ORIGINAL ARTICLE

Effect of CYP3A inhibitors on the pharmacokinetics of pevonedistat in patients with advanced solid tumours

Hélène Faessel[1] [ID] | John Nemunaitis[2] | Todd M. Bauer[3] | A. Craig Lockhart[4] |
Douglas V. Faller[1] | Farhad Sedarati[1] | Xiaofei Zhou[1] | Karthik Venkatakrishnan[1] [ID] |
R. Donald Harvey[5]

[1] Millennium Pharmaceuticals, Inc., Cambridge, MA, USA, a wholly owned subsidiary of Takeda Pharmaceutical Company Limited

[2] University of Toledo, Toledo, OH, USA

[3] Sarah Cannon Research Institute/Tennessee Oncology, PLLC, Nashville, TN, USA

[4] Sylvester Comprehensive Cancer Center, Miller School of Medicine, University of Miami, Miami, FL, USA

[5] Winship Cancer Institute, Emory University, Atlanta, GA, USA

Correspondence
Hélène Faessel, PharmD, PhD, Millennium Pharmaceuticals, Inc., 40 Landsdowne Street, Cambridge, MA 02139, USA.
Email: Helene.Faessel@Takeda.com

Aims: This phase I study evaluated the effects of the moderate cytochrome P450 (CYP) 3A inhibitor fluconazole and the strong CYP3A/P-glycoprotein (P-gp) inhibitor itraconazole on the pharmacokinetics of the investigational neural precursor cell expressed, developmentally downregulated 8 (NEDD8)-activating enzyme inhibitor pevonedistat in patients with advanced solid tumours.

Methods: Patients received single doses of intravenous pevonedistat 8 mg m^{-2}, alone and with fluconazole (loading: 400 mg; maintenance: 200 mg once daily), or pevonedistat 8, 15 or 20 mg m^{-2}, alone and with itraconazole 200 mg once daily. Serial blood samples for pevonedistat pharmacokinetics were obtained pre- and post-infusion on days 1 (alone) and 8 (with fluconazole/itraconazole). After completing the pharmacokinetic portion, patients remaining on study received pevonedistat with docetaxel or carboplatin and paclitaxel.

Results: The ratios of geometric mean area under the concentration–time curves (n; 90% confidence interval) of pevonedistat in the presence vs. absence of fluconazole or itraconazole were 1.11 (12; 1.03–1.19) and 1.14 (33; 1.07–1.23), respectively. Fifty patients (98%) experienced at least one adverse event (AE), with maximum severity of grade 1–2 in 28 patients (55%) and of grade ≥3 in 22 patients (43%). The most common drug-related AEs were vomiting (12%), diarrhoea (10%) and nausea (8%). No new safety findings were observed for pevonedistat.

Conclusions: Fluconazole or itraconazole had insignificant effects on pevonedistat pharmacokinetics, indicating minor contributions of CYP3A/P-gp to pevonedistat clearance. The safety profile of single doses of pevonedistat plus steady-state fluconazole or itraconazole was consistent with prior clinical experience, with no new safety signals observed.

KEYWORDS

anticancer drugs, drug interactions, patient safety, pharmacokinetics

The authors confirm that the PI for this paper is Dr R. Donald Harvey and that he had direct clinical responsibility for patients.

1 | INTRODUCTION

The ubiquitin–proteasome system plays an important role in cell survival, proliferation and apoptosis by regulating protein homeostasis through ubiquitination.[1] Proteins are targeted for degradation by the proteasome through the addition of a polyubiquitin chain by E3 ubiquitin ligases, of which the largest sub-family are the **cullin-Really Interesting New Gene (RING) ligases (CRLs)**.[2] CRLs are activated via the binding of the small ubiquitin-like protein NEDD8 (neural precursor cell expressed, developmentally downregulated 8), which is facilitated by NEDD8-activating enzyme (NAE). As several substrates for CRLs are linked to cancer pathogenesis, including p27, cMYC, p-IκBα, hypoxia-inducible factor-1 (HIF1), and mammalian target of rapamycin (mTOR), CRLs are attractive targets for the development of antitumour agents.[2,3]

Pevonedistat (TAK-924/MLN4924) is a first-in-class, investigational, small molecule inhibitor of NAE. Pevonedistat inhibits the activity of NAE by forming a pevonedistat–NEDD8 adduct, which remains tightly bound to active NAE, preventing it from processing NEDD8 for CRL conjugation and resulting in CRL substrate accumulation and apoptotic cell death.[4,5] Pevonedistat has demonstrated antitumour activity in preclinical studies of solid tumour, lymphoma, and acute myelogenous leukaemia (AML) xenograft mouse models, as well as single-agent activity in patients with advanced solid tumours and haematologic malignancies.[2,4,6-12]

The pharmacokinetics (PK) of pevonedistat administered as a 1-h intravenous (IV) infusion have been evaluated following single and multiple dosing in patients with advanced solid tumours, metastatic melanoma, AML, myelodysplastic syndromes (MDS), multiple myeloma and lymphoma.[6,10,11,13] PK results indicate a linear increase in area under the plasma concentration *vs.* time curve (AUC) from 0 to 24 h (AUC_{24h}) over the examined pevonedistat dose range of 25–261 mg m^{-2}. The maximum observed concentration (C_{max}) generally appears to increase in a dose-proportional manner. The plasma concentration of pevonedistat declines in a bi-exponential manner at the end of infusion following once-daily (QD) dosing on consecutive or intermittent days, with little or no notable drug accumulation.[10,12,13] This is consistent with a mean terminal elimination half-life ($t_{1/2}$) of approximately 10–11 h estimated across doses and schedules.[6]

Preclinical studies demonstrated that hepatic metabolism appeared to be the major route of elimination for pevonedistat, with metabolisation predominantly by cytochrome P450 (CYP) 3A4 (data on file). The contribution of **CYP3A4** to pevonedistat biotransformation was estimated to be 97%, which was above the 25% threshold of potential clinical relevance for drug–drug interactions (DDIs) according to the guidelines set by the US Food and Drug Administration (FDA) and European Medicines Agency (EMA).[14,15] These in vitro metabolism studies suggested that CYP3A4 inhibition could lead to a clinically meaningful increase in pevonedistat exposure. Additionally, pevonedistat was a substrate of the transporter multidrug resistance protein 1 (MDR1)/P-glycoprotein (**P-gp**) in vitro (data on file).

Based on these results, we conducted a study to assess the potential DDI of pevonedistat with the moderate CYP3A inhibitor

What is already known about this subject
- Pevonedistat is a first-in-class inhibitor of NEDD8-activating enzyme within the ubiquitin-proteosome system, which regulates cullin-RING ligases (CRL), a family of E3 ligases controlling the ubiquitination and proteasomal degradation of CRL-dependent substrates, including proteins with important roles in cell cycle progression, DNA replication, oxidative stress response, and signal transduction. These cellular processes are relevant to tumour cell growth, proliferation, and survival, thereby providing a rationale for investigating pevonedistat as an anticancer agent.
- Pevonedistat was predominantly metabolised by CYP3A4 in vitro, and was shown to be a P-gp substrate, suggesting the potential for a clinically meaningful drug–drug interaction with strong inhibitors of CYP3A or P-gp.

What this study adds
- The moderate CYP3A inhibitor fluconazole and the strong CYP3A/P-gp inhibitor itraconazole had no clinically meaningful effects on pevonedistat pharmacokinetics.
- Pevonedistat can be co-administered without dose modifications with strong CYP3A/P-gp inhibitors including azole antifungal agents, which are commonly used treatments in patients with haematological malignancies.

fluconazole and the strong CYP3A and P-gp inhibitor itraconazole in patients with solid tumours to inform strategies for managing potential DDIs with moderate and strong CYP3A inhibitors in future clinical studies of pevonedistat and, ultimately, support adequate labelling.

2 | METHODS

2.1 | Study design

This was an open-label, multicentre, parallel-group, two-arm, phase I clinical pharmacology study in adult patients with advanced solid tumours. The study was composed of two parts (Figure 1): a 24-day part that examined potential DDI of pevonedistat with CYP3A inhibitors (Part A), followed by an (optional) extension where participating patients received (after a washout period of at least 2 weeks) pevonedistat in combination with the standard-of-care chemotherapy agents, docetaxel and carboplatin plus paclitaxel until they experienced symptomatic deterioration or progressive disease, or until their treatment was discontinued for another reason (Part B). Here we report the results from Part A. As the impact of CYP3A inhibition on pevonedistat systemic exposure was unknown at the time of
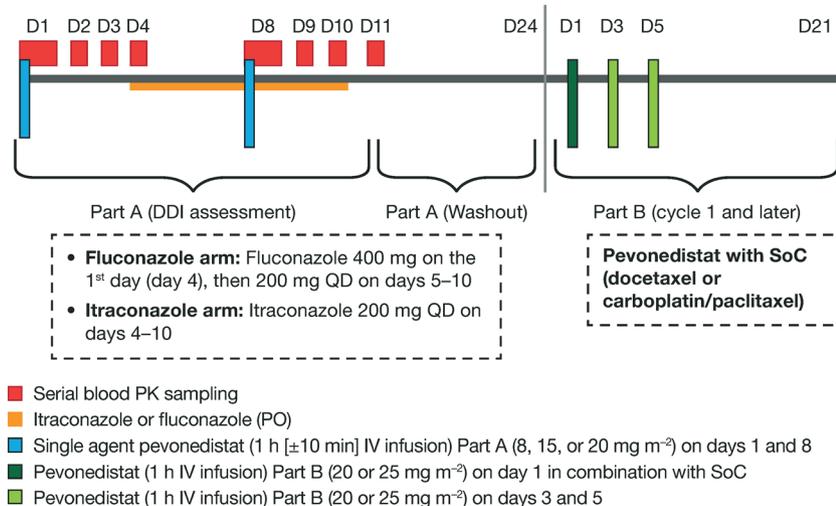
FIGURE 1 Study design. After completion of Part A (DDI assessment), eligible patients had the opportunity to continue in Part B (optional) of the study. DDI, drug–drug interactions; IV, intravenous; QD, once daily; PK, pharmacokinetics; PO, by mouth

designing this DDI study, the effect of fluconazole, a moderate CYP3A inhibitor, was evaluated first, before proceeding to conduct the DDI assessment with itraconazole, a strong CYP3A inhibitor, as a conservative measure in the interest of patient safety. Secondary objectives were to assess the safety and tolerability of pevonedistat in combination with fluconazole or itraconazole.

Pevonedistat was supplied as 10 mg mL^{-1} vials (BSP Pharmaceuticals), fluconazole as a 200 mg oral tablet (Teva)[16] and itraconazole as a 10 mg mL^{-1} oral solution (Janssen).[17] All doses of fluconazole and itraconazole were administered on an empty stomach with the patient fasting from food and fluids, except water and prescribed medications, for 2 hours before and 1 hour after each dose. The dose of pevonedistat (8 mg m^{-2}) selected for the study was approximately 14-fold lower than the 110 mg m^{-2} dose shown to result in an increased frequency of severe adverse events (AEs) in prior phase I studies.[6,10] This low dose was anticipated to provide an adequate safety margin for conduct of this DDI assessment with a strong CYP3A inhibitor.

Patients in the pevonedistat plus fluconazole group received a single 1-h IV infusion of pevonedistat 8 mg m^{-2} on days 1 and 8 with concomitant oral fluconazole at a 400 mg loading dose on day 4 and then 200 mg QD on days 5–10. Fluconazole treatment was based on the prescribing information of the drug.[16]

Patients in the pevonedistat plus itraconazole group initially received a single 1-h IV infusion of pevonedistat 8 mg m^{-2} on days 1 and 8 with concomitant oral itraconazole 200 mg QD on days 4–10, dosed per the prescribing information.[17] On the basis of the emerging data from the fluconazole and itraconazole arms, the pevonedistat 20 mg m^{-2} dose was subsequently selected for further clinical investigation because it was within the clinically relevant dose range evaluated in two phase Ib combination studies with standard-of-care therapy in solid tumours[18] and haematologic malignancies.[19] Based on an observed 23% increase in pevonedistat plasma systemic exposure with concomitant itraconazole at the 8 mg m^{-2} dose, it could be inferred that exposures of pevonedistat when administered at a single 20 mg m^{-2} dose with itraconazole could be expected to be well below exposures seen at doses of ≥100 mg m^{-2}. Conservatively,

before any patients could be enrolled in the pevonedistat 20 mg m^{-2} plus itraconazole group, the intermediate dose of pevonedistat 15 mg m^{-2} plus itraconazole was included as a safety lead-in group with the purpose of confirming that the PK and safety were adequate, based on data from three PK-evaluable patients. The administration of the last dose of fluconazole or itraconazole was followed by at least a 2-week, and up to 8-week, drug washout period. The concomitant medications that patients were prohibited from taking during the study are listed in Supplementary Table S1.

The study protocol and amendments were approved by the institutional review boards at all participating centres. The trial was conducted in accordance with the Declaration of Helsinki and the International Conference on Harmonisation Guideline for Good Clinical Practice and was registered at www.clinicaltrials.gov (NCT02122770).

2.2 | Patients

To be eligible for enrolment, adult patients were required to have a histologically or cytologically confirmed metastatic or locally advanced solid tumour for which no effective standard treatment was available. They also required an Eastern Cooperative Oncology Group performance status (ECOG PS) of 0 or 1, an absolute neutrophil count (ANC) ≥1500 mm^{-3}, a platelet count ≥100 000 mm^{-3}, total bilirubin ≤ the upper limit of normal (ULN), alanine aminotransferase (ALT) and aspartate aminotransferase (AST) ≤2.5 times ULN, and serum creatinine ≤1.2 mg dl^{-1} or a calculated creatinine clearance ≥50 mL min^{-1}.

Patients were excluded if they had: prior treatment with pevonedistat; treatment with any systemic antineoplastic therapy or investigational products within 21 days; radiotherapy or major surgery within 14 days of the study treatment; or received systemic treatment with moderate or strong CYP3A inhibitors or inducers within 14 days of the study treatment. Patients were also excluded if they had persistent diarrhoea (grade ≥2) lasting >3 days within 2 weeks of the study treatment.

2.3 | Assessments

Blood samples were collected from each patient for the determination of pevonedistat plasma concentrations. Samples were obtained before and after pevonedistat infusion on day 1 (when administered alone) and day 8 (when co-administered with fluconazole or itraconazole) at the following timepoints: within 1 h predose, at the end of infusion, and 0.5, 1, 2, 3, 4, 8, 12, 24, 48 and 72 h postdose. Plasma samples (K_2EDTA) were analysed for pevonedistat concentrations using two Good Laboratory Practice (GLP)-validated liquid chromatography/ tandem mass spectrometry (LC–MS/MS) methods. The dynamic range was 0.0500 ng mL^{-1} to 25.0 ng mL^{-1} for the low range method (precision [%CV]: 3.1–5.1%; accuracy [%bias]: −1.8% to −0.5%) and 1.00–500 ng mL^{-1} for the medium range method (precision: 2.5–3.1%; accuracy: −1.5–0.0%).

AEs were graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events (NCI CTCAE), version 4.03, and monitored throughout the study from the first dose of any study drug up to 30–40 days after the last dose of a study drug.

2.4 | PK and statistical analyses

The PK-evaluable population was defined as all patients who received the protocol-specified doses of pevonedistat and fluconazole or itraconazole. The safety population comprised all patients who received at least one dose of pevonedistat.

The sample size calculation was based on the expected two-sided 90% confidence interval (CI) for the difference in the paired, log-transformed AUC (or C_{max}) means of pevonedistat on day 8 (pevonedistat co-administered with CYP3A inhibitor) and day 1 (pevonedistat alone). Based on the PK information obtained from previous studies, the within-subject coefficient of variation (CV) was estimated to be 0.12 for AUC and 0.17 for C_{max}, respectively. Assuming the AUC (or C_{max}) ratio in the presence of fluconazole (or itraconazole) vs. in the absence of fluconazole (or itraconazole) was 2.0, with a sample size of 12 evaluable patients per arm, the 90% CI of the ratio of geometric means was expected to be (1.833, 2.182) for AUC and (1.759, 2.274) for C_{max} based on the above-mentioned variance assumptions.

In the fluconazole arm, approximately 12 PK-evaluable patients were to be enrolled. In the itraconazole arm, approximately 12 PK-evaluable patients were to be enrolled in the 8 mg m^{-2} and 20 mg m^{-2} pevonedistat cohorts, and three additional PK-evaluable patients were to be enrolled in the 15 mg m^{-2} safety lead-in cohort. Assuming 25% of the enrolled cancer patients may not be PK-evaluable, approximately 52 patients in total were planned for the study.

PK parameters for pevonedistat in the presence or absence of fluconazole or itraconazole in individual patients were calculated using noncompartmental methods (Phoenix WinNonlin version 6.3). For the estimation of the effect of fluconazole or itraconazole on the PK of pevonedistat, the ratios of geometric mean C_{max}, AUC from time zero to the time of the last quantifiable concentration (AUC$_{last}$), and

AUC from time zero to infinity (AUC$_\infty$), calculated using the observed value of the last quantifiable concentration, in the presence vs. absence of fluconazole or itraconazole, and the associated two-sided 90% CIs were calculated based on the within-patient variance calculated via an analysis of variance (ANOVA). Subject was treated as a random effect in the model. Point estimates and adjusted 90% CIs for the difference in the log means were calculated and then exponentially back-transformed to provide point and CI estimates for the ratios of interest. Lack of a clinically relevant DDI could be claimed if the 90% CIs for the systemic exposure ratios fell entirely within the equivalence limits of 80.00–125.00%.

2.5 | Nomenclature of targets and ligands

Key protein targets and ligands in this article are hyperlinked to corresponding entries in http://www.guidetopharmacology.org, the common portal for data from the IUPHAR/BPS Guide to PHARMACOLOGY,[20] and are permanently archived in the Concise Guide to PHARMACOLOGY 2017/18.[21,22]

3 | RESULTS

3.1 | Patients and treatment exposure

A total of 51 patients were enrolled in the study and received pevonedistat plus fluconazole (n = 13) or pevonedistat plus itraconazole (n = 38). Overall, 47% of the patients were male, and most were white (80%). Median age was 64 years (range 30–88 years), median weight was 72 kg (range, 39–202), and median body surface area was 1.83 m^2 (range, 1.29–3.19 m^2). The most common cancer types were pancreatic (12%), breast, oesophageal, non-small cell lung and sarcoma (8% each) and ovarian (6%). All 51 patients had received at least one prior therapy, including chemotherapy (100%), surgery (90%) and radiation therapy (43%). Detailed patient baseline demographics and disease characteristics are summarized in Supplementary Table S2.

Forty-five patients (88%) received both scheduled doses of pevonedistat. Six patients received only one of the two scheduled pevonedistat doses: one had pevonedistat held in the pevonedistat plus fluconazole group, and one had pevonedistat held and four discontinued study drug in the pevonedistat 20 mg m^{-2} plus itraconazole group. Fifteen patients (29%) discontinued study treatment due to symptomatic deterioration (n = 7, 14%), AEs (n = 6, 12%), withdrawal of consent (n = 1, 2%), or ineligibility to participate in Part B because of alkaline phosphatase abnormality (n = 1, 2%).

3.2 | Pharmacokinetics

Data from 12 patients who were PK-evaluable were assessed for the effects of fluconazole on the PK of pevonedistat administered at 8 mg m^{-2}. The mean plasma concentration–time profiles and summary statistics of PK parameters for pevonedistat in patients treated on

day 1 with pevonedistat alone (reference condition) and on day 8 with pevonedistat plus fluconazole (test condition) are shown in Figure 2A and Table 1, respectively. The results of the statistical analysis of C_{max}, AUC_{last} and AUC_∞ for the estimation of the effect of fluconazole on the PK of pevonedistat are summarized in Table 2. Following IV administration of pevonedistat in the presence of fluconazole, pevonedistat systemic exposure, measured as geometric mean of AUC_∞, was similar to that observed in the absence of fluconazole (geometric mean ratio of 1.11 [90% CI, 1.03–1.19]).

Data from 33 patients who were PK-evaluable were assessed for the effects of itraconazole on the PK of pevonedistat administered

at 8, 15 or 20 mg m^{-2}. The mean plasma concentration–time profiles and summary of PK parameters for pevonedistat in patients treated with pevonedistat with and without itraconazole are presented in Figure 2B–D and Table 1, respectively. The results of the statistical analysis of C_{max}, AUC_{last} and AUC_∞ for estimation of the effect of itraconazole on the PK of pevonedistat are summarized in Table 2. Following IV administration of pevonedistat 8 mg m^{-2} in the presence of itraconazole, pevonedistat systemic exposure, measured as geometric mean of AUC_∞, was 123% of that observed in the absence of itraconazole (geometric mean ratio of 1.23 [90% CI, 1.12–1.35]) (Table 2). On the basis of these observations, additional patients were
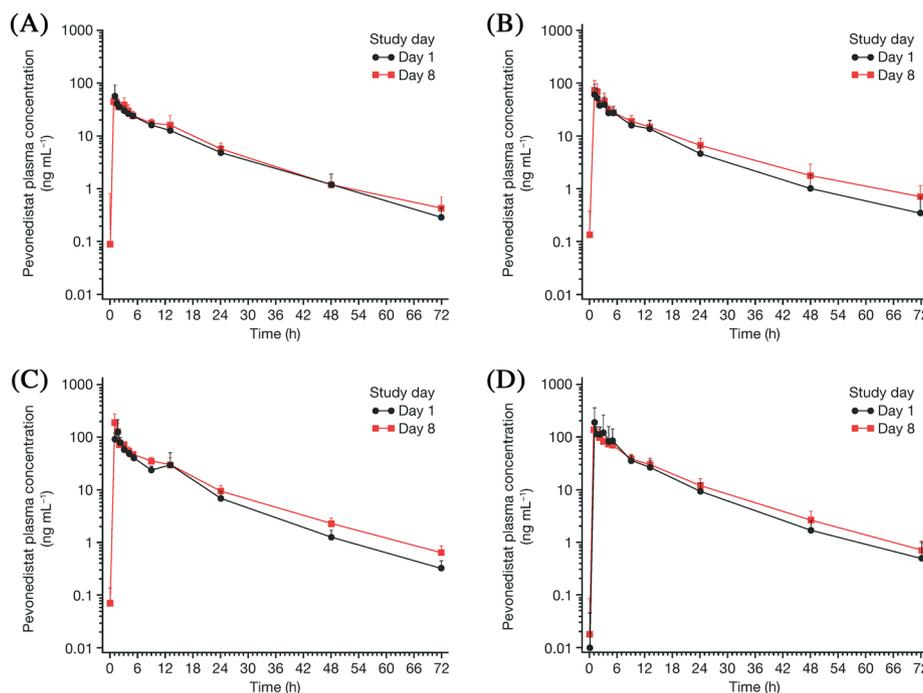
FIGURE 2 Mean pevonedistat plasma concentration-time profile in the absence (day 1) or presence (day 8) of CYP3A inhibitor: A, pevonedistat 8 mg m^{-2} ± fluconazole; B, pevonedistat 8 mg m^{-2} ± itraconazole; C, pevonedistat 15 mg m^{-2} ± itraconazole; D, pevonedistat 20 mg m^{-2} ± itraconazole

TABLE 1 Summary statistics of PK parameters of single-dose pevonedistat with and without co-administration of fluconazole or itraconazole

Co-administration	Pevonedistat dose, mg m^{-2}	n	C_{max}, ng mL^{-1a}	AUC_{last}, h*ng mL^{-1a}	AUC_∞, h*ng mL^{-1a}	$t_{1/2}$, h^b	CL, l h^{-1a}
– Fluconazole (day 1)	8	12	51.6 (61)	445 (16)	450 (16)	11.1 (1.9)	31.7 (25%)
+ Fluconazole (day 8)	8	12	51.0 (30)	491 (15)	498 (15)	11.7 (1.7)	28.6 (28%)
– Itraconazole (day 1)	8	13	59.1 (57)	459 (23)	465 (23)	10.8 (2.4)	29.2 (26%)
+ Itraconazole (day 8)	8	13	66.8 (52)	571 (24)	585 (24)	13.5 (2.6)	23.2 (29%)
– Itraconazole (day 1)	15	6	121 (68)	793 (18)	798 (18)	9.8 (0.74)	35.2 (17%)
+ Itraconazole (day 8)	15	6	193 (40)	1030 (18)	1060 (16)	11.0 (0.87)	26.5 (18%)
– Itraconazole (day 1)	20	14	178 (90)	1110 (34)	1120 (34)	10.0 (1.5)	35.8 (39%)
+ Itraconazole (day 8)	20	14	137 (33)	1140 (22)	1130 (23)	10.8 (1.0)	35.5 (36%)

^{a}Geometric mean (% CV).

^{b}Mean (standard deviation).

AUC_{last}, area under the plasma concentration–time curve from time 0 to time of the last quantifiable concentration; AUC_∞, area under the plasma concentration–time curve from 0 to infinity, calculated using the value of the last quantifiable concentration; C_{max}, maximum observed plasma concentration; CL, clearance; CV, coefficient of variation; PK, pharmacokinetics; $t_{1/2}$, terminal elimination half-life.

TABLE 2 Statistical analysis of plasma PK parameters for estimation of effect of fluconazole or itraconazole on pevonedistat PK

DDI cohort/PK parameter (units)	Geometric LS mean test condition (day 8)	Geometric LS mean reference condition (day 1)	Geometric LS mean ratio (%) (test/reference) (90% CI)
Pevonedistat (8 mg m^{-2}) + fluconazole	n = 12		
C_{max}, (ng mL^{-1})	50.995	51.642	98.75 (82.60–118.05)
AUC$_{last}$, (h*ng mL^{-1})	490.951	445.462	110.21 (102.34–118.69)
AUC$_\infty$, (h*ng mL^{-1})	498.422	450.412	110.66 (102.53–119.43)
Pevonedistat (8 mg m^{-2}) + itraconazole	n = 13		
C_{max}, (ng mL^{-1})	66.830	59.117	113.05 (85.35–149.74)
AUC$_{last}$, (h*ng mL^{-1})	558.062[a]	459.053	121.57 (110.92–133.24)
AUC$_\infty$, (h*ng mL^{-1})	571.589[a]	464.887	122.95 (112.13–134.82)
Pevonedistat (15 mg m^{-2}) + itraconazole	n = 6		
C_{max}, (ng mL^{-1})	192.690	120.769	159.55 (89.97–282.96)
AUC$_{last}$, (h*ng mL^{-1})	1031.923[a]	792.816	130.16 (106.99–158.35)
AUC$_\infty$, (h*ng mL^{-1})	1058.338	797.568	132.70 (111.10–158.49)
Pevonedistat (20 mg m^{-2}) + itraconazole	n = 14		
C_{max}, (ng mL^{-1})	137.395	177.824	77.26 (55.19–108.17)
AUC$_{last}$, (h*ng mL^{-1})	1129.221[a]	1114.116	101.36 (90.72–113.23)
AUC$_\infty$, (h*ng mL^{-1})	1131.593	1121.626	100.89 (91.14–111.68)
Pevonedistat dose-normalized PK + itraconazole[c]	n = 33		
C_{max}, (ng mL^{-1})	4.480	4.374	102.41 (83.12–126.19)
AUC$_{last}$, (h*ng mL^{-1})	34.172[b]	30.077	113.61 (105.65–122.18)
AUC$_\infty$, (h*ng mL^{-1})	34.668[a]	30.346	114.24 (106.54–122.50)

[a]Value calculated based on data from n − 1 patients.

[b]Value calculated based on data from n − 2 patients.

[c]Natural log-transformed dose-normalized PK parameters were fit using a mixed effects model with study day as a fixed effect and patient as a random effect. Geometric LS means and geometric LS mean ratio were back-transformed LS mean and treatment mean differences.

AUC$_{last}$, area under the plasma concentration–time curve from time 0 to time of the last quantifiable concentration; AUC$_\infty$, area under the plasma concentration–time curve from 0 to infinity, calculated using the value of the last quantifiable concentration; C_{max}, maximum observed plasma concentration; CI, confidence interval; DDI, drug–drug interaction; LS, least-squares; PK, pharmacokinetics.

enrolled to assess the effects of itraconazole on pevonedistat PK at higher and clinically relevant doses. While not an objective of the study, the ANOVA results for the 15 mg m^{-2} safety lead-in cohort showed that the mean AUC$_\infty$ ratio was 133%; some caution should, however, be applied when interpreting these data given the small sample size of six patients for reliable estimation, as evidenced by the wide 90% CIs, at this intermediate dose level prior to escalating to 20 mg m^{-2}. Pevonedistat systemic exposure following administration of the clinically relevant dose of pevonedistat 20 mg m^{-2} with itraconazole was similar to that without itraconazole (geometric mean AUC$_\infty$ ratio of 1.01 [90% CI, 0.911–1.12]). Furthermore, as no consistent trends in geometric mean ratios of pevonedistat AUC (co-administered with itraconazole vs. administered alone) were observed in relation to dose in the study, dose-normalized pevonedistat PK parameters were used for an integrated assessment of the effects of itraconazole on pevonedistat PK across the dose range of 8–20 mg m^{-2} in order to leverage the strength of all available data. Dose-normalization used the actual daily dose of pevonedistat received on each study day, which was calculated using the assigned dose level, patient body surface area

at baseline, and adjusted volume of the IV bag actually infused. This integrated analysis indicated that the exposure of pevonedistat was only minimally increased by itraconazole (14% increase; geometric mean AUC$_\infty$ ratio of 1.14 [90% CI, 1.07–1.23]); a summary of the statistical analysis results is presented in Table 2.

3.3 | Safety

All patients were included in the safety population, of which 50 patients (98%) experienced at least one AE and 28 (55%) experienced at least one drug-related AE. The most common AEs by preferred term are listed in Table 3. Across all study groups, the most common drug-related AEs included vomiting (n = 6, 12%), diarrhoea (n = 5, 10%), nausea (n = 4, 8%), and increased AST, fatigue, pruritus, anaemia and decreased appetite (n = 3, 6% each). Fifty-five percent (n = 28) of patients experienced AEs with a maximum intensity of only grade 1 or 2, while 43% (n = 22) of patients reported at least one grade ≥3 AE. Two patients (5%) in the pevonedistat plus itraconazole group

TABLE 3 Most common (≥10% overall) AEs by preferred term

| Adverse event | Pevonedistat 8 mg m^{-2} + fluconazole (n = 13) | Pevonedistat + itraconazole | | | | Total (N = 51) |
		Pevonedistat 8 mg m^{-2} (n = 13)	Pevonedistat 15 mg m^{-2} (n = 6)	Pevonedistat 20 mg m^{-2} (n = 19)	Total (n = 38)	
Fatigue	3 (23)	3 (23)	1 (17)	8 (42)	12 (32)	15 (29)
Anaemia	3 (23)	3 (23)	2 (33)	6 (32)	11 (29)	14 (27)
Decreased appetite	4 (31)	3 (23)	1 (17)	4 (21)	8 (21)	12 (24)
Vomiting	3 (23)	2 (15)	1 (17)	6 (32)	9 (24)	12 (24)
Diarrhoea	0	4 (31)	1 (17)	6 (32)	11 (29)	11 (22)
Headache	3 (23)	0	0	6 (32)	6 (16)	9 (18)
Constipation	3 (23)	2 (15)	0	3 (16)	5 (13)	8 (16)
Dehydration	1 (8)	3 (23)	2 (33)	2 (11)	7 (18)	8 (16)
Dizziness	1 (8)	2 (15)	1 (17)	4 (21)	7 (18)	8 (16)
Dyspnoea	2 (15)	1 (8)	2 (33)	3 (16)	6 (16)	8 (16)
Abdominal pain	1 (8)	3 (23)	1 (17)	2 (11)	6 (16)	7 (14)
Asthenia	3 (23)	2 (15)	0	2 (11)	4 (11)	7 (14)
Cough	2 (15)	4 (31)	0	1 (5)	5 (13)	7 (14)
Hypokalaemia	0	2 (15)	2 (33)	3 (16)	7 (18)	7 (14)
Hyponatraemia	2 (15)	1 (8)	1 (17)	2 (11)	4 (11)	6 (12)
Nausea	3 (23)	1 (8)	0	2 (11)	3 (8)	6 (12)

All data are shown as n (%).

had drug-related grade ≥3 AEs of diarrhoea (n = 1) and increased ALT and increased AST (n = 1). Thirteen patients (25%) had serious AEs, which were not deemed to be drug-related, and seven patients (14%) discontinued the study due to AEs. Overall, seven patients (14%) died during Part A of the study due to sarcoma, metastatic squamous cell carcinoma, intestinal obstruction, metastatic pancreatic carcinoma, metastatic endometrial carcinoma, ovarian cancer and oesophageal haemorrhage. All fatal AEs were related to the respective diseases and not to the study drug. Qualified researchers may request data from Takeda. Complete details are available at the following: https://www.takedaclinicaltrials.com/approach#commitment.

4 | DISCUSSION

In patients with MDS, leukaemias and other neutropenic cancers at risk of developing infections, azole antifungals are widely used in both prophylaxis and treatment due to their efficacy and limited AE profile.[23] However, many of these, such as posaconazole and voriconazole, are potent CYP3A4 inhibitors and must be used with caution in patients who require concurrent treatment with CYP3A4 substrates such as simvastatin, midazolam, amlodipine and other agents sensitive to CYP3A4 inhibition.

In vitro metabolism studies suggested that CYP3A4 plays a major role in pevonedistat elimination pathways. This open-label, multicentre, parallel-group, two-arm, phase I study was conducted to evaluate the effect of a moderate CYP3A inhibitor, fluconazole, and a strong CYP3A/P-gp inhibitor, itraconazole, on pevonedistat PK in

patients with advanced solid tumours. After patients completed the DDI assessment, they were given the opportunity to participate in the optional Part B portion of the study after an adequate washout period. Both regimens, docetaxel and carboplatin plus paclitaxel, are approved standard-of-care therapies for various malignancies in front-line or relapsed/refractory settings. The dose of pevonedistat used in Part B in these chemotherapeutic combinations has been previously established as the maximum tolerated dose and recommended phase II dose for these combinations based on results from a phase Ib dose-finding study.[18]

Pevonedistat, which is currently being investigated as an anticancer agent, targets the NEDD8-conjugation pathway within the ubiquitin-proteosome system. It inhibits NAE activity, thereby disrupting proteasomal degradation of a variety of critical regulatory proteins integral to tumour cell growth, proliferation and survival, resulting in DNA damage response and cell death.[4,5] Preclinical studies show that pevonedistat is cytotoxic to a range of solid and haematopoietic tumour cell lines,[5,9,24] and changes in pharmacodynamic biomarkers, indicative of target and pathway inhibition following pevonedistat treatment, were detected at all doses studied in humans, thus precluding the conduct of this study in healthy volunteers.

Fluconazole and itraconazole are among the choices of moderate and strong inhibitors of CYP3A, respectively, recommended in regulatory guidelines.[14,15] Itraconazole was chosen instead of ketoconazole based on FDA communications advising against the use of ketoconazole for DDI studies due to serious side effects.[25] Because of the known overlap in CYP3A and P-gp specificity, several azole

antifungals effectively inhibit the human P-gp transport function; however, they do not necessarily share similar inhibition potency on the enzyme and transporter. While itraconazole is a strong dual inhibitor of CYP3A and P-gp, fluconazole is classified as a moderate CYP3A inhibitor, but devoid of P-gp inhibitory effect.[26,27] As the extent of DDIs between CYP3A and/or P-gp inhibitors and pevonedistat was uncertain at the time of designing this study, in the interest of patient safety we utilized a conservative approach and analysed the effects of fluconazole before proceeding to conduct the DDI assessment with itraconazole. Additionally, the pevonedistat dose was started at 8 mg m^{-2} and proceeded to 15 mg m^{-2} and then the clinically relevant dose of 20 mg m^{-2}, which is being used in ongoing phase II and phase III studies of pevonedistat in combination with azacitidine in patients with higher-risk MDS, chronic myelomonocytic leukaemia or low-blast AML (NCT02610777 and NCT03268954, clinicaltrials.gov). The results from this study are intended to inform concomitant use of CYP3A/P-gp inhibitors in cancer patients receiving pevonedistat.

Data from 12 PK-evaluable patients were obtained to assess the effects of fluconazole on the PK of pevonedistat administered at 8 mg m^{-2}. Following IV administration in the presence of fluconazole, the pevonedistat systemic exposure, measured as geometric mean of AUC$_\infty$, was similar to that observed in the absence of fluconazole (geometric mean ratio of 1.11 with an associated 90% CI of 1.03–1.19), indicating that multiple-dose administration of fluconazole had no clinically relevant effects on pevonedistat PK. Similarly, pevonedistat systemic exposures at a single dose of 8 mg m^{-2} increased by 23% on average in the presence of itraconazole. Given these modest drug interaction effects with two validated inhibitor probes and apparent discrepancy with the in vitro metabolism data indicating a major role of CYP3A4 in pevonedistat metabolism, the additional clinically relevant dose of pevonedistat 20 mg m^{-2} was subsequently selected, with an intermediate dose of 15 mg m^{-2} as a safety lead-in step, for further clinical investigation. As clearance estimates of pevonedistat in this study were comparable across the dose range of 8–20 mg m^{-2}, and consistent with previous reports that pevonedistat PK is linear up to 261 mg m^{-2} in cancer patients, it was deemed appropriate to pool the PK information generated to assess the effects of itraconazole on pevonedistat PK. This integrated analysis used dose-normalized pevonedistat PK parameters from 33 PK-evaluable patients across the pevonedistat dose range of 8–20 mg m^{-2}. The systemic exposure of pevonedistat in the presence of itraconazole was only minimally increased compared with that in the absence of itraconazole (geometric mean dose-normalized AUC$_\infty$ ratio of 1.14, with an associated 90% CI of 1.07–1.23). The 90% CI of the AUC$_\infty$ geometric mean ratio was contained within the 80–125% statistical equivalence range.[15] In addition, the 14% increase in the pevonedistat geometric mean dose-normalized AUC$_\infty$ was not considered clinically meaningful when viewed in the context of overall pevonedistat PK variability (CV of 15–34% in AUC$_\infty$, Table 1), supporting inference of the lack of a clinically relevant effect of itraconazole on total systemic exposure (AUC) of pevonedistat. These findings with established moderate and strong CYP3A inhibitor probes indicate a minor contribution of

CYP3A/P-gp to pevonedistat metabolism in humans. As a result, the restriction on the use of moderate/strong CYP3A inhibitors was lifted in the ongoing pevonedistat phase III PANTHER trial (NCT03268954, clinicaltrials.gov).

As noted above, in vitro metabolism studies suggested a major role for CYP3A in pevonedistat elimination pathways, and yet the results of this DDI trial are consistent with only a minor contribution of CYP3A/P-gp in humans. It should be noted that while the Km for metabolism of pevonedistat by CYP3A4 could not be determined, due to its low metabolic turnover in in vitro experimental systems, the lack of sensitivity of pevonedistat in vivo clearance to itraconazole is not believed to be related to a very low Km for CYP3A4 compared to the Ki for itraconazole. Itraconazole and its metabolites are highly potent inhibitors of CYP3A4 with unbound IC$_{50}$ values of 0.4–7 nM.[28] Furthermore, the unbound maximum plasma concentrations of pevonedistat at the clinical dose of 20 mg m^{-2} is 8.25 ng mL^{-1} (18.6 nM) which is well below the typical range of Km values ($\geq$0.35 μM) for metabolism by CYP3A4.[29] A human radiolabelled mass balance study (NCT03057366), clinicaltrials.gov is currently ongoing and is expected to characterize pevonedistat metabolism and elimination pathways in cancer patients. The results of this study is expected to provide a potential explanation on the discordance between in vitro metabolism data and clinical observations in this DDI study with moderate/strong CYP3A inhibitors. Based on the preliminary data from this ongoing study (data on file), approximately 80% of the administered dose of pevonedistat is accounted for by metabolites. The hydroxylation/oxidation pathway leading to the major metabolites accounts for approximately 70% of the total dose. In vitro enzymology studies of the primary human biotransformation pathways of pevonedistat, including reaction phenotyping, at clinically relevant concentrations are ongoing to identify hepatic and extrahepatic enzymes involved in pevonedistat metabolism.

The toxicity profile of pevonedistat in combination with fluconazole or itraconazole was consistent with that reported in previous single-agent pevonedistat studies.[6,10,11,13] More than 50% of patients experienced AEs with a maximum intensity of grade 1 or 2, and the only drug-related grade 3 AEs, observed in two patients who received pevonedistat plus itraconazole, were diarrhoea (n = 1) and increased ALT and increased AST (n = 1). Although the patient numbers were small in each subgroup, there were no clinically relevant differences in the frequency or severity of AEs between the fluconazole and itraconazole groups or among the pevonedistat dose subgroups in the pevonedistat plus itraconazole group.

In conclusion, fluconazole, a moderate CYP3A inhibitor, and itraconazole, a strong CYP3A/P-gp inhibitor, had no clinically meaningful effects on pevonedistat PK. The clinical PK profile of pevonedistat is comparable in patients with haematological or solid tumour malignancies[6,10,11,13,30] and therefore, the study results indicate that the use of moderate/strong CYP3A inhibitors and P-gp inhibitors is permitted in patients receiving pevonedistat, regardless of the cancer type. Of particular relevance, no dose adjustment of pevonedistat is needed when co-administered with CYP3A-inhibitory azole antifungal agents (e.g., itraconazole, posaconazole), which are

commonly used medications for systemic fungal infections in patients with haematologic malignancies.

CONTRIBUTORS

All authors contributed to the writing and critically revised the manuscript, reviewed and approved the final version of the manuscript, and agreed to be accountable for all aspects of the work.

ORCID

Hélène Faessel https://orcid.org/0000-0002-3007-3345
Karthik Venkatakrishnan https://orcid.org/0000-0003-4039-9813

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SUPPORTING INFORMATION

Additional supporting information may be found online in the Supporting Information section at the end of the article.