. Nat Rev Cancer 2008;8:299–308.
30. Phan GQ, Weber JS, Sondak VK. CTLA-4 blockade with monoclonal antibodies in patients with metastatic cancer: surgical issues. Ann Surg Oncol 2008;15:3014–21.
31. Gemlo BT, Palladino MA Jr, Jaffe HS, Espevik TP, Rayner AA. Circulating cytokines in patients with metastatic cancer treated with recombinant interleukin 2 and lymphokine-activated killer cells. Cancer Res 1988;48:5864–7.
32. Romo de Vivar Chavez A, de Vera ME, Liang X, Lotze MT. The biology of interleukin-2 efficacy in the treatment of patients with renal cell carcinoma. Med Oncol 2009;26 Suppl 1:3–12.
33. Ahmadzadeh M, Rosenberg SA. IL-2 administration increases CD4+ CD25(hi) Foxp3+ regulatory T cells in cancer patients. Blood 2006;107:2409–14.
34. van der Vliet HJ, Koon HB, Yue SC, Uzunparmak B, Seery V, Gavin MA, et al. Effects of the administration of high-dose interleukin-2 on immunoregulatory cell subsets in patients with advanced melanoma and renal cell cancer. Clin Cancer Res 2007;13:2100–8.
35. Miyara M, Sakaguchi S. Human FoxP3(+)CD4(+) regulatory T cells: their knowns and unknowns. Immunol Cell Biol 2011;89:346–51.

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