An open-label, single-dose bioavailability study of the pharmacokinetics of CAT-354 after subcutaneous and intravenous administration in healthy males

Chad K. Oh,[1] Raffaella Faggioni,[1] Feng Jin,[1] Lorin K. Roskos,[1] Bing Wang,[1] Claire Birrell,[1] Rosamund Wilson[2] & Nestor A. Molfino[1]

[1]MedImmune, LLC, Gaithersburg, MD, USA and [2]SPICA Consultants, Ltd, Marlborough, UK

WHAT IS ALREADY KNOWN ABOUT THIS SUBJECT

• The safety profile and effective dose of CAT-354 have been determined with intravenous (i.v.) administration, but no information is available on subcutaneous (s.c.) administration.

WHAT THIS STUDY ADDS

• This study characterized the pharmacokinetics of CAT-354 following s.c. administration of 150 mg and 300 mg doses, and indicated a bioavailability of approximately 60%.

AIM

To assess the bioavailability and pharmacokinetics of CAT-354, an anti-IL-13 human monoclonal IgG4 antibody, following subcutaneous (s.c.) and intravenous (i.v.) administration.

METHODS

This was a single-dose, randomized, open-label, parallel-group bioavailability study. Healthy male subjects aged 20–54 years were randomly assigned to one of three dose groups ($n = 10$/group) to receive CAT-354: 150 mg i.v.; 150 mg s.c. or 300 mg s.c. (two 150 mg injections). Serum pharmacokinetics, adverse events (AEs), vital signs, electrocardiograms and laboratory parameters were assessed.

RESULTS

CAT-354 showed bioavailability of 62% and 60% after 150 mg and 300 mg s.c. doses, respectively, and linear pharmacokinetics over the dose range tested. Peak serum concentrations in the s.c. groups occurred after 3–9 (median 5) days, with a mean elimination half-life of 19.2 ± 3.1 days (150 mg) and 19.4 ± 3.59 days (300 mg) after s.c. and 21.4 ± 2.46 days after i.v. administration. Volume of distribution at steady state (V_{ss}) was 4960 ± 1440 ml kg^{-1} after i.v. (slightly greater than plasma volume). Average apparent clearances (CL/F) were 292 ± 82.3 and 307 ± 109 ml day^{-1} after 150 and 300 mg s.c., respectively; systemic CL of 188 ± 84.0 ml day^{-1} after i.v. dosing was consistent with endogenous IgG and reticuloendothelial elimination. No severe or serious AEs occurred. Among 40 reported AEs, 25 were headache, sinus disorders/respiratory symptoms and changes in body temperature perception.

CONCLUSIONS

CAT-354 exhibited bioavailability of approximately 60% when given s.c. to healthy male subjects.

Introduction

Asthma is characterized by airway hyper-responsiveness (AHR), airway inflammation and underlying structural changes to the airways. Almost 30% of the study population in the Gaining Optimal Asthma controL (GOAL) study failed to maintain control despite regular use of high-dose fluticasone and salmeterol [1]. Omalizumab, a humanized monoclonal antibody that binds to immunoglobulin E (IgE), has been shown to decrease asthma exacerbations and corticosteroid requirements and reduce hospitalizations and emergency room visits in moderate-to-severe allergic asthmatics [2]. Despite these benefits, however, approximately 40% of allergic asthmatics failed to have an optimal clinical response [2]. This suggests that treatment with corticosteroids or IgE pathway blockade may not be sufficient to control some asthmatics and indicates that there is clearly an unmet need for the effective and safe treatment of asthma.

The pathogenesis of asthma involves cytokines produced by the T-helper-2 (TH-2) cells that express interleukin (IL)-4, IL-5 and IL-13 [3, 4]. IL-13 receptors are expressed on a multitude of cell types, and the action of IL-13 on these cells is to promote the acute inflammatory process, AHR and underlying structural changes to the airways [5]. In animal models, IL-13 has been shown to be both necessary and sufficient to induce a phenotype that closely resembles allergic asthma [6].

Elevated IL-13 production has been detected in the airways and sputum of patients with asthma [7, 8], and mast cell-derived IL-13 (as well as IL-4) has been demonstrated in airway smooth muscle of patients with asthma [5, 9]. Animal model data show that IL-13 causes AHR, eosinophilic inflammation and mucus hypersecretion [6, 10–13]. Taken together, these findings suggest an important role for IL-13 in the pathophysiology of asthma. Therefore, antagonizing the function of IL-13 may offer therapeutic benefit to patients with asthma.

CAT-354 is a human monoclonal IgG4 antibody which has been shown to neutralize IL-13 in both the murine airway [14] and human lung mast cells [15]. Phase 1 data in subjects with mild-to-moderate asthma have demonstrated safety and tolerability profiles that support continued clinical development of CAT-354 [16]. However, all human studies to date have utilized an i.v. infusion of CAT-354 over a period of 30 min. CAT-354 has demonstrated good bioavailability, linear pharmacokinetics over the dose range tested and an acceptable safety profile and tolerability after i.v. administration in another phase 1 study in patients with mild-to-moderate asthma [16]. Simpler injection protocols of s.c. administration may offer patients advantages in ease of use.

The primary objective of this single-dose study in healthy male subjects was to assess the bioavailability of single-dose administration of CAT-354 by s.c. and i.v. routes at two s.c. dose levels (150 mg and 300 mg) in comparison with that following a single 150 mg i.v. dose. Additional objectives were to compare other measures of the pharmacokinetics of CAT-354 and to assess the tolerability, safety and immunogenicity profiles of these single i.v. and s.c. doses of CAT-354 in these subjects.

Compartmental pharmacokinetic modelling was also conducted to characterize further the absorption kinetics and define pharmacokinetic parameters to support future modelling and simulation of repeat-dose s.c. regimens.

Methods

Subject eligibility

This study was conducted at a single centre in the United States, MDS Pharma Services, Lincoln, NE. The study was conducted in accordance with the ethical principles set forth in the Declaration of Helsinki, the International Conference on Harmonisation Guidance for Good Clinical Practice and the US Code of Federal Regulations (21 CFR). The study protocol and informed consent documents were approved by the MDS Pharma Services (US) Inc. Institutional Review Board prior to subject enrolment.

Males aged 19–55 years with a body mass index (BMI) between 18 and 30 kg m^{-2}, inclusive, who had no clinically significant abnormality on clinical examination, medical history, electrocardiogram (ECG), clinical chemistry, haematology or urinalysis results (negative screen for drug abuse and alcohol use was also required) were eligible to participate in this study.

Subjects were excluded from participation if they had experienced any acute illness in the 2 weeks before study day 0 (study visit 2) or any active concomitant disease (including psychological disorders), had donated blood or had significant loss of blood within 56 days (or donated plasma within 7 days) of study day 0, had a positive test for or received treatment for hepatitis B, hepatitis C, HIV and/or other immunodeficiency disorders, or had a history of receiving any medication that might have had carryover effects into this study. Specifically, subjects who had previously received any monoclonal antibody or similar related protein that might have sensitized them to CAT-354 were excluded. In addition, subjects could not have participated in another investigational product study within 3 months of study day 0 (or five half-lives of the previously administered investigational product, whichever was longer).

Study design

This was a phase 1, single-dose, randomized, open-label, parallel-group bioavailability and pharmacokinetic study of CAT-354 in healthy male subjects. The study design is summarized in Figure 1.

Following confirmation of eligibility and completion of a 14-day screening phase, subjects were randomly assigned at the second study visit to one of three dose
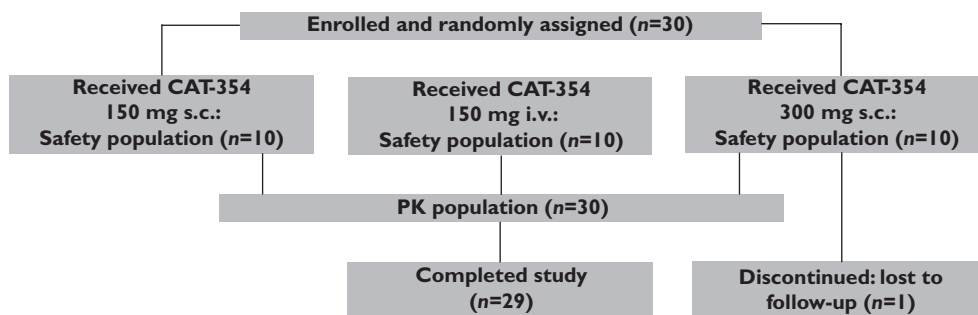
Figure 1

Flowchart of subject assignment and treatment through the study

groups: 150 mg CAT-354 given i.v. (one 30-min infusion), 150 mg CAT-354 given s.c. (one injection into the thigh) or 300 mg CAT-354 given s.c. (one injection of 150 mg given into the anterior aspect of one thigh immediately followed by a second injection of 150 mg into the anterior aspect of the contralateral thigh). Doses of the assigned treatment were administered at visit 2, following screening and randomization. Subjects remained in the study for assessments of safety characteristics and blood sampling for pharmacokinetics and immunogenicity until at least day 56 post-dose.

CAT-354 was manufactured on behalf of MedImmune under current good manufacturing practice, supplied by MedImmune in identifiable bulk packs containing medication vials and labelled in accordance with good clinical practice and any other local regulatory requirements. CAT-354 was formulated at a nominal concentration of 150 mg ml^{-1} in an acetate buffer (pH 5.5). It was presented as a sterile, opalescent solution in 2-ml clear glass vials, each containing a nominal volume of 1 ml.

Study procedures

Pharmacokinetic and immunogenicity assessments Venous blood samples for quantitation of CAT-354 in serum were obtained before dosing, at the end of infusion (~30 min) or immediately following injection(s), at 30 min (± 10 min), 1 h (± 10 min), 3, 8 and 24 h (± 30 min), and at 3, 5, 7, 9, 14, 21, 28, 35, 42 and 56 days post-infusion stop/injection. Additional blood samples were collected from all subjects before dosing and at day 56 for CAT-354 immunogenicity determination. The total blood volume drawn from an individual subject for all assessments during the study was less than 100 ml. Each serum sample was divided into three aliquots and stored at −80°C until shipped to MedImmune for analysis.

Pharmacokinetic assay The concentration of CAT-354 in the serum samples was determined using a qualified immunoassay performed on the Gyrolab assay platform (Gyros AB, Uppsala, Sweden) as follows: biotinylated CAT-375 (antibody against the idiotypic region of CAT-354) was

captured onto streptavidin-coated columns of the Gyros compact disc. Prediluted samples and CAT-354 standards were flowed through the columns. CAT-354 that bound to the immobilized CAT-375 was quantified with a sheep anti-human IgG4 antibody labelled with Alexa Fluor® 647 (Invitrogen Corp., Carlsbad, CA) and detected by fluorescence. The CAT-375 anti-idiotype antibody only binds to free CAT-354 (i.e. CAT-354 where one or both variable regions are not bound to serum IL-13); therefore, by design the assay only detects free CAT-354, and gives results equivalent to using IL-13 antigen capture. This method exhibits accuracy of ≤25% absolute relative error and precision of ≤20% coefficient of variation (CV).

Immunogenicity assays The anti-drug antibody (ADA) screening assay was a validated, homogeneous, drug-tolerant, double-bridging immunoassay with detection by electrochemiluminescence (ECL). All samples, including positive and negative controls, were treated with 300 mM acetic acid for 1 h at room temperature, and then neutralized by 1 M Tris-HCl. The acid-treated samples, biotinylated CAT-354, and ruthenium-conjugated CAT-354 were mixed with equal volumes to form an immunocomplex. The ADA-bridged immunocomplex was incubated at 2–8°C overnight. Streptavidin-coated standard MA 2400 96 plates were blocked with MSD Blocker A buffer (Blocker A) for at least 1 h at room temperature. The immunocomplex was captured by the streptavidin-coated standard MSD plate. The ECL signal was captured with an MSD Sector Imager 2400 (all MSD assay components supplied by Meso-Scale Discovery, Gaithersburg, MD). CAT-375 prepared in 100% pooled normal human serum at a concentration of 250 ng ml^{-1} was used as the assay positive control; 100% pooled normal human serum served as the assay negative control. The assay cut point signal-to-background ratio was 1.4 with a 1% false positive rate at the cut point. The assay limit of detection was 244 ng ml^{-1} in 100% pooled normal human serum with 1% false positive rate and 5% false negative rate at the limit-of-detection concentration [17]. The assay was drug tolerant, and could detect 244 ng ml^{-1} and 488 ng ml^{-1} of

CAT-375 in the presence of 1 µg ml^{-1} and 10 µg ml^{-1} CAT-354, respectively.

The confirmatory ADA assay was a validated immunoassay conducted using the same immunoassay format as the screening assay. Samples that were positive in the screening assay were pre-incubated with and without 300 µg ml^{-1} CAT-354. Assay positive controls were 250 ng ml^{-1} of CAT-375 with and without 300 µg ml^{-1} CAT-354 in 100% pooled normal human serum; 100% pooled normal human serum with and without 300 µg ml^{-1} CAT-354 was used as the assay negative control. The confirmatory cut point was calculated based on the percent inhibition of signal by CAT-354 pre-incubated with 50 normal human serum samples and 10 asthma patient serum samples. With a 1.0% false-positive error rate, the confirmatory cut point was 38.8%. Samples with a percent inhibition greater than or equal to the cut point would be confirmed positive for the presence of ADA to CAT-354.

Noncompartmental analysis Pharmacokinetic parameters for CAT-354 in serum were estimated using noncompartmental methods with WinNonlin® (version 5.2) software (Pharsight Corporation, St Louis, MO). WinNonlin® noncompartmental models 202 and 200 with default settings were used to analyze the i.v. and s.c. serum pharmacokinetic data, respectively. Pharmacokinetic modelling and simulation were performed using the NONMEM software program, Version VI, Level 1.0 (Regents of the University of California, Oakland, CA).

The AUC(0,τ) where τ = 56 days, was estimated by the linear and log-linear trapezoidal rules. The AUC(0,∞) was estimated by extrapolating the concentration–time curve from time zero to infinity. After i.v. or s.c. administration, the maximum observed concentration (C_{max}) and time of C_{max} (t_{max}) were recorded as observed. Systemic clearance (CL) after i.v. dosing and apparent clearance (CL/F) after s.c. dosing were calculated as Dose/AUC(0,∞). Volume of distribution at steady state (V_{ss}) after i.v. dosing was estimated by the formula V_{ss} = mean residence time from time 0 to infinity (MRT(0,∞)) CL, where MRT(0,∞) = AUCM(0,∞) (area under the moment curve)/AUC(0,∞). The elimination rate constant, λ_z, was determined by least-squares regression of the log-transformed concentration data using the terminal phase, identified by inspection. The elimination half-life was calculated as: $t_{1/2}\lambda_z = \ln 2/\lambda_z$.

Compartmental modelling

All pharmacokinetic data were fitted simultaneously using the first-order conditional estimation plus interaction (between inter-/intra-individual and residual variability) method. Data processing and graphical analyses were done using S-PLUS (version 7.0, Insightful/TIBCO, Palo Alto, CA).

A population pharmacokinetic model that involved a linear, two-compartmental design was developed to characterize the disposition of CAT-354. During model development, various model structures (one and two compartments; one, two and three absorption compartments; with or without absorption lag time) were sequentially evaluated. The final model was selected based on comparison in diagnostic plots and the objective function values between different structural models. In the final model, for s.c. groups, the absorption was described with two absorption compartments, one of which had an absorption lag time. Parameters that characterize the fraction of the dose that enters into both absorption compartments were also included into the final model. The whole model was described by the following equations:

$$\frac{dA_{ab1}}{dt} = -k_{a1} \times A_{ab1} \tag{1}$$

$$\frac{dA_{ab2}}{dt} = -k_{a2} \times A_{ab2} \tag{2}$$

$$\frac{dA_{central}}{dt} = k_{a1} \times A_{ab1} + k_{a2} \times A_{ab2} - CL \times C_{central} + Q \times C_{peripheral} - Q \times C_{central} \tag{3}$$

$$\frac{dA_{peripheral}}{dt} = Q \times C_{central} - Q \times C_{peripheral} \tag{4}$$

A_{ab1} and A_{ab2} represent the amount of CAT-354 present in two separate absorption compartments (ab1 and ab2) after s.c. administration. Fractions of CAT-354 absorbed from both compartments are set as: Fraction Abs1 + Fraction Abs$_2$ = 1. An absorption lag time (t_{lag}) in ab2 was also incorporated to describe the absorption delay. $A_{central}$, $A_{peripheral}$, $C_{central}$ and $C_{peripheral}$ represent the amount and concentration in both central and peripheral compartments, respectively. The concentrations in the central and peripheral compartments were calculated by dividing the amounts in the central and peripheral compartments by the central volume of distribution, $V_{central}$, and the peripheral volume of distribution, $V_{peripheral}$, respectively. CL and Q represent systemic clearance and distributional clearance between central and peripheral compartments.

Interindividual variability for the fixed-effect parameters was modelled using an exponential error term:

$$\theta_i = \theta \times \exp(\eta_i) \tag{5}$$

where θ_i is the ith individual's variable, θ is the typical value of the population parameter, and η_i represents the individual-specific random effects for the ith individual parameter symmetrically distributed with zero mean and variance ω^2. A combination of proportional and additive models was used to characterize the residual error variability:

$$Y_{ij} = F \times (1 + \varepsilon_{ij1}) + \varepsilon_{ij2} \tag{6}$$

where Y_{ij} represents the jth observation for the ith subject, F is the predicted concentration and ε_{ij1} and ε_{ij2} represent the proportional and additive residual error, respectively.

To evaluate the model results, visual predictive checks were performed to simulate the pharmacokinetic profiles using the parameter estimates generated from the original data. The 5th, 50th and 95th quantiles were calculated from the simulated profiles for the predictive checks and were compared with the raw data to allow assessment of model predictability.

Safety assessments

The safety of subjects in this study was monitored through ongoing assessments of local tolerability, vital signs, clinical laboratory tests, physical examinations, 12-lead ECGs, adverse events (AEs) and serious adverse events (SAEs), and CAT-354 immunogenicity testing. All subjects who were randomly assigned to treatment and received CAT-354 were included in the safety population.

Safety observations were recorded from the time informed consent was obtained until 56 days post-dose. Venous blood samples were obtained for haematology and biochemistry testing at screening and at visits 5, 8, 10 and 12 (termination). Vital signs were assessed at screening and at visits 2, 5, 8, 10 and 12. ECGs were performed at screening and at visits 2 and 12.

All AEs and SAEs were described using MedDRA (Medical Dictionary for Regulatory Activities) version 11.0 and graded by severity (mild, moderate, severe) and attributional relationship to CAT-354 (none, remote, possible, probable, definite).

Statistical analysis

The number of subjects studied was based on the desire to obtain adequate pharmacokinetic data whilst exposing as few healthy males as possible to CAT-354 and study procedures. A total of 24 evaluable subjects completing the study (8 per group) was considered sufficient to provide adequate information.

Log-transformed dose-normalized $AUC(0,\infty)$ ($AUC(0, \infty)/D$) was analyzed using WinNonlin® (version 5.2) software with an analysis of variance (ANOVA) model, fitting treatment as the main effect. The results of these analyses are presented in terms of geometric least-square means (LSMs) for each treatment, the treatment effect (ratio of each s.c. treatment geometric LSM/i.v. treatment geometric LSM) and the corresponding 90% confidence intervals.

Pharmacokinetic parameters were summarized by dose group using descriptive statistics, including n, mean and standard deviation (SD). For t_{max} only n, min, median, and max were reported. All CAT-354 concentration data were transformed from ng ml^{-1} to μg ml^{-1} for pharmacokinetic analyses and presentation. Final results were rounded and recorded to three significant figures for all pharmacokinetic parameters except t_{max}.

Results

Demographics and baseline characteristics

A total of 30 subjects were enrolled in the study between April and June 2008. All 30 subjects received their planned doses of study medication. One subject in the CAT-354 300 mg s.c. group did not complete the study because he was lost to follow-up, but his results were included in the pharmacokinetic and safety analyses.

All subjects were male and were predominantly white (77%). The mean age of the subjects was 32.5 years, the mean weight was 83.7 kg, the mean height was 180.0 cm and the mean BMI was 25.8 kg m^{-2}. These characteristics were comparable across treatment groups and are summarized in Table 1.

Pharmacokinetics and bioavailability results

Pharmacokinetic parameters for CAT-354 are listed in Table 2. The mean serum concentration–time profiles of CAT-354 over the 56-day study period (with detailed focus on period from infusion/injection through day 7) are illustrated in Figure 2. The highest sustained concentration was achieved with the 300 mg s.c. dose (see Figure 2).

Following i.v. dosing, CAT-354 exhibited biphasic kinetics (see Figure 2). The mean steady-state volume of distribution was 4960 ± 1440 ml (63.6 ± 16.6 ml kg^{-1}), which is about 50% greater than plasma volume. The mean systemic clearance was 188 ± 84.0 ml day^{-1} (2.40 ± 0.984 ml kg^{-1}). The mean elimination half-life was 21.4 ± 2.46 days.

Following s.c. administration, the pharmacokinetics of CAT-354 were linear over the dose range tested. $AUC(0,\infty)$ and C_{max} increased approximately twofold with an increase

Table 1

Demographic characteristics of subjects at study entry

|	Treatment group		
	CAT-354 150 mg i.v.	CAT-354 150 mg s.c.	CAT-354 300 mg s.c.
Number of subjects	10	10	10
Age (years)	30.4 ± 8.34	39.3 ± 8.55	27.8 ± 10.4
Race			
American Indian or Alaska Native	0%	0%	10%
Asian	0%	10%	0%
Black or African American	10%	20%	10%
Other: Arabic	10%	0%	0%
White	80%	70%	80%
Ethnicity			
Hispanic or Latino	10%	0%	10%
Not Hispanic or Latino	90%	100%	90%
Height (cm)	179 ± 7.8	179 ± 6.0	182 ± 5.9
Weight (kg)	78.7 ± 14.7	87.3 ± 8.1	85.3 ± 12.2
BMI (kg m^{-2})	24.6 ± 3.47	27.1 ± 1.86	25.6 ± 2.99

Averages are presented as mean ± SD.

Table 2

Noncompartmental average pharmacokinetic parameters: CAT-354 administered s.c. or i.v.

	Treatment group 150 mg i.v.	150 mg s.c.	300 mg s.c.
Number of subjects	10	10	10
C_{max} (µg ml^{-1})	58.3 ± 14.4	17.1 ± 5.91	36.6 ± 13.1
C_{max}/D (µg ml^{-1} mg^{-1})	0.389 ± 0.096	0.114 ± 0.039	0.122 ± 0.044
t_{max} median (min, max) (days)	0.063 (0.042, 1.02)	5 (3, 9)	5 (3, 9)
AUC(0,56 days) (µg ml^{-1} day)	765 ± 220	467 ± 122	881 ± 287
AUC(0,∞) (µg ml^{-1} day)	903 ± 291	548 ± 143	1080 ± 315
AUC extrapolated (%)	14.3 ± 4.45	14.7 ± 4.98	18.2 ± 10.6
AUC(0,∞)/D (µg ml^{-1} day mg^{-1})	6.02 ± 1.94	3.66 ± 0.953	3.59 ± 1.05
CL or CL/F (ml day^{-1})	188 ± 84.0	292 ± 82.3	307 ± 109
CL or CL/F (ml day^{-1} kg^{-1})	2.40 ± 0.984	3.34 ± 0.828	3.60 ± 1.07
V_{SS} (ml)	4960 ± 1440	ND	ND
V_{SS} (ml kg^{-1})	63.6 ± 16.6	ND	ND
$t_{1/2}$ (days)	21.4 ± 2.46	19.2 ± 3.1	19.4 ± 3.59

Averages are presented as mean ± SD except for t_{max}, which is presented as median (min, max); ND, not determined; C_{max}, maximum observed concentration after infusion/injection; t_{max}, time at which C_{max} is observed after infusion/injection; AUC(0,56 days), area under the serum concentration–time curve from time 0 to study day 56 (last observation); AUC(0,∞), area under the serum concentration–time curve from time 0 to infinity; CL or CL/F, systemic clearance; V_{ss}, volume of distribution at steady state; $t_{1/2}$, terminal phase elimination half-life.
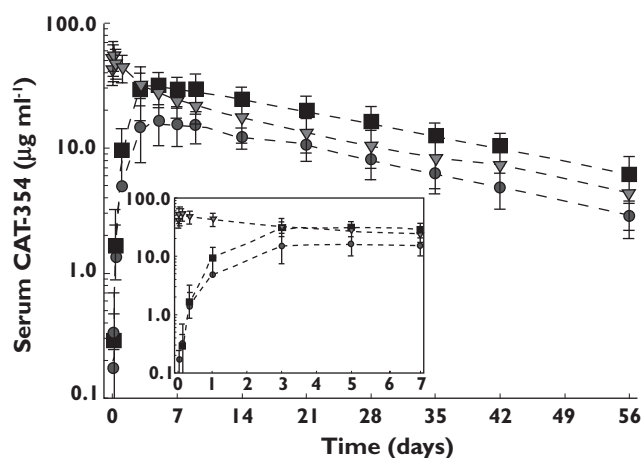
Figure 2

Serum concentration vs. time profile of single doses of CAT-354 at doses of 150 mg s.c. (– ⬤ –), 300 mg s.c. (– ⬛ –), and 150 mg i.v. (– ▽ –) administered by s.c. injection or i.v., infusion

in dose from 150 mg to 300 mg. The percent coefficients of variation (CV%) for AUC(0,∞) and C_{max} were 35% and 26%, respectively, for the 150 mg s.c. dose and 36% and 29%, respectively, for the 300 mg s.c. dose. By comparison, the CV% after the 150 mg i.v. dose were 25% and 32%, respectively, for AUC(0,∞) and C_{max}. Absorption of CAT-354 was slow, with a median t_{max} of 5 days (range, 3–9 days) observed in both s.c. dose groups.

The mean elimination half-life of CAT-354 was 19.2 ± 3.1 days and 19.4 ± 3.59 days following the 150 mg and 300 mg s.c. doses, respectively, which was similar to the half-life after i.v. dosing. The absolute bioavailability of the s.c. doses, as assessed by the geometric LSM ratios of

AUC(0,∞)/D, is presented in Table 3. The bioavailability of CAT-354 was 62% and 60% following the 150 mg and 300 mg s.c. doses, respectively.

Compartmental modelling results

The model-estimated parameters are summarized in Table 4. Figure 3 shows these plots for the 150 mg s.c. (Panel A), 300 mg s.c. (Panel B), and 150 mg i.v. (Panel C) groups. The model predicted mean (50%, solid line) and the 90% prediction interval (5% and 95%, dashed lines) were plotted together with observed data to allow visual predictive checks of the model performance.

The absorption kinetics of CAT-354 were described by parallel mechanisms: (i) a first-order absorption process and (ii) a delayed first-order absorption process. The delayed absorption process accounted for approximately 85% of the absorbed dose. The central volume of distribution was approximately equal to plasma volume. The steady-state volume of distribution was 4830 ml, consistent with the V_{ss} determined by noncompartmental analysis. Clearance and bioavailability determined by compartmental analysis also were similar to the values determined by noncompartmental methods.

Safety assessment results

No AEs were considered by the investigator to be probably or definitely related to CAT-354. Four AEs were assessed as possibly related to CAT-354: three AEs in the 150 mg i.v. treatment group (headache, dizziness and feeling hot), and one AE in the 150 mg s.c. treatment group (feeling cold). Six additional AEs were assessed as remotely related: one AE in the 150 mg i.v. treatment group (headache) and five AEs in the 150 mg s.c. treatment group (three AEs of head-

Table 3

Absolute bioavailability of CAT-354

Parameter	Geometric LSM 150 mg i.v.	150 mg s.c.	300 mg s.c.	Ratio of LSM % (90% CI) 150 mg s.c. : i.v.	300 mg s.c. : i.v.
C_{max}/D (µg ml^{-1} mg^{-1})	0.379	0.109	0.115	28.7 (22.5, 36.6)	30.3 (23.7, 38.6)
$AUC(0,\infty)/D$ (µg ml^{-1} day mg^{-1})	5.70	3.54	3.43	Absolute bioavailability* (90% CI) 62.1 (48.5, 79.6)	60.1 (46.9, 77.1)

*Assessed from geometric LSM ratios of $AUC(0,\infty)/D$. LSM, least-square means; C_{max}/D, dose-normalized maximum concentration after each infusion/injection; $AUC(0.\infty)/D$, dose-normalized area under the serum concentration–time curve from time 0 to infinity.

Table 4

Two-compartment model-estimated parameters for CAT-354 pharmacokinetics

Parameter	Final estimates	SEE (%)
CL (ml day^{-1})	186	13.5
$V_{central}$ (ml)	3060	8.17
Q (ml day^{-1})	294	37.4
$V_{peripheral}$ (ml)	1770	24.9
F (%)	66.2	12.4
t_{lag} (days)	0.245	35.3
k_{a1} (day^{-1})	0.110	221
k_{a2} (day^{-1})	0.270	35.3
Fraction Abs$_2$ (%)	0.847	69.4

CL, systemic clearance; $V_{central}$, volume of central compartment; Q, distributional clearance between central and peripheral compartments; $V_{peripheral}$, volume of peripheral compartment; F, absolute bioavailability; t_{lag}, absorption lag time; k_{a1}, first-order absorption rate constant; k_{a2}, delayed first-order absorption rate constant; Fraction Abs$_2$, fraction of CAT-354 absorbed by the delayed absorption process; SEE, standard error of the estimate.

ache, and one AE each of dizziness and sinus headache). The remaining 30 AEs were assessed as unrelated to CAT-354.

Fourteen subjects reported a total of 40 treatment-emergent AEs. Thus, approximately half of all subjects in all treatment groups experienced at least one AE. The number of subjects experiencing AEs was comparable across treatment groups. There were more AEs reported in the 150 mg i.v. treatment group (17) compared with either of the s.c. treatment groups (150 mg s.c.: 13; 300 mg s.c.: 10). The incidence of all AEs reported by one or more subjects after administration of CAT-354 is summarized in Table 5.

Across all subjects, AEs from the MedDRA System Organ Class (SOC) 'Nervous system disorders' were reported by the largest number of subjects (9/30), and consisted of headache (most frequent), dizziness and sinus headache. More subjects in the 150 mg i.v. (4/10) and 150 mg s.c. (4/10) treatment groups experienced nervous system disorders compared with the 300 mg s.c. treatment group (1/10).

Subjects also reported AEs from MedDRA SOC 'Respiratory, thoracic and mediastinal disorders' (6/30), including paranasal sinus hypersecretion and sinus congestion. Respiratory, thoracic and mediastinal disorders were reported by more subjects in the 150 mg i.v. group (six events, three subjects), compared with the 150 s.c. group (three events, two subjects) and the 300 mg s.c. group (one event, one subject).

Of note, no subject in any treatment group experienced a local reaction to infusion or injection of CAT-354. No deaths or SAEs occurred in any treatment group during the study, and no subject discontinued the study because of an AE.

Immunogenicity testing results

The immunogenicity of CAT-354 was evaluated by analysis of samples collected pre-dose and on day 56. On day 56, the maximum concentrations of CAT-354, which could potentially interfere with detection of ADA, measured in samples from individual volunteers were 8.9 µg ml^{-1}, 4.5 µg ml^{-1} and 10 µg ml^{-1} in the 150 mg i.v., 150 mg s.c. and 300 mg s.c. treatment groups, respectively. The ADA assay could measure less than 500 ng ml^{-1} ADA in the presence of 10 µg ml^{-1} CAT-354; therefore, the ADA assay was sufficiently sensitive and drug tolerant to detect clinically relevant concentrations of ADA on day 56 [18]. All samples were negative for the presence of ADA. All individual pharmacokinetic profiles were consistent with the absence of an ADA response.

Discussion

Despite adequate treatment with inhaled corticosteroids, long-acting β$_2$-adrenoceptor agonists, and other controller medications such as leukotriene antagonists and omalizumab, a significant medical need still remains for alternative treatments for patients with poorly controlled asthma. Alternative therapies for asthma targeting IL-13-expressing cells probably constitute a reasonable goal for advancement in the treatment of asthma.
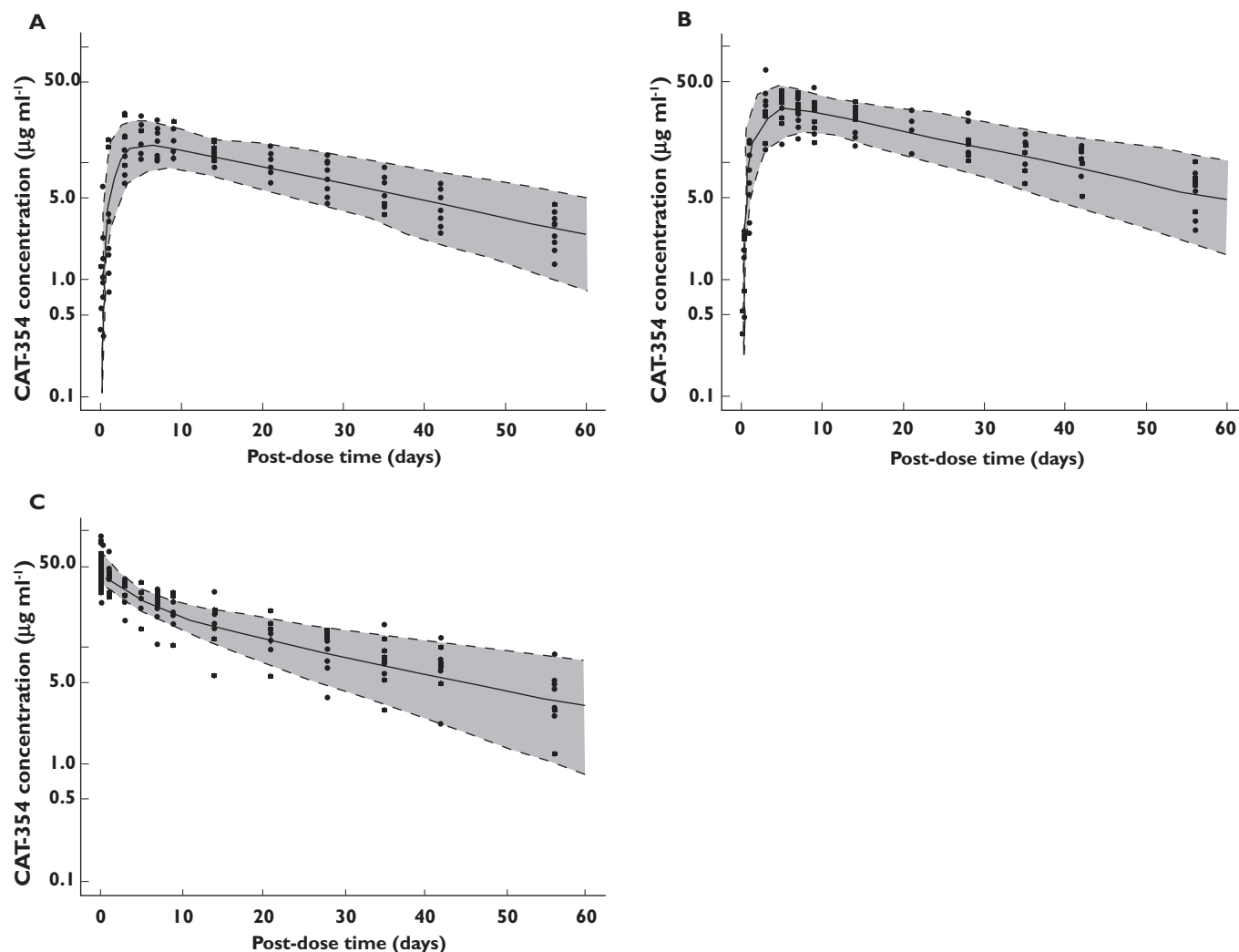
Figure 3

(A) Observed and model-predicted profiles of CAT-354 following 150 mg s.c. administration. Solid line: mean prediction. Dashed lines: 5th and 95th quantiles. Shaded area: 90% prediction interval. (B) Observed and model-predicted profiles of CAT-354 following 300 mg s.c. administration. Solid line: mean prediction. Dashed lines: 5th and 95th quantiles. Shaded area: 90% prediction interval. (C) Observed and model-predicted profiles of CAT-354 following 150 mg i.v. administration. Solid line: mean prediction. Dashed lines: 5th and 95th quantiles. Shaded area: 90% prediction interval

The targeting of IL-13 in the management of pulmonary diseases has been recently reviewed by Blease, with a summary specific to CAT-354 [19]. Early therapeutic potential of CAT-354 for asthma was demonstrated by Blanchard *et al*. via a murine model in which CAT-354 significantly inhibited IL-13-induced AHR and airway eosinophilia [14]. Kaur *et al*. demonstrated that human lung mast cells (HLMCs) expressed IL-13, that IL-13 can up-regulate allergic functions of HLMCs and, specifically, that CAT-354 inhibited all of these IL-13-mediated effects in HLMCs [15].

Preclinical pharmacokinetics and interspecies scaling analyses by Vugmeyster *et al*. suggested that i.v. or s.c. administration of anti-IL-13 antibodies with safe and favourable pharmacokinetic profiles was feasible [20]. Recent data from a study of three dose levels of CAT-354 (1, 5 and 10 mg kg^{-1}) presented by Bhowmick *et al*. demon-

strated linear pharmacokinetics and an acceptable safety profile after multiple i.v. doses in 23 patients with asthma; most AEs observed were mild or moderate and not related to CAT-354 [16]. Overall, the CAT-354 pharmacokinetic parameters demonstrated in the current study were very similar to those of the single-dose pharmacokinetic behaviour of CAT-354 as reported by those authors, although its half-life in that study was reported as somewhat shorter (12–17 days) [16]. Thus, the required characterization of the safety and pharmacokinetic profile of CAT-354 during its development as an asthma therapeutic has been encouraging.

The current study is the first to compare s.c. injection with i.v. infusion of single doses of CAT-354 with regard to bioavailability, safety and pharmacokinetics in healthy subjects. The safety profile of CAT-354 was encouraging in this

Table 5

Summary of all reported adverse events (AEs) by relationship to CAT-354

	Subjects with AEs	Number of AEs	Relationship to CAT-354*				
			None	Remote	Possible	Probable	Definite
150 mg i.v. (n = 10)							
All AEs	5	17	13	1	3	0	0
Headache	4	4	2	1	1	0	0
Back pain	1	1	1	0	0	0	0
Cough	1	1	1	0	0	0	0
Dizziness	1	1	0	0	1	0	0
Eye pruritus	1	1	1	0	0	0	0
Feeling hot	1	1	0	0	1	0	0
Joint swelling	1	1	1	0	0	0	0
Nasal congestion	1	1	1	0	0	0	0
Pain in extremity	1	1	1	0	0	0	0
Pharyngolaryngeal pain	1	1	1	0	0	0	0
Post-traumatic pain	1	1	1	0	0	0	0
Rhinorrhoea	1	1	1	0	0	0	0
Sinus congestion	1	1	1	0	0	0	0
Sneezing	1	1	1	0	0	0	0
150 mg s.c. (n = 10)							
All AEs	5	13	7	5	1	0	0
Headache	4	6	3	3	0	0	0
Paranasal sinus hypersecretion	2	2	2	0	0	0	0
Abdominal pain	1	1	1	0	0	0	0
Dizziness	1	1	0	1	0	0	0
Feeling cold	1	1	0	0	1	0	0
Sinus congestion	1	1	1	0	0	0	0
Sinus headache	1	1	0	1	0	0	0
300 mg s.c. (n = 10)							
All AEs	4	10	10	0	0	0	0
Headache	1	3	3	0	0	0	0
Sunburn	1	2	2	0	0	0	0
Breast mass	1	1	1	0	0	0	0
Breast tenderness	1	1	1	0	0	0	0
Epistaxis	1	1	1	0	0	0	0
Melanocytic naevus	1	1	1	0	0	0	0
Procedural pain	1	1	1	0	0	0	0

*All AEs were described using MedDRA (Medical Dictionary for Regulatory Activities) Version 11.0 and graded by severity (mild, moderate, severe) and attributional relationship to CAT-354.

study, and the most commonly reported AEs (headache, sinus hypersecretion) were similar to those observed by Bhowmick et al. in patients with asthma (nasopharyngitis, headache [also reported by that study's placebo arm]) [16].

The most common AEs overall in the current study were reported with the lowest frequency in the 300 mg s.c. group; this was especially apparent with AEs coded under the MedDRA 'Respiratory, thoracic and mediastinal disorders' SOC, which occurred more frequently in the 150 mg i.v. group (six AEs compared with one AE in the 300 mg s.c. group).

Of particular note was the absence of injection site or infusion reactions observed in this study, together with the absence of immunogenicity demonstrated by CAT-354. Again, a monoclonal antibody administered subcutaneously should ideally produce as few injection-site reactions as possible.

CAT-354 demonstrated linear pharmacokinetics over the dose range tested, with a half-life that was essentially independent of dose and route of administration (ranging between 19.2 and 21.4 days across groups and routes) and an AUC that was dose-proportional within the s.c. route. The systemic clearance and elimination half-life after i.v. dosing were consistent with endogenous IgG, elimination of CAT-354 by the reticuloendothelial system and the absence of an antigen sink [21]. The apparent clearances and elimination half-lives observed were similar between the two s.c. dose groups.

Whilst the AUC associated with the 150 mg s.c. dose was slightly greater than half that achieved with the 150 mg i.v. dose, the 300 mg s.c. dose produced mean and median AUCs that were in fact higher than that achieved by the 150 mg i.v. dose. Since the 300 mg s.c. dose demonstrated a bioavailability of 60%, the simpler route of admin-

istration could provide therapeutic benefit equivalent to that afforded by a 150 mg i.v. infusion and thus an advantage for patients. A comparison of the interpatient pharmacokinetic variability following i.v. and s.c. dosing suggests that s.c. administration will not increase variability compared with the i.v. route.

A bisegmental absorption model has been used to describe the s.c. absorption of other therapeutic proteins; this model reflects direct absorption into the blood and absorption through the lymphatics [22–24]. Compartmental modelling was used in the current study to describe the absorption, distribution and elimination kinetics of CAT-354. After s.c. dosing, the absorption kinetics of CAT-354 were modelled by a two-phase absorption process. The bioavailability, clearance and V_{ss} parameters derived from compartmental modelling were similar to parameters derived by noncompartmental analysis. The compartmental pharmacokinetics model can be used for future simulation of repeat-dose s.c. dosing regimens of CAT-354. CAT-354 exhibited a pharmacokinetic profile consistent with endogenous human IgG, and an absolute bioavailability of approximately 60% when given s.c. to healthy male subjects.

In conclusion, CAT-354 showed good bioavailability and a good tolerability profile in this study of healthy male subjects, with encouraging results shown with s.c. administration. The similar safety profile demonstrated by this study to that already demonstrated by CAT-354 in patients with asthma [16] might be expected to predict an acceptable safety profile in larger efficacy studies that would provide further confirmation of the bioavailability results demonstrated in the current study.

Competing interests

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