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ORIGINAL ARTICLE

Effect of multiple-dose osimertinib on the pharmacokinetics of simvastatin and rosuvastatin

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Principal Investigator information: The International Co-ordinating Investigators of the CYP3A study and the BCRP study were Professor Suresh S. Ramalingam and Dr Nicolas Isambert, respectively. Professor Ramalingam invited Dr Donald Harvey to take his place as an author on the manuscript.

Keywords CYP3A, BCRP, NSCLC, osimertinib

AIM

We report on two Phase 1, open-label, single-arm studies assessing the effect of osimertinib on simvastatin (CYP3A substrate) and rosuvastatin (breast cancer resistance protein substrate [BCRP] substrate) exposure in patients with advanced epidermal growth factor receptor (EGFR)-mutated non-small cell lung cancer who have progressed after treatment with an EGFR tyrosine kinase inhibitor, to determine, upon coadministration, whether osimertinib could affect the exposure of these agents.

METHODS

Fifty-two patients in the CYP3A study (pharmacokinetic [PK] analysis, $n = 49$), and 44 patients in the BCRP study were dosed (PK analysis, $n = 44$). In the CYP3A study, patients received single doses of simvastatin 40 mg on Days 1 and 31, and osimertinib 80 mg once daily on Days 3–32. In the BCRP study, single doses of rosuvastatin 20 mg were given on Days 1 and 32, and osimertinib 80 mg once daily on Days 4–34.

RESULTS

Geometric least squares mean (GLSM) ratios (90% confidence intervals) of simvastatin plus osimertinib for area under the plasma concentration–time curves from zero to infinity (AUC) were 91% (77–108): entirely contained within the predefined no relevant effect limits, and C_{max} of 77% (63, 94) which was not contained within the limits. GLSM ratios of rosuvastatin plus osimertinib for AUC were 135% (115–157) and C_{max} were 172 (146, 203): outside the no relevant effect limits.

CONCLUSIONS

Osimertinib is unlikely to have any clinically relevant interaction with CYP3A substrates and has a weak inhibitory effect on BCRP. No new safety concerns were identified in either study.

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WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT

- Osimertinib is a potent, oral, central nervous system-active, irreversible EGFR-TKI selective for both EGFR-TKI sensitizing (EGFRm) and T790M resistance mutations.
- *In vitro* studies show that osimertinib can inhibit or induce CYP3A4/5 enzymes, and inhibit breast cancer resistance protein (BCRP) transporter.

WHAT THIS STUDY ADDS

- Osimertinib is unlikely to have any clinically relevant interaction with CYP3A substrates and has a weak inhibitory effect on BCRP substrates.

Introduction

Epidermal growth factor receptor (EGFR)-tyrosine kinase inhibitors (TKIs) are the standard first-line treatment for non-small cell lung cancer (NSCLC) patients with TKI-sensitizing mutations in EGFR (EGFRm) [1–3]. However, the majority of patients who initially respond to EGFR-TKIs ultimately develop resistance, with over 50% of tumours harbouring the EGFR T790M resistance mutation [4–10]. **Osimertinib** is a potent, oral, central nervous system active, irreversible EGFR-TKI selective for EGFRm and T790M resistance mutations [11–13]. Osimertinib is approved and also recommended for the treatment of patients with metastatic EGFR T790M-positive advanced NSCLC [1, 3]. In the Phase 3 AURA3 trial, osimertinib provided a higher objective response rate (71% *vs*. 31%) and significantly longer progression-free survival than platinum-based doublet chemotherapy (median 10.1 *vs*. 4.4 months; hazard ratio [HR] 0.30; 95% confidence interval [CI] 0.23, 0.41; $P < 0.001$) [14].

As part of treatment with osimertinib, it is important to understand potential drug–drug interactions (DDI) due to the risk of comorbidities requiring concomitant therapy in this patient population. *In vitro* studies have shown that osimertinib has potential to be a competitive inhibitor and inducer of **CYP3A** and that it is a competitive inhibitor of the **breast cancer resistance protein (BCRP)** transporter [15]. CYP3A is the most important enzyme involved in the metabolism of drugs [16], while BCRP is involved in the elimination of certain widely prescribed medicines with relatively narrow therapeutic margins, including rosuvastatin at the higher dose [17, 18]. Comorbidities commonly associated with NSCLC, such as chronic obstructive pulmonary disease or diabetes [19], may need to be treated with concomitant medications that are metabolized through CYP3A or transport-mediated elimination via BCRP. Moreover, statins are a common co-medication in this patient population. Therefore, it is important to understand any potential implications osimertinib could have on the exposure and, thereby, the efficacy and safety of these agents when co-administered.

We report two clinical studies designed to investigate the impact of multiple doses of osimertinib on the pharmacokinetics (PK) of **simvastatin** and simvastatin acid (a sensitive CYP3A substrate and its metabolite; [NCT02197234]), and **rosuvastatin** (a substrate for BCRP and a medication likely to be administered concomitantly with osimertinib; [NCT02317016]). The two active metabolites of osimertinib (AZ5104 and AZ7550), which are also substrates of BCRP and formed via CYP3A4, and represent approximately 10% each of osimertinib exposure [20–22], were also monitored, though were not considered likely to contribute to any DDI. 4β-hydroxy-cholesterol (4BHC) concentration ratios were measured in order to understand the overall effect of CYP3A modulation following multiple-dose administration of osimertinib. Both studies were conducted in patients with advanced EGFRm NSCLC after disease progression during or after a prior EGFR-TKI. Herein, we report results that show the PK-mediated potential for DDI between these agents.

Methods

Details of *in vitro* CYP inhibition, transporter inhibition and CYP induction potential of osimertinib are provided in the Supplementary information.

Clinical trial design

Both studies were Phase 1, open-label, single-arm studies in patients with EGFRm NSCLC with disease progression during or after treatment with an EGFR-TKI. They were conducted in accordance with International Conference on Harmonization–Good Clinical Practice guidance, and protocols were reviewed and approved by an Independent Ethics Committee and Institutional Review Board prior to implementation. Written informed consent was obtained from all participants.

Each study consisted of two parts. Part A was designed to assess the effect of osimertinib on simvastatin and simvastatin acid (CYP3A study) or rosuvastatin (BCRP study) exposure and was split into three segments: Periods 1–3. Part B allowed patients to have continued access to osimertinib after the PK phase (Part A) and provided additional safety data. Only Part A results are described in this report.

In the CYP3A and BCRP studies, patients received a single oral dose of simvastatin 40 mg or rosuvastatin 20 mg, respectively, alone on Day 1 (Period 1) and remained in the clinic for approximately 32 to 34 h, during which time blood samples for PK analysis and safety information were collected. Patients then received osimertinib 80 mg orally once daily for 28 days (Period 2, Days 3–30 in the CYP3A study, and Days 4–31 in the BCRP study) and returned to the clinic at weekly intervals for collection of osimertinib and metabolite (AZ5104 and AZ7550) trough levels. In Period 3 on Day 31 of the CYP3A study and Day 32 of the BCRP study, patients received a single oral dose of simvastatin 40 mg, or rosuvastatin 20 mg, in combination with osimertinib 80 mg. In the CYP3A study, this was followed by a final oral dose of osimertinib 80 mg on Day 32, whereas in the BCRP study this dosing

was followed by subsequent daily doses of osimertinib 80 mg on Days 33 and 34. Patients remained in the clinic for approximately 32 to 34 h, during which time blood samples for PK analysis and safety information were collected.

In both studies, patients fasted from at least 2 h before dosing to at least 2 h after dosing on simvastatin and rosuvastatin dosing days. Osimertinib was to be given with 1 h of fasting before to 2 h after dosing.

Data underlying the findings described in this paper may be obtained in accordance with AstraZeneca's data sharing policy described at https://astrazenecagrouptrials.pharmacm.com/ST/Submission/Disclosure.

Participants

Adult patients with a histological or cytological confirmed diagnosis of EGFRm NSCLC, and radiological confirmation of disease progression during previous continuous treatment with an EGFR-TKI, were enrolled. Inclusion criteria included local confirmation that tumours harboured an EGFR mutation known to be associated with EGFR-TKI sensitivity, an Eastern Cooperative Oncology Group performance status 0–1 with no deterioration over the previous 2 weeks, and a life expectancy of ≥12 weeks as estimated at the time of screening.

Exclusion criteria included inadequate bone marrow reserve or organ function and unresolved toxicities from any prior therapy exceeding CTCAE Grade 1. In both studies, patients were required to avoid any food/drugs with known CYP3A inducer/inhibitor effects; if patients were taking CYP3A inhibitors/inducers, a sufficient wash out was required before enrolment. Based on the prescribing information of simvastatin and rosuvastatin, patients treated with concomitant medications likely to cause PK interaction, or another statin, were excluded. The BCRP study was limited to patients of non-Asian ethnicity to avoid BCRP polymorphism [17, 23]. Intake of Seville oranges or grapefruits was prohibited in both studies as these act as potent inhibitors of CYP3A [24].

Objectives

The primary objective of both studies was to assess the exposure (AUC and C_{max}) of simvastatin or rosuvastatin when administered as a single dose alone and in combination with osimertinib. Secondary objectives were to assess the PK of simvastatin (and simvastatin acid) and rosuvastatin, respectively, when administered as a single dose alone and in combination with osimertinib, and to assess the PK of osimertinib (and metabolites) when administered in combination with simvastatin and rosuvastatin, respectively. Safety and tolerability of osimertinib alone and in combination with simvastatin and rosuvastatin, respectively, were also evaluated. The potential for osimertinib to induce CYP3A through changes in post-dose to pre-dose ratios for 4BHC concentration was assessed as an exploratory objective.

Statistical methods

The PK analysis set was defined as dosed patients with at least one quantifiable plasma concentration collected post-dose without any important deviations or events that could alter the evaluation of the PK. Important deviations or events included dosing deviations, vomiting following oral dosing, and administration of or changes in concomitant medications thought to affect simvastatin or rosuvastatin PK. With respect to osimertinib, any deviations or events resulting in osimertinib AUC_{τ} (AUC during the dosing interval) falling below the 10th percentile of exposure of the overall patient population resulted in exclusion of the patients' simvastatin or rosuvastatin PK data from the analyses.

To evaluate the effect of osimertinib on simvastatin, simvastatin acid or rosuvastatin exposure, natural log-transformed AUC (and AUC from zero to the last quantifiable concentration at time t [AUC_{0-t}]) and C_{max}, were compared between treatments using a mixed-effects analysis of variance, with treatment as a fixed effect and patient as a random effect. The mean differences and the CIs were back transformed to the original scale in order to give estimates of the geometric mean ratios ([osimertinib + simvastatin/rosuvastatin] *vs*. simvastatin/rosuvastatin alone) and the associated 90% CIs. No effect on the PK of simvastatin/rosuvastatin after co-administration of osimertinib was concluded if the two-sided 90% CIs for the ratios of simvastatin/rosuvastatin AUC (or AUC_{0-t}) and C_{max} were within the range of 70% to 143%. For simvastatin/rosuvastatin and simvastatin acid, analyses of time to maximum concentration (t_{max}) were performed using the Wilcoxon signed rank test. The Hodges–Lehman median estimator of the difference in treatments ([osimertinib + simvastatin/rosuvastatin] – simvastatin/rosuvastatin alone) and 90% CIs are presented.

A sufficient number of patients were enrolled to address the primary PK study objectives, as measured by AUC and C_{max}. The studies were powered based on a within-subject coefficient of variation of 45% for simvastatin and 41% for rosuvastatin, assuming an increase of approximately 20% in the coefficient of variation observed in healthy subjects. No change in exposure for simvastatin and rosuvastatin when given with osimertinib was assumed. It was estimated that 40 and 34 patients would be needed to ensure evaluation for PK analysis in the CYP3A and BCRP studies, respectively. These sample sizes were expected to provide 90% power for the 90% CIs for both AUC and C_{max} ratios to be within 70% to 143%. The relevant no-effect boundary was determined based on the high variability of simvastatin and rosuvastatin. Also, with the exposure response of simvastatin and rosuvastatin, a change of 0.7 to 1.43-fold is unlikely to alter its benefit risk, and hence this margin was used [25].

The safety analysis set included all patients who received at least one dose of osimertinib or either statin. Safety assessments in both studies included adverse event (AE) reporting graded by CTCAE v4.0, physical examination, vital signs, electrocardiogram, ophthalmic examination, clinical chemistry, coagulation, haematology and urinalysis. For additional information, see the Supporting Information Appendix S1.

Bioanalysis

Samples for the determination of simvastatin, simvastatin acid, rosuvastatin, 4BHC, and osimertinib and its metabolites (AZ5104 and AZ7550) in plasma were analysed by Covance Laboratories at their sites globally using validated bioanalytical methods. Simvastatin, simvastatin acid, and

4BHC were detected in plasma containing K_2EDTA using high performance liquid chromatography (HPLC) followed by tandem mass spectrometric (MS/MS) detection. Rosuvastatin was detected in plasma containing lithium heparin using supported-liquid extraction, and analysed using HPLC-MS/MS. Calibration, quality control and clinical study samples (40 µL) were spiked with (^{13}C, $^{2}H_3$) osimertinib as an internal standard, processed by protein precipitation and then simultaneously assayed for osimertinib, AZ5104 and AZ7550 using reversed-phase HPLC with Turbo Ion Spray® MS/MS. Drug-to-internal standard peak area ratios for the standards were used to create a calibration curve using $1/x^2$ weighted least-squares regression analysis. Concentrations of each analyte were quantified by comparing ratios in trial samples with the relevant calibration curve. During validation of all assays, no analytically significant interferences from endogenous matrix components were observed. All methods demonstrated acceptable selectivity with mean normalized matrix factors of 1.00 ± 0.08 observed at the concentrations tested. The lower limit of quantification of the method was 16 nM for osimertinib, 1.65 nM for AZ5104 and AZ7550, 0.04 ng ml^{-1} for rosuvastatin, 0.05 ng ml^{-1} for simvastatin and simvastatin acid and 4 ng ml^{-1} for 4BHC. Accuracy ranged from 93% to 112% and precision from 2.5% to 10.1% for all analytes in both studies.

PK parameters for plasma osimertinib, AZ5104, AZ7550, simvastatin, simvastatin acid and rosuvastatin non-compartmental methods were calculated and summarized with Phoenix® WinNonlin® Version 6.4 (Pharsight Corp., A Certara Company, Princeton, NJ, USA). PK and safety summaries, as well as the inferential analyses for simvastatin/rosuvastatin and simvastatin acid, were performed by IQVIA using SAS® Version 9.2 (SAS Institute, Inc., Cary, NC, USA).

Nomenclature of targets and ligands

Key protein targets and ligands in this article are hyperlinked to corresponding entries in the http://www.guidetopharmacology.org, the common portal for data from the IUPHAR/BPS Guide to Pharmacology [26], and are permanently archived in the Concise Guide to Pharmacology 2017/18 [27, 28].

Results

In vitro studies

In human liver microsomes, only CYP3A4/5 using nifedipine as the substrate showed inhibition at less than 25 µM (IC_{50} = 5.1 µM with nifedipine as substrate and >25 µM for midazolam as substrate). Osimertinib is not an inhibitor (IC_{50} > 30 µM) for CYP1A2, 2A6, 2B6, 2C8, 2C9, 2C19, 2D6 and 2E1 (Table S1). No time-dependent inhibition was observed for any of the enzymes.

No induction in mRNA or activity was observed for CYP2B6 and up to 16% of positive control for CYP1A2 was observed. A concentration-dependent maximal induction of up to 173-fold (89% of positive control) in one lot and 4.9-fold (45% of positive control) in the other two lots in mRNA and activity was observed for CYP3A4/5.

For transporter inhibition, the inhibition values and the potential for interaction are shown in Table S2. The results indicate that BCRP inhibition (mostly intestinal) is likely. Based on *in vitro* data, osimertinib is not likely to be a clinically relevant inhibitor of Pgp, OATP1B1, OATP1B3, OCT2, OAT1, OAT3, MATE1 and MATE2K transporters.

Patients

In the CYP3A study, 57 patients were enrolled across 17 centres in Asia, North America and Western Europe. Of these patients, 52 were assigned to and received treatment, of whom 49 were included in the PK analysis set. Of the three patients excluded from PK analyses, two were excluded as their clinical imaging showed excessive hepatic metastases which was significantly reduced after 4 weeks of treatment with osimertinib, which likely confounds the DDI results, and one was excluded due to changes in concomitant medication (a CYP3A4 inducer) dosing during the treatment period. In the BCRP study, 55 patients were enrolled from 13 centres across Western Europe and North America (no Asian patients in the BCRP study). Of these, 44 patients were assigned to and received treatment, all of whom were included in the PK analysis set. Baseline demographics, disease characteristics and allowed concomitant medications are shown in Table 1.

CYP3A study: simvastatin PK

Geometric mean plasma concentrations of simvastatin are shown in Figure 1. Geometric mean simvastatin concentrations were slightly lower following co-administration of osimertinib over the initial 4 h while the terminal concentrations appeared to exhibit a similar decline. The simvastatin acid profiles were similar to each other following administration of simvastatin alone and simvastatin with osimertinib throughout the time course. Administration of osimertinib with simvastatin decreased the area under the plasma concentration–time curve from zero to infinity (AUC) for simvastatin by approximately 9%, and the maximum plasma concentration (C_{max}) by approximately 23%, compared with administration of simvastatin alone (Table 2). Table 2 shows that exposure of simvastatin acid relative to simvastatin was similar across treatments, based on arithmetic mean metabolite-to-parent ratios (MR) for AUC and C_{max}. Individual and geometric mean AUCs of simvastatin and simvastatin acid alone, versus in combination with osimertinib, are shown in Figure S1.

The geometric least squares mean (GLSM) ratios of evaluable patients receiving simvastatin plus osimertinib to simvastatin alone for AUC and C_{max} are shown in Table 2: the 90% CI of GLSM ratio for AUC was entirely contained within the no relevant effect limits of 70% to 143%, but the reduction seen for C_{max} was not entirely contained within these limits. No effect of osimertinib on AUC or C_{max} of simvastatin acid was observed.

Osimertinib did not affect the time to maximum concentration (t_{max}) or the half-life of simvastatin or simvastatin acid (Table 2). The mean apparent plasma clearance (CL/F) was slightly higher with osimertinib and simvastatin *vs*. simvastatin alone (Table 2).

Table 1

Baseline demographics and disease characteristics (safety analysis set)

	CYP3A study (*n* = 52)	BCRP study (*n* = 44)
Age (years), median (range)	61.5 (44–83)	64.5 (36–79)
Gender, *n* (%)		
Male	15 (29)	14 (32)
Female	37 (71)	30 (68)
Race, *n* (%)		
White	35 (67)	41 (93)
Asian	17 (33)	1 (2)[a]
Other	0	2 (5)[b]
Height (cm), mean (SD)	161 (9)	165.5 (9)
Weight (kg), mean (SD)	60 (13)	66 (13)
Body mass index (kg m^{-2}), mean (SD)	23 (4)	24 (5)
ECOG performance status, *n* (%)		
0	11 (21)	20 (45)
1	41 (79)	24 (55)
Overall disease classification NSCLC (%)		
Metastatic	45 (87)	37 (84)
Locally advanced	7 (13)	7 (16)
Histology type, NSCLC *n* (%)		
Adenocarcinoma	49 (94)	44 (100)
Adenosquamous carcinoma	1 (2)	0
Large cell carcinoma	1 (2)	0
Squamous cell carcinoma	1 (2)	0
Prior EGFR-TKI, *n* (%)		
Gefitinib	29 (56)	21 (48)
Erlotinib	29 (56)	23 (52)
Afatinib	6 (12)	9 (20)
Dacomitinib	1 (2)	2 (5)
Prior platinum-chemotherapy, *n* (%)	32 (62)	23 (52)
Median number of prior treatments	2	2
Allowed concomitant medications, >10% pts, *n* (%)		
Calcium carbonate + colecalciferol	6 (12)	–
Denosumab	7 (13)	–
Ibuprofen	6 (12)	≤10% pts
Paracetamol	15 (29)	5 (11)
Prednisolone sodium sulfobenzoate	≤10% pts	5 (11)
Tramadol	≤10% pts	6 (14)

ECOG, Eastern Cooperative Oncology Group; EGFR-TKI, epidermal growth factor receptor tyrosine kinase inhibitor; NOS, not otherwise specified; SD standard deviation

[a]Patients of certain Asian ethnicities (e.g. Chinese, Filipino, Japanese, Korean and Vietnamese) were excluded from the BCRP study due to higher rosuvastatin exposures observed in these populations. Asian Indian ethnicity was acceptable

[b]Black or African American

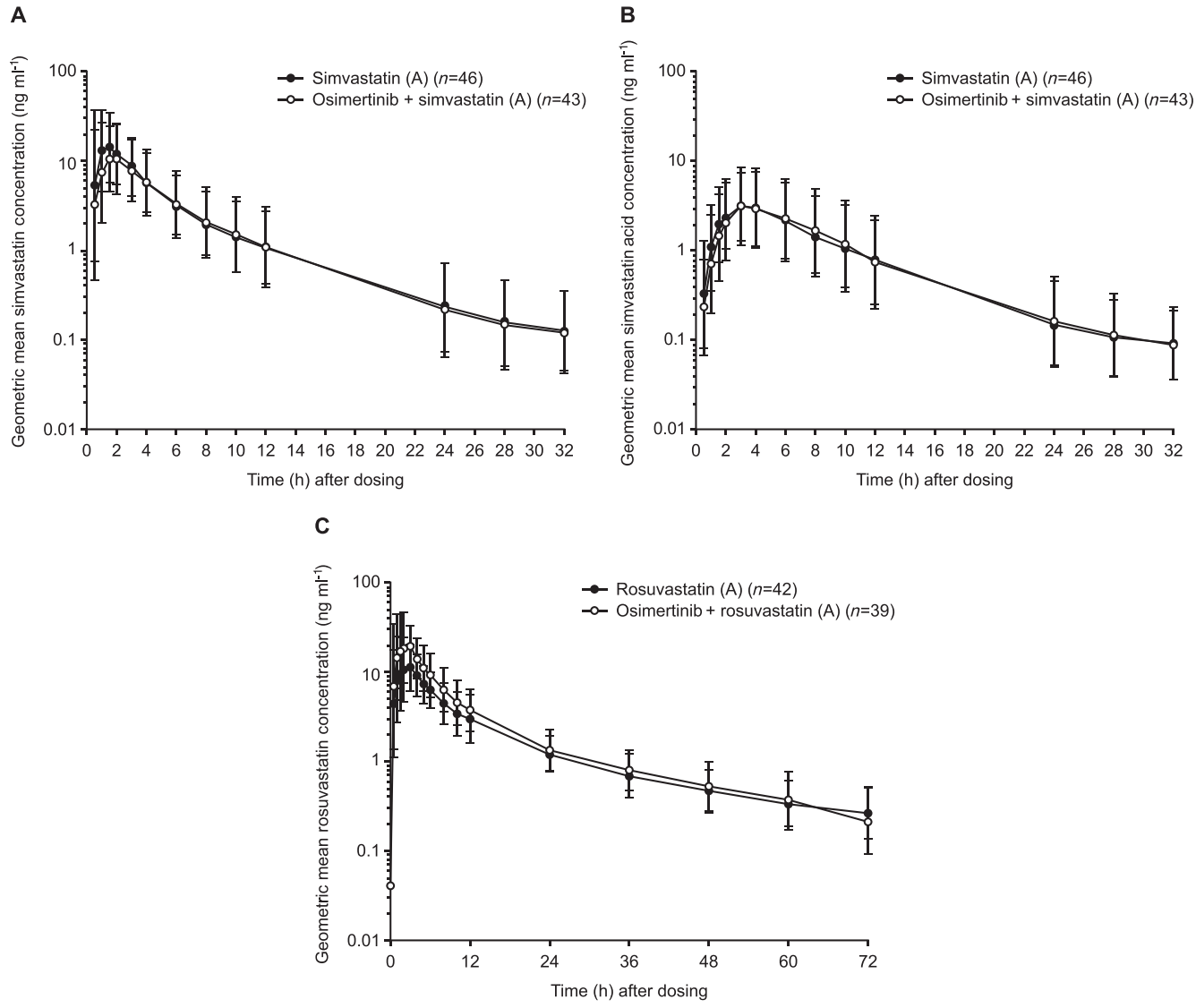

Figure 1

Geometric mean plasma concentration (ng ml^{-1}) *vs.* time by treatment [semi-log scale] (pharmacokinetic analysis set). A, simvastatin; B, simvastatin acid; C, rosuvastatin

BCRP study: rosuvastatin PK

Geometric mean rosuvastatin plasma concentration–time profiles are shown by treatment in Figure 1. AUC, AUC_{0-t} and C_{max} of rosuvastatin were higher with osimertinib and rosuvastatin *vs.* rosuvastatin alone (Table 2). With rosuvastatin, the concentrations were higher for the first 24 h, following administration of osimertinib and rosuvastatin, compared with rosuvastatin alone. After 24 h, both rosuvastatin concentrations appeared to exhibit a similar decline. Individual and geometric mean AUCs of rosuvastatin alone *vs.* in combination with osimertinib are shown in Figure S2. GLSM ratios of rosuvastatin plus osimertinib to rosuvastatin alone for AUC and C_{max} were 135% (115–157) and 172% (146–203), respectively (Table 2). The 90% CIs of the GLSM ratios for these parameters were not contained within the predefined no relevant effect range of 70% to 143%. Co-administration of osimertinib had no effect on rosuvastatin t_{max} (Table 2). The half-life of rosuvastatin was similar: 19.8 h when given with osimertinib *vs.* 19.5 h with rosuvastatin alone.

CL/F and volume of distribution (Vz/F) were both lower with rosuvastatin plus osimertinib compared with rosuvastatin alone, as shown in Table 2.

Osimertinib and metabolites PK

PK parameters for osimertinib and the metabolites AZ5104 and AZ7550 after 29 days of dosing are shown in Table 3. In both studies, visual observations indicated that steady state was attained for osimertinib and its metabolites at the time of Period 3 evaluation of PK interaction. Across the two studies, the metabolite-to-parent ratio for AUC during the dosing

Table 2

Pharmacokinetic parameters by simvastatin or rosuvastatin treatment group (pharmacokinetic analysis set)

Pharmacokinetic parameter	CYP3A4 study: *n* = 49				BCRP study: *n* = 44	
	Simvastatin		Simvastatin acid		Rosuvastatin	
Treatment	Simvastatin	Simvastatin + osimertinib	Simvastatin	Simvastatin + osimertinib	Rosuvastatin	Rosuvastatin + osimertinib
AUC (ng h^{-1} ml^{-1}), geometric mean (%GCV) *n*	80.3 (67) 46	73.5 (76) 41	31.2 (106) 42	30.2 (117) 37	139.1 (49) 31	185.7 (61) 32
Geometric LSM[a]	80.4	73.5	32.4	30.7	138.7	186.7
Ratio, % (90% CI)[a]	91.5 (77.2, 108.4)		94.7 (80.0, 112.1)		134.63 (115.4, 157.1)	
AUC_{0-t} (ng h^{-1} ml^{-1}), geometric mean (%GCV) *n*	78.0 (67) 46	70.2 (76) 43	29.3 (110) 46	29.6 (125) 43	130.6 (51) 42	183.7 (58) 37
C_{max} (ng ml^{-1}), geometric mean (%GCV) *n*	24.5 (82) 46	18.7 (75) 43	4.2 (115) 46	4.2 (126) 43	14.0 (67) 42	24.0 (71) 39
Geometric LSM[a]	24.6	19.0	4.3	4.2	13.9	23.8
Ratio, % (90% CI)[a]	77.1 (63.4, 93.7)		97.9 (81.9, 117.2)		171.9 (145.9, 202.5)	
t_{max} (h), median (min, max) *n*	1.5 (0.5, 4.0) 46	1.5 (0.5, 10.0) 43	3.1 (0.5, 10.0) 46	3.1 (1.5, 12.5) 43	2.1 (0.5, 6.0) 42	2.1 (0.5, 6.0) 39
Median difference (90% CI), *P*-value[b]	0.06 (−0.3, 0.5) 0.5501		−0.02 (−0.5, 0.4) 0.7617		−0.2 (−0.5, 0.3) 0.6033	
$t_{1/2\lambda z}$ (h), arithmetic mean (SD) *n*	5.8 (2.4) 46	5.8 (3.5) 42	5.1 (2.6) 43	4.9 (2.9) 37	19.5 (5.9) 31	19.8 (7.5) 32
CL/F (l), arithmetic mean (SD) *n*	589 (343) 46	655 (363) 41	N/A	N/A	160 (82.8) 31	125 (72.5) 32
Vz/F (l h^{-1}), arithmetic mean (SD) *n*	4723 (3905) 46	4753 (2790) 41	N/A	N/A	4645 (3233) 31	3617 (2543) 32
MRAUC, arithmetic mean (SD) *n*	N/A	N/A	0.6 (0.9) 42	0.6 (0.9) 36	N/A	N/A
MRC_{max}, arithmetic mean (SD) *n*	N/A	N/A	0.3 (0.7) 46	0.3 (0.5) 43	N/A	N/A

AUC, area under the plasma concentration–time curve from zero to infinity; AUC_{0-t}, area under the plasma concentration–time curve from zero to the last quantifiable concentration; CL/F, apparent plasma clearance; C_{max}, maximum plasma concentration; %GCV, percent geometric coefficient of variation; MRAUC, metabolite-to-parent ratio for AUC; MRC_{max}, metabolite-to-parent ratio for C_{max}; N/A, not applicable; SD, standard deviation; $t_{1/2\lambda z}$, terminal half-life; t_{max}, time of maximum concentration; Vz/F, apparent volume of distribution

[a]Results are based on linear mixed-effects model with treatment as fixed effect and subject as a random effect

[b]Median difference and confidence intervals calculated using the Hodges–Lehmann median estimator. *P*-value for treatment difference in median t_{max} calculated using the Wilcoxon signed rank test

interval ($MRAUC_\tau$) and MRC_{max} for AZ5104 and AZ7550 were approximately 10% of osimertinib.

4β-hydroxy-cholesterol

Following multiple doses of osimertinib, plasma concentrations of 4BHC increased by approximately 10% relative to baseline (Day 1 pre-dose) in the CYP3A study and approximately 15% in the BCRP study, following 4 weeks of osimertinib dosing. Geometric mean (90% CI) post/pre-dose 4BHC concentration ratios were 1.139 (1.10, 1.22) and 1.087 (1.04, 1.19) on Day 24 and Day 31 in the CYP3A study, and 1.147 (1.08, 1.22) and 1.153 (1.08, 1.23) on Day 25 and Day 32 in the BCRP study.

Safety

Mean (standard deviation) total treatment duration of osimertinib in the CYP3A study was 29.3 (2.93) days, with a median of 30.0 days (range 14–35 days). In the BCRP study, mean total treatment duration of osimertinib was 27.4 (3.77) days, with a median of 26.0 days (range 22–47 days); mean of 4.2 (1.78) days for Period 3 (osimertinib plus rosuvastatin). The actual treatment duration (excluding dose interruptions) was similar to total treatment duration in both studies.

The number and percentage of patients with an AE in any category during Part A (see Methods section) is summarized in Table 4. Across treatment periods, 44 patients (85%) in the CYP3A study and 40 patients (91%) in the BCRP study experienced AEs. Of the all causality AEs in both studies, the majority were mild or moderate in severity; three (6%) and seven (16%) reported Grade ≥3 AEs in the CYP3A and BCRP studies respectively, none of which were considered related to study treatment. There were no possibly causally related AEs leading to death or

Table 3

Summary of osimertinib and metabolites pharmacokinetic parameters by study (pharmacokinetic analysis set)

Pharmacokinetic parameter	CYP3A4 study (*n* = 49)			BCRP study (*n* = 44)		
	Osimertinib (*n* = 44)	AZ5104 (*n* = 44)	AZ7550 (*n* = 44)	Osimertinib (*n* = 37)	AZ5104 (*n* = 37)	AZ7550 (*n* = 37)
AUC_{τ} (nM h^{-1}), geometric mean (%GCV)	11 530 (37)	1252 (48)	1119 (38)	15 800 (45)	1655 (62)	1418 (45)
$C_{ss,max}$ (nM), geometric mean (%GCV)	620.1 (34)	62.0 (48)	54.8 (39)	897.9 (47)	86.2 (60)	73.7 (47)
$t_{ss,max}$ (h), median (min, max)	6.0 (0.5, 10.1)	6.0 (0.5, 23.9)	6.1 (0.0, 12.0)	5.0 (2.0, 8.2)	5.0 (0.5, 24.3)	5.1 (1.5, 12.1)
$C_{ss,min}$ (nM), geometric mean (%GCV)	381.7 (39)	44.1 (49)	39.3 (39)	485.6 (47)	54.6 (64)	46.5 (46)
CL_{ss}/F (l h^{-1}), mean (SD)	14.7 (5.1)	N/A	N/A	11.0 (4.2)	N/A	N/A
$MRAUC_{\tau}$, mean (SD)	N/A	0.1 (0.03)	0.1 (0.03)	N/A	0.1 (0.03)	0.1 (0.03)
$MRC_{ss,max}$, mean (SD)	N/A	0.10 (0.03)	0.09 (0.03)	N/A	0.10 (0.029)	0.09 (0.03)

AUC_{τ}, area under the plasma concentration–time curve over the dosing interval; CL_{ss}/F, apparent plasma clearance after multiple dosing; $C_{ss,max}$, maximum plasma concentration after multiple dosing; $C_{ss,min}$, minimum plasma concentration after multiple dosing; %GCV, percent geometric coefficient of variation; $MRAUC_{\tau}$, metabolite-to-parent ratio for AUC_{τ}; $MRC_{ss,max}$, metabolite-to-parent ratio for C_{max}; N/A, not applicable; SD, standard deviation; $t_{ss,max}$, time of maximum concentration after multiple dosing

Table 4

Adverse events (safety analysis set)

n (%)[a]	CYP3A study (*n* = 52)			BCRP study (*n* = 44)		
	Simvastatin (Day 1–2)	Osimertinib (Day 3–30)	Simvastatin + osimertinib (Day 31–32)	Rosuvastatin (Day 1–3)	Osimertinib (Day 4–31)	Rosuvastatin + osimertinib (Day 32–35)
Any AE	11 (21.2)	43 (82.7)	17 (34.7)	10 (22.7)	36 (81.8)	20 (46.5)
Any AE causally related to treatment[b]	0	26 (50.0)	10 (20.4)	1 (2.3)	18 (40.9)	9 (20.9)
Any AE causally related to osimertinib[b]	0	26 (50.0)	10 (20.4)	0	18 (40.9)	8 (18.6)
Any AE causally related to statin[b]	0	2 (3.8)	1 (2.0)	1 (2.3)	5 (11.4)	2 (4.7)
Any AE causally related to both treatments[b]	0	2 (3.8)	1 (2.0)	0	5 (11.4)	1 (2.3)
Any AE of CTCAE grade 3 or higher	0	2 (3.8)	1 (2.0)	2 (4.5)	6 (13.6)	0
Any AE leading to death	0	0	0	0	0	0
Any SAE (including death)	0	3 (5.8)	1 (2.0)	1 (2.3)	1 (2.3)	0
Any SAE causing discontinuation of osimertinib	0	0	0	0	0	0
Any AE leading to discontinuation of osimertinib	0	0	0	0	0	1 (2.3)

AE, adverse event, CTCAE, Common Terminology Criteria for Adverse Events; SAE, serious adverse event

[a]Patients with multiple events in the same category are counted only once in that category; patients with events in more than one category are counted once in each of those categories

[b]AEs assessed by the investigator

discontinuation of osimertinib, simvastatin or rosuvastatin. Two patients died due to disease progression in the BCRP study.

The most common all causality AEs in the CYP3A study were dry skin (grouped term, 11 patients [21%]), rashes and acnes (grouped term, 10 patients [19%]) and diarrhoea

(eight patients [15%]). In the BCRP study they were dyspnoea (11 patients [25%]), decreased appetite and diarrhoea (nine patients [20%] each). In the CYP3A study there was one AE of a cardiac event: a non-serious, Grade 1 event of electrocardiogram QT prolonged that was considered possibly causally related to osimertinib by the investigator. There were no cases of interstitial lung disease reported in either study.

More details on patient safety can be found in the Supporting Information Appendix S1.

Discussion

Based on *in vitro* data, osimertinib was shown to have potential to be an inhibitor and inducer of CYP3A and an inhibitor of intestinal BCRP transport. Hence, we evaluated the impact of osimertinib on the PK of simvastatin, a sensitive CYP3A substrate, and rosuvastatin, a BCRP substrate, in patients with EGFRm NSCLC following progression on an EGFR-TKI. For further details of the *in vitro* data, see the Supporting Information Appendix S1. Baseline demographics in both studies were consistent with other osimertinib clinical trials, except with regard to race in the BCRP study [14, 29, 30].

Simvastatin is particularly sensitive to CYP3A inhibition due to high first-pass metabolism, leading to very low bioavailability [31]. Simvastatin was chosen as the sensitive substrate in the CYP3A study, rather than midazolam, as the study was performed in patients who would be at risk of impaired respiratory function if treated with midazolam [32]. Moreover, the common use of simvastatin in the NSCLC patient population makes the use of simvastatin a more relevant substrate to study the CYP3A interaction potential of osimertinib. In this study, a small decrease in C_{max} of simvastatin and no effect on the AUC of simvastatin, or on the AUC and C_{max} of simvastatin acid (all within the predefined limits) when dosed with osimertinib was observed. Although the decrease in C_{max} was not within the predefined no relevant effect limits, the changes in C_{max} are unlikely to be of clinical relevance as AUC is considered the PK parameter of interest for efficacy of most compounds. Simvastatin acid, which is also formed predominately via CYP3A in the liver, showed no effect after osimertinib treatment; therefore, no clinically meaningful impact on CYP3A substrate exposure is expected when co-dosed with osimertinib. This lack of change in the PK of simvastatin and simvastatin acid suggests that there is a lack of effect on CYP3A by osimertinib. These observations support the assumption made from the *in vitro* data that although a potential inhibition of CYP3A4 was shown, this reflects what the CYP3A4 inhibition would be in the intestinal lumen rather than the liver, where free (unbound) osimertinib is more limited. As bioavailability of simvastatin is so low (5%), in comparison to other statins that utilize the CYP3A pathway (such as atorvastatin, bioavailability: 12%), it is probable that other statins that use this pathway are less likely to have any clinically meaningful impact when co-dosed with osimertinib [31].

In the BCRP study, rosuvastatin was chosen as the BCRP substrate as it is another statin that is likely to be co-administered with osimertinib. Rosuvastatin is eliminated mostly through an efflux-mediated process in the gut and in the bile (minimal elimination via metabolism). This study showed an effect on the exposure of rosuvastatin after co-administration with osimertinib; AUC of rosuvastatin was increased by approximately 35% and C_{max} by approximately 72%, compared with the administration of rosuvastatin alone; the 90% CIs of AUC and C_{max} were not contained within the predefined range. These changes are likely due to inhibition of BCRP-mediated efflux by osimertinib during the first pass (osimertinib is not an inhibitor of OATP1B1 or OATP1B3 and does not cause any clinically relevant DDI via this pathway) [15, 33]. Based on our results, the inhibition of BCRP by osimertinib most likely occurs in the absorption/distribution phase, as opposed to the elimination phase. As BCRP is found in both efflux from the blood to the intestines and efflux from the liver to bile ducts to the intestines [34], and rosuvastatin is largely eliminated by faeces [35], it is likely that osimertinib-mediated BCRP inhibition increased rosuvastatin absorption by both blocking efflux into bile, which allowed recirculation into blood, and blocking efflux from blood back to the intestines. This leads to a notable extension of time taken for rosuvastatin to be eliminated through efflux into the gut and, thereby, an increased absorption and/or slower elimination due to reduced efflux by the intestinal mucosa. Though Vz/F was lower with rosuvastatin co-administration, compared with rosuvastatin alone, there was no difference in the half-life of rosuvastatin with and without osimertinib, suggesting that any inhibition of the elimination of the circulating rosuvastatin levels by osimertinib (after first pass) is negligible. The decrease in Vz/F is likely a byproduct of non-compartmental analysis, where because AUC was greater, CL was lower, and thus so too was Vz/F (due to the elimination rate being similar with and without osimertinib); therefore, this result should be interpreted with caution. These small (<2-fold) changes to the PK of rosuvastatin suggest that osimertinib acts as a weak inhibitor of BCRP transporter. This outcome is consistent with the data produced in the *in vitro* studies, in which it was concluded that osimertinib is an inhibitor of BCRP (IC_{50} = 2 μM) and that an 80 mg dose of osimertinib may result in a DDI via the intestine.

4BHC levels were measured in an exploratory capacity in order to gauge the induction potential of osimertinib on CYP3A. In both studies, an increase in 4BHC levels of 10–15% relative to baseline following 28 days of osimertinib administration was observed. As 4BHC is the product of a CYP3A-catalysed reaction, plasma concentrations of 4BHC are expected to increase when CYP3A induction occurs [36]. However, it is important to note that 4BHC has a half-life of approximately 17 days and the length of dosing in these studies was 4 weeks, compared with a dosing period of around 2 weeks in similar studies [37, 38]. Even with a longer dosing duration, this increase was not deemed to be clinically significant and the data reported here suggest a low potential for CYP3A induction.

The exclusion of two patients from the CYP3A study's PK analysis was due to their PK results. Both had higher (~10 fold) simvastatin exposure in Period 1 (simvastatin

alone) compared with all other patients dosed in that period and computed tomography scans prior to study entry indicated significant tumour burden in the liver. By week 6 of the study, there were reductions of approximately 50% and 80% in liver metastases from baseline and the patients returned to within normal simvastatin exposure ranges. It is possible that treatment with osimertinib reduced this tumour burden. A limitation of this study was that due to its fixed sequence design, patients could have clinically improved during the intervening period between the two doses of simvastatin and efficacy determination was not an objective in this study. Therefore, liver function could have been slightly different between the doses as occurred with the two patients discussed here.

In the CYP3A study, steady-state exposures observed for osimertinib and its metabolites were similar to those observed in other osimertinib clinical trials [20]. Slightly higher mean exposures were observed in the BCRP study, but were within the expected exposures of osimertinib across clinical studies; however, overall PK parameter ranges and geometric mean metabolite-to-parent ratios for the metabolites (approximately 10%) were similar to other clinical trials [20]. The higher exposure of osimertinib in the BCRP study may have resulted in increased inhibition of BCRP, potentially presenting an overestimation of the DDI between the two drugs. The numbers of AEs reported here were lower, the majority of AEs were mild or moderate and similar to those reported in the AURA studies [14, 30, 39]. Overall, in both studies, osimertinib was well tolerated in patients with EGFRm-positive NSCLC whose disease had progressed during treatment with an EGFR-TKI and for whom no new safety concerns were identified.

In conclusion, as osimertinib neither induces nor inhibits CYP3A to a clinically relevant extent, PK-mediated interactions are unlikely and hence osimertinib can be used concomitantly with CYP3A substrates. Osimertinib had a <2-fold change inhibitory effect on rosuvastatin exposure; therefore, caution is recommended when using osimertinib with sensitive BCRP substrates with a narrow therapeutic index.

Competing Interests

All authors have completed the Unified Competing Interest form at http://www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author) and declare: J.V. reports honoraria for AstraZeneca, during the conduct of the study. P.A.D. is a former employee of, and shareholder in, AstraZeneca; his current organization provides services to AstraZeneca. K.B. and K.T. declare contract work for AstraZeneca. D.W. is an employee of IQVIA, Clinical Research Organization, which was contracted to execute the two studies on behalf of AstraZeneca. K.S. and K.V. are employees of, and shareholders in, AstraZeneca. The other authors have nothing to disclose. The studies (NCT02197234; NCT02317016) were sponsored by AstraZeneca, Cambridge, UK, the manufacturer of osimertinib.

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Contributions

P.A.D., R.D.H., N.I., N.R.A., K.S., K.T., J.V. and K.V. wrote the manuscript. P.A.D., R.D.H., K.S., K.T., K.V. and D.W. designed the research. T.A., R.D.H., N.I., J.-S.L., N.R.A., J.V. and K.V. performed the research. T.A., K.B., R.D.H., K.S., K.T., K.V. and D.W. analysed the data. K.V. contributed new reagents/analytical tools.

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Supporting Information

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

http://onlinelibrary.wiley.com/doi/10.1111/bcp.13753/suppinfo

Appendix S1 Additional method details
Figure S1 Individual and geometric mean AUC. A, simvastatin *vs*. treatment. B, simvastatin acid *vs*. treatment (pharmacokinetic analysis set)
Figure S2 Individual and geometric mean AUC of rosuvastatin *vs*. treatment (pharmacokinetic analysis set)
Table S1 Interpretation of osimertinib CYP inhibition data and evaluating the potential for DDI based on *in vitro* data
Table S2 Evaluating the potential for DDI of osimertinib based on *in vitro* data and static modelling