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ARTICLE

A Physiologically Based Pharmacokinetic Model to Predict Potential Drug–Drug Interactions and Inform Dosing of Acumapimod, an Oral p38 MAPK Inhibitor

Alex Agyemang[1], Colm Farrell[2], William Moore[3] and Jacqueline Parkin[3,*]

Acumapimod, an investigational oral p38 mitogen-activated protein kinase inhibitor for treatment during severe acute exacerbations of chronic obstructive pulmonary disease, is metabolized primarily by cytochrome P450 3A4 (CYP3A4) and is a P-glycoprotein (P-gp) substrate. Concerns about drug–drug interactions (DDIs) have meant patients receiving drugs that inhibit CYP3A4 were ineligible for acumapimod trials. We report on how 2 acumapimod clinical DDI studies and a physiologically-based pharmacokinetic (PBPK) model assessing how co-administration of a weak (azithromycin) and strong (itraconazole) CYP3A4 inhibitor affected acumapimod systemic exposure, informed decision making and supported concomitant use of CYP3A4 and P-gp inhibitors. Studies MBCT102 and MBCT103, respectively, demonstrated that co-administration of azithromycin or itraconazole had no clinically meaningful impact on acumapimod pharmacokinetics. Findings were consistent with PBPK model results. Safety profiles were similar when acumapimod was co-administered with azithromycin or itraconazole. These studies highlight the value of PBPK modeling in drug development, and its potential to inform DDI investigations.

Study Highlights

WHAT IS THE CURRENT KNOWLEDGE ON THE TOPIC?

☑ Acumapimod is an oral p38 mitogen-activated protein kinase inhibitor for acute exacerbations of chronic obstructive pulmonary disease treatment. Such inhibitors can be hepato-/neurotoxic, so safety concerns exist regarding their increased exposure. Acumapimod is metabolized by CYP inhibitors, and many drugs it may be co-administered with are CYP3A4 inhibitors, which could increase its exposure.

WHAT QUESTION DID THIS STUDY ADDRESS?

☑ To investigate whether CYP3A4 inhibitors impact acumapimod pharmacokinetics (PKs), requiring its dose to be modified during co-administration to avoid drug–drug interactions (DDIs), a physiologically-based pharmacokinetic (PBPK) model was used to predict DDIs and provide confidence that the therapeutic acumapimod dose was safe for ongoing clinical studies.

WHAT DOES THIS STUDY ADD TO OUR KNOWLEDGE?

☑ CYP3A4 inhibitor co-administration had little impact on acumapimod PKs. PBPK model results provided confidence to safely co-administer strong CYP3A4 inhibitors with acumapimod in the clinical DDI studies.

HOW MIGHT THIS CHANGE DRUG DISCOVERY, DEVELOPMENT, AND/OR THERAPEUTICS?

☑ The study shows the development value of clinical pharmacology and simulation science; through rapid understanding of acumapimod drug interactions, it was possible for use of CYP3A4 inhibitors to be permitted in an ongoing clinical chronic obstructive pulmonary disease study to enhance recruitment of a population on expected concomitant medications.

Acumapimod is an investigational oral p38 mitogen-activated protein kinase inhibitor being developed to be given on top of standard care (including systemic steroids and antibiotics) for the treatment of severe acute exacerbations of chronic obstructive pulmonary disease (AECOPD).[1] A 5-day acumapimod regimen suppresses inflammation, and there is some evidence for a reduction of recurrent exacerbation risk.[1,2] Because p38 mitogen-activated protein kinase inhibitors can cause hepatotoxicity, neurotoxicity, and acneiform skin rash, there are theoretical but specific safety concerns regarding increased exposure to this drug class.[3,4]

Acumapimod is metabolized primarily by cytochrome P450 3A4 (CYP3A4) and is a substrate of P-glycoprotein (P-gp). Drugs frequently used for chronic obstructive pulmonary disease exacerbations and commonly associated comorbidities (e.g., macrolide antibiotics and calcium channel blockers) may inhibit CYP3A4 and, therefore, may increase acumapimod exposure close to or beyond the

[1]Barts and The London School of Medicine and Dentistry, London, UK; [2]ICON Clinical Research, Marlow, UK; [3]Mereo BioPharma Group plc, London, UK. *Correspondence: Jacqueline Parkin (jp@mereobiopharma.com)

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maximum safety exposure limits.[5,6] As a result, participants receiving these drugs were ineligible to enter acumapimod trials. One such acumapimod trial in which recruitment from the wider patient population was not initially possible was study MBCT206, a phase II study that evaluated acumapimod plus standard of care vs. placebo plus standard of care in the treatment of patients with AECOPD.[7] Hence, it was important to determine the extent to which co-administration of CYP3A4 and P-gp inhibitors would impact the pharmacokinetics (PKs) of acumapimod.

A strategy was used to gain timely initial evidence to support concomitant use of macrolide antibiotics in the MBCT206 study, and to inform subsequent investigations regarding whether the acumapimod dose would require modification to avoid drug–drug interactions (DDIs) in planned phase III trials. Study MBCT102 assessed the effect on acumapimod PK when acumapimod was co-administered with azithromycin, a weak CYP3A4 and P-gp inhibitor commonly used in the treatment of AECOPD.[5] The results from MBCT102 enabled azithromycin to be co-administered in the MBCT206 study, facilitating recruitment. However, there remained the need to determine potential interaction(s) with moderate and strong inhibitors of CYP3A4 and/or P-gp. Moving directly to a strong inhibitor for the next DDI study was the most efficient approach, therefore, study MBCT103 was planned to assess the effect of acumapimod co-administration with itraconazole, a strong CYP3A4 inhibitor.[8,9] However, without evidence to assess the potential extent of elevation of acumapimod systemic exposure following co-administration with a strong CYP3A4 inhibitor, the selection of an appropriate and safe acumapimod dose for study MBCT103 would be challenging. Consequently, a physiologically-based PK (PBPK) model was developed to predict how co-administration of a moderate or strong CYP3A4 inhibitor with acumapimod affects acumapimod systemic exposure, to inform decision making and increase confidence that toxicity margins would not be exceeded. The PBPK model was verified by comparing its predictions for the interaction between the weak CYP3A4 inhibitor azithromycin and acumapimod, with the PK parameters from the azithromycin-acumapimod DDI clinical study (MBCT102). Following successful verification, the model was prospectively applied to predict the acumapimod exposure when co-administered with a moderate or strong CYP3A4 inhibitor.[9] The PBPK model predictions obviated the need for a clinical study with a moderate CYP3A4 inhibitor and provided confidence to proceed directly to assess for potential interactions when acumapimod is co-administered with itraconazole in a clinical setting (study MBCT103).

PBPK modeling has been used in other studies to predict DDIs, including to predict possible DDIs upon concomitant administration of ixazomib, osimertinib, and evofosfamide with CYP3A inhibitors or inducers,[10–12] and its use in this context is increasing. According to current US Food and Drug Administration (FDA) and European Medicines Agency guidance, PBPK modeling is a powerful tool to explore and quantitatively predict DDIs, and may offer an alternative to dedicated clinical trials.[13]

We report here on the results of 2 clinical DDI studies on acumapimod (MBCT102 and MBCT103) and the strategy that was applied, including integration of a PBPK model to inform decision making and expedite/facilitate the development process.

METHODS

Study MBCT102

Study design and subjects. This phase I, open-label, 2-period, single-sequence, crossover, single-center study was conducted between August 26, 2016, and October 20, 2016, in the UK (ClinicalTrials.gov: NCT02926326).[14] The study was conducted in accordance with the Declaration of Helsinki and International Conference on Harmonisation (ICH) Guideline for Good Clinical Practice (GCP). The study protocol and any documents and/or information given to the subjects to recruit or record data were reviewed and approved by an appropriate independent ethics committee. All subjects provided written informed consent before screening.

Eligible subjects were healthy men, aged 18−65 years, with a body mass index between 18 and 30 kg/m^2 inclusive. Subjects were nonsmokers for ≥ 6 months before receiving the first dose of study drug, and for the study duration.

The main exclusion criteria were clinically relevant laboratory values or conditions, including liver function tests or abnormal electrocardiogram, pre-existing active skin disease, testing positive for HIV, hepatitis B or hepatitis C, and contraindication or hypersensitivity to the treatments or excipients used during the studies. Subjects were also excluded for use of any other medication within 14 days prior to study drug administration until the end of the study, with the exception of paracetamol. Use of P-gp inhibitors, CYP3A4 inhibitors and inducers, and consumption of products containing grapefruit or Seville oranges were not permitted within 14 days prior to study drug administration, during study confinement, and during washout periods.

Study treatment. Subjects participated in a screening visit and two in-house study periods (period 1 and period 2), separated by a ≥ 14-day washout period between acumapimod doses. In period 1, subjects were admitted to the study unit on the evening before dosing with a single dose of acumapimod 14 mg (2 × 7 mg capsules) on day 1 and observed in the unit until discharge on the morning of day 3. In period 2, subjects were admitted to the unit on the evening of day –2 before receiving 3 daily doses of azithromycin 500 mg (days –1 to day 2) and a single acumapimod dose on day 1 (24 hours after the first dose of azithromycin). Subjects were discharged on the morning of day 3 (**Figure 1**). The overall study duration was 38 days (day –1 to follow-up telephone call), which included the washout period but excluded the screening period.

End points and assessments Primary objective and end points. The primary objective was to estimate the magnitude of effect caused by multiple doses of azithromycin on the PKs of a single dose of acumapimod. The primary end points were maximum plasma concentration (C_{max}), area under the plasma concentration–time curve (AUC) from drug administration

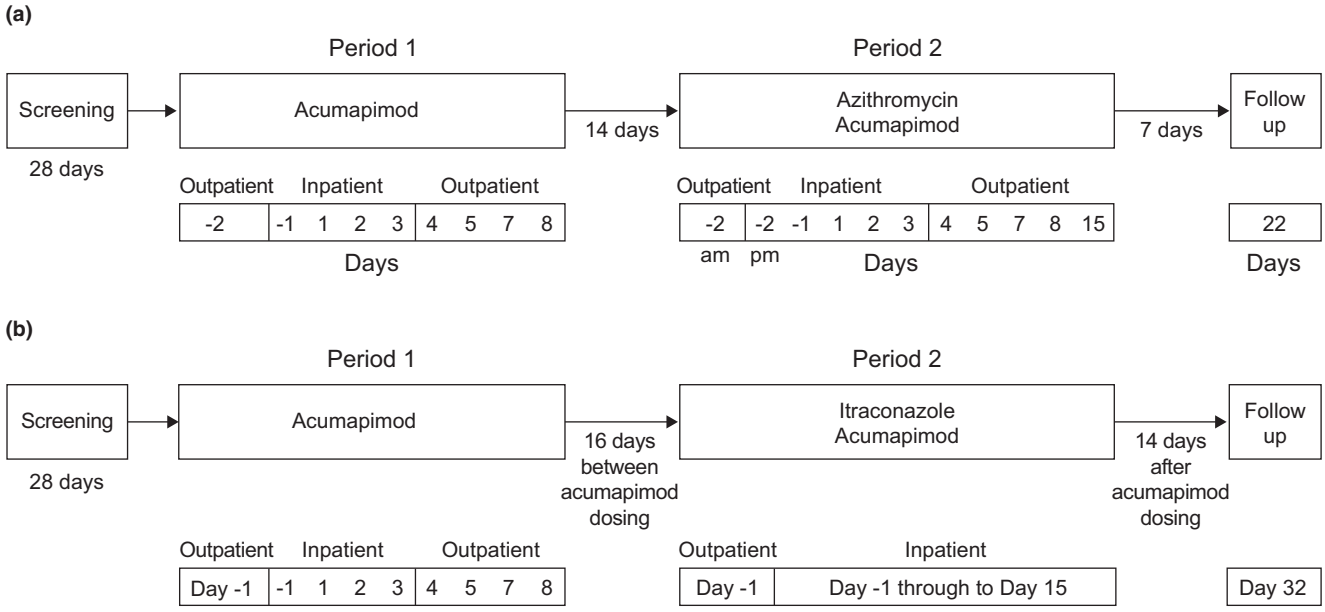

Figure 1 Drug–drug interaction study designs. Study treatment and pharmacokinetic sampling for periods 1 and 2 of azithromycin (study MBCT102) (**a**) and itraconazole (study MBCT103) (**b**) treatment.

to last observed concentration at time t (AUC_{0-t}), and AUC extrapolated to infinite time ($AUC_{0-\infty}$) of acumapimod.

Pharmacokinetic analysis. Blood samples for the acumapimod PK analysis were collected pre-acumapimod dose and at 1, 2, 4, 6, 12, 24, 36, 48, 72, 96, 144, and 168 hours postdose for both treatment periods, with a further postdose sample at 336 hours during period 2. Both periods included 4 outpatient visits from days 4 to 8 for PK and safety assessments. After period 2, subjects returned for a final PK sample and post-study physical examination on day 15 and received a follow-up telephone call 22 days after the second period. Adverse events (AEs) were captured from check-in to the study unit on day –1 until the post-study physical examination. Plasma samples were separated in a refrigerated centrifuge (4°C) at 1008 g for 10 minutes. Plasma samples were analyzed using validated liquid chromatography followed by tandem mass spectrometry detection methods. PK was tested using K2 EDTA plasma samples, using aliquots of 25 µL extracted by protein precipitation through an Ostro plate (Ostro Protein Precipitation & Phospholipid Removal Plate, 25 mg; Waters, Milford, MA) and detected by liquid chromatography with tandem mass spectrometry. The calibration range of the assay for lower and upper limits of quantitation was 1.0 to 1,000 ng/mL. The quality control levels were at 1.0 (lower limits of quantitation), 3.0 (low), 40.0 (medium), 800 (high), and 1,000 (upper limits of quantitation) ng/mL. Inter-run and intra-run precision percentage coefficient of variation and accuracy percentage relative error are shown in **Table S1**.

Safety and tolerability. The safety and tolerability of the study treatment was evaluated by recording AEs and serious AEs (SAEs). An SAE was defined as an AE that resulted in death, or persistent or significant disability or incapacity, was life-threatening, required (or prolonged) hospitalization, or was a congenital abnormality or birth defect. A treatment-emergent AE (TEAE) was defined as an AE not present before treatment initiation and/or an AE already present that worsened in intensity or frequency following drug(s) exposure. AEs were classified as mild, moderate, or severe. Mild AEs were an awareness of sign, symptom, or event, but easily tolerated. Moderate AEs caused enough discomfort to result in interference with usual activity and may have warranted intervention. Severe AEs were either incapacitating, with inability to do usual activities, or significantly affected clinical status and warranted intervention.

Statistical analyses. A planned enrollment of 16 healthy subjects allowed for 4 withdrawals and 12 subjects to complete the study. The sample size rationale for PK analysis was based on consideration of the precision of the estimate of the geometric mean ratio (GMR) of C_{max} of acumapimod with and without co-administered azithromycin. With a sample size of 12 subjects completing the study, there was a 90% probability for the 90% confidence interval (CI) to be within 80% and 125% of the point estimate of the GMR for C_{max}.

All subjects who received at least one dose of the study drug without a major protocol deviation were included in the PK set for PK analysis. Those who received ≥ 1 dose of the study drug, and for whom a safety assessment was available, were included in the safety set.

The PK analysis was performed using Phoenix WinNonLin (version 6.3.1; Certara, Princeton, NJ). Standard noncompartmental methods were used for the calculation of the plasma parameters from the plasma drug concentration–time data.

Primary PK parameters (AUC_{0-t}, $AUC_{0-\infty}$, and C_{max}) were ln-transformed and analyzed using an analysis of variance. Therefore, the calculation of the GMR (acumapimod plus azithromycin or acumapimod alone) with its 90% two-sided CI was performed based on analysis of ln-transformed data of these primary PK parameters for acumapimod. Back-transformed 90% CIs for the ratios of the geometric means were used to assess the magnitude of the effect of azithromycin on the PKs of acumapimod. The mean, geometric mean, SD, percentage coefficient of variation, SEM, and range (minimum, median, and maximum) were calculated for plasma concentrations at each sampling time. All statistical procedures were performed using SAS (version 9.2 or a higher version; Cary, NC).

Physiologically-based pharmacokinetic modeling. The Simcyp PBPK Simulator parameter module (Certara, Princeton, NJ) was used to optimize key parameter values (first-order absorption rate constant (k_a) and lag time (T_{lag})). This enabled a link between the "bottom-up" and "top-down" PK modeling approaches, allowing more accurate DDI prediction simulations to be carried out. The PBPK model (**Figure S1**) was developed using Simcyp version 16.1 by incorporating data describing the contribution of CYP3A4 and carboxylesterase-1-mediated hydrolysis.

Data were derived from *in vitro* studies, and human absorption, distribution, metabolism, and excretion, and mass balance studies. First-order absorption was assumed in all simulations, and the minimal PBPK model with a single adjusting compartment was applied in all simulations of the plasma concentration–time profiles for acumapimod. A single adjusting compartment considers metabolism in the liver, intestines, and kidneys, and combines other tissues together, as incorporated in the Simcyp PBPK Simulator.

The DDI mechanism incorporated in the model was that of CYP3A4. The locations of the DDIs included the liver, kidneys, and biliary system. Data from the human absorption, distribution, metabolism and excretion study (CBCT197A2205) were used to estimate the extent of absorption, as well as the relative contributions of CES1-mediated hydrolysis, CYP3A4-mediated metabolism (fm_{CYP3A4}), and renal clearance to the disposition of acumapimod. Clinical PK profiles suggested potential enterohepatic circulation of acumapimod and, based on the mass balance data, several scenarios were possible depending on whether or not biliary clearance was a significant clearance component. Due to the uncertainty in fm_{CYP3A4} (as a result of uncertainty in biliary clearance), 2 models were developed to assess the best (fm_{CYP3A4} 26%) and worst (fm_{CYP3A4} 50%) case scenarios for the DDI assessment (**Table S3**). Model 1 assumed 33.9% of the dose recovered in the feces to be mediated by biliary clearance (without assigning to a specific hepatic efflux transporter; e.g., P-gp), followed by enterohepatic recirculation of drugs excreted in bile. Model 2 assumed 33.9% of the dose recovered in the feces to be unabsorbed drug. In both models, 97.8% of CYP-mediated metabolism was assigned to CYP3A4 based on *in vitro* phenotyping data.

The PBPK model for acumapimod was verified against clinical data arising from single and multiple dosing regimens of acumapimod (i.e., 10–75 mg), in addition to a DDI study in healthy subjects after a single oral dose of 14 mg with and without azithromycin treatment. The model was considered to be acceptable when the ratio of the predicted vs. observed parameter was not > 1.25-fold.[15]

For the DDI assessment, concentration–time profiles were simulated using 10 virtual trials of 16 male subjects aged 27–37 years. Simulations were based on subjects receiving a single oral dose of 14 mg acumapimod on day 2, in the presence and absence of azithromycin treatment (500 mg q.d. for 3 days) under fasted conditions (**Figure 2**). The simulation duration was 336 hours after the first dose of azithromycin in line with the study protocol.

For single and multiple oral dosing regimens of acumapimod (10–75 mg) in healthy volunteers, both models 1 and 2 predicted the PK parameters of $AUC_{0-\infty}$ and C_{max} within 1.4-fold of the reported values at all dose levels (**Tables S4** and **S5**). A discrepancy was observed between the simulated and observed terminal half-life at dose levels higher than 14 mg due to the simulated profiles not capturing the secondary peak at 24–48 hours postdose; however, this limitation was not expected to significantly impact the predicted DDI effects due to CYP3A4 inhibition.

The acumapimod PBPK model was applied prospectively to predict the plasma concentrations and PK parameters of a single oral dose of acumapimod (75 mg) following coadministration of a strong CYP3A4/P-gp inhibitor itraconazole (200 mg q.d.), or a moderate CYP3A4 inhibitor verapamil (240 mg q.d.), for 20 days.[9,16] For both itraconazole and verapamil, these predictions were based on 10 virtual trials of 10 healthy male subjects aged 27–37 years.[17] Each virtual subject received a single 75 mg dose of acumapimod orally on day 5, in the presence and absence of itraconazole, simulated under fasted conditions (**Figures 3** and **4**). The input parameters for the azithromycin model can be found in **Table S2** and those for the mechanism-based inhibition used for verapamil are in **Table S12**.

Study MBCT103

Study design and subjects. This phase I, open-label, 2-period, single-sequence, crossover, single-center study was conducted between March 19, 2018, and May 11, 2018, in the United States (ClinicalTrials.gov: NCT03498170).[18] Study design and subject inclusion/exclusion criteria were as for study MBCT102, except that itraconazole was coadministered with acumapimod instead of azithromycin. The study was conducted in accordance with the Declaration of Helsinki and ICH Guideline for GCP and the requirements of the Code of Federal Regulations, the Institutional Review Board/IEC, FDA, and all other applicable local regulatory requirements. The clinical study protocol, any relevant associated documents, and informed consent forms were reviewed and approved by Chesapeake Institutional Review Board Services, an independent service provider.

Treatment. Period 1 was as for study MBCT102. In period 2, subjects were admitted to the study unit on the morning of day –1 before receiving once-daily doses of itraconazole 200 mg as an oral solution (day 1 to day 14) and a single

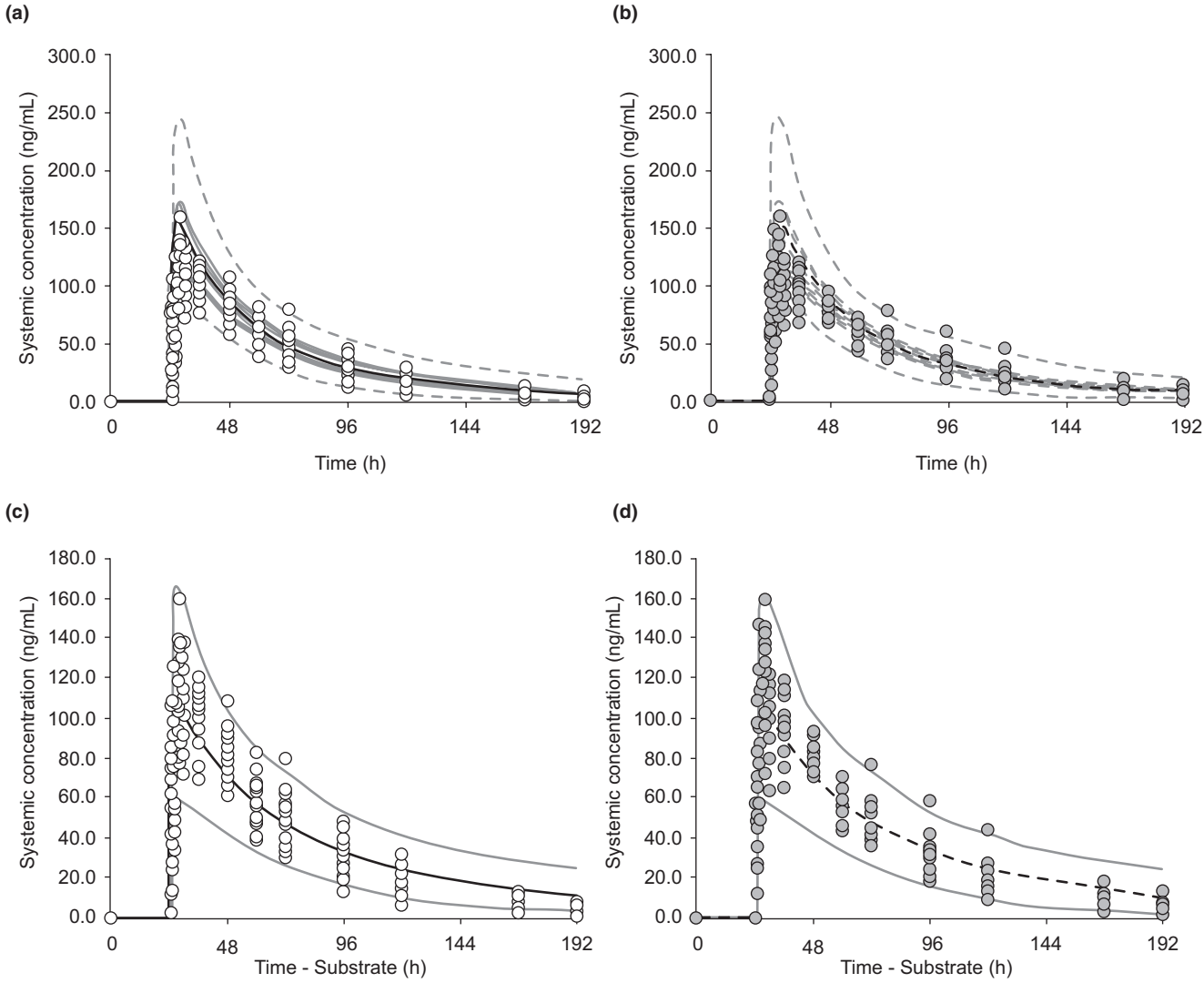

Figure 2 Simulated and observed (data points; from study MBCT102) mean plasma concentration–time profiles of acumapimod. After a single oral dose (14 mg) in the absence (**a** and **c**) and presence (**b** and **d**) of multiple oral daily doses of azithromycin (500 mg for 3 days). In **a** and **b**, model 1 was used; in **c** and **d**, model 2 was used. The grey lines represent the outcomes of simulated individual trials (10 × 16 virtual individuals) and the black line is the mean data for the simulated population (n = 160). The grey dashed line represents the 95th and 5th percentile of the simulated data. It is not possible to see the early phase of the profile (between 0 and 24 hours) due to the scale of the x-axis in this figure.

dose of acumapimod on day 7 (2 hours post-itraconazole dosing). Subjects were discharged on the morning of day 15 (**Figure 1**). The overall study duration was ~35 days (day –1 to follow-up assessment), which included a washout period of at least 16 days between the acumapimod doses but excluded the screening period.

End points and assessments Primary objective and end points. These were as for study MBCT102, except that itraconazole was co-administered with acumapimod instead of azithromycin.

Pharmacokinetic analysis. Blood samples for PK analysis of acumapimod were collected pre-acumapimod dose and at 1, 2, 4, 6, 12, 24, 36, 48, 72, 96, 144, 168, and 192 hours postdose. In period 1, subjects made 4 outpatient visits from day 4 to day 8 for PK and safety assessment, and received a follow-up telephone call 14 days after the last acumapimod dose in period 2. AEs were captured from check-in to the study unit on day –1 until the post-study follow-up call. Plasma analysis techniques were as for study MBCT102.

Safety and tolerability. The same methods were used as for study MBCT102.

Statistical analyses. The same methods were used as for study MBCT102, except that itraconazole was co-administered with acumapimod instead of azithromycin. Version 6.4 of Phoenix WinNonLin was used.

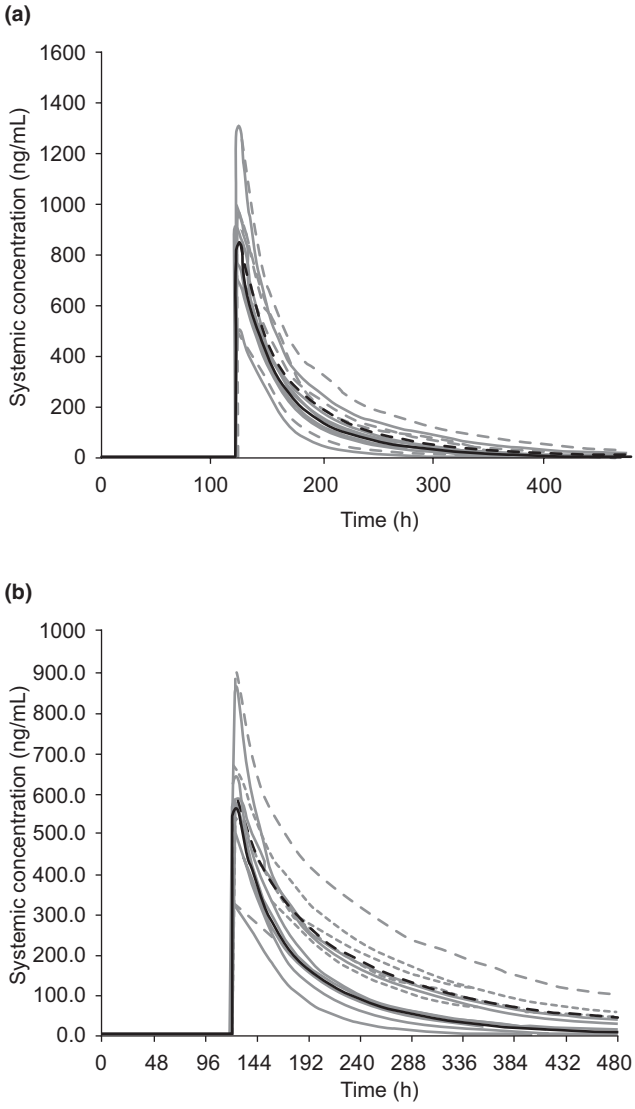

Figure 3 Simulated plasma concentration–time profiles of acumapimod. After a single oral dose (75 mg) in the presence (dashed line) and absence (solid line) of multiple oral doses of itraconazole oral solution (200 mg q.d. for 20 days), using model 1 (**a**), and model 2 (**b**). The grey lines represent the outcomes of simulated individual trials (10 × 10 virtual individuals) and the solid/dashed black line is the mean data for the simulated population (n = 100).

RESULTS

Study MBCT102

Subject characteristics. Thirty-six subjects were screened in total; of these, 21 subjects were enrolled and 16 were dosed. Overall, 14 subjects (87.5%) completed the study (who received acumapimod alone and acumapimod with azithromycin) and were analyzed in the PK set. Baseline demographics are summarized in **Table 1**. Participants were white men, with a mean age of 37.3 years (range 29.3–45.3 years).

Primary end point. The adjusted GMRs (90% CI) for C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for acumapimod co-administered with azithromycin (period 2) relative to acumapimod alone (period 1) were 106.34% (98.68–114.60), 105.65% (100.05–111.56), and 104.32% (99.24–109.65), respectively (**Table 2**). Thus, the geometric mean for acumapimod exposure ($AUC_{0-\infty}$) increased by only 4.3% and C_{max} by 6.3% when acumapimod was co-administered with azithromycin vs. acumapimod administered alone. The upper limits of the interactions were 115% (C_{max}), 112% (AUC_{0-t}), and 110% ($AUC_{0-\infty}$); therefore, co-administration of azithromycin was not considered a risk for subjects receiving acumapimod.

Safety and tolerability. Safety analysis was performed on the 16 subjects who received ≥ 1 dose of acumapimod, and for whom a safety assessment was available. Fifteen TEAEs were reported in 8 subjects (50%); 7 in subjects receiving acumapimod alone (period 1), and 8 in those receiving acumapimod with azithromycin (period 2). All were mild or moderate in intensity. One TEAE was considered by the investigator to be likely related to acumapimod (acneiform rash). This occurred on the first exposure (acumapimod alone), and the second dosing (acumapimod with azithromycin) was not progressed in this subject. No SAEs or deaths were reported. No clinically relevant findings were observed for safety laboratory, vital signs, electrocardiogram, or physical examination parameters.

Physiologically-based pharmacokinetic modeling results: azithromycin. Predicted GMRs (95% CI) for simulations using model 1 were 1.01 (1.01–1.01) for $AUC_{0-\infty}$ and 1.00 (1.0–1.0) for C_{max}, and using model 2 were 1.01 (1.01–1.02) for $AUC_{0-\infty}$ and 1.0 (1.0–1.0) for C_{max} (**Table 3**, **Tables S6** and **S7**). Predictions were consistent with the observed GMRs (90% CI) for $AUC_{0-\infty}$ (1.04 (0.99–1.10)) and C_{max} (1.06 (0.99–1.15)), and were fully contained within the 80–125% confidence limit. Therefore, the geometric mean predictions signified a negligible DDI effect on the kinetics of a single oral dose of acumapimod (14 mg) following azithromycin treatment, as observed in the clinical study MBCT102.

Study MBCT103

Subject characteristics. Forty-one subjects were screened in total; of whom 22 were enrolled; 16 were dosed with acumapimod alone during treatment period 1. Overall, 15 subjects completed treatment period 1 following the withdrawal of 1 subject during this period. These 15 subjects were dosed with itraconazole and acumapimod during treatment period 2. One subject discontinued the study during this treatment period and a total of 14 subjects completed both treatment periods and were analyzed in the PK set. Baseline demographics are summarized in **Table 1**. Participants were Hispanic or Latino men, with a mean age of 40.6 years (range 28.1–53.1 years).

Primary end points. The adjusted GMRs (90% CI) for C_{max}, AUC_{0-t}, and $AUC_{0-\infty}$ for acumapimod co-administered with itraconazole (period 2) vs. acumapimod alone (period 1) were 99.44% (91.05–108.59), 110.09% (101.04–119.94), and 110.22% (101.13–120.12), respectively (**Table 2**). Thus, the

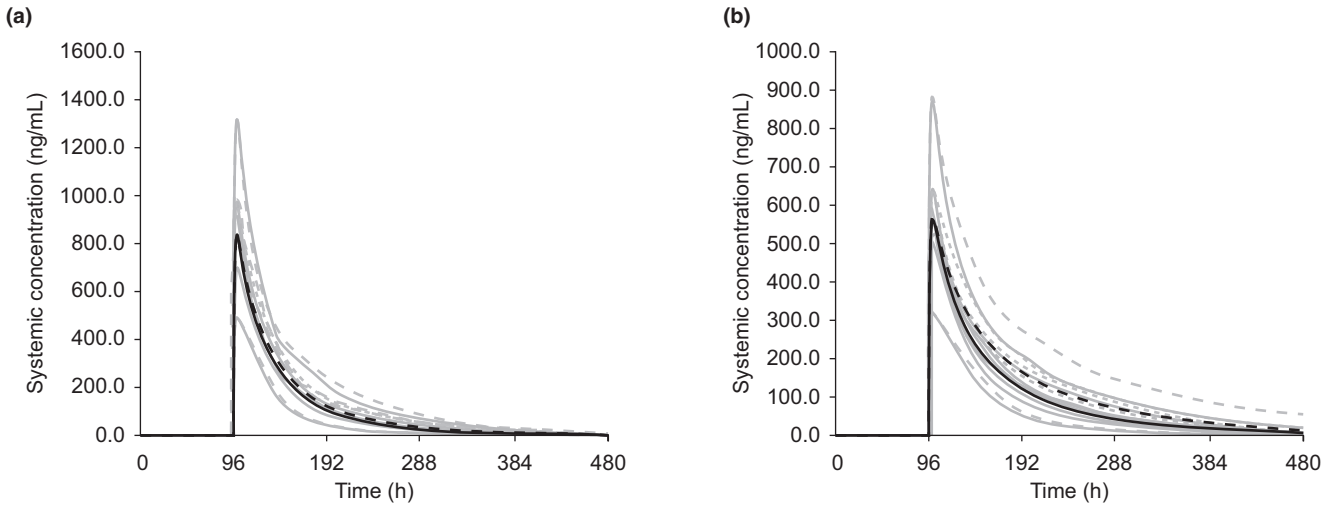

Figure 4 Simulated plasma concentration–time profiles of acumapimod. After a single oral dose (75 mg) in the presence (dashed line) and absence (solid line) of multiple oral doses of verapamil (240 mg q.d. for 20 days) (**a**) or itraconazole oral solution (200 mg q.d. for 20 days) (**b**). Model 1 was used. The grey lines represent the outcomes of simulated individual trials (10 × 10 virtual individuals), and the solid/dashed black line is the mean data for the simulated population (n = 100). It is not possible to see the early phase of the profile (between 0 and 96 hours) due to the scale of the x-axis in the figure.

geometric mean for acumapimod exposure (both AUC_{0-t} and $AUC_{0-\infty}$) increased by only 10% with no change in C_{max} when acumapimod was co-administered with itraconazole relative to when acumapimod was administered alone. The upper limits of interactions were 109% (C_{max}) and 120% (AUC_{0-t} and $AUC_{0-\infty}$), therefore, itraconazole co-administration was not considered a risk for acumapimod-treated subjects.

Safety and tolerability. Safety analyses was performed on all 16 subjects who received ≥ 1 dose of acumapimod. Overall, 10 TEAEs were reported by 6 subjects (37.5%). Two TEAEs were observed in subjects receiving acumapimod alone (period 1) and 8 in those receiving acumapimod and itraconazole (period 2). No TEAEs were judged to be treatment related. No deaths, SAEs, or severe TEAEs were reported, and only 1 subject discontinued from the study due to an AE (TEAE (blood pressure increased) in period 2 prior to acumapimod administration). No safety concerns were raised from vital sign measurements, electrocardiogram results, or laboratory parameters.

Physiologically-based pharmacokinetic modeling results: itraconazole. Application of the PBPK model to predict the impact of co-administration of itraconazole on the PKs of acumapimod showed weak-to-moderate DDI effects.[9] Simulations of the predicted GMRs (95% CI) for $AUC_{0-\infty}$ and C_{max} of 1.31 (1.29–1.33) and 1.03 (1.02–1.03), respectively, for model 1, and 1.69 (1.65–1.73) and 1.04 (1.03–1.04), respectively, for model 2 are presented in **Figure 3**, **Tables S8** and **S9**. The reported GMRs (90% CI) for $AUC_{0-\infty}$ and C_{max} from study MBCT103 were 1.10 (1.01–1.20) and 0.99 (0.91–1.08). The PBPK model was consistent with the observed C_{max} ratio being close to 1 and falling within the equivalence range of 80–125% confidence limits, indicating no clinically significant DDI effects based on FDA guidelines[15] (**Table 3**). However, the

Table 1 Baseline demographics (safety set and pharmacokinetic set)

	Azithromycin study		Itraconazole study	
	SS (n = 16)	**PKS (n = 14)**	**SS (n = 16)**	**PKS (n = 14)**
Mean age, year (range)	37.3 (29.3–45.3)	38.4 (30.5–46.3)	40.6 (28.1–53.1)	39.6 (27.0–52.2)
Sex				
Male, *n* (%)	16 (100.0)	14 (100.0)	16 (100)	14 (100)
Mean BMI, kg/m^2 (range)	26.19 (24.53–27.85)	26.53 (25.07–27.99)	26.83 (25.04–28.62)	26.80 (25.20–28.40)
Ethnicity, *n* (%)				
Unknown	16 (100)	14 (100)	0 (0)	0 (0)
Hispanic or Latino	0 (0)	0 (0)	16 (100)	14 (100)
Race, *n* (%)				
White	16 (100)	14 (100)	14 (87.5)	13 (92.9)
Black	0 (0)	0 (0)	2 (12.5)	1 (7.1)

BMI, body mass index; PKS, pharmacokinetic set; SS, safety set.

Table 2 Comparison of geometric mean plasma acumapimod pharmacokinetic parameters between treatment period 1 and period 2 – PK population

	MBCT102			MBCT103		
PK parameter	**Acumapimod alone (%CV)**	**Acumapimod + azithromycin (%CV)**	**Ratio (%)[a]**	**Acumapimod alone (%CV)**	**Acumapimod + itraconazole (%CV)**	**Ratio (%)[a]**
N	14	14		14	14	
C_{max} (ng/mL)	116 (16.2)	123 (16.9)	106.34	111 (13.22)	110 (26.48)	99.44
AUC_{0-t} (ng/mL.h)	5,733 (15.3)	6,057 (20.4)	105.65	6,029 (12.86)	6,637 (28.98)	110.09
$AUC_{0-\infty}$ (ng/mL.h)	5,954 (16.5)	6,211 (19.9)	104.32	6,254 (12.90)	6,894 (30.36)	110.22

%CV, coefficient of variation; AUC_{0-t}, area under the plasma concentration–time curve from drug administration to last observed concentration at time t; $AUC_{0-\infty}$, area under the plasma concentration–time curve extrapolated to infinite time; C_{max}, peak plasma concentration; PK, pharmacokinetic.
[a]Calculated using least-square means according to the formula: $\exp^{(DIFFERENCE)} * 100$.

CI for the predicted geometric mean $AUC_{0-\infty}$ values fell outside of the 80–125% confidence limit.

Physiologically-based pharmacokinetic modeling results: verapamil. The application of the verified PBPK model to predict the impact of the moderate CYP3A4 inhibitor verapamil[16] (240 mg q.d. for 20 days) on the PKs of acumapimod (75 mg on day 5) in healthy volunteers showed weak DDI effects. From model 1, the predicted acumapimod $AUC_{0-\infty}$ and C_{max} ratios were 1.13 (95% CI 1.11–1.16) and 1.01 (95% CI 1.01–1.01), respectively. Based on model 2, the predicted acumapimod $AUC_{0-\infty}$ and C_{max} ratios were 1.27 (95% CI 1.23–1.32) and 1.02 (95% CI 1.02–1.02), respectively (**Table 3**, **Tables S10** and **S11**).

DISCUSSION

Acumapimod is being developed as a short treatment regimen, 3 doses given over 5 days during AECOPD, to better control the inflammatory response and thereby reduce the risk of recurrent exacerbations that cluster after the index event.[19] The initial acumapimod therapeutic dose (75 mg) is at the maximum limit of safety exposure margins, leading to clinical concerns about potential toxicity and presenting 2 challenges to its development. First, CYP3A4 inducers were prohibited concomitant medications in acumapimod clinical trials, precluding the recruitment of many AECOPD subjects from trials. Second, without evidence to assess the potential extent of elevation in acumapimod plasma exposure with CYP3A4 inhibitors, it was challenging to select an acumapimod dose for clinical DDI studies owing to concern that increased plasma exposure may result in toxicity.

Azithromycin, a commonly used treatment for AECOPD, was chosen for the initial DDI study (MBCT102).[20] Because the potential extent of elevation of acumapimod exposure upon co-administration with CYP3A4 inhibitors was unknown, a low dose of acumapimod (14 mg, once daily) was chosen to minimize the risk of exceeding toxicity limits. The results of this first DDI study enabled subjects taking azithromycin to be enrolled in the phase IIa study (MBCT206).

Study MBCT102 demonstrated that co-administration of azithromycin with acumapimod had no clinically meaningful impact on acumapimod PK based on FDA guidelines for clinical drug interaction studies.[15] Results from study MBCT102 were also consistent with PBPK model results,

Table 3 Geometric mean model predicted and observed exposure of acumapimod in the presence and absence of a CYP3A4 inhibitor (PK population)

	Control		+Inhibitor		Ratio[a]	
	C_{max}, ng/mL	**$AUC_{0-\infty}$, ng/mL.h**	**C_{max}, ng/mL**	**$AUC_{0-\infty}$, ng/mL.h**	**C_{max}[b]**	**$AUC_{0-\infty}$[b]**
Azithromycin						
Model 1	149.2	6,957.3	149.3	7,015.1	1.00 (95% CI 1.0–1.0)	1.01 (95% CI 1.01–1.01)
Model 2	99.6	6,491.1	99.7	6,585.1	1.00 (95% CI 1.0–1.0)	1.01 (95% CI 1.01–1.02)
Observed	116.0	5,954.0	123.0	6,211.0	1.06 (90% CI 0.99–1.15)	1.04 (90% CI 0.99–1.10)
Itraconazole						
Model 1	810.9	37,820.7	832.0	49,691.6	1.03 (95% CI 1.02–1.03)	1.31 (95% CI 1.29–1.33)
Model 2	539.2	34,639.0	559.3	58,561.0	1.04 (95% CI 1.03–1.04)	1.69 (95% CI 1.65–1.73)
Observed	111.12	6,254.38	110.49	6,893.51	0.99 (90% CI 0.91–1.08)	1.10 (90% CI 1.01–1.20)
Verapamil						
Model 1	810.1	37,441.7	820.2	42,479.4	1.01 (95% CI 1.01–1.01)	1.13 (95% CI 1.11–1.16)
Model 2	539.2	34,797.4	548.7	44,297.5	1.02 (95% CI 1.02–1.02)	1.27 (95% CI 1.23–1.32)

AUC_{0-t}, area under the plasma concentration–time curve from drug administration to last observed concentration at time t; $AUC_{0-\infty}$, area under the plasma concentration–time curve extrapolated to infinite time; CI, confidence interval; C_{max}, peak plasma concentration; PK, pharmacokinetic.
[a]Calculated using least-square means according to the formula: $\exp^{(DIFFERENCE)} * 100$.
[b]90% geometric CI calculated according to the formula: $\exp^{(DIFFERENCE \pm t_{(dfResidual)} * SE_{DIFFERENCE})} * 100$.

which predicted negligible DDI effects when azithromycin and acumapimod were co-administered.

As it remained necessary to determine potential interaction(s) with moderate and strong inhibitors, a PBPK modeling strategy (based on all available acumapimod clinical data, including data from study MBCT102) was used to assess the impact of co-administration of a moderate (verapamil, 240 mg q.d.) and a strong (itraconazole, 200 mg q.d.) CYP3A4/P-gp inhibitor on the PKs of acumapimod, following a single oral dose of acumapimod (75 mg).[9,16] The model predicted weak-to-moderate DDI effects and provided confidence that a second DDI study (MBCT103) could be progressed with acumapimod and the strong CYP3A4 inhibitor itraconazole, without exceeding safety exposure margins.[9] Accuracy of the PBPK model predictions was confirmed by the MBCT103 study results, which indicated that itraconazole had no meaningful effect on the acumapimod PKs. The absence of clinically meaningful sensitivity to strong inhibitors is not unprecedented and is consistent with enzyme kinetic principles of inhibition DDIs. For example, co-administration of steady-state ketoconazole (400 mg/day) with the investigational anticancer agent tivozanib did not alter tivozanib AUC, with 90% CIs for the GMRs contained within the 80–125% equivalence range.[21,22]

It is noteworthy that the CI from the PBPK model for the predicted geometric mean $AUC_{0-\infty}$ values fell outside of the 80–125% confidence limit. The reported GMRs for study MBCT103 were lower than the ratios predicted by the models, particularly for $AUC_{0-\infty}$. Overall, there was no clinically meaningful difference in safety profiles when acumapimod was co-administered with either of the CYP3A4 inhibitors azithromycin or itraconazole, in studies MBCT102 and MBCT103. This is particularly significant given that subjects with AECOPD who take acumapimod may also be taking CYP3A4 inhibitors for the treatment of exacerbations and commonly associated comorbidities.[5,6] The studies also highlight the value of PBPK modeling in the efficient progression of investigational products during drug development, and demonstrate the potential for PBPK models to inform decision making for DDI assessment.

A limitation of the model was that it did not evaluate the potential impact of itraconazole inhibition of P-gp on acumapimod concentrations in the brain and liver. Any potential clinically meaningful impact of itraconazole inhibition of P-gp on acumapimod concentrations in the brain and liver would have manifested as AEs in the clinical DDI study (MBCT103). There were no clinically meaningful changes in liver function tests or central nervous system AEs following co-administration of acumapimod and itraconazole in the clinical DDI study, suggesting that there no clinically relevant DDI effects in the brain or liver.

Supporting Information. Supplementary information accompanies this paper on the *CPT: Pharmacometrics & Systems Pharmacology* website (www.psp-journal.com).

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1. Wedzicha, J., MacKinnon, A. & Parkin, J. Effectiveness of acumapimod oral p38 inhibitor in the treatment of acute severe exacerbations of COPD: results of the AETHER phase II trial. *Am. J. Resp. Crit. Care Med.* **197**, A7710 (2018).
2. Gross, N. The COPD pipeline XXXVI. *Chronic Obstr. Pulm. Dis.* **4**, 320–324 (2017).
3. Lee, J. *et al.* p38 MAPK activity is associated with the histological degree of interstitial fibrosis in IgA nephropathy patients. *PLoS One* **14**, e0213981 (2019).
4. MacNee, W., Allan, R.J., Jones, I., De Salvo, M.C. & Tan, L.F. Efficacy and safety of the oral p38 inhibitor PH-797804 in chronic obstructive pulmonary disease: a randomised clinical trial. *Thorax* **68**, 738–745 (2013).
5. Westphal, J.F. Macrolide - induced clinically relevant drug interactions with cytochrome P-450A (CYP) 3A4: an update focused on clarithromycin, azithromycin and dirithromycin. *Br. J. Clin. Pharmacol.* **50**, 285–295 (2000).
6. National Institute for Health and Care Excellence. Chronic obstructive pulmonary disease (acute exacerbation): antimicrobial prescribing. NICE Guideline (NG114) <https://www.nice.org.uk/guidance/ng114>.
7. ClinicalTrials.gov. Efficacy and safety of BCT197 in subjects with acute respiratory exacerbations of chronic obstructive pulmonary disease <https://clinicaltrials.gov/ct2/show/NCT02700919>.
8. Fleet, J.L. *et al.* Comparing two types of macrolide antibiotics for the purpose of assessing population-based drug interactions. *BMJ Open* **3**, e002857 (2013).
9. Vermeer, L.M.M., Isringhausen, C.D., Ogilvie, B.W. & Buckley, D.B. Evaluation of ketoconazole and its alternative clinical CYP3A4/5 inhibitors as inhibitors of drug transporters: the in vitro effects of ketoconazole, ritonavir, clarithromycin, and itraconazole on 13 clinically-relevant drug transporters. *Drug Metab. Dispos.* **44**, 453–459 (2016).
10. Gupta, N. *et al.* Effects of strong CYP3A inhibition and induction on the pharmacokinetics of ixazomib, an oral proteasome inhibitor: results of drug-drug interaction studies in patients with advanced solid tumors or lymphoma and a physiologically based pharmacokinetic analysis. *J. Clin. Pharmacol.* **58**, 180–192 (2018).
11. Reddy, V.P., Walker, M., Sharma, P., Ballard, P. & Vishwanathan, K. Development, verification, and prediction of osimertinib drug-drug interactions using PBPK modeling approach to inform drug label. *CPT Pharmacometrics Syst. Pharmacol.* **7**, 321–330 (2018).
12. Lüpfert, C. *et al.* A novel PBPK modeling approach to assess cytochrome P450 mediated drug-drug interaction potential of the cytotoxic prodrug evofosfamide. *CPT Pharmacometrics Syst. Pharmacol.* **7**, 829–837 (2018).
13. Hanke, N. *et al.* PBPK models for CYP3A4 and P-gp DDI prediction: a modeling network of rifampicin, itraconazole, clarithromycin, midazolam, alfentanil, and digoxin. *CPT Pharmacometrics Syst. Pharmacol.* **7**, 647–659 (2018).
14. ClinicalTrials.gov. The effect of azithromycin on BCT197 exposure in healthy male volunteers <https://clinicaltrials.gov/ct2/show/NCT02926326>.
15. US Food and Drug Administration Center for Drug Evaluation and Research (CDER). Clinical drug interaction studies—cytochrome P450 enzyme- and transporter-mediated drug interactions. Guidance for industry <https://www.fda.gov/media/134581/download> (2020).
16. Martin, P. *et al.* Effects of CYP3A4 inhibitors ketoconazole and verapamil and the CYP3A4 inducer rifampicin on the pharmacokinetic parameters of fostamatinib: results from in vitro and Phase I clinical studies. *Drugs R D* **16**, 81–92 (2016).

17. Zhao, P., Rowland, M. & Huang, S.-M. Best practice in the use of physiologically based pharmacokinetic modeling and simulation to address clinical pharmacology regulatory questions. *Clin. Pharmacol. Ther.* **92**, 17–20 (2012).
18. ClinicalTrials.gov. The effect of itraconazole on BCT197 exposure in healthy male participants <https://clinicaltrials.gov/ct2/show/NCT03498170>.
19. Hurst, J.R. *et al*. Temporal clustering of exacerbations in chronic obstructive pulmonary disease. *Am. J. Respir. Crit. Care Med.* **179**, 369–374 (2009).
20. Cazzola, M. *et al*. Treatment of acute exacerbation of severe-to-very severe COPD with azithromycin in patients vaccinated against Streptococcus pneumoniae. *Respir. Med.* **99**, 663–669 (2005).
21. Cotreau, M., Siebers, N., Miller, J., Strahs, A. & Slichenmyer, W. Effects of ketoconazole or rifampin on the pharmacokinetics of tivozanib hydrochloride, a vascular endothelial growth factor receptor tyrosine kinase inhibitor. *Clin. Pharmacol. Drug Dev.* **4**, 137–142 (2015).
22. Venkatakrishnan, K. DDI Risk Assessment and Evaluation in Clinical Development: Interfacing Drug Metabolism and Clinical Pharmacology. In Encyclopedia of Drug Metabolisms and Interactions. (Lyubimov, A.) (John Wiley & Sons, Hoboken, NJ, 2012).