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PHASE I STUDIES

A phase 1 study of PF-06840003, an oral indoleamine 2,3-dioxygenase 1 (IDO1) inhibitor in patients with recurrent malignant glioma

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Summary

Background PF-06840003 is a highly selective indoleamine 2, 3-dioxygenase (IDO1) inhibitor with antitumor effects in preclinical models. This first-in-human phase 1 study evaluated safety, pharmacokinetics/pharmacodynamics, and preliminary efficacy in recurrent malignant glioma to determine the maximum tolerated dose (MTD) or recommended phase 2 dose (RP2D).
Methods Patients ($N = 17$) received oral PF-06840003 in four dose-escalation groups: 125 mg once-daily (QD; $n = 2$); 250 mg QD ($n = 4$); 250 mg twice-daily (BID; $n = 3$); 500 mg BID ($n = 8$). A modified toxicity probability interval method determined the MTD.
Results Four patients experienced serious adverse events (SAEs); one with treatment-related SAEs (grade 4 alanine and aspartate aminotransferase elevations). The dose-limiting toxicity (DLT) rate at 500 mg BID was 12.5% ($n = 1/8$); the MTD was not reached. Following PF-06840003 dosing, median time to maximum plasma concentration for the active enantiomer PF-06840002 was 1.5–3.0 hr and mean elimination half-life was 2 to 4 hr (Cycle 1 Day 1). Urinary recovery of PF-06840002 was low (< 1%). At 500 mg BID, maximum mean percentage inhibition of ^{13}C10 kynurenine vs endogenous kynurenine was 75% vs 24%. PF-06840002 CSF-to-plasma ratio was 1.00. Disease control occurred in eight patients (47%). Mean duration of stable disease (SD) was 32.1 (12.1–72.3) weeks. Two patients with SD discontinued the study at 450 and 561 days and continued PF-06840003 on compassionate use.
Conclusion PF-06840003 up to 500 mg BID was generally well tolerated with evidence of a pharmacodynamic effect and durable clinical benefit in a subset of patients with recurrent malignant glioma. ClinicalTrials.gov, NCT02764151, registered April 2016.

Keywords Glioblastoma · IDO1 · Immuno-oncology · Kynurenine · Malignant glioma, tryptophan catabolism

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Introduction

Malignant gliomas (World Health Organization [WHO] grade III–IV tumors), such as anaplastic oligodendroglioma, anaplastic astrocytoma, and glioblastoma (GBM), are associated with poor long-term survival and there is a clear unmet need for new therapies [1–5].

Indoleamine 2, 3-dioxygenase (IDO1) is a heme-containing dioxygenase that catalyzes the oxidation of tryptophan (trp) to N-formyl kynurenine (kyn) in the first and rate-limiting step of trp catabolism [6]. N-formyl kyn is converted to kyn and other immunologically active metabolites via the kyn pathway. Kyn triggers inflammatory signals via its binding to the aryl hydrocarbon receptor, a transcription factor involved in pro-inflammatory pathways and xenobiotic responses associated with cancer and inflammatory carcinogenesis [7].

IDO1 expression is often high in the tumor microenvironment, typically in response to inflammatory stimuli [8]. The enzymatic activity of IDO1 is associated with suppression of T-cell responses [9] both through depletion of trp and production of kyn [10, 11], and is correlated with poor prognosis in several cancer indications [12, 13]. As such, IDO1 is a target of interest for cancer immunotherapy. In a study of 75 gliomas, IDO1 was reported to be expressed in 72 (96%) [14]. IDO1 expression level correlated with prognosis, as observed in other studies [15]. The selective nature of IDO1 expression in malignant glioma provides a high potential for targeting specificity.

PF-06840003, a highly selective IDO1 inhibitor, is a racemic mixture of active (PF-06840002) and inactive (PF-06840001) enantiomers that spontaneously epimerize to each other in plasma [16]. PF-06840003 monotherapy demonstrated an antitumor benefit in humanized mice bearing a human breast tumor and in syngeneic tumor models, although inhibition of tumor growth in some models was variable or transient [16]. Collectively, in vitro and in vivo pharmacokinetic (PK)/pharmacodynamic (PD) tumor growth inhibition studies in BALB/c mice suggested that near maximal IDO1 inhibition (90% relative to baseline) was required to achieve maximum tumor growth inhibition (Pfizer, data on file). PF-06840003 in combination with anti–programmed cell death-ligand 1 (PD-L1) demonstrated greater in vivo efficacy than PF-06840003 or anti-PD-L1 alone [16]. Good central nervous system (CNS) penetration in rats has also been reported for PF-06840003 [17] and data from preclinical studies led to a predicted half-life in humans of approximately 19 hr, providing justification for initiating evaluation in humans using a once-daily (QD) dosing regimen [17].

In this first-in-human phase 1 study, we evaluated the safety, tolerability, PK, PD, and preliminary antitumor activity of PF-06840003 in patients with recurrent malignant glioma. As the efficacy of PF-06840003 monotherapy was expected to be limited, the use of PD biomarkers alongside PK to determine the recommended phase 2 dose (RP2D) was an important component of the phase 1 study.

Patients and methods

Study design

This study was a phase 1, open-label, multicenter, dose-escalation study of single-agent PF-06840003 sponsored by Pfizer (ClinicalTrials.gov NCT02764151). Patients were enrolled between September 9, 2016 and September 6, 2017, and the study was completed (last patient/last visit) on December 26, 2018. The study was approved by the institutional review board or independent ethics committee of each participating center and followed the Declaration of Helsinki and International Council for Harmonization Good Clinical Practice guidelines. All patients or legally authorized representatives provided written informed consent.

Patients

Adults (≥ 18 years of age) with Karnofsky performance score ≥ 70% and a recurrent malignant glioma (GBM at the time of 1st or 2nd recurrence, or anaplastic glioma at 1st–4th recurrence) were eligible if they had adequate renal, hepatic, and hematopoietic function and were on a stable/decreasing dose of corticosteroids for at least 7 days prior to registration.

Exclusion criteria included a history of CNS hemorrhage within 6 months of registration, prior anti-angiogenic therapy (eg, bevacizumab) within 12 months of registration, or dexamethasone > 2 mg/day. Patients were required to be at least 3 months from prior radiotherapy (unless there was a new area of enhancement consistent with recurrent tumor outside the radiation field or unequivocal histologic confirmation of tumor progression), and 4 weeks from either surgery or cytotoxic chemotherapy (6 weeks for nitrosoureas). Patients on strong cytochrome P450 (CYP)1A2 inhibitors or inducers were excluded.

Treatment

Sequential cohorts of patients received escalating doses of PF-06840003, with a starting dose of 125 mg orally QD. PF-06840003 was administered QD in the 125-mg dose group, QD or twice daily (BID) in two separate 250-mg dose groups, and BID in the 500-mg dose group. Treatment was to continue until disease progression, unacceptable toxicity, or patient refusal, whichever occurred first.

Endpoints

The primary goal was determination of safety and RP2D. The primary endpoint was the incidence and grade of treatment-emergent adverse events (TEAEs), including dose-limiting toxicities (DLTs). Secondary and exploratory endpoints included laboratory abnormalities, vital signs, PK parameters, PD effects, progression-free survival (PFS), overall survival, response rate and disease control rate (DCR) at 8 and 24 weeks using both Macdonald [18] and immunotherapy response assessment in neuro-oncology (iRANO) criteria [19].

Study assessments

Safety

Adverse events (AEs) were graded according to National Cancer Institute Common Terminology Criteria for Adverse Events, version 4.03. The following AEs were considered DLTs if they were attributable to study drug and occurred in the first cycle of treatment (28 days): grade 4 neutropenia; febrile neutropenia (defined as absolute neutrophil count < 1000/mm^3 with a single temperature > 38.3 °C or a sustained temperature ≥ 38 °C for more than 1 hr); grade ≥ 3 neutropenia with infection; grade ≥ 3 thrombocytopenia with bleeding; grade 4 thrombocytopenia; any toxicity attributable to PF-06840003 that resulted in administration of less than 80% of the planned doses during Cycle 1; grade 4 non-hematologic AE; grade 3 AE lasting > 7 days despite optimal supportive care; grade 3 CNS AE regardless of duration; grade 3 corrected QT prolongation (QTc > 500 msec) if persisting after correction of any reversible causes; concurrent aspartate aminotransferase (AST) or alanine aminotransferase (ALT) over three times the upper limit of normal (ULN); and total bilirubin over twice the ULN.

Pharmacokinetics

Blood samples for the evaluation of PK parameters were collected at predefined time points during each 28-day cycle as follows: Cycle 1 Day 1 (predose; 1-, 2-, 4-, 6-, 8-, and 24-hr postdose), Day 4 (predose), Day 8 (predose), and Day 15 (predose; 1-, 2-, 4-, 6-, 8-, and 24-hr postdose); Cycle 2 Day 1 (predose); Cycle 3 onwards, Day 1 (predose) and Day 15 (predose); and at the end of treatment (EOT). Blood samples were assayed for PF-06840002 and PF-06840001 using a validated analytical method in compliance with Pfizer standard operating procedures. The lower limits of quantitation (LLOQ) for PF-0684002 and PF-06840001 were 4.91 ng/mL and 5.02 ng/mL, respectively. Standard PK parameters, including the maximum plasma concentration, time to maximum plasma concentration (T_{max}), and area under the plasma concentration–time curve (AUC) for PF-06840002 and PF-06840001 were estimated using non-compartmental analysis. Where data permitted, AUC to infinity (AUC_{inf}), terminal elimination half-life ($t_{1/2}$), oral plasma clearance, apparent volume of distribution, accumulation ratio, and linearity ratio were also estimated. Urine was analyzed to estimate the fraction of drug excreted unchanged in urine (Ae%) and renal clearance for PF-6840002 and PF-06840001. Cerebrospinal fluid (CSF) samples were collected by lumbar puncture at screening and Cycle 1 Day 15 (predose) for estimation of steady-state trough level ratio between CSF and plasma in willing patients.

Pharmacodynamic assessments

Blood samples (~ 4 mL of whole blood) for the evaluation of circulating trp and kyn were collected at predefined time points in Cycles 1 and 2 and EOT. A liquid chromatography with tandem mass spectrometry assay was validated for quantitation of endogenous trp, endogenous kyn, $^{13}C_{11}$ trp, and $^{13}C_{10}$ kyn levels in human plasma. The LLOQ was 0.8 μM for trp ($^{13}C_{11}$ trp or endogenous trp) and 0.02 μM for kyn ($^{13}C_{10}$ kyn or endogenous kyn).

Efficacy

Magnetic resonance imaging scans were collected at screening and every 8 weeks and repeated following completion of treatment if > 4 weeks had passed since the last scan. Responses were designated by the treating investigator (locally) using both Macdonald criteria (primarily) [18] and iRANO criteria (additionally) [19].

Statistical analyses

A modified toxicity probability interval method was used to determine the maximum tolerated dose (MTD), where target toxicity rate (pT) was defined as 27.5% with 5% uncertainty (0.225 < pT < 0.325). The target sample size for each cohort was 2 to 4 and the actual number of patients treated at each dose varied. Enrollment of no more than 72 DLT-evaluable patients was planned to provide a reliable and accurate estimate of the MTD.

Baseline characteristics and patient demographics were summarized by dose level using the full analysis set (all enrolled patients). Efficacy was assessed using the full analysis set. Binary endpoints (including overall response rate, DCR) were summarized descriptively. Time to event analyses (PFS and overall survival) were calculated by the Kaplan-Meier method [20].

Safety assessment was performed for enrolled patients who received at least one dose of study treatment. A

binary variable was created at the patient level to indicate whether or not a patient had experienced a DLT. Safety endpoints were summarized descriptively by dose level. PK analysis was performed for enrolled patients who were treated and had sufficient information to estimate at least one of the PK parameters of interest.

Results

Patients and treatment

A total of 17 patients received PF-06840003 in four dose-escalation groups: 125 mg QD ($n = 2$), 250 mg QD ($n = 4$),

Table 1 Patient demographics and baseline characteristics

	PF-06840003 dose			
	125 mg QD 125 mg QD (n = 2)	**250 mg QD250 mg QD (n = 4)**	**250 mg BID250 mg BID (n = 3)**	**500 mg BID (n = 8)**
Gender, n				
Male	1	4	2	5
Female	1	0	1	3
Age subgroup, n				
18–44 years	1	2	1	1
45–64 years	1	1	2	3
≥ 65 years	0	1	0	4
Age, mean (StdD), years	44.5 (19.1)	51.8 (14.5)	49.3 (12.1)	58.1 (14.6)
Range	31–58	36–65	38–62	36–79
KPS score at screening, n				
90–100	1	3	0	5
80	1	1	3	3
Disease, n				
Anaplastic astrocytoma (grade III)	1	0	1	0
Anaplastic oligodendroglioma (grade III)	0	1	0	0
Glioblastoma (grade IV)	1	3	2	8
Time from original high-grade glioma diagnosis, mean, years				
Anaplastic astrocytoma	1.8	NA	0.1	NA
Anaplastic oligodendroglioma	NA	7.0	NA	NA
Glioblastoma	0.9	1.9	0.7	1.1
No. of surgeries prior to enrollment				
None	0	0	0	0
1	1	1	0	4
2	1	3	2	4
3	0	0	1	0
Prior VEGF therapies				
Yes	0	1	0	0
No	0	0	0	0
Baseline dexamethasone use				
Yes	1	0	0	0
No	1	4	3[a]	8
Measurable disease present[b], n (%)				
Yes	2 (100.0)	3 (75.0)	3 (100.0)	6 (75.0)
No	0	1 (25.0)	0	2 (25.0)

[a] One patient stopped dexamethasone use 22 days before onset of study treatment.

[b] At least one target lesion with two perpendicular diameters ≥ 10 mm.

BID, twice daily; KPS, Karnofsky Performance Status; N, not available; QD, once daily; StdD, standard deviation; VEGF, vascular endothelial growth factor

250 mg BID ($n = 3$), and 500 mg BID ($n = 8$). Most patients were male (71%) and enrolled after multiple prior therapies (Table 1). Median duration of treatment was 14.1 weeks, 10.1 weeks, 24.1 weeks, and 8.1 weeks in the 125-mg QD, 250-mg QD, 250-mg BID, and 500-mg BID cohorts, respectively.

Safety

All patients experienced TEAEs, the most common of which were anemia and fatigue ($n = 9$ [52.9%] for each; Table 2); 14 patients experienced treatment-related AEs, the most common of which were fatigue ($n = 8$; 47.1%) and anemia ($n = 6$; 35.3%). Most treatment-related AEs were grade ≤ 2 (Table 3). One patient (250-mg QD group) had a treatment-related grade 3 increase in ALT and one patient (500-mg BID group) had a grade 4 increase in both ALT and AST that was also considered treatment-related. Two cases of decreased ventricular ejection fraction (one each in the 250-mg QD [grade 2] and 500-mg BID [grade 3] groups) were recorded, neither of which required a PF-06840003 dose adjustment.

Serious AEs were reported in four patients, but only one patient had serious AEs considered treatment-related, which were grade 4 increases in ALT and AST. This grade 4 increase, which occurred in a patient in the 500-mg BID group, was the only DLT; the same patient also had a lung infection and was receiving levofloxacin and paracetamol, which were temporarily withdrawn. The DLT rate at 500 mg BID was 12.5% (one DLT out of eight patients), which was less than the targeted DLT rate of 27.5%. There were no reports of permanent study drug discontinuations or deaths during the study. Due to the low number of DLTs at the highest dose tested, the MTD was not reached.

Pharmacokinetics

Median plasma concentration-time profiles for the active enantiomer PF-06840002 following single and multiple oral doses of PF-06840003 are shown in Fig. 1. Following single oral administration of PF-06840003 (125 mg QD to 500 mg

Table 2 Treatment-emergent adverse events (≥ 10% of total patients)

	PF-06840003 dose				
TEAE (all grades), n (%)	**125 mg QD (n = 2)**	**250 mg QD (n = 4)**	**250 mg BID (n = 3)**	**500 mg BID (n = 8)**	**Total (N = 17)**
Anemia	1 (50.0)	2 (50.0)	3 (100.0)	3 (37.5)	9 (52.9)
Fatigue	1 (50.0)	3 (75.0)	1 (33.3)	4 (50.0)	9 (52.9)
Aphasia	1 (50.0)	0	0	4 (50.0)	5 (29.4)
Headache	0	2 (50.0)	1 (33.3)	2 (25.0)	5 (29.4)
Hyperglycemia	0	3 (75.0)	0	2 (25.0)	5 (29.4)
Hypocalcemia	0	0	3 (100.0)	1 (12.5)	4 (23.5)
Hypokalemia	0	0	2 (66.7)	3 (37.5)	5 (29.4)
WBC count decreased	0	1 (25.0)	2 (66.7)	1 (12.5)	4 (23.5)
ALK increased	0	2 (50.0)	0	1 (12.5)	3 (17.6)
Decreased appetite	1 (50.0)	0	0	2 (25.0)	3 (17.6)
Dysarthria	0	0	1 (33.3)	2 (25.0)	3 (17.6)
Fall	0	0	2 (66.7)	1 (12.5)	3 (17.6)
Hyponatremia	0	0	2 (66.7)	1 (12.5)	3 (17.6)
Lymphocyte count decreased	0	0	2 (66.7)	1 (12.5)	3 (17.6)
Muscular weakness	1 (50.0)	0	0	2 (25.0)	3 (17.6)
Pyramidal tract syndrome	0	0	2 (66.7)	1 (12.5)	3 (17.6)
Upper respiratory tract infection	1 (50.0)	0	0	2 (25.0)	3 (17.6)
Blood creatinine increased	0	0	0	2 (25.0)	2 (11.8)
Confusional state	0	0	0	2 (25.0)	2 (11.8)
Diarrhea	0	0	0	2 (25.0)	2 (11.8)
Myalgia	0	0	0	2 (25.0)	2 (11.8)
Pain in extremity	0	0	0	2 (25.0)	2 (11.8)
Tinnitus	0	0	0	2 (25.0)	2 (11.8)

ALK, alkaline phosphatase; BID, twice daily; QD, once daily; TEAE, treatment-emergent adverse event; WBC, white blood cell

Table 3 Incidence and grade of treatment-related TEAEs (≥ 10% of total patients)

	PF-06840003 dose				
Treatment-related TEAE, n	**125 mg QD (n = 2)**	**250 mg QD (n = 4)**	**250 mg BID (n = 3)**	**500 mg BID (n = 8)**	**Total, n (%) (N = 17)**
Fatigue	1 (G2)	1 (G1), 2 (G2)	1 (G1)	2 (G1), 1 (G2)	8 (47.1)
Anemia	0	2 (G1)	2 (G1)	2 (G1)	6 (35.3)
Vomiting	1 (G1)	0	1 (G1)	1 (G1)	3 (17.6)
ALK increased	0	2 (G1)	0	1 (G1)	3 (17.6)
ALT increased	0	1 (G3)	0	1 (G4)	2 (11.8)
AST increased	0	1 (G2)	0	1 (G4)	2 (11.8)
Decreased appetite	1 (G2)	0	0	1 (G1)	2 (11.8)
Ejection fraction decreased	0	1 (G2)	0	1 (G3)	2 (11.8)
Hyperglycemia	0	2 (G1)	0	0	2 (11.8)

ALK, alkaline phosphatase; ALT, alanine aminotransferase; AST, aspartate aminotransferase; BID, twice daily; G, grade; QD, once daily

BID), PF-06840002 was absorbed rapidly, with median T_{max} ranging from 1.5 to 3.0 hr postdose. Following multiple oral doses, the median T_{max} ranged from 2.0 to 3.0 hr postdose (Table 4).

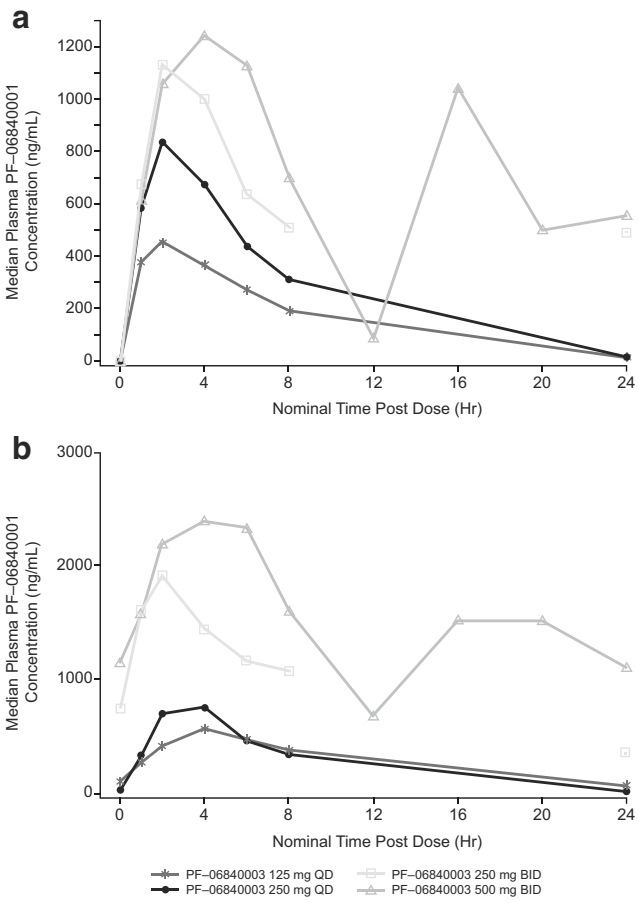

Fig. 1 Median plasma PF-06840002 concentration-time profiles following (A) single oral doses (Cycle 1 Day 1) and (B) multiple oral doses (Cycle 1, Day 15)
BID, twice daily; QD, one daily

Plasma exposure of PF-06840002, measured by AUCτ and maximum plasma concentration, appeared to increase in a less than proportional manner from 125 mg QD to 500 mg BID (Table 4). This may be due to a low number of dose levels and patients. In general, there did not appear to be accumulation following multiple dosing across the 125 mg QD to 250 mg QD and BID dose range (Table 4). The mean elimination half-life was approximately 2 to 4 hrs. PK parameters for PF-06840001 were similar to those for PF-06840002 (Online Resource 1).

Predose CSF-to-plasma ratios indicate evidence of CNS penetration by Cycle 1 Day 15 for both PF-06840002 and the inactive enantiomer PF-06840001. Where CSF was measurable for PF-06840002, predose CSF-to-plasma ratio approximated a mean of 1.0 for both 250 mg BID ($n = 2$) and 500 mg BID ($n = 4$) dose levels. Similarly, predose CSF-to-plasma mean ratios for PF-06840001 were 0.89 and 0.88 for the 250-mg BID and 500-mg BID dose levels, respectively.

Urinary recovery of PF-06840002 was low, with < 1% of the dose recovered unchanged (Aeτ%) in urine following the 500 mg BID dose (Cycle 1 Day 15). Renal clearance was 103 mL/hr for PF-06840002 (Table 4).

Pharmacodynamics

Mean percentage inhibition of ^{13}C10 kyn and endogenous kyn in peripheral blood versus nominal time postdose on Day 15 are shown in Fig. 2. On Cycle 1 Day 15, the mean plasma ^{13}C10 kyn percentage inhibition increased at predose in the 125-mg QD (12%), 250-mg BID (62%), and 500-mg BID (61%) cohorts. The mean endogenous kyn percentage inhibition increased at predose in the 250-mg BID (6%) and 500-mg BID (10%) cohorts. At the highest dose tested in the

Table 4 Summary of PF-06840002 pharmacokinetic parameters following single and multiple oral doses

Parameter, unit[a]	Summary statistics by PF-06840002 dose			
	125 mg QD (n = 2)	**250 mg QD (n = 4)**	**250 mg BID (n = 3)**	**500 mg BID (n = 8)**
Cycle 1 Day 1 (Single Dose)				
AUC_{τ}, ng·hr/mL	3380 (2900–3860)	5356 (60)	7606 (11)	8653 (57)
AUC_{inf}, ng·hr/mL	3960[b]	5549 (85)[c]	NC	5343 (40)[c]
AUC_{last}, ng·hr/mL	3155 (2450–3860)	4517 (80)	13340 (21)	17720 (78)
C_{max}, ng/mL	580.5 (468–693)	775.0 (37)	1135 (6)	1407 (23)
T_{max}, hr	1.58 (1.00–2.15)	1.51 (1.00–2.08)	1.95 (1.00–2.05)	3.02 (1.00–4.05)
CL/F, mL/min	527.0[b]	751.6 (85)[c]	NC	1561 (40)[c]
V_z/F, L	186.0[b]	228.4 (27)[c]	NC	277.2 (20)[c]
$t_{½}$, hr	4.08[b]	3.78 ± 1.59[c]	ND	2.09 ± 0.45[c]
Cycle 1 Day 15 (Multiple Dose)				
AUC_{τ}, ng·hr/mL	6115 (3320–8910)	6680 (75)	12730 (50)	20410 (96)
C_{av}, ng/mL	254.5 (138–371)	278.0 (75)	1060 (50)	1702 (96)
C_{max}, ng/mL	623.0 (463–783)	779.3 (9)	1763 (25)	2474 (66)
C_{min}, ng/mL	63.95 (11.9–116)	15.85 (136)	687.3 (60)	1611 (109)
T_{max}, hr	3.00 (2.00–4.00)	3.03 (2.08–4.07)	2.00 (2.00–2.10)	3.02 (1.00–4.00)
CL/F, mL/min	431.0 (234–628)	624.2 (75)	327.6 (50)	408.3 (96)
V_z/F, L	184.5 (140–229)[d]	216.0 (202–230)[d]	128.0[b]	270.5 (269–272)[d]
$t_{½}$, hr	5.580 (4.22–6.94)[d]	2.900 (2.16–3.64)[d]	2.680[b]	2.885 (1.97–3.80)[d]
R_{ss}	2.250[b]	1.289 (37)[c]	NC	1.961 (72)[c]
R_{ac}	1.725 (1.14–2.31)	1.244 (31)	1.670 (45)	2.359 (45)
A_e, ng	NA	NA	NA	3271000 (94)[e]
A_e%	NA	NA	NA	0.6539 (95)[e]
CL_r, mL/hr	NA	NA	NA	103.0 (56)[e]
CSF:plasma ratio	NA	NA	1.00 (0.99–1.00)[d]	1.00 (69)[e]

[a] Geometric mean (geometric %CV) for all except: median (range) for T_{max}; arithmetic mean (± StdD) for $t_{½}$ and arithmetic mean (%CV) for C_{min} and CSF-to-plasma ratio. Median and range presented for $n < 3$. Individual value presented for $n = 1$.

[b] n=1; [c] n=3; [d] n=2; [e] n=4

A_e, cumulative amount of drug recovered unchanged in urine; AUC_{inf}, area under the concentration-time profile from time 0 extrapolated to infinite time; AUC_{last}, area under the concentration-time profile from time 0 to the time of the last quantifiable concentration; AUC_{τ}, area under the concentration-time profile from time 0 to time tau (τ), the dosing interval, where tau was 24 hours for QD dosing and 12 hours for BID dosing. When $t_{last} < \tau$ data was extrapolated; BID, twice daily; C_{av}, average concentration for the dosing interval; CL/F, apparent clearance; CL_r, renal clearance; C_{max}, maximum plasma concentration; C_{min}, lowest concentration observed during the dosing interval; CSF, cerebrospinal fluid; CV, coefficient of variation; NA, not available; NC, not calculated; ND, not determined; QD, once daily; R_{ac}, observed accumulation ratio; R_{ss}, steady-state accumulation ratio; StdD, standard deviation; $T_{½}$, terminal half-life; T_{max}, time to maximum plasma concentration; V_z/F, apparent volume of distribution

study (500 mg BID), the maximum mean percentage inhibition over the 12-hr dosing interval was 75% (^{13}C10 kyn) and 24% (endogenous kyn).

At Cycle 1 Day 15, CSF-to-plasma mean ratio ranged from 0.01 to 0.05 for kyn and from 0.03 to 0.05 for trp across the 250-mg QD to 500-mg BID dose levels (Table 5).

Efficacy

Disease control occurred in eight patients (47%), 7 of which showed sustained stable disease (SD) > 6 weeks after treatment initiation, but there were no partial or complete responses (Macdonald criteria). Patients with SD had a mean duration of treatment before progression and/or discontinuation of 32.1 weeks (range 12.1 to 72.3 weeks) (Fig. 3). One patient received PF-06840003 for 20.1 weeks and discontinued due to global deterioration of health status, leading to a switch to new anticancer treatment.

Two patients treated at the 500-mg BID dose level discontinued from the study but continued to receive PF-06840003 through an expanded access program, with ongoing SD at 450 + days (Patient A) and 561 + days (Patient B) since starting study therapy. Patient A, a 36-year-old man, was

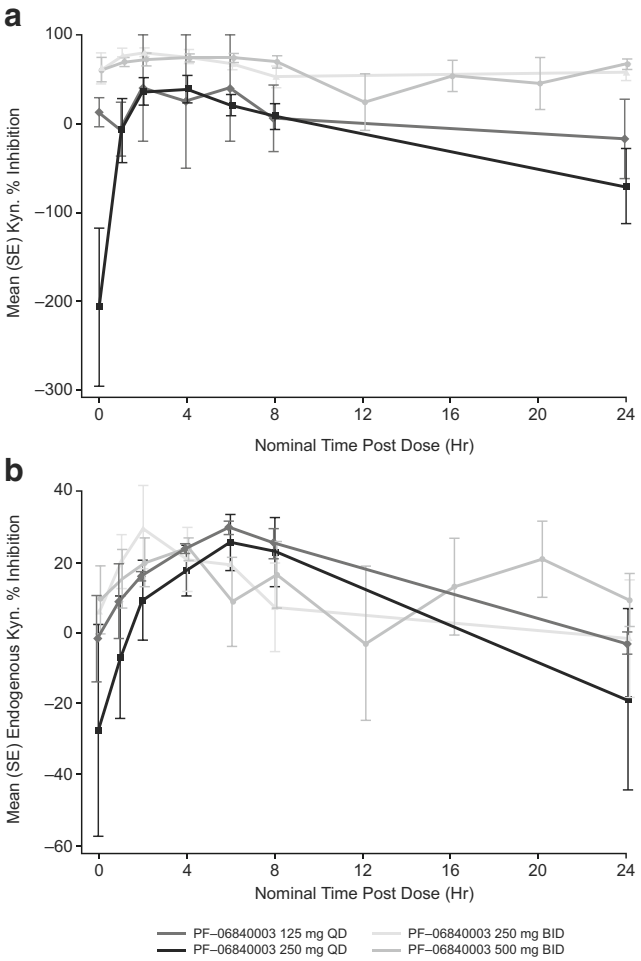

Fig. 2 Mean percentage inhibition of (A) ^{13}C10 kynurenine, and (B) endogenous kynurenine in peripheral blood versus nominal time postdose on Cycle 1 Day 15
SE, standard error

originally diagnosed with a left temporal anaplastic astrocytoma (*IDH1R132H* mutant, *MGMT* promoter unmethylated). Following subtotal resection, he underwent standard radiation with daily temozolomide (TMZ) followed by one cycle of adjuvant TMZ. Due to worsened headaches and progressive imaging findings, he underwent a gross total resection 2 months after completion of radiation/daily TMZ, with pathology revealing unequivocal recurrent tumor consistent with transformation to GBM. He began study therapy approximately 1 month post operatively. Patient B, a 48-year-old female developed recurrent GBM, confirmed histologically from gross-total resection of recurrent disease. She was originally treated with surgery, radiation, and standard TMZ chemotherapy 4.7 years prior to enrolling in the study. Of note, neither were on corticosteroids at study initiation.

PFS was 1.9 months in the PF-06840003 125-mg QD cohort (median value was not estimable due to only one patient with event) and the median PFS ranged from 1.9 to 2.8 months for other cohorts.

Based on iRANO criteria [19], disease control was observed in six (35.3%) patients at Week 8, five (29.4%) patients at Week 16, and two (11.8%) patients at Week 24. No meaningful differences in DCR were observed across the treatment cohorts.

Discussion

The highly selective IDO1 inhibitor PF-06840003 has demonstrated promising antitumor activity in preclinical models [16]. In this first-in-human phase 1 study, we evaluated the safety, tolerability, PK, PD, and preliminary antitumor activity of PF-06840003 in patients with recurrent malignant glioma in order to determine the RP2D.

Sequential cohorts of patients with recurrent malignant glioma received escalating doses of PF-06840003 (125 mg QD, 250 mg QD, 250 mg BID, and 500 mg BID). PF-06840003

Table 5 Pharmacodynamic parameters following PF-06840003 dosing

Parameter, unit	PF-06840003 dose			
	125 mg QD (n = 2)	**250 mg QD (n = 4)**	**250 mg BID (n = 3)**	**500 mg BID (n = 8)**
Cycle 1 Day 15				
Mean CSF tryptophan, μM	–	1.050[a]	1.500[b]	1.508[c]
Mean CSF kynurenine, μM	–	0.06570[a]	0.03540[b]	0.07150[c]
Mean plasma endogenous tryptophan, μM	48.15	29.55	36.77	33.10
Mean plasma endogenous kynurenine, μM	1.545	2.185	1.144	1.523
CSF:plasma ratio (tryptophan)	–	0.03009[a]	0.0458[b]	0.04918[c]
CSF:plasma ratio (kynurenine)	–	0.01386[a]	0.03302[b]	0.04538[c]

[a] n=1; [b] n=2; [c] n=4;
BID, twice daily; CSF, cerebrospinal fluid; QD, once daily

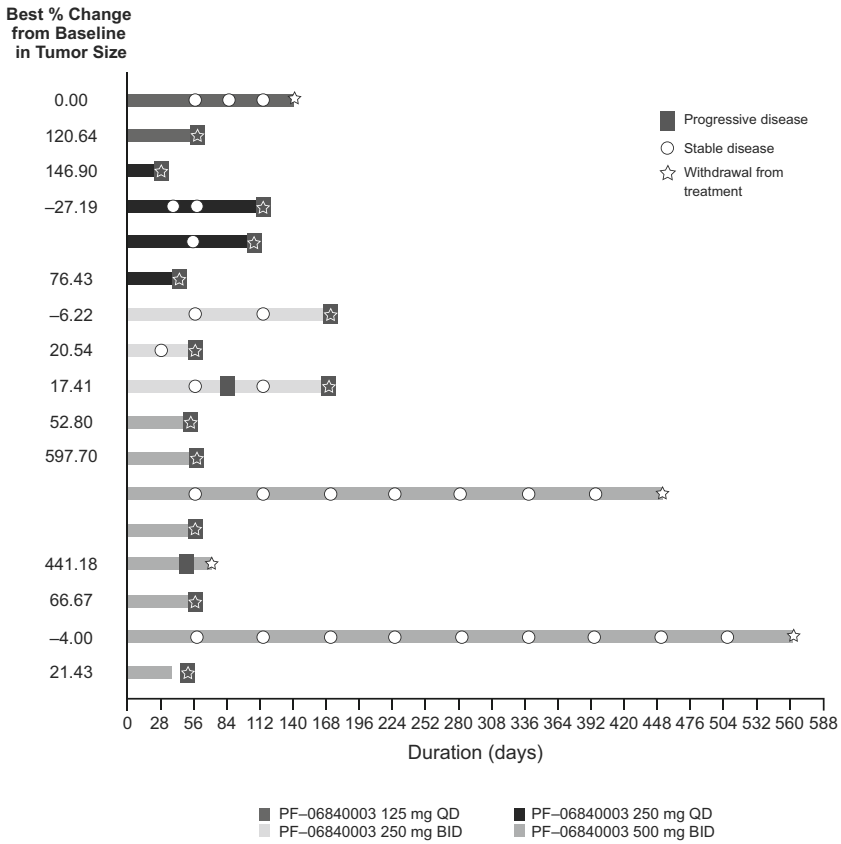

Fig. 3 Overall response and best percent change from baseline in tumor size (Macdonald criteria [18])

was generally well tolerated. One of the 17 patients had serious AEs reported as DLT. However, the MTD was not determined due to the low number of DLTs at the highest dose (500 mg BID) evaluated in the study. There was no notable trend in rates of AEs across treatment cohorts. Most of the events were non-serious, manageable, and did not lead to treatment discontinuation.

Urinary recovery of PF-06840002 was limited (< 1%), consistent with findings in preclinical species, including rat and dog [17]. These data are consistent with PF-06840002 being cleared primarily via metabolism in humans. However, it is unclear why the human half-life was shorter than predicted given the excellent in vitro and in vivo correlations observed between clearance scaled from hepatocyte data and in vivo clearance across various species (data not shown). Emerging PK/PD data from the first (125 mg QD) and second (250 mg QD) cohorts were evaluated during dose-escalation and it was predicted that dosing under a QD regimen would not maintain trough concentrations sufficient for maximal IDO1 inhibition over the dosing interval. Therefore, a BID dosing regimen was initiated for the third cohort (250 mg BID) and evaluated for further dose escalation towards the MTD. Subsequently, increases in drug exposure were small when dosing increased from 250 mg BID to 500 mg BID, which suggested a potential maximum absorption and limited benefit of higher dosages with respect to PK/PD.

As highlighted, the targeted level of IDO1 inhibition based on preclinical models of efficacy was 90%, or near maximal inhibition. The most relevant measures are kyn inhibition in the tumor microenvironment, but post-treatment biopsy data were not collected. Multiple PD assays were validated to measure plasma kyn and used to monitor target inhibition. A minimal circulating kyn inhibition needed for clinical efficacy has not been established. However, epacadostat, an inhibitor of IDO1, was shown to reduce endogenous kyn by 46% at steady-state (predose) at clinically active doses (100 mg BID), which were subsequently selected for evaluation of efficacy in phase 3 studies [21]. Another potent IDO1 inhibitor, BMS-986205, showed greater than 60% inhibition of endogenous kyn observed at 100 mg QD [22]. These data provided a basis for selecting the RP2D for PF-06840003, relying on the endogenous PD measure, with the minimal clinical goal of achieving at least 46% inhibition of endogenous kyn at steady-state. At the highest dose evaluated in the current first-in-patient study, the inhibition of endogenous kyn at steady-state was modest, reaching a maximum of 24% at 4 hr postdose and 10% at the predose level.

It should be noted that although the goal based on external information was not reached, this study provides evidence of a PD effect of PF-06840003 in patients with GBM. Furthermore, preclinical evidence of CNS penetration in rat models, along with data supporting the hypothesis that IDO1 activity correlates with clinical outcome in GBM [15], provided a strong rationale for assessing safety and PK/PD in this patient population. In this study, CSF-to-plasma ratios provided evidence of CNS penetration at steady-state for both the active and inactive enantiomers, with mean ratios of 0.9 to 1.0, indicating equivalent unbound concentrations of both enantiomers in CSF and plasma.

Limitations

Our data are limited by the nonrandomized design and small sample size and that no responses were observed, although 47% achieved disease stabilization. This is consistent with PF-06840003 monotherapy that had a heterogeneous antitumor efficacy in syngeneic tumor models [16] and suggests that IDO1-targeting therapy may cytostatically benefit a subset of GBM patients. The enhanced in vivo efficacy of PF-06840003 in combination with anti-PD-L1 observed previously [16] suggests that combinatorial therapies may be worthy of further study.

Additional limitations included the small study sample size, especially for CSF PK determinations. Furthermore, although CSF PK data were reassuring, they may not accurately reflect actual intra-parenchymal drug levels, particularly in the non-enhancing component of tumors where the blood–brain barrier is relatively intact. In addition, although, a subset of patients with durable benefit were identified, correlative data on tumor or systemic biomarkers were not available and therefore future studies may be required to better identify patients with malignant glioma who are most likely to benefit from IDO1 inhibitor therapy.

Conclusions

The highly selective IDO1 inhibitor PF-06840003 had a tolerable safety profile in patients with recurrent malignant glioma. The DLT rate at the 500-mg BID dose was low (12.5%) and the MTD was not reached. PF-06840003 led to inhibition of ^{13}C10 kyn and endogenous kyn, indicating a PD effect in this patient population. This finding combined with durable tumor control for > 450 days observed in two patients treated at the 500-mg BID dose level suggest that further studies of IDO inhibitors in this subject population may be warranted.

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Data availability Upon request, and subject to certain criteria, conditions, and exceptions (see https://www.pfizer.com/science/clinical-trials/trial-data-and-results for more information), Pfizer will provide access to individual de-identified participant data from Pfizer-sponsored global interventional clinical studies conducted for medicines, vaccines, and medical devices: (1) for indications that have been approved in the US and/or EU; or (2) in programs that have been terminated (i.e., development for all indications has been discontinued). Pfizer will also consider requests for the protocol, data dictionary, and statistical analysis plan. Data may be requested from Pfizer trials 24 months after study completion. The de-identified participant data will be made available to researchers whose proposals meet the research criteria and other conditions, and for which an exception does not apply, via a secure portal. To gain access, data requestors must enter into a data access agreement with Pfizer.

Compliance with ethical standards

Conflict of interest DA Reardon declares research support (paid to DFCI): Acerta Phamaceuticals, Agenus, Celldex, EMD Serono, Incyte, Inovio, Midatech, Omniox, and Tragara; Advisory/consultation (paid to Dr Reardon): AbbVie, Advantagene, Agenus, Amgen, Bayer, Bristol-Myers Squibb, Celldex, DelMar, EMD Serono, Genentech/Roche, Inovio, Merck, Merck KGaA, Monteris, Novocure, Oncorus, Oxigene, Regeneron, Stemline, and Taiho Oncology Inc; Honoraria (paid to Dr Reardon): AbbVie, Advantagene, Agenus, Bristol-Myers Squibb, Celldex, EMD Serono, Genentech/Roche, Inovio, Merck, Merck KGaA, Monteris, Novocure, Oncorus, Oxigene, Regeneron, Stemline, and Taiho Oncology Inc; A Desjardins declares research research support (paid to Duke): Orbus Therapeutics, Genentech/Roche, Triphase Accelerator, Symphogen A/S; has served as consultant/participated in advisory boards (paid to Dr Desjardins: Orbus Therapeutics and Istari Oncology; as stock options in Istari Oncology; and patent on the use of the oncolytic poliovirus for the treatment of solid tumors; T Cloughesy declares research support from AstraZeneca, honoraria from Roche for serving on the Speakers Bureau, has served as a consultant/participated in advisory boards for VBL, Bayer, GW Pharma, Tocagen, Del Mar, Amgen, QED, Merck, Karyopharm, Odonate, Pascal Biosciences, Agios, is a Board member for Global Coalition for Adaptive Research 501 ©3, has stock options with Notable Labs, has a provisional patent application for compositions and methods for treating cancer; O Rixe declares research support from: Kyowa/Kirin, Oxford Biotherapeutics, Nanobiotix, IMV Inc, Newlink, Seattle Genetics, has served on the advisory committee for Kyowa/Kirin, Bexion, and is a stock shareholder in Bexion; S Alekar is an employee of Pfizer and hold stock in Pfizer, JH Williams is an employee of Pfizer and hold stock in Pfizer, CT Taylor is an employee of Pfizer and hold stock in Pfizer, R Li is an employee of Pfizer and hold stock in Pfizer; AB Lassman declares grants and non-financial support from Pfizer, during the conduct of the study; personal fees and non-financial support from Bioclinica as an expert blinded independent reviewer of clinical and imaging data for a BMS-sponsored trial, grants, personal fees and non-financial support from Karyopharm, personal fees from Sapience, personal fees from Magnolia Innovation, personal fees and non-financial support from Guidepoint Global, personal fees and non-financial support from Abbott Molecular, grants, personal fees and non-financial support from QED Therapeutics, personal fees

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Ethical approval *Research involving human participants*: The study was approved by the institutional review board or independent ethics committee of each participating center and followed the Declaration of Helsinki and International Council for Harmonization Good Clinical Practice guidelines.

Informed consent All subjects or legally authorized representatives provided written informed consent.

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