Clinical Cancer Research

First-in-Human Phase I Study of the Oral Inhibitor of Indoleamine 2,3-Dioxygenase-1 Epacadostat (INCB024360) in Patients with Advanced Solid Malignancies

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Abstract

Purpose: Indoleamine 2,3-dioxygenase-1 (IDO1) catalyzes the degradation of tryptophan to N-formyl-kynurenine. Overexpressed in many solid malignancies, IDO1 can promote tumor escape from host immunosurveillance. This first-in-human phase I study investigated the maximum tolerated dose, safety, pharmacokinetics, pharmacodynamics, and antitumor activity of epacadostat (INCB024360), a potent and selective inhibitor of IDO1.

Patients and Methods: Fifty-two patients with advanced solid malignancies were treated with epacadostat [50 mg once daily or 50, 100, 300, 400, 500, 600, or 700 mg twice daily (BID)] in a dose-escalation 3 + 3 design and evaluated in 28-day cycles. Treatment was continued until disease progression or unacceptable toxicity.

Results: One dose-limiting toxicity (DLT) occurred at the dose of 300 mg BID (grade 3, radiation pneumonitis); another DLT occurred at 400 mg BID (grade 3, fatigue). The most common adverse events in >20% of patients overall were fatigue, nausea, decreased appetite, vomiting, constipation, abdominal pain, diarrhea, dyspnea, back pain, and cough. Treatment produced significant dose-dependent reductions in plasma kynurenine levels and in the plasma kynurenine/tryptophan ratio at all doses and in all patients. Near maximal changes were observed at doses of $\geq$100 mg BID with >80% to 90% inhibition of IDO1 achieved throughout the dosing period. Although no objective responses were detected, stable disease lasting $\geq$16 weeks was observed in 7 of 52 patients.

Conclusions: Epacadostat was generally well tolerated, effectively normalized kynurenine levels, and produced maximal inhibition of IDO1 activity at doses of $\geq$100 mg BID. Studies investigating epacadostat in combination with other immunomodulatory drugs are ongoing. *Clin Cancer Res; 1–8.* *©2017 AACR.*

Introduction

Recently, immunotherapy with checkpoint inhibitors has produced potent long-lasting antitumor activity in patients with advanced cancer (1–4). However, the majority of patients across a wide range of malignancies do not respond (5). This observation emphasizes the likely existence of additional immunoregulatory pathways that control the effectiveness of immunosurveillance in cancer (6, 7). Strategies designed to derail these pathways may be critical for broadening the application of cancer immunotherapy.

Indoleamine 2,3-dioxygenase-1 (IDO1) is a heme-containing, monomeric oxidoreductase rate-limiting enzyme that catalyzes the degradation of the amino acid tryptophan to kynurenine (8). IDO has been shown to play an important role in immune regulation. During fetal development, IDO inhibition using the competitive inhibitor 1-methyltryptophan leads to fetal rejection by maternal T cells (9). In autoimmunity, IDO appears to be a key regulator involved in suppressing T-cell responses toward apoptotic cell-associated antigens (10) and in controlling T-cell activity at sites of graft-versus-host disease (11). In cancer, IDO1 expression can block the development of a productive antitumor immune response (12–16).

Indoleamine 2,3-dioxygenase-1 is expressed by various cell types, including malignant epithelial and myeloid cells such as dendritic cells and macrophages that localize in tumor sites and within tumor-draining lymph nodes. IDO1 expression by malignant and nonmalignant cells can inhibit T-cell immune responses, leading to immune evasion and tumor outgrowth (17–19). Tryptophan metabolites such as kynurenine, produced by IDO1, inhibit immunosurveillance in cancer by arresting T cells in the G_1 phase of the cell cycle (18), promoting T-cell and

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Translational Relevance

Immunotherapy has emerged as a promising treatment option for advanced cancer; however, many patients do not receive adequate benefit, possibly because of immune checkpoint pathway redundancies. Here, we report results of a first-in-human study of epacadostat, an oral selective inhibitor of indoleamine 2,3-dioxygenase (IDO1), in patients with treatment-refractory advanced carcinoma. IDO1 is a rate-limiting enzyme that catalyzes the degradation of the amino acid tryptophan to kynurenine and, in doing so, can inhibit antitumor immune activity. In this phase I study, epacadostat was generally well tolerated and was associated with stable disease lasting $\geq$16 weeks in 7 of 52 patients. Reductions in plasma kynurenine levels and the kynurenine/tryptophan ratio were found to be relevant blood-based biomarkers for monitoring IDO1 inhibition *in vivo*. Our findings support further evaluation of epacadostat in combination with other treatment approaches for improving antitumor immune activity in patients with advanced cancer.

dendritic cell apoptosis, and supporting regulatory T-cell generation (12, 19–22). In addition, tryptophan metabolites have been found to negatively affect natural killer cell function (23, 24).

Indoleamine 2,3-dioxygenase-1 is increased in tumors that are actively infiltrated by effector T cells (13). This enhanced tumor expression of IDO1 is thought to be driven by IFNγ (12). In patients with advanced cancer, IDO1 activity is increased with elevated levels of tryptophan metabolites detected in the serum and is associated with a poor prognosis (12, 25–29). Based on these findings, IDO1 inhibition using epacadostat (INCB024360), a selective small-molecule inhibitor of IDO1, was initially evaluated in a mouse model of cancer, where it was found to induce T-cell–dependent antitumor immunity (12). This activity was related to the ability of epacadostat to block regulatory T-cell activity and promote dendritic cell maturation and function. Here, we report on the safety and activity of single-agent epacadostat in patients with advanced solid malignancies. The primary objective was to define the safety, tolerability, and maximum tolerated dose (MTD) of epacadostat. Secondary objectives were to evaluate clinical activity, pharmacokinetics (PK), pharmacodynamics, and the effect on blood biomarkers of inflammation.

Patients and Methods

Patients

Fifty-two patients with a neoplastic disease that was refractory to currently available therapies or for which no effective treatment was available were enrolled at the Abramson Cancer Center, University of Pennsylvania (Philadelphia, PA), and the University of Chicago (Chicago, IL). Inclusion criteria were age $\geq$18 years, Eastern Cooperative Oncology Group (ECOG) performance status of 0 to 2, life expectancy $\geq$12 weeks, and adequate end organ function. Exclusion criteria included receipt of an investigational study drug within 28 days (42 days for monoclonal antibodies) before study entry and prior anticancer medication within 21 days of the first dose of study medication or 6 weeks for mitomycin-C or nitrosoureas. Hormonal treatments were allowed and could be continued, and there were no restrictions on the number of prior lines of therapy. Additional exclusion criteria were active viral hepatitis or other active chronic or acute systemic infection, active autoimmune disorder, receipt of any compound known to be a potent inducer or inhibitor of CYP3A4, prior bone marrow or solid organ transplant, receipt of serotonin reuptake inhibitor within 3 weeks before study entry, major surgery within 4 weeks of study entry, active cardiovascular disease, history of gastrointestinal condition causing malabsorption or obstruction, history of brain metastases or spinal cord compression, and active infection. All patients provided written informed consent. The study was approved by the institutional review boards of the participating institutions.

Study design and treatment

This was an open-label, multicenter, phase I dose-escalation study using a $3 + 3$ design. The primary objective was to determine safety, tolerability, and MTD of epacadostat. Secondary objectives were to characterize PK, pharmacodynamics, biomarkers, and tumor response rates.

Epacadostat was supplied by Incyte Corporation (Wilmington, DE; 25-, 100-, and 300-mg tablets) and was administered orally once or twice daily after a 2-hour fast [50 mg once daily or 50, 100, 300, 400, 500, 600, or 700 mg twice daily (BID)] in 28-day cycles. The predetermined doses of 50, 100, and 300 mg BID were predicted based on preclinical studies to achieve 50%, 75%, and 90% inhibitory concentrations (IC_{50}, IC_{75}, and IC_{90}) of IDO1 at trough serum levels in humans. The starting dose of 50 mg daily was proposed to minimize risk while still providing some inhibitory effect. Patients were instructed to fast for an additional hour following dosing. Additionally, the food effect of a standardized high-fat meal on steady-state PK was evaluated for the epacadostat 600 mg BID group on cycle 2, day 1. Patients continued treatment until disease progression or unacceptable toxicity.

Safety and efficacy assessments

All patients who received at least 1 dose of epacadostat were evaluated for safety and efficacy. Safety assessments included the incidence of all adverse events (AE), irrespective of relationship to study drug, according to the National Cancer Institute Common Terminology Criteria for Adverse Events, version 4.0, and the incidence of patients experiencing dose modifications and/or premature discontinuation of study drug. AEs were monitored at screening; on days 1, 8, 15, and 22 during cycle 1; and on days 1 and 15 for all subsequent cycles. A dose-limiting toxicity (DLT) was defined as a treatment-related grade $\geq$4 hematologic toxicity of any duration except for grade 4 neutropenia without fever lasting less than 7 days or any grade $\geq$3 nonhematologic toxicity that occurred during the first 28 days of treatment (cycle 1). The MTD was estimated as the dose level below that at which one third or more patients in a group (minimum 6) experienced DLTs related to treatment with epacadostat. Tumor response was assessed by computed tomography at baseline and every 8 weeks according to Response Evaluation Criteria in Solid Tumors, version 1.1.

Pharmacokinetics assessment

Plasma samples were collected on days 1 and 15 (steady state) of cycle 1 before dosing and at defined time points after dosing. Samples were analyzed for epacadostat by a validated liquid chromatography with tandem mass spectrometry (LC/MS/MS)

assay, with a lower limit of quantitation of 2 nmol/L (30). Noncompartmental PK analysis was performed using Phoenix WinNonlin (Certara USA, Inc.). Where available, the actual sample collection times were used for PK analysis. For a small number of samples for which actual collection times were not recorded, the scheduled collection times were used instead.

Pharmacodynamic assessments

Plasma levels of tryptophan and kynurenine at different time points were evaluated by LC/MS/MS. In addition, whole blood samples, collected at times correlating with PK assessments, were evaluated for IDO1 inhibition by stimulating the blood with IFNγ (100 ng/mL) and lipopolysaccharide (LPS; 100 ng/mL) for 18 hours and then analyzing the plasma from the samples for tryptophan and kynurenine levels by LC/MS/MS as previously described (30). For each patient, the percentage inhibition of IDO1, as determined by the decrease in kynurenine levels, was calculated by comparing predose values with values obtained at different times after dosing.

Changes in the plasma levels of proteins related to immunity or inflammation were monitored using Evidence Investigator Biochip Array technology, a custom-designed multiplex biochip array based on sandwich chemiluminescent immunoassays (Randox Laboratories).

Statistical analysis

Descriptive statistical analyses were used to evaluate all applicable outcomes.

Results

Patient characteristics

Fifty-two patients with advanced solid malignancies were enrolled (Table 1). The predominant tumor sites were colorectal ($n = 29$, 55.8%) and melanoma ($n = 6$, 11.5%), and the most frequently reported locations of metastatic disease were lung, liver, and lymph nodes. Each patient had received at least 1 prior treatment for metastatic disease before study entry, and all patients except 1 had ECOG performance status of 0 or 1.

Table 1. Patient demographics and disease characteristics at baseline

Characteristic	Total ($N = 52$)
Age, y	
Median	59.5
Min, max	38.0, 78.0
≤65 y, *n* (%)	36 (69.2)
Gender, *n* (%)	
Male	31 (59.6)
Female	21 (40.4)
Race, *n* (%)	
White	45 (86.5)
Black/African American	4 (7.7)
Asian	2 (3.8)
Native Hawaiian/Other Pacific Islander	1 (1.9)
ECOG status, *n* (%)	
0	20 (38.5)
1	31 (59.6)
2	1 (1.9)
Tumor site, *n* (%)	
Colorectal	29 (55.8)
Melanoma	6 (11.5)
Renal cell carcinoma	3 (5.8)
Other	14 (26.9)
Locally advanced disease, *n* (%)	
Yes	33 (63.5)
No	19 (36.5)
Locations of metastatic disease, *n* (%)	
Lung	38 (73.1)
Liver	30 (57.7)
Lymph nodes	27 (51.9)
Bone	6 (11.5)
Ascites	3 (5.8)
Skin or subcutaneous tissue	2 (3.8)
Pancreas	1 (1.9)
Pleural effusion	1 (1.9)
Other[a]	22 (42.3)

Abbreviation: ECOG, Eastern Cooperative Oncology Group.
[a]Includes lesions detected in or invading the spleen, adrenals, abdominal wall, ovary, peritoneum, kidney, small bowel, mesentery, bladder, chest wall, mediastinum, pleura, pelvis, uterus, and ureters.

Dose-limiting toxicities

Two patients experienced DLTs: grade 3 radiation pneumonitis and grade 3 fatigue occurred in 1 patient each at the 300- and 400-mg BID dose levels, respectively. In light of the projection from preclinical animal modeling data (30) that all BID doses administered during dose escalation (50–700 mg) would be efficacious, no other DLT occurred with dosing up to 700 mg BID; thus, the MTD was not established for epacadostat.

Safety and tolerability

The median (range) duration of epacadostat exposure was 51.5 (7–284) days with a median (range) total daily dose of 800 (43.2–1400) mg. Irrespective of association with therapy, the most common AEs (all grades) occurring throughout the study period were fatigue (69.2%), nausea (65.4%), decreased appetite (53.8%), and vomiting (42.3%; Table 2). These AEs were managed by investigators using routine supportive care measures. Seven patients (13.5%) discontinued therapy because of AEs (50 mg once daily, $n = 1$; 100 mg BID, $n = 1$; 300 mg BID, $n = 2$; 400 mg BID, $n = 3$), including pain, hepatic infection, pneumonia, radiation recall pneumonitis (DLT), fatigue (DLT), dyspnea and hypoxia, and nausea and vomiting. Only radiation pneumonitis and fatigue were considered DLTs and possibly related, but these dose levels were expanded and determined to not exceed the MTD. Liver enzymes were monitored closely throughout treatment in all patients. No grade 4 elevations were observed in aspartate aminotransferase (AST) or alanine aminotransferase (ALT) levels. Grade 3 AST/ALT elevation was observed in 1 patient but was attributed to biliary duct obstruction consistent with progressive disease. A second patient also experienced grade 3 AST/ALT elevations, but this was determined to be most likely related to acetaminophen ingestion over the maximum recommended daily dose for tumor fevers.

Pharmacokinetics

Following oral dose administration in the fasted state, the PK of epacadostat was characterized by a time of maximum observed concentration at approximately 2 hours and a biphasic disposition with an apparent terminal-phase disposition half-life of 2.9 hours, which appeared to be dose-independent. Systemic accumulation following BID dosing increased mean epacadostat maximum observed concentration (C_{max}) and area under the concentration versus time curve over 1 steady-state dosing interval ($AUC_{0-\tau}$) by 16% and 33%, respectively, suggesting a relatively longer effective half-life of 4 to 6 hours. Increases in epacadostat C_{max} and $AUC_{0-\tau}$ were slightly less

Table 2. Treatment-emergent adverse events

AE, *n* (%)	50 mg QD (*n* = 3)	50 mg BID (*n* = 4)	100 mg BID (*n* = 5)	300 mg BID (*n* = 6)	400 mg BID (*n* = 11)	500 mg BID (*n* = 5)	600 mg BID (*n* = 14)	700 mg BID (*n* = 4)	Total (*N* = 52)
All-grade AEs in ≥10% of patients									
Fatigue	2 (66.7)	2 (50.0)	4 (80.0)	3 (50.0)	10 (90.9)	5 (100.0)	8 (57.1)	2 (50.0)	36 (69.2)
Nausea	3 (100.0)	4 (100.0)	3 (60.0)	4 (66.7)	5 (45.5)	4 (80.0)	8 (57.1)	3 (75.0)	34 (65.4)
Decreased appetite	0	2 (50.0)	1 (20.0)	3 (50.0)	7 (63.6)	4 (80.0)	8 (57.1)	3 (75.0)	28 (53.8)
Vomiting	2 (66.7)	2 (50.0)	0	3 (50.0)	3 (27.3)	2 (40.0)	8 (57.1)	2 (50.0)	22 (42.3)
Constipation	1 (33.3)	2 (50.0)	1 (20.0)	4 (66.7)	2 (18.2)	0	6 (42.9)	3 (75.0)	19 (36.5)
Abdominal pain	1 (33.3)	1 (25.0)	0	2 (33.3)	5 (45.5)	1 (20.0)	4 (28.6)	1 (25.0)	15 (28.8)
Diarrhea	1 (33.3)	2 (50.0)	1 (20.0)	1 (16.7)	4 (36.4)	1 (20.0)	3 (21.4)	1 (25.0)	14 (26.9)
Back pain	1 (33.3)	0	0	3 (50.0)	3 (27.3)	0	4 (28.6)	2 (50.0)	13 (25.0)
Dyspnea	1 (33.3)	1 (25.0)	1 (20.0)	1 (16.7)	2 (18.2)	1 (20.0)	5 (35.7)	1 (25.0)	13 (25.0)
Cough	1 (33.3)	0	2 (40.0)	2 (33.3)	1 (9.1)	2 (40.0)	3 (21.4)	0	11 (21.2)
Hypokalemia	0	2 (50.0)	2 (40.0)	1 (16.7)	2 (18.2)	0	2 (14.3)	1 (25.0)	10 (19.2)
Weight decreased	2 (66.7)	0	1 (20.0)	1 (16.7)	3 (27.3)	0	2 (14.3)	1 (25.0)	10 (19.2)
Pain	1 (33.3)	1 (25.0)	0	1 (16.7)	1 (9.1)	2 (40.0)	1 (7.1)	2 (50.0)	9 (17.3)
Pyrexia	1 (33.3)	1 (25.0)	0	1 (16.7)	3 (27.3)	0	2 (14.3)	0	8 (15.4)
Dehydration	0	1 (25.0)	0	1 (16.7)	3 (27.3)	0	2 (14.3)	0	7 (13.5)
Dizziness	1 (33.3)	0	0	0	1 (9.1)	0	2 (14.3)	2 (50.0)	6 (11.5)
Dysgeusia	0	0	1 (20.0)	1 (16.7)	1 (9.1)	0	2 (14.3)	1 (25.0)	6 (11.5)
Grade 3 or 4 AEs occurring in ≥2 patients									
Fatigue	0	0	0	0	1 (9.1)	1 (20.0)	3 (21.4)	1 (25.0)	6 (11.5)
Abdominal pain	0	1 (25.0)	0	0	3 (27.3)	0	1 (7.1)	0	5 (9.6)
Hypokalemia	0	1 (25.0)	1 (20.0)	0	1 (9.1)	0	1 (7.1)	1 (25.0)	5 (9.6)
Nausea	0	1 (25.0)	0	1 (16.7)	1 (9.1)	0	2 (14.3)	0	5 (9.6)
Dehydration	0	1 (25.0)	0	1 (16.7)	1 (9.1)	0	0	0	3 (5.8)
Dyspnea	0	0	0	0	1 (9.1)	0	2 (14.3)	0	3 (5.8)
Hypotension	0	0	1 (20.0)	1 (16.7)	1 (9.1)	0	0	0	3 (5.8)
Vomiting	0	1 (25.0)	0	0	1 (9.1)	0	1 (7.1)	0	3 (5.8)
ALT increased	0	1 (25.0)	0	0	0	0	1 (7.1)	0	2 (3.8)
Anemia	0	0	1 (20.0)	0	1 (9.1)	0	0	0	2 (3.8)
Back pain	0	0	0	0	1 (9.1)	0	1 (7.1)	0	2 (3.8)
Bacterial infection	0	0	0	0	0	0	2 (14.3)	0	2 (3.8)
Hyponatremia	0	0	0	0	0	0	2 (14.3)	0	2 (3.8)
Pneumonia	0	0	0	1 (16.7)	0	0	1 (7.1)	0	2 (3.8)
Small intestinal obstruction	0	2 (50.0)	0	0	0	0	0	0	2 (3.8)

Abbreviations: AE, adverse event; ALT, alanine aminotransferase; BID, twice daily; QD, once daily.

than proportional to dose within the range of 50 to 700 mg BID. Moderate intersubject variability was observed for epacadostat plasma exposures (Table 3). Administration of a high-fat meal with epacadostat 600 mg BID decreased the geometric mean C_{max} by approximately 10% and increased the geometric mean area under the curve from 0 to 12 hours (AUC_{0-12h}) by 22%. The 90% CIs of the geometric mean ratio point estimates for C_{max} and AUC_{0-12h} were 0.645 to 1.25 and 0.952 to 1.57, respectively, both including the value of 1 and indicating that the effect on epacadostat plasma exposures from a high-fat meal was not statistically significant.

Pharmacodynamics

Treatment with epacadostat did not produce any significant changes in plasma proteins related to immune function, including IL-4, IL-10, IL12, CCL22 (macrophage-derived chemokine), or CCL5 (RANTES). In addition, no changes were observed with treatment in plasma proteins related to inflammation, including C-reactive protein, intercellular adhesion molecule 1, IL-6, tumor necrosis factor (TNF)-α, TNF receptor 2, and vascular cell adhesion protein 1.

In the published literature, kynurenine levels are generally elevated in patients with cancer (12), suggesting activation of the

Table 3. Summary of epacadostat steady-state (day 15) pharmacokinetic parameters

	Mean ± SD			
Dose group	C_{max}, μmol/L	t_{max}, h[a]	$t_{1/2}$, h	$AUC_{0-\tau}$, μmol/L × h
50 mg QD (*n* = 3)	0.396 ± 0.172	2.0 (1.0–4.0)	2.4 ± 0.26	1.58 ± 0.31
50 mg BID (*n* = 4)	0.742 ± 0.212	2.0 (1.0–3.9)	2.4 ± 0.56	3.05 ± 1.36
100 mg BID (*n* = 5)	1.23 ± 0.348	2.0 (1.0–2.2)	3.3 ± 0.75	5.77 ± 2.34
300 mg BID (*n* = 5)	2.48 ± 0.515	2.0 (1.0–2.0)	3.9 ± 2.1	9.78 ± 0.86
400 mg BID (*n* = 8)	4.39 ± 2.02	2.0 (1.0–6.0)	2.7 ± 0.62	19.6 ± 7.43
500 mg BID (*n* = 5)	4.82 ± 2.26	2.0 (2.0–2.4)	2.4 ± 0.37	20.6 ± 6.82
600 mg BID (*n* = 12)	4.82 ± 2.16	2.0 (1.0–2.1)	3.3 ± 0.97	22.9 ± 10.0
700 mg BID (*n* = 4)	6.23 ± 2.09	3.0 (2.0–4.5)	3.0 ± 1.2	35.8 ± 15.5

Abbreviations: $AUC_{0-\tau}$, area under the concentration versus time curve over 1 steady-state dosing interval; BID, twice daily; C_{max}, maximum observed concentration; QD, once daily; $t_{1/2}$, half-life; t_{max}, time of maximum observed concentration.
[a]t_{max} is reported as median (range).

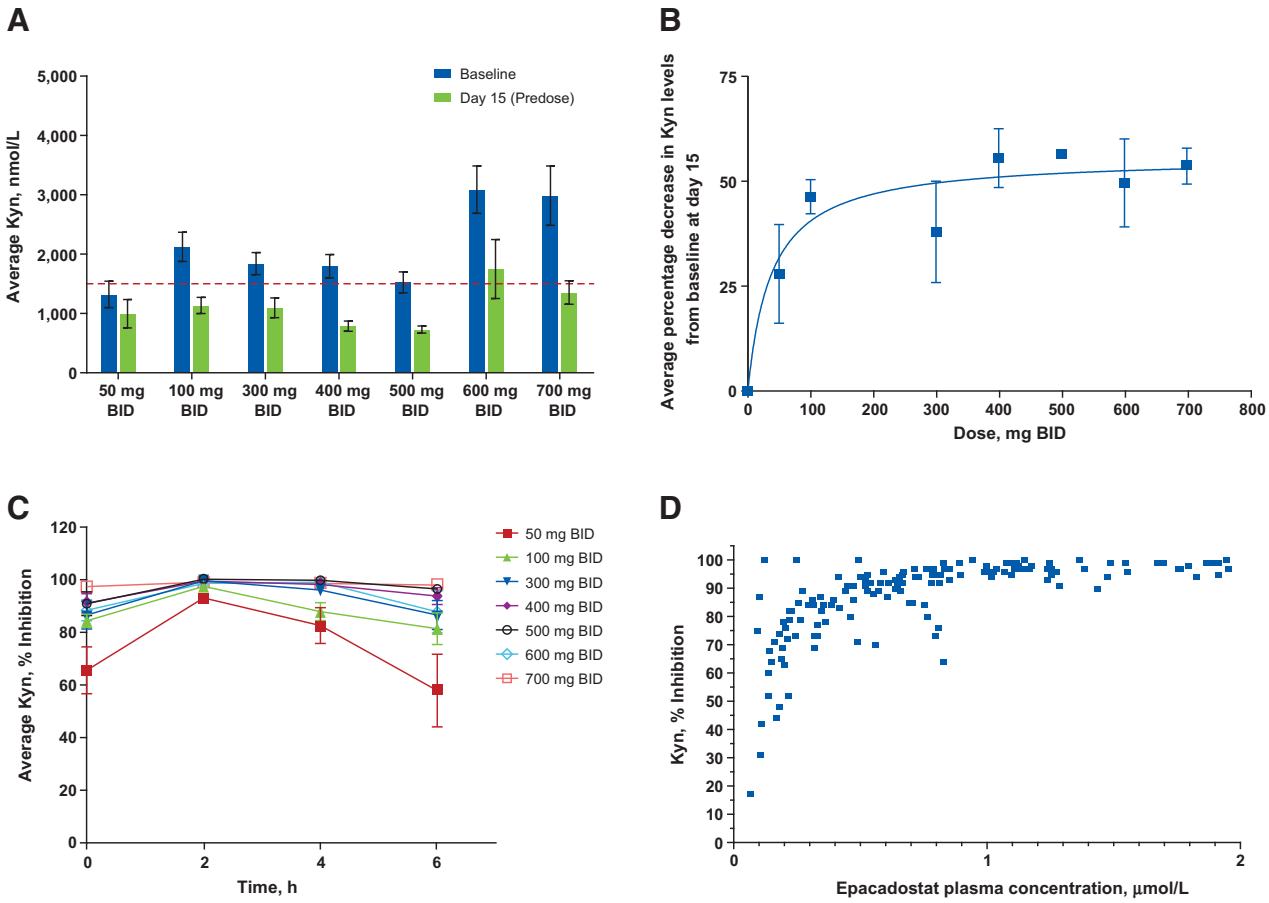

Figure 1.

Pharmacodynamic effects of epacadostat BID dosing *in vivo*. **A,** Kyn levels by dose group at baseline and day 15. The dashed line represents the median Kyn value observed in normal, healthy volunteers (1,499 nmol/L). **B,** Percentage decrease in Kyn levels from baseline by dose. **C,** Average Kyn inhibition over time by dose on day 15 (relative to day 1) following IDO1 induction with IFNγ and LPS. Time 0 represents the trough levels before the next dose of epacadostat treatment. **D,** Epacadostat plasma concentrations relative to percentage Kyn inhibition. BID, twice daily; IDO1, indoleamine 2,3-dioxygenase-1; IFNγ, interferon gamma; Kyn, kynurenine; LPS, lipopolysaccharide.

IDO1 pathway. Treatment with epacadostat reduced plasma kynurenine levels by 2 hours (data not shown) and, at doses ≥100 mg BID, was shown to reduce kynurenine levels to within the range observed in normal, healthy volunteers (median value of healthy subjects, 1499 nmol/L; Fig. 1A). In patients treated with epacadostat, plasma kynurenine levels were found to decrease at all doses, with near maximal inhibition achieved at doses ≥100 mg BID (Fig. 1B). Similar findings were also observed for kynurenine to tryptophan ratios (data not shown).

Basal kynurenine levels in the plasma can be influenced by both IDO1 and tryptophan 2,3-dioxygenase (TDO), a liver enzyme that controls dietary tryptophan catabolism (31). To determine the ability of epacadostat to inhibit IDO1 activity *in vivo* and minimize the TDO component, whole blood was stimulated for 18 hours with IFNγ and LPS to induce IDO1 expression and then assayed for kynurenine levels. Within 2 hours of dosing, a rapid onset of IDO1 inhibition was seen with >90% inhibition achieved at all dose levels. Maximal inhibition at trough was observed at doses ≥100 mg BID (Fig. 1C). The 90% inhibitory concentration was achievable at plasma concentrations greater than 500 nmol/L (Fig. 1D), which is consistent with the *in vitro* potency of epacadostat (12).

Efficacy

The overall objective response rates are summarized in Table 4. Stable disease, seen in 18 patients (34.6%), was the best overall response. Eleven patients (21.2%), including 2 melanoma patients who had progressed on ipilimumab immunotherapy before study entry, showed prolonged stable disease compared with their last prior therapy (Fig. 2). Stable disease lasting ≥16 weeks was seen in 7 patients (13.5%).

Table 4. Best overall tumor response by RECIST, version 1.1

n (%)	Total (*N* = 52)
Complete response	0
Partial response	0
Stable disease	18 (34.6)
Progressive disease	22 (42.3)
Not evaluable	12 (23.1)

Abbreviation: RECIST, Response Evaluation Criteria in Solid Tumors.

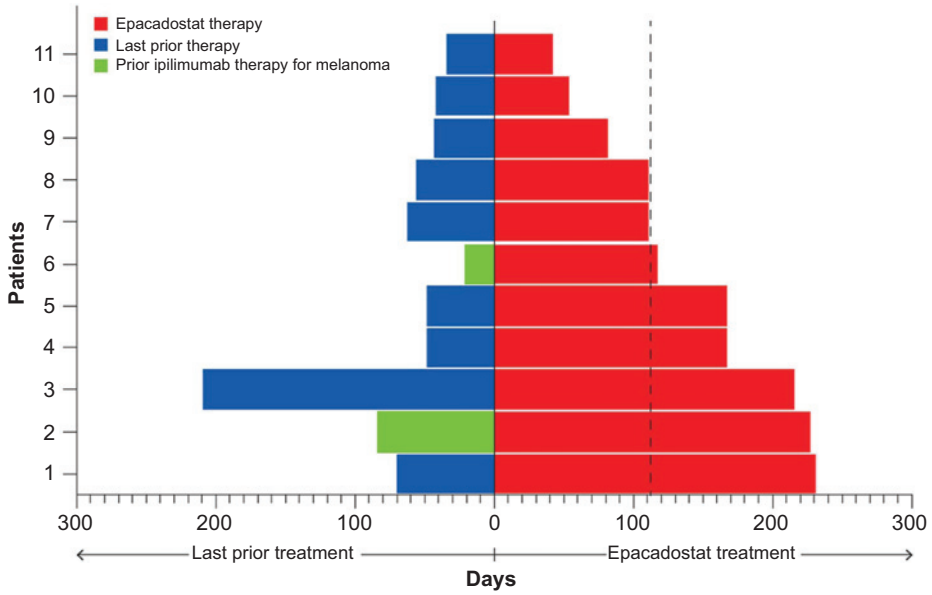

Figure 2.
Prolonged stable disease compared with last prior therapy was achieved in a subset of patients ($n = 11$). Two patients with melanoma refractory to ipilimumab achieved stable disease with epacadostat. The dashed line represents at least 16 weeks (112 days) on epacadostat therapy.

Discussion

A key hallmark of cancer is the capacity of malignant cells to evade immune elimination. This premise is supported by the impressive responses and long-term disease remissions achieved in some patients through the disruption of important immunoregulatory pathways (i.e., PD-1/PD-L1 and CTLA-4) involved in limiting the antitumor potential of the adaptive immune system (32). However, because responses are observed in only a fraction of patients and treatment responses are sometimes transient, additional strategies to reverse elements of immune suppression imposed by cancer are needed (5). IDO1 is a key enzyme involved in immunoregulation that is overexpressed in the majority of cancers (33). In this study, we conducted a first-in-human investigation of epacadostat, a novel and selective small-molecule inhibitor of IDO1, to assess safety, MTD, and biological correlatives.

Epacadostat was generally well tolerated at doses of up to 700 mg BID and can be administered without regard to food. Typical immune-related AEs, such as colitis, pneumonitis, rash, and liver chemistry test abnormalities, now recognized as complications of therapy with checkpoint inhibitors, were uncommon in this study. Because IDO1 expression may be limited to sites of inflammation (including tumor inflammation), IDO1 inhibition with epacadostat was predicted to be well tolerated. This was confirmed in preclinical toxicology studies and in this first-in-human study. An MTD was not reached even at dose levels capable of suppressing >90% of IDO1 activity *in vivo*. Based on PK and pharmacodynamic data achieving predetermined goals related to target inhibition and available tablet strengths, 300 mg BID was selected as the recommended phase II monotherapy dose to provide an exposure to epacadostat that would inhibit >90% of IDO1 activity despite variation in PK among individuals. It should be noted, however, that maximal efficacy as monotherapy in preclinical models is seen with continued coverage above the IC_{50}, suggesting that the 50-mg BID dose would also provide significant target inhibition and could be considered to be an active dose in future clinical studies (30).

The tumor microenvironment (TME) is characterized by multiple mechanisms of evasion from immune surveillance. For example, within the TME, the immune responses elicited by PD-L1 blockade might be limited by additional upregulation of IDO1 (34). Likewise, targeting IDO1 inhibition alone may be insufficient for an antitumor effect in humans because of the continued presence of immune blockade through PD-L1. This is consistent with our findings with single-agent epacadostat in this study, in which the best overall response was stable disease. In addition, the majority of patients in this study had chemotherapy-refractory colorectal cancer, a malignancy that has demonstrated marked resistance to immunotherapy, with the exception of a subset of patients with microsatellite instability who have recently been found to be responsive to PD-1 blockade (35). Nonetheless, the prolonged disease stabilization seen in some patients is encouraging.

Strategies to inhibit IDO1 are currently being evaluated in combination with vaccines (NCT02166905 and NCT01961115), cellular therapy (NCT02118285), and checkpoint inhibitors (NCT02178722, NCT02327078, NCT02318277, and NCT02298153). In preclinical models, single-agent IDO1 inhibition was found to only slow tumor outgrowth, whereas in combination with chemotherapy or additional immune checkpoint inhibitors (e.g., CTLA-4 blockade), tumor regressions were observed (12, 15, 16, 34). Preliminary results from a phase I study of daily oral epacadostat plus the immune checkpoint inhibitor pembrolizumab (200 mg intravenous every 3 weeks) in patients with treatment-naïve advanced melanoma ($n = 19$) have shown favorable objective response rates (58%), with complete responses seen in 26% of patients receiving combination therapy (36). Responses were observed in all dose cohorts receiving $\geq$50 mg BID and in 5 of 6 patients receiving $\geq$100 mg BID epacadostat (36, 37). These results informed the decision to use epacadostat 100 mg BID as the recommended phase II dose for use in combination with pembrolizumab (37). In a prior phase I study in patients with unresectable metastatic melanoma, oral epacadostat in combination with intravenous ipilimumab (38) produced an overall response rate of 31.3% for patients who were immunotherapy naïve. Together, these data suggest the potential

of epacadostat to combine with immune checkpoint inhibitors to stimulate antitumor activity.

Biomarkers that aid in assessing target inhibition of IDO1 and in selecting patients who are most likely to achieve benefit from immunotherapy are critical to the development of immunomodulatory drugs. In this study, kynurenine metabolite formation from tryptophan was used as a biomarker of IDO1 inhibition by epacadostat. All patients responded to epacadostat with a decrease in tryptophan metabolism, which has been found to be increased in many human malignancies (12). Thus, kynurenine metabolite formation is an effective biomarker for confirmation of target inhibition, but its usefulness for patient selection remains unclear. However, even at doses that resulted in effective IDO1 inhibition, we did not detect significant changes in plasma biomarkers of inflammation and immune function. This finding would be consistent with a role for IDO1 in regulating antitumor immune activity locally within tumor tissues and secondary lymphoid organs. In this regard, it is possible that IDO1 overexpression within the TME could serve as a potential biomarker, as can be seen in colorectal cancer patients with mismatch repair deficiency (39) or in patients with a robust T-cell infiltration (13, 34). Therefore, patients who demonstrate T-cell infiltrates but resistance to immune checkpoint inhibitors may be a particularly relevant population for receiving IDO1 inhibitors. Ongoing clinical studies are currently evaluating this hypothesis.

In conclusion, the results from this first-in-human study identify a biologically active and highly selective inhibitor of IDO1 that is well tolerated in patients with advanced solid malignancies. Although objective responses were not observed with single-agent epacadostat, potent inhibition of IDO1 was achieved *in vivo* at tolerable doses. As a result, further investigation of epacadostat in combination with other therapeutics, such as vaccines, cellular therapy, and immune checkpoint inhibitors, is being pursued to maximize the potential therapeutic benefit of targeting IDO1 in cancer.

Disclosure of Potential Conflicts of Interest

G.L. Beatty and P. J. O'Dwyer report receiving a commercial research grant from Incyte. P.A. Scherle is VP, Preclinical at Incyte Corporation. R.C. Newton is VP, Translational Sciences at Incyte Corporation and has ownership interest (including patents) in Incyte Corporation. L. Leopold is VP, Development at Incyte Corporation, has ownership interest (including patents), and is a stockholder in Incyte Corporation. T.F. Gajewski reports receiving a commercial research grant from Incyte. No potential conflicts of interest were disclosed by the other authors.

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