Supplementary Online Content

Reardon DA, Brandes AA, Omuro A, et al. Effect of nivolumab vs bevacizumab in patients with recurrent glioblastoma: the CheckMate 143 phase 3 randomized clinical trial [published online May 21, 2020]. *JAMA Oncol*. doi: 10.1001/jamaoncol.2020.1024

This supplementary material has been provided by the authors to give readers additional information about their work.

eMethods

Additional inclusion/exclusion criteria
Baseline laboratory values must meet the following criteria: white blood cells ≥ 2000/μL, neutrophils ≥ 1500/μL, platelets ≥ 100 × 10^3/μL, hemoglobin ≥ 9.0 g/dL, serum creatinine ≤ 1.5 × upper limit of normal (ULN) or creatinine clearance ≥ 40 mL/min (using the Cockcroft-Gault formula), aspartate aminotransferase and alanine aminotransferase ≤ 3 × ULN, and bilirubin ≤ 1.5 × ULN. Patients were also required to have a resting baseline oxygen saturation of ≥ 92%. Patients were not excluded if they were using NovoTTF-100L during the study.

Patients with prior or current autoimmune disease; extracranial metastatic or leptomeningeal disease; or prior inhibitor therapy targeting vascular endothelial growth factor, PD-1, or cytotoxic T-lymphocyte–associated protein were excluded. Patients with known tumor isocitrate dehydrogenase mutations; evidence of grade > 1 central nervous system hemorrhage; inadequately controlled hypertension, history of hypertensive crises, or clinically significant cardiovascular disease; known or suspected autoimmune disease; and history of gastrointestinal diverticulitis, perforation, or abscess were excluded. Other exclusion criteria were central nervous system disease unrelated to cancer unless adequately controlled, significant vascular disease (within 6 months prior to start of study treatment or any previous grade > 3 venous thromboembolism), history of pulmonary hemorrhage/hemoptysis (grade ≥ 2) within 1 month prior to randomization, or history or evidence of inherited bleeding diathesis. Additionally, patients with a history of intracranial abscess within 6 months prior to randomization or an active ulcer, untreated bone fracture, or serious, nonhealing wound were excluded. Patients with hepatitis B virus surface antigen or detectable hepatitis C virus RNA, HIV infection, or AIDS were excluded. Patients unable or unwilling to undergo contrast-enhanced magnetic resonance imaging of the head, with current or recent use of anticoagulants, or who had a surgical procedure within 28 days before the first study treatment were also excluded.

Randomization and masking
Patients were randomly assigned (1:1) via an interactive web response system with randomization stratified with a block size of 2 to control for the presence or absence of measurable disease as determined by the investigator. Bevacizumab was chosen as the comparator due to its approved use in the United States and several other countries in the recurrent setting based on the phase 2 noncomparative AVF3708g study.[1]

PD-L1 testing methods
Tumor tissue from diagnosis or from recurrence before initiation of study therapy was requested for all patients but not mandated per protocol. Fresh tumor biopsy was not required, and archival samples were accepted. For patients with available tumor samples, PD ligand-1 (PD-L1) tumor expression was determined retrospectively by a central laboratory using immunohistochemistry (PD-L1 IHC 28-8 pharmDx assay [Dako, Santa Clara, CA]). PD-L1 positivity was defined as membranous staining in ≥ 1% of tumor cells.

Statistical analysis
All randomized patients were included in baseline demographic and efficacy analyses; however, only patients with measurable disease at baseline were evaluable for best overall response. Patients who did not have measurable disease at baseline were not evaluable for response. PFS was estimated for each treatment group using Kaplan-Meier methodology, and ORR estimates and corresponding 95% CIs were provided by treatment group. CIs for ORR estimates were derived using the Clopper-Pearson method. Baseline patient characteristics and safety were characterized using descriptive statistics. Two-sided 95% CI for median OS were computed by the Brookmeyer and Crowley method. OS rates were derived from the Kaplan Meier estimate, along with their corresponding log-log transformed 95% CIs. A stratified Cox proportional hazards regression model was used to estimate hazard ratio, along with the 95% CI, between treatment groups. The software used for statistical analyses was SAS version 9.2.

A multivariable Cox regression model, stratified by the presence or absence of measurable disease, was used to estimate the treatment effect of the following covariates measured at baseline were included: age, sex, corticosteroid use (yes/no), Karnofsky performance status (≤ 80, > 80), MGMT status, log (baseline tumor load), and time from glioblastoma diagnosis to recurrence. Age, baseline tumor load, and time from diagnosis to recurrence were treated as continuous variables. These were modeled on linear scale, with the exception of baseline tumor load, which was analyzed on log scale. For this particular study, other transformations were not explored. Note that for time from

diagnosis to recurrence, there were 0 values so a log transformation may not be appropriate. Backward selection was used to eliminate covariates with a *P* value < 0.15 using a Wald statistic. Baseline factors (except for baseline tumor load) were prespecified as relevant to the assessment of prognostic effect on OS.

Sensitivity analyses

In the "assignment of not reported to methylated" analysis, the group of patients with *MGMT* status not reported was assigned to the *MGMT*-methylated group and the Kaplan-Meier analysis was reassessed; in the "assignment of not reported to unmethylated" analysis, the group of patients with *MGMT* status not reported was assigned to the *MGMT*-unmethylated group and the Kaplan-Meier analysis was reassessed. Similar sensitivity analyses were also performed to test associations of *MGMT* methylation status and baseline corticosteroid use with OS in both study groups.

eResults

Treatments
The median duration of study treatment was 2.3 months (range, 0-26.3 months) in the nivolumab group and 3.7 months (range, 0-25.8 months) in the bevacizumab group. The median number of doses received was 6.0 (range, 1-56 doses) with nivolumab and 9.0 (range, 1-53 doses) with bevacizumab.

Sensitivity analyses
Sensitivity analyses demonstrated a trend toward a longer mOS in patients with a methylated *MGMT* promoter treated with nivolumab rather than bevacizumab (eFigure 3 in the Supplement). Sensitivity analyses also supported the longer mOS observed in patients with a methylated *MGMT* promoter and no baseline corticosteroid use treated with nivolumab (eFigure 4 in the Supplement).

Objective response rates
With nivolumab, 2 patients (1.3%) achieved a CR, and 10 patients (6.5%) achieved a PR. Of the 12 patients who achieved a response with nivolumab, 10 were not receiving corticosteroids at baseline, 1 was receiving ≥ 4 mg/day of dexamethasone equivalent at baseline, and 1 was receiving < 4 mg/day of dexamethasone equivalent at baseline. Three responders had methylated *MGMT* promoter tumors, 1 had an unmethylated *MGMT* promoter tumor, and 8 had tumors with unknown *MGMT* status. In the bevacizumab group, 4 patients (2.6%) achieved a CR, and 32 patients (20.5%) achieved a PR. Of these responders, 21 were not receiving corticosteroids at baseline, whereas 3 were receiving ≥ 4 mg/day and 12 were receiving < 4 mg/day of dexamethasone equivalent. Nine responders had methylated *MGMT* promoter tumors, 12 had unmethylated *MGMT* promoter tumors, and 15 had tumors with unknown *MGMT* status. The median time to response was 3.0 months (range, 1.4-12.0 months) (Table 2) with nivolumab and 1.5 months (range, 1.2-6.5 months) with bevacizumab.

Subsequent therapy
Most patients in the nivolumab (66.3%) and bevacizumab (55.7%) groups received subsequent therapy (eTable 3 in the Supplement); 39.1% (nivolumab) and 38.4% (bevacizumab) of patients received an alkylating agent, and 42.4% (nivolumab) and 22.7% (bevacizumab) received bevacizumab. The mOS of patients in the nivolumab group who received subsequent bevacizumab therapy was comparable to the mOS of those who received subsequent alkylating agents (12.3 months [95% CI, 10.4-15.2 months] vs 14.6 months [95% CI, 12.3-17.0 months]). For patients in the bevacizumab arm, the mOS was 11.0 months (95% CI, 9.7-12.9 months) in patients receiving subsequent alkylating therapy.

Safety
Any-grade serious TRAEs occurred in 11.5% (nivolumab) and 9.1% (bevacizumab) of patients. The only serious TRAE reported in > 2% of patients was pulmonary embolism, which occurred in 3% of patients in the bevacizumab group and none in the nivolumab group.

Baseline corticosteroid use was similar between treatment groups (Table 1); patients in each group received a median dose of 2 mg/day in dexamethasone equivalents (interquartile ranges, 1.6-4.0 with nivolumab and 1.1-4.0 with bevacizumab) (eFigure 5 in the Supplement). However, nivolumab-treated patients received higher corticosteroid doses (for all purposes) than bevacizumab-treated patients to manage the disease while receiving treatment (eFigure 5 in the Supplement).

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eTable 1. Response Rates, Time to Response, and Duration of Response per Investigator Assessment

	Nivolumab n = 153[a]	Bevacizumab n = 156[a]
Objective response rate, n (%)	12 (7.8)	36 (23.1)
95% CI, %	4.1-13.3	16.7-30.5
Best overall response, No. (%)		
Complete response	2 (1.3)	4 (2.6)
Partial response	10 (6.5)	32 (20.5)
Stable disease	33 (21.6)	73 (46.8)
Progressive disease	107 (69.9)	26 (16.7)
Unable to determine	1 (0.7)	21 (13.5)
Not treated	1 (0.7)	16 (10.3)
Discontinued early due to toxicity	0	3 (1.9)
Other	0	2 (1.3)
Time to response Median (range), months	2.97 (1.4-12.0)	1.48 (1.2-6.5)
Duration of response Median (range), months	11.07 (0.6[b]-18.7)	5.29 (3.1-24.9[b])

[a] Patients who were evaluable for response.
[b] Censored observation.

eTable 2. Multivariable Analysis of Baseline Factors Associated With Overall Survival[a]

	Nivolumab	Bevacizumab
	HR (95% CI)[b]	
Corticosteroid use No vs yes	0.59 (0.36-0.95)	NS
Karnofsky performance status ≤ 80 vs > 80	2.25 (1.37-3.71)	1.70 (1.02-2.83)
MGMT status Methylated vs unmethylated	0.47 (0.29-0.78)	0.54 (0.32-0.89)
Age	NS	1.02 (1.00-1.05)

Abbreviations: HR, hazard ratio; MGMT, O^6-methylguanine DNA methyltransferase; NS, corresponding covariate not significant.
[a] Adjusted for age, sex, corticosteroid use (yes or no), performance status per the Karnofsky scale (≤ 80, > 80), *MGMT* status, log(baseline tumor load), and time from glioblastoma diagnosis to recurrence. Backward selection was used to eliminate covariates with a *P* value < 0.15 using a Wald statistic.
[b] HRs were estimated using a multivariable Cox regression model stratified by the presence or absence of measurable disease at baseline.

eTable 3. Subsequent Therapies

Patients, No. (%)	Nivolumab (n = 184)	Bevacizumab (n = 185)
Any subsequent therapy	122 (66.3)	103 (55.7)
Surgery	28 (15.2)	14 (7.6)
Radiotherapy	12 (6.5)	13 (7.0)
Systemic therapy	112 (60.9)	91 (49.2)
Immunotherapy	2 (1.1)	12 (6.5)
Nivolumab	2 (1.1)	5 (2.7)
Pembrolizumab	0	3 (1.6)
Investigational immunotherapy	0	3 (1.6)
Investigational antineoplastic drug	0	1 (0.5)
Bevacizumab	78 (42.4)	42 (22.7)
Alkylating agent	72 (39.1)	71 (38.4)
Lomustine	51 (27.7)	34 (18.4)
Temozolomide	18 (9.8)	22 (11.9)
Fotemustine	10 (5.4)	18 (9.7)
Carmustine	2 (1.1)	3 (1.6)
Procarbazine	2 (1.1)	2 (1.1)
Other cytotoxic therapy	28 (15.2)	24 (13.0)
Carboplatin	11 (6.0)	12 (6.5)
Etoposide	14 (7.6)	8 (4.3)
Irinotecan	6 (3.3)	6 (3.2)
Vincristine	2 (1.1)	2 (1.1)
Carboplatin/etoposide	0	1 (0.5)
Teniposide	0	1 (0.5)
Topotecan	0	1 (0.5)
Other targeted therapy	4 (2.2)	3 (1.6)
Axitinib	2 (1.1)	0
Vorinostat	0	2 (1.1)
Lapatinib	0	1 (0.5)
Temsirolimus	1 (0.5)	0
Vemurafenib	1 (0.5)	0
Other	11 (6.0)	8 (4.3)
Investigational antineoplastic/investigational drug	9 (4.9)	7 (3.8)
Mibefradil	0	1 (0.5)
Valganciclovir	1 (0.5)	0
Unassigned	1 (0.5)	0

eTable 4. Immune-Mediated Adverse Events Reported in ≥ 2% of Patients in Either Group

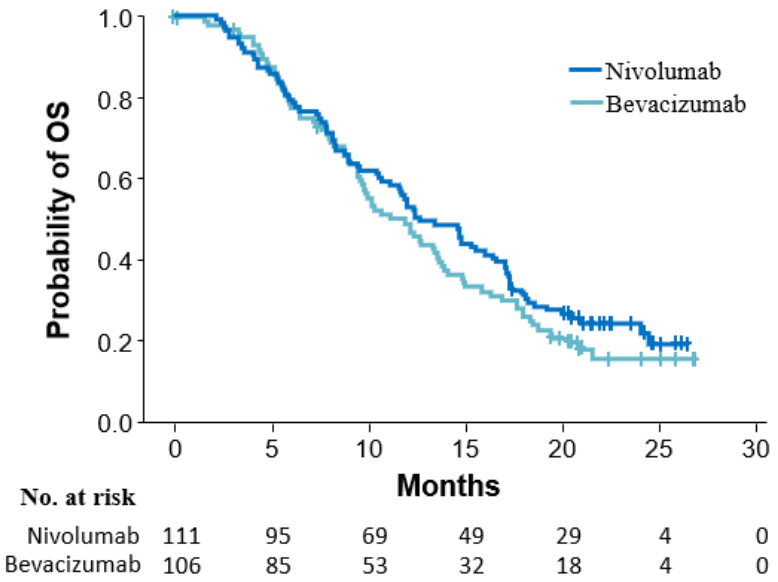
Patients, No. (%)	Nivolumab (n = 182)[a]		Bevacizumab (n = 165)[a]	
	Any Grade	Grade 3/4	Any Grade	Grade 3/4
Increased alanine aminotransferase	15 (8.2)	5 (2.7)	9 (5.5)	2 (1.2)
Increased aspartate aminotransferase	5 (2.7)	3 (1.6)	3 (1.8)	0
Diarrhea	27 (14.8)	1 (0.5)	13 (7.9)	0
Hyperthyroidism	5 (2.7)	0	0	0
Hypothyroidism	9 (4.9)	0	2 (1.2)	0
Maculopapular rash	7 (3.8)	0	3 (1.8)	0
Pneumonitis	6 (3.3)	2 (1.1)	3 (1.8)	2 (1.2)
Rash	17 (9.3)	0	7 (4.2)	0

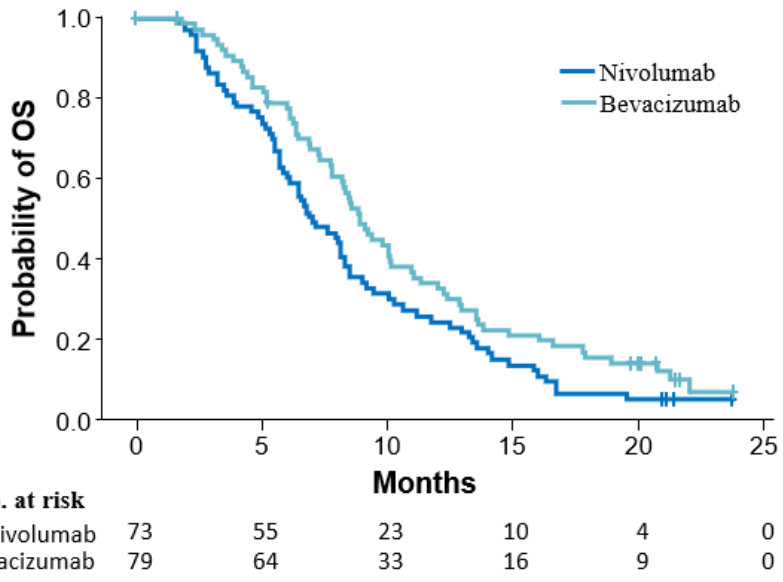
[a] Patients who received study treatment.

eFigure 1.

	No. of Events/ No. of Patients	Median OS (95% CI), months
Nivolumab	85/111	12.6 (10.7-16.3)
Bevacizumab	79/106	11.8 (9.5-13.6)

	No. of Events/ No. of Patients	Median OS (95% CI), months
Nivolumab	69/73	7.0 (5.8-8.2)
Bevacizumab	68/79	8.9 (7.8-10.2)





eFigure 1. OS Based on Baseline Corticosteroid Use. Panels A and B show the number of events, median OS, and the Kaplan-Meier curves for OS in patients treated with nivolumab or bevacizumab who did not receive corticosteroids at baseline (A) and in those who received corticosteroids at baseline (B). CIs were estimated using a Cox proportional hazards model. OS, overall survival.

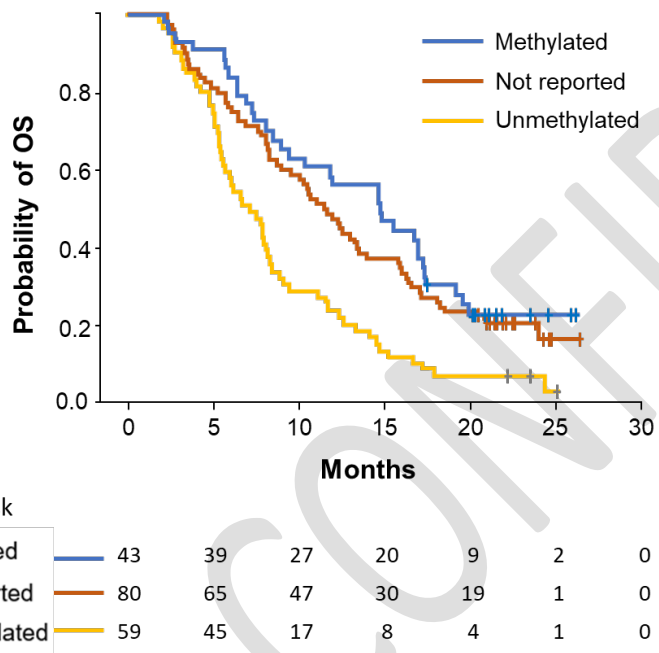
eFigure 2.

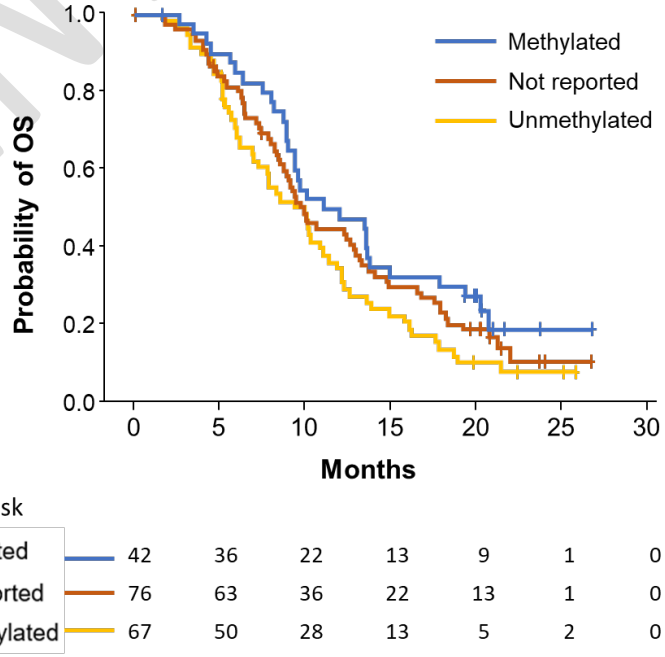
A. Nivolumab

	No. of Events/ No. of Patients	Median OS (95% CI), months
MGMT **methylated**	33/43	14.7 (8.9-17.2)
Not reported	64/80	11.5 (8.7-13.4)
MGMT **unmethylated**	56/59	7.2 (5.5-8.2)

B. Bevacizumab

	No. of Events/ No. of Patients	Median OS (95% CI), months
MGMT **methylated**	31/42	11.5 (8.9-13.8)
Not reported	63/76	9.9 (8.5-12.9)
MGMT **unmethylated**	53/67	9.7 (6.9-11.3)





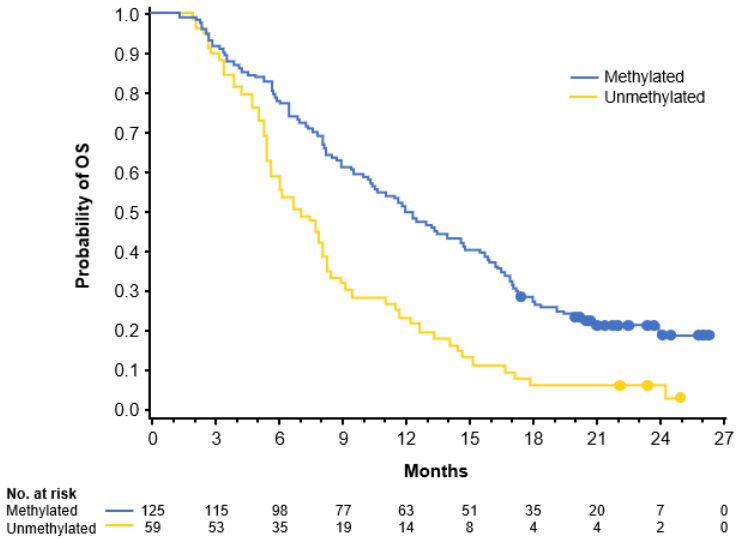
eFigure 2. OS based on *MGMT* Methylation Status in Either Group. Panels A and B show the Kaplan-Meier curves for OS by *MGMT* methylation status in patients treated with nivolumab (A) or bevacizumab (B). Symbols indicate censored observations. CIs were estimated using a Cox proportional hazards model. *MGMT*, O6-methylguanine DNA methyltransferase; and OS, overall survival.

eFigure 3.

Nivolumab

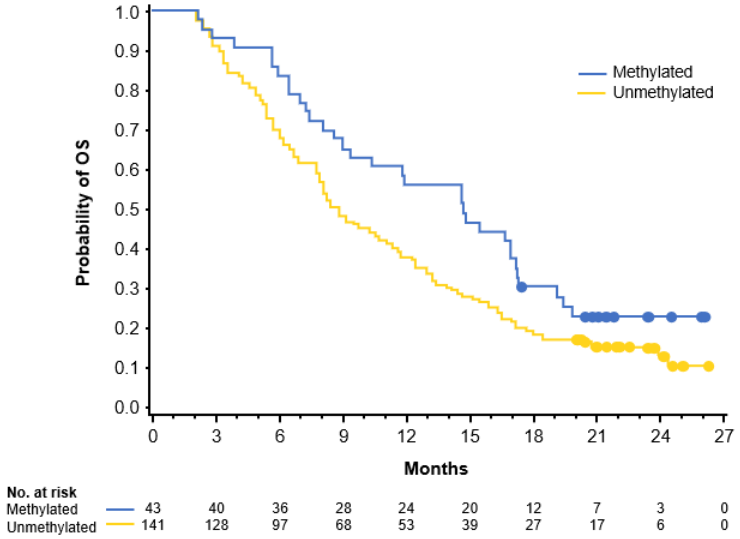
A.

	No. of Events/ No. of Patients	Median OS (95% CI), months
MGMT **methylated**	98/125	12.3 (10.3-14.8)
MGMT **unmethylated**	56/59	7.2 (5.5-8.2)



B.

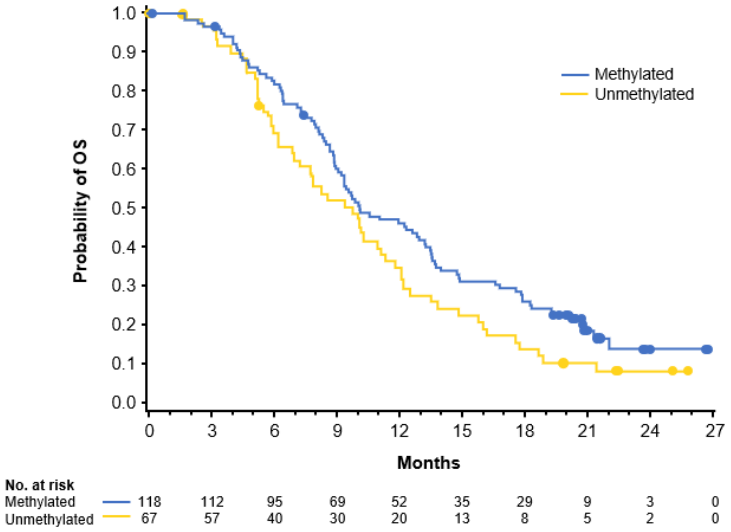
	No. of Events/ No. of Patients	Median OS (95% CI), months
MGMT **methylated**	33/43	14.7 (8.9-17.2)
MGMT **unmethylated**	121/141	8.7 (7.9-11.0)



Bevacizumab

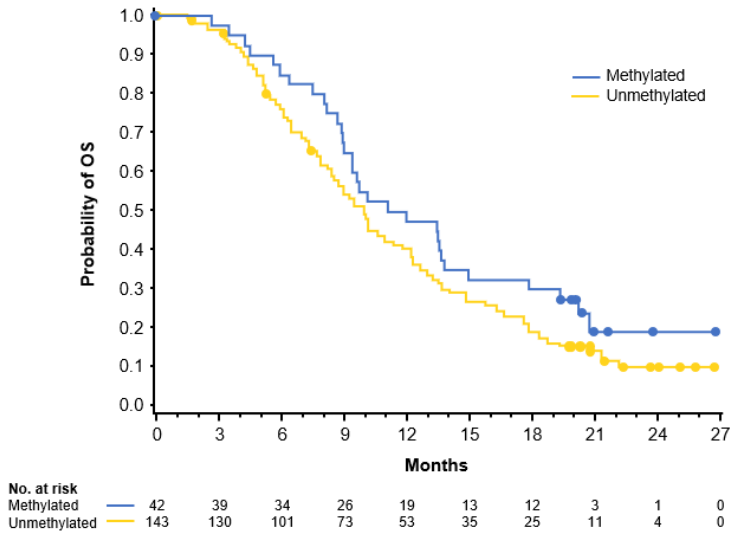
C.

	No. of Events/ No. of Patients	Median OS (95% CI), months
MGMT **methylated**	94/118	10.1 (9.1-13.2)
MGMT **unmethylated**	53/67	9.8 (6.9-11.3)



D.

	No. of Events/ No. of Patients	Median OS (95% CI), months
MGMT **methylated**	31/42	11.5 (8.9-13.8)
MGMT **unmethylated**	116/143	9.8 (8.4-11.3)



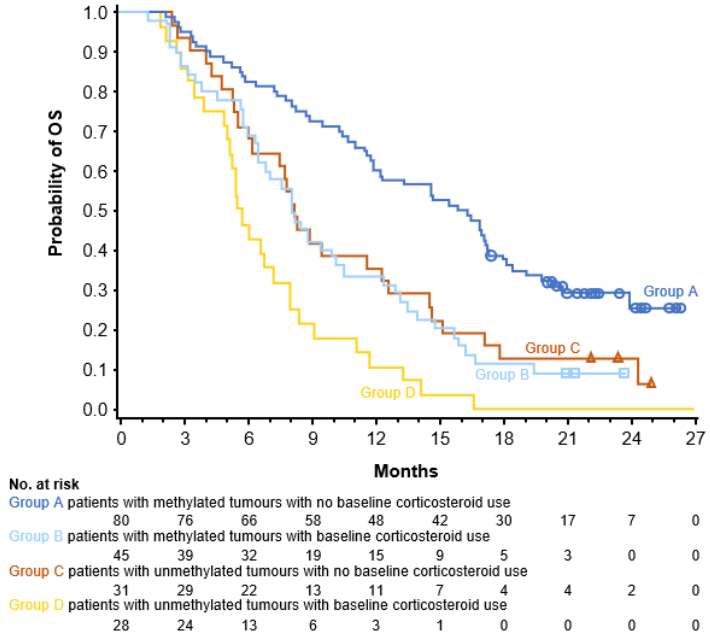
eFigure 3. Sensitivity Analysis of the Association of *MGMT* Promoter Methylation Status With OS. Kaplan-Meier curves for OS per *MGMT* sensitivity analyses. Panel A shows the "assignment of not reported to methylated" analysis, in which the *MGMT* not reported group was assigned to the *MGMT*-methylated group and the Kaplan-Meier analysis was reassessed in the nivolumab group. Panel B shows the "assignment of not reported to unmethylated" analysis, in which the *MGMT* not reported group was assigned to the *MGMT*-unmethylated group and the Kaplan-Meier analysis was reassessed in the nivolumab group. Panel C shows the "assignment of not reported to methylated" analysis, in which the *MGMT* not reported group was assigned to the *MGMT*-methylated group and the Kaplan-Meier analysis was reassessed in the bevacizumab group. Panel D shows the "assignment of not reported to unmethylated" analysis, in which the *MGMT* not reported group was assigned to the *MGMT*-unmethylated group and the Kaplan-Meier analysis was reassessed in the bevacizumab group. Symbols indicate censored observations. CIs were estimated using a Cox proportional hazards model. *MGMT* indicates O^6-methylguanine DNA methyltransferase and OS, overall survival.

eFigure 4.

Nivolumab

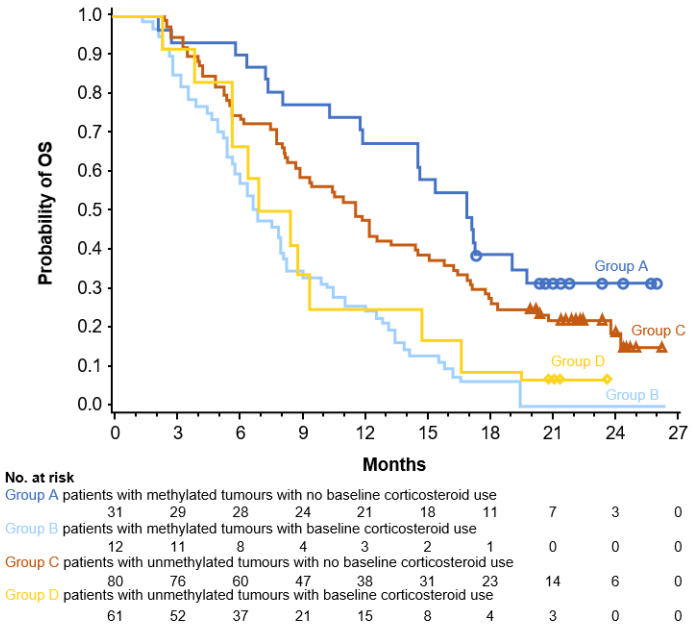
A.

	No. of Events/No. of Patients	Median OS (95% CI), months
MGMT **methylated; no baseline corticosteroid use**	57/80	16.1 (11.9-17.3)
MGMT **methylated; baseline corticosteroid use**	41/45	8.2 (6.5-10.3)
MGMT **unmethylated; no baseline corticosteroid use**	28/31	8.2 (6.1-12.3)
MGMT **unmethylated; baseline corticosteroid use**	28/28	5.6 (5.0-7.2)



B.

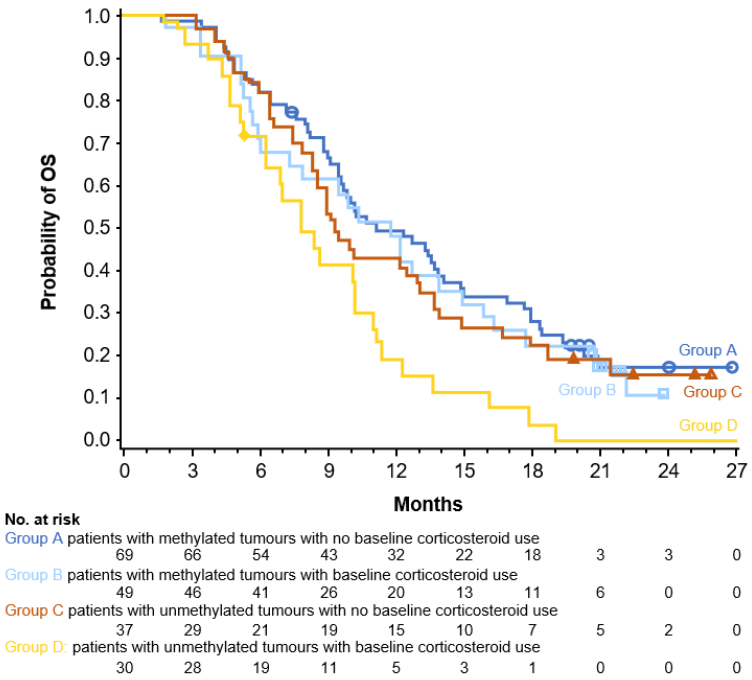
	No. of Events/No. of Patients	Median OS (95% CI), months
***MGMT* methylated; no baseline corticosteroid use**	21/31	17.0 (11.9-19.8)
***MGMT* methylated; baseline corticosteroid use**	12/12	7.7 (3.9-14.8)
***MGMT* unmethylated; no baseline corticosteroid use**	64/80	11.6 (8.7-14.7)
***MGMT* unmethylated; baseline corticosteroid use**	57/61	6.9 (5.7-8.2)



Bevacizumab

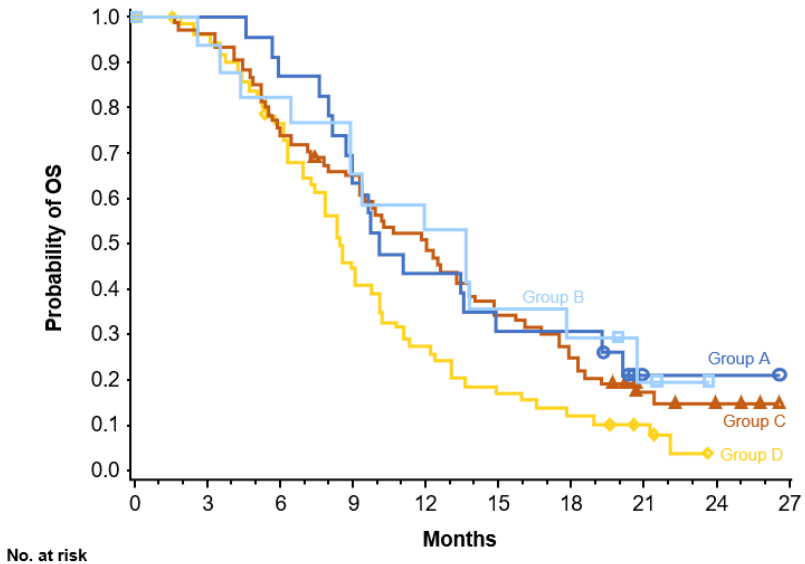
C.

	No. of Events/No. of Patients	Median OS (95% CI), months
***MGMT* methylated; no baseline corticosteroid use**	53/69	11.1 (9.4-14.0)
***MGMT* methylated; baseline corticosteroid use**	41/49	9.2 (8.3-12.9)
***MGMT* unmethylated; no baseline corticosteroid use**	26/37	11.8 (6.0-14.9)
***MGMT* unmethylated; baseline corticosteroid use**	27/30	7.8 (6.1-10.2)



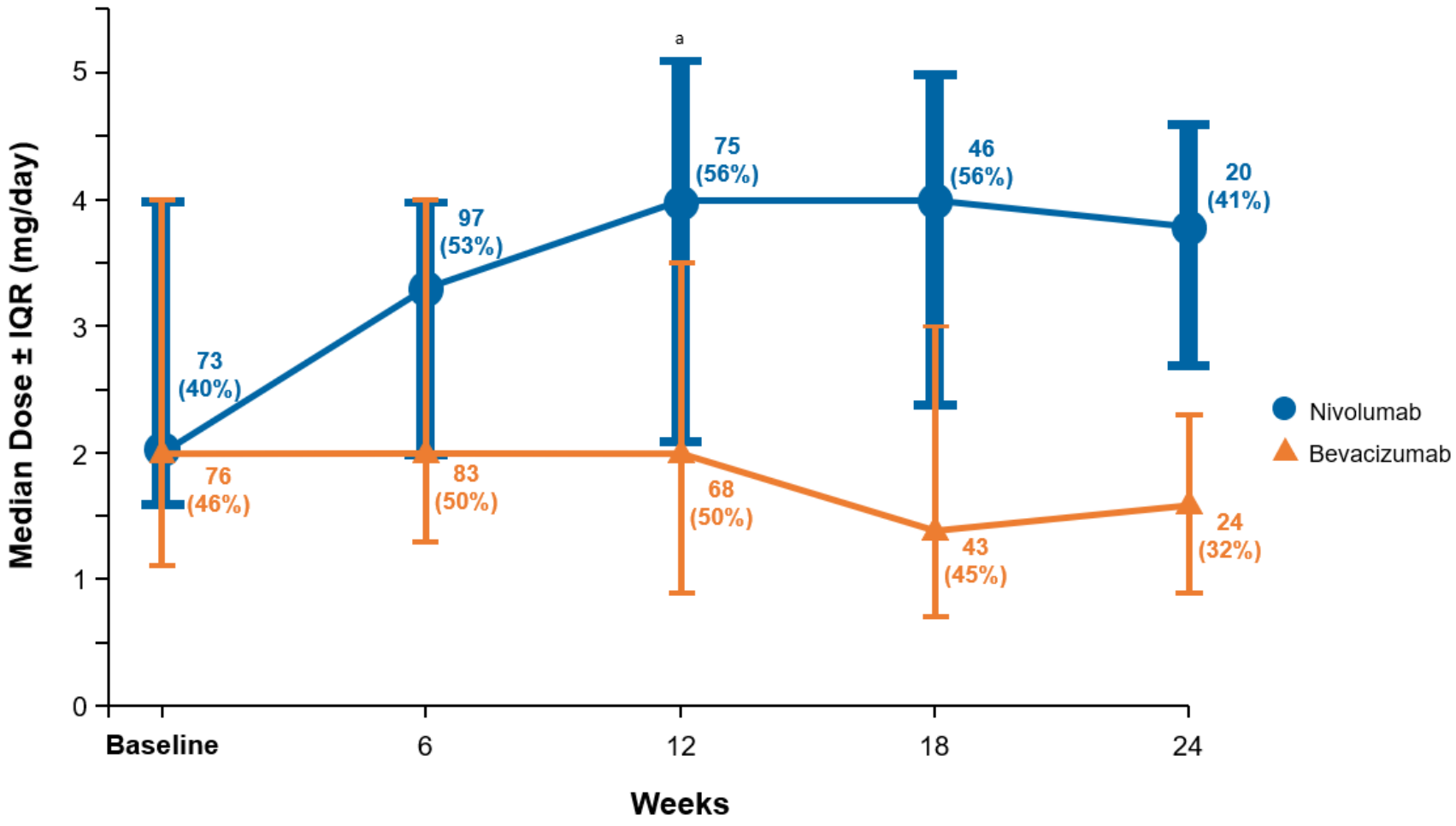
D.

	No. of Events/No. of Patients	Median OS (95% CI), months
***MGMT* methylated; no baseline corticosteroid use**	18/25	10.1 (8.7-14.9)
***MGMT* methylated; baseline corticosteroid use**	13/17	13.5 (6.4-20.7)
***MGMT* unmethylated; no baseline corticosteroid use**	61/81	12.1 (9.4-13.8)
***MGMT* unmethylated; baseline corticosteroid use**	55/62	8.4 (7.3-10.0)



eFigure 4. Sensitivity Analysis of the Association of *MGMT* Promoter Methylation Status and Baseline Corticosteroid Use With OS. Kaplan-Meier curves for OS per *MGMT* sensitivity analyses. Panel A shows the "assignment of not reported to methylated" analysis, in which the *MGMT* not reported group was assigned to the *MGMT*-methylated group and the Kaplan-Meier analysis was reassessed in the nivolumab group. Panel B shows the "assignment of not reported to unmethylated" analysis, in which the *MGMT* not reported group was assigned to the *MGMT*-unmethylated and the Kaplan-Meier analysis was reassessed in the nivolumab group. Panel C shows the "assignment of not reported to methylated" analysis, in which the *MGMT* not reported group was assigned to the *MGMT*-methylated group and the Kaplan-Meier analysis was reassessed in the bevacizumab group. Panel D shows the "assignment of not reported to unmethylated" analysis, in which the *MGMT* not reported group was assigned to the *MGMT*-unmethylated group and the Kaplan-Meier analysis was reassessed in the bevacizumab group. Symbols indicate censored observations. CIs were estimated using a Cox proportional hazards model. *MGMT* indicates O^6-methylguanine DNA methyltransferase and OS, overall survival.

eFigure 5.



eFigure 5. Corticosteroid Use in Dexamethasone Equivalents in the Nivolumab and Bevacizumab Treatment Groups. Plot shows median dose ± IQR (mg/day) of corticosteroid use (dexamethasone equivalents) by patients at baseline and during treatment with nivolumab (circles) or bevacizumab (triangles). The number of patients who received corticosteroids and the proportion of on-treatment patients who received corticosteroids are indicated at each data point. [a] Patients in the nivolumab group were given higher doses of corticosteroids—most notably around week 12—than patients in the bevacizumab group, which could be attributed to immuno-oncology–related adverse events or disease progression. Most immuno-oncology–related adverse events were observed around week 12 of treatment and may have led to increased use of corticosteroids for management. IQR indicates interquartile range.

Reference
1. Friedman HS, Prados MD, Wen PY, et al. Bevacizumab alone and in combination with irinotecan in recurrent glioblastoma. *J Clin Oncol*. 2009;27(28):4733-4740.