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ORIGINAL RESEARCH

Anti-PD-1 antibody monotherapy versus anti-PD-1 plus anti-CTLA-4 combination therapy as first-line immunotherapy in unresectable or metastatic mucosal melanoma: a retrospective, multicenter study of 329 Japanese cases (JMAC study)

Y. Nakamura[1*], K. Namikawa[2], S. Yoshikawa[3], Y. Kiniwa[4], T. Maekawa[5], O. Yamasaki[6], T. Isei[7], S. Matsushita[8], M. Nomura[9], Y. Nakai[10], S. Fukushima[11], S. Saito[12], T. Takenouchi[13], R. Tanaka[14], H. Kato[15], A. Otsuka[16], T. Matsuya[17], N. Baba[18], K. Nagase[19], T. Inozume[20], N. Fujimoto[21], Y. Kuwatsuka[22], M. Onishi[23], T. Kaneko[24], T. Onuma[25], Y. Umeda[1,14], D. Ogata[2], A. Takahashi[2], M. Otsuka[3], Y. Teramoto[1] & N. Yamazaki[2]

[1]Department of Skin Oncology/Dermatology, Saitama Medical University International Medical Center, Saitama; [2]Department of Dermatologic Oncology, National Cancer Center Hospital, Tokyo; [3]Department of Dermatology, Shizuoka Cancer Center, Shizuoka; [4]Department of Dermatology, Shinshu University, Matsumoto; [5]Department of Dermatology, Jichi Medical University, Tochigi; [6]Department of Dermatology, Okayama University Graduate School of Medicine, Dentistry and Pharmaceutical Sciences, Okayama; [7]Department of Dermatologic Oncology, Osaka International Cancer Institute, Osaka; [8]Department of Dermato-Oncology/Dermatology, National Hospital Organization Kagoshima Medical Center, Kagoshima; [9]Department of Clinical Oncology, Kyoto University, Kyoto; [10]Department of Dermatology, Mie University, Tsu; [11]Department of Dermatology and Plastic Surgery, Faculty of Life Sciences, Kumamoto University, Kumamoto; [12]Department of Dermatology, Gunma University, Maebashi; [13]Department of Dermatology, Niigata Cancer Center, Niigata; [14]Department of Dermatology, Kawasaki Medical School, Kurashiki; [15]Department of Geriatric and Environmental Dermatology, Nagoya City University Graduate School of Medical Sciences, Nagoya; [16]Department of Dermatology, Kyoto University, Kyoto; [17]Department of Dermatology, Asahikawa Medical University, Asahikawa; [18]Department of Dermatology, Fukui University, Fukui; [19]Division of Dermatology, Department of Internal Medicine, Saga University, Saga; [20]Department of Dermatology, Chiba University, Chiba; [21]Department of Dermatology, Shiga University of Medical Science, Otsu; [22]Department of Dermatology, Nagasaki University, Nagasaki; [23]Department of Dermatology, Iwate Medical University, Morioka; [24]Department of Dermatology, Juntendo University Urayasu Hospital, Urayasu; [25]Department of Dermatology, Yamanashi University, Kofu, Japan

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Background: Anti-programmed cell death protein 1 (PD-1) antibody monotherapy (PD1) has led to favorable responses in advanced non-acral cutaneous melanoma among Caucasian populations; however, recent studies suggest that this therapy has limited efficacy in mucosal melanoma (MCM). Thus, advanced MCM patients are candidates for PD1 plus anti-cytotoxic T lymphocyte-associated antigen-4 (CTLA-4) combination therapy (PD1 + CTLA4). Data on the efficacy of immunotherapy in MCM, however, are limited. We aimed to compare the efficacies of PD1 and PD1 + CTLA4 in Japanese advanced MCM patients.

Patients and methods: We retrospectively assessed advanced MCM patients treated with PD1 or PD1 + CTLA4 at 24 Japanese institutions. Patient baseline characteristics, clinical responses (RECIST), progression-free survival (PFS), and overall survival (OS) were estimated using Kaplan−Meier analysis, and toxicity was assessed to estimate the efficacy and safety of PD1 and PD1 + CTLA4.

Results: Altogether, 329 patients with advanced MCM were included in this study. PD1 and PD1 + CTLA4 were used in 263 and 66 patients, respectively. Baseline characteristics were similar between both treatment groups, except for age (median age 71 versus 65 years; $P < 0.001$). No significant differences were observed between the PD1 and PD1 + CTLA4 groups with respect to objective response rate (26% versus 29%; $P = 0.26$) or PFS and OS (median PFS 5.9 months versus 6.8 months; $P = 0.55$, median OS 20.4 months versus 20.1 months; $P = 0.55$). Cox multivariate survival analysis revealed that PD1 + CTLA4 did not prolong PFS and OS (PFS: hazard ratio 0.83, 95% confidence interval 0.58-1.19, $P = 0.30$; OS: HR 0.89, 95% confidence interval 0.57-1.38, $P = 0.59$). The rate of ≥grade 3 immune-related adverse events was higher in the PD1 + CTLA4 group than in the PD1 group (53% versus 17%; $P < 0.001$).

Conclusions: First-line PD1 + CTLA4 demonstrated comparable clinical efficacy to PD1 in Japanese MCM patients, but with a higher rate of immune-related adverse events.

Key words: mucosal melanoma, anti-PD-1 antibody, anti-CTLA-4 antibody, nivolumab, pembrolizumab, ipilimumab

*Correspondence to: Prof. Yasuhiro Nakamura, Department of Skin Oncology/Dermatology, Comprehensive Cancer Center, Saitama Medical University International Medical Center, 1397-1 Yamane, Hidaka, Saitama 350-1298, Japan. Tel: +81-42-984-4111

E-mail: ynakamur@saitama-med.ac.jp (Y. Nakamura).

INTRODUCTION

Malignant melanoma originates from melanocytes located in the basal layer of the epidermis (non-acral and acral cutaneous melanoma: NACM and ACM, respectively), the uveal tract (uveal melanoma), and the mucosal epithelium (mucosal melanoma: MCM). MCM is a rare clinical subtype of melanoma, which accounts for ∼1% of all melanoma subtypes in the United States.[1] In Asia, MCM accounts for ∼15% to 27% of all melanoma subtypes, and it is the second most common subtype, followed by ACM.[2-4] Notably, advanced MCM (unresectable primary or metastatic) is the most common advanced-stage subtype in Japan, accounting for 28%-38% of all melanoma subtypes at this stage.[5-7] MCM is genetically, molecularly, and clinically different from NACM in that it has distinct oncogenic drivers with a 3%-15% infrequent rate of *BRAF* V600 mutation,[8] a lower tumor mutational burden,[9] and a highly aggressive phenotype with a poorer prognosis.[10]

Recent phase III, randomized trials investigating the effect of immune checkpoint blockades (ICBs), including anti-PD-1 antibodies (PD1) and PD1 combined with anti-CTLA-4 antibodies (PD1 + CTLA4), have described a good response and prolonged survival with these treatments, primarily for NACM.[11-13] Based on these clinical trials, ICBs are commonly applied for advanced melanoma as a standard of care. Conversely, it has been expected that the use of ICBs for MCM may have limited clinical efficacy due to the lower tumor mutational burden.[14,15] Clinically, several studies have indicated that PD1 exhibits a lower efficacy in MCM patients than in NACM patients [objective response rate (ORR): 23.3% versus 40.9%, median progression-free survival (PFS): 3.0 versus 6.2 months].[16] Other studies have reported similar trends for MCM.[16-18] Therefore, advanced MCM patients are strong candidates for PD1-based combined immunotherapy, such as PD1 + CTLA4,[16,17] or PD1 plus another combined treatment modality aimed at improving the low clinical efficacy of PD1. These studies have included small sample sizes, however, mainly from Caucasian populations. In this study, we therefore focused on comparing the efficacies of PD1 and PD1 + CTLA4 in Japanese patients with advanced MCM, using a large sample size from real-world settings to investigate whether PD1 + CTLA4 is superior to PD1 in terms of efficacy.

PATIENTS AND METHODS

Patients and study design

This multicenter, retrospective, observational study included Japanese patients with advanced, histologically diagnosed MCM who were ≥18 years of age. Patients with MCM arising from the head and neck (nasal cavity, nasal sinus, oral mucosa), genitourinary tract, and gastrointestinal tract including anorectum were included; however, patients with ocular melanomas involving the conjunctiva and uvea were excluded. Patients treated with PD1 or PD1 + CTLA4 as a first-line immunotherapy for advanced disease, between July 2014 and July 2020 at 24 Japanese institutions, were included. Patients who had a history of receiving PD1 as an adjuvant therapy were excluded. Melanomas were staged according to the 8th American Joint Committee on Cancer (AJCC) staging system for cutaneous melanoma,[19] as there is no integrated staging system for MCM at all anatomical sites. Data on patient baseline characteristics before ICBs, including age, gender, primary tumor site, Eastern Cooperative Oncology Group performance status (ECOG PS), AJCC stage, presence or absence of brain metastases, lactate dehydrogenase (LDH) level, the number of organ sites involved, and *BRAF* status, were extracted from medical charts. The ORR, PFS, and overall survival (OS) were considered as treatment outcomes.

This study was reviewed and approved by the institutional review boards and human research ethics committees of Saitama Medical University International Medical Center (IRB approval number: 20-109) and each participating institution. The study was conducted according to the ethical guidelines outlined in the Declaration of Helsinki. The requirements for written informed consent were waived, because this study retrospectively analyzed anonymized patient data.

Efficacy assessment

The co-primary outcomes were the ORR, PFS, and OS. Radiologic response or progression was assessed by board-certified radiologists or independent investigators at each institution, based on RECIST version 1.1.[20] The ORR was defined as the proportion of patients who showed a complete response (CR) or a partial response (PR). PFS and OS were defined from the time at which ICB administration was initiated to the radiologic or clinical progression of any tumors (PFS), death from any cause (OS), or last follow-up (PFS and OS).

Statistical analysis

The patient baseline characteristics are presented as frequencies and percentages and were compared between MCM patients treated with PD1 (PD1 group) and those treated with PD1 + CTLA4 (PD1 + CTLA4 group) using Fisher's exact test or Pearson's chi-square test for categorical variables, as appropriate, and the Mann−Whitney *U*-test for continuous variables. The ORR was calculated as the proportion in the all-patient cohort and each primary site cohort (head and neck, genitourinary tract, and gastrointestinal tract), and the difference between the PD1 and PD1 + CTLA4 groups was compared using Fisher's exact test. Patients who were alive at the last follow-up were censored. Kaplan−Meier analysis was used to estimate PFS and OS, which were expressed as median values with a two-sided 95% confidence interval (CI). The log-rank test was carried out to compare patient survival between the PD1 and PD1 + CTLA4 groups. The Cox multivariate proportional hazards model was used to analyze the variables that may influence the survival of patients. All data were analyzed using JMP version 14.2.0 or JMP Pro version16

(SAS Institute, Cary, NC). A P value <0.05 was considered statistically significant.

RESULTS

Patient baseline characteristics at ICB treatment initiation

The final study cohort included 329 Japanese patients with advanced MCM (head and neck 184 patients; urogenital tract 76 patients; gastrointestinal tract, 69 patients) (Supplementary Figure S1, available at https://doi.org/10.1016/j.esmoop.2021.100325 and Table 1). The numbers of patients treated with PD1 or PD1 + CTLA4 as a first-line immunotherapy were 263 and 66, respectively. The median age at the initiation of ICB administration was 70 years (interquartile range, 63-76 years), and there were more female (199, 60%) than male (130, 40%) patients. More patients with an ECOG PS of 0 (235, 71%) were included than patients with a PS $\geq$1. Before immunotherapy, half of the patients (163) had stage IV-M1c disease; there were very few patients with stage IV-M1d, including brain metastasis (13, 4%). There were more patients with LDH levels within the upper limit of normal (203, 62%) than patients with LDH levels over the normal upper limit. Approximately half of the cohort (150 patients, 46%) exhibited disease at a single organ site only. *BRAF* mutations were identified in only 7 patients (2%), although 37 patients (11%) did not undergo molecular testing.

Most patient characteristics in the cohort were similar between the PD1 and PD1 + CTLA4 groups, except for age ($P < 0.001$) (Table 1). In each primary site cohort, the patient characteristics were also similar between the PD1 and PD1 + CTLA4 groups (Supplementary Tables S1-S3, available at https://doi.org/10.1016/j.esmoop.2021.100325), apart from the higher age in the PD1 + CTLA4 group within the head and neck ($P = 0.004$) and gastrointestinal tract ($P = 0.02$) cohorts and a lower number of organs with tumor involvement ($P = 0.01$) in the PD1 + CTLA4 group in the head and neck cohort. No significant differences in patient characteristics between the two groups were observed in the urogenital cohort.

Overall response

The ORR in the patient cohort was 26% (CR, 7%; PR, 19%). Analysis of the ORRs between the PD1 and PD1 + CTLA4 groups revealed no statistical significance [26% (CR, 8%; PR, 18%) versus 29% (CR, 5%; PR, 24%), $P = 0.32$] (Table 2). With respect to ORRs in each primary site cohort, again, no

Table 1. Patient baseline characteristics

Baseline characteristics	No. of patients (%)			*P*
	Overall $n = 329$	PD1 $n = 263$	PD1 + CTLA4 $n = 66$	
Age				
Median age at ICB treatment initiation, years [interquartile range]	70 [63-76]	71 [65-77]	65 [58-73]	<0.001
Sex				
Female	199 (60)	161 (61)	38 (58)	0.67
Male	130 (40)	102 (39)	28 (42)	
Location				
Head and neck	184 (56)	152 (58)	32 (48)	0.32
Urogenital tract	76 (23)	57 (22)	19 (29)	
Gastrointestinal tract	69 (21)	54 (20)	15 (23)	
ECOG performance status				
0	235 (71)	185 (70)	50 (76)	0.45
≥1	94 (29)	78 (30)	16 (24)	
AJCC-TNM 8th Stage				
Unresectable stage II or III	70 (21)	58 (22)	12 (18)	0.83
Stage IV (M1a)	32 (10)	25 (10)	7 (11)	
Stage IV (M1b)	51 (16)	38 (14)	13 (20)	
Stage IV (M1c)	163 (50)	131 (50)	32 (48)	
Stage IV (M1d)	13 (4)	11 (4)	2 (3)	
Brain metastasis				
Absent	319 (97)	254 (97)	65 (98)	0.69
Present	10 (3)	9 (3)	1 (2)	
LDH level				
≤ULN	203 (62)	163 (62)	40 (61)	0.89
>ULN	126 (38)	100 (38)	26 (39)	
No. of organs involved				
1 Organ site	150 (46)	114 (43)	36 (55)	0.22
2-3 Organ sites	86 (26)	70 (27)	16 (24)	
≥4 Organ sites	93 (28)	79 (30)	14 (21)	
BRAF mutation				
Wild type	285 (87)	228 (87)	57 (86)	0.23
Mutation	7 (2)	4 (2)	3 (5)	
Not investigated	37 (11)	31 (12)	6 (9)	

AJCC-TNM, American Joint Committee on Cancer-Tumor—Node—Metastasis; ECOG, Eastern Cooperative Oncology Group; ICB, immune checkpoint blockade; LDH, lactate dehydrogenase; PD1, anti-programmed cell death protein 1 monotherapy; PD1 + CTLA4, anti-programmed cell death protein 1 plus anti-cytotoxic T lymphocyte-associated antigen-4 combination therapy; ULN, upper limit of normal.

Table 2. Overall response between the anti-PD-1 antibody monotherapy and anti-PD-1 plus anti-CTLA-4 combination therapy groups in the all-patient cohort

	No. of patients (%)			P
	Overall $n = 329$	PD1 $n = 263$	PD1 + CTLA4 $n = 66$	
Best overall response				
Complete response	24 (7)	21 (8)	3 (5)	0.32
Partial response	63 (19)	47 (18)	16 (24)	
Stable disease	80 (24)	60 (23)	20 (30)	
Progressive disease	157 (48)	130 (49)	27 (41)	
Unable to determine	5 (2)	5 (2)	0	

PD1, anti-programmed cell death protein 1 antibody monotherapy; PD1 + CTLA4, anti-programmed cell death protein 1 plus anti-cytotoxic T lymphocyte-associated antigen-4 combination therapy.

Table 3. Overall response between the anti-PD-1 antibody monotherapy and anti-PD-1 plus anti-CTLA-4 combination therapy groups in each primary site cohort

	No. of patients (%)			P
	Overall $n = 329$	PD1 $n = 263$	PD1 + CTLA4 $n = 66$	
Head and neck				
Complete response	12 (7)	11 (7)	1 (3)	0.36
Partial response	37 (20)	30 (20)	7 (22)	
Stable disease	50 (27)	37 (24)	13 (41)	
Progressive disease	83 (45)	72 (48)	11 (34)	
Unable to determine	2 (1)	2 (1)	0	
Urogenital tract				
Complete response	8 (11)	6 (11)	2 (10)	0.95
Partial response	10 (13)	7 (12)	3 (16)	
Stable disease	17 (22)	14 (25)	3 (16)	
Progressive disease	39 (51)	28 (49)	11 (58)	
Unable to determine	2 (3)	2 (3)	0	
Gastrointestinal tract				
Complete response	4 (6)	4 (7)	0	0.25
Partial response	16 (23)	10 (19)	6 (40)	
Stable disease	13 (19)	9 (17)	4 (27)	
Progressive disease	35 (51)	30 (55)	5 (33)	
Unable to determine	1 (1)	1 (2)	0	

PD1, anti-programmed cell death protein 1 antibody monotherapy; PD1 + CTLA4, anti-programmed cell death protein 1 plus anti-cytotoxic T lymphocyte-associated antigen-4 combination therapy.

statistical significance was observed between the PD1 and PD1 + CTLA4 groups [head and neck: 27% (CR, 7%; PR, 20%) versus 25% (CR, 3%; PR, 22%), $P = 0.36$; urogenital tract: 23% (CR, 11%; PR, 12%) versus 26% (CR, 10%; PR, 16%), $P = 0.95$; gastrointestinal tract: 26% (CR, 7%; PR, 19%) versus 40% (CR, 0%; PR, 40%), $P = 0.25$] (Table 3).

Estimation of PFS and OS

In the all-patient cohort, there was no significant difference in PFS or OS between the PD1 and PD1 + CTLA4 groups (median PFS, 5.9 versus 6.8 months, $P = 0.55$; median OS, 20.4 versus 20.1 months; $P = 0.55$) (Figure 1). The same result was observed in each primary tumor site cohort (head and neck: median PFS, 6.0 versus 7.7 months, $P = 0.47$; median OS, 18.9 versus 19.8 months, $P = 0.41$; urogenital tract: median PFS, 5.9 versus 4.6 months, $P = 0.87$; median OS, 25.6 months versus not-reached, $P = 0.72$; gastrointestinal tract: median PFS, 5.7 versus 5.0 months, $P = 0.72$; median OS, 17.8 versus 20.1 months, $P = 0.86$) (Figure 2). There was also no significant difference in survival between the three primary site cohorts treated with PD1 (PFS: $P > 0.99$; OS: $P > 0.99$) (Supplementary Figure S2 and B, available at https://doi.org/10.1016/j.esmoop.2021.100325) or PD1+CTLA4 (PFS: $P > 0.99$; OS: $P > 0.99$) (Supplementary Figure S2C and D, available at https://doi.org/10.1016/j.esmoop.2021.100325).

Cox multivariate analysis for PFS and OS

As there were several differences in patient baseline characteristics between the PD1 and PD1 + CTLA4 (Table 1) groups, a multivariate analysis using a Cox proportional hazards model was carried out. The results of the multivariate analysis indicated that younger age, poorer ECOG PS, and elevated LDH levels negatively affected the PFS [age of 1-year increase: hazard ratio (HR), 0.98, $P = 0.006$; ECOG PS $\geq$1: HR, 1.75, $P < 0.001$; elevated LDH: HR, 2.12, $P = 0.001$]. PD1 + CTLA4, however, did not positively impact PFS (selection of PD1 + CTLA4: HR, 0.83, $P = 0.32$) (Table 4).

Poorer ECOG PS, elevated LDH levels, and stage IV-M1b disease negatively affected the OS (ECOG PS $\geq$1: HR, 2.11, $P < 0.001$; elevated LDH: HR, 2.05, $P < 0.001$, stage IV-M1b: HR, 1.85, $P = 0.03$). PD1 + CTLA4, however, did not positively impact OS (PD1 + CTLA4: HR, 0.89, $P = 0.61$) (Table 4).

Second- or later-line salvage therapy after disease progression

After disease progression, second- or later-line therapy was initiated in 177 patients (67%) in the PD1 group and 20 patients (30%) in the PD1 + CTLA4 group (Supplementary Table S4, available at https://doi.org/10.1016/j.esmoop.2021.100325). Immunotherapy was the most common treatment approach (used for 48% of patients in the PD1 group and 11% of patients in the PD1 + CTLA4 group), followed by cytotoxic agents (used for 14% of patients in the PD1 group and 9% of patients in the PD1 + CTLA4 group). Ipilimumab (24%) or nivolumab plus ipilimumab combination therapy (15%) was commonly used as second- or later-line immunotherapy in the PD1 group. Very few patients received small molecular targeted therapies, including BRAF inhibitor-based therapy or KIT inhibitor therapy (used for 2% of patients in the PD1 group and 1.5% of patients in the PD1 + CTLA4 group) (Supplementary Table S4, available at https://doi.org/10.1016/j.esmoop.2021.100325).

Estimation of PFS and OS following disease progression in the PD1 group

As the selection of second-line salvage therapy after failure of PD1 may impact survival, Kaplan—Meier analysis was carried out for the evaluation of the efficacy of second-line ipilimumab, nivolumab plus ipilimumab, and cytotoxic

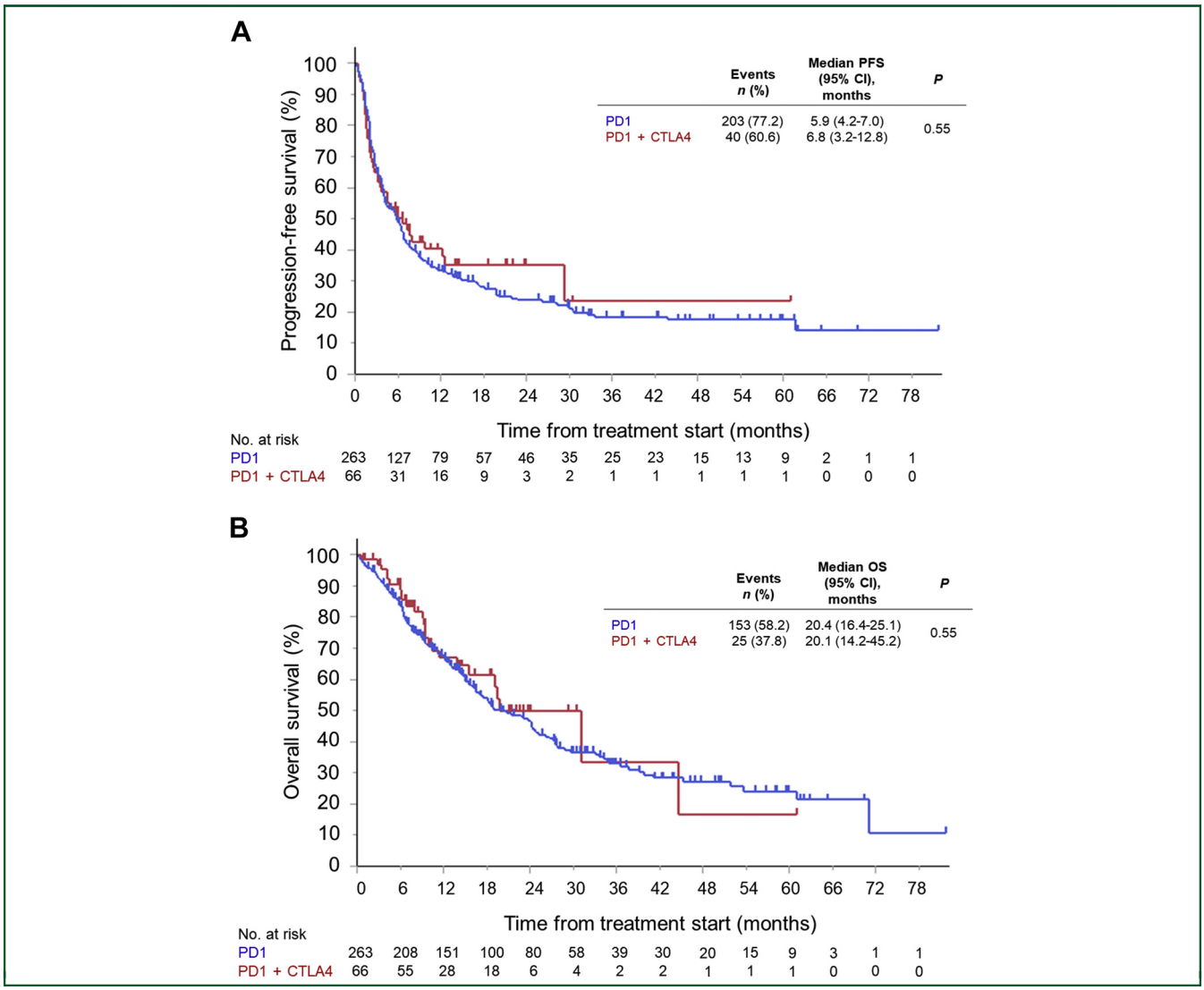

Figure 1. Kaplan–Meier survival estimates for the all-patient cohort treated with anti-PD-1 antibody monotherapy or anti-PD-1 plus anti-CTLA-4 combination therapy.

(A) Progression-free survival. (B) Overall survival.

CI, confidence interval; OS, overall survival; PD1, anti-programmed cell death protein 1 antibody monotherapy; PD1 + CTLA4, anti-programmed cell death protein 1 plus anti-cytotoxic T lymphocyte-associated antigen-4 combination therapy; PFS, progression-free survival.

agents. There was no significant difference in PFS or OS between the three treatment groups (PFS: nivolumab plus ipilimumab versus ipilimumab, ipilimumab versus cytotoxic agents, nivolumab plus ipilimumab versus cytotoxic agents, $P > 0.99$; OS: nivolumab plus ipilimumab versus ipilimumab, ipilimumab versus cytotoxic agents, $P > 0.99$, nivolumab plus ipilimumab versus cytotoxic agents, $P = 0.88$) (Supplementary Figure S3, available at https://doi.org/10.1016/j.esmoop.2021.100325).

Toxicity

In this study cohort, 17% (46 events) and 53% (35 events) of ≥grade 3 immune-related adverse events (irAEs) occurred in the PD1 and PD1 + CTLA4 groups ($P < 0.001$), respectively, which required treatment discontinuation. The irAEs of ≥grade 3 with an incidence rate of 5% or higher included pneumonitis (5%) in the PD1 group and increased aspartate aminotransferase/alanine aminotransferase level (30%), pneumonitis (6%), diarrhea/colitis (6%), and hypophysitis (6%) in the PD1 + CTLA4 group (Supplementary Table S5, available at https://doi.org/10.1016/j.esmoop.2021.100325).

DISCUSSION

This retrospective study analyzing real-world data demonstrated that, as a first-line immunotherapy, PD1 + CTLA4 did not improve ORR, PFS, or OS compared with PD1 in either the all-patient cohort or the various primary site cohorts in Japanese MCM patients. Our Cox multivariate analysis indicated that the selection of immunotherapy (PD1 or PD1 + CTLA4) did not have a positive impact on PFS or OS, with only ECOG PS and LDH level affecting PFS and OS among the patients in our cohort. Furthermore, the use of PD1 + CTLA4 as a second-line salvage therapy after failure of PD1 did not improve survival compared with that

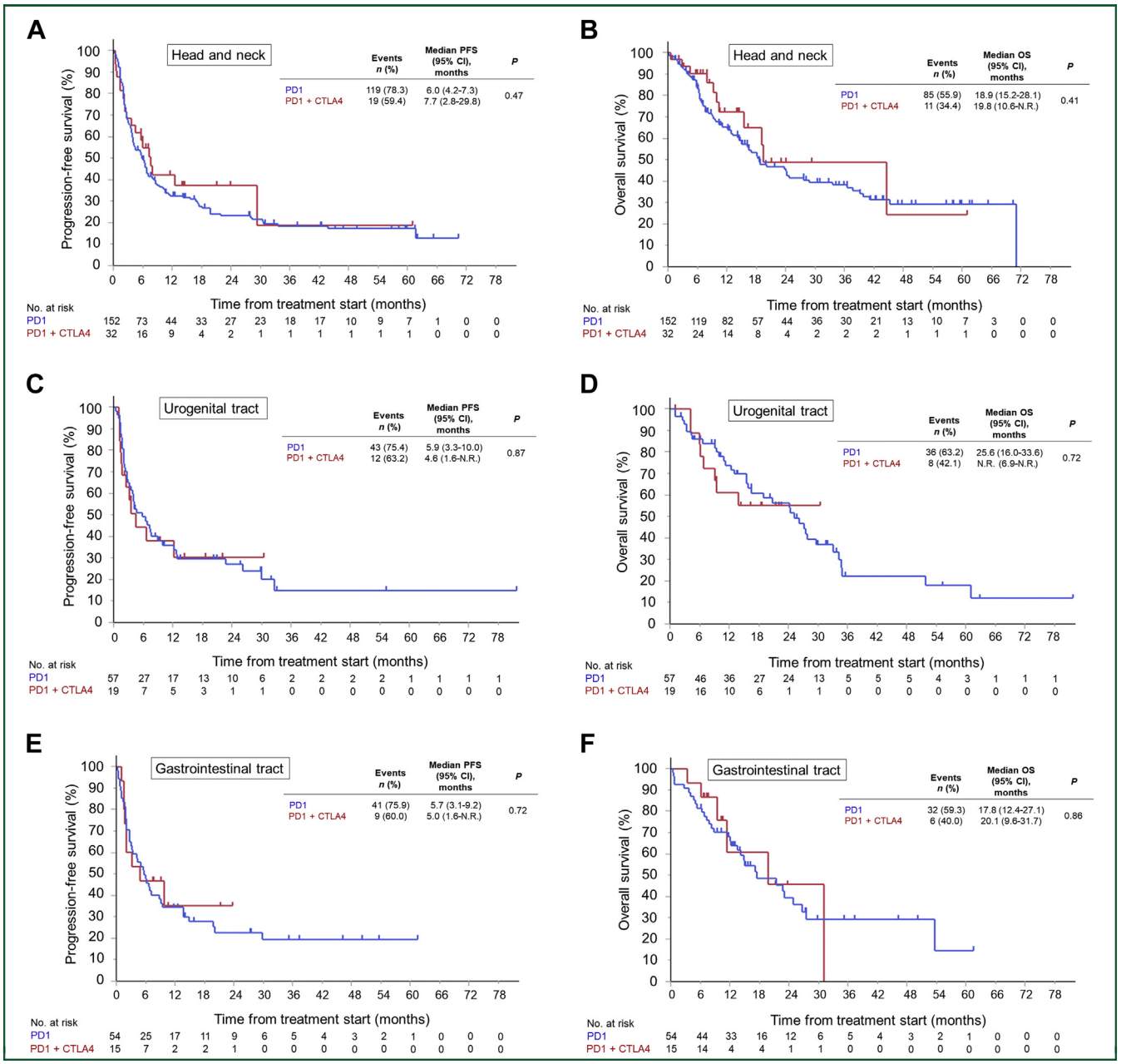

Figure 2. Kaplan–Meier survival estimates for each primary site cohort treated with anti-PD-1 antibody monotherapy or anti-PD-1 plus anti-CTLA-4 combination therapy.

(A) Progression-free survival in the head and neck cohort. (B) Overall survival in the head and neck cohort. (C) Progression-free survival in the urogenital tract cohort. (D) Overall survival in the urogenital cohort. (E) Progression-free survival in the gastrointestinal tract cohort. (F) Overall survival in the gastrointestinal tract cohort.
CI, confidence interval; OS, overall survival; PFS, progression-free survival; PD1, anti-programmed cell death protein 1 antibody monotherapy; PD1 + CTLA4, anti-programmed cell death protein 1 plus anti-cytotoxic T lymphocyte-associated antigen-4 combination therapy; N.R., not reached.

observed upon treatment with ipilimumab or cytotoxic agents.

ICBs are currently the preferred and standard procedure for treating advanced cutaneous melanoma worldwide, particularly for NACM patients. The outcomes of the randomized, phase III study CheckMate-067,[11] which compared the efficacy of nivolumab with that of nivolumab plus ipilimumab in the treatment of advanced melanoma (most being NACM cases), demonstrated that the combination therapy tended to exhibit a higher efficacy than nivolumab alone. In the nivolumab and nivolumab plus ipilimumab arms, the ORR was 45% and 58%, with a median PFS of 6.9 and 11.5 months and a median OS of 36.9 and >60 months, respectively.[11]

In comparison, evidence regarding the efficacy of ICBs for advanced MCM is limited due to a poor/fair study design and poor evidence quality as a result of small sample sizes (which are typically related to the rarity of MCM). Thus far, 24 studies have evaluated the efficacy of PD1 in MCM.[7,17,18] These studies (which used nivolumab, pembrolizumab, or toripalimab) had sample sizes ranging from 6 to 208 and described varying results: ORRs ranging from 9.5% to 50%, a

Table 4. The Cox multivariate proportional hazards model for progression-free survival and overall survival

	Progression-free survival			Overall survival		
	Hazard ratio	95% CI	*P*	Hazard ratio	95% CI	*P*
Age at ICB treatment initiation	0.98	0.97-1.00	0.006	1.00	0.98-1.01	0.74
Male sex	0.92	0.69-1.23	0.58	0.85	0.60-1.19	0.34
Location						
Head and neck	Reference					
Urogenital tract	1.28	0.89-1.84	0.18	1.12	0.74-1.69	0.58
Gastrointestinal tract	1.08	0.77-1.53	0.65	1.17	0.79-1.74	0.43
ECOG PS ≥1	1.75	1.31-2.34	<0.001	2.11	1.52-2.93	<0.001
Elevated LDH	2.12	1.60-2.79	0.001	2.05	1.48-2.82	<0.001
AJCC-TNM 8th stage						
Unresectable II or III	Reference					
Stage IV (M1a)	0.92	0.54-1.56	0.75	0.98	0.51-1.89	0.95
Stage IV (M1b)	1.11	0.69-1.78	0.68	1.85	1.07-3.23	0.03
Stage IV (M1c)	1.21	0.76-1.92	0.41	1.46	0.85-2.50	0.17
Stage IV (M1d)	1.07	0.50-2.30	0.86	1.22	0.52-2.86	0.65
No. of organs involved						
1 Organ site	Reference					
2-3 Organ sites	0.89	0.60-1.32	0.56	0.78	0.50-1.22	0.28
≥4 Organ sites	1.47	0.98-2.23	0.07	1.45	0.91-2.30	0.12
PD1 + CTLA4	0.83	0.58-1.20	0.32	0.89	0.57-1.39	0.61

AJCC-TNM, American Joint Committee on Cancer-Tumor—Node—Metastasis; CI, confidence interval; ECOG PS, Eastern Cooperative Oncology Group performance status; ICB, immune checkpoint blockade; LDH, lactate dehydrogenase; PD1 + CTLA4, anti-programmed cell death protein 1 plus anti-cytotoxic T lymphocyte-associated antigen-4 combination therapy.

median PFS of 1.4-10.2 months, and a median OS of 8.2-20.1 months. In a study with a larger sample size, a retrospective analysis of 208 Japanese MCM patients treated with nivolumab demonstrated a median OS of 11.3 months.[5] *Post hoc* analysis of KEYNOTE-001, 002, and 006, which included 84 Caucasian MCM patients treated with pembrolizumab, revealed an ORR of 19%, a median PFS of 2.8 months, and a median OS of 11.3 months.[21] These data suggest that the clinical efficacy of PD1 in MCM is clearly inferior to its efficacy in NACM.

To improve the lower clinical efficacy of PD1 in MCM, additional treatment options with PD1 should be explored; however, very few studies have examined PD1 in combination with other treatment modalities, and they have used small sample sizes. Four studies have tested the efficacy of PD1 plus radiotherapy, including a study recently reported by our group,[22-25] two studies have tested PD1 plus a vascular endothelial growth factor receptor (VEGFR) inhibitor,[26,27] and four studies explored the efficacy of PD1 + CTLA4.[6,16,17,28,29]

A retrospective study investigating the efficacy of pembrolizumab plus radiotherapy in 12 Korean MCM patients demonstrated a significantly higher 1-year infield local control (ILC) rate (94.1%) than that observed upon treatment with pembrolizumab alone (25%), but there was no significant increase in PFS and OS.[22] Another retrospective study of 10 Japanese MCM patients that evaluated pembrolizumab plus radiotherapy showed an ILC rate of 100% and a median PFS of 7.4 months.[24] A retrospective study that evaluated the efficacy of PD1 plus radiotherapy in seven Japanese MCM patients noted an ORR of 57.1% and a 1-year PFS of 50%.[23] These studies mainly focused on ILC rate and did not include sufficient data regarding survival compared with PD1. Conversely, our previous retrospective analysis of 171 Japanese MCM patients (PD1: 115 patients, PD1 plus radiotherapy: 56 patients) focused on survival and reported no significant difference in ORR, PFS, and OS between the PD1 and PD1 plus radiotherapy groups (ORR, 26% versus 27%, $P > 0.99$; median PFS, 6.2 versus 6.8 months; $P = 0.79$; median OS, 19.2 versus 23.1 months; $P = 0.70$).[25]

Assessments of PD1 plus the VEGFR inhibitor axitinib have included a single-institutional phase Ib trial that evaluated the efficacy of toripalimab plus axitinib in 33 Chinese patients with advanced MCM. This trial described an ORR of 48.3% and a median PFS of 7.5 months; the median OS was not reached after 18 months of follow-up.[26] In parallel, a retrospective study evaluated the efficacy of this combination as a first- or salvage-line treatment with larger sample sizes of 81 and 66 Chinese MCM patients, respectively. The ORR for all treatment-line and first-line therapy was 24.5% and 30%, respectively, with a median OS of 11.1 months.[27] These outcomes were lower than those from the phase Ib trial,[26] although more patients had poorer ECOG (ECOG ≥ 1: 51.7%) and elevated LDH levels (44.2%) in this retrospective study[27] than in the phase Ib trial.[26] It is unclear whether the combination of PD1 plus axitinib is superior to PD1 alone in terms of efficacy.

Similar to the small pool of studies that revealed the limited survival benefit of PD1 in combination with radiotherapy or axitinib, only a few studies have evaluated PD1 + CTLA4, and all of them had small sample sizes. A single-arm prospective study of 12 Japanese MCM patients treated with PD1 + CTLA4 showed an ORR of 33.3%, but the median PFS and OS were not reached during a median follow-up of 20.8 months.[28,29] A single-institutional retrospective study that included 16 Japanese MCM patients showed an ORR of 12.5% (25% among the 8 patients who received PD1 + CTLA4 as a first-line therapy) and a median PFS of 2.6 months; the median OS was not reached.[6] In a

Caucasian population, a pooled analysis of the CheckMate-067 and CheckMate-069 cohorts (35 MCM patients) showed that nivolumab plus ipilimumab displayed more favorable activity than nivolumab monotherapy, with an ORR of 37.1% and a median PFS of 5.9 months, although statistical analysis was not carried out.[16] Shoushtari et al.[17] analyzed the survival of advanced MCM patients who were included in the CheckMate-067 trial. In this study, 28 patients treated with nivolumab plus ipilimumab demonstrated more favorable survival outcomes than the 23 MCM patients treated with nivolumab only (median PFS, 5.8 versus 3.0 months; median OS, 22.7 versus 20.2 months), but these differences were not statistically significant.[17] Overall, the study data described above imply that the use of PD1 + CTLA4 for MCM may be more efficacious than the use of PD1; however, the present study (which evaluated a far larger sample size) suggests that the clinical efficacy of PD1 + CTLA4 is unlikely to exceed that of PD1, particularly as a higher rate of severe irAEs was still observed in the PD1 + CTLA4 group. To the best of our knowledge, the present study is the largest data analysis comparing the efficacy of PD1 with that of PD1 + CTLA4 in an Asian MCM cohort to date. Furthermore, the efficacy of ICBs for Japanese MCM patients was quite different from the outcomes for these drugs in the treatment of NACM in Caucasians.

There were several limitations in the present study. The study design was based on a retrospective analysis, with several uneven patient characteristics between the groups, both in the all-patient cohort and in each primary site cohort. The PD1 group included different doses and treatment intervals for the two ICBs (nivolumab and pembrolizumab). Data regarding the *KIT* and *NRAS* mutation status of patients were missing because molecular testing for these mutations is not covered by health insurance in Japan. In addition, use of the KIT inhibitors imatinib and sunitinib is not approved in Japan and is thus unavailable to MCM patients, although *KIT* mutations have been detected in 18% of Japanese MCM patients.[30] The status of programmed death-ligand 1 expression at the tumor cell surface was not included in our analyses because many of the patients in this study cohort were missing these data. The sample size was still small, and the follow-up period was short, particularly in the PD1 + CTLA4 group.

Despite these limitations, we did not identify a prolonged survival benefit with the use of PD1 + CTLA4 in Japanese patients with advanced MCM. Future studies will need to analyze larger sample sizes and longer follow-up periods in evaluations of PD1 + CTLA4. Research investigating immunotherapies in combination with novel agents will be essential to improve treatment outcomes for advanced MCM patients.

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